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A REVIEW AND EVALUATION  
OF THE AMPHIBIAN  
TOXICOLOGICAL LITERATURE

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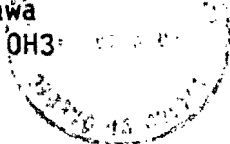


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## ABSTRACT

The impacts of environmental contaminants on amphibian populations have not been extensively studied. Amphibians, with the exception of a few specialized species, utilize both the terrestrial and aquatic ecosystems during their life cycles.

This report reviews the published literature on the effects of environmental contaminants on amphibians. Acute toxicity studies, other laboratory studies, field studies and residue data are summarized in Tables 1, 2, 3, and 4 respectively.

Amphibians are particularly sensitive to metals and to acidification. They are considered to be useful indicator species for measuring the effects of local changes in environmental studies. Several species are readily reared in the laboratory so that experimental studies can be undertaken.

## RÉSUMÉ

Les répercussions des contaminants sur les populations amphibiennes n'ont pas été étudiées à fond. Les amphibiens, à l'exception de quelques espèces spécialisées, ont besoin des deux écosystèmes durant les étapes de leur vie.

Ce rapport passe en revue les écrits sur les effets des contaminants environnementaux sur les amphibiens. Des études de pointe sur la toxicité, différentes recherches de laboratoire, des observations sur le terrain et des données sur les résidus sont relevés sommairement dans les tableaux 1, 2, 3 et 4 respectivement.

Les amphibiens sont particulièrement vulnérables aux métaux et à l'acidification. Ils sont considérés comme des sujets témoins tout indiqué pour mesurer les effets des changements locaux dans les études environnementales. On peut facilement en élever plusieurs espèces en laboratoire à des fins de recherches expérimentales.



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## I INTRODUCTION

The impact of environmental contaminants on amphibians is a subject that has received limited attention. This review is an attempt to present the toxicological data that exists. The majority of the toxicological research conducted using amphibians is centered around the anurans and urodeles. The apodans, due to a lack of knowledge concerning the biology and secretive lifestyle have not been studied in relation to toxic chemicals.

Amphibians, with the exception of a few specialized species, require moisture in some form, whether it be a permanent lake or leaf that has collected dew, in order to complete their life cycle. It is this requirement and their subsequent metamorphosis to the combined terrestrial/aquatic adult stage that is the major route of exposure for amphibians to toxic contaminants. Thus, in order to fully comprehend the possible implications of environmental contaminants on amphibians a brief discussion of their life cycle is necessary.

Amphibians fall into three general categories with respect to their breeding habitats. The first category is composed of those that breed in permanent water; the second breed in temporary or seasonal pools; and the third group breeds out of the water (Porter 1972). The latter group still requires a moist location to deposit its eggs in order to ensure the development of the embryos. Thus all amphibians rely upon seasonal and geographical patterns of precipitation which govern their distribution (Romer 1959, Porter 1972). In general, the amphibians of North America breed in spring or early summer (Romer 1959, Porter 1972). The rate of development of the eggs and embryos is species dependent. Eggs deposited in seasonal pools or other moist areas develop more quickly to avoid dessication. Eggs laid in permanent water bodies need not develop as fast due to the unlikelyhood of dessication (Porter 1972).

Anurans congregate in leks in a pond where the males call to attract reproductively active females. The female, upon hearing the mating call of her species, will move in the direction of the call. The male mounts the female and clasps her inducing the deposition of the eggs, which are immediately fertilized. The number of eggs deposited by a female during a breeding season, ranges from a single egg (Genus Sminthillus) to 30,000 or more (Genus Bufo) (Porter 1972). Anuran egg masses are of three generalized types and positions enabling them to maximize oxygen availability in different habitats and water temperatures. One type is deposited as a flattened egg mass at the surface of the water such as those of R. catesbeiana; the second type is globular and attached to submerged vegetation as in P. triseriata egg masses; and the third type is characteristic of the true toad species (Genus Bufo), and are deposited in elongated strings (Otto and Towle 1947, Romer 1959, Porter 1972). Depending upon the species, the eggs take between 2 and 30 days to hatch.

Other species exhibit more specialized breeding behaviour. Some female anurans protect their eggs by carrying them on their backs or under the skin. Others build nests while some lay eggs enveloped in a gelatinous material that hardens and reduces moisture loss. For the most part these specialized adaptations are found in amphibians inhabiting the tropics, while the generalized life cycle described here is more characteristic of the species located in North America (Villemant et al. 1958).

The embryo hatches out as a larvae equipped with a well developed tail for swimming. This aids it in foraging for submerged vegetation upon which it browses (Villemant et al. 1958, Romer 1959, Porter 1972). The anuran larvae respire by gulping water and forcing it over the gills located in the future neck region. Some gaseous exchange is also accomplished through the skin (Romer 1959, Porter 1972). The larvae or tadpoles transform to the adult stage via metamorphosis. This occurs two weeks to three years after hatching, depending upon the species (Porter 1972).

Metamorphosis involves radical structural changes of the body: the gills disappear, lungs and limbs develop rapidly (hind limbs first), the tail is absorbed and the digestive tract shortens. Eventually the adult frog becomes an air breather (Romer 1959). Respiration is accomplished via internal lungs as well as via the skin.

The length of the metamorphic period is also species dependent. In the case of the Spadefoot toad (Scaphiopus holbrookii), metamorphosis into a terrestrial toad takes only twelve to thirteen days from fertilization. Spadefoot toads breed in temporary pools and must develop quickly to avoid dessication (Porter 1972). Some species require a year, (R. catesbeiana) (Cecil and Just 1979), while others require still more time. Several amphibian species are non-metamorphosing and retain both larval characteristics and their aquatic lifestyle (genus Necturus) (Porter 1972).

Adult anurans, unlike the larvae, are carnivorous, consuming almost any moving prey of the right size. This includes worms, insects, other amphibians, reptiles and occasionally mammals. Throughout its life cycle an anuran can occupy several different positions within the food web (Curtis 1968, Porter 1972).

The urodeles also follow a general pattern of breeding behaviour. During courtship the male either clasps or blocks the path of the female and begins lashing with his tail, bumping or rubbing to stimulate the female. The male then deposits a spermatophore (a capsule containing sperm). The female either follows the male or is pulled by him into a position where she can pick up the spermatophore



with the cloaca, where it is stored (Romer 1959). The eggs are fertilized internally and may be deposited in decaying vegetation, logs, pools or other moist environments. The eggs can take up to two months to hatch. Aquatic salamander larvae differ from anuran larvae in that they develop adult appearance quickly, they possess external gills and their diet is primarily insectivorous (Romer 1959). After several months and under the correct environmental conditions the larvae transform into the adult stage and may leave the water. Some species have lungs whereas the Plethontidae, known as the "lungless salamanders", respire via the skin. Another group, the Necturidae, are neotonic, retaining their external gills and aquatic lifestyle even after sexual maturity.

Adult urodeles, like adult anurans, are carnivorous. Their diet consists of arachnids, annelids, insects, amphibians, small fish and their eggs, and in the case of Cryptobranchus, mammals. Adult urodeles inhabit moist locations under logs, rocks, and leaf litter, returning to the water in spring to breed (Porter 1972).

Interruptions of this life cycle by toxic contaminants can have disastrous effects on amphibian populations (Paulov 1977). Amphibians are of significant economic importance to man. Due to their carnivorous adult stage they consume insect pests harmful to crops (Schwabe 1977). Some cultures use amphibian skins for leather goods, amphibian venom for hunting and medicines, while frogs legs (R. catesbeiana) are considered a delicacy by some humans. Amphibians are also the perennial biological specimen in institutions due to their relative abundance and availability (Otto and Towle 1947). Several species have actually benefitted from man's agricultural practices by taking advantage of irrigation ditches and new habitats that have been created, but many more species have been detrimentally affected by habitat destruction and the use of toxic chemicals. Amphibians are not only important to man, but also to other animals within the community. They make major contributions to community biomass (Burton and Likens 1975a, 1975b, Cecil and Just 1979, Debendictis 1974), as well as playing significant roles in competitive and predator-prey relationships (Orser and Shure 1972, Burton and Likens 1975b, Lynch 1979). It is of interest to note that amphibians are the only class of vertebrates that do not include any pests or species harmful to man. Amphibians do not compete for harvests, nor are there any ferocious or destructive species (Porter 1972). Even so amphibians are victimized by man's attempts to control pests with toxic chemicals, primarily insecticides and herbicides (Fashingbauer 1957, Hazelwood 1969, Porter 1972, Cooke 1973a, Curtis 1968).

The toxicity of these contaminants to amphibians in lab bioassays and field experiments as well as their general effects on populations, will be reviewed in order to identify specific substances that pose

potential hazards to amphibians. From this information an evaluation will be made as to whether amphibians can be employed as useful indicators of environmental quality.

## II INSECTICIDES

### a) ORGANOCHLORINES

#### ALDRIN AND DIELDRIN

##### Short-Term Effects

In static bioassays dieldrin was highly toxic to B. woodhousii tadpoles with a 96 hour LC50 of 0.15 mg/L (Sanders 1970) (Table 1).

In static bioassays, complete mortality of adult R. cyanophlyctis (an Asian frog species) occurred within 215 minutes of exposure to 0.006 mg/L aldrin (Rane and Mathur 1978). Although sample sizes were small (2 to 4 frogs per treatment with 6 different treatments) no frogs survived longer than 120 minutes when exposed to 0.125 mg/L (the highest concentration tested) (Table 2). Preceding death, the frogs swam erratically exhibiting a loss of equilibrium. A coagulation of mucous occurred over the body with the belly becoming bloated and the skin turning pale (Ibid 1978). These results indicate that aldrin is very toxic to the adults of R. cyanophlyctis and that the 96 hour LC50 would be much lower than 0.006 mg/L considerably less than the 96 hour LC50 reported for B. woodhousii by Sanders (1970) (Table 1).

Mulla (1962, 1963) found R. catesbeiana tadpoles experienced 100% mortality within 24 hours when maintained in ponds and treated with 0.1 kg/ha dieldrin (Table 3). Since the actual concentrations of dieldrin in the ponds were not measured it is difficult to compare these results with other studies.

##### Long-Term Effects

Short-term exposures to dieldrin can have long-term effects on developing amphibians. When groups of 150 Limnodynastes tasmaniensis (an Australian frog) embryos were exposed to 0.0, 0.01 and 0.1 mg/L dieldrin for 7 hours, the 0.1 mg/L exposure resulted in accelerated growth and abnormalities later in development (Brooks 1981) (Table 2). No mortality occurred during the exposure, but 21 of the tadpoles treated with 0.1 mg/L dieldrin as eggs, exhibited some degree of deformity later in development. Nineteen days after exposure some

severe cases had cephalic pigmentation disruption resulting in two large transparent spots on the head of the tadpole. These spots were swellings situated over the otolith which was also abnormal. The affected tadpoles also had constricted guts indicating lack of feeding. In general the tadpoles exposed to 0.1 mg/L tended to feed less and frequently rested on the bottom of the aquaria (Ibid 1981).

At a concentration of 0.30 ppm aldrin in solution, R. pipiens suffered 40% mortality after thirty days (Kaplan and Overpeck 1964) (Table 2). In the same study at 0.10 ppm dieldrin in solution the frogs suffered 50% mortality after thirty days (Table 2). Neuromuscular changes were evident in all insecticide concentrations studied, but were more pronounced in aldrin and dieldrin. These changes included coarse tremors, rigidity, abnormal reactions to stimulation, convulsions and a reduction in the ability to orient themselves (Kaplan and Overpeck 1964).

Vinson et al. (1963) examined a number of populations of A. crepitans and A. gryllus to determine if these species had developed cross-resistance to aldrin after being exposed to DDT, toxaphene, methyl parathion and endrin. Six populations of A. crepitans from six localities in Mississippi were chosen, with three localities being free of pesticides and three heavily treated with pesticides in the past fifteen years. Adult frogs were exposed to 1 ml of either 30,000 mg/L or 50,000 mg/L aldrin. The aldrin was pipetted onto filter paper and then the adult frog was placed on the paper for 36 hours. For A. crepitans, mortality varied from 0% at both 30,000 and 50,000 mg/L for individuals from populations that had been treated with other pesticides, to 30% mortality at 30,000 mg/L, and 57% mortality at 50,000 mg/L for individuals from populations that had never been treated with other pesticides. These results indicate a cross-resistance to aldrin which was thought to have occurred by natural selection for a physiological mechanism to confer resistance to pesticides. This resistance was non-specific enough to be effective against aldrin (Ibid 1963) (Table 2).

## Residues

Aldrin can be bioaccumulated by amphibians and passed up the food chain (Korschgen 1970). Korschgen (1970) measured aldrin residues in soils, plant and animal tissues from a Missouri corn field that had been treated with aldrin at 1 kg/ha for at least 15 years. Adult B. americanus had 0.03 mg/kg aldrin (wet weight) although young of the year had no measurable aldrin residues. The garter snake (Thamnophis sirtalis), a species that preys upon B. americanus, had very high residue levels ranging from 10.3-14.4 mg/kg indicating that residues bioaccumulate as they are passed up the food web (Ibid 1970) (Table 4).

Niethammer et al. (1984) also found evidence of accumulation up the food chain in an area of Louisiana characterized by oxbow lakes. This area had been subjected to heavy organochlorine pesticide use due to the fertile alluvial soil in the area. Ninety-five percent of all animals tested including R. catesbeiana, R. pipiens and the Bronze frog had residues of one or more organochlorine insecticides (Table 4). Ohlendorf et al. (1981) reported a die-off of herons (Ardea herodias) probably due to insecticide poisoning. Frogs represent a large percentage of a heron's diet and thus, even though frogs appear to be able to mobilize and excrete these insecticides, there is the danger of serious effects occurring through bioaccumulation (Niethammer et al. 1984).

Collections from agricultural areas in Iowa revealed residues of dieldrin and other organochlorine insecticides in the viscera of B. bufo and R. pipiens (Punzo et al. 1979) (Table 4). The values were low or absent due to the method of analysis employed and thus may not represent accurate values contained in the amphibians from this area (Punzo et al. 1979).

Aldrin and dieldrin's moderate water solubility was used by Kaiser and Dunham (1972) to explain the passage of these two insecticides through R. pipiens skin, explaining the rapid excretion of these compounds by amphibians.

### **Mechanism for Toxicity**

The primary site of action of aldrin-transdiol (the active metabolite of dieldrin) is somewhere in the central nervous system (Akkermans et al. 1975b). In preparations of spinal cords of X. laevis, Akkermans et al. (1975b) measured an augmentation of polysynaptic reflex activity and a reduction of orthodromic postsynaptic inhibition in dieldrin poisoned toads. Direct application of aldrin-transdiol to spinal cord preparations resulted in a potentiation of spinal reflex activity and a marked reduction of spinal inhibitory mechanism (Table 2). These excitatory effects were followed by a reduction of spinal excitability. Similar effects have been measured on the motor end-plate of the sartorius nerve with increased synaptic transmission (Akkermans et al. 1974) (Table 2).

Akkermans et al. (1975a) examined the effect of dieldrin and aldrin-transdiol on the sense organs of X. laevis. These two substances failed to induce any sign of repetitive activity (Table 2). Aldrin-transdiol caused an increase in the rate of firing of the lateral-line organ followed by a complete blockage of this firing (Ibid 1975a).

Webb et al. (1979) examined the short circuit current and resistance in isolated intact R. pipiens skin to determine if dieldrin was affecting the sodium balance. Dieldrin caused a significant increase in short circuit current while resistance was decreased. These results were interpreted as indicating that dieldrin was producing an increase in permeability of the outer skin membrane (Ibid 1979) (Table 2).

## CHLORDANE

### Long-Term Effects

Kaplan and Overpeck (1964) examined the survival of R. pipiens adults in response to varying concentrations of chlordane. At a concentration of 0.50 ppm, 40% of the frogs were dead after thirty days (Table 2). Some of the symptoms of poisoning were neuromuscular changes, excessive thrashing and tremors (Kaplan and Overpeck 1964).

Field studies to examine the potential threat of chlordane to amphibians were conducted by Mulla (1962, 1963). When applied at 0.56 kg/ha a.i. tadpoles in the pond reached maximum mortality within 48 hours (30%) (Mulla 1963) (Table 3). At a lower application (0.11 kg/ha a.i.) no mortality was observed (Ibid 1963).

### Residues

Up to 1,036 days after a lake in British Columbia was sprayed with technical grade chlordane at 0.010 mg/L, residue levels were measured in T. torosa (Albright et al. 1980). Residues were high within the first 14 days (8.9 ppm mean whole body without liver and stomach, 26.3 ppm liver, 2.2 ppm stomach) (Table 4). After 1,036 days residue levels declined to 0.2 ppm body without liver and stomach, 0.3 ppm liver, and only a trace in the stomach. Up to 93 days after treatment residue levels in T. torosa were much greater than in the water (Table 4). Of the chlordane constituents analyzed trans-nonachlor was most persistent. Although trans-nonachlor was present in the technical grade chlordane at 5.26% it accounted for 10-17% of the residues in newts after 14 days. Residue levels in T. torosa were compared with Salmo clarki and it appeared that the newts were more capable of metabolizing and eliminating chlordane residues (Ibid 1980).

### Mechanism for Toxicity

Several chlorinated hydrocarbon insecticides have been shown to inhibit ATPase enzymes including those activated by sodium and potassium (Webb et al. 1979). Webb et al. (1979) measured sodium (Na<sup>+</sup>) transport by measuring the short-circuit current across R. pipiens skin. At 2 X

$10^{-4}$  M  $\alpha$ -chlordane and  $\gamma$ -chlordane produced no significant change in the short-circuit current and thus it was concluded that sodium transport is not severely affected by chlorinated hydrocarbons based on suggested application rates (Webb et al. 1979) (Table 2).

## CHLORDIMEFORM

### Short-Term Effects

Chlordimeform was used extensively in Japan against rice stem borer larvae. Most of the research conducted on amphibians has been in physiological or biochemical studies to elucidate the mechanism of toxicity (Martin 1968, Wang et al. 1975).

Watanabe et al. (1975) conducted toxicity experiments by injecting adult R. nigromaculata with chlordimeform. At 300 mg/kg, 80% mortality was observed, and at 100 mg/kg no mortality was recorded (Table 2). Watanabe et al. (1975, 1976) also showed that chlordimeform at concentrations of  $10^{-4}$  to  $10^{-3}$  M causes the contraction of the rectus abdominus muscle (Table 2).

### Mechanism for Toxicity

The effect of chlordimeform on the muscle of the frog is the suppression of contraction induced by acetylcholine and potassium. This occurs due to a blockage of neuromuscular transmission by the depression of motor end-plate sensitivity to the transmitter (Wang et al. 1975, Watanabe et al. 1975, 1976) (Table 2).

## DDT

### Short-Term Effects

In static bioassays with B. woodhousii tadpoles and P. triseriata tadpoles, DDT was one of the least toxic insecticides tested, resulting in 24 hour LC50s of 2.4 mg/L and 1.4 mg/L respectively (Sanders 1970) (Table 1). Examination of DDT toxicity to B. woodhousii at different lifestages indicated an increasing sensitivity with age with the most sensitive stages being tadpoles at 3 to 7 weeks of age. The 24 hour LC50 for two-three week old tadpoles was 5.3 mg/L but by seven weeks this had dropped to 1.4 mg/L (Table 1). The increased sensitivity to DDT was thought to be related to developmental changes associated with metamorphosis. These changes include the disappearance of external gills as the internal lungs begin to function; an increase in thyroid activity leading to an increased rate of metabolism, the alteration in the permeability and ion transport of the integument and an increase in weight (Ibid 1970).

In contrast Marchal-Segault (1976) reported DDT to be slightly more toxic to B. bufo tadpoles with the 95% confidence limit of the 24 hour LC50 lying between 0.7 and 2.3 mg/L, although some mortality occurred at concentrations as low as 0.03 mg/L (Table 1). This worker confirmed the finding of Sanders (1970) that the sensitivity of tadpoles diminished after metamorphosis.

R. temporaria tadpoles exposed to 0.1 to 10.0 mg/L DDT exhibited a lack of coordination and hyperactivity. This behaviour returned to normal when the tadpoles were removed from the test medium, older tadpoles recovering more rapidly. At concentrations of 1.0 and 10.0 mg/L the tadpoles suffered mortality of 20 and 60-70 percent respectively (Table 2). In all tadpoles, growth was reduced and large tadpoles frequently surfaced to breathe, sometimes developing a hole in the snout between the nostrils and upper mandible (Cooke 1970, 1979). These abnormal behaviours after exposure to DDT were in part dependent on tadpole size and density. Small tadpoles at high density (50 tadpoles/L) exhibited more severe effects than larger tadpoles reared at a density of 10 tadpoles/L (Cooke 1979).

R. temporaria adults force-fed DDT dissolved in olive oil had a 96 hour LD50 of 7.6 mg/kg (Harri et al. 1979) (Table 1).

Ellis et al. (1944) reported one hundred percent mortality after injections of 150 mg/kg of DDT. Some mortality was observed following injections of as little as 10 mg/kg (Table 2).

Treated frogs displayed hyperexcitability, tremors, and a lack of locomotion and co-ordination. Frogs that survived for at least 10 days, lost all observable signs of poisoning and their behaviour returned to normal (Harri et al. 1979).

Issacson (1968) conducted experiments to examine peripheral nervous system involvement in the behaviour associated with DDT intoxication (Table 2) and found R. pipiens exhibited increased activity until the time of death. He theorized that an increase in activity of this type would place the frogs at a selective disadvantage. In studies exposing DDT intoxicated R. temporaria tadpoles to a predator Triturus cristatus, the newt made significantly more lunges at hyperactive tadpoles (Cooke 1971) (Table 2).

Mortality of amphibians following field applications of DDT varies and is dependent upon the amount of chemical, the weather at the time of application, the depth of pond, mode of application and the amount of canopy cover. These parameters render field results difficult to compare (Herald 1949, Cooke 1973b, Pearce and Price 1977). In field applications of DDT to ponds, 0.11 kg/ha caused no mortality of R. catesbeiana tadpoles (of varying sizes and stages) whereas 1.0 kg/ha

caused 80% mortality within 48 hours (Mulla 1963) (Table 3). In Minnesota, Fashingbauer (1957) reported a serious reduction in a local population of R. sylvatica following an aerial spray operation aimed at the reduction of forest tent caterpillars (Table 3). He noted no immediate adverse effects, but 60 hours after the application discovered large numbers of dead frogs. It was not known if the accumulation of DDT via the caterpillars or contact with the surface film caused the mortality but 94% of frogs had ingested caterpillars. Routine spraying of a forest in Georgia with 0.11 and 0.56 kg/ha DDT resulted in some tadpole and frog mortality (Tarzwell 1950) (Table 3).

### Long-Term Effects

R. temporaria tadpoles exposed to 0.001 mg/L DDT exhibited uncoordinated hyperactive behaviour during the period day 5 to day 8 post-exposure (Cooke 1970, 1971, 1972a). The onset of the behaviour coincides with DDT levels having reached 2 ppm in the tissues. These hyperactive tadpoles developed at a slower rate than the controls, presumably due to the elevated pesticide residues in their tissues (Cooke 1973a) (Table 4).

When R. temporaria tadpoles were exposed to 0.0001 mg/L they behaved normally even though, from day 15 onward, the tadpoles and small frogs contained between 2 and 5 ppm DDT (Table 4). This is in contrast to tadpoles receiving an acute dose of DDT, which resulted in hyperactivity when tissue levels reached 2 ppm (Cooke 1973a). An acute dose can result in high residue levels in the blood and thus a build up in the nervous system, causing hyperactive behaviour. The slower rate of intake induced by chronic exposure, results in lower residue levels in the blood and does not cause an overload (Cooke 1973a). Differences in response to acute and chronic dosing were considered to be related to the storage of residues in the fatty tissues (eg. tail) (Cooke 1973a).

Long term effects of DDT in the field were examined by Pearce and Price (1977) in New Brunswick forests (Table 3). Substantial residues were discovered in four species of amphibians while other species had lower levels. No long-term effects on the populations of these amphibians were reported.

The effect of DDT on tail regeneration of R. pipiens and R. catesbeiana was examined by Weiss (1975). The two species exhibited both different rates of regeneration and vulnerability to DDT in terms of direct mortality. R. pipiens experienced slower and retarded tail regeneration at 0.005 ppm with 30% mortality after 12 days, while regeneration in R. catesbeiana at this concentration, was not different from controls (Table 2). At 0.025 ppm, R. pipiens suffered 95% mortality and retarded tail regeneration after 12 days. R. catesbeiana



experienced 60% mortality and a reduced regenerative capability at this concentration. Weiss (1975) suggested that a retardation of regeneration may increase the susceptibility of the organism to predation, causing an increase in the concentration of a contaminant at higher levels in the food chain.

## Residues

A large portion of the research into residues in amphibians has centered around DDT. The level of residues accumulated in amphibians is dependent upon a number of factors. One factor is the stage of development during exposure. Cooke (1972a) was unable to detect DDT residues in R. temporaria, B. bufo, and Triturus vulgaris spawn after 24 to 48 hour exposures to DDT (Cooke 1972a). R. temporaria and B. bufo tadpoles exposed to DDT prior to or during hind limb bud development, had tissue concentrations of 2-3 and 3-4 mg/kg respectively. During tail resorption the frog was susceptible to levels that had been accumulated and stored during exposure at the larval stage. In general R. temporaria was more sensitive to DDT than B. bufo, indicating species differences in the ability to accumulate residues (Cooke 1972a) (Table 2).

The absolute amount of DDT taken in by R. temporaria tadpoles exposed to 0.1, 1.0 or 10.0 mg/L DDT was independent of size. Smaller tadpoles had higher tissue content of DDT, and higher mortality than larger individuals (Cooke 1970).

Licht (1976a) studied the effect of length of exposure and temperature on accumulation. Subacute exposure of R. sylvatica tadpoles for 7 days to 0.001 to 0.002 mg/L DDT reached maximum tissue levels in 24 hours (Licht 1976a) (Tables 2 and 4). After 24 hours exposure to 0.003 mg/L DDT at 15°C the liver had the highest level with 27.04 mg/kg (Table 4) while whole body residues were 0.7 mg/kg. These levels declined to less than 18 mg/kg in the liver and 0.6 mg/kg whole body after 80 hours. When treated with 0.003 mg/L DDT for 7 days R. sylvatica maintained higher DDT levels for longer at 15°C than tadpoles at 21°C (Licht 1976a) (Table 4).

Feeding also influences DDT accumulation. Fed R. temporaria adults had lower DDT residues than starved frogs in tissues other than fat (Harri et al. 1979). Loss of DDT residues from body tissues can vary depending upon previous exposure to DDT and the physical environment (Harri et al. 1979, Licht 1979). Previous exposure to DDT can increase the rate of loss of DDT after re-exposure in R. temporaria adults (Harri et al. 1979). It was speculated that previous DDT exposure leads to increased activity of drug-metabolizing enzymes that increases DDT excretion (Ibid 1979). R. sylvatica adults treated as tadpoles for 1 week with 0.003 mg/L DDT lost DDT much more slowly when maintained in

an aquatic environment for 3 to 4 weeks than adults maintained in a dry terrestrial environment (Licht 1976a, b, Licht 1979). DDT in the fat of the terrestrial adults was 37.4 mg/kg compared to 4.93 mg/kg in the aquatic tadpoles (Ibid 1976b). Adult R. temporaria, however, were found to eliminate DDT at the same rate in both aquatic and terrestrial environments (Harri et al. 1979).

Niethammer et al. (1984) found frogs from a contaminated area in Louisiana had low levels of DDT and suggested that this was due to efficient excretion and their mobility, which allows them to flee unsuitable areas (Table 4).

Finley and Pillmore (1963) found traces of DDD in R. pipiens in an area where only DDT had been sprayed. This suggests that DDT is converted to DDD by animal tissues and thus possibly establishes another route of excretion.

Residues vary greatly in different parts of the body (Harri et al. 1979, Licht 1976a, b). Fat had the highest residue concentration in R. sylvatica tadpoles exposed to 0.001 to 0.003 mg/L DDT for 7 days with up to 60 mg/kg, the liver had up to 20 mg/kg while the heart, head, gut and lungs contained only a trace (Licht 1976b). R. temporaria also had maximum DDT residues in fat, tissues with high concentrations of fat and the liver (Harri et al. 1979).

DDT residues have been correlated with mortality and behavioural and developmental abnormalities (Cooke 1970, 1972b, 1973a, Harri et al. 1979). The LD50 for adult R. temporaria was 7.6 mg/kg (Harri et al. 1979) (Table 1). Chronic dosing of R. temporaria tadpoles in the field and lab both showed that residues up to 5 mg/kg DDT resulted in mild hyperactivity (Cooke 1973a) (Table 2). Acute doses of 2 to 5 mg/kg caused severe hyperactivity, weight loss and delay in development (Cooke 1970, 1972b, 1973a).

Size may also influence the severity of effects from residues. Cooke (1979) found R. temporaria tadpoles of the same age, but larger weight ( $x = 689 \pm 44$  mg) had no behavioural abnormalities at 2.5 mg/kg DDT while much smaller tadpoles ( $x = 363 \pm 11$  mg) exhibited hyperactivity with only 2.2 mg/kg DDT (Table 2).

DDT residues can be retained for a long period of time after applications are stopped (Dimond et al. 1968). Sampling of P. cinereus in sprayed forests in Maine showed that it took 8 to 9 years before DDT residues in adult salamander returned to pre-spray levels (Ibid 1968) (Table 4).

## Mechanism for Toxicity

DDT enters the organism at different life stages via several routes; at the early spawn stage (Cooke 1972a) (Table 2), via prey species and water intake both orally and via absorption through the skin (Kaiser and Dunham 1972-73) (Table 2). Having entered the body, the mechanism for toxicity of DDT, although mainly due to a physiological response, can in part be due to behavioural aberrations. The morphological abnormalities observed in R. temporaria tadpoles after exposure to DDT were due to a disruption of glandular development in the external skin of the snout (Cooke 1970, Osborn et al. 1981). The disruption of glandular development coupled with the hyperactive behaviour, causes the lower mandible to strike the inner surface of the upper mandible. This damage, in conjunction with the disrupted external skin gland, can cause the loss of the upper mandible in some individuals. These deformities were sometimes permanent, resulting in a blunt snout and brain deformities (Ibid 1981) (Table 2).

DDT has been shown to significantly decrease the shortening time, twitch duration, contraction and relaxation time of the skeletal muscles of Rana hexadactyla (Rajendra et al. 1980). DDT may also increase nerve acetylcholine by enhancing liberation from bound reserves which play an important role in the dynamics of contractile tissues (Rajendra et al. 1980, Craciun et al. 1981) (Table 2).

Akkermans et al. (1975a) studied another physiological response to DDT. They reported DDT caused repetitive activity in the cutaneous touch receptors of X. laevis (Table 2) which may affect the toad's reactions and behaviour to external stimuli.

## ENDOSULFAN

### Short-Term Effects

In static bioassays with endosulfan, R. tigrina tadpoles had LC50 an order of magnitude lower than juvenile catfish (Clarias batrachus) and damselfly nymphs (Enallagma spp.) (Gopal et al. 1981) (Table 1). A reduced swimming capacity, lack of physical stamina and a thinning and transparency of the gills were the observed symptoms of endosulfan poisoning (Ibid 1981).

### Long-Term Effects

In an aerial application of 0.014 kg/ha a.i. endosulfan on a savannah woodland in Zimbabwe-Rhodesia, no mortality was recorded for adult Chiromantis xampelina adults maintained in cages for 19 days after treatment (Cockbill 1979) (Table 3), although mortality was recorded to some shallow water fish (Tilapia spp.). There are no data for residue accumulations of endosulfan in amphibians.

## ENDRIN

### Short-Term Effects

Endrin toxicity to amphibians varies depending upon the species and the lifestage. R. sphenoccephala eggs in continuous flow bioassays were sensitivity of R. temporaria tadpoles to endrin in static bioassays gradually increased during development with two-day old tadpoles having a 72 hour LC50 of 0.425 mg/L while for 107-day old tadpoles (with fore and hind limbs developed) the 72 hour LC50 was 0.015 mg/L (Wohlgemuth 1977) (Table 1). The increase in sensitivity to endrin during development was thought to be due to changes in respiration and metabolism (Ibid 1977).

Wohlgemuth (1977) conducted a comparison of the toxicity of endrin to seven species of amphibians. The results are presented on Table 1. All species were in the tadpole stage except Ambystoma opacum. R. sylvatica and A. opacum were most resistant to endrin but the high LC50 for A. opacum was thought to be due to the older age of these individuals. R. catesbeiana was the most sensitive to endrin toxicity. Behavioural abnormalities during the exposure were all associated with mortality (Ibid 1977).

In static bioassays with 4-5 week old B. woodhousii tadpoles, endrin had a 96 hour LC50 of 0.12 mg/L and was the second most toxic pesticide tested (Sanders 1970) (Table 1). One week old P. triseriata tadpoles had a similar 96 hour LC50 of 0.18 mg/L endrin (Table 1). This 96 hour LC50 cannot be compared directly with those for the species used by Hall and Swineford (1980, 1981) because the former used continuous flow dosing, while the latter used static dosing (Table 1).

Field applications of endrin have also been shown to be highly toxic to amphibians. Applications of 0.11 and 0.56 kg/ha endrin caused 100% mortality to R. catesbeiana, B. boreas and S. hammondi tadpoles within 5 days of treatment (Mulla 1962, 1963) (Table 3). Endrin was one of the most toxic of the organochlorine insecticides tested (Ibid 1963).

In an experiment designed to study the effects of endrin on an artificially evacuated pool with no inlet, 0.5L of 20% endrin was poured in at one end. Immediately a white turbid cloud was formed which moved off due to the wind. Concentrations of endrin were found to be 0.047 mg/L 10 metres from the point of entry (Wohlgemuth and Trnkova 1979) (Table 3). Aquatic invertebrates died within five minutes of contact with the endrin. The indigenous population of R. esculenta adults began to emit high shrieks and experience severe cramping and paralysis. This was followed by death. Caged R. temporaria tadpoles died within thirteen minutes of contact with the endrin. The concentration of endrin over the volume of the pool was calculated to be 2.5 mg/L (Ibid 1979) (Table 3).

## Long-Term Effects

Kaplan and Overpeck (1964) placed R. pipiens adults in varying concentrations of endrin and recorded their survival after 30 days (Table 2). Frogs exposed to 0.03 ppm endrin in solution suffered 30% mortality after 30 days. Endrin caused neuromuscular changes to occur including excessive thrashing and tremors (Ibid 1964).

Webb et al. (1979) examined the effect of endrin of the short-circuit current and active sodium transport across the skin of R. pipiens adults (Table 2). They found endrin did not produce a significant effect on the short-circuit current or resistance of the frog skin at a concentration of  $2 \times 10^{-4}$  M.

## Residues

R. sphenoccephala accumulated endrin residues up to 2.8 mg/kg which was 94 times water levels (Hall and Swineford 1980). Frogs which survived endrin exposures however never accumulated more than 0.06 mg/kg which was a concentration factor of only six (Ibid 1980).

Accumulation of endrin residues in amphibian foods can cause mortality when ingested. An endrin resistant population of mosquito fish (Gambusia affinis) exposed to 2 mg/L endrin for 7 days and then force fed to R. catesbeiana adults resulted in 100% frog mortality within 24 hours (Rosato and Ferguson 1968) (Table 2). A pooled sample (N=4) of field collected mosquito fish contained 9.65 mg/kg DDT and a pooled sample (N=4) of endrin treated mosquito fish exhibited residues of 890 mg/kg of endrin.

Niethammer et al. (1984) analyzed three species of frog from alluvial cropland in Louisiana that had been subjected to heavy pesticide use (Table 2). The low levels reported for amphibians were considered to be due to an efficient ability to excrete these contaminants.

## LINDANE

### Short-Term Effects

In static bioassays lindane was the least toxic of 14 insecticides to 1 week old P. triseriata and 4 week old B. woodhousii tadpoles (Sanders 1970) (Table 1). Mortality was preceded by irritability and loss of equilibrium. These effects were noted at concentrations lower than the 96 hour LC50 value and were irreversible (Ibid 1970).

### Long-Term Effects

In chronic exposures to lindane, the egg stage of X. laevis had lower hatching success at all the lindane concentrations tested (Marchal-Segault and Ramade 1981) (Table 2). When the tadpole stage was exposed to the same lindane concentrations, mortality increased with time and after 6 weeks' exposure was 100% (Table 2). Tadpoles exposed to lindane at lower weights, showed morphological deformities in comparison with the controls. The morphological deformities included lateral deviation of the body axis and a dorsal lump on the cranial area. A darkening of pigmentation also occurred in treated tadpoles and persisted in the surviving adults (Ibid 1981).

Mulla (1962, 1963) found lindane caused the lowest mortality of R. catesbeiana tadpoles in comparison with ten other organochlorine insecticides. Lindane caused only 10% mortality after 7 days at an application rate of 0.56 kg/ha (Table 3).

### Residues

Whitacre and Ware (1967) examined the chronic effects of vaporized lindane on R. pipiens adults. Lindane residues in the brain, liver and fat were measured during 38 day exposures. Residue accumulation increased with increased exposure to lindane (Table 4). Residues contained in fat actually declined over the exposure period and were not detectable after day 38 (Table 4). No frog mortality occurred during the exposure but residues were much greater than in carp, guppies, mice or rats. The high residue levels in the frogs were thought to have been caused by the frogs not eating during exposure. This could have lowered the rate of metabolism and excretion resulting in accumulation of high levels (Ibid 1967).

Lindane has also been shown to readily pass through the skin of R. pipiens (Kaiser and Dunham 1972) (Table 2). The high water solubility of lindane and simple diffusion through the skin were speculated to be the cause of lindane being readily accumulated (Ibid 1972).

### Mechanism for Toxicity

Subacute chronic exposures to lindane during growth and development have effects analogous to those reported for eggs and tadpoles intoxicated with fungicides (Bancroft and Prahlad 1973), herbicides (Cooke 1977), and fenthion (an organophosphorous insecticide) and so may be caused by similar mechanisms (Marchal-Segault and Ramade 1981). Marchal-Segault and Ramade (1981) suggested that the delays in growth and development they observed due to chronic exposures of lindane, indicate some endocrine dysfunction of the hypothalamo-hypophyseal axis. This dysfunction did not appear to be complete, since some surviving animals attained the final stage of metamorphosis. An increase in the pigmentation, which was also noted during the tadpole stage, indicated a dysfunction of the intermediate lobe of the hypophysis that controls pigmentation (Ibid 1981).

In adult frogs lindane has been shown to cause damage to skeletal muscle (Publicover et al. 1979). The damage was caused by a rise in calcium concentration resulting in the dissolution of muscle myofilaments. In vivo, the increased calcium also caused a marked increase in the miniature endplate potential frequency at the neuromuscular junction with the threshold for an observable response at 0.0003 M (Ibid 1979) (Table 2). Publicover and Duncan (1979) speculated that if lindane has similar effects on other synapses of the central nervous system then hyperexcitability, tremors and convulsions could be produced. These effects, however, have not been reported for amphibians exposed to lindane.

The increase in calcium concentration appears to be caused by lindane decreasing the permeability of the outer skin membrane to all ions except sodium. Sodium permeability is actually increased, promoting calcium release from storage sites such as the mitochondria (Publicover et al. 1979, Publicover and Duncan 1979, Webb et al. 1979).

## METHOXYCHLOR

### Short-Term Effects

Methoxychlor was moderately toxic to P. triseriata tadpoles in static bioassays with a 48 hour LC50 of 0.42 mg/L while B. woodhousii tadpoles had a 48 hour LC50 of 0.11 mg/L (Sanders 1970) (Table 2).

### Residues

Methoxychlor was much more readily accumulated in B. americanus adults after exposure in water than through ingestion of mealworms containing methoxychlor residues (Hall and Swineford 1979). Feeding tests in which adult toads were fed mealworms containing 0.024 mg/kg of methoxychlor for 6 days revealed an accumulation of 0.008 mg/kg in the whole body of the toads. Conversely, toads that were exposed to water containing 0.069±0.036 mg/L methoxychlor for 6 days accumulated 0.244 mg/kg in the body (Table 4). Residue levels were not correlated with duration of exposure suggesting equilibrium is reached rapidly. There were no changes in organ weights, feeding, behaviour or survival due to the exposure to methoxychlor (Hall and Swineford 1979).

## MIREX

### Residues

Mirex is a chlorinated hydrocarbon insecticide used specifically for control of the fire ants (Salenopsis saevissima) in the southeastern United States. In separate studies in Florida (Wheeler et al. 1977), Georgia (Wojcik et al. 1975), Mississippi (Naqvi and de la Cruz 1973) and Louisiana (Niethammer et al. 1984) reported the levels

of mirex in amphibians. Wojcik et al. (1975) reported that the highest levels occurred one to three months after treatment and Wheeler et al. (1977) found that amphibians still showed low levels a year after a single application of mirex. Naqvi and de la Cruz (1973) found residues in some organisms in an area not treated with mirex, suggesting widespread movement in the environment and evidence of some biological magnification. Niethammer et al. (1984) considered that the low residue levels in frogs might be due to their high turn-over of fatty acids (Brown 1964) which could increase their ability to mobilize and excrete chlorinated hydrocarbon compounds.

After an application of mirex to a fish pond in Mississippi the residues found in A. gryllus reached a maximum after 6 months and declined to 0.05 ppm within 16 months of treatment. The initial increase in residues was partly attributed to an accumulation through the food chain (Collins et al. 1973) (Table 4).

## TOXAPHENE

### Short-Term Effects

Differences in toxicity of toxaphene to amphibians have been shown to vary with life stage and species. In continuous flow bioassays, R. sphenoccephala eggs were resistant to toxaphene in comparison with the tadpole stage, which in turn was more sensitive than the two sub-adults tested (Hall and Swineford 1980) (Table 1). Behavioural aberrations (mainly hyperirritability and prolonged stupor) and growth retardation were observed for toxaphene treated tadpoles with effects on growth in 96 hour exposures, occurring at 0.013-0.018 mg/L (Ibid 1980).

Species differences in sensitivity to toxaphene were observed in continuous flow bioassays for seven species, although the species were at varying lifestages (Hall and Swineford 1981). The species and 96 hour LC50s are presented on Table 1. A. opacum and R. sylvatica were most tolerant while A. crepitans and A. maculatum were most sensitive to toxaphene. The high tolerance of A. opacum was thought to be partly due to age as they were field collected as adults as all other species were in the larval stages (Ibid 1981).

In static bioassays Sanders (1970) measured the 96 hour LC50 of toxaphene for B. woodhousii and P. triseriata (Table 1). The variation in sensitivity may have been a result of species differences and/or differences in age (Sanders 1970).



The seven species exhibited a range in severity of behavioural abnormalities which were caused by exposure to different concentrations of toxaphene (studied by Hall and Swineford 1981). R. catesbeiana exhibited only subtle behavioural abnormalities with mild loss of equilibrium and disorientation where A. opacum displayed debilitating contortions and immobilization. Some species exhibited behavioural abnormalities at concentrations much lower than their LC50 values. The independence of the behavioural abnormalities and mortality led Hall and Swineford (1981) to conclude that the mechanisms for these two effects were acting separately.

In field trials, using R. catesbeiana, Mulla (1962, 1963) found toxaphene to be among the most toxic substances that were tested. At an application rate of 0.5 kg/ha the tadpoles suffered 100% mortality within 24 hours (Table 3). No mortality was observed when toxaphene was applied at 0.1 kg/ha (Mulla 1963) (Table 3).

### Long-Term Effects

Kaplan and Overpeck (1964) examined the survival of R. pipiens at different concentrations of toxaphene (Table 2). After 30 days the frogs exposed to the highest level (0.60 ppm) suffered 25% mortality. Neuromuscular changes were observed in treated frogs in conjunction with thrashing and tremors (Kaplan and Overpeck 1964).

### Residues

Organochlorine residues were measured in the fauna of oxbow lakes in Louisiana that have acted as sumps in accumulating residues from nearby agricultural land (Niethammer et al. 1984) (Table 4). Toxaphene was found to be one of the principal residues detected. The levels in R. catesbeiana, R. sphenoccephala and the Bronze frog were low (Table 4) when compared to their trophic level. These workers related this to the high turnover of fatty acids in these species (Brown 1964).

Hall and Swineford (1980) reported that frogs (R. sphenoccephala) were able to concentrate toxaphene residues by a factor of one hundred. These results are based on animals under constant exposure to the toxicant and for comparison, it was also noted that some fish concentrate residues by factors of one hundred thousand under these conditions (Hall and Swineford 1980).

### OTHERS

This section deals with several organochlorine insecticides for which there is little information available concerning their toxicity to amphibians.

Kepone EC2 is utilized primarily for agricultural pest control. When applied to ponds at 0.11 and 0.56 kg/ha it produced no mortality among R. catesbeiana tadpoles (Mulla 1963) (Table 3).

Thiodan I is also an agricultural pest control insecticide. An application of 0.56 kg/ha to a pond resulted in 100 percent mortality of tadpoles within 24 hours (Mulla 1962, 1963) (Table 3). When Thiodan II was applied at the identical rate the result was the same to tadpoles of R. catesbeiana (Mulla 1963).

Trichlorphon injected into R. temporaria adults at a dose of 50-300 mg/kg caused a reduced red blood cell count and shifts in the composition of white blood cells (Szubartowska 1979) (Table 2).

Benzene hexachloride, an insecticide closely related to lindane, was reported as being moderately toxic to Bufo woodhousii tadpoles (Sanders 1970) (Table 1). Kaplan and Overpeck (1964) observed the survival of R. pipiens adults in varying concentrations of BHC and reported that they suffered 40% mortality after exposure to the highest concentration (Table 2).

Residues of BHC were not detected in any amphibian sample from contaminated oxbow lakes in Louisiana (Niethammer et al. 1984) (Table 4).

Heptachlor, another insecticide, was also found to be moderately toxic to B. woodhousii tadpoles (Sanders 1970) (Table 1). At an application rate of .56 kg/ha heptachlor caused 80% mortality two days after treatment (Mulla 1962, 1963) (Table 3). No further mortality was observed 5 days after treatment. Albright et al. (1980) analyzed Tarichia torosa in British Columbia and found heptachlor levels in the newts 4 days after treatment with chlordane. Subsequent samples taken from the same location had non-detectable amounts of heptachlor epoxide (Albright et al. 1980) (Table 4).

## b) CARBAMATES

### Short-Term Effects

In static bioassays using Rana clamitans tadpoles, aminocarb had a 24 hour LC50 of 247 mg/L for small tadpoles and 234 mg/L for larger tadpoles. The 96 hour LC50 for small tadpoles was 118 mg/L (Lyons et al. 1976) (Table 2). Treated tadpoles had extensive hemorrhagic regions and exhibited jaw-twitching, ecdysis, difficulty in swimming, buoyancy problems, colour change, mouth deformities, edema and bent tails. The mouth deformities were thought to have occurred through direct contact of the snout with aminocarb that had adhered to the sides of the treatment container (Ibid 1976).

In other toxicity tests involving carbamates, Marking and Chandler (1981) reported a 96 hour LC50 of 8.7 mg/L for methiocarb, employing R. sphenoccephala larvae (Table 1). Marchal-Segault (1976), exposing B. bufo tadpoles to various concentrations of sevin, found a 72 hour LC50 of between 16.8 and 20.6 mg/L (Table 1). Tucker and Crabtree (1969) estimated an LD50 of 283-800 mg/kg in a single oral dose of zectran to adult R. catesbeiana (Table 1). They considered that zectran would not be toxic to adult amphibians at the current field application rates.

Marian et al. (1983) obtained a 96 hour LC50 value of 6.2 mg/L of carbaryl for tadpoles of R. tigrina. Rzehak et al. (1977) conducted static bioassays using different concentrations of carbaryl and both R. temporaria and X. laevis tadpoles. Exposure to a concentration of 0.1% caused an increase in mobility of X. laevis tadpoles. When placed in tap water after exposures of 30, 60 and 120 minutes the tadpoles gradually returned to normal. Histological examination of the group exposed for two hours revealed contraction of miomers; partial separation of muscle fibers and oedema of the intestinal epithelium (Rzehak et al. 1977). Tadpoles of R. temporaria when exposed to a concentration of 0.1% and then returned to tap water after 15 minutes, reacted in the same manner as X. laevis. None of these exposures seem to affect the development of the organism. Marian et al. (1983) found R. tigrina tadpoles to be moderately resistant to carbaryl in 96 hour toxicity tests (Table 1). However, prolonged exposures using 0.01% and 0.001% carbaryl produced different results (Table 2).

After an exposure of 24 hours to 0.01% carbaryl tadpoles of R. temporaria were smaller than the corresponding controls and exhibited bent tails, causing the tadpoles to swim in tight circles (Rzehak et al. 1977) (Table 2). When transferred into tap water, 90% of the tadpoles survived, though some with severe abnormalities that restricted their movements died later. Under the same conditions, tadpoles of X. laevis exhibited a reduced heartbeat, bent tails, partial separation and contraction of muscle fibres and appeared moribund. Eighty percent of these tadpoles died within a week after exposure. Exposures of 10 days for R. temporaria and 8 days for X. laevis to 0.001% carbaryl led to permanent tail malformation in both species. R. temporaria also exhibited growth inhibition, with 90% of the tadpoles dying between day 7 and day 10 of the treatment and the remainder dying within 2 days following transferral to tap water. After 8 days of exposure to 0.001% carbaryl, further growth and development of X. laevis tadpoles was inhibited. Upon transferral to tap water many individuals died and mortality for the first week after treatment was 20% (Ibid 1977) (Table 2).

In general both species exhibited behavioural changes immediately after exposure to carbaryl. Their activity increased and then, after a period of time, their movement slowed until they lay motionless on the bottom of the test vessel. The length of time for this behaviour varied with the concentration of carbaryl (Rzehak et al. 1977).

Application of carbofuran to rice fields in Texas had severe effects on A. crepitans blanchardi adults (Flickinger et al. 1980). Frogs were found paralyzed or exhibiting abnormal behaviour as early as 15 minutes after application. Four out of 19 frogs observed one half hour after treatment were paralyzed. Only one frog was found dead (Ibid 1980) (Table 3).

R. temporaria tadpoles maintained in cages and suspended in seasonal ponds in ditches near farm fields sprayed with oxamyl exhibited a significantly higher incidence of vertical curvature deformities and tail tip defects than tadpoles maintained in ditches near fields not sprayed with oxamyl (Cooke 1981). In the lab after exposure for 1 hour to 100 mg/L oxamyl, 90% of the tadpoles exhibited vertical curvature deformities similar to those observed in the field. The deformities appeared not to be spinal, but rather of muscular origin (Ibid 1981) (Tables 2 and 3).

### Long Term Effects

Field applications of zectran in New Brunswick over several years produced no adverse effects on the population of R. clamitans and posed no serious hazard to either larval or adult frogs (Pearce and Rick 1969, Pearce and Price 1977, Rick and Price 1974) (Tables 2 and 3). Matacil (aminocarb) produced no adverse effects on individual amphibians or their populations. Bracher and Bider (1982) found no significant change in populations of R. sylvatica and R. pipiens following aerial application of matacil in Quebec (Table 3). Similarly, following spraying in New Brunswick Rick and Price (1974) observed no mortality of frogs, toads or tadpoles and Pearce and Price (1977) concluded adult and larval amphibians were unaffected by matacil (Table 3).

In experiments to determine the effects of carbaryl on growth and metamorphosis of R. tigrina, Marian et al. (1983) raised tadpoles from hatching to metamorphosis in sublethal concentrations. It was found that carbaryl does not affect the larval or metamorphic period. During metamorphosis mortality was significant at the 2 mg/L level (Table 2). It was concluded that any level below 5 ppm may be considered as sublethal (Marian et al. 1983).

## Residues

Bullfrog tadpoles were analyzed for residues of carbofuran following an application of 0.025 ppm in conjunction with an application of atrazine at 0.3 ppm. Residues of carbofuran were not detectable (less than 0.4 ppb) 77 days post-treatment (Klaassen and Kadoum 1979).

## Mechanism for Toxicity

Carbamates are systemic insecticides that act by inhibiting cholinesterase activity (Corbett 1974, Przędziecki 1976). The cholinesterase inhibition caused by carbaryl led to contraction and partial separation of muscle fibres and the eventual immobilization of tadpoles (Rzehak et al. 1977). Carbaryl also had an effect on frog skin where it increased the sodium ( $\text{Na}^+$ ) uptake suggesting that the outer border of transporting cells is the area of action of externally applied insecticides (Webb et al. 1979) (Table 2). Takeno et al. (1977) found the response of R. pipiens nerve muscle to indirect stimulation was suppressed more effectively than the response to direct stimulation in the presence of  $2 \times 10^{-4}$  M carbofuran. Various concentrations of carbofuran also suppressed muscle contractions in frog nerve-muscle (Table 2).

Prolonged exposures to carbaryl leading to long-term contraction of trunk and tail muscles may lead to secondary changes in other organs such as the tail bones (Rzehak et al. 1977). Carbaryl derivatives can be detoxified, which explains why tadpoles recovered from short exposures in the experiment conducted by Rzehak et al. (1977) (Table 2).

## c) ORGANOPHOSPHATES

### FENITROTHION

#### Short-Term Effects

Lyons et al. (1976) using R. clamitans tadpoles, established a 96 hour LC50 of 4.9 ppm (Table 1). Fenitrothion was more toxic to amphibians than two other compounds tested simultaneously (Matacil and Orthene). Symptoms of fenitrothion poisoning were characterized by swimming difficulties, bent tails, jaw twitching, ecdysis and gulping, apparently in an effort to move more water over the gills (Lyons et al. 1976). The authors speculated that altered physical behaviour stimulated by reduced cholinesteratic activity induced by organophosphates would place the tadpoles at a selective disadvantage.

#### Long-Term Effects

Folithion (fenitrothion) drastically altered the length of the larval period of R. tigrina and reduced the number that metamorphosed (Mohanty-Hejmadi and Dutta 1981) (See Folithion, Table 2). Concentra-

tions of 4 ppm reduced the number of larvae that metamorphosed and extended the larval period by approximately 40 days compared to the control group (Table 2).

In field studies on the effect of fenitrothion on amphibian populations two distinct results have been reported. In two separate studies conducted in New Brunswick no mortality or unusual behaviour was observed in frogs, toads and tadpoles (Pearce and Price 1977, Rick and Price 1974) (Table 3). However, Thirumurthi et al. (1973) observed 100% mortality of frogs in Indian rice fields (Table 3). The application rate on the rice fields was 0.5 kg/ha and in New Brunswick it was 0.14 kg/ha applied twice, in one study, and 0.057 kg/ha, applied twice, in the second study.

## FENTHION

### Short-Term Effects

Using 26 to 37 day old B. bufo tadpoles, Marchal-Segault (1976) reported a 48 hour LC50 of 2.0-2.2 ppm for fenthion (Table 1). It was also reported that development of the larvae was retarded by exposure to fenthion. (Ibid 1976). Hall and Kolbe (1980) calculated a 96 hour LD50 of 4.9 ppm for R. catesbeiana tadpoles (Table 1).

### Long-Term Effects

A single aerial application of fenthion at 0.5 kg/ha on Indian rice fields produced 100% mortality of frogs in the field (Thirumurthi et al. 1973) (Table 3).

Johnson and Prine (1976) reported that fenthion, when used at one-half of the usual field concentrations, produced a pronounced reduction in the temperature tolerance range of B. boreas (Table 2). Sublethal effects of this type may render the organism more susceptible to environmental temperature shifts causing death within a much narrower range than non contaminated healthy toads. Fenthion produced the largest reduction of the six insecticides tested (Johnson and Prine 1976).

### Residues

In two experiments to test whether Rana catesbeiana tadpoles could accumulate fenthion to levels that would be lethal to Mallard (Anas platyrhynchos) ducklings, Hall and Kolbe (1980) found that 8 of 10 ducklings died after being fed exposed tadpoles (Table 2). In the first experiment, tadpoles placed in water containing 2.2 ppm of fenthion and then fed to ducklings killed 4 of 6 ducklings. Mortality was 100% within 5 hours in ducklings fed tadpoles dosed at 5 ppm fenthion (Table 2). Significant reductions in cholinesteratic activity were reported for the ducklings (Hall and Kolbe 1980).

## GUTHION

### Short-Term Effects

In static bioassays using B. woodhousii tadpoles Sanders (1970) reported a 96 hour LC50 of 0.13 ppm for guthion (Table 1), the organisms went through a period of irritability followed by a loss of equilibrium and finally death.

### Long-Term Effects

Mulla (1962) reported that guthion at an application rate of 1.8 kg/ha, resulted in 100% mortality of R. catesbeiana. Mulla et al. (1963) also reported that at rates of .11 and .45 kg/ha a.i. no mortality was observed within cages containing B. boreas and S. hammondi (Table 3).

## MALATHION

### Short-Term Effects

In static toxicity tests Sanders (1970) found 96 hour LC50's of 0.42 and 0.20 ppm to B. woodhousii and P. triseriata respectively. When exposed to a concentration of approximately 5 ppm malathion for 96 hours, forty percent of R. catesbeiana tadpoles died (Hall and Kolbe 1980) (Table 1).

Malathion also caused abnormal development in Microhyla ornata at the yolk plug stage when exposed to concentrations of between 1 and 20 ppm (Pawar et al. 1983) (Table 2). Observed abnormalities included head, trunk and tail defects, behavioural aberrations, loss of balance, poor pigmentation and retarded growth. Mortality increased with increasing concentrations of malathion (Ibid 1983).

### Long-Term Effects

Mohanty-Hejmadi and Dutta (1981) also reported that malathion affects growth of amphibians. In tests using R. tigrina it was found that malathion caused a prolongation of life history and smaller animals after metamorphosis (Table 2). The concentrations used in this experiment, though introduced via chronic exposure, were much lower than levels recommended for field applications of malathion.

The effects of malathion at different concentrations ranged from a decrease in white cell counts, and depression of red blood cells to physical lethargy causing reduced avoidance response (Kaplan and Glaczenski 1965) (Table 2).

Aerial applications using Ambithion (malathion and fenitrothion) effectively eliminated frog populations in sprayed areas (Thirumurthi et al. 1973) (See Ambithion, Table 3).

## Residues

Hall and Kolbe (1980) studied the accumulation of several organophosphate insecticides by amphibians. R. catesbeiana tadpoles were dosed with malathion at 5 mg/L and then force fed to Mallard ducklings. No mortality among the ducklings was observed and it was concluded that symptoms of malathion intoxication would only be observed if the insecticide was massively accumulated (Ibid 1980) (Table 2).

## PARATHION

### Short-Term Effects

In static toxicity bioassays using P. triseriata the 96 hour LC50 for parathion was 1.0 ppm (Sanders 1970) (Table 1). Hall and Kolbe (1980) calculated the LD50 for R. catesbeiana tadpoles to be 1.32 ppm and the LD50 reported by Ederly and Schatzberg-Porath (1960) following injection into the dorsal lymphatic sac, was 967 mg/kg (Table 1). In the latter study B. vulgaris was found to be more resistant to parathion than either the frog or mouse used as comparative species.

Guzman and Guardia (1978) conducted experiments on the short-term and long-term effects of exposure to parathion. B. arenarum were exposed to parathion in two groups. Group 1 received constant exposure and Group 2 was exposed for 2 hours. Both groups exhibited an initial loss of cholinesteratic activity and subsequently recovered when removed from the contaminated medium (Ibid 1978) (Table 2).

### Long-Term Effects

Kaplan and Glaczenski (1965) noted the same results in hematological tests with parathion as those exhibited by frogs exposed to malathion, though to a slightly more severe degree (Table 2).

Field studies examining the effect of parathion on populations of amphibians revealed that aerial applications of 0.11 to 1.12 kg/ha did not adversely affect amphibian populations in several different habitats (Mulla 1962, Mulla et al. 1963, 1966) (Table 3).

## Residues

Amphibians are generally resistant to cholinesterase inhibitors (Potter and O'Brien 1964). This coupled with the fact that organophosphates have a low persistence in the environment may indicate



that adverse effects from utilizing these insecticides may be a result of the ability of certain organisms to accumulate the contaminants and move large concentrations up through the food chain (Hall and Kolbe 1980). Several studies have been undertaken to test this idea using amphibians and organisms located at higher trophic levels.

Hall and Kolbe (1980) fed Mallard ducklings tadpoles that had been exposed to 1 ppm parathion. Accumulation in the tadpoles was estimated to be 64 times the level at which they were exposed. The ducklings fed these tadpoles all died. Similarly all ducklings died within one half hour after ingesting tadpoles dosed to 5 ppm of parathion (Ibid 1980) (Table 3).

Fleming et al. (1982), fed American kestrels (Falco sparverius) frogs (A. crepitans) that had been exposed to concentrations of 0.1-10 ppm parathion for 96 hours. One bird of four died 3 hours after eating 5 frogs exposed to 10 ppm. No mortality of kestrels was recorded from any other group (Table 2). The authors noted that 10 ppm was an unrealistic exposure rate for field applications (Fleming et al. 1982). However, frogs can accumulate parathion from water as Fleming et al. (1982) found that after 96 hours parathion was not detectable in the water but was present in frogs (Ibid 1982) (Table 2). This may be due to the constant availability of parathion to the frogs which is a situation that is unlikely to be duplicated in the field.

## ORTHENE (Acephate)

### Short-term Effects

In tests using R. clamitans tadpoles Lyons et al. (1976), from the maximum likelihood regression analysis, established a 24 hour LC50 of 6433 ppm (Table 1). Similarly, Geen et al. (1984) reported an equally low toxicity for the salamander Ambystoma gracile. Sixty-nine day old larvae had a 96 hour LC50 of 8816 mg/L (Table 1). Amphibians would not be affected by control programs using Orthene as the maximum surface residue from current field application rates would be less than 1 ppm (Ibid 1984).

### Long-Term Effects

Geen et al. (1984) found that 798 mg/L of Orthene did not produce significant differences in mortality to Ambystoma gracile (Table 2). There was some decrease in the growth rate and the appearance of abnormalities in salamanders exposed to 382 and 798 mg/L (Table 2), but these are well above the levels likely to be encountered in the field (Geen et al. 1984).

Buckner and McLeod (1975) monitored R. sylvatica, Hyla crucifer and Ambystoma laterale for 7 days after an aerial application of Orthene in two mixed wood stands in Ontario. It was concluded that the amphibians "were not affected by the insecticide treatment" (Ibid 1975) (Table 3).

### Residues

When ducklings were force fed tadpoles exposed to 5 ppm Orthene no adverse effects or mortality were observed (Hall and Kolbe 1980) (Table 1).

### OTHERS

#### Short-Term Effects

Using P. triseriata in static toxicity bioassays Sanders (1970) determined the 96 hour LC50's of a number of organophosphate insecticides. Carbophenthion was found to be the most toxic of the sixteen substances tested with a 96 hour LC50 of 0.028 mg/L (Table 1). Naled, another insecticide, was moderately toxic with a 96 hour LC50 of 1.7 mg/L (Sanders 1970) (Table 1).

In 96 hour flow through experiments exposing R. catesbeiana tadpoles to 5 mg/L of dicrotophos no mortality was observed though there was evidence of adverse cholinesterase effects (Hall and Kolbe 1980).

Injection into the dorsal lymphatic sac of amphibians yielded the following LD50's for B. viridis; 188 mg/kg paraoxon, 540 mg/kg TEPP, 1,410 mg/kg Dimefox and 1,450 mg/kg DFP. LD50's for R. ridibunda were only conducted with paraoxon and TEPP and were 91 mg/kg and 34 mg/kg respectively (Edery and Schatzberg-Porath 1960) (Table 1). Symptoms of poisoning were reported as a generalized paralysis followed by a loss of ability to right themselves and finally coma. Recovery was observed after removal from the contaminated medium. Edery and Schatzberg-Porath (1960) stated that amphibians show an unusually strong resistance to organophosphorus intoxication.

#### Long-Term Effects

Chronic exposures to Abate, chlorpyrifos-ethyl, chlorpyrifos-methyl (0.03 or 0.06 ppm) and methyl-parathion (0.025 or 0.05 ppm) caused a reduction in the thermal tolerance of B. boreas (Johnson and Prine 1976) (Table 2). The levels employed only represented fifty percent of field concentrations (Ibid 1976). Johnson (1980) also tested temperature tolerance in the presence of temephos and chlorpyrifos and found it was significantly reduced even at concentrations less than recommended field application rates (Table 2).

Development of R. tigrina affected by DDT and methyl-parathion was studied by Mohanty-Hejmadi and Dutta (1981) (Table 2). The toxic effects caused by exposures of 1-7 mg/L varied depending upon the tadpole's developmental stage at the time of treatment. The limb-bud and well developed hind limb stages were the most sensitive to these insecticides. Complete developmental arrest occurred at higher concentrations and all treated frogs were smaller than controls at metamorphosis. In general, all the insecticides caused an extension of the life histories, even at concentrations lower than recommended field application rates (Ibid 1981). These effects were also observed by Dutta and Mohanty-Hejmadi (1978) in studies using different concentrations of Rogor (dimethoate) and R. tigrina (Table 2).

In field studies conducted by Mulla et al. (1963) insecticides were applied to ponds and the resulting mortality of amphibians recorded. Applications of 0.11 and 0.45 kg/ha of methyl-parathion resulted in no mortality to B. boreas and S. hammondii populations (Table 3). No mortality was observed for R. catesbeiana tadpoles following an application 0.56 kg/ha of naled, however applications of 1.8 and 0.45 kg/ha carbophenthion resulted in 100 percent mortality within twenty-four hours (Mulla et al. 1963) (Table 3).

Several other insecticides were tested (See Table 3 for data on Bayer 38920, G-27365, Bayer 34042, Ethyl Guthion, G.C.-3582, Bayer 44831, Bayer 29952, Bayer 22408, G-28029, Bayer 37289, Methyl-parathion, G-30494, G-30493 and Ronnel) and only G.C.-3582 was toxic to R. catesbeiana with 100 percent mortality when applied at 1.8 kg/ha (Mulla et al. 1963) (Table 3).

Field studies involving leptophos and phosphamidon concluded that these insecticides were highly toxic to frogs, causing 100 and 75 percent mortality respectively (Thirumurthi et al. 1973) (Table 3). In contrast Pearce and Price (1977) concluded that phosphamidon, due to relatively low toxicity observed in toxicity tests, would not present a major direct hazard to amphibians. Pearce and Teeple (1969) also examined the effects of sumithion spraying on amphibians in New Brunswick and concluded that at 0.14 kg/ha it had no effect on frogs and toads (Table 3).

## Residues

Hall and Kolbe (1980) studied the ability of R. catesbeiana to accumulate dicrotophos (Bidrin) to levels lethal to Mallard ducklings. Tadpoles dosed with 5 ppm and fed to ducklings caused some reduction in cholinesterase activity but resulted in no mortality of ducklings suggesting the tadpoles could not accumulate dicrotophos to levels lethal to duckling consuming one meal (Table 2).

## Mechanism of Toxicity

Organophosphate insecticides are toxic to most organisms due to the inhibition of acetylcholinesterase enzymes that are essential for nerve impulse conduction and transmission (Corbett 1974). This inhibition can lead to erratic altered behaviour (Lyons et al. 1976, Pawar et al. 1983, Sanders 1970). Erratic behaviour may place the organism at a selective disadvantage and lead to increased predation (Cooke 1971, Hall and Kolbe 1980, Lyons et al. 1976, Mohanty-Hejmadi and Dutta 1981). Direct effects of this reduction of cholinesterase activity have been studied by Dekin et al. (1978) who observed inhibition of muscle response caused by exposure to organophosphates (Table 2). Guy et al. (1977) showed a reduced amplitude of the response to stimulation of the sartorius muscle in the presence of OMPA (Table 2). These studies suggest that organophosphates may exert their toxicity by affecting the cholinergic receptor-channel complex.

Several studies have shown that amphibians can recover from this reduction of cholinesterase activity if returned to contaminant free media (Dekin et al. 1978, Guy et al. 1977, Guzman and Guardia 1978).

Potter and O'Brien (1964) attempting to explain the relative low toxicity of organophosphates to frogs in contrast with some insects and mammals concluded that the insensitivity of frogs to paraoxon was due to its insensitive cholinesterase which may account for the low relative toxicities to many organophosphate insecticides (Ibid 1963).

## d) PYRETHROIDS

### Short-Term Effects

Pyrethroids are a group of insecticides developed from a natural substance (Pyrethrum) and are of particular interest due to a short half life in the environment and their high toxicity to insects.

In static bioassays, permethrin had a 96 hour LC50 of 7.03 mg/L for two week old R. catesbeiana tadpoles (Jolly et al. 1978) (Table 1). Cole and Casida (1983) found frogs highly sensitive to eleven different pyrethroids. In general the trans-isomers were less toxic than the cis-isomers. The cis-isomers of permethrin (IR S)-Cypermethrin and Deltamethrin were the most toxic to adult R. pipiens (Ibid 1983) (Table 1). (S)-Fenpropathin and (SS)-Fenvalerate were also highly toxic to adult frogs (Table 1). Tadpoles of R. catesbeiana were especially sensitive to (IR, S)-Cypermethrin with an LD50 of 0.04 mg/kg (Ibid 1983) (Table 1), (See Table 1 for data on Pyrethrins, S-Bioallethrin, (IR)-Tetramethrin, Kadethrin, (IRS)-Resmethrin and (IR)-Phenothrin).

## Mechanism for Toxicity

Pyrethroid insecticides are recognized as typical neurotoxicants (Vijverberg et al. 1982).

Allethrin, decamethrin and 5-benzyl-3-furylmethyl(+)-cischrysanthemate (NRDC 119) acts on the peripheral nervous system inducing repetitive activity in sensory nerve fibres, sense organs and the distal portion of motor fibres resulting in repetition in the motor end-plate (Evans 1976, van den Bercken 1977, Wouters et al. 1977, Vijverberg and van den Bercken 1979) (Table 2). The repetitive activity of nerves occurs due to a transient increase in the prolongation of sodium permeability. The effect of pyrethroids on nerve fibres is temperature dependent with more pronounced effects at lower temperatures (Wouters et al. 1977, Vijverberg and Ruigt 1981) (Table 2). The effect of pyrethroids on nerve fibres can account for the symptoms of hyperexcitability, convulsive tremors and paralysis prior to death, noted in pyrethroid treated frogs (van den Bercken 1977) (Table 2). Pyrethroids have no direct effect on muscle fibres (Wouters et al. 1977) (Table 2).

Akkermans et al. (1975a) compared the effects of allethrin and several organochlorines and found DDT produced effects similar to those of allethrin (Table 2). DDT and allethrin produced repetitive firing in both the lateral-line organ and cutaneous touch receptors (Akkermans et al. 1975a, van den Bercken et al. 1973).

The frog nerve is not as sensitive to pyrethroids as crayfish nerves (Takeno et al. 1977), which could explain the lower sensitivity of R. catesbeiana tadpoles to permethrin in acute bioassays reported by Jolly et al. (1978) (Table 1).

## III HERBICIDES AND FUNGICIDES

### a) CARBAMIC ACIDS

#### MANEB

#### Short-Term Effects

Maneb (Manganese Ethylene bis dithiocarbamate) is a widely used fungicide in several parts of the world. Dithiocarbamates are known to contaminate plants, soil and water to certain degrees and thus present a hazard to public health (Arias and Zavanella 1979). Little is known of their toxicity and toxic effects to aquatic vertebrates. In tests to examine the survival of T. cristatus in maneb, Zaffaroni et al. (1978) found the fungicide caused 100% mortality within 15 days at 50 ppm (Table 2). It was also noted that males were slightly less susceptible than females (Ibid 1978).

### Long-Term Effects

The effects of maneb on the forelimb regenerative ability of newts T. cristatus was examined by Arias and Zavanella (1979) and Zavanella et al. (1979). Both studies found that limbs regenerated in maneb exhibited delayed growth, reduced melanogenesis and malformations (Table 2). An overall delay in growth was also observed in newts exposed to maneb (Zavanella et al. 1979) (Table 2).

Delay in development was also noted by Bancroft and Prahlad (1973) for X. laevis embryos placed in maneb solutions (Table 2). At levels of 1-5 ppm, maneb caused reduced melanogenesis, shortened tail and notochord waviness in developing embryos (Ibid 1973). At these levels no tumors were found in any test animals (T. cristatus) though the spleen weight increased and there was some edema of the soft tissues (Zavanella et al. 1979) (Table 2).

### Mechanism for Toxicity

Though little is known about the precise mechanism of lethal effects, Zaffaroni et al. (1978) hypothesized that the toxic effects of maneb are caused by the impairment of cutaneous respiration and osmoregulatory function of the skin.

### NABAM

#### Long-Term Effects

In experiments to determine nabam's effect upon amphibians, it was found that X. laevis embryos exhibited reduced melanogenesis, growth delays, shortened tail and notochord and a difficulty in swimming at various concentrations (Bancroft and Prahlad 1973) (Table 2). Ultrastructural changes were also induced by exposure to nabam (Prahlad et al. 1974) (Table 2). Anderson and Prahlad (1976) examined the synergistic effect of diquat (a herbicide) and nabam and noted many ultrastructural changes and a high mortality rate (See Diquat, Table 2). Thus, nabam, when applied at low concentrations may represent a serious embryotoxic and teratogenic hazard to developing organisms (Bancroft and Prahlad 1973, Anderson and Prahlad 1976).

### OTHER CARBAMIC ACID FUNGICIDES AND HERBICIDES

It was reported that preparations of carbamic acids were the most toxic substances in comparison with nine groups of herbicides and insecticides (Zazhivilov 1972).

Sodium diethyldithiocarbamate (NaDEDC) at concentrations of 1-3 mg/L caused severe embryo malformation, growth delay, notochord abnormalities and curvature of the body axis. At levels higher than 3 mg/L, NaDEDC was lethal within 24 hours (Ghate and Mulherkar 1980a). Yalan and eptam are two herbicides used to control weeds in agricultural fields. Though they both exhibit relatively low toxicities to tadpoles (Table 1), it was noted that they produced a decrease in acetylcholinesterase activity in fish similar to the effects induced by sevin (Perevozchenko 1977).

## b) DIQUAT AND PARAQUAT

### Short-Term Effects

Paraquat was the least toxic of a number of pesticides and herbicides tested using B. woodhousii and P. triseriata tadpoles (Sanders 1970) (Table 1). Johnson (1976) also reported that paraquat had a low toxicity in assays employing two Australian frog species (Adelotus brevis and Limnodynastes peroni) (Table 1). The large difference between the two species demonstrates the wide range in species sensitivity to paraquat.

Exposure to diquat during the egg stage can reduce survival later in development. R. pipiens eggs exposed to 100 mg/L diquat during fertilization or later in development had higher mortality post-hatching than controls (Bimber and Mitchell 1978) (Table 2).

Anderson and Prahlad (1976) also reported diquat to be highly embryotoxic to X. laevis embryos (Table 2). Concentrations higher than 2 ppm caused distortion in body shape and 78% mortality while levels lower than 2 ppm were embryotoxic (Ibid 1976).

### Long-Term Effects

No mortality, behavioural or developmental abnormalities were observed in R. temporaria or B. bufo tadpoles or T. vulgaris adults exposed in ponds treated with 1 mg/L diquat (Cooke 1977) (Table 3). Thirty-two days after the pond was treated with diquat, R. temporaria and B. bufo tadpoles were heavier than control tadpoles (Cooke 1977). The increase in tadpole weight was attributed to an algal bloom which had occurred due to the diquat. The algae appeared to be an acceptable food source for the tadpoles. The algae was, however, thought to have reduced the suitability of the pond as a newt breeding site (Ibid 1977).

Dial and Bauer (1984) speculated that paraquat could represent a serious hazard to natural populations of R. pipiens. Field application rates vary from 0.1 to 2.0 ppm for aquatic weed control and it was observed that after 12 days no frogs survived when exposed to 2.0 ppm paraquat (Table 2). Paraquat also caused growth delays, tail abnormalities and poor head development.

### c) OTHER HERBICIDES

Toxicity data for eleven herbicides have been reported by Sanders (1970) and Johnson (1976) for North American and Australian amphibians respectively (Table 1). The herbicides tested were: 1) 2,2 DPA, 2) Amitrole-T, 3) DEF, 4) DSMA, 5) Dicamba, 6) Fenoprop, 7) Hydrothol 191, 8) Molinate, 9) Picloram, 10) Sodium arsenite, 11) Trifluralin.

Johnson (1976) noted that fenoprop was the most toxic of the substances he tested against anurans. The thermal tolerance of some species was reduced when exposed to all of the herbicides. Both Sanders (1970) and Johnson (1976) found adult frogs to be more resistant than tadpoles due to physiological and morphological differences.

Monosodium methanearsonate (MSMA) was used in bioassays with Scaphiopus couchii juveniles and adults (Judd 1977) (Table 2). At 477 mg/L MSMA mortality of juveniles was 87% in 1 week, although at 4770 mg/L 100% of the juveniles died within 72 hours, while adults experienced only 20% mortality. High juvenile toad mortality was expected to occur when MSMA is used as a herbicide since the recommended application rate is 8.3 mg/L (Ibid 1977).

A triazine compound, cyanatryn had a 96 hour LC50 of 30 mg/L for R. temporaria tadpoles in static bioassays (Scorgie and Cooke 1979) (Table 1). At 20 mg/L tadpoles were lethargic with occasional spasmodic twitching and did not feed. Body weight declined in increasing concentrations of triazine. In 20 day exposures to cyanatryn within the range of recommended treatment rates, tadpole body weight declined by half from 240 mg to 114 mg (Ibid 1979) (Table 2).

### Long-Term Effects

Long-term effects of dichlobenil were found to be similar to those for diquat (Cooke 1977). In field applications, dichlobenil caused no mortality of B. bufo or R. temporaria tadpoles, but weights were increased as a result of elevated food resources due to an algal bloom caused by the dichlobenil (Table 3).

Two compounds with both herbicidal and fungicidal properties, Gesagard 50 and Miedzian 50, were introduced to X. laevis tadpoles to examine the effects on tadpole development (Jordan et al. 1977). At both concentrations tested, all tadpoles in Gesagard 50 died within 72 hours. Those exposed to Miedzian 50 suffered high mortality at the two concentrations. Transferal to tap water did not result in recovery



from the induced effects (Table 2). Application rates for these two compounds are up to 300 times greater than the concentrations used in the tests (Ibid 1977).

Chloranil is a herbicide that causes embryotoxic and other detrimental effects on developing X. laevis embryos (Anderson and Prahlad 1976) (Table 2). At 2.0 ppm chloranil, only 5 percent of the embryos used survived more than 24 hours and these exhibited alterations in development (Anderson and Prahlad 1976).

Defenuron (N-phenyl-N'-methyl urea) is a wide spectrum herbicide that has low toxicity for vertebrates but has been shown to inhibit development of mosquitoes (Paulov 1977). When R. temporaria tadpoles were exposed to 100 ppm defenuron development and metamorphosis were delayed (Table 2). One observation made by Paulov (1977) was the development of "giant" tadpoles. This was due to a multiplication of cells coupled with the inhibition of metamorphosis (Paulov 1977) (Table 2).

#### d) OTHER FUNGICIDES

Malachite green, a fungicide frequently used in fish culture, was tested in flow-through bioassays with adult Notophtalmus viridescens, R. pipiens and Bufo sp. tadpoles (Bills et al. 1977) (Table 1). Adult newts were more resistant to malachite green than either frog tadpoles or toad tadpoles (Table 1). The tadpoles had a sensitivity to malachite green similar to most fish species tested (Ibid 1977).

Afugan (a fungicide containing pyrasophos) was lethal at 1.0 mg/L to R. temporaria tadpoles (Paulov 1981). Exposures to afugan during larval development resulted in stimulation of growth but inhibition of metamorphosis. Tadpoles exposed to sublethal doses exhibited alterations in enzymatic activity of transaminase (Ibid 1981) (Table 2).

The effects of a number of chlorinated bisphenols were tested on grass frog blood (species not stated) (Flores and Buhler 1974). Most bisphenols were highly toxic to frog blood in vitro, resulting in hemolysis with the degree of hemolysis being directly related to incubation. In vivo exposures to lethal doses of bisphenols did not, however, cause hemolysis (Ibid 1974) (see Hexachlorophene, Table 2).

Anderson and Prahlad (1976) found dichlone to be extremely toxic to X. laevis embryos (Table 2). Concentrations above 0.2 ppm caused 100% mortality within two to four days. Additionally there was inhibition of growth and disruptions of the cranial end resulting in headless individuals.

#### e) PHENOXY ACID HERBICIDES

The Phenoxy acid herbicides are related to natural plant growth regulators and are widely used in agriculture and park maintenance to control weeds. The LC50 toxicity data for 2,4-D, 2,4,5-T, silvex and Weedar 64 are found on Table 1 (Sanders 1970, Johnson 1976).

Studies examining the effects of 2,4-dichloro-phenoxyacetic acid on developing R. temporaria tadpoles revealed no visible effects or tissue residues even after treatment with 50 ppm (Cooke 1972a). In contrast Buslovich and Borushko (1976) found that 1-2 mg/L of 2,4-dichloro-phenoxyacetic acid significantly inhibits metamorphosis when in solution with its sodium and diethylamine salts (Table 2).

TCDD (2,3,7,8-tetrachlorodibenzo-p-dioxin) is a highly toxic contaminant of commercial supplies of 2,4,5-T and silvex. TCDD causes teratogenic effects in mammals and fish, yet Beatty et al. (1976) and Neal et al. (1979) reported little if any effect on R. catesbeiana development and survival (Table 2). Following an accident in Italy which released TCDD, Fanelli et al. (1980) found the TCDD level in a toad to be the lowest of the five animal species examined (Table 4).

#### IV BACTERICIDES

##### FURANACE

Furanace, a nitrofuran used as a fish bactericide, was highly toxic to R. pipiens tadpoles in static bioassays, compared to the fish and invertebrate species that were tested (Marking et al. 1977) (Table 1). The egg stage was not as sensitive as treatments up to 4 ppm failed to produce any effect on development. Above 4.0 mg/L few embryos survived, either failing to develop or not escaping from the egg mass during hatching (Ibid 1977).

#### V LAMPRICIDES

##### TFM

The lampricide TFM (3-trifluoromethyl-4-nitrophenol) has been used to control the sea lamprey in the Great Lakes. In toxicity tests using Hyla versicolor, R. pipiens and R. catesbeiana tadpoles Chandler and Marking (1975) found little difference in species sensitivity (Table 1). TFM is applied in twelve hour exposures and it is doubtful that this would have any effect on amphibians (Chandler and Marking 1975).

#### VI ORGANICS

##### a) AROMATIC AMINES

##### Short-Term Effects

Toxicity studies revealed that of four aromatic amines and two methyl derivatives, acridine was the most toxic followed by quinoline, and its two methyl derivatives, (2-methyl-quinoline, 2,6-dimethyl-quinoline), aniline and pyridine (Dumont et al. 1979, Sloof and Baerselman 1980, Davis et al. 1981) (Table 1). All of the compounds caused embryos to become malformed in various ways. These included exogastrulation, edema, formation of blisters and at concentrations

near the 96 hour LC50's, axial lengthening resulting in short, fat larvae with underdeveloped digestive tracts. These effects were more severe in exposures to acridine than the three other amines (Davis et al. 1981). Electron micrographs of the exposed larvae revealed damage to the spinal chord and muscle alterations. In general, larvae, when exposed to sublethal concentrations, were immobile, slower to develop and had abnormal pigmentation (Dumont et al. 1979, Davis et al. 1981) (Table 1).

Greenhouse (1976a,b,c, 1977) assessed the toxicity of a group of organic amines found in jet fuels employed by the United States Air Force. Only N-phenyl-a-naphthylamine was found to be toxic to developing frogs (Greenhouse 1976a). Greenhouse (1976c) also found this compound to be teratogenic, whereas octyl-phenyl-a-naphthylamine and dioctyldiphenylamine were neither teratogenic nor otherwise toxic to frog embryos probably due to their insolubility (Table 2).

In studies to examine the effect of hydrazine and its methylated derivatives on development of X. laevis Greenhouse (1976b, 1977) determined that these compounds were teratogenic (Table 2). Two types of malformation were observed: i) malformations of the axial skeleton and tail and ii) those of the head and trunk.

This group of compounds is readily soluble in water and were found to be toxic to X. laevis embryos. At a concentration of 1 mg/L hydrazine, methylhydrazine, and symmetrical dimethylhydrazine caused irreversible toxic effects and 100 percent mortality to Xenopus larvae (Greenhouse 1976a) (Table 2). Greenhouse (1976a,b,c, 1977) noted that embryos were able to withstand much higher concentrations of these compounds than were the larval stages.

### **Mechanism for Toxicity**

The mechanism for toxicity of acridine, aniline, pyridine and quinoline was speculated by Davis et al. (1981) to be through the association of the amines with hydrophobic components of cells and tissue (Ibid 1981).

## **b) AROMATIC HYDROCARBONS**

### **Long-Term Effects**

Triturus cristatus adults when exposed to benzene in air at varying frequencies during a 43 day period exhibited a decrease in leukocyte number, while the number of white blood cells increased (Garavini and Seren 1978) (Table 2). Preceding a decrease in red blood cell numbers the red blood cells became enlarged and immature red blood cell numbers increased. These results were interpreted to indicate that benzene was inhibiting blood cell maturation with the degree of inhibition dependent on frequency of exposure (Ibid 1978).

Toluene had some of the same effects on the blood as benzene (Ibid 1978) (Table 2). T. cristatus, after inhaling varying doses of toluene, exhibited initially an increase in red blood cells followed by a reduction in number. There was no change in white blood cell count, but there was a reduction in the phagocytic activity of the neutrophils (Ibid 1978).

Aromatic hydrocarbons can lead to high incidences of cancer (Busbee et al. 1978). A. tigrinum collected from a sewage polluted pond with high concentration of polycyclic aromatic hydrocarbons were found to have abnormal metabolite ratios and a high rate of cancer (Ibid 1978).

### c) PHENOLS

#### Residues

Residues in tissues of R. temporaria adults after injections of 3 mg/kg of either phenol, 3-nitrophenol, 3,5-diethylphenol or 4-aminophenol were similar for all four phenol compounds (Nagel and Urich 1981). Residues declined with time except in the gallbladder and gut, indicating that biliary excretion was minor in comparison to renal elimination (Ibid 1981).

#### Mechanism for Toxicity

One mechanism of phenol toxicity has been demonstrated for the surfactant nonilphenoethoxylate (Celentano et al. 1979). In R. esculenta skin, the surfactant inhibits active ion transport-coupled water flow. The mechanism for the change in skin permeability was thought to occur as a result of the surfactant affecting protein structure and activity (Ibid 1979).

### d) DIOXIN

(See also III e) Phenoxy acid Herbicides)

#### Long-Term Effects

No mortality of R. catesbeiana tadpoles could be attributed to dosages of 0.025-1.000 mg/kg dioxin applied intra-peritoneally (Neal et al. 1979). Histopathological examination of the tadpoles after metamorphosis revealed no lesions. After adult R. catesbeiana were administered dosages of up to 0.500 mg/kg i.p. dioxin, no mortality occurred during the 35 day observation period. Also no histopathological lesions occurred due to the dioxin (Ibid 1979).

## VII METALS

### a) CADMIUM

#### Short-Term Effects

Bioassays to determine the toxicity of cadmium have produced contrasting results (Birge et al. 1979, Canton and Slooff 1982) (Table 1). These discrepancies may be due to inherent differences in flow-through and static techniques (Birge and Just 1975, Birge et al. 1979, Hall and Swineford 1980, 1981), species sensitivity and life stage sensitivity.

Miller and Landesman (1978) exposed X. laevis to varying concentrations of cadmium and observed at 0.001 mg/L 92% of the embryos survived but were severely malformed. In the presence of magnesium (2 mg/L) the toxic effects of cadmium were greatly reduced (Ibid 1978).

#### Long-Term Effects

Development and survival were severely affected by lengthy exposures to cadmium (Birge et al. 1977, Manson and O'Flaherty 1978) (Table 2). The most severe effects were noted by Birge et al. (1977) when R. pipiens eggs were exposed to cadmium at the cleavage stage (Table 2). No further development took place when the eggs were exposed to cadmium at this stage. At lower concentrations of cadmium survival was severely reduced (Table 2).

At low concentrations (0.03 mg/L) no mortality occurred, though at levels above 0.09 mg/L cadmium, development was inhibited (Canton and Slooff 1982) (Table 1).

Long-term exposures of Notophthalmus viridescens to 2.0-6.75 mg/L of cadmium caused mortality and retarded forelimb regeneration sometimes leading to abnormal limb regeneration (Manson and O'Flaherty 1978) (Table 2). Mortality was 82% at 6.75 mg/L after 51 days exposure, compared to no mortality in the controls. The deaths were partly attributed to weight loss, since all salamanders exposed to cadmium frequently refused food. All salamanders had their forelimbs removed at the elbow before exposure, in order to assess the effect of cadmium on regeneration. Limb regeneration was retarded, but after 76 days, development was similar to controls although severe abnormalities of the forelimbs were more common in the cadmium treated groups (Ibid 1978).

## Residues

In areas located near a zinc mill in Poland, five species of amphibians were analyzed for cadmium residues (Dmowski and Karolewski, 1979) (Table 4). The residues in organisms from contaminated areas were higher than the control location. Food chain accumulation was observed through the different trophic levels in this study.

Canton and Slooff (1982) observed a concentration factor of 130 times in X. laevis exposed for 50 days to a solution of cadmium in water (Table 4).

## Mechanism for Toxicity

Cadmium exerts its toxicity on amphibians in a number of ways. There is strong evidence to suggest that cadmium effects epithelial cells by inhibiting active ion transport by combining with sulfhydryl groups in the cell membrane and decreasing membrane potential (Kanno et al. 1978, Arita et al. 1979, Arhem 1980, Takada and Hayashi 1981a,b) (Table 2). The degree of inhibition of ion transport however, varies between different epithelial tissue (Hillyard and Gonick 1978, Takada and Hayashi 1978a,b) (Table 2). Cadmium appears to affect nerve tissue, at least in the heart, by suppressing the release of acetylcholine (Hayashi and Takayama 1978) (Table 2). Cadmium has been shown to cause a depression in the rod receptor cells in the retina of R. catesbeiana (Fox and Sillman 1979). Cadmium had a much greater effect on the rod cells than either Pb or Hg (Ibid 1979) (Table 2).

Treatment of R. nigromaculata eggs, prior to the first cleavage, at a level of 4 mg/L or higher, has been shown to reduce the number of primordial germ cells in the later developmental stages (Hah 1978) (Table 2). Reduction of primordial germ cells can lead to smaller gonads and partial or complete sterility (Bounoure et al. 1954).

## b) LEAD

### Short-Term Effects

Lead was highly toxic to Gastrophryne carolinensis eggs between fertilization and 4 days posthatching in continuous flow bioassays (Birge et al. 1979) (Table 1).

X. laevis, exposed during the embryo stage to 0.001-10 mg/L lead in static bioassays, resulted in 100% mortality at 10 mg/L and 82% deformities with 18% mortality at 0.001 mg/L (Miller and Landesman 1978). The toxic effect however was reduced when 2.0 mg/L or greater of magnesium was added (Ibid 1978).

Adult anurans appear to be much less sensitive to lead (Kaplan et al. 1967). R. pipiens adults exposed to lead nitrate in daily renewable bioassays had a 96 hour LC50 of 105 mg/L (Table 1). Frogs

exposed to lead had a permanent loss of semi-erect posture (Table 2). Other toxic effects observed were a sloughing of the integument, muscular twitching, salivation, sluggishness and death (Table 2).

### Long-Term Effects

Exposure of R. nigromaculata eggs to lead, prior to the first cleavage, resulted in a reduction of the number of primordial germ cells later in development (Hah 1978) (Table 2).

Dilling and Healey (1926) reported lead salts had a severe inhibitory effect on the development of frogs causing a slow down in growth and abnormal development (Table 2).

### Residues

Field collected R. esculenta from the River Toce had higher lead levels in tadpole tissues than the adult stage (Baudo 1976). He interpreted the difference in lead levels in the tadpole and adult to be the result of the differences in the lead levels of their foods. Tadpoles primarily eat vegetation which had higher lead levels than the insects eaten by adults. Adult females had high levels of lead in their liver (7.56 mg/kg) while the highest levels in males were found in the skin (3.66 mg/kg) (Ibid 1976).

Other studies have found similar results for lead accumulation. The liver and heart were found to have the highest lead concentrations in frogs (species not reported) in Katowice, Poland (Dec et al. 1979) (Table 2). Similarly, after X. laevis had ingested worms containing lead for four weeks, adults had the highest lead residues in the liver (30.75 mg/kg wet wt), kidney (81.32 mg/kg) and bone (55.98 mg/kg) with lower levels in the skin and muscle (less than 12.0 mg/kg) (Ireland 1977) (Table 4). Ireland (1977) compared lead levels in X. laevis with Peromyscus and concluded that since amphibians had much higher levels, amphibians may be important in concentrating lead in the food web.

Dmowski and Karolewski (1979) suggested amphibians pose a potential hazard due to food chain accumulation. Residue levels in amphibians from an area close to a zinc mine in Poland were high due to accumulation via prey species (Ibid 1979) (Table 4).

### Mechanism for Toxicity

Binding of lead in amphibian skin has been shown to be the result of the affinity of melanin for metals (Ireland et al. 1979) (Table 2). X. laevis adults exposed to similar lead levels were either dark adapted to increase the melanophore number, or light adapted to decrease melanophore number. Toads that were dark adapted accumulated significantly more lead than light adapted toads, implicating melanin as the lead binding site in the skin (Ibid 1979) (Table 2).

Lead exerts its toxic effects on amphibians in a number of ways. Kaplan et al. (1967) concluded that a reduction in red and white blood cells in R. pipiens adults exposed to lead nitrate was likely the result of an inhibition of the hematopoietic tissues.

Discolouration of the liver reported for Necturus (Dawson 1933), R. catesbeiana tadpoles (Barrett 1947) and R. pipiens adults (Kaplan et al. 1967), is thought to indicate either that phagocytosis of red blood cells is occurring or that lead is being metabolized and excreted through the gallbladder (Kaplan et al. 1967) (Table 2).

Lead can also affect vision. Dissected R. catesbeiana retina treated with lead caused a depression of the amplitude of the rod receptor cells (Fox and Sillman 1979). The effect was reversible when lead levels were reduced (Ibid 1979).

The main physiological mechanism of lead toxicity is thought to be the binding of lead to sulfhydryl groups and disruption of sulphur bridges (Celentano et al. 1979). This would change the structure of membrane proteins leading to changes in ionic channels (Ibid 1979).

#### c) MAGNESIUM

##### Short-Term Effects

Magnesium is an essential element that can compete with other metal ions and reduce their toxicity to developing embryos (Miller and Landesman 1978). Magnesium at 2.0-200 mg/L greatly reduced teratogenicity in X. laevis tadpoles exposed to mercury, lead, cadmium and manganese. Since magnesium levels in amphibians are constant regardless of environmental levels, there may be competition for some common carrier mechanism that transports divalent cations into the embryos, thereby reducing metal toxicity (Ibid 1978).

#### d) MERCURY

##### Short-Term Effects

Tests to determine the toxicity of twenty-two metals to amphibians revealed mercury to be the most toxic to Gastrophryne carolinensis eggs (Birge et al. 1979) (Table 1). The toad was more sensitive to mercury than three species of fish that were also tested. Ghate and Mulherkar (1980b) reported 100% mortality of Microhyla ornata eggs at the gastrula stage when exposed to mercuric chloride at concentrations above 3 mg/L (Table 1). Mercuric chloride also retarded development,



caused blisters on the body and curvature of the spine. The tadpole stage had a slightly lower resistance to mercuric chloride. Sublethal doses caused the tadpoles to be sluggish and distension of the body cavity was observed (Ibid 1980b) (Table 2). X. laevis exhibited many of these symptoms when they were treated with mercury at the egg stage (Miller and Landesman 1978).

Birge et al. (1977) examined the survival of G. carolinensis eggs treated with mercury before hatching. At the lowest concentration of mercury (0.146 ppm) 61% of the hatched embryos died within four days of hatching. Eleven percent of the hatched embryos exhibited gross congenital deformities at hatching (Table 2).

Studies using R. pipiens embryos at different stages revealed death and serious developmental defects can be induced by trace amounts of methylmercury (Dial 1976) (Table 2). Frog gastrulae appeared to be the most sensitive embryonic stage of the three stages tested.

### Long-Term Effects

Mercury caused severe damage to R. nigromaculata germ cells in eggs prior to the first cleavage (Hah 1978) (Table 2). Further proliferation of primordial germ cells was much reduced. Loss of primordial germ cells can result in reduced gonads and complete or partial sterility (Bounoure 1954).

### Residues

Mercury residues can be accumulated by most aquatic biota (Cox et al. 1975). Cox et al. (1975) found Ranidae tadpoles to accumulate moderately high levels (up to 6.41 mg/kg) when exposed to low levels of mercury. Less than 1% of the mercury residues in the tadpoles was in the methyl form (Ibid 1975) (Table 4).

In Yugoslavia mercury residues were measured in a number of amphibian species from sites with varying levels of mercury contamination (Byrne et al. 1975). The liver had the highest concentrations ranging from 0.19 mg/kg in frogs collected in uncontaminated areas, to 25.9 mg/kg in R. temporaria from a site near a mercury plant. The kidney also had high residue levels with lung, egg and muscle tissue accumulating almost an order of magnitude less mercury. The liver and kidney contained only a small fraction of the mercury in the methyl form, whereas the muscle contained almost 100% methylmercury (Ibid 1975) (Table 4).

Dustman et al. (1972) analyzed organisms from lake St. Clair to determine their mercury content. Great Blue herons (Ardea herodias) and common terns (Sterna hirundo) had the highest levels while frogs

(R. pipiens and R. catesbeiana) had the lowest (Table 4). Though frogs form part of the diet of herons, the high residue levels were believed to have been caused by the fish they consumed which contained high levels of total mercury (Ibid 1972).

Accumulation of mercury in the liver and kidneys is common in other animals. In mammals mercury binds to metallothionien and there is evidence that this is the primary binding site in amphibians as well (Mehra et al. 1980).

### **Mechanism for Toxicity**

Mercury has a high affinity for sulfhydryl groups and may affect enzymes and proteins associated membranes with the most severe effects occurring at nerve membranes (Dial 1976, Arhem 1980, Ghatte and Mulherkar 1980b).

Mercury has been reported to exert its toxic effect on amphibians in a number of ways. Lesions in the rods in eyes have been shown to occur in the perfused retina of R. catesbeiana when exposed to mercury (Fox and Sillman 1979) (Table 2).

Chang et al. (1975) after observing complete mortality of R. pipiens tadpoles within 48 hours of exposure to low levels of mercury, (0.05 ppm), speculated that the distention of the body cavity (Chang et al. 1975, Ghatte and Mulherkar 1980b) and mortality was the result of a disturbance of the osmotic regulator system caused by mercury.

Mercury at low concentrations (0.01-0.07 mg/L) can inhibit cell growth while at higher concentrations (0.1 mg/L) actually promote cell growth and tissue development (Chang et al. 1976). The actual mechanism for this dose dependent effect is not yet understood.

## **e) SELENIUM**

### **Short-Term Effects**

Although selenium is an essential trace element it can be acutely toxic in high concentrations. X. laevis embryos (at the gastrula stage) and tadpoles exposed to low levels of sodium selenite in daily renewal bioassays had increased survival compared to controls, but mortality rapidly increased at 2.0 mg/L and higher (Browne and Dumont 1979). Although toxicity of selenium to tadpoles was not high (Table 1) 50% of those hatched from exposed eggs exhibited abnormalities in development. Exposures to selenium during the egg stage caused spine curvatures, tail flexures and malformed heads. Tadpoles treated with selenium exhibited erratic swimming behaviour and sluggishness as well as epithelial blisters, abdominal edema and degeneration of muscle cells (Browne and Dumont 1979, 1980).

Although toxicity for the egg stage was not measured, eggs experienced a lower rate of mortality than tadpoles. The gelatinous coating of the egg was thought to present a permeability barrier affording partial protection to the egg.

In continuous flow bioassays, G. carolinensis eggs were much more sensitive to selenium with a 96 hour LC50 of 0.09 mg/L (Birge et al. 1979) (Table 1).

### Residues

X. laevis eggs exposed to 5 mg/L of selenium exhibited a rapid uptake for the first 24 hours (Browne and Dumont 1979). Up to 50% of these residues were retained in tadpoles for extended periods of time (Ibid 1979).

## f) ZINC

### Short-Term Effects

Zinc was the second most toxic metal of twenty-two metals tested using G. carolinensis eggs (Birge et al. 1979) (Table 1). B. boreas tadpoles exhibited higher resistance to zinc concentrations. Concentrations of 39 mg/L of zinc caused complete mortality within 12 hours whereas exposure to 0.1 mg/L caused no mortality even after 61 days of exposure (Porter and Hakanson 1976) (See Acid Mine Drainage Table 2).

Birge et al. (1977) found G. carolinensis eggs exposed to sediment bound zinc, did not suffer high mortality or teratogenesis at hatching (Table 2).

### Residues

In laboratory exposures, the zinc residues accumulated by T. granulosa had a half life of 3 1/3 years (Willis and Valett 1978). Loss of zinc was temperature dependent with higher temperatures shortening the half life. The skin, muscle, blood and liver had the highest residue levels and together these tissues accounted for 70% of the total body burden (Ibid 1978) (Table 2). Dec et al. (1979) stated that zinc accumulated in the heart of frogs taken from the environs of a steel plant in Poland (Table 3).

### Mechanism for Toxicity

Zinc appears to exert its toxic effect in a manner similar to other metals. In myelinated nerve fibres zinc has been shown to slow the kinetics of the potassium system and alter sodium permeability (Arhem

1980). Zinc can also change the sodium and potassium kinetics across epithelial cell membranes (Kanno et al. 1978) (Table 2). These results were thought to indicate that changes in membrane permeability are in part due to zinc combining with sulfhydryl groups in the cell membrane (Kanno et al. 1978) and in part due to the tendency of the zinc to form complexes with certain ligands.

## g) OTHER METALS

### Short-Term Effects

In continuous flow bioassays Birge et al. (1979) measured the 96 hour LC50s of a number of metals to G. carolinensis eggs (Table 1).

Components of acid mine drainage (iron, zinc, copper) were tested to determine their toxicity to amphibians (Porter and Hakanson 1976). Larvae were able to tolerate 20 mg/L of iron but at 30 mg/L there was complete mortality. Copper was toxic to B. boreas at 3.7 mg/L. When zygotes were placed in a solution of mine drainage 100% mortality occurred within 12 hours (See Acid Mine Drainage, Table 2).

R. pipiens eggs exposed in static bioassays to copper sulfate at low levels, exhibited no toxic effects (Landé and Guttman 1973) (Table 2). The tadpole stage was much more sensitive (Ibid 1973) (Table 1). Higher mortality in the tadpole stage has also been reported by Kaplan and Yoh (1961).

### Residues

Residues of chromium, manganese and copper measured in R. esculenta from the Toce River, Italy, were 0.42-9.22 mg/kg, 23-958 mg/kg and 7.63-32.4 mg/kg respectively in the tadpole stage (mean annual concentrations: chromium 0.01 mg/L; copper 0.005 mg/L; manganese 0.06 mg/L) (Baudo 1976). Neometamorphosed frogs had significantly lower chromium and manganese levels (0.47 mg/kg and 25.0 mg/kg respectively) than the tadpole stage. The significant drop in chromium and manganese residues in the neometamorphosed frog was attributed to a change in the metal levels in their food, as the tadpoles shift from being primarily herbivorous to being mainly insectivorous adults. In adults copper was most highly concentrated in the liver (61-63 mg/kg) while chromium and manganese had the highest concentrations in the skin (1.27-2.47 mg/kg and 30.3-67.9 mg/kg respectively) (Ibid 1976).

### Mechanism for Toxicity

Nickel and copper have been shown to slow down the kinetics of the potassium system in myelinated nerve fibres with copper having the greatest negative effect (Arhem 1980). Manganese and cobalt had no

effect. All four metals, however did alter sodium permeability with copper causing the greatest decrease and nickel the smallest. The toxicity of these metals is thought to be due to a similar mechanism as described for zinc, where the metals combine with sulfhydryl groups in cell membranes and alter membrane permeability (Ibid 1980).

Pytasz et al. (1980) reported a decrease in the general metabolic rate of frogs in an area close to a metallurgic worksite (See Metals, Table 2).

## VIII RADIOACTIVE ISOTOPES

### a) CESIUM

#### Residues

Radiocesium content was measured in Hyla cinerea adults collected from a contaminated swamp in South Carolina (Dapson and Kaplan 1975) (Table 3). Variability in body burdens was attributed to an inverse relationship between body burden and body size plus a large degree of variability in radiocesium content of food items. The biological half-life for unfed adult frogs at 20-30°C was 30.1 days (Ibid 1975) (Table 3).

### b) COBALT

#### Long-term Effects

Two B. canorus adults affixed with tags containing cobalt<sup>60</sup> and released in the field for 41 weeks exhibited no ill-effects from the exposure (Karlstrom 1957) (Table 3).

### c) STRONTIUM

#### Short-Term Effects

Birge et al. (1979) reported strontium to be one of the less toxic metals tested using G. carolinensis in flow-through toxicity tests (Table 1).

#### Long-Term Effects

T. granulosa adults exposed to radiostrontium for 25 days at 10°C exhibited an initial rapid uptake for 72 hours which declined without reaching equilibrium at day 21 (Willis and Valett 1971, 1976) (Table 2). The biological half-life was 136 days which was similar to the half-life in acute exposures using intraperitoneal injections. Significant concentrations were found in the blood, liver, skin and

muscle although the bone contained the largest portion. The biological half-life of radiostrontium for R. pipiens at 10°C after intraperitoneal injections was 22 days (Ibid 1971) (Table 2).

#### d) IONIZING RADIATION

The effects of ionizing radiation on amphibian development have been comprehensively reviewed by Brunst (1965).

### IX OTHER COMPOUNDS

#### a) INDUSTRIAL EFFLUENTS

X. laevis embryos exposed to sour water from a coal-gasification process exhibited high mortality and teratogenic effects (Dumont and Schultz 1980) (Table 1). The teratogenic effects ranged from tail flexure to gross distortions of the longitudinal axis, as well as the optic, cephalic, visceral and pericardial tissues. Loss of balance and reduced pigmentation were also common. The sour water was typical of most coal gasification processes and contained straight chain carboxyl acids, and varying amounts of aromatic hydrocarbons, aromatic amines and sulphur compounds (Ibid 1980).

R. ridibunda collected near the Katowice ironworks in Poland had significantly reduced metabolic rates that were inversely related to the distance from the mill (Pytasz et al. 1980). The reduction in metabolism was directly related to metal residues in tissues (Ibid 1980) (See Metals, Table 2).

#### b) ACID DRAINAGE

##### Short-Term Effects

Runoff from some rock types (particularly those containing pyrite), can be very acidic and have elevated metal levels which are highly toxic to amphibians (Huckabee et al. 1975, Mathews et al. 1976, Mathews and Morgan 1982). Eighty-three percent mortality of Leurognathus marmoratus larvae occurred in Anakeesta acid metal leachate (Huckabee et al. 1975). The 72 hour LC50 for L. marmoratus larvae exposed to the leachate from the same rock deposit was 33% (Mathews and Morgan 1982) (Table 1). The combined effects of acidity plus elevated metals, particularly zinc, copper and aluminum, were considered to be the toxic components of the leachate (Huckabee et al. 1975, Mathews and Morgan 1982). The toxicity of the leachate was reduced by the addition of sodium hydroxide (NaOH) (Mathews et al. 1976). The sodium hydroxide raised the pH and reduced the metal levels by decreasing solubility and inducing the formation of metal complexes. Neutralized leachate had a 72 hour LC50 of 113% (Mathews et al. 1976).

Porter and Hakanson (1976) attributed the absence of B. boreas from a creek in Colorado to a combination of toxic effects from acid mine drainage that was entering the stream. The stream water was non-toxic only after a 1,000 fold dilution. The toxic components of the drainage were hydrogen ion, copper, zinc and iron (Ibid 1976) (See Acid Mine Drainage, Table 2).

## X WATER QUALITY

### a) NITRITE

#### Short-Term Effects

Toxicity values for Ambystoma texanum larvae suggest that the salamander is extremely sensitive to nitrite (Huey and Beitinger 1980) (Table 1). Nearly 100% mortality was observed at levels as low as 2.5 mg/L nitrite. However, when chloride concentrations were increased, larvae suffered no mortality in exposures to 10 mg/L nitrite. Chlorides appeared to compete with nitrite in ionic uptake on respiratory surfaces, resulting in reduced uptake of nitrite (Huey and Beitinger 1980).

### b) pH

Acidity is an important water quality parameter that can affect amphibians directly and indirectly by influencing the toxicity of other contaminants. Only the direct effects will be discussed in this section. Indirect effects of pH are referred to in the section for the chemical where an effect has been noted.

#### Short-Term Effects

The direct short term effects of elevated hydrogen ion concentration have been observed at all lifestages. Fertilization of R. pipiens eggs was reduced below pH 6.5 (Schlichter 1981) (Table 2). Below pH 4.8 complete failure of egg development was attributed to low sperm motility. Fertilization was more susceptible to depressed pH than either activation or cleavage. The optimal pH for normal egg development was concluded to be above pH 6.0 (Ibid 1981). Gosner and Black (1957) estimated levels of pH that would allow 50% or more of embryos to survive in lab bioassays with tap water acidified with hydrochloric acid. The species and pH levels were as follows:

<u>Acris crepitans</u>	4.7
<u>H. versicolor</u>	4.3
<u>Rana catesbeiana</u>	4.3
<u>R. palustris</u>	4.3
<u>H. crucifer</u>	4.2
<u>R. clamitans melanota</u>	4.1

<u>R. pipiens</u>	4.1
<u>Pseudacris nigrata</u>	4.1
<u>R. sylvatica</u>	3.9
<u>Hyla andersoni</u>	3.8
<u>R. virgatipe</u>	3.8

Gosner and Black (1957) also reported the observed lethal level for all these species (Table 2).

Acid stressed embryos exhibited morphological and behavioural abnormalities that appeared to be a result of shrinkage of the perivitelline membrane. Embryos were cramped in the eggs and had difficulty hatching (Ibid 1957). Short-term pH depressions were also toxic to developing embryos. R. sylvatica eggs exposed for short periods to pH 3.5-3.7 exhibited increased mortality with the degree of toxicity dependent upon the lifestage and length of exposure (Ibid 1957).

A. maculatum and A. jeffersonianum had marked reductions in hatching success in lab and field exposures to pH less than 6.0 (Pough 1976, Pough and Wilson 1977) (Table 3, Table 2). Tolerable pH for A. maculatum was 6 to 10 while A. jeffersonianum tolerated pH 7 to 9 (Pough and Wilson 1977) (Table 2). A. maculatum experienced greater than 60% mortality in ponds with pH 6.0. Developmental abnormalities at pH 6.0 included stunted gills, swelling near the heart, failure of yolk plug retraction and curved spines (Pough 1976).

Optimal development of amphibians is dependent upon both pH and temperature (Punzo 1983). Punzo (1983) by exposing Ambystoma texanum to varying temperature and pH values discovered that optimum hatching occurred within a temperature range of 10 to 15°C with a pH range of 7 to 9 (Table 2). Beyond these ranges hatching success dropped to no survival at the extremes tested (Punzo 1983).

Dunson and Connell (1982) observed a marked inhibition of hatching of X. laevis eggs at a pH of 3.9 (sulfuric acid) and a pH of 4.3 (bog water) (Table 2). In natural breeding ponds A. maculatum hatching was completely arrested at a pH of 3.6 caused by acid precipitation (Nielsen et al. 1977) (Table 3). However, Cook (1983) found no significant correlation between pond pH and embryonic mortality for two species of salamander inhabiting ponds in an area of acidic rainfall (Table 3).

R. catesbeiana eggs and larvae exposed at sites in a stream draining a bog which exhibited a natural pH gradient from 3.9 to 6.6, showed a positive relationship between mortality and pH (Saber and Dunson 1978) (See Bog Water, Table 3). R. catesbeiana embryos and larvae between stage 18 and 26 (Gosner 1960) were extremely sensitive to pH, whereas R. catesbeiana and R. clamitans larvae beyond stage 26



were not sensitive. Mortality of R. catesbeiana and X. laevis was also greater in coloured water than clear water at the same pH, indicating some other toxic component of the bog water (Saber and Dunson 1978) (Table 3).

B. bufo and R. Temporaria exposed to pH 3.75-8.0 exhibited dramatic increases in mortality between pH 4.25 and 4.75 (Beebee and Griffin 1977) (Table 3). B. calamita was much more sensitive with increased mortality of eggs and larvae below pH 6.0 and complete mortality at pH 4.75. Two abnormalities were observed preceding some instances of mortality in the acidic exposures, an inhibition of growth beyond stage 5 (Taylor and Kollros 1946) and a bloating of the body late in development (Beebee and Griffin 1977) (Table 3).

### Long-Term Effects

pH affects the distribution of many amphibian species (Gosner and Black 1957, Cooke and Frazer 1976, Pough 1976, Beebee and Griffin 1977, Pough and Wilson 1977, Hagstrom 1977, 1980, Strijbosch 1979, Beebee 1983) (Tables 2 and 3). Most species avoid acid waters, although since there is a range in species sensitivities to acidity, some species such as R. sylvatica and R. clamitans which are more tolerant, will be found in more acidic habitats (Gosner and Black 1957, Saber and Dunson 1978). Other species such as A. jeffersonianum will be found only in circumneutral habitats (Pough and Wilson 1977). Although vegetation and other physical/chemical parameters are important in amphibian distribution, pH was found by Strijbosch (1979) to be one of the most important habitat characteristics. Amphibians can detect soil pH gradients and so can actively select acceptable habitat acidity (Mushinsky and Brodie 1975).

### Mechanism for Toxicity

Fromm (1981) examined osmoregulation across R. pipiens skin in vitro at varying pH levels. He found at low pH that the outer skin surface experiences a decrease in the flux of sodium ions into the skin, reducing overall sodium transport. He concluded that there was an ionoregulatory failure as a result of decreased uptake rather than an excess loss of ions.

Disruptions of ionic balance were speculated by Gosner and Black (1957) and Pough (1976) to be a cause of egg mortality. Gosner and Black (1957) suggested that this was the cause of shrinkage of the perivitelline membrane during egg development (Table 2).

### c) SALTS

#### Short-Term Effects

The toxicity of three chloride salts (potassium chloride, sodium chloride, calcium chloride) were measured using R. breviceps tadpoles in static bioassays (Mahajan et al. 1979). Tadpoles were most sensitive to potassium chloride with 73% mortality in 72 hours when exposed to 3000 mg/L. Sodium chloride above 5000 mg/L was highly toxic. Calcium chloride did not produce significant mortality until exposure to concentrations of 6000 mg/L and higher. All observed mortality was associated with respiratory failure and possible osmoregulatory imbalances resulting in sluggishness and loss of balance (Ibid 1979).

Sea water is highly toxic to amphibians. R. pipiens adults died within one hour of being placed in sea water (Bentley and Schmidt-Nielsen 1971) (Table 3). Post mortem analysis revealed that the frogs had accumulated large amounts of sodium through the skin and by drinking (Ibid 1971).

## XI EVALUATION OF AMPHIBIANS AS MONITORS

### a) CHEMICALS REPRESENTING A POTENTIAL HAZARD TO AMPHIBIANS

Wildlife toxicological concerns can be divided into two broad areas, direct concerns for wildlife health and the use of wildlife as indicators of environmental quality. In both cases, it is important to be able to carry out controlled laboratory experiments and as well as field studies.

The relative toxicity of various chemicals to amphibians are summarized in Table 5. Among the most toxic are the salts of heavy metals. The most sensitive life stage to metal toxicity is the egg followed by the larval and then adult stage (Birge et al. 1979). Birge et al. (1979) reported that treatment of the egg stage with metals (mercury, cadmium) produced mortality and teratogenesis in larvae at hatching (Table 2). Mercuric chloride, at less than 1 ppm, caused mortality, abnormal development and retarded growth (Ghate and Mulherkar 1980b) (Table 2). Though differences in species sensitivity have been observed, tolerance to pollutants must also be a function of the sensitivity of specific life stages. Birge et al. (1979) reported the sensitivity of G. carolinensis eggs to cadmium, but X. laevis larvae resistance was two orders of magnitude greater (Canton and Slooff 1982) (Table 2). Adult R. pipiens were more resistant to lead than G. carolinensis eggs as reported by Birge et al. (1979) (Kaplan et al. 1967) (Table 2). Thus different life stages display greater resistance to contaminants than others within the life cycle of one species.

Metals are not only toxic to amphibians but are also readily accumulated in body tissues and can be concentrated as they are passed up the food web (Byrne et al. 1975, Baudo 1976, Browne and Dumont 1979). Metals can be concentrated in amphibian tissues many times environmental levels and in some instances the metals are retained for long periods of time (Ireland 1977, Browne and Dumont 1979, Canton and Slooff 1982). Dial (1976) stated that in most environmental situations mercury stress would not be a factor due to low environmental levels, but that at point sources, such as industrial effluent disposal, populations R. pipiens could be seriously affected due to their acute sensitivity to mercury.

Of the insecticides the cyclodienes are the most toxic (Table 5). In particular endosulfan (Korschgen 1970, Gopal et al. 1981) endrin, toxaphene (Sanders 1970, Hall and Swineford 1981, 1982), and dieldrin (Sanders 1970) are the most toxic (Table 1). Cyclodiene toxicity not only can result in mortality but can have long-term effects on behaviour, morphology and development (Rane and Mathur 1978, Hall and Swineford 1980, Marchal-Segault and Ramade 1981). There is some indication that the tadpole stage is more sensitive to cyclodienes than the egg stage. For lindane X. laevis experienced greater mortality in exposures as tadpoles than during egg stage (Marchal-Segault and Ramade 1981) (Table 2). Hall and Swineford (1980) reported R. sphenoccephala experienced no mortality at the egg stage but increased mortality at the tadpole stage when exposed to endrin (Hall and Swineford 1980).

Larval amphibians have exhibited an ability to store organochlorine insecticides in their fatty tissues, most importantly the tail (Cooke 1970, 1973a, 1974, Licht 1976b, Lyons et al. 1976). During resorption of the tail at metamorphosis, these residues are mobilized and result in mortality among young frogs (Cooke 1970). Thus the hazard presented by some organochlorines affects all stages of the amphibian life cycle up to the adult stage, where mobilization and excretion of residues is more efficient (Niethammer et al. 1984).

Organophosphate insecticides presented the widest range in toxicity to amphibians (Table 5). Many organophosphates had extremely high toxicity (carbophention, parathion, malathion) (Sanders 1970) while others such as orthene were toxic only in large amounts (Lyons et al. 1976, Geen et al. 1984) (Table 1).

Symptoms of organophosphate poisoning were abnormal behaviour and development (Lyons et al. 1976, Marchal-Segault 1976), and alterations in thermal resistance (Johnson and Prine 1976) (Table 2).

Organophosphate residues can be concentrated above background levels (Lyons et al. 1976). Parathion and fenthion can be accumulated to levels lethal to ducks (Hall and Kolbe 1980) suggesting that organophosphate concentration in the food web could represent a serious hazard at higher trophic levels.

Carbamate, organochlorine (except the cyclodienes) and pyrethroid insecticides were all moderately toxic to amphibians resulting in behavioural and developmental abnormalities as well as mortality (Cooke 1970, 1979, Sanders 1970, Rzehak 1977, Jolly et al. 1978, Harri et al. 1979) (Tables 1 and 2).

Cooke (1972b) suggested that the levels of some insecticides necessary to produce slight hyperactivity in amphibian larvae are not encountered in the field in Britain and that hazards from insecticides would only occur due to misuse or accidents.

Acidification alone can impair egg and larval development (Gosner and Black 1957, Pough 1976, Pough and Wilson 1977, Schlichter 1981). The lowest pH that embryos can survive are listed on page 82. In view of the acidification of lakes and other smaller water bodies in eastern North America and much of Europe the effect on a number of species of amphibians should be a matter of concern. In addition to the direct effects of increasing acidification there are chronic effects on the ecology of the pond, changes in habitat structure, predator-prey relationships, food resources etc., which may profoundly affect anuran populations.

Amphibians which breed in temporary ponds may be the group most vulnerable to acidic precipitation. These temporary bodies of water often refill with rainwater and snowmelt during the early spring. Acidic water entering a dry pond is not diluted and may encounter little buffering as it enters through surface flow rather than as ground water. Semkin and Jeffries (1986) found that ionic concentration in the snowmelt was 2 to ten times greater than that of pre-melt material, the so-called "acid-shock".

The herbicides and fungicides include chemicals with a wider range in their toxicity to amphibians (Table 5). Malachite green had a 96 hour LC of 0.068 mg/L while DSMA had a 96 hour TL50 of 271 mg/L. Behavioural and developmental abnormalities have been observed after short or long term exposures to some herbicides (Arias and Zavanella 1979, Scorgie and Cooke 1979, Ghatge and Mulherkar 1980) (Table 2). Herbicides may exert indirect effects by altering the environment, for example the use of 2,4-D, which has low toxicity to amphibians, has been considered to reduce the diversity of prey species in an area and thus cause a reduction in amphibian numbers (Cooke 1972b).

A symptom of toxicity common to many of the compounds discussed in this review is an alteration to some degree of normal behaviour. This has been shown to put amphibians at a selective disadvantage which could result in depletion of some natural populations by overpredation (Cooke 1971, Lyons et al. 1976, Hall and Kolbe 1980, Mohanty-Hejmadi and Dutta 1981) (Table 2).

TABLE 5  
RANKING OF TOXICITY OF POLLUTANTS BASED ON THE 96 HR LC50

CHEMICAL	SECTION	SPECIES	STAGE	96 HR LC50 mg/L	RE
Mercury	Metals	<u>Gastrophryne carolinensis</u>	egg	0.001	19
Endosulfan	Insecticide	<u>Rana tigrina</u>	tadpole	0.0018	5
Endrin	Insecticide	<u>Rana catesbeiana</u>	tadpole	0.002	10
Zinc	Metals	<u>Gastrophryne carolinensis</u>	egg	0.01	19
Carbophenthion	Insecticide	<u>Pseudacris triseriata</u>	tadpole	0.028	2
Chromium	Metals	<u>Gastrophryne carolinensis</u>	egg	0.03	19
Arsenic	Metals	<u>Gastrophryne carolinensis</u>	egg	0.04	19
Cadmium	Metals	<u>Gastrophryne carolinensis</u>	egg	0.04	19
Copper	Metals	<u>Gastrophryne carolinensis</u>	egg	0.04	19
Lead	Metals	<u>Gastrophryne carolinensis</u>	egg	0.04	19
Aluminum	Metals	<u>Gastrophryne carolinensis</u>	egg	0.05	19
Cobalt	Metals	<u>Gastrophryne carolinensis</u>	egg	0.05	19
Germanium	Metals	<u>Gastrophryne carolinensis</u>	egg	0.05	19
Nickel	Metals	<u>Gastrophryne carolinensis</u>	egg	0.05	19
Toxaphene	Insecticide	<u>Rana sphenoccephala</u>	larval	0.032-0.054	8
Malachite green	Fungicide	<u>Bufo sp.</u>	tadpole	0.068	13
Selenium	Metals	<u>Gastrophryne carolinensis</u>	egg	0.09	19
Thallium	Metals	<u>Gastrophryne carolinensis</u>	egg	0.09	19
Tin	Metals	<u>Gastrophryne carolinensis</u>	egg	0.09	19
Dieldrin	Insecticide	<u>Pseudacris triseriata</u>	tadpole	0.10	2
Trifluralin	Herbicide	<u>Bufo woodhousii</u>	tadpole	0.10	2
Guthion	Insecticide	<u>Bufo woodhousii</u>	tadpole	0.13	2
TDE	Insecticide	<u>Bufo woodhousii</u>	tadpole	0.14	2
Aldrin	Insecticide	<u>Bufo woodhousii</u>	tadpole	0.15	2
Strontium	Metals	<u>Gastrophryne carolinensis</u>	egg	0.16	19
Malathion	Insecticide	<u>Pseudacris triseriata</u>	tadpole	0.20	2
Antimony	Metals	<u>Gastrophryne carolinensis</u>	egg	0.30	19
Methoxychlor	Insecticide	<u>Pseudacris triseriata</u>	tadpole	0.33	2
DEF	Herbicide	<u>Bufo woodhousii</u>	tadpole	0.42	2
Heptachlor	Insecticide	<u>Bufo woodhousii</u>	tadpole	0.44	2
Furanace	Fungicide	<u>Rana pipiens</u>	larval	0.77	17
DDT	Insecticide	<u>Pseudacris triseriata</u>	tadpole	0.80	2
Parathion	Insecticide	<u>Pseudacris triseriata</u>	tadpole	1.0	2
Piperonyl butoxide	Insecticide	<u>Pseudacris triseriata</u>	tadpole	1.0	2
Nitrite	Nitrite	<u>Ambystoma texanum</u>	larval	1.090	6
Hydrothol 191	Herbicide	<u>Bufo woodhousii</u>	tadpole	1.2	2
Manganese	Metals	<u>Gastrophryne carolinensis</u>	egg	1.42	19
Naled	Insecticide	<u>Pseudacris triseriata</u>	tadpole	1.7	2
2,2-DPA	Herbicide	<u>Limnodynastes peroni</u>	tadpole	2.0	1
Lindane	Insecticide	<u>Pseudacris triseriata</u>	tadpole	2.7	2
Tungsten	Metals	<u>Gastrophryne carolinensis</u>	egg	2.90	19
Amitrole-T	Herbicide	<u>Adelotus brevis</u>	tadpole	3.0	1

Benzene hexachloride	Insecticide	<u>Bufo woodhousii</u>	tadpole	3.2	2
Acridine	Organics	<u>Xenopus laevis</u>	egg	3.6	18
Fenitrothion	Insecticide	<u>Rana clamitans</u>	tadpole	4.9	7
Permethrin	Insecticide	<u>Rana catesbeiana</u>	tadpole	7.033	16
Molinate	Herbicide	<u>Bufo woodhousii</u>	tadpole	14.0	2
Fenoprop	Herbicide	<u>Limnodynastes peroni</u>	tadpole	22.0	1
Paraquat	Herbicide	<u>Bufo woodhousii</u>	tadpole	26.0	2
Quinoline	Organics	<u>Xenopus laevis</u>	egg	79.0	18
Picloram	Herbicide	<u>Adelotus brevis</u>	tadpole	95.0	1
Sodium arsenate	Herbicide	<u>Adelotus brevis</u>	tadpole	96.0	1
Dicamba	Herbicide	<u>Limnodynastes peroni</u>	tadpole	106.0	1
Matacil	Insecticide	<u>Rana clamitans</u>	tadpole	118.0	7
Aniline	Organics	<u>Xenopus laevis</u>	tadpole	150.0	18
2,4,5-T amine	Herbicide	<u>Limnodynastes peroni</u>	tadpole	169.0	1
2,4-D amine	Herbicide	<u>Adelotus brevis</u>	tadpole	200.0	1
DSMA	Herbicide	<u>Limnodynastes peroni</u>	tadpole	271.0	1
Pyridine	Organics	<u>Xenopus laevis</u>	tadpole	1090.0	18

References:

1=Johnson 1976, 2=Sanders 1970, 3=Marchal-Segault 1976, 4=Dumont and Schultz 1980, 5=Gopal et al. 1981, 6=Huey and Beitingger 1980, 7=Lyons et al. 1976, 8=Hall and Swineford 1980, 9=Wohlgemuth 1977, 10=Hall and Swineford 1981, 11=Mathews et al. 1976, 12=Landé and Guttman 1973, 13=Bills et al. 1977, 14=Browne and Dumont 1979, 15=Canton and Slooff 1982, 16=Jolly et al. 1978, 17=Marking et al. 1977, 18=Davis et al. 1981, 19=Birge et al. 1979.

## b) TOXICITY TESTING TECHNIQUES

Several species of amphibians are readily available from the field or from commercial distributors. Adult anurans can be housed at low temperatures, without food, until they are to be used. They can be induced to spawn readily (Rugh 1962, Browne and Dumont 1979). Spawn can be collected from the field and developed under laboratory conditions. Development in lab aquaria can easily be observed and amphibians do not generally require a large amount of water or space. Some studies have shown that anurans require less attention during these processes than urodeles. Urodeles require live food and can be difficult to handle, making anurans more attractive as a test organism (Slooff and Baerselman 1980).

Amphibians have several other attributes that make them useful test organisms. A short life cycle which involves physiological, histological and anatomical changes (Cooke 1981), allow observations of the complete cycle over a short period of time (Birge et al. 1975). They respond to environmental contaminants in many different ways and these can be monitored over the life of the organism.

Some concerns that are evident after reviewing the amphibian toxicological literature about methods and techniques that should be evaluated prior to any toxicity testing are as follows:

1. Section of species. There is a certain degree of species variability in response to environmental pollutants (e.g. Sanders 1970, Cooke 1972a,b, Rzehak et al. 1977, Birge et al. 1979) (Table 1). Although it is generally recommended that the most sensitive species be used in any toxicity testing, so little is known about the sensitivity of most amphibians that it is difficult to choose a single indicator species. X. laevis and R. pipiens are often used in toxicity testing. X. laevis does not naturally occur in Canada and may not reflect the sensitivity of native species. Whenever possible a number of species should be tested and results from single species should not be generalized to cover all amphibians until species sensitivities have been more extensively investigated (Hall and Swineford 1980, 1981).
2. Lifestages sensitivity. There is a wide range in sensitivity of lifestages to the same toxicant (e.g. Cooke 1972a,b, Wohlgenuth 1977, Lyons et al. 1976, Hall and Swineford 1980, Davis et al. 1981). In general, adults are more tolerant than either the egg or larval stages. The egg and larval stages exhibit wide ranges in sensitivity (Hall and Swineford 1980, Davis et al. 1981). The sensitive stages vary greatly between toxicants presumably dependent upon the physiological processes that are occurring in the organisms during the developmental stage and the physiological

mechanism causing toxicity (Davis et al. 1981). One process that has not been considered, except with respect to pH, is toxic effects on fertilization and egg cleavage. Schlichter (1981) found fertilization of R. pipiens eggs to be highly sensitive to pH 5.8. Species may be experiencing high mortality during fertilization which is not measured in standard egg bioassays. In all bioassays as many lifestages as possible should be tested and in all instances, the exact stages when exposures were initiated and terminated should be reported.

3. Toxic effect. Mortality, as measured in bioassays, is not the only toxic effect that can be induced by a pollutant. Behavioural, morphological and developmental aberrations that occur during chronic dosing at levels much lower than the LC50, can also be important measures of toxicity (e.g. Cooke 1970, 1973a, 1981, Lyons et al. 1976, Rzehak 1977, Harri et al. 1979, Hall and Swineford 1980). Residues are an indirect toxic effect that should be considered, particularly for pollutants that have little effect on amphibians but that may accumulate and pass up the food web (Rosato and Ferguson 1968, Collins et al. 1973, Baudo 1976).
4. Dilution water. Although dilution waters should vary to represent conditions where the pollutant is a concern, the water characteristics should always be reported, particularly pH, alkalinity and oxygen content. Generalizations cannot be made about the effects in all water qualities. pH is an important parameter that can be toxic by itself (Gosner and Black 1957, Pough 1976) and also can influence toxicity (Hashimoto and Nishiuchi 1981). Dissolved organic carbon levels are also important because some toxicants, such as metals, can interact with organics and modify the toxicity (Baker and Schofield 1980). Hardness or alkalinity is closely linked with pH, but alone it can influence toxicity (Porter and Hakanson 1976). For example, metal toxicity is greater in soft water than in hard water at the same pH (Ibid 1976).
5. Temperature. Toxicity can also vary with temperature during exposures (Licht 1976b, Pough and Wilson 1977). Temperature should always be reported and experiments should be conducted at a temperature comparable to what would be encountered in field conditions.
6. Holding technique. With only a few exceptions, aquatic amphibians inhabit lentic waters, often small temporary pools or ponds with very little water flow. Flow-through experiments may therefore not represent an accurate field situation. To maintain toxicant levels and avoid build up of waste product a flow-through system is often preferred to static bioassays. Hall and Swineford (1981) found



flowing water, with a five hour replacement time, caused moderate mortality to R. septentrionalis tadpoles and in later experiments reduced the flow to a 24 hour replacement (Ibid 1981). In comparison with Sanders' (1970) methods Hall and Swineford (1980) found that continuous-flow methods yielded lower LC50 estimates than static methods. However, flow-through methods with a rapid renewal time do not duplicate conditions in the field. Further investigation is necessary to fully assess the implications of static and flow-through bioassays on different life stages.

7. Rearing density and body size. Rearing density can affect size of tadpoles with high densities (50 tadpoles/L) producing tadpoles half the size of those reared at low densities (10 tadpoles/L) (Cooke 1979). The significance of size is that large individuals can be more resistant to pollutants than small individuals. For example body size was negatively correlated with T. cristatus sensitivity to Maneb (a fungicide) (Zaffaroni et al. 1978) and copper toxicity to R. pipiens tadpoles (Landé and Guttman 1973). Bioassays should therefore include either a random sample of sizes or should be designed to incorporate any effects of body size.
8. Observation period. The observation period to observe lethality after exposure to different chemicals needs to be determined for different groups of pollutants. Hall and Swineford (1980) reported much lower LC50s for toxaphene based on a 30 day observation period than Sanders (1970) did based on a 4 day observation period. Hall and Swineford (1980) attributed their lower LC50 mainly to the longer observation period because most mortalities were observed after the 4 day dosing between 1 and 26 days after exposure.

There are no generally accepted standardized techniques for toxicity testing using amphibians. The wide range of bioassay methodologies used in measuring LC50s makes comparison between studies very difficult. The United States EPA (1975) has published a detailed methodology which is recommended for use in toxicity testing of amphibians. However, it is designed mainly for fish, and to a lesser extent, aquatic invertebrate testing, and it does not fully address amphibian toxicity testing.

### c) AMPHIBIANS AS MONITORS OF ENVIRONMENTAL POLLUTANTS

Ideally indicator species should be both representative of a specific trophic level within the ecosystem and capable of being used, at reasonable cost, under laboratory conditions. The latter requirement has already been considered under section XI b).

The requirements for indicator species were listed by Moore (1966) as follows:

- (a) The species should be widely distributed, relatively abundant and easy to collect.
- (b) If monitoring is to be carried out by chemical analysis of organs, these should be large enough for adequate samples.
- (c) It should be possible to ascertain the age of the individual.
- (d) The level of residues in the species should be between the limit of detection and the limit of crude toxicological effect.
- (e) If measurements of local changes are required, the species must be sedentary. If the degree of contamination of a large area is to be measured, species with more extensive ranges can be used. In either case the range of the indicator species must be known.

Dumpert and Zietz (1984) considered that the platanna (X. laevis) could be used as an indicator species for determining embryotoxic effects of environmental chemicals. It met their criteria that it was available in sufficient numbers and at an acceptable cost at any time of the year and was adequately sensitive.

Cooke (1972a) has stated that under normal usage patterns that most insecticides would probably not be likely to reach levels high enough to induce toxic effects in the field. Meeks (1968) and Niethammer et al. (1984, 1985) believed that adult anurans would not be useful as indicators of organochlorine compounds as they do not accumulate residues in proportion with environmental levels.

The toxicity and toxic effects of organophosphate insecticides to amphibians varies greatly (Table 1) and due to the low persistence of these compounds in the environment, results are difficult to compare. Pearce and Price (1977) reported that spraying operations using fenitrothion represented little hazard to amphibians when used according to recommended field application rates. In contrast, Johnson and Prine (1976) reported that fenthion even when used at half the usual concentration caused marked physiological changes. These results, the apparent tolerance to cholinesterase inhibitors by amphibians (Potter and O'Brien 1964) and the generally low toxicity of organophosphates (Edery and Schatzberg-Porath 1960), indicates amphibians would not be adversely affected by many organophosphate compounds in the environment.

Arias and Zavanella (1979) presented data indicating the feasibility of using forelimb regeneration in newts as a model for assessing the risks incurred by the use of pesticides. However, the applications of this method may yield results difficult to apply to environmental situations.

Birge et al. (1973, 1975, 1976, 1977, 1979) have studied the effects of metal contamination on amphibians and the possibility of using amphibians as bioassay and bioindicator species based on the sensitivity of some amphibian species. Birge et al. (1975) considered that amphibians are an ideal test organism. The authors noted that due to differences in species sensitivity, that conclusions should be based on results from two or three test species. Birge et al. (1979) noted significant differences between urodele and anuran sensitivity based on individual ecological requirements, and concluded that sensitivity to environmental stress may be related to individual life cycles and requirements.

The sensitivity of amphibians to metallic contaminants may allow environmental monitoring of these compounds using amphibians as indicators, once standardized techniques and representative species have been established (Birge et al. 1975, 1979, Niethammer et al. 1985).

Amphibians can be used to monitor the effects of acidification. The survival of embryos has been related to pH, under both experimental and field conditions (see Section X b)). Fish are widely used monitor species for the impact of acidification. Amphibians could be used to broaden the approach and might be particularly valuable for assessing the impact on temporary water bodies.

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TABLE 1  
ACUTE TOXICITY STUDIES

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>				Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	72 hr	96 hr			
Acephate (OP)	- <u>Rana clematis</u>	tadpoles	A	housed at 21 ± 1	-	-	6433 (5857-6775)	-	-	-	-	95% fiducial limits in parentheses.	Lyons et al. 1976
	- <u>Achyrotace gracilis</u>	69 day old larvae	forced-daily	22 (7.0)	-	6 mg/L hardness as CaCO <sub>3</sub>	-	-	-	8816			Gren et al. 1984
Acetone	- <u>Achyrotace saxatilis</u>	3-4 week post hatch	A	20 ± 1	-	-	-	20,000	-	-		N = 10 for each species. Organisms not fed during expt.	Slooff and Boorsloot, 1980
	<u>Xenopus laevis</u>	3-4 week post hatch	"	"	"	"	-	24,000	-	-			
Acridine	- <u>Xenopus laevis</u>	mid-blastula embryos	A	Room Temp.	-	98.5 mg/L	-	10.9 (3.0-39.3)	8.7 (4.1-18.6)	3.6 (1.9-6.7)	teratogenicity EC50 for midblastula embryos: 48 hr-6.5 (3.1-13.7); 72 hr-2.6 (0.7-10.2); 96 hr-2.4 (0.8-7.0). Abnormalities included exogastrulation, edema, formation of blisters.	95% confidence limits in brackets.	Davis et al. 1981

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>				Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	72 hr	96 hr			
Acridine (cont'd)	- <u>Xenopus laevis</u>	tailbud embryos	A	Room Temp.	-	98.5 mg/L	13.2 (8.5-20.6)	7.1 (5.3-9.5)	5.5 (4.6-6.5)	-		95% confidence limits in brackets.	Davis et al. 1981 (cont'd)
		free swimming larvae	"	"	"	"	6.2 (5.5-7.0)	5.4 (4.7-6.3)	4.9 (4.1-5.9)	4.5 (3.7-5.5)			
Acrolein	- <u>Xenopus laevis</u>	tadpoles	B	17.2 ± 5 (7.39 ± .22)	-	40.4-49.5 mg/L as CaCO <sub>3</sub>	-	-	-	0.007 (.006-.008)		95% confidence interval in parentheses.	Holcombe et al., 1987
Aroazine-50 (propellant)	- <u>Ambystoma maculatum</u> , <u>A. opacum</u>	larvae	A	20.0-24.0 (7.8-8.2)	APHA et al. 1971	185-232 mg/L as CaCO <sub>3</sub>	>10	6.7	2.5				Slonie, 1986
			"	"	"	16-18 mg/L as CaCO <sub>3</sub>	>10	>10	5.2				

Table 1 - Acute toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References	
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr				
Aldrin (OC)	- <u>Bufo woodhousii</u> <u>foxierei</u>	4-5 week old tadpoles	A	15.5 ± .5 (7.1)	-	30 ppm methyl orange	2.0 (1.6-3.1)	0.68 (0.17-2.7)	0.15 (0.03-0.78)		N = 10. 95% confidence limits in brackets.	Sanders, 1970	
Allylamine	- <u>Amygdalus</u> <u>mexicanus</u>	3-4 weeks post hatch	A	20 ± 1	-	-	-	1.8	-		N = 10 for each species. Organisms not fed during expt.	Slooff and Baerseman, 1980	
	- <u>Xenopus laevis</u>	3-4 weeks post hatch	-	"	"	"	-	5.0	-				
Aluminum (M)	- <u>Gastrophryne</u> <u>carolinensis</u>	embryo-larval	B	-	-	Birge & Black 1977a,b	195.0 ± 5.4 ppm hardness as CaCO <sub>3</sub>				Exposure from fertilization to 4 days posthatch.	N = 14.	Birge et al., 1979a
									7 day				

Table 1 - Acute toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References	
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr				
Aminocarb (C)	- <u>Rana clamitans</u>	tadpoles: small 8.2 ± 1.1 mm snout body length; large 10.9 ± .1 mm body length	A	housed at 21 ± 1	-	-		267 (232-262)	206 (191-220)	118 (112-125)		95% fiducial limits in parentheses.	Lyons et al., 1976
2-Amino-ethanol	- <u>Xenopus laevis</u>	3-4 week old larvae	A	20 ± 1	-	-	-	-	220	-		N = 10 per concentration.	de Zwart and Slooff, 1987
1-Amino-2-propanol	- <u>Xenopus laevis</u>	3-4 week old larvae	A	20 ± 1	-	-	-	-	420	-		N = 10 per concentration.	de Zwart and Slooff, 1987
4-Aminopyridine (avian frightening agent)	- <u>Rana sphenoccephala</u>	larvae	A	16	ASTM Committee E-35 on Pesticides 1980	hardness 24 mg/L as CaCO <sub>3</sub>	7.2 (6.6-7.8)	-	2.4 (2.0-2.9)			95% confidence intervals in brackets.	Marking and Chandler, 1981

Table 1 - Acute toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>				Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	72 hr	96 hr			
Amitrole-T (H)	- <u>Adelotus brevicaudus</u>	1-2 week old tadpoles	A	21-22	Sanders 1970	-	5.2 g/L	5.0 g/L	-	3.0 g/L			Johnson, 1970
Aniline	- <u>Ambystoma mexicanum</u>	3-4 weeks post hatch	A	20 ± 1	-	-	-	660	-	-			N = 10 for each species. Organisms not fed during expt.
	- <u>Xenopus laevis</u>	3-4 weeks post hatch	"	"	"	"	-	560	-	-			
Anthracene	- <u>Xenopus laevis</u>	mid-blastula embryos	A	Room Temp.	-	98.5 mg/L	1600 (320-6100)	660 (470-980)	490 (330-730)	550 (350-850)	Teratogenicity EC50 for midblastula embryos: 24 hr-360 (180-740); 48 hr-560 (340-910); 72 hr-460 (280-750); 96 hr-370 (150-900). Abnormalities included exogastrulation, edema, formation of blisters.	95% confidence limits in brackets.	Davis et al 1981
		tailbud embryos	"	"	"	"	1620 (1100-2380)	1350	1150	980			
		free swimming larvae	"	"	"	"	-	1400 <sup>d</sup>	540 (350-830)	150 (100-200)			
	- <u>Rana pipiens</u>	embryo - stage 25	A	-	-	-	0.065	0.025	-	-			Kagan et al 1984

Table 1 - Acute toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	30 min	5 hr	7 day			
Antimony (M)	- <u>Gastrophryne carolinensis</u>	embryo-larval	B	- (7.0-7.8)	Birge & Black 1977a,b	195.0 ± 5.4 ppm hardness as CaCO <sub>3</sub>	-	-	0.30	Exposure from fertilization to 4 days post hatch.	N = 16.	Birge et al., 1979
Atprocarb (C)	- <u>Bufo bufo japonicus</u>	tadpoles	-	-	-	-	-	35	-			
Arsenic (M)	- <u>Gastrophryne carolinensis</u>	embryo-larval	B	- (7.0-7.8)	Birge & Black 1977a,b	195.0 ± 5.4 ppm hardness as CaCO <sub>3</sub>	-	0.04	-	Exposure from fertilization to 4 days post hatch.	N = 16.	Birge et al., 1979
Azinphos-methyl (OP)	- <u>Bufo woodhousei fowleri</u>	4-5 week old tadpoles	A	15.5 ± .5 (7.1)	-	30 ppm methyl orange	0.68 (0.45-1.6)	0.31 (0.17-0.56)	0.13 (0.05-0.33)		N = 10. 95% confidence limits in brackets.	Sanders, 1970
							24 hr	48 hr	96 hr			

Table 1 - Acute Toxicity (cont'd)

Contaminant*	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr			
Acetaminophen 593M	- <u>Rana bufo</u>	tadpoles 5.2 mg 16 mg	A	- (7.2)	American Public Health Assoc. 1971	-	220 470	- -	- -		N = 10 per concentration. LC50 given in concentration of protein (µg/ml).	Mathavan and Velupandi, 1984
Acetaminophen 562	- <u>Rana bufo</u>	tadpoles 5.2 mg 16 mg	A	- (7.2)	American Public Health Assoc. 1971	-	125 300	- -	- -		N = 10 per concentration. LC50 given in concentration of protein (µg/ml).	Mathavan and Velupandi, 1984
Anthracene (H)	- <u>Bufo bufo</u> <u> japonicus</u>	tadpoles	-	-	-	-	-	3.5	-			Hashimoto and Nishiuchi, 1981
Benzene	- <u>Ambystoma</u> <u> mexicanum</u>	3-4 weeks post hatch	A	20 ± 1	-	-	-	370	-		N = 10 for each species. Organisms not fed during expt.	Slooff and Baerelman, 1980
	<u>Xenopus laevis</u>	3-4 weeks post hatch	"	"	"	"	-	190	-			
	<u>Xenopus laevis</u>	3-4 week old larvae	A	20 ± 1	-	-	-	190	-		N = 10 per concentration.	de Zwart and Slooff, 1987

Table 1 - Acute Toxicity (cont'd)

Contaminant*	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr			
Benzene Hexachloride	- <u>Bufo woodhousii</u> <u> fowleri</u>	4-5 week old tadpoles	A	15.5 ± .5 (7.1)	-	30 ppm methyl orange	13 (4.3- 40)	7.1 (4.8- 10)	3.2 (0.30- 6.3)		N = 10. 95% confidence limits in brackets.	Senders, 1970
Beryllium Sulfate	- <u>Ambystoma</u> <u> opacum</u>	larvae	A	23.5 ± 1.5	APHA 1965	hard = 400 mg/L; soft = 20 - 25 mg/L	31.5 23.7	31.5 4.21	31.5 3.15	In ppm Be	N = 120.	Sionas and Kay, 1975
	<u>Ambystoma</u> <u> maculatum</u>	larvae	"	"	"	hard soft	18.2 21.2 ≥10 ≥12	18.2 18.2 ≥10 ≥7	18.2 18.2 8.02 5.65		N = 80.	
g-BHC	- <u>Bufo bufo</u> <u> japonicus</u>	tadpoles	-	-	-	-	-	24	-		Obtained using formulated product.	Hashimoto and Nishiuchi, 1981
S-Bioallethrin (PY)	- <u>Rana pipiens</u>	adults 20-30 g	-	20	subcuta- neous injection into the dorsum	-	-	-	-	24 hr LD50 = 1.7 (1.3-2.4)	N = 28-34. 95% confidence interval in parentheses.	Cole and Casida, 1985

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr			
3-Bromopropano	- <u>Xenopus laevis</u>	3-4 week old larvae	A	20 ± 1	-	-	-	0.66	-	N = 10 per concentration.	de Zwart and Slooff, 1987	
1-Butanol	- <u>Xenopus laevis</u>	3-4 week old larvae	A	20 ± 1	-	-	-	1200	-	N = 10 per concentration.	de Zwart and Slooff, 1987	
2-Butanol	- <u>Xenopus laevis</u>	3-4 week old larvae	A	20 ± 1	-	-	-	1530	-	N = 10 per concentration.	de Zwart and Slooff, 1987	
Cadmium (M)	- <u>Bufo calomastictus</u>	tadpoles 1.8-2.2 cm	A	29-30 (7.1- 7.6)	APHA et al. 1976	120-160 mg/L as CaCl <sub>2</sub>	19.81 (17.39- 29.15)	11.91 (9.99- 13.99)	8.18 (6.96- 9.53)	95% confidence limits in brackets. Animals not fed 24 hr prior to or during tests.	Khanjari and Ray, 1987	
	- <u>Microhylis ornata</u>	tadpoles	re-nosed daily	25.5- 26.0 (6.86- 6.90)	-	97.0-98.0 ppm				Ranges in parentheses. Animals not fed during test.	Rao and Madhyastha, 1987	
		1 week old					2.62 (2.0- 2.8)	2.48 (2.0- 2.8)	1.58 (1.4- 1.8)			
		4 weeks old					2.78 (2.0- 3.0)	2.66 (2.2- 2.8)	1.81 (1.4- 2.0)			

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr			
Cadmium (cont'd)	- <u>Bufo regularis</u>	adult females 25 ± 4.3 g	-	-	Single i.m. injection CdCl <sub>2</sub>	-	-	-	-	24 hr - 22 48 hr - 18 96 hr - 6.2		Hiley et al., 1986a
	- <u>Gastrophryne carolinensis</u>	embryo-larval	B	- (7.0- 7.8)	Birge & Black 1977a,b	195.0 ± 5.4 ppm hardness as CaCl <sub>2</sub>		0.06		Exposure from fertilization to 4 days post hatch	N = 10.	Birge et al., 1979a
	- <u>Xenopus laevis</u>	3-4 weeks	C	20 ± 1	Dutch Standardization Organization 1980	1.7 mmol/L hardness	4 (16)	3.2 (12.8)	-	No Toxic Effect Level for 24 or 48 hr -2.2.	N = 10. Values are corrected (based on actual test concentrations); uncorrected values are in parentheses.	Canton and Slooff, 1982a
Cadmium Nitrate	- <u>Ambystoma mexicanum</u>	3-4 weeks post hatch	A	20 ± 1	-	-	-	1.3	-		N = 10 for each species. Organisms not fed during expt.	Slooff and Baerelman, 1980
	- <u>Xenopus laevis</u>	3-4 weeks post hatch	"	"	"	"	-	32	-			

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>				Effects <sup>c</sup>	Remarks	References	
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	72 hr	96 hr				
Cadmium Nitrate (cont'd)	- <u>Xenopus laevis</u>	3-4 weeks old larvae	A	20 ± 1	-	-	-	20.2	-	-		N = 10 per concentration.	de Zwart and Slooff, 1987	
Captao (F)	- <u>Bufo bufo japonicus</u>	tadpoles	-	-	-	-	-	3.0	-	-		Obtained using formulated product.	Hoshimoto and Nishiyuchi, 1981	
Carbaryl (C)	- <u>Rana tigrina</u>	tadpoles weight: 0.1 g 0.02 g 1.2 g	A	-	-	-	12.8	8.25	-	6.3	Median Resistance time for 0.5 g tadpoles: 5250 min at 10 ppm; 110 min at 30 ppm.	N = 10 per group. LC <sub>50</sub> is weight dependent factor.	Marian et al., 1983	
	- <u>Bufo bufo</u>	26-37 day old tadpoles	A	20 ± 1	-	-	20.5- 21.8	18.2- 20.8	16.8- 20.6	-	Development slowed.	95% confidence intervals.	Marchei-Séguin, 1976	
	- <u>Bufo bufo japonicus</u>	tadpoles	-	-	-	-	-	7.2	-	-		Obtained using formulated product.	Hoshimoto and Nishiyuchi, 1981	
Carbophenothion (OP)	- <u>Pseudacris triseriata</u>	1 week old tadpoles	A	15.5 ± 5 (7.1)	-	30 ppm methyl orange	24 hr	48 hr	96 hr					
							0.10 (0.030-0.20)	0.05 (0.010-0.25)	0.028 (0.006-0.09)					N = 10. 95% confidence limits in brackets.

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr			
Carboxymethyl-succinate	- <u>Xenopus laevis</u>	3-4 weeks	semi-static	21 ± 2	-	-	-	-	1,800- 3,200		Test solutions corrected for oxygen content.	Canton and Slooff, 1982b
Cartap	- <u>Bufo bufo japonicus</u>	tadpoles	-	-	-	-	-	5.6	-		Obtained using formulated product.	Hoshimoto and Nishiyuchi, 1981
1-Chloro-3-bromopropane	- <u>Xenopus laevis</u>	3-4 week old larvae	A	20 ± 1	-	-	-	41	-		N = 10 per concentration.	de Zwart and Slooff, 1987
3-Chloro-2-methylpropene	- <u>Xenopus laevis</u>	3-4 week old larvae	A	20 ± 1	-	-	-	10	-		N = 10 per concentration.	de Zwart and Slooff, 1987
6-Chloro-2-picolinic Acid (H)	- <u>Pseudacris triseriata</u>	1 week old tadpoles	A	15.5 ± 5 (7.1)	-	30 ppm methyl orange	18 (9.0-27)	12 (6.0-24)	6.0 (2.0-10.0)		N = 10. 95% confidence limits in brackets.	Sanders, 1970
3-Chloro-1,2-propanediol	- <u>Xenopus laevis</u>	3-4 week old larvae	A	20 ± 1	-	-	-	581	-		N = 10 per concentration.	de Zwart and Slooff, 1987
3-Chloro-1-propanol	- <u>Xenopus laevis</u>	3-4 week old larvae	A	20 ± 1	-	-	-	2750	-		N = 10 per concentration.	de Zwart and Slooff, 1987

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References		
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr					
3-Chloropropene	- <u>Xenopus laevis</u>	3-4 week old larvae	A	20 ± 1	-	-	-	0.34	-	Exposure from fertilization to 4 days post hatch.	N = 10 per concentration.	de Zwart and Slooff, 1987		
Chromium (VI)	- <u>Gastrophryna carolinensis</u>	embryo-larval	B	-	Birge & Bick 1977a,b	195.0 ± 5.4 ppm hardness as CaCO <sub>3</sub>	7 day				0.03	In high concentrations surfacing, erratic body movement, loss of equilibrium noted.	N = 14.	Birge et al., 1979a
							24 hr	48 hr	96 hr					
	- <u>Bufo alvarius</u>	larval	A	29-30 (7.1-7.6)	APHA et al. 1976	120-160 mg/L as CaCO <sub>3</sub>	57.97 (49.14-65.48)	53.43 (49.21-61.67)	49.29 (43.63-56.39)		95% confidence limits in brackets. Animals not fed 24 hr prior to or during tests.	Khengarot and Ray, 1987		
Citric-5-5	- <u>Xenopus laevis</u>	3-4 weeks	semi static	21 ± 2	-	-	-	-	10,000	Exposure from fertilization to 4 days post hatch.	Test solutions corrected for pH and oxygen content.	Canton and Slooff, 1982b		
							7 day							
Cobalt (VI)	- <u>Gastrophryna carolinensis</u>	embryo-larval	B	- (7.0-7.8)	Birge & Bick 1977a,b	195.0 ± 5.4 ppm hardness as CaCO <sub>3</sub>		0.05			N = 14.	Birge et al., 1979a		

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr			
Cobalt (60Co)	- <u>Liamodynetes tennesseensis</u>	fertilized egg (.75 hr)	-	25	Cobalt-60 gamma radiation from a liberation unit at dose rate of 0.3 Gy/min.	-	-	-	-	LD50 values (Gy):	N = 100 - 120.	Panter, 1986
		late cleavage (10 hr)								0.6	40 day observation.	
		tail bud (33 hr)								3.3	40 day observation.	
		heartbeat (3 days)								9.9	40 day observation.	
		early limb bud (9-10 days)								10.4	60 day observation.	
		toe development (40-50 days)								20.2	60 day observation.	
									20.9	60 day observation.		



Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr			
Cobalt (60Co) (cont'd)		metamorphosis ( 60 days)	-	25	0.3 Gy/min	-	-	-	-	18.3	160 day observation.	Panter, 1986 (cont'd)
		young frog ( 80 days)								18.7	160 day observation.	
Copper (M)	- <u>Bufo melanostictus</u>	tadpoles 1.8-2.2 cm	A	29-34 (7.1-7.6)	APHA et al. 1976	120-160 mg/L as CaCO <sub>3</sub>	.845 (.731-.936)	.446 (.36-.55)	.32		95% confidence limits in brackets. Animals not fed 24 hr prior to or during tests.	Khangerol and Ray, 1987
		tadpoles	re-nerved daily	25.5-26.0 (6.86-6.94)	-	97.0 98.0 ppm					Range in brackets. Animals not fed during tests.	Mao and Madhyastha, 1987
	1 week old					5.61 (5.4-5.8)	5.31 (5.0-5.4)	5.74 (4.7-5.1)				
	4 weeks old					6.04 (5.6-6.4)	5.74 (5.4-6.2)	5.74 (5.0-5.8)				

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References	
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity		7 day					
Copper (cont'd)	- <u>Gastrophysa carolinensis</u>	embryo-larval	B	- (7.0-7.8)	Birge & Black 1977a,h	195.0 ± 5.4 ppe hardness as CaCO <sub>3</sub>		0.04		Exposure from fertilization to 4 days post hatch.	N = 14.	Birge et al., 1979a	
Copper Sulfate	- <u>Rana pipiens</u>	larval-immediately post hatch	A	19.4 ± 1.2 (7.73 ± 0.5)	-	290 ± 32 mg/L as CaCO <sub>3</sub>		72 hr				N = 35 per concentration. Survival time was positively correlated with weight of tadpole.	Lundé and Guttman, 1973
								30 day					
	- <u>Rana pipiens</u>	adults	A	20	-	-		.0016%				Kepler and Yoh, 1961	
	- <u>Xenopus laevis</u>	3-4 week old larvae	A	20 ± 1	-	-		24 hr 48 hr 96 hr				N = 10 per concentration.	de Zwart and Slooff, 1987

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr			
o-Cresol	- <u>Achyrocline satureioides</u>	3-4 weeks post hatch	A	20 ± 1	-	-	-	40	-		N = 10 for each species. Organisms not fed during expt.	Slooff and Baersemans, 1980
	- <u>Xenopus laevis</u>	3-4 weeks post hatch	"	"	"	"	-	38	-			
Cyanotryn (H)	- <u>Rana temporaria</u>	tadpoles	No test methodology specified				-	-	30			Hoddae et al., 1978
1,5,9 - Cyclohexadecatrieno	- <u>Xenopus laevis</u>	3-4 week old larvae	A	20 ± 1	-	-	-	1.6	-		N = 10 per concentration.	de Zwart and Slooff, 1987
1,3,5 - Cycloheptatrieno	- <u>Xenopus laevis</u>	3-4 week old larvae	A	20 ± 1	-	-	-	41	-		N = 10 per concentration.	de Zwart and Slooff, 1987
1,5 - Cyclo-octadecano	- <u>Xenopus laevis</u>	3-4 week old larvae	A	20 ± 1	-	-	-	24	-		N = 10 per concentration.	de Zwart and Slooff, 1987

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr			
1R, aS - Cypermethrin (PY)	- <u>Rana catesbeiana</u>	tadpoles 10-15 g	-	20	intrapertoneal injection	-	-	-	-	24 hr LD50: <u>trans</u> <u>cis</u> 0.20        0.04 (0.06-    (0.01- 0.41)    0.24)	N = 32-45. 95% confidence limits in parentheses.	Cole and Casida, 1983
	- <u>R. pipiens</u>	adults 20-30 g	-	20	subcutaneous injection into the dorsum	-	-	-	-	24 hr LD50: <u>trans</u> <u>cis</u> 0.65        0.16 (0.41-    (0.12- 1.0)        0.24)	N = 28-34. 95% confidence limits in parentheses.	
2,4-D (H) (2,4-dichlorophenoxyacetic acid)	- <u>Bufo melanostictus</u>		A	Approx. 25 (8.3)	APHA 1980	220 mg/L total hardness	13.77 (11.81-16.05)	9.05 (8.23-9.91)	8.05 (7.29-8.81)	Incipient Lethal Level = 6.1.	95% fiducial limits in parentheses.	Verste et al., 1984
2,4-D amine	- <u>Adelotus brevis</u>	1-2 week old tadpoles	A	21-22	Sanders 1970	-	255	228	200			Johnson, 1976
	- <u>Limnodynastes peroni</u>	1-2 week old tadpoles	"	"	"	"	321	300	287			
	- <u>Bufo marinus</u>	2 week old tadpoles	"	"	"	"	346	335	288			

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References															
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr																		
2,4-D base (cont'd)	- <u>Pseudacris triseriata</u>	1 week old tadpoles	A	15.5 ± .5 (7.1)	-	30 ppm methyl orange	100	-	100		N = 10.	Sanders, 1970															
2,4-D isocytal ester (Agoxone 5)	- <u>Iriturus cristatus</u> - <u>Iriturus carnifex</u>	adults	renewed daily	-	-	-	-	-	-	L50 values (lethal time for 50% of individuals) hrs <table border="1"> <thead> <tr> <th>conc</th> <th>male</th> <th>female</th> </tr> </thead> <tbody> <tr> <td>150</td> <td>14 (6.8-28.7)</td> <td>not determined</td> </tr> <tr> <td>125</td> <td>not determined</td> <td>52 (34.3-78.9)</td> </tr> <tr> <td>75</td> <td>102 (85.1-122.3)</td> <td>132 (105.6-165.0)</td> </tr> <tr> <td>50</td> <td>440 (321.9-601.5)</td> <td>-</td> </tr> </tbody> </table>	conc	male	female	150	14 (6.8-28.7)	not determined	125	not determined	52 (34.3-78.9)	75	102 (85.1-122.3)	132 (105.6-165.0)	50	440 (321.9-601.5)	-	Animals immersed in Agoxone 5 (57% 2,4-D; 63% diluents and emulsifiers); concentrations for L50's are 2,4-D concentrations. N = 10 per concentration 95% confidence limits in parentheses.	Zaffarini et al., 1986
conc	male	female																									
150	14 (6.8-28.7)	not determined																									
125	not determined	52 (34.3-78.9)																									
75	102 (85.1-122.3)	132 (105.6-165.0)																									
50	440 (321.9-601.5)	-																									

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>				Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	72 hr	96 hr			
2,4 - D sodium	- <u>Bufo bufo japonicus</u>	tadpoles	-	-	-	-	-	>40	-	-			Heshimoto and Nishiuchi, 1981
DDT (OC)	- <u>Rana temporaria</u>	adults	-	10-15	Oral administration of DDT dissolved in olive oil.	-	-	-	-	-	96 hr LD50 (estimated from graph) - 24; 20 day LD50 - 7.6.	Toxicity was increased by starving.	Harris et al., 1979
	- <u>Bufo bufo</u>	26-37 day old tadpoles	A	20 ± 1	-	-	0.7-1.3	0.3-0.8	0.3-0.5	-	95% confidence intervals.	Development is slowed. DDT more toxic than Sevin or Fenthion.	Marchal-Segault, 1976
	- <u>Pseudacris triseriata</u>	one week old tadpoles	A	15.5 ± .5 (7.1)	-	30 ppm methyl orange	1.4 (0.91-2.8)	0.90 (0.40-1.5)	-	0.80 (0.50-2.3)		N = 10. 95% confidence limits in brackets.	Sanders, 1970

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr			
DDT (cont'd)	<u>Bufo woodhousii fowleri</u>	tadpoles: 1 week old weight $\bar{x}$ = 15 mg  2-3 weeks old weight $\bar{x}$ = 56 mg  4-5 weeks old weight $\bar{x}$ = 76 mg  6 weeks old weight $\bar{x}$ = 350 mg  7 weeks old weight $\bar{x}$ = 600 mg	A	15.5 ± .5 (7.1)	-	30 ppm methyl orange	5.3 (2.9- 9.9)	1.8 (0.95- 3.3)	0.75 (0.28- 2.0)		N = 10.	Sanders, 1970 (cont'd)
	- <u>Bufo bufo japonicus</u>	tadpoles	-	-	-	-	31			Obtained using formulated product.	Hashimoto and Nishizuchi, 1981	

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr			
DEF (H)	- <u>Bufo woodhousii fowleri</u>	4-5 week old tadpoles	A	15.5 ± .5 (7.1)	-	30 ppm methyl orange	1.2 (0.90- 2.6)	0.76 (0.46- 0.82)	0.42 (0.16- 1.1)		N = 10. 95% confidence limits in brackets.	Sanders, 1970
cis - Deltamethrin (PY)	- <u>Rana pipiens pipiens</u>	adults 20-30 g	-	20	subcuta- neous injection into the dorsum	-	-	-	24 hr LD50: 0.35 (0.19-0.62)		N = 28-34. 95% confidence limits in parentheses.	Cole and Centis, 1981
	- <u>R. catesbeiana</u>	tadpoles 10-15 g	-	20	intraperi- toneal injection	-	-	-	24 hr LD50: 0.13 (0.05-0.34)		N = 32-45. 95% confidence limits in parentheses.	
DFP (OP) (diisopropyl fluorophosphate)	- <u>Bufo varidis</u>	adults 18-45 g	-	25	injection into dorsal lymphatic sac	-	-	-	7 day LD50: 1450.		N = 25 minimum.	Ebery and Schatzberg- Porath, 1960
Diazinon (OP)	- <u>Bufo bufo japonicus</u>	tadpoles	-	-	-	-	14	-		Obtained using formulated product.	Hashimoto and Nishizuchi, 1981	

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr			
Picamba (H)	- <u>Adelotus brevis</u>	1 to 2 week old tadpoles	A	21-22	Sanders 1970	-	220	202	185			Johnson, 1976
	- <u>Limnodynastes peroni</u>	1 to 2 week old tadpoles	"	"	"	"	205	166	106			
1,4-Dichlorobenzene	- <u>Xenopus laevis</u>	< 2 days	semi-static	20 ± 1	-	-	-	-	-	100 day exposure No Effect Levels (the highest concentration at which no effect or lethality was observed.): Mortality - 1 Development - 0.32 Growth - 1		Slooff and Canton, 1983
1,3-Dichloropropane	- <u>Xenopus laevis</u>	3-4 week old larvae	A	20 ± 1	-	-	-	63	-		N = 10 per concentration.	de Zwart and Slooff, 1987
1,3-Dichloropropanol	- <u>Xenopus laevis</u>	3-4 week old larvae	A	20 ± 1	-	-	-	450	-		N = 10 per concentration.	de Zwart and Slooff, 1987

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr			
Dieldrin (DC)	- <u>Bufo woodhousei fowleri</u>	4-5 week old tadpoles	A	15.5 ± .5 (7.1)	-	30 ppe methyl orange	1.1 (1.4-2.2)	0.40 (0.09-1.2)	0.15 (0.02-0.47)		N = 10. 95% confidence limits in brackets.	Sanders, 1970
	- <u>Pseudacris triseriata</u>	one week old tadpoles	"	"	-	"	0.25 (0.083-0.41)	0.22 (0.080-0.40)	0.10 (0.03-0.28)			
Dimefox (OP)	- <u>Bufo viridis</u>	adults 18-45 g	-	25	Injection into dorsal lymphatic sac	-	-	-	-	7 day LD50-1,410.	N = 25 minimum.	Edey and Schatzberg-Parath, 1960
Dimethoate (OP)	- <u>Xenopus laevis</u>	< 2 days	semi-static	20 ± 1	-	-	-	-	-	100 day exposure No Effect Levels (the highest concentration at which no effect or lethality was observed.): Mortality - 1 Development - 32 Growth - 32		Slooff and Canton, 1983

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr			
Dioxinates (cont'd)	<i>Rana cymbophryctis</i>	adults	renewed daily	23 ± .2 (7.3-7.8)	-	60-70 mg/L hardness	-	-	-	Significant glycogen elevation in liver, muscle and kidney noted.	95% confidence limits in parentheses.	Mudgill and Patil, 1987
		male	-	-	-	51.4 (49.0-53.5)	43.3 (41.6-45.1)	39.0 (37.5-40.5)				
		female	-	-	-	-	46.3 (45.0-47.1)	36.0 (35.8-36.2)				
1,2-Dioxethyl-benzene	<i>Xenopus laevis</i>	3-4 week old larvae	A	20 ± 1	-	-	-	73	-	N = 10 per concentration.	de Zwart and Sloof 1987	
Dioxethylhydrazine (unsymmetrical, UDMH) (propellant)	<i>Ambystoma maculatum</i> , <i>A. opacum</i>	larvae	A	20.5-23.5 (7.8-8.2)	APHA et al. 1971	185-232 mg/L as CaCO <sub>3</sub>	>100	55	26			Siano, 1986
			-	20.5-23.5 (6.5-6.9)	-	16-18 mg/L as CaCO <sub>3</sub>	>135	>135	108			

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr			
1,1-Dimethyl-hydrazine	<i>Xenopus laevis</i>	embryos	A	-	-	-	-	-	-	E050 (teratogenesis) for exposure from blastula to hatching: 7-9.		Greenhouse, 1977
2,6-Dimethyl-quinoline	<i>Xenopus laevis</i>	early cleavage to mid-blastula embryos	A	20	-	-	-	-	6.5 (5.6-7.5)		N = 25. 95% confidence limits in brackets. Reduced motility, development retarded, pigmentation lighter.	Dumont et al., 1979
Dinitro-o-cresol	<i>Xenopus laevis</i>	<2 days	Semi-static	20 ± 1	-	-	-	-	-	100 day exposure No Effect Levels (the highest concentration at which no effect or lethality was observed): Mortality - 0.32 Development - 0.32 Growth - 0.32	N = 75.	Slooff and Canton, 1983
Dinoseb (H)	<i>Bufo bufo japonicus</i>	tadpoles	-	-	-	-	-	0.55	-			Hashiwato and Nishizuchi, 1981

Table 1 - Acute Toxicity (cont'd)

Contaminant*	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr			
2,4-Dioxo-1,8-naphthol	<u>Xenopus laevis</u>	3-4 week old larvae	A	20 ± 1	-	-	-	3047	-		N = 10 per concentration.	de Zwart and Stooff, 1987
2-OPA (H)	<u>Adelotus brevis</u>	1-2 week old tadpoles	A	21-22	Sandera 1970	-	11.1 g/L	5.2 g/L	4.2 g/L			Johnson, 1976
	<u>Limnodynastes peroni</u>	1-2 week old tadpoles	"	"	"	"	3.3 g/L	2.5 g/L	2.0 g/L			
TC-1339 (avian control chemical)	<u>Rana sphenocephala</u>	larvae	A	16	ASTM Committee E-35 on Pesticides 1980	hardness 24 mg/L as CaCO <sub>3</sub>	63 (60-66)	-	44 (42-47)		95% confidence intervals in brackets.	Marking and Chandler, 1981
TC-1347 (avian control chemical)	<u>Rana sphenocephala</u>	larvae	A	16	ASTM Committee E-35 on Pesticides 1980	hardness 24 mg/L as CaCO <sub>3</sub>	41 (40-42)	-	32 (30-34)		95% confidence intervals in brackets.	Marking and Chandler, 1981

Table 1 - Acute Toxicity (cont'd)

Contaminant*	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr			
TC-2698 (avian control chemical)	<u>Rana sphenocephala</u>	larvae	A	16	ASTM Committee E-35 on Pesticides 1980	hardness 24 mg/L as CaCO <sub>3</sub>	-	-	> 30			Marking and Chandler, 1981
SMA (H) disodium methyl arsonate)	<u>Adelotus brevis</u>	1-2 week old tadpoles	A	21-22	-	-	600	525	453			Johnson, 1976
	<u>Limnodynastes peroni</u>	1-2 week old tadpoles	"	"	"	"	324	310	271			
Diosulfan (DC)	<u>Rana tigrina</u>	tadpoles	A	20 ± 2 (6.9-7.2)	-	95-100 mg/L	0.0021 (0.0018-0.0025)	0.0020 (0.0017-0.0025)	0.0018 (0.0014-0.0022)	96 hr presumable harmless conc. - .00055.	95% confidence limits in brackets.	Gopal et al., 1981
	<u>Bufo melanostictus</u>	tadpoles	A	Approx. 25 (8.3)	APHA 1980	220 mg/L total hardness	0.1419 (.1659-.1213)	0.1344 (.1370-.1310)	0.1230 (.1278-.1184)	Incipient Lethal Level = 0.105.	95% fiducial limits in parentheses.	Vardie et al., 1984
	<u>Bufo bufo japonicus</u>	tadpoles	-	-	-	-	-	9.0	-		Obtained using formulated product.	Hoshimoto and Nishizumi, 1981

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr			
Endothall (H) (canso (N,N-dimethylolkylicaine) salt)	- <u>Bufo woodhousii</u>	4-5 week old tadpoles	A	15.5 ± .5 (7.1)	-	30 ppm methyl orange	3.2 (1.7-5.5)	1.8 (0.93-3.2)	1.2 (0.40-3.4)		N = 10. 95% confidence limits in brackets.	Sanders, 1970
Endrin (GC)	- <u>Rana sphenoccephala</u>	eggs	B	20 (7.2-7.5)	-	total hardness 100 ppm as CaCO <sub>3</sub>	0.025	-	-	Exposed for 24 hr, observed at 96 hr.	N = 20-30 per level.	Hall and Swineford, 1980
		sub-adulto	"	"	"	"	-	-	0.005		N = 5-10 per level.	
		larvae	"	"	"	"	-	-	0.006	Exposed for 96 hr, observed at 192 hr.	N = 20-30 per level.	
	- <u>Bufo bufo japonicus</u>	tadpoles	-	-	-	-	-	12	-		Obtained using formulated product.	Hashimoto and Nishizuchi, 1981
	- <u>Rana sphenoccephala</u>	larvae	B	20 (7.2-7.5)	Hall and Swineford 1980	hardness 100 ppm as CaCO <sub>3</sub>	-	-	0.009 (0.006-0.014)	24 hr EC50 (conc. producing behavioral aberrations in 50% of animals): 0.013 (0.009-0.020)	Animals were exposed for 96 hr, mortality was determined after 192 hr. 95% confidence limits in brackets.	Hall and Swineford, 1981

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr			
Endrin (cont'd)	<u>R. catesbeiana</u>	larvae	"	"	"	"	-	-	0.002 (0.001-0.005)	>0.040	Some differences between results using static or flow through test methods noted.	
	<u>R. sylvatica</u>	larvae	"	"	"	"	-	-	0.034 (0.025-0.055)	<0.016	Removal of pesticide from water by test animals was significant.	
	<u>Bufo americanus</u>	larvae	"	"	"	"	-	-	0.010 (0.005-0.016)	0.008 (0.006-0.010)		
	<u>Acris crepitans</u>	larvae	"	"	"	"	-	-	0.010 (0.008-0.011)	0.023 (0.016-0.031)		
	<u>Ambystoma opacum</u>	larvae	"	"	"	"	-	-	0.018 (0.0-0.031)	0.018 (0.014-0.021)		
	<u>Ambystoma maculatum</u>	larvae	"	"	"	"	-	-	0.056 (0.039-0.089)	0.048 (0.043-0.053)		



Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>				Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	72 hr	96 hr			
Endrin (cont'd)	- <u>Bufo woodhousei</u>	4-5 week old tadpoles	A	15.5 ± .5 (7.1)	-	50 ppm methyl orange	0.57 (0.33-1.3)	0.46 (0.31-0.68)	-	0.12 (0.30-0.66)		N = 10. 95% confidence limits in brackets.	Sanders, 1970
	- <u>Pseudacris triseriata</u>	1 week old tadpoles	-	"	"	"	0.29 (0.18-0.45)	0.29 (0.18-0.45)	-	0.18 (0.09-0.50)			
	- <u>Rana catesbeiana</u>	tadpoles 2-3 g	B	17.4-21.2 (8.00-8.04)	APHA 1980; USEPA 1974					0.0025 (.0021-.0029)		Range in parentheses.	Thurston et al., 1985

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	72 hr			
Endrin (cont'd)	- <u>Rana temporaria</u>	tadpoles: with external gills (2 days)	A	20 (6.0)	-	2.21-2.39 ml 1N-HCl/L	2.0086 (1.2104-3.1947)	0.7098 (0.3982-1.2555)	0.4255 (0.2995-0.6061)		tadpoles lay paralyzed on the bottom of aquaria.	Mohiqueuth, 1977
		with internal gills, no limbs (8 days)					0.9886 (0.5898-1.7181)	0.2268 (0.1504-0.3932)	0.505 (0.0207-0.0903)		fully mobile. Initial period of agitation, thereafter a loss of balance and paralysis, lying on bottom of aquaria.	
		with internal gills, no limbs (16 days)					1.1375 (0.7217-1.8508)	0.6149 (0.3635-1.1796)	0.2875 (0.1537-0.4682)			
		with internal gills, (45 days)					0.2417 (0.1165-0.4691)	0.0740 (0.0300-0.1480)	0.0243 (0.0151-0.0351)			
		with pelvic limbs (86 days)					0.1605 (0.0705-0.3187)	0.0396 (0.0182-0.0774)	0.0172 (0.007-0.0257)			
		with pelvic and thoracic limbs (107 days)					0.1133 (0.0440-0.2206)	0.0289 (0.0150-0.0453)	0.0147 (0.0047-0.0220)			

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr			
Eptam (H)	- Unspecified	tadpoles 1-1.5cm	re-nosed daily	17-20 (7.7-8.2)	-	-	-	16.8	-	Value is LC <sub>50</sub> of the active ingredient.	Perevozchenko, 1975	
1,2-Ethanediol	- <u>Xenopus laevis</u>	3-4 week old larvae	A	20 ± 1	-	-	-	326	-		N = 10 per concentration.	de Zwart and Slooff, 1987
Ethyl Acetate	- <u>Ambystoma macrodactylum</u>	3-4 weeks post hatch	A	20 ± 1	-	-	-	150	-			N = 10 for each species. Organisms not fed during expt.
	- <u>Xenopus laevis</u>	3-4 weeks post hatch	"	"	"	"	-	160	-			
Ethylene Dichloro	- <u>Xenopus laevis</u>	tadpoles 5-12 days old	A	22 ± 1	-	-	10 day			Treatment was continuous for 10 days; LC50 was determined at 10 days post exposure.	Birch and Prahalad, 1986	
							250					

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr			
Ethylene Thiourea	- <u>Xenopus laevis</u>	tadpoles 5-12 days old	A	22 ± 1	-	-	10 day			Treatment was continuous for 10 days; LC50 was determined at 10 days post exposure.	Birch and Prahalad, 1986	
							100					
Ethyl Propionate	- <u>Ambystoma macrodactylum</u>	3-4 weeks post hatch	A	20 ± 1	-	-	-	56	-	N = 10 for each species. Organisms not fed during expt.	Slooff and Baerends, 1980	
	- <u>Xenopus laevis</u>	3-4 weeks post hatch	"	"	"	"	-	56	-			
Fenitrothion (OP)	- <u>Rana catesbeiana</u>	tadpoles 22.4 mm	A	housed at 21 ± 1	-	-	9.9 (8.9-7)	7.8 (7.1-8.5)	4.9 (4.2-5.3)	95% fiducial limits in brackets.	Lyons et al., 1976	
	- <u>Bufo bufo japonicus</u>	tadpoles	-	-	-	-	-	9.0	-			Obtained using formulated product.

Table 1 - Acute toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr			
Fenoprop (H)	- <u>Adeletus brevis</u>	1-2 week old tadpoles	A	21-22	Sanders 1970	-	77	60	54			Johnson, 1976
	- <u>Limnodynastes peroni</u>	1-2 week old tadpoles	"	"	"	"	55	27	22			
	- <u>Bufo marinus</u>	2 week old tadpoles	"	"	"	"	60	42	34			
S - Fenpropathrin (PY)	- <u>Rana pipiens</u>	adults 20-30 g	-	20	subcutaneous injection into the dorsum	-	-	-	-	24 hr LD50: 0.27 (0.12-0.63)	N = 28-34. 95% confidence limits in parentheses.	Cole and Casida, 1983
Fenthion (OP)	- <u>Bufo bufo</u>	26-37 day old tadpoles	A	20 ± 1	-	-	2.1-2.6	2.0-2.2	1.8-2.2	95% confidence intervals.	Development slowed.	Marchal-Séguin, 1976
	- <u>Rana pipiens</u> <u>Bufo boreas</u>	1 day old larvae	-	-	-	-	-	-	5.0			Lewis et al., 1985

Table 1 - Acute toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr			
Fentin-acetate (F)	- <u>Bufo bufo japonicus</u>	tadpoles	-	-	-	-	-	0.33	-			Hashimoto and Nishiuchi, 1981
SS - Fenvalerate (PY)	- <u>Rana pipiens</u>	adults 20-30 g	-	20	subcutaneous injection into the dorsum	-	-	-	-	24 hr LD50: 0.13 (0.09-0.19)	N = 28-34. 95% confidence limits in parentheses.	Cole and Casida, 1983
Forbam (F)	- <u>Bufo bufo japonicus</u>	tadpoles	-	-	-	-	-	4.2	-		Obtained using formulated product.	Hashimoto and Nishiuchi, 1981
Fluorometamide (rodenticide)	- <u>Bufo bufo japonicus</u>	tadpoles	-	-	-	-	-	>40	-			Hashimoto and Nishiuchi, 1981
Fluoranthene	- <u>Rana pipiens</u>	embryo stage 25	A	-	-	-	-	1 hr 0.09	-		LC50 determined for 1 hour of exposure to sunlight.	Kagan et al., 1985

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr			
Fossil Fuel	- <u>Xenopus laevis</u>	mid-late blastula embryo	A	-	-	-				EC50 (conc. that induced terato in 50% of survivors):		Dumont et al., 1983
Coal-derived fuel oil blend							-	-	1.48%	0.96%		
Shale-derived crude							-	-	6.97%	3.36%		
Coal-gasifier electrostatic-precipitator tar							-	-	0.83%	0.48%		
Aromatic petroleum crude							-	-	33.38%	31.10%	Little effect on growth noted; pigmentation and motility reduced by some of the materials.	

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr			
Fossil Fuels (Coal-gasification Electrostatic Precipitator Tar Fractions):	- <u>Xenopus laevis</u>		A	Room temp.	-	-				EC50 (conc. which causes malformations in 50% of survivors); ECO (no effect conc.):	95% confidence intervals in parentheses.	Schultz et al., 1983
Raw tar		blastula embryo					-	-	3.13- (2.32-4.23)	96 hr EC50=0.70 (0.51-1.57); ECO=0.50.		
		newly hatched froglets					-	-	163			
Ether Soluble Acid Fraction		blastula embryo					-	-	5.26 (4.22-6.55)	96 hr EC50=2.48 (1.49-4.11); ECO=1.00.		
		froglets					-	-	425			
Ether Soluble Base Fraction		blastula embryo					-	-	5.34 (1.67-17.07)	96 hr EC50=1.02 (0.62-1.67); ECO=0.50		
		froglets					-	-	299			

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr			
Gasol Fuels (ter reactions) (cont'd)			A	Room temp.	-	-						Schultz et al., 1983 (cont'd)
kro/di aromatic Fraction		blastula embryo					-	-	17.91 (13.20-59.00)	96 hr EC50-21.47 (15.79-29.20); EC0 < 10.00.		
		froglets					-	-	776			
Poly Aromatic Fraction		blastula embryo					-	-	6.76 (5.21-8.77)	96 hr EC50-2.55 (1.48-4.38); EC0 < 1.00.	Malformations in head and gut, edema and growth retardation observed.	
		froglets					-	-	529			
Furazone (fish bactericide)	<i>Rana pipiens</i>	larvae	A	16	Committee on Methods for Toxicity Tests with Aquatic Organisms 1975	total hardness 21 mg/L as CaCO <sub>3</sub>	6.90 (5.55-8.57)	-	0.770 (0.590-1.01)		95% confidence limits in brackets. Larvae were more sensitive than eggs.	Harking et al., 1977

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References		
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr					
Germanium (M)	<i>Gastrophryne carolinensis</i>	embryo-larvel	B	- (7.0-7.8)	Birge & Black 1977a,b	hardness 195.5 ± 5.4 ppm as CaCO <sub>3</sub>			0.05		Exposure from fertilization to 4 days post hatch.	N = 10.	Birge et al., 1979a	
Heptachlor (OC)	<i>Bufo woodhousii fowleri</i>	4-5 week old tadpoles	A	15.5 ± .5 (7.1)	-	30 ppm methyl orange			0.85 (0.19-3.8)	0.76 (0.23-2.7)	0.66 (0.30-0.65)		N = 10. 95% confidence limits in brackets.	Sanders, 1970
n-Heptanal	<i>Ambystoma mexicanum</i>	3-4 weeks post hatch	A	20 ± 1	-	-				52			N = 10 for each species.	Slooff and Baerelman, 1980
	<i>Xenopus laevis</i>	3-4 weeks post hatch	"	"	"	"				66			Organisms not fed during expt.	

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>b</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>d</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr			
Hydrozine (propellant)	- <u>Ambystoma maculatum</u> , <u>A. opacum</u>	larvae	A	22.2-24.0 (7.8-8.2)	APHA et al. 1971	185-232 mg/L as CaCO <sub>3</sub>	>10	8.0	5.3			Slonim, 1986
			*	22.2-24.0 (6.3-6.9)	-	16-18 mg/L as CaCO <sub>3</sub>	>10	5.2	2.3			
	- <u>Xenopus laevis</u>	embryo	A	-	-	-	-	-	ED50 (teratogenesis) for exposure from blastula to hatching - 11.48-12.5.			Greenhouse, 1977
Bis(2-Hydroxyethyl) amine	- <u>Xenopus laevis</u>	3-4 week old larvae	A	20 ± 1	-	-	-	1174	-	N = 10 per concentration.		de Zwart and Slooff, 1987
Bis(2-Hydroxyethyl) ether	- <u>Xenopus laevis</u>	3-4 week old larvae	A	20 ± 1	-	-	-	1065	-	N = 10 per concentration.		de Zwart and Slooff, 1987

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>b</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>d</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr			
Bis(2-Hydroxyethyl) ether	- <u>Xenopus laevis</u>	3-4 week old larvae	A	20 ± 1	-	-	-	3181	-	N = 10 per concentration.		de Zwart and Slooff, 1987
Bis(2-Hydroxypropyl) amine	- <u>Xenopus laevis</u>	3-4 week old larvae	A	20 ± 1	-	-	-	410	-	N = 10 per concentration.		de Zwart and Slooff, 1987
[BP(S-benzyl diisopropyl) phosphorothioate	- <u>Bufo bufo japonicus</u>	tadpoles	-	-	-	-	-	10	-	Obtained using formulated product.		Hashimoto and Nishiuchi, 1981
[Iron Methanoarsenate (H)	- <u>Bufo bufo japonicus</u>	tadpoles	-	-	-	-	-	>40	-			Hashimoto and Nishiuchi, 1981
cis - Kadethrin (PT)	- <u>Rene pipiens</u>	adulte 20-30 g	-	20	subcutaneous injection into the dorsum	-	-	-	-	24 hr LD50: 1.2 (0.65-2.4)	N = 28-34. 95% confidence limits in parentheses.	Cole and Casida, 1983
Kasugamycin (F)	- <u>Bufo bufo japonicus</u>	tadpoles	-	-	-	-	-	6.4	-	Obtained using formulated product.		Hashimoto and Nishiuchi, 1981

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity		7 day				
Lead (H)	<i>Gestrophryne carolinensis</i>	embryo-larval	8	-	Birge & Black 1977 a, b	195.0 ± 5.4 ppm hardness as CaCO <sub>3</sub>		0.04		Exposure from fertilization to 4 days posthatch.	N = 14.	Birge et al., 1979a
							24 hr	48 hr	96 hr			
Linear Alkylbenzene Sulfonate	<i>Xenopus laevis</i>	3-4 weeks	semi-static	21 ± 2	-	-	-	-	56-100	test solutions corrected for pH and oxygen content.	Canton and Slooff, 1982b	
Lindane (OC)	<i>Bufo woodhousei fowleri</i>	4-5 weeks old tadpoles	A	15.5 ± .5 (7.1)	-	30 ppm methyl orange	14 (5.0-41)	5.4 (3.1-9.6)	4.4 (1.8-5.6)	N = 10. 95% confidence limits in brackets.	Sanders, 1970	
	<i>Pseudacris triseriata</i>	1 week old tadpoles	"	"	"	"	4.0 (2.7-6.1)	3.8 (2.50-5.7)	2.7 (1.4-4.3)			
Malachite Green (F)	<i>Notophtalmus viridescens</i>	adults	8	16 (7.5)	Committee on Methods for Toxicity Tests with Aquatic Organisms 1975	total hardness 20 mg/L as CaCO <sub>3</sub>	3.90 (3.47-4.38)	-	1.03 (0.672-1.98)	95% confidence limits in brackets.	Bills et al., 1977	

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr			
Malachite Green (cont'd)	<i>Rana pipiens</i>	larvae	8	16 (7.5)	Committee on Methods for Toxicity Tests with Aquatic Organisms 1975	total hardness 20 mg/L CaCO <sub>3</sub>	0.380 (0.351-0.412)	-	0.173 (0.149-0.200)		Bills et al., 1977 (cont'd)	
	<i>Bufo</i> sp.	larvae	"	"	"	"	0.355 (0.235-0.276)	-	0.0680 (0.0530-0.0860)			
Malathion (OP)	<i>Bufo woodhousei fowleri</i>	4-5 week old tadpoles	A	15.5 ± .5 (7.1)	-	30 ppm methyl orange	1.9 (1.4-3.5)	0.50 (0.25-1.3)	0.42 (0.09-0.98)	N = 10. 95% confidence limits in brackets.	Sanders, 1970	
	<i>Pseudacris triseriata</i>	1 week old tadpoles	"	"	"	"	0.56 (0.28-0.94)	0.32 (0.18-0.68)	0.20 (0.09-0.27)			
	<i>Rana pipiens</i>	adults approx. 9 cm	renewed daily	24 (5.3-6.5)	-	-	-	150				

Table 1 - Acute toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr			
Manganese (F) (Manganese ethylenediamine dithiocarbamate)	- <u><i>Triturus cristatus</i></u>	adults median length - 7.4 cm (males), 7.8 cm (females)	renewed daily	18 ± 2 (7.4)	-	hardness 300 mg/L as CaCO <sub>3</sub>	-	-	-	LT50 (lethal time for 50% of individuals) hr:  Conc.    Male    Female 125    8.4    28.5 (7.6-    (16.4- 9.2)    49.6 100    28.0    19.5 (16.0-    (9.5- 49.0)    40.2) 75    19.0    25.5 (7.0-    (11.6- 51.3)    55.8) 50    76.0    168.0 (57.6-    (85.7- 100.3)    329.3) 25    255.0 (147.4- 441.1)	N = 12 (6 males, 6 females) for each concentration. 95% confidence limits in parentheses. Females appeared less susceptible than males possibly due to larger body size.	Zaffaroni et al., 1978

Table 1 - Acute toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References	
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity		7 day					
Manganese (H)	- <u><i>Gastrophryne carolinensis</i></u>  - <u><i>Microhylis ornata</i></u>	embryo-larval	8	- (7.0- 7.8)	Birge & Black 1977 a, b	195.0 ± 5.4 ppm hardness as CaCO <sub>3</sub>		1.42		Exposure from ferti- lization to 4 days posthatch.	N = 14.	Birge et al., 1979a	
		tadpoles: 1 week old	renewed daily	25.5- 26.0 (6.96- 6.94)			97.0- 98.0 ppm	24 hr	48 hr				96 hr
								16.62 (16.2- 17.0)	16.03 (15.6- 16.4)				14.84 (14.6- 15.4)
4 weeks old				17.56 (17.0- 17.8)	16.52 (16.0- 16.8)	14.33 (14.0- 14.8)							
MCPA (H) Sodium salt (2-methyl- 4-chlorophenoxy acetic acid)	- <u><i>Triturus cristatus</i></u> <u><i>carolinensis</i></u>	adults	semi- static	18 ± 2	-	-	-	-	LT50 (lethal time for 50% of individuals) hr:  Conc.    Male    Female 3200    17    21 (16.0-    (19.4- 18.0)    22.7) 1600    35    45.5 (31.0    (41.0- 40.9)    50.0)	95% confidence limits in parentheses. Animals not fed during test.	Zaffaroni et al., 1986b		



Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity		7 day				
Mercury	<u>Gastrophryne carolinensis</u>	embryo-larval	B	- (7.0-7.8)	Birge & Black 1977 a, b	195.0 ± 5.4 ppm hardness as CaCO <sub>3</sub>		0.001		Exposure from fertilization to 4 days posthatch.	N = 14. Toad is more sensitive than goldfish and rainbow trout.	Birge et al., 1979a
	- <u>Bufo melanostictus</u>	tadpoles 1.8-2.2 cm	A	29-34 (7.1-7.6)	APHA et al. 1976	120-160 mg/L as CaCO <sub>3</sub>	.0528 (.0436-.0615)	.0456 (.0409-.0567)	.0436 (.0368-.0585)	In high concentrations surfacing, erratic body movement, loss of equilibrium noted.	95% confidence limits in brackets. Animals not fed 24 hr prior to or during test.	Khanjarat and Ray, 1987
	- <u>Microhyla ornata</u>	tadpoles: 1 week old  4 weeks old	renewed daily	25.5-26.0 (6.86-6.94)	-	97.0-98.0 ppm	2.04 (1.8-2.2)	1.68 (1.4-1.8)	1.12 (0.9-1.3)		Range in brackets. Animals not fed during test.	Rao and Madhyestha, 1987
	- <u>Bufo regularis</u>	adult females 25-30 g	-	-	-	-	-	-	-	96 hr LD50 following single i.m. injection of Hg <sup>2+</sup> - 5.60		Hiley et al., 1986b

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity		7 day				
Mercury (cont'd)	<u>Gastrophryne carolinensis</u>	embryo-larval	renewed every 12 hr	19 - 22 (7.0-7.8)	-	hardness 90-105 ppm CaCO <sub>3</sub>		1.3 (0.9-1.9)		Exposure from fertilization to 4 days post-hatch.	95% confidence limits in brackets.	Birge et al., 1979b
	<u>Hyla chrysoeclis</u>	"	"	"	"	"		2.4 (1.5-3.4)				
	<u>H. squirella</u>	"	"	"	"	"		2.4 (1.5-3.8)				
	<u>H. gratiosa</u>	"	"	"	"	"		2.5 (1.7-3.4)				
	<u>H. varicolor</u>	"	"	"	"	"		2.6 (1.2-4.2)				
	<u>H. crucifer</u>	"	"	"	"	"		2.8 (1.9-3.9)				
	<u>Rana pipiens</u>	"	"	"	"	"		7.3 (4.8-10.0)				

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References	
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity		7 day					
Mercury (cont'd)	<u>Acris crepitans blanchardi</u>	embryo-larval	renewed every 12 hr	19-22 (7.0-7.8)	-	hardness 90-105 ppm CaCO <sub>3</sub>		10.4 (8.5-12.6)		Exposure from fertilization to 4 days post-hatch.	95% confidence limits in brackets.	Birge et al., 1979b (cont'd)	
	<u>Bufo punctatus</u>	"	"	"	"	"		36.8 (18.3-51.1)					
	<u>B. d. debilis</u>	"	"	"	"	"	"		40.0 (25.6-52.2)				
	<u>R. hakochoxi</u>	"	"	"	"	"	"		59.9 (55.8-65.9)				
	<u>B. foxiari</u>	"	"	"	"	"	"		65.9 (44.0-84.0)				
	<u>R. gryllis</u>	"	"	"	"	"	"		67.2 (54.3-79.5)				
	<u>Ambystoma opacum</u>	"	"	"	"	"		107.5 (72.5-151.5)					

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr			
Mercury (Mercuric chloride)	<u>Ambystoma mexicanum</u>	3-4 weeks post hatch	A	20 ± 1	-	-	-	0.4	-	-	N = 10 for each species. Organisms not fed during expt.	Slooff and Baerselman, 1987
	<u>Xenopus laevis</u>	3-4 weeks post hatch	"	"	"	"	-	0.1	-			
	<u>Microhyla ornata</u>	embryos in gastrulation	A	21-25 (7.1)	-	51 mg/L as CaCO <sub>3</sub>	-	-	0.1704		N = 50 embryos per concentration.	Chate and Mithner, 1987
		tadpoles 8-10 days post-hatch	"	"	"	"	-	-	0.1184			
	<u>Xenopus laevis</u>	3-4 week old larvae	A	20 ± 1	-	-	-	0.1	-	N = 10 per concentration.	de Zwart and Slooff, 1987	
Methiocarb (avian repellent)	<u>Rana sphenoccephala</u>	larvae	A	16	ASTM Committee E-35 on Pesticides 1980	hardness 24 mg/L as CaCO <sub>3</sub>	8.5 (7.9-9.2)	-	8.7 (7.8-9.6)	95% confidence intervals in brackets.	Marking and Chandler, 1981	
Methoxy	<u>Bufo bufo japonicus</u>	tadpoles	-	-	-	-	-	>40	-		Hoshimoto and Mitsuuchi, 1981	

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr			
Methoxychlor (OC)	- <u>Bufo woodhouseii fowleri</u>	4-5 week old tadpoles	A	15.5 ± .5 (7.1)	-	50 ppm methyl orange	0.76 (0.52-1.1)	0.11 (0.02-0.60)	-	Significant variations observed in muscle, liver and kidney glycogen levels.	N = 10. 95% confidence limits in brackets.	Sanders, 1970
	- <u>Pseudacris triseriata</u>	1 week old tadpoles	"	"	"	"	0.44 (0.30-0.65)	0.42 (0.29-0.62)	0.33 (0.19-0.57)			
1-Methyl-4 (tert)butylbenzene	- <u>Xenopus laevis</u>	3-4 week old larvae	A	20 ± 1	-	-	-	5.0	-		N = 10 per concentration.	de Zwart and Slooff, 1987
Methyl Parathion	- <u>Rana cyanophlyctis</u>	adults	renewed daily	23 ± .2 (7.3-7.8)	-	60-70 mg/L hardness	-	-	-		95% confidence intervals in parentheses.	Mirkall and Patil, 1987
		male					30.0 (21.4-42.0)	17.7 (16.65-18.8)	8.0 (7.7-8.3)			
		female					37.0 (30.2-45.4)	24.0 (22.0-26.2)	11.5 (11.2-12.2)			

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr			
4-Methyl-2-pentanol	- <u>Xenopus laevis</u>	3-4 week old larvae	A	20 ± 1	-	-	-	656	-		N = 10 per concentration.	de Zwart and Slooff, 1987
2-Methyl-1-propanol	- <u>Xenopus laevis</u>	3-4 week old larvae	A	20 ± 1	-	-	-	18.3	-		N = 10 per concentration.	de Zwart and Slooff, 1987
2-Methyl-2-propanol	- <u>Xenopus laevis</u>	3-4 week old larvae	A	20 ± 1	-	-	-	2450	-		N = 10 per concentration.	de Zwart and Slooff, 1987
2-Methyl-quinoline	- <u>Xenopus laevis</u>	early cleavage to mid-blastula embryos	A	20	-	-	-	-	26.4 (22.4-31.1)		95% confidence limits in brackets. N = 25. Development retarded, reduction in motility.	Dumont et al., 1979
Hevinphos (OP)	- <u>Bufo arenarum</u>	adults	-	-	injected via lymphatic sac	-	-	-	-	96 hr LD50: 850		Juarez and Guzman, 1984a
Hexacarbate (C)	- <u>Rana catesbeiana</u>	adults male	-	-	single dose - insertion of gelatin capsule to the level of the proventriculus	-	-	-	-	Estimated 14 day LD50 = 283-800.	Animals fasted for 2-24 hr prior to test. N = 14 males. Bullfrog more resistant than birds and mammals.	Fucker and Crabtree, 1969

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>				Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	72 hr	96 hr			
Mine Drainage (acidic)	- <u>Leurognathus nigricaratus</u>	larvae	B	12 (3.9)	-	-	53% leachate	47% leachate	33% leachate	-	LF50 ± SE (time of exposure causing 50% mortality): 33% leachate - 41 ± .93 hr; 45% leachate - 35 ± .66 hr; 63% leachate - 21 ± 1.82 hr; 100% leachate - 19 ± 2.15 hr.  96 hr EC50 confirmation:  13.1-13.68  27.7-34.38	Mortality rate was a function of exposure time and concentration.	Mathews and Morgan, 1982; Mathews et al., 1976
	- <u>Xenopus laevis</u>	embryos	A	- (5.4)	modified Ducrest et al. 1983	-	-	-	13.1-15.88	13.1-13.68			
				- (5.7-5.9)			-	-	27.7-34.38	30.3-53.38		Fe: 414.0-533.0; Zn: 47.2-73.0; Pb: < 0.005-0.008; Cd: < 0.005-0.004; Cu: < 0.04-0.04	

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr			
Molinate (M)	- <u>Bufo woodhousii fowleri</u>	4-5 week old tadpoles	A	15.5 ± .5 (7.1)	-	30 ppm methyl orange	33 (18-60)	28 (12-55)	14 (4.2-36)		N = 10. 95% confidence limits in brackets.	Sanders, 1970
Nabam (F)	- <u>Xenopus laevis</u>	tadpoles 5-12 days old	A	22 ± 1	-	-		2		Treatment was continuous for 10 days; LC50 was determined at 10 days post exposure.	Birch and Prabhakar, 1986	
							24 hr	48 hr	96 hr			
							2.2 (0.80-4.0)	2.0 (0.90-5.0)	1.7 (0.50-3.2)		N = 10. 95% confidence limits in brackets.	Sanders, 1970
Naled (OP)	- <u>Pseudacris triseriata</u>	1 week old tadpoles	A	15.5 ± .5 (7.1)	-	30 ppm methyl orange						
Naphthalene	- <u>Xenopus laevis</u>	larvae 3 weeks old	B	28 (7.0-7.1)	-	-	-	-	2.1 (1.3-3.8)	6 hour EC50 for loss of pigment - 3.7 (3.1-4.5).	95% confidence intervals in brackets.	Edvisten and Bantle, 1982
									2.1 (1.4-2.9)	6 hour EC50 for absence of swimming - 2.3 (0.8-5.3), 1.7 (1.2-2.3).		

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	7 day					
Nickel (M)	- <u>Gastrophysa carolinensis</u>	embryo-larval	B	- (7.0-7.8)	Birge & Black 1977 a, b	195.0 ± 5.4 ppm hardness as CaCO <sub>3</sub>		0.05		Exposure from fertilization to 4 days poethatch.	N = 14.	Birge et al., 1979a
							24 hr	48 hr	96 hr			
	- <u>Bufo melanostictus</u>	tadpoles 1.8-2.2 cm	A	29-34 (7.1-7.6)	APHA et al. 1976	120-160 mg/L as CaCO <sub>3</sub>	53.21 (49.56-58.37)	34.3 (32.9-37.21)	25.32 (22.8-28.62)	Exposure greater than 10 ppm produced immediate stress, air gulping and mucus production.	95% confidence limits in brackets. Animals not fed 24 hr prior to or during tests.	Khargart and Ray, 1987
Nitrite	- <u>Ambystoma texanum</u>	larvae 0.45 ± 0.08 g	A	25 (7.0)	E.P.A. 1975	140 mg/L total hardness	-	-	1.09 (0.48-0.2)			
Nitrofen	- <u>Bufo bufo japonicus</u>	tadpoles	-	-	-	-	-	3.8	-	Obtained using formulated product.		Hashimoto and Nishiyuki, 1981
Nitrotriacetic Acid	- <u>Xenopus laevis</u>	3-4 weeks	semi-static	21 ± 2	-	-	-	-	560-1000	Test solutions corrected for pH and oxygen content.		Canton and Slooff, 1982b

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr			
p-Nitro-Toluene	- <u>Xenopus laevis</u>	42 days	semi-static	20 ± 1	-	-	-	-	-	100 day exposure no effect levels (the highest concentration at which no effect or lethality was observed): Mortality - 10 Development - 3.2 Growth - 32	N = 75.	Slooff and Canton, 1983
DNPA (OP)	- <u>Rana pipiens</u>	adults approx. 9 cm	renewed daily	24 (6.2 - 6.6)	-	-		2,900				

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr			
PA-14 (evich streamer agent)	<u>Rana sphenoccephala</u>	larva	A	12	ASIM Committee E-35 on Pesticides 1980	hardness 24 mg/L as CaCO <sub>3</sub>	7.5 (6.3-9.0)	-	5.9 (5.3-6.5)		95% confidence intervals in brackets.	Marking and Chandler, 1981
				16			4.5 (4.2-4.7)	-	4.2 (3.6-4.7)			
				22			2.6 (2.0-3.0)	-	2.3 (1.8-2.9)			
Paraxon (OP)	<u>Bufo viridis</u>	adults 18-45 g	-	16	injection into dorsal lymphatic sac	-	-	-	-	7 day LD50: 188	N = 25 animals.	Edery and Schotzberg-Porath, 1960
	<u>Rana ridibunda</u>						-	-	-			

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr			
Paraquat (H)	<u>Adelotus brevis</u>	1-2 week old tadpoles	A	21-22	Sanders 1970	-	320	315	262		N = 10. 95% confidence limits in brackets.	Johnson, 1976
	<u>Limnodynastes peroni</u>	1-2 week old tadpoles	"	"	"	"	204	153	100			
	<u>Bufo woodhousei fowleri</u>	4-5 week old tadpoles	A	15.5 ± .5 (7.1)	-	30 mg/L methyl orange	54 (30-100)	25 (9.4-64)	26 (11-43)			
	<u>Pseudacris triseriata</u>	1 week old tadpoles	"	"	"	"	43 (18-56)	17 (28-52)	28 (21-36)			
	<u>Bufo bufo japonicus</u>	tadpoles	-	-	-	-	-	14	-			

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr			
Parathion (OP)	- <u>Bufo viridis</u>	adults 18-45 g	-	16	injection into dorsal lymphatic sac	-	-	-	7 day LD50: 967	N = 25 minimum.	Edey and Schatzberg-Porath, 1960	
	- <u>Pseudacris triseriata</u>	1 week old tadpoles	A	15.5 ± .5 (7.1)	-	30 ppm methyl orange	1.6 (0.40-3.0)	1.4 (0.91-2.8)	1.0 (0.30-2.0)	N = 10. 95% confidence limits in brackets.	Sanders, 1970	
	- <u>Rana pipiens</u>	adults approx. 9 cm	renewed daily	24 (5.0-6.8)	-	-	-	15 day	10.	N = 24 per concentration.	Kaplan and Glaczinski, 1965	
	- <u>Bufo arenarum</u>	adults	-	-	injected via lymphatic sac	-	-	-	96 hr LD50: 3,352	-	Juarez and Guzman, 1984a	
	- <u>Bufo bufo japonicus</u>	tadpoles	-	-	-	-	-	7.2	-	-	Hashimoto and Nishiuchi, 1981	

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr			
Pentachlorophenol	- <u>Ambystoma mexicanum</u>	3-4 weeks post hatch	A	20 ± 1	-	-	-	0.30	-	N = 10 for each species. Organisms not fed during expt.	Slooff and Baerselman, 1980	
	- <u>Xenopus laevis</u>	3-4 weeks post hatch	"	"	"	"	-	0.26	-	-	-	
	- <u>Rana catesbeiana</u>	tadpoles 2-5 g	B	17.2-18.2 (8.01-8.04)	APHA 1980, USEPA 1974	-	-	-	0.207 (.185-.231)	Range in parentheses.	Hurlston et al., 1985	
	- <u>Xenopus laevis</u>	< 2 days	Semi-static	20 ± 1	-	-	-	-	-	100 day exposure no effect levels (the highest concentration at which no effect or lethality was observed): Mortality - 0.032 Development - 0.032 Growth - 0.032	N = 75.	Slooff and Canton, 1983
Pentachlorophenol Sodium	- <u>Bufo bufo japonicus</u>	tadpoles	-	-	-	-	-	0.25	-	Obtained from formulated product.	Hashimoto and Nishiuchi, 1981	

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>b</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>d</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr			
Paraothenin (PY)	- <u>Rana catesbeiana</u>	tadpoles 2-5 g	B	17.3 18.0 (7.98- 8.01)	APHA 1980, USEPA 1974	-	-	-	0.115 (.0558- .245)		Range in parentheses.	Thurston et al., 1985
	- <u>Rana catesbeiana</u>	tadpoles 6-8 cm 0.01 g	A	24 ± 1 (8.4)	Doudoroff et al. 1951	100 mg/L hardness	-	-	7.033 (4.129- 8.733)		N = 10 per concentration. 95% confidence limits in brackets. Frogs more tolerant than fish or crayfish.	Jolly et al., 1978
(1R-paraothenin)	- <u>Rana pipiens</u>	adults 20-30 g	-	20	subcutaneous injection into the dorsum	-	-	-	24 hr LD50: <u>trans</u> <u>cis</u> 7.5        0.14 (5.2-     (0.11- 10.9)    0.17)		N = 28-34. 95% confidence limits in parentheses.	Cole and Casida, 1983
Phenol	- <u>Xenopus laevis</u>	tadpoles	B	17.2 ± 5 (7.39 ± .22)	-	40.0-49.5 mg/L as CaCO <sub>3</sub>	-	-	> 51.1			Holcombe et al., 1987

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>b</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>d</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr			
1R-Phenothenin (PY)	- <u>Rana pipiens</u>	adults 20-30 g	-	20	subcutaneous injection into the dorsum	-	-	-	24 hr LD50: <u>trans</u> <u>cis</u> greater    6.0 than        (3.9 - 20         9.2)		N = 28-34. 95% confidence limits in parentheses.	Cole and Casida, 1983
Phenyl Mercury Acetate	- <u>Bufo bufo japonicus</u>	tadpoles	-	-	-	-	-	0.12			Obtained using formulated product.	Hanamoto and Nishiyuchi, 1981
N-Phenyl-ε-naphthylamine	- <u>Xenopus laevis</u>	larvae	A	-	-	-	-	2.1- 2.3				Greenhouse, 1977
		embryos	-	-	-	-	-	-	ED50 (teratogenesis) for exposure from blastula to hatching- 4.57-4.8.			
Phosdrin (OP)	- <u>Rana pipiens</u>	adults approx. 9 cm	daily renewal	24 (6.0- 6.4)	-	-	-	12			N = 24 per concentration.	Keplan and Glaczinski, 1965
Phosphamidon (OP)	- <u>Bufo arenarum</u>	adults	-	-	injected via lymphatic sac	-	-	-	96 hr LD50: 1,195			Juarez and Guzman, 1984a



Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>d</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr			
Picloram (H)	<u>Adelotus brevis</u>	1-2 week old tadpoles	A	21-22	Sanders 1970	-	143	123	95			Johnson, 1976
		4 week old tadpoles	"	"	"	"	210	182	154			
	<u>Limodynastes peroni</u>	1-2 week old tadpoles	"	"	"	"	120	116	105			
Piperonyl Butoxide (symargiet)	<u>Pseudacris triseriata</u>	1 week old tadpoles	A	15.5 ± .5 (7.1)	-	30 ppm methyl orange	1.8 (0.40-8.2)	1.3 (0.30-12)	1.0 (0.10-9.0)		N = 10. 95% confidence limits in brackets.	Sanders, 1970
Potassium Dichromate	<u>Xenopus laevis</u>	~2 days	semi-static	20 ± 1	-	-	-	-	-	100 day exposure No Effect Levels (the highest concentration at which no effect or lethality was observed): Mortality - 1 Development - 3.2 Growth - 3.2	N = 75.	Slooff and Canton, 1983

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>d</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr			
Propanil (H)	<u>Bufo bufo japonicus</u>	tadpoles	-	-	-	-	-	2.5	-			Hashimoto and Nishiuchi, 1981
n-Propanol	<u>Ambystoma mexicanum</u>	3-4 weeks post hatch	A	20 ± 1	-	-	-	4,000	-		N = 10 for each species. Organisms not fed during expt.	Slooff and Berven, 1980
	<u>Xenopus laevis</u>	3-4 weeks post hatch	"	"	"	"	-	4,000	-			
2-Propenyl Azine	<u>Xenopus laevis</u>	3-4 week old larvae	A	20 ± 1	-	-	-	12.4	-		N = 10 per concentration.	de Iwert and Slooff, 1987
Bis (2-Propenyl) Azine	<u>Xenopus laevis</u>	3-4 week old larvae	A	20 ± 1	-	-	-	25.5	-		N = 10 per concentration.	de Iwert and Slooff, 1987
Pyrene	<u>Rana pipiens</u>	embryos stage 25	A	-	-	-	-	1 hr	0.14		LC50 determined for 1 hour of exposure to sunlight.	Kagan et al., 1985
trans - Pyrethrins (PY)	<u>Rana pipiens</u>	adults 20-30 g	-	20	subcutaneous injection into the dorsum	-	-	-	-	24 hr LD50: 5.8 (4.9-7.0) N = 28-34.	95% confidence limits in parentheses.	Cole and Coside, 1983

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>				Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	72 hr	96 hr			
Pyridine	- <u>Ambystoma</u> <u>barbicarpus</u>	3-4 weeks post hatch	A	20 ± 1	-	-	-	950	-	Teratogenicity EC50 for mid-blastula embryos: 24 hr - 2190 (1340-3570); 48 hr - 1350 (600-4010); 72 hr - 1350 (440-4140); 96 hr - 1200 (730-1980).	N = 10 for each species. Organisms not during expt.	Slooff and Boersma, 1980	
	- <u>Xenopus</u> <u>laevis</u>	3-4 weeks post hatch	*	"	"	"	-	1,400	-				
	- <u>Xenopus</u> <u>laevis</u>	mid-blastula embryos	A	Room Temp.	-	98.5 µg/L	3800 (2520-5740)	2570 (1880-3520)	2340				-
	- <u>Xenopus</u> <u>laevis</u>	tailbud embryos					9550 (1710-35000)	3190 (3000-3830)	2820 (2350-3380)				2460 (1710-3540)
	- <u>Xenopus</u> <u>laevis</u>	free swimming larvae					1660 <sup>d</sup>	1590 (1190-2110)	1200 (980-1460)	1090 (900-1340)			

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>				Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	72 hr	96 hr			
Quinoline	- <u>Xenopus</u> <u>laevis</u>	mid-blastula embryo	A	Room Temp.	-	98.5 µg/L	219	115 (53-247)	87 (45-167)	79 (51-123)	Teratogenicity EC50 for mid-blastula embryos: 24 hr - 71 (38-154); 72 hr - 36 (15-90); 96 hr - 29 (14-61).	95% confidence limits in brackets.	Davis et al., 1981
	- <u>Xenopus</u> <u>laevis</u>	tailbud embryo					200 (157-245)	148 (117-186)	129 (102-164)	129 (98-169)			
	- <u>Xenopus</u> <u>laevis</u>	free swimming larvae					135 (91-200)	117 (78-176)	107 (90-127)	95 (75-120)			
	- <u>Xenopus</u> <u>laevis</u>	early cleavage to mid-blastula embryos	A	20	-	-	-	-	-	26.3 (22.1-31.3)			

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>d</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr			
IRS - Resmethrin (PY)	- <u>Rana catesbeiana</u>	tadpoles 10-15 g	-	20	intraperitoneal injection	-	-	-	24 hr LD50: Trans C18 5.6 1.2 (3.6-8.8) (0.81-1.6)	N = 32-45. 95% confidence limits in parentheses.	Cole and Caside, 1983	
	- <u>Rana pipiens</u>	adults 20-30 g	"	"	subcutaneous injection into the dorsum	"	-	-	24 hr LD50: Trans C18 greater than 60 (1.0-1.6)	N = 28-34. 95% confidence limits in parentheses.		
Rotenone	- <u>Bufo bufo japonicus</u>	tadpoles	-	-	-	-	0.33	-			Hashimoto and Nishiuchi, 1981	
Rotenone (Maxfish-5% rotenone)	- <u>Rana sphenoccephala</u>	larvae	A	16 ± 1 (6.6)	Committee on Methods for Toxicity Tests with Aquatic Organisms 1973	20 mg/L	0.380 (.498-.680)	-	0.500 (.423-.591)	95% confidence intervals in brackets.	Chandler and Marking, 1982	
Rotenone (Rotenoids)	- <u>Xenopus laevis</u>	tadpoles	B	17.2 ± 5 (7.39 ± .22)	-	40.4-49.5 mg/L as CaCO <sub>3</sub>	-	-	>0.040		Holcombe et al., 1987	

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>d</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr			
Salicylaldehyde	- <u>Ambystoma mexicanum</u>	3-4 weeks post hatch	A	20 ± 1	-	-	-	7.0	-	Exposure from fertilization to 4 days posthatch.	N = 10 for each species. Organisms not fed during expt.	Slooff and Baerelman, 1980
	- <u>Xenopus laevis</u>	3-4 weeks post hatch	"	"	"	"	-	7.7	-			
Selenium (M)	- <u>Gastrophryne carolinensis</u>	embryo-larval	B	- (7.0-7.8)	Birge & Black 1977 a, b	195.0 ± 5.4 ppm hardness as CaCO <sub>3</sub>	-	0.09	-	72 hr	N = 14.	Birge et al., 1979a
	- <u>Xenopus laevis</u>	tadpoles	renewed daily	23	-	-	-	8.04 (6.99-9.25)	-			
Selenium (sodium selenite)	- <u>Xenopus laevis</u>	tadpoles	renewed daily	23	-	-	-	8.04 (6.99-9.25)	-	LD50 (median survival time): 4.7 days in 2 ppm, 4.0 days in 5 ppm, 2.54 days in 10 ppm.	95% confidence limits in brackets. ≥ 2 ppm produced abnormalities.	Browne and Dumont, 1979
Silver (M)	- <u>Bufo melanostictum</u>	tadpoles 1.8-2.2 cm	A	29-34 (7.1-7.6)	APHA et al. 1976	120-160 mg/L as CaCO <sub>3</sub>	.0073 (.00637-.00809)	.0062 (.00504-.00708)	.0041 (.00364-.00461)	In high concentrations surfacing, erratic body movement, loss of equilibrium noted.	95% confidence limits in brackets. Animals not fed 24 hr prior to or during tests.	Khargserot and Ray, 1987
							24 hr	48 hr	96 hr			

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr			
Simezino (H)	- <u>Bufo bufo japonicus</u>	tadpoles	-	-	-	-	-	>100	-		Obtained from formulated product.	Hoshimoto and Nishizuchi, 1981
Sodium Aluminum Silicate	- <u>Xenopus laevis</u>	3-4 weeks	semi-static	21 ± 2	-	-	-	-	5,600-10,000		Test solutions corrected for pH and oxygen content.	Canton and Slooff, 1982b
Sodium Arsenite (H)	- <u>Adelotus brevicaudatus</u>	1-2 week old tadpoles	A	21-22	-	-	152	119	96			Johnson, 1976
	- <u>Limnodynastes peronii</u>	1-2 week old tadpoles	"	"	"	"	100	92	60			
	- <u>Bufo aspinosus</u>	2 week old tadpoles	"	"	"	"	195	150	123			
Sodium Bromide	- <u>Xenopus laevis</u>	<2 days	semi-static	20 ± 1	-	-	-	-	-	100 day exposure No Effect Levels (the highest concentration at which no effect or lethality was observed): Mortality - 32 Development - 320 Growth - 320	N = 75.	Slooff and Canton, 1983

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr			
Sodium Fluoroacetate (1080)	- <u>Limnodynastes tasmanicus</u>	adults 2-5 g	-	-	Single i.p. injection	-	-	-	-	LD50 approx. 60 ppm. Time until death (median and range) - 78.4 hr (36.8-98.3)		Hellroy et al., 1985
Sodium or Methyl Fluoroacetate	- <u>Rana pipiens</u>		-	-	Subcutaneous injection	-	-	-	-	LD50 = 1%.		Chenoweth, 1969
	- <u>Xenopus laevis</u>		-	-	Subcutaneous or i.p. injection	-	-	-	-	LD50 > 500.0.		
Sodium o-Fluorocrotonate	- <u>Rana pipiens</u>		-	-	Subcutaneous injection	-	-	-	-	LD50 = 25.0.		Chenoweth, 1969
Sodium Thiocyanate	- <u>Xenopus laevis</u>	tadpoles 5-12 days old	A	22 ± 1	-	-						
								10 day				
								2,000		Treatment was continuous for 10 days; LC50 was determined at 10 days post exposure.		Birch and Prehler 1986

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr			
Sour water from coal gasification plant	<i>Ienopus leavis</i>	early cleavage to mid-blastula embryos (stages 2-6.5)	A	20 (6.4 for controls; 6.7 for 1% sour water)	-	-	0.69* (0.61-0.77)	0.36* (0.36-0.38)	-	*Levels in percent sour water content.	N = 200 per treatment. 95% confidence intervals in brackets. Reduction in motility, pigmentation and rate of embryonic development noted.	Dumont and Schultz, 1980
									7 day			
Strontium (M)	<i>Gastrophryne carolinensis</i>	embryo-larval	B	-	Birge & Black 1977 a, b	195.0 ± 5.4 ppm hardness as CaCO <sub>3</sub>		0.16		Exposure from fertilization to 4 days posthatch.	N = 14.	Birge et al., 1979a
							24 hr	48 hr	96 hr			
2,4,5-T amine (H) (2,4,5-trichlorophenoxyacetic acid)	<i>Adelotus brevis</i>	1-2 week old tadpoles	A	21-22	Sanders 1970	-	228	205	200			Johnson, 1976
	<i>Lithodyastes peroni</i>	1-2 week old tadpoles	"	"	"	"	210	190	169			
	<i>Bufo marinus</i>	2 week old tadpoles	"	"	"	"	425	382	340			

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr			
TDE (OC)	<i>Bufo woodhousii fowleri</i>	4-5 week old tadpoles	A	15.5 ± .5 (7.1)	-	30 ppm methyl orange	0.70 (0.25-2.0)	0.32 (0.21-0.45)	0.14 (0.10-0.21)	N = 10. 95% confidence limits in brackets.		Sanders, 1970
	<i>Pseudacris triseriata</i>	1 week old tadpoles	"	"	"	"	0.61 (0.41-0.82)	0.50 (0.21-0.75)	0.40 (0.21-0.75)			
TEPP (OP)	<i>Bufo viridis</i> <i>Rana ridibunda</i>	adults 18-45 g	-	25	Injection into dorsal lymphatic sac	-	-	-	-	24 hr LD50: 540 JA	N = 25 minimum.	Fidry and Schatzberg-Porath, 1960
	<i>Rana pipiens</i>	adults approx. 9 cm	daily renewal	24 (5.3-6.4)	-	-	-	15 day	60			
								30 min	2 hr			
a-fertihenyl	<i>Rana pipiens</i>	embryos stage 25	A	-	-	-	0.11	0.018		LC50 determined for given length of exposure to sunlight.		Kagen et al., 1984

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr			
D-thorstenyl (cont'd)	- <u>Hyla crucifer</u>	tadpoles	-	-	-	-	.003	-	-	Initial incubation time was 1 hr, irradiation time (sunlight and UV) was 1 hr; survival determined 24 hr after treatment.		Kagan et al., 1987
Tetrachloro- phtalide	- <u>Bufo bufo</u> <u>iguanicus</u>	tadpoles	-	-	-	-	-	>40	-	Obtained using formulated product.		Hashimoto and Nishiyuki, 1981
1R tetracethers (Py)	- <u>Rana pipiens</u>	adults 20-30 g	-	20	subcutaneous injection into the dorsum	-	-	-	-	24 hr LD50: $\frac{LC}{LD50}$ greater than 20 1.8 (1.2-2.6)	N = 28-36. 95% confidence limits in parentheses.	Cole and Casida, 1983
Tetrapropylene Benzene Sulphonate	- <u>Xenopus</u> <u>laevis</u>	~2 days	semi- static	20 ± 1	-	-	-	-	-	100 day exposure No Effect Levels (the highest concentration at which no effect or lethality was observed): Mortality - 3.2 Development - 10 Growth - 10	N = 75.	Slooff and Canton, 1983

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr			
TFM (Impricide) (3-trifluoro- methyl-4- nitrophenol)	- <u>Hyla</u> <u>versicolor</u>	larvae	A	17 ± 1 (6.8- 7.0)	-	hardness 44 mg/L as CaCO <sub>3</sub>	-	-	1.98 (1.77- 2.22)	Exposure from ferti- lization to 4 days posthatch.	59.4% TFM tested. 95% confidence intervals in parentheses.  Difficult to compare results of static vs flow through assays.	Chandler and Marking, 1975
	- <u>Rana pipiens</u>	larvae	"	"	"	"	-	-	2.76 (2.45- 3.11)			
	- <u>R.</u> <u>catobryana</u>	larvae	B	16- 17	-	"	-	-	3.55 (2.62- 4.82)			
Thallium (M)	- <u>Gastrophysa</u> <u>carolinensis</u>	embryo- larval	B	- (7.0- 7.8)	Birge & Black 1977 a, b	195.0 ± 5.4 ppm hardness as CaCO <sub>3</sub>		0.11		N = 14.	Birge et al., 1979a	
Thiram (F)	- <u>Xenopus</u> <u>laevis</u>	47	A	20 ± .5 (7.5)	-	230 ppm CaCO <sub>3</sub> hardness				0.017 0.014 0.013	Seugé et al., 1983	
		53	"	"	"	"						0.025 0.022 0.021

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity		7 day				
Lin (M)	<u>Gastrophryne carolinensis</u>	embryo-larval	B	- (7.0-7.8)	Birge & Black 1977 a, b	195.0 ± 5.4 ppm hardness as CaCO <sub>3</sub>		0.09		Exposure from fertilization to 4 days post hatch.	N = 14.	Birge et al., 1979a
Toxaphene (OC)	<u>Rana sphenoccephala</u>	eggs	B	20 (7.2-7.5)	-	total hardness 100 ppm as CaCO <sub>3</sub>	-	-	0.651	Exposed for 96 hr, mortality determined at 15 days.	N = 20-30 per concentration.	Hall and Swineford, 1980
		larvae	"	"	"	"	-	-	0.065 (0.055-0.083)	"	N = 20-30 per concentration. 95% fiducial limits in brackets.	
		sub-adults	"	"	"	"	-	-	0.378 (0.130-1.125)	Exposed for 96 hr, mortality determined at 8 days.	N = 5-10 per concentration. Flow through method produced lower results than static method.	
	<u>Bufo woodhousei</u>	4-5 week old tadpoles	A	15.5 ± 0.5 (7.1)	-	30 ppm methyl orange	.60 (0.30-1.2)	0.29 (0.20-0.42)	0.14 (0.06-0.33)		N = 10. 95% confidence limits in brackets.	Sanders, 1970
<u>Pseudacris triseriata</u>	1 week old tadpoles	"	"	"	"	1.7 (0.50-3.2)	0.70 (0.40-1.2)	0.50 (0.10-1.1)				

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr			
Toxaphene (cont'd)	<u>Rana sphenoccephala</u>	larvae	B	20 (7.2-7.5)	Hall and Swineford 1980	100 ppm as CaCO <sub>3</sub> hardness	-	-	0.130 (0.095-0.184)	24 hr EC50 (behavioural aberrations): 0.193 (0.138-0.298)	95% confidence limits in brackets.	Hall and Swineford, 1981
		larvae	"	"	"	"	-	-	0.099 (0.071-0.166)	0.312 (0.225-0.435)	Disturbed equilibrium, abnormal posture, erratic swimming noted.	
		larvae	"	"	"	"	-	-	0.195 (0.176-0.220)	0.036 (0.023-0.053)		
		larvae	"	"	"	"	-	-	0.034 (0.027-0.042)	0.038 (0.014-0.051)		
		larvae	"	"	"	"	-	-	0.076 (0.048-0.116)	>1.00		
		larvae	"	"	"	"	-	-	0.342 (0.245-0.470)	0.170 (0.122-0.235)		

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr			
fosphono (cont'd)	<u>Achyrotes</u> <u>oculatus</u> <u>fowleri</u>	larvae	B	20 (7.2- 7.5)	Hall and Swineford 1980	100 ppm as CaCO <sub>3</sub> hardness	-	-	0.034 (0.020- 0.057)	24 hr EC50: 0.227 (0.215- 0.254)	95% confidence limits in brackets	Hall and Swineford, 1981 (cont'd)
TPH (tetrachloro- phtalocyanine)	- <u>Bufo bufo</u> <u>japonicus</u>	tadpoles	-	-	-	-	-	0.16	-	-	-	Hashimoto and Nishiuchi, 1981
Trichloroethylene	- <u>Achyrotes</u> <u>oculatus</u>	3-4 weeks post hatch	A	20 ± 1	-	-	-	48	-	-	N = 10 for each species. Organisms not fed during exp.	Slooff and Boorcoorn, 1980
	- <u>Xenopus</u> <u>laevis</u>	3-4 weeks post hatch	-	-	-	-	-	45	-	-	-	-
2,4,6-trichloro- phenol	- <u>Xenopus</u> <u>laevis</u>	tadpoles	B	17.2 ± 5 (7.39 ± .22)	40.0- 49.5 mg/L as CaCO <sub>3</sub>	-	-	-	1.20 (1.08- 1.40)	-	95% confidence interval in parentheses.	Holcombe et al. 1987

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity	24 hr	48 hr	96 hr			
2-(2,4,5-trichloro- phenoxy) Propionic Acid, butyl ether ester (H)	- <u>Bufo</u> <u>woodhousei</u> <u>fowleri</u>	4-5 week old tadpoles	A	15.5 ± .5 (7.1)	-	30 ppm methyl orange	22 (15-32)	20 (13-28)	-	N = 10. 95% confidence limits in brackets.	-	Sanders, 1970
	- <u>Pseudacris</u> <u>triseriata</u>	1 week old tadpoles	-	-	-	-	20 (14-32)	18 (10-31)	10 (4.0-18)			
Trifluralin (H)	- <u>Bufo</u> <u>woodhousei</u> <u>fowleri</u>	4-5 week old tadpoles	A	15.5 ± .5 (7.1)	-	30 ppm methyl orange	0.18 (0.10- 0.30)	0.17 (0.10- 0.31)	0.10 (0.08- 0.49)	N = 10. 95% confidence limits in brackets.	-	Sanders, 1970
	- <u>Bufo bufo</u> <u>japonicus</u>	tadpoles	-	-	-	-	-	14	-			
3,5,5-trimethyl- 1-hexanol	- <u>Xenopus</u> <u>laevis</u>	3-4 week old larvae	A	20 ± 1	-	-	-	13.5	-	N = 10 per concentration.	-	de Zwart and Slooff, 1987
								15 day				
Trithion (OP)	- <u>Rana</u> <u>pipiens</u>	adults approx. 9 cm	renewed daily	24 (6.7)	-	-	-	155	-	N = 24 per concentration.	-	Kaplan and Glaczinski, 1960



Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity		7 day				
Fungeton (H)	<u>Gastrophysa carolinensis</u>	embryo-larval	B	-	Birge & Black 1977 a, b	195.0 ± 5.4 ppm hardness as CaCO <sub>3</sub>	2.90			Exposure from fertilization to 4 days posthatch.	N = 14.	Birge et al., 1979a
							24 hr	48 hr	96 hr			
Yolan (H)		tadpoles 1-1.5 cm	renewed daily	17-20 (7.7-8.2)	-	-	15.1			Value is the LC <sub>50</sub> of the active ingredient.	Perevozchikova, 1975	
							7 day					
Zinc (H)	<u>Gastrophysa carolinensis</u>	embryo-larval	B	-	Birge & Black 1977 a, b	195.0 ± 5.4 ppm hardness as CaCO <sub>3</sub>	0.01			Exposure from fertilization to 4 days posthatch.	N = 14.	Birge et al., 1979a
	<u>Rana pipiens</u>	adulte approx. 9 cm	renewed daily	24 (6.7)	-	-	155			N = 24 per concentration.	Kaplan and Glazowski, 1965	

Table 1 - Acute Toxicity (cont'd)

Contaminant <sup>a</sup>	Species	Stage/age	Experimental Conditions				LC 50 <sup>c</sup>			Effects <sup>c</sup>	Remarks	References
			Flow <sup>b</sup>	Temp. °C (pH)	Methodology	Alkalinity		7 day				
Zinc (cont'd)	<u>Bufo melanostictus</u>	tadpoles 1.8-2.2 cm	A	29-34 (7.1-7.6)	APHA et al. 1976	120-160 mg/L as CaCO <sub>3</sub>	47.26	25.65	19.86	95% confidence limits in brackets.  Animals not fed 24 hr prior to or during tests.	Khangarot and Ray, 1987	
							(35.48-58.19)	(23.73-27.74)	(17.68-23.90)			
	<u>Microhylis ornata</u>	tadpoles 1 week old	renewed daily	25.5-26.0 (6.86-6.94)	-	97.0-98.0 ppm	24.06	23.42	22.41	Ranges in parentheses. Animals not fed during test.	Reo and Madhyastha, 1987	
(23.6-24.4)							(23.0-25.8)	(22.0-22.8)				
	<u>Xenopus laevis</u>	embryo blastula	static renewal	22-24 (7)	-	100 mg/L CaCO <sub>3</sub> hardness	-	-	34.5 ± 1.2	EC50 (malformation)-3.6 ± 0.5.	Mean ± confidence interval.	Dawson et al., 1988
Zinc (F)	<u>Bufo bufo japonicus</u>	tadpoles	-	-	-	-	40	-	Obtained using formulated product.	Mashimoto and Nishiyuchi, 1981		

TABLE 2  
GENERAL EFFECTS - LABORATORY STUDIES

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE
Aboto (OP)	- <u>Bufo boreas</u>	20	Juveniles (X snout-vent length = 19.8 mm)	Exposure of hydrated toads to 60 ppb significantly lowered temperature tolerance.	23-24°C; 24 hr exposure.	Johnson and Prime, 1976
Acophate (OP)	- <u>Rana satesbawiana</u>		tadpoles	All tadpoles exposed to 3 ppm survived.	Continuous flow apparatus for 96 hr. Brain cholinesterase activity of tadpoles fed dosed tadpoles was 109.7% of controls; none died.	Hall and Kolbe, 1980
	- <u>Ambystoma gracile</u>	200-600 per treatment	eggs, larvae	Egg hatch was not significantly affected by concentrations up to 798 µg/L; in first week post-hatch, mortality was directly related to concentration to which larvae exposed.  Growth rate decreased and abnormalities increased when larvae were exposed to 382 and 798 µg/L.	The maximum surface water concentration of acophate used in insect control programs should be <1 µg/L and should have little effect on populations of <u>A. gracile</u>	Geen et al., 1986
	- <u>Rana clamitans</u>	7 per treatment	larvae	Significant decrease in mean activity time recorded at 1000 ppm but not at 500 ppm.	Temp. 21 ± 1°C. 24 hr. acclimation.	Lyons et al., 1976
Acetone Extract	- <u>Rana temporaria</u>		tadpoles	>400 ppm caused 100% mortality, 300 ppm caused 50% mortality, 200 ppm produced no lethal effect during 1 week observation. At 10 ppm accelerated metamorphosis by 2 days in 80% animals observed; 20% did not complete metamorphosis, and were malformed.		Paulov and Paulovova, 1983

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS					REMARKS	REFERENCE
				Exp#	Wght of frog (gm)	Lgth of frog (mm)	TLM % concentration (ppm)	Length of time until death (min)		
Aldrin (OC)	- <u>Rana cyanophytica</u>	4		1	3-4.5	26-35	0.006	180-215	pH - 7.9. Room temp. - 19-26°C. Water temp. - 20-24°C.	Rane and Mathur, 1979
				2	2	21-25.5	0.03	75-95		
				3	6-8	32-43	0.06	140-180		
				4	3-4	30-35	0.09	85-155		
				5	4-5	36-38	0.1	90-110		
				6	3-10	27-46	0.125	55-120		
Aldrin (OC)	- <u>Acris crepitans</u>	360	frogs 18-23 mm snout-vent length	Frogs taken from areas treated or untreated with pesticides and exposed to 30,000 or 50,000 µg/L aldrin in lab. Mortality recorded after 24 and 36 hr.  Percent mortality was lower in frogs from treated fields than in frogs from areas with no or minimum prior treatment.					Areas treated with DDT, endrin, toxaphene, methyl parathion. Aldrin not used. Thus, resistance must represent a cross-resistance.  Temp. 21-24°C.	Vinson et al., 1965
	- <u>A. gryllus</u>	50		10,000 µg/L killed 40% of frogs. 50,000 µg/L killed 70% in 36 hours.						
Aldrin (OC)	- <u>Rana pipiens</u>		abdominal skin	No changes in short-circuit current at 2 x 10 <sup>-6</sup> M.						Webb et al., 1979

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE														
Aldrin (cont'd)	- <u>Bufo arenarum</u>	20 10 10	embryos	5 and 15 ppm produced 100% mortality on day 15 and 10, respectively. 1 ppm was not toxic.	Exposed from fertilization, medium replaced daily.	Juarez and Guzman, 1980b														
	- <u>Rana pipiens</u>		liver microsomes (adults)	Microsomal metabolism (apparent $V_{max}$ and $K_m$ values $\pm$ SE): App $V_{max}$ = $0.056 \pm 0.01$ nmol/min/mg. App $K_m$ = $4.3 \pm 3$ nmol/mg.			Ronia and Walker, 1985													
	- <u>Rana pipiens</u>		adults 65 g, 8.9 cm long	<table border="1"> <thead> <tr> <th>Aldrin (ppm)</th> <th>pH of solution at 25°C</th> <th>No. of frogs dead at 30 days</th> </tr> </thead> <tbody> <tr> <td>0.30</td> <td>5.90</td> <td>8</td> </tr> <tr> <td>0.23</td> <td>5.71</td> <td>0</td> </tr> <tr> <td>0.15</td> <td>5.68</td> <td>0</td> </tr> </tbody> </table>			Aldrin (ppm)	pH of solution at 25°C	No. of frogs dead at 30 days	0.30	5.90	8	0.23	5.71	0	0.15	5.68	0	Frogs placed in 200 cc of test solution. Neuromuscular changes produced; excessive thrashing and abnormal reactivity to stimulation were observed.	Kaplan and Overpeck, 1964
	Aldrin (ppm)		pH of solution at 25°C	No. of frogs dead at 30 days																
0.30	5.90	8																		
0.23	5.71	0																		
0.15	5.68	0																		
Aldrin-transdiol	- <u>Xenopus laevis</u>		lateral-line organ	$2.5 \times 10^{-6}$ and $7.5 \times 10^{-6}$ M failed to induce repetitive activity; caused marked increase in the rate of spontaneous firing, followed by a blockade.	Temp. 19-21°C.	Akkermans et al., 1975a														
			cutaneous touch receptors	Exposure to $10^{-5}$ M failed to induce repetitive nerve spikes; possible blocking effect.	No effects of temperature changes noted.															

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE
Aldrin - transdiol (cont'd)	- <u>Rana sacculenta</u> <u>R. temporaria</u>		sartorius nerve-muscle	$2.5 \times 10^{-5}$ M produced increase in end-plate potential to a time control level at 20 min, after which amplitude declined and transmission completely blocked.	Temp. $18 \pm 0.2^\circ\text{C}$ .  Hypothesis that aldrin-transdiol increases amount of calcium entering nerve terminal during nerve impulse.	Akkermans et al., 1975b
	- <u>Rana sacculenta</u> <u>R. pipiens</u> <u>R. temporaria</u>		motor end-plate	Concentrations of $10^{-5}$ to $10^{-4}$ exerted both pre- and post-synaptic actions; caused increase in end-plate potential frequency and decrease in their amplitude.	Temp $18 \pm 0.2^\circ\text{C}$ .	Akkermans et al., 1974
	- <u>Xenopus laevis</u>		spinal chord	Application of 1 to $5 \times 10^{-5}$ M in vitro caused potentiation of spinal reflex activity, increase in spontaneous activity of ventral and dorsal roots and reduction of spinal inhibitory mechanisms.	Temp. $10 \pm 2^\circ\text{C}$ .	Akkermans et al., 1975c
Allethrin (PV)	- <u>Xenopus laevis</u>		lateral-line organ	Repetitive activity induced by application of 1-3 ppm allethrin for 20-40 min.		Akkermans et al., 1975e
			cutaneous touch receptors	Produced repetitive activity after 5 min exposure to $10^{-5}$ M or 15 min to $10^{-6}$ M.	Slight increase in number of repetitive spikes observed when temperatures of preparation lowered from 19-21°C to 10-15°C. Negative temperature co-efficient of activity.	

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE
Allethrin (cont'd)	- <u>Xenopus laevis</u>		myelinated fibre from sciatic nerve	$2 \times 10^{-5}$ M hardly affected rate of rise, amplitude and declining phase of sodium current during test pulse. Upon repolarisation, large, slow sodium tail current apparent.	Temp. $15 \pm 1^\circ\text{C}$ . Concluded that allethrin acts on open sodium channels only.	van den Bercken and Vijverberg, 1980
	- <u>Rana pipiens</u>		nerve-muscle	Muscle contraction evoked by nerve stimulation was augmented somewhat at low concentrations ( $10^{-6}$ - $10^{-5}$ M) but was suppressed at higher concentrations ( $1-5 \times 10^{-4}$ M).	Temp. $22^\circ\text{C}$ .	Iokano et al., 1977
	- <u>Rana esculenta</u> <u>R. temporaria</u> <u>Xenopus laevis</u>		peripheral nervous system	Induced repetitive activity in sensory nerve fibres (0.33 - 3.3 $\mu\text{M}$ ), sensory organs (1-10 $\mu\text{M}$ ) and in distal part of the motor fibres, leading to pronounced repolarization in the motor end plate (0.1 - 10 $\mu\text{M}$ ).	May account for hyperexcitation and tremors leading to paralysis and death. Negative temperature coefficient of activity in lateral-line sense organ and motor nerve terminal.	van den Bercken, 1977
	- <u>Xenopus laevis</u>		adults  lateral line organ	Exposure to 5 ppm caused excitation and convulsions within 10 min; excitation also occurred following exposure to 1 ppm for 30 min.  2 ppm ( $6.6 \times 10^{-6}$ M) in vitro caused pronounced repetitive activity.	  Similarity between DDI and allethrin noted.	van den Bercken et al., 1975

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE
Allethrin (cont'd)	- <u>Rana temporaria</u> <u>R. esculenta</u>		motor-end plate	$10^{-7}$ M produced pronounced repetitive activity in the motor end plate; a single nerve stimulus could provoke up to 15 end plate potentials.	Showed negative temperature coefficient.	Wouters et al., 1977
Altoacid (IGR)	- <u>Bufo boreas</u>	20	juveniles ( $\bar{x}$ snout-vent length = 19.8 mm)	Exposure of hydrated toads to 100 ppb significantly lowered temperature tolerance.	$23-24^\circ\text{C}$ ; 24 hr exposure.	Johnson and Price, 1976
Aluminum (M)	- <u>Rana catesbeiana</u>	26 7	adults 100-300 g brain	$\text{Al}^{3+}$ enriched water environment produced no measurable changes in AChE levels in brain, <i>in vivo</i> or <i>in vitro</i> .	pH 4.6-6.6.	Marquis, 1982
	- <u>Ambystoma maculatum</u>	18-46 per treatment	embryos late blastula stage	Between pH 4.5 - 6.0 Al had no effects on hatching success; at pH 4.0, hatching success decreased from 11% at 0.1 ppm total Al to 3% at 0.4 ppm and 2% at 0.7 and 1.1 ppm. All larvae hatched at pH 4.0 died within a few days.	Exposure for 31 days at $21^\circ\text{C}$ .	Dale et al., 1985
	- <u>Hyla crucifer</u>	11-34 per treatment	embryos mid-blastula	No consistent effect on hatching success at any pH where hatching occurred (pH 4.5, 5.0, 6.0).	Exposure for 16 days at $21^\circ\text{C}$ .	
	- <u>Bufo americanus</u>	47-52 per treatment	embryos early gastrula stage	Hatching >90% at all pH's and total Al concentrations from pH 4.0 - 6.0.	Exposure for 11 days at $21^\circ\text{C}$ .	

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE																															
Aluminum (cont'd)	<u>Xenopus laevis</u>	10 per treatment	embryos prior to 4 cell stage	% hatching (% of hatched larvae dead at end of expt. in parentheses) in oligotrophic brown water:  Al Concentration (ppm)  <table border="1"> <thead> <tr> <th>pH</th> <th>0.15</th> <th>0.20</th> <th>0.40</th> <th>0.90</th> </tr> </thead> <tbody> <tr> <td>3.5</td> <td>0 (-)</td> <td>0 (-)</td> <td>0 (-)</td> <td>0 (-)</td> </tr> <tr> <td>4.0</td> <td>0 (-)</td> <td>0 (-)</td> <td>0 (-)</td> <td>0 (-)</td> </tr> <tr> <td>4.5</td> <td>88 (100)</td> <td>100 (38)</td> <td>100 (0)</td> <td>100 (100)</td> </tr> <tr> <td>5.0</td> <td>100 (0)</td> <td>100 (0)</td> <td>100 (0)</td> <td>90 (100)</td> </tr> <tr> <td>6.0</td> <td>100 (0)</td> <td>100 (0)</td> <td>100 (0)</td> <td>100 (50)</td> </tr> </tbody> </table>	pH	0.15	0.20	0.40	0.90	3.5	0 (-)	0 (-)	0 (-)	0 (-)	4.0	0 (-)	0 (-)	0 (-)	0 (-)	4.5	88 (100)	100 (38)	100 (0)	100 (100)	5.0	100 (0)	100 (0)	100 (0)	90 (100)	6.0	100 (0)	100 (0)	100 (0)	100 (50)	Exposure for 11 days at 21°C. At .05 - .15 ppm Al, high frequency deformed larvae found at pH 4.5; malformed larvae also found at pH >5.0 and high Al (=55).	Odeh et al., 1985 (cont'd)	
	pH	0.15	0.20	0.40	0.90																																
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6.0	100 (0)	100 (0)	100 (0)	100 (50)																																	
	<u>Rana sylvatica</u>	100 per treatment	eggs within 24 hr of oviposition	Percent hatching success:  <table border="1"> <thead> <tr> <th rowspan="2">Nominal Al (ug/L)</th> <th colspan="3">Nominal pH</th> </tr> <tr> <th>5.75</th> <th>4.75</th> <th>4.14</th> </tr> </thead> <tbody> <tr> <td>0</td> <td>91</td> <td>93</td> <td>*81</td> </tr> <tr> <td>10</td> <td>89</td> <td>95</td> <td>*74</td> </tr> <tr> <td>20</td> <td>93</td> <td>87</td> <td>*67</td> </tr> <tr> <td>50</td> <td></td> <td>92</td> <td>*56</td> </tr> <tr> <td>100</td> <td></td> <td>96</td> <td>*67</td> </tr> <tr> <td>200</td> <td></td> <td></td> <td>*40</td> </tr> </tbody> </table> * indicates significant difference between treatments on either side of asterisk.	Nominal Al (ug/L)	Nominal pH			5.75	4.75	4.14	0	91	93	*81	10	89	95	*74	20	93	87	*67	50		92	*56	100		96	*67	200			*40	Aluminum toxicity dependent on pH.  Egg mortality due to hydrogen ion stress was correlated with delay in time to hatch and reduction in egg size.	Clark and LaZerte, 1985
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Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE																																																							
Aluminum (cont'd)	<u>Bufo americanus</u>	89-160 per treatment	eggs within 24 hr of oviposition	Percent hatching success:  <table border="1"> <thead> <tr> <th rowspan="2">Nominal Al (ug/L)</th> <th colspan="3">Nominal pH</th> </tr> <tr> <th>4.75</th> <th>4.32</th> <th>4.14</th> </tr> </thead> <tbody> <tr> <td>0</td> <td>97</td> <td></td> <td>*75</td> </tr> <tr> <td>10</td> <td>100</td> <td></td> <td>*56</td> </tr> <tr> <td>20</td> <td>94</td> <td></td> <td>*61</td> </tr> <tr> <td>50</td> <td>98</td> <td></td> <td>*53</td> </tr> <tr> <td>100</td> <td>99</td> <td></td> <td>*55</td> </tr> <tr> <td>200</td> <td></td> <td></td> <td>34</td> </tr> <tr> <td>0</td> <td>90</td> <td>84</td> <td>*40</td> </tr> <tr> <td>5</td> <td></td> <td>89</td> <td>*41</td> </tr> <tr> <td></td> <td></td> <td>*</td> <td>*</td> </tr> <tr> <td>10</td> <td></td> <td>64</td> <td>*18</td> </tr> <tr> <td>20</td> <td></td> <td>69</td> <td>*21</td> </tr> <tr> <td>50</td> <td>97</td> <td>74</td> <td></td> </tr> </tbody> </table> * significant difference of treatments on either side of asterisk in that row or column.	Nominal Al (ug/L)	Nominal pH			4.75	4.32	4.14	0	97		*75	10	100		*56	20	94		*61	50	98		*53	100	99		*55	200			34	0	90	84	*40	5		89	*41			*	*	10		64	*18	20		69	*21	50	97	74		Aluminum toxicity dependent on pH.  Egg mortality due to hydrogen ion stress was correlated with delay in time to hatch and reduction in egg size.	Clark and LaZerte, 1985 (cont'd)
	Nominal Al (ug/L)	Nominal pH																																																											
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	<u>R. sylvatica</u> <u>B. americanus</u>	50 per treatment	tadpoles	Survival not affected by exposure to 0-200 ug/L Al in combination with pH 4.14 - 5.75.	Tadpoles which had also been exposed during egg-stage survived as well as those which developed in egg-stage in non-acidic, low-aluminum water.																																																								

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE
Aluminum (cont'd)	- <u>Rana temporaria</u>	12 per treatment	tadpoles	Growth depressed and metamorphosis delayed by 800 ug/L and several tadpoles died at foreleg emergence. At 1600 ug/L small tadpoles died, large tadpoles metamorphosed at small size but without delay.	Tadpoles raised in given concentration Al to metamorphosis.	Cuccina, 1986
Aminocarb (C)	- <u>Rana catesbeiana</u>	7 per treatment	larvae	Significant decrease in mean activity time recorded at 10 ppm but not at 5 ppm.	Temp. 21 ± 1° C. 24 hr acclimation.	Lyons et al., 1976
Arsenic (non-ionized)	- <u>Xenopus laevis</u>		embryos	Gastrulation is most resistant stage.		Cotte Reusino, 1980
Aniline	<u>Xenopus laevis</u>	60 per treatment	tadpoles stage 58	All tadpoles dead within 30 sec and 2 days at 10,000 and 1000 ppm, respectively.	Exposure for 2 weeks.	Duempert, 1987
		30 per treatment		Animals exposed to 90 ppm died within 12 days; only those held at ≤ 30 ppm developed into toads. 1 ppm resulted in reduced body size in toads.	Exposure for 90 days.	
Aroclor 66-E <sub>2</sub> (nanomolecular organic surface film)	- <u>Hyla cinerea</u>		tadpoles	Progressed normally from tadpole to adult in tank with 0.68 ml/m <sup>2</sup> added.		Webber and Cochran, 1984
Atrazine (H) (Weedex A)	- Frog	25 per test	spoon	Spoon developed normally in various waters used except in water containing Weedex (0.03 g/L in 3 litres) or water containing mud from a contaminated pond.	Thought that Weedex had drained into a pond resulting in no hatching success for two years.	Hazelwood, 1969

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE	
<u>Bacillus thuringiensis</u> (var. <u>terrestris</u> )	- <u>Rana temporaria</u>	10 per treatment	tadpoles	Exposure to 10 mg/L decreased weight, delayed metamorphosis by four days.	Lyophilized endotoxins from B.t.i. (28 x 10 <sup>10</sup> sp./g) used.	Paulov, 1987a	
Benzene	- <u>Rhinophrynus cristatus</u>	35	blood	Exposed for 1 minute to 250 ppm benzene in air:  Daily exposure for 43 days produced transitory alterations in hematological picture followed by a return to normal.  Twice weekly exposure for 59 days or alternate day exposure for 38 days produced same results as above but slower.  Alterations included increase in number of circulating white blood cells, decrease in red blood cells preceded by decrease in hemoglobin percentage and increase in size of red blood cells.	Intensity of alterations was proportional to frequency of treatments. Onset of a modification was related to time interval following first treatment.	Garavini and Seren, 1978	
Benzene Hexachloride (OC)	- <u>Rana pipiens</u>		adults 65 g, 8.9 cm long		Frogs placed in 200 cc of test solution. Frogs lethargic; made no attempt to escape handling.	Kaplan and Overpeck, 1964	
		20		BHC (ppm)	pH of solution at 25°C	No of frogs dead at 30 days	
		10		17.0	6.23	3	
		10		8.0	5.85	0	
		10		6.0	5.82	0	

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE
Benzo(a)pyrene	- <u>Rana pipiens</u>		marrow cells	Three-fold increase in sister chromatid exchanges at 0.01 ug/ml in ambient water. Mitotic index reduced and cell cycling time lengthened in dose-dependent manner.		Geard and Soutter, 1986
	- frog		eye lens epithelium	No micronuclei present in the epithelium; cell cycle appeared unaffected in frogs immersed in $\leq 20.0$ ug/ml for 1 week.	No evidence of genotoxicity.	Kung et al., 1987
	- <u>Pleurodeles waltl</u>	30 per dose	erythrocytes from larvae stage 53	Incidence of micronuclei in larvae exposed for 8 days markedly increased with dose up to 0.075 ppm, then more gradually reaching 158 per 1000 at 0.75 ppm, minimum dose which produced a significant increase was 0.01 ppm.	Exposure for $< 2$ days did not result in increased incidence of micronuclei.	Grinfeld et al., 1986
Beryllium (M)	- Frogs		embryos	Development not interfered with until N/5000 to N/1000.		Dilling and Healey, 1926
Bidrin (OP)	- Frogs		sartorius muscle	Inhibition of membrane voltage response of muscle to carbamylcholine which was dependent upon concentration and temperature.		Dekin et al., 1978
Dioxmethrin (PY)	- <u>Xenopus laevis</u>		lateral-line sense organ	$5 \times 10^{-6}$ M for 2-3 hr <i>in vivo</i> caused only weak repetitive activity.		Vijverberg et al., 1982
			peripheral nerves	$5 \times 10^{-6}$ M caused repetitive activity within first 2 hr of exposure.		

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE																					
Cadmium (M)	- <u>Rana catesbeiana</u>		abdominal skin	Decreased skin resistance upon epidermal application of 2 uM.		Arita et al., 1979																					
	- <u>Rana catesbeiana</u>		heart, cardiac nerves	Cd reduced acetylcholine release from cardiac nerve. 10 uM did not alter the compound action potential of nerve trunk or affect pacemaker activity of heart.	Cd may act on the cardiac nerve terminals where it suppresses the release of acetylcholine.	Hayashi and Takayama, 1978																					
	- <u>Gastrophysa carolinensis</u>	100	eggs	Percent mortality and teratogenesis at hatching (H) and 4 days posthatching (PH) of eggs exposed to sediment-bound cadmium:	Hardness - 200 ppm as CaCO <sub>3</sub> . pH = 7.5 - 8.0. Percent mortality expressed as frequency in experimental population/controls.	Biroe et al., 1977																					
				<table border="1"> <thead> <tr> <th rowspan="2">Cd added to sediment (ppm)</th> <th rowspan="2">Concentration in sediment (ppm)</th> <th colspan="2">% mortality</th> </tr> <tr> <th>H</th> <th>PH</th> </tr> </thead> <tbody> <tr> <td>0.1</td> <td>1.36</td> <td>27 (1)*</td> <td>33</td> </tr> <tr> <td>1.0</td> <td>2.18</td> <td>76 (9)</td> <td>33</td> </tr> <tr> <td>10.0</td> <td>14.8</td> <td>31 (7)</td> <td>41</td> </tr> <tr> <td>100.0</td> <td>127.8</td> <td>36 (11)</td> <td>52</td> </tr> </tbody> </table>	Cd added to sediment (ppm)	Concentration in sediment (ppm)	% mortality		H	PH	0.1	1.36	27 (1)*	33	1.0	2.18	76 (9)	33	10.0	14.8	31 (7)	41	100.0	127.8	36 (11)	52	* Percentage of survivors bearing gross congenital deformities at hatching.
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Cadmium (cont'd)	- <u>Xenopus laevis</u>	5 per treatment	lung	Histopathological lesions on lungs pneumonia found in animals from 0.3 ppm Cd <sup>2+</sup> .		Canton and Siooff, 1982a
	- <u>Bufo arenarum</u>	60-120 per treatment	embryos 2-cell stage	In 0.50 - 4.00 ppm Cd <sup>2+</sup> range high mortality, delayed development, significant alterations in the gastrulation and neurulation processes observed.  In 0.03 - 0.25 ppm range lethality much lower, retarded growth, reduced body size, behavioural disorders, and malformations observed.	Exposure was continuous from 2-cell stage onward.	Perez-Coll et al., 1985
	- <u>Xenopus laevis</u>		embryo	Cannot tolerate >2 ppm; stages of organogenesis are most sensitive.		Cotta Reusino, 1980
	- <u>Rana temporaria</u>		adults	Subcutaneous injections of 0.12 - 0.24 mg/100 g/day for 10 days did not cause death.		Vasili'eva et al., 1987
				skin epithelium	High external concentrations CdCl <sub>2</sub> (0.005%) inhibited activity of Na, K-ATPase in skin epithelium.	
	- <u>Rana japonica</u> <u>R. nigromaculata</u> <u>Rhacophorus schlegelii</u>	5 per treatment	adults	Injection (i.v.) of 0.225 mg Cd/kg body weight 11 times during 15 days induced metallothionein (MT): - induced MT consisted of a single isoform - induced MT was Cd, Cu-MT in contrast to native Cu-MT.	No effect on concentrations of 10 elements in the livers and kidneys except Cu.	Suzuki et al., 1986

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE
Cadmium (cont'd)	- <u>Bufo regularis</u>	10 per group	adults female	A single i.v. injection of 6.2 mg Cd <sup>2+</sup> /kg produced elevated serum activities of alanine aminotransferase, aspartate aminotransferase, alkaline phosphatase, lactic dehydrogenase.  Ca serum not affected; serum phosphorus, total protein, total bilirubin increased.	6.2 mg/kg = 96 hr LD50.  0.2 mmole/kg EDTA protected toads from mortality in up to 20 mg Cd <sup>2+</sup> /kg.	Hilmy et al., 1986a
	- <u>Bufo regularis</u>		liver, kidney, spleen, muscle, whole blood	Following a single injection of 6.2 mg Cd/kg:  - Zn increased significantly in liver, kidney; decreased significantly in whole blood, no change in muscle, spleen  - Mg increased in liver, spleen, whole blood; no change in kidney, muscle  - Cu decreased in liver, muscle, whole blood but not in kidney, spleen  - Ca increased in liver, muscle, spleen; decreased in whole blood; no change in kidney  - Fe decreased in kidney, spleen, whole blood; no change in muscle; increased then decreased in liver.	Co-administration of 40 mg EDTA/kg caused Zn, Cu to return to normal in most tissues and organs. EDTA therapy caused increase in Mg, Ca, Fe in most tissue and organs.  6.2 mg Cd/kg = 96 hr LD50.	Hilmy et al., 1986b



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Cadmium (cont'd)	- <u>Rana pipiens</u>	100 per treatment	embryo-larval	Survival rates at hatching and 4 days posthatching were at or above 99% following exposure to Cd-enriched sediments (1.04 - 1074 ppm).	Concentrations in water were 1.0 - 76.5 ug/L.	Francis et al., 1984																																																																																				
	- <u>Bufo regularis</u>	20 per group	adult female, 25 ± 4.3 g	6.2 mg Cd <sup>2+</sup> /kg i.m. injection caused decrease in erythrocyte count, haemoglobin content, haematocrit value, reaching minimum after 48 hr; then gradual increase after 96 hr.	6.2 mg/kg = 96 hr LD50. EDTA therapy caused immediate recovery.	Hilmy et al., 1986c																																																																																				
	- <u>Rana pipiens</u>	100	embryos	Percent survival with treatment initiated at cleavage (C1), Neurula (N) and tail Bud (TB) stages:	Temp. 21.1 - 22.2°C.	Birge and Just, 1975b																																																																																				
				<table border="1"> <thead> <tr> <th rowspan="2">Days</th> <th colspan="2">2.5 ppm</th> <th colspan="2">0.5 ppm</th> <th colspan="2">0.1 ppm</th> <th colspan="2">0 ppm</th> </tr> <tr> <th>C1</th> <th>N</th> <th>C1</th> <th>N</th> <th>C1</th> <th>N</th> <th>C1</th> <th>N</th> </tr> </thead> <tbody> <tr> <td>1</td> <td>73</td> <td>77</td> <td>53</td> <td>90</td> <td>93</td> <td>74</td> <td>97</td> <td>85</td> <td>100</td> </tr> <tr> <td>2</td> <td>48</td> <td>70</td> <td>53</td> <td>90</td> <td>85</td> <td>74</td> <td>94</td> <td>85</td> <td>100</td> </tr> <tr> <td>3</td> <td>36</td> <td>50</td> <td>51</td> <td>90</td> <td>78</td> <td>74</td> <td>85</td> <td>85</td> <td>100</td> </tr> <tr> <td>4</td> <td>8</td> <td>33</td> <td>51</td> <td>90</td> <td>68</td> <td>74</td> <td>84</td> <td>85</td> <td>100</td> </tr> <tr> <td>5</td> <td>3</td> <td>30</td> <td>51</td> <td>90</td> <td>63</td> <td>72</td> <td>74</td> <td>85</td> <td>100</td> </tr> <tr> <td>6</td> <td>0</td> <td>30</td> <td>51</td> <td>90</td> <td>63</td> <td>70</td> <td>70</td> <td>85</td> <td>100</td> </tr> <tr> <td>7</td> <td></td> <td>30</td> <td>51</td> <td>90</td> <td>63</td> <td>70</td> <td>70</td> <td>85</td> <td>100</td> </tr> </tbody> </table>			Days	2.5 ppm		0.5 ppm		0.1 ppm		0 ppm		C1	N	C1	N	C1	N	C1	N	1	73	77	53	90	93	74	97	85	100	2	48	70	53	90	85	74	94	85	100	3	36	50	51	90	78	74	85	85	100	4	8	33	51	90	68	74	84	85	100	5	3	30	51	90	63	72	74	85	100	6	0	30	51	90	63	70	70	85	100	7		30	51	90	63	70
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- <u>Rana catesbeiana</u>			eye	12.5 µM Cd <sup>2+</sup> depressed the amplitude of rod receptor potential in the retina by 50%, while leaving cone response unaffected. 5.0 µM decreased rod response 27%.	Effect reversible.	Fox and Sillman, 1979																																																																																				

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE												
Cadmium (cont'd)	- <u>Rana nigromaculata</u>		eggs, tadpoles	Exposure of eggs to 4 ppm Cd <sup>2+</sup> resulted in partial reduction of primordial germ cells at the 9-12 mm body length stage; caused abnormalities in many embryos.	Treatment prior to the first cleavage.	Hah, 1978												
	- <u>Rana pipiens</u>		epithelial membranes	10 <sup>-3</sup> M Cd <sup>2+</sup> did not inhibit active sodium transport across isolated epithelial cell layer from frog skin or when applied to basal-lateral surface; inhibited transport across frog urinary bladder and large intestine.	Effects vary according to epithelium studied and presence of potential carrier molecules.	Hillyard et al., 1979												
	- <u>Iriturus pyrrhogaster</u>		stomach mucous epithelial cells	10 <sup>-4</sup> M Cd <sup>2+</sup> decreased membrane potential to 66% of the control value; effective membrane resistance and electrical coupling ratio not appreciably affected. 10 <sup>-7</sup> - 10 <sup>-5</sup> M did not affect electrical properties.	Action antagonized by cysteine and acetylpenicillamine.	Kanno et al., 1978												
	- <u>Notophthalmus viridescens</u>	12 per group	adult	<table border="1"> <thead> <tr> <th>Nominal Cd<sup>2+</sup> concentration (ppm)</th> <th>Mortality (%) on Day 51</th> <th>Limb Regeneration</th> </tr> </thead> <tbody> <tr> <td>2.25</td> <td>35</td> <td>onset delayed</td> </tr> <tr> <td>4.5</td> <td>45</td> <td>onset delayed</td> </tr> <tr> <td>6.75</td> <td>80</td> <td>onset delayed - longer than groups above</td> </tr> </tbody> </table>	Nominal Cd <sup>2+</sup> concentration (ppm)	Mortality (%) on Day 51	Limb Regeneration	2.25	35	onset delayed	4.5	45	onset delayed	6.75	80	onset delayed - longer than groups above	No mortality in controls. Cd caused an increase in frequency and severity of abnormal limb regeneration.	Manson and O'Flaherty, 1978
	Nominal Cd <sup>2+</sup> concentration (ppm)	Mortality (%) on Day 51	Limb Regeneration															
2.25	35	onset delayed																
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6.75	80	onset delayed - longer than groups above																
- <u>Xenopus laevis</u>		myelinated nerve fibres	Decreased permeability constant in K and Na systems; reversibly shifted Na activation curve in positive direction along potential axis.		Arhee, 1980													

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT*	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE																																																																													
Cadmium (cont'd)	- <u>Rana catesbeiana</u>		abdominal skin	Activity of ouabain - sensitive, ATPase completely inhibited by 1 $\mu$ M Cd <sup>2+</sup> . From 10 <sup>-7</sup> -10 <sup>-3</sup> M larger decreases found in activities of ouabain - sensitive ATPase and ouabain - insensitive ATPase as Cd concentration increased.	Uptake of Cd into whole skin was about 150 $\mu$ M/kg wet weight with 2 $\mu$ M treatment for 20 min.	Takado and Hayashi, 1978a																																																																													
	- <u>Rana catesbeiana</u>		abdominal skin	2 $\mu$ M epidurally applied for 20 minutes caused 41.5% increase in short circuit current; no change in SCC when 2 $\mu$ M dermally applied.		Takado and Hayashi, 1978b																																																																													
	- <u>Rana catesbeiana</u>		abdominal skin	1 $\mu$ M increased short circuit current to 126%. Cd <sup>2+</sup> acted uncompetitively on the binding reaction of Na <sup>+</sup> with a Na <sup>+</sup> entry channel.	No interaction between Cd <sup>2+</sup> and ouabain effects on Na transport.	Takado and Hayashi, 1981a,b																																																																													
	- <u>Xenopus laevis</u>		embryo		Exposure for 6 days at 18°C.	Miller and Landmann, 1978																																																																													
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				D - Death; S - Severely deformed; M - Moderately deformed; SL - Slight effects; N - No effects; numbers = % survival from blastula to feeding stage.																																																																															

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT*	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE																																																										
Cadmium Chloride (see also Cadmium, Zinc sulphate)	- <u>Rana tigrina</u>	10 per group	ovaries	Effect of i.p. injections of 200 $\mu$ g each at 10-day interval:	In Cd-treated frogs, ovaries contained large number of yolky atretic follicles.	Prasad and Sridapur, 1986																																																										
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	- <u>Xenopus laevis</u>	20 per test	tadpoles stage 54-58	Mortality (%) of pre-treated (P) and non pretreated (C) tadpoles:	Pretreatment with 5.0 mg/l zinc sulphate or 2.5 mg/l cadmium chloride for 96 hr.	Woodall et al., 1988																																																										
				<table border="1"> <thead> <tr> <th rowspan="2">Pre-treat./ Exposure Metal</th> <th rowspan="2">Cd conc. (mg/l)</th> <th colspan="3">Time (hr)</th> </tr> <tr> <th>15</th> <th>45</th> <th>75</th> </tr> </thead> <tbody> <tr> <td rowspan="6">Cd/Cd</td> <td>50 P</td> <td>0</td> <td>0</td> <td>0</td> </tr> <tr> <td>C</td> <td>55</td> <td>75</td> <td>80</td> </tr> <tr> <td>80 P</td> <td>21</td> <td>30</td> <td>30</td> </tr> <tr> <td>C</td> <td>84</td> <td>100</td> <td>100</td> </tr> <tr> <td>100 P</td> <td>26</td> <td>35</td> <td>35</td> </tr> <tr> <td>C</td> <td>90</td> <td>100</td> <td>100</td> </tr> <tr> <td rowspan="6">Zn/Cd</td> <td>50 P</td> <td>4</td> <td>35</td> <td>36</td> </tr> <tr> <td>C</td> <td>13</td> <td>40</td> <td>44</td> </tr> <tr> <td>80 P</td> <td>4</td> <td>5</td> <td>5</td> </tr> <tr> <td>C</td> <td>35</td> <td>80</td> <td>81</td> </tr> <tr> <td>100 P</td> <td>0</td> <td>5</td> <td>5</td> </tr> <tr> <td>C</td> <td>48</td> <td>70</td> <td>77</td> </tr> </tbody> </table>	Pre-treat./ Exposure Metal	Cd conc. (mg/l)	Time (hr)			15	45	75	Cd/Cd	50 P	0	0	0	C	55	75	80	80 P	21	30	30	C	84	100	100	100 P	26	35	35	C	90	100	100	Zn/Cd	50 P	4	35	36	C	13	40	44	80 P	4	5	5	C	35	80	81	100 P	0	5	5	C	48	70	77		
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Carbaryl (C) (Kerbatox 75)	- <u>Rana temporaria</u>	50-75 per treatment	tadpoles stage 25	Exposure to 0.1 - 0.05% caused death within 10-24 hr; short exposure (30 or 15 min) caused rapid frantic swimming.  Exposure to .01% for 24 hr or .001% for 10 days caused tail malformations, increased mortality. Tadpoles increased activity for 2 hr followed by gradual immobilization.		Rzehak et al., 1977																																																										

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE
Carbaryl (cont'd)	- <u>Xenopus laevis</u>	50-75 per treatment	tadpoles stage 19-20	Exposure to 0.1% for up to 2 hr caused contraction and separation of muscle fibres.  Exposure to 0.05% for 4 hr or 0.01% for 10 hr produced bent tails, retarded growth; after 24 hr 0.01% group appeared moribund and heart beat was slow - 80% died in first week and remainder died in second week.  Exposure to 0.0001% for 8 days produced bent tails, oedema, inhibited growth and development. Mortality was 20% in first week, 65% in second week.		
	- <u>Rana tigrina</u>	6 per group	adults 80 ± 5 g	Hepatic vitamin A storage significantly lowered and serum vitamin A levels elevated at 0.006% but not at 0.004%.	Starved for 24 hr before exposure.	Keshaven and Deshaikh, 1984
	- <u>Rana pipiens</u>		abdominal skin	$2 \times 10^{-4}$ M produced significant increases in the short-circuit current and decreases in the resistance.	Likely that the sodium permeability of the outer barrier was altered.	Webb et al., 1979
	- <u>Rana tigrina</u>	4-7 per treatment	tadpoles	Rates of feeding, defecation, excretion increased with increasing concentrations (0-5 mg/L); conversion rate decreased leading to production of small froglets. No mortality until appearance of forelimb; during metamorphosis mortality was significant at 2 mg/L.	Older and larger tadpoles were more resistant.	Marian et al., 1983
	- <u>Rana tigrina</u>		pancreas serum	Exposure of frogs to 0.002-0.006% had no effect on amylase or lipase activities of either tissue.		Deshaikh and Keshaven, 1987
Carbofuran (C)	- <u>Rana pipiens</u>		nerve-muscle	Muscle contractions evoked by nerve or muscle stimulation were suppressed by $1 \times 10^{-5}$ to $1 \times 10^{-3}$ M.  Response to indirect nerve stimulation was suppressed more effectively than response to direct muscle stimulation by $2.5 \times 10^{-4}$ M.	Temp. 22°C.	Iakeno et al., 1977

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS			REMARKS	REFERENCE
Chloranil (H,F)	- <u>Xenopus laevis</u>	50	embryos tail bud stage	Concentration (ppm)		24 hr Survival	Surviving embryos (1.25 - 1.75 ppm) showed alterations in the development of the otolith, optic cup and pigmentation. Movement was sporadically convulsive.	Anderson and Prahlad, 1976
				control		100%		
				0.50		100%		
				0.75		95%		
				1.00		90-95%		
				1.25		90%		
				1.50		65%		
				1.75		40%		
				2.00		5%		
				* taken from mortality curve.				
Chlordane (OC)	- <u>Rana pipiens</u>	20 10 10	adults 65 g, 8.9 cm long	Chlordane (ppm)	pH of solution at 25°C	No. of frogs dead at 30 days	Frogs placed in 200 cc of test solution. Neuromuscular changes produced; excessive thrashing and abnormal reactivity to stimulation were observed.	Kaplan and Overpeck, 1964
				0.50	5.79	8		
				0.38	5.75	0		
			0.25	5.70	0			
	- <u>Bufo arenarum</u>		embryos	5 and 15 ppm produced 100% mortality on day 20 and 14, respectively. 1 ppm was not toxic.			Exposed from fertilization, medium replaced daily.	Juarez and Guzman, 1984b
Chlordane (α- and γ-)	- <u>Rana pipiens</u>		abdominal skin	Produced no significant changes in short circuit current or resistance at $2 \times 10^{-4}$ M.				Webb et al., 1979

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE
Chlordaneform	- <u>Rana nigromaculata</u>		rectus abdominis muscle	Chlordaneform and its metabolites caused a slow contraction at $10^{-6}$ - $10^{-3}$ M. Chlordaneform and 1 metabolite strongly inhibited the acetylcholine-induced contraction of the muscle whereas 2 other metabolites were inactive to the ACh-induced contraction at $10^{-3}$ M.		Watanabe et al., 1976
			adults 20 g	i.p. injection of 300 mg/kg chlordaneform produced 80% mortality within 24 hr.		
	- <u>Rana pipiens</u>		sciatic nerve - sartorius muscle	0.1 $\mu$ M suppressed amplitude of spontaneous miniature end-plate potentials (45% of controls after 22-25 min) without changing their frequency. 1 $\mu$ M blocked miniature end-plate potentials completely. Resting membrane potential not affected.	Concluded that the block of neuromuscular transmission by chlordaneform is due primarily to a depression of the end-plate sensitivity to the transmitter.	Hong et al., 1975
	- <u>Rana catesbeiana</u> <u>R. nigromaculata</u>		rectus-abdominis muscle	$10^{-3}$ M caused a slow contraction and $10^{-5}$ - $10^{-3}$ M inhibited the acetylcholine-induced contraction in a noncompetitive manner. In <u>R. catesbeiana</u> $K^{+}$ -induced contraction was also inhibited in a noncompetitive manner.	Effect probably due to depression not only of sensitivity of endplate but also of excitability of cell membrane.	Watanabe et al., 1975

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE		
p-Chloroaniline	- <u>Xenopus laevis</u>	40 per treatment	embryo	All embryos in 100 ppm died within 3 weeks. Development in controls and $\leq$ 0.1 ppm was complete after 13 weeks; at this time 85% and 20% of animals held at 1 and 10 ppm, respectively, had completed larval development.	Exposure for 90 days.	Duspert, 1987		
Chlorpyrifos (OP)	- <u>Hyla regilla</u>		tadpoles 3 weeks old	Thermal tolerance significantly lowered:	24 hr exposure. Recommended dosage rate for mosquito control in California-50 ppb.	Johnson, 1980		
				Conc. (ppb)			Onset of Spasms ( $^{\circ}$ C)	
							Mean	Range
				Control			37.5	36.9-37.8
25	35.3	33.4-36.9						
50	34.1	30.8-36.2						
Chlorpyrifos-ethyl (OP)	- <u>Bufo boreas</u>	20	juveniles ( $\bar{x}$ snout-vent length 19.8 mm)	Exposure of hydrated toads to 30 ppb significantly lowered temperature tolerance.	23-24 $^{\circ}$ C; 24 hr exposure.	Johnson and Prince, 1976		
Chlorpyrifos-methyl (OP)	- <u>Bufo boreas</u>	20	juveniles ( $\bar{x}$ snout-vent length 19.8 mm)	Exposure of hydrated toads to 30 ppb significantly lowered temperature tolerance.	23-24 $^{\circ}$ C; 24 hr exposure.	Johnson and Prince, 1976		
Chromium (VI) (M)	- <u>Rana tigrina</u>	80 per treatment	tadpoles	All died within 72 hours at $\geq$ 2 ppm (lowest concentration tested). Abnormalities observed in pigmentation, tail fin and alimentary canal.	Tadpoles intolerant to levels permitted in irrigation water.	Abbasi and Sani, 1984		
Cismethrin (PY)	- <u>Xenopus laevis</u>		lateral-line sense organ	$1.5 \times 10^{-6}$ M for 1-3 hr caused repetitive activity.	Temp. 8-22 $^{\circ}$ C. Number of impulses per train and train duration increased with cooling.	Vijverberg et al., 1982		
			peripheral nerves	$5 \times 10^{-6}$ M caused repetitive activity within first 2 hr of exposure.				

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT*	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE
Cadmium (Cd <sup>2+</sup> ) (M)	- <u><i>Iriturus pyrrhogaster</i></u>		stomach mucous epithelial cells	Concentrations of 10 <sup>-7</sup> M to 10 <sup>-4</sup> M did not affect the electrical properties of the cells.		Kanno et al., 1978
	- <u><i>Xenopus laevis</i></u>		myelinated nerve fibres	Decreased permeability constant in K and Na systems; reversibly shifted Na activation curve in positive direction along potential axis.		Arhem, 1980
Copper (M)	- Frogs		embryos	In N/500,000, few tadpoles survived many days after leaving the spawn.		Dilling and Hesley, 1926
	- <u><i>Iriturus pyrrhogaster</i></u>		stomach mucous epithelial cells	10 <sup>-4</sup> M Cu <sup>2+</sup> decreased membrane potential to 67% of the control value. Effect of ion increased as concentration increased from 10 <sup>-7</sup> to 10 <sup>-4</sup> M. Slight changes in the effective membrane resistance and electrical coupling ratio.	Action antagonized by cysteine and acetylpenicillamine.	Kanno et al., 1978
	- <u><i>Bufo boreas</i></u>		larvae	3.7 mg/L Cu <sup>2+</sup> - all died within 12 hours. 0.02 mg/L - all metamorphosed.	Up to 61 days exposure.	Porter and Hekanson, 1976
	- <u><i>Xenopus laevis</i></u>		myelinated nerve fibres	Slowed down the kinetics of the K system; decreased permeability constant in K and Na systems; reversibly shifted Na activation curve in a positive direction along potential axis.		Arhem, 1980

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT*	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE															
Copper (cont'd)	- <u><i>Rana pipiens</i></u>		rectus abdominus muscle	10 <sup>-6</sup> M Cu had little effect; 10 <sup>-6</sup> M Cu + 10 <sup>-5</sup> ACh caused larger contractions than ACh alone. Exposure to Cu + ACh for 10-15 min resulted in spontaneous spasmodic contractions.		Miller and Mackay, 1983															
	- <u><i>Rana temporaria</i></u>	500 per treatment	tadpoles just after hatching; after completion of operculum	Effect of Misdren 50 on tadpoles: <table border="1" data-bbox="582 1451 1059 1934"> <thead> <tr> <th>Conc. (suspension)</th> <th>Exposure (days)</th> <th>Age</th> <th>Result</th> </tr> </thead> <tbody> <tr> <td>0.05%</td> <td>3</td> <td>Young</td> <td>Inhibition of growth, accumulation of pigment in liver and stomach cells. Most died; after transferred to tap water, some of survivors returned to normal.</td> </tr> <tr> <td>0.05%</td> <td>5</td> <td>Young</td> <td>Stoppage of growth, malformations.</td> </tr> <tr> <td>0.01% and 0.05%</td> <td>3 and 5</td> <td>Old</td> <td>Inhibition of growth usually leading to death. After transferred to tap water all surviving tadpoles died within 3 days.</td> </tr> </tbody> </table>	Conc. (suspension)	Exposure (days)	Age	Result	0.05%	3	Young	Inhibition of growth, accumulation of pigment in liver and stomach cells. Most died; after transferred to tap water, some of survivors returned to normal.	0.05%	5	Young	Stoppage of growth, malformations.	0.01% and 0.05%	3 and 5	Old	Inhibition of growth usually leading to death. After transferred to tap water all surviving tadpoles died within 3 days.	Static tests.
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Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS				REMARKS	REFERENCE
				Conc. %	pH	No. of frogs surviving			
		6 days	30 days						
Copper Sulfate	- <u>Rana pipiens</u>							Frogs in high concentrations increased their cutaneous output of mucus. Temp. 20°C.	Kaplan and Yoh, 1961
		18		0.1	4.9	0	0		
		16		0.025	5.2	0	0		
		11		0.005	5.3	3	0		
		12		0.0025	5.6	2	0		
		17		0.0015	5.6	17	10		
		8		0.001	5.5	8	8		
		8		0.0005	5.6	8	8		
	8		0.0003	5.7	8	8			
	8		0.0001	5.6	8	8			
	- <u>Rana pipiens</u>		eggs, newly-hatched tadpoles	Fertilized eggs were not affected by 0.04-1.56 mg/L. 0.31 mg/L or more was fatal to tadpoles. Heights of tadpoles grown in 0.06 or 0.16 mg/L were lower than controls.				Landé and Gutman, 1973	
	- <u>Rana cyanophlyctis</u>	15 per treatment	adults	Progressive reduction in total number blood cells following i.m. injections of 100 and 200 µg CuSO <sub>4</sub> for 40 days. White blood cells increased for first 7 days then gradually decreased. Haemoglobin content reduced.				Frogs sacrificed at end of 7th, 14th, 30th, 40th day.	Patil and Shivaraj, 1984

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS		REMARKS	REFERENCE
				oil (ml/L)	% embryonic mortality		
Crude Oil	- <u>Rana arvalis</u>		embryos			Skeleton deformities and edema of abdominal cavity noted in hatched larvae.	Pyastolova and Danilova, 1986
		363		control	30.5		
		294		0.25	28.7 <sup>**</sup>		
		253		0.50	33.9 <sup>**</sup>		
		322		0.75	69.2 <sup>**</sup>		
		240		1.00	60.8 <sup>**</sup>		
		40 per treatment	larvae just post-hatch	* all hatched larvae died within a few days. ** 88% of hatched larvae died within first week. All exposed to 0.25 and 0.5 ml/L died within 20 days; 35% and 75%, respectively, had anomalies. Reduction in growth noted at 0.15 ml/L.			
		30 per treatment	larvae rear limb buds present	All exposed to 0.5 ml/L died within 22 days; those in 0.05 ml/L delayed metamorphosis and anomalies found in juveniles.			
Cyanatryn (H)	- <u>Rana temporaria</u>	20 per treatment	tadpoles weight 40 ± 5 mg hind limb buds present	No effect at 0.2 or 2 ppm pure cyanatryn for 24 hr; at 20 ppm behaviour changed; lethargic, displayed spasmodic twitching and a tendency to remain at the water surface, did not feed.			Scorgie and Cooks, 1979
				Exposure to 20 or 200 ppm as slow release pellets led to behaviour similar to above; more active than controls for first 3 hr.			

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE
pyrethrin (PY)	- <u>Xenopus laevis</u>		lateral-line sense organ sciatic nerves	$5 \times 10^{-6}$ M induced long-lasting trains of nerve impulses. Up to $10^{-5}$ M for >24 hr did not cause repetitive activity after a single stimulus.		Vijverberg et al., 1982
4-D (H)	- <u>Rana temporaria</u>		tadpoles hind limb paddles or hind legs	No mortality or behavioural changes at concentrations up to 50 ppm for 48 hr.		Cooke, 1972
	- <u>Rana temporaria</u>		tadpoles	1 mg/L of the sodium salt and 2 mg/L of the diethylamine salt significantly inhibited metamorphosis. An addition to thyroxin in a conc. of 1-5 mg/L hinders the action of the hormone stimulating metamorphosis.	24 hr exposure.	Buslovich and Borushko, 1976
DDO (UL)	- <u>Rana pipiens</u>		abdominal skin	$2 \times 10^{-4}$ M caused a decrease in skin resistance and an increase in the short-circuit current.	Suggests that DDO increased the sodium permeability of the outer barrier.	Webb et al., 1979
DDT (DC) (see also metacid)	- Frog		single muscle fibres	Membrane hyperpolarization reduced and delayed in $K^+$ -free medium in presence of $5.10^{-6}$ .	Action pH dependent.	Cracium and Agricoraei, 1978

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE
DDT (cont'd)	- <u>Rana ridibunda</u>		sartorius muscle single fibres	$5.10^{-4}$ prevented $Ca^{+2}$ binding in the globular phospholipidic micelle and facilitated $Ca^{+2}$ removal from the lamellar phospholipidic micelle.		Cracium et al., 1981
	- <u>Rana pipiens</u>	50 - Controls; 50 - Exp.		Injection with 0.2 ml DDT dissolved in alcohol to 100 ppm based on body weight caused increased activity (violent leg extensions) after 6 hr; increase maintained until death.	Controls injected with 0.2 ml ethyl alcohol. Controls and experimental frogs exhibited low activity levels for first 6 hr.	Isaacson, 1968
	- <u>Xenopus laevis</u>		lateral-line organ	Marked repetitive activity after exposure to 2-5 ppm for 18 hr.	Negative temperature coefficient of activity. Temp. 19-21°C.	Akkerma et al., 1975a
			cutaneous touch receptors	Showed short trains of spikes in response to a single mechanical stimulus after treatment with $10^{-5}$ M for 30-90 min.	Slight increase in number of repetitive spikes when temperature lowered from 19-21°C to 10-15°C.	
	- <u>Xenopus laevis</u>		myelinated nerve fibre from sciatic nerve	$4 \times 10^{-5}$ M induced a sodium tail current which was similar to that induced by exposure to allethrin.	Temp. $15 \pm 1^\circ C$ . Suggests that allethrin and DDT share common mechanism of action.	van den Bercken and Vijverberg, 1980

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE
DDT (Cont'd)	- <u>Bufo arenarum</u>	10 expt., 10 control	tadpoles 31 days	Activity of beta-glucuronidase in tail fin treated with 1 ppm DDT (mean + SE, $\mu\text{mol/g/min}$ ): Treated = $0.02694 \pm 0.00289$ Ctrl = $0.00485 \pm 0.00063$ . Plasma levels of thyroid hormones similar in 2 groups.	Exposed from fertilization to 31 days, medium replaced daily. Activity in treated toads significantly greater than that in controls.	Juarez and Guzman, 1986
	- <u>Bufo arenarum</u>		embryos	5 and 15 ppm produced 100% mortality on day 16 and 12, respectively. 1 ppm reduced time to metamorphosis.	Exposed from fertilization, medium replaced daily. Room temp.	Juarez and Guzman, 1984b
	- <u>Rana tigrina</u>	6 per group	adults $80 \pm 5$ g	Exposure to 0.1, 0.2 and 0.3% solutions caused decreased vitamin A stores in liver; greater decrease with increase in time of exposure (24-96 hr). Increased serum vitamin A levels with increased DDT noted.	Starved for 24 hr before exposure.	Keshavan and Deshpande, 1984

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE			
DDT (cont'd)	<u>Rana pipiens</u>	20 per group	tadpoles at limb bud stage	Tail regeneration (mm; mean + S.E.):					Weis, 1975
				Exposure (days)	Controls	5 ppb	25 ppb		
				5	$1.3 \pm .07$	$1.2 \pm .05$	$0.9 \pm .04^*$		
				8	$2.7 \pm .21$	$1.8 \pm .08^*$	$1.6 \pm .14^*$ (60%)**		
				12	$4.6 \pm .5$	$2.6 \pm .1^*$ (30%)	2.5 (95%)		
				<u>R. catesbeiana</u>	10 per group	tadpoles at limb bud stage	5		
	7	$2.9 \pm .14$	$2.8 \pm .16$ (30%)				$2.3 \pm .12^*$ (30%)		
	9	$4.5 \pm .21$	$4.5 \pm .22$ (30%)				$3.8 \pm .09^*$ (30%)		
	- <u>Rana hexadactyla</u>			gastrocnemius muscle	Soaking in 20-80 ppm for 10 min caused significant decrease in shortening length, twitch duration, half contraction time, half relaxation time.		Rajendra et al., 1980		
					* Significantly different from controls. ** % mortality.				



Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE																									
DDT (cont'd)	- <u>Rana temporaria</u>		spawn	Fresh spawn exposed to 0.5 nominal ppm for 24 hr 5 days prior to hatch were hyperactive 8-13 days after hatching. Development and weight gain were retarded.	During tail resorption small frogs susceptible to tissue residues as DDT released from the fat.	Cooke, 1972																									
			tadpoles	Tadpoles most susceptible either just before or just after developing hind limb buds and at these and later stages they became hyperactive when tissues contained 2-4 ppm.  Toads were more resistant than frogs at all developmental stages.  Some toads survived despite tissue concentrations >300 ppm.  Abnormal snouts noted in frogs treated with 0.02 or 0.5 ppm for 48 hr.																											
	- <u>Rana temporaria</u> <u>Bufo bufo</u>																														
	- <u>Lasiurus vulgaris</u>		tadpoles snout-anus 10-12 mm	<table border="1"> <thead> <tr> <th>Conc. (nominal ppm)</th> <th>Exposure (hr)</th> <th>Mortality %</th> <th>Behaviour</th> </tr> </thead> <tbody> <tr> <td>0</td> <td>24</td> <td>0</td> <td>normal</td> </tr> <tr> <td></td> <td>48</td> <td>0</td> <td>normal</td> </tr> <tr> <td>0.005</td> <td>24</td> <td>0</td> <td>normal/frantic</td> </tr> <tr> <td></td> <td>48</td> <td>0</td> <td>frantic</td> </tr> <tr> <td>0.5</td> <td>24</td> <td>10</td> <td>moribund</td> </tr> <tr> <td></td> <td>48</td> <td>33</td> <td>moribund</td> </tr> </tbody> </table>			Conc. (nominal ppm)	Exposure (hr)	Mortality %	Behaviour	0	24	0	normal		48	0	normal	0.005	24	0	normal/frantic		48	0	frantic	0.5	24	10	moribund	
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Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE
DDT (cont'd)	- <u>Rana sylvatica</u>	50 per treatment except 30 for stage 20 embryos	embryos	No abnormal effects on morphology or hatching success of embryos in stages 15, 16 or 18 exposed to 0.025 ppm for 24 hr; signs of DDT poisoning observed in embryos treated at stage 20.		Licht, 1985
	- <u>Rana temporaria</u>	160	tadpoles weight 363 ± 11 mg, snout-anus 10.5 - 12.5 mm	29% of frogs treated with 0.1 ppm DDT for 48 hr developed snout abnormalities; 3% died.  Stages of poisoning: I - skin has rough appearance; II - appearance of hole in centre of snout; III - loss of mandible and subsequent metamorphosis halted.	All affected individuals that reached tail resorption stage died. 2 factors caused snout abnormality: - disruptive effect on development of skin glands above upper mandible - lower mandible striking inner surface above upper due to hyperactivity.	Osborn et al., 1981
	- Frogs, toads			6 of 13 animals which ate insects poisoned by 10 % DDT spray died.		Lozier, 1949
	- <u>Rana temporaria</u>	24	adults weight 13-31 g, snout-urostyle 50-67 mm	Frogs subcutaneously injected exhibited sluggishness, irritability, extension and trembling of hind limbs, abnormal posturing, colour change and an increase in croaking. Treated frogs did not hide as often as controls. No significant changes were noted in food consumption.	These poisoning symptoms may increase predation in the field.	Cooke, 1974

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE
DDT (cont'd)	- <u>Rana temporaria</u> <u>triturus cristatus</u>			Treated tadpoles became hyperactive. Nests made significantly more lungee at hyperactive tadpoles than at controls. Accumulation of DDT in nests had no visible effect.	Possibly more predation of tadpoles by nests in contaminated areas due to altered behaviour of frogs. Nests may be more resistant than frogs.	Cooke, 1971
	- <u>Rana temporaria</u>	160 large; 160 small	tadpoles with external gills ̄ weight (mg): large - 6.89 ± 46 small 3.63 ± 11	Exposure to 0.1 ppm for 2 days: - no toxic effects in large tadpoles. - hyperactivity, tendency to float near surface, deformation in small tadpoles	Small tadpoles had higher residues.	Cooke, 1979
	- <u>Rana tigrina</u>		pancreas  serum	Amylase activity decreased significantly in frogs exposed to 0.2 and 0.3 % for 24-96 hr; no effect at 0.1% exposure. Lipase activity increased significantly in frogs exposed to 0.1, 0.2, 0.3% for 24-96 hr.  Amylase activity increased significantly in frogs exposed to 0.2, 0.3 % for 24-96 hr; no effect at 0.1% exposure. No effect on lipase activity at any concentration.		Deshmukh and Keshavan, 1987

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE	
DDT (cont'd)	- <u>Rana temporaria</u>		Tadpoles	Mortality of tadpoles exposed to various concentrations of DDT:  Normal Concentration (ppm)   Mortality (%)	Temp. 20 ± 5°C. Exposure - 1 hr. Length of expt. - 14 days.	Cooke, 1970	
		40 per group	hind limb bud stage; snout-anus- 6.5 - 9.5 mm	0.0 0.01 0.1 1.0 10.0			< 3 0 0 20 70
		40 80 80	small-medium hind legs; snout-anus- 10-12 mm	0.0 1.0 10.0			< 3 10 50
	- <u>Rana pipiens</u>		abdominal skin	2 x 10 <sup>-4</sup> M did not produce changes in short circuit current or resistance.		Webb et al., 1979	
	- Frogs		adulte	All frogs treated with 150 mg/kg dissolved in olive oil and injected into the dorsal lymph sac died in 4-72 hr. Some died following injection of 10 mg/kg.		Ellis et al., 1966	
DDVP (DP) (phosphoric acid 2,2 - dichlorovinyl dimethyl ester)	- Frog		sartorius muscle	Inhibition of membrane voltage response of muscle to carbamylcholine which was dependent upon concentration and temperature.		Dekin et al., 1978	

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE
fenuron (H)	- <u>Rana temporaria</u>	20 control; 20 expt.	tadpoles 2 days old	Delayed development found in tadpoles kept in 100 ppm for 80 days: controls - developed forelegs at 30 days, metamorphosed by 40 days. experimental - developed forelegs at 50 days, only 40% underwent complete metamorphosis, 40% developed into giant tadpoles without forelegs, 20% died.		Paulov, 1977
EHP Di-2-ethylhexyl- nthalate)	- <u>Rana arvalis</u>	Approx. 100 eggs per treatment	2-3 day old embryos	Hatching success decreased with increasing levels DEHP; approximately 50% hatched when exposed to sediments with 150 mg/g fresh weight. Survival rates of tadpoles did not differ.	DEHP added to sediment of model system. No abnormalities in development, no delays in hatching observed.	Larsson and Thurén, 1987
deltamethrin (PY)	- <u>Xenopus laevis</u>		lateral-line sense organ  sciatic nerves	Produced long trains of impulses <u>in vitro</u> at $1-5 \times 10^{-6}$ M for 3 hr.  Up to $10^{-5}$ M for more than 24 hr did not induce repetitive activity after a single stimulus.		Vijverberg et al., 1982
	- <u>Rana sacculenta</u>		skin	Under open circuit conditions, $10^{-6}$ M did not provoke changes in $Na^+$ fluxes; at $10^{-5}$ M a slight inhibition of the $J(a)Na^+$ after 30 min noted, no change in $Cl^-$ fluxes.		Salibian, 1983
	- <u>Xenopus laevis</u>		myelinated nerve fibres	$10^{-6}$ M caused a frequency dependent depression of the action potential which was associated with a progressive membrane depolarisation brought about by summation of depolarising after-potentials.	Specifically affects the sodium channels of the nerve membrane.	Vijverberg and van den Bercken, 1979

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE														
DDP (OP) (diisopropyl fluorophosphate)	- <u>Bufo viridis</u>	3	adulte  blood nervous tissue	Residual cholinesterase at time of death following injection of 2 LD50 into dorsal lymphatic sac:  20% 64%	LD50 - 1450 ppm.	Ebery and Schatzberg- Paroth, 1960														
Dichlofenthion (OP)	- <u>Rana pipiens</u>		nerve-muscle	Only a small depressive effect on muscle contraction evoked by direct or indirect stimulation at $1 \times 10^{-3}$ M. No effect at lower concentrations.	Temp. 22°C.	Iakeno et al, 1977														
Dichloro (F,H)	- <u>Xenopus laevis</u>	30	embryos tail-bud stage	<table border="1"> <thead> <tr> <th>Concentration (ppm)</th> <th>24 hr Survival</th> </tr> </thead> <tbody> <tr> <td>control</td> <td>100%</td> </tr> <tr> <td>0.075</td> <td>100%</td> </tr> <tr> <td>0.1</td> <td>95%</td> </tr> <tr> <td>0.125</td> <td>50-55%</td> </tr> <tr> <td>0.15</td> <td>15-20%</td> </tr> <tr> <td>&gt;0.2</td> <td>0%</td> </tr> </tbody> </table> <p>* taken from mortality curve</p>	Concentration (ppm)	24 hr Survival	control	100%	0.075	100%	0.1	95%	0.125	50-55%	0.15	15-20%	>0.2	0%	Overall inhibition of growth and development; 0.1 - 0.15 ppm disrupted development of cephalic end of embryo.	Anderson and Prahlad, 1976
Concentration (ppm)	24 hr Survival																			
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0.15	15-20%																			
>0.2	0%																			
Dicrotophos (OP)	- <u>Rana catesbeiana</u>		tadpoles	All tadpoles exposed to 5 ppm survived.	Continuous flow apparatus for 96 hr. Brain cholinesterase activity of Hellards fed dosed tadpoles was 70.1% of that of controls. None died.	Hall and Kolbe, 1980														
Dieldrin (HEOD) (DC)	- <u>Rana pipiens</u>		adulte 65 g, 8.9 cm long	<table border="1"> <thead> <tr> <th>Dieldrin (ppm)</th> <th>pH of solution at 25° C</th> <th>No. of frogs dead at 30 days</th> </tr> </thead> <tbody> <tr> <td>0.10</td> <td>5.77</td> <td>10</td> </tr> <tr> <td>0.08</td> <td>5.61</td> <td>0</td> </tr> <tr> <td>0.05</td> <td>5.59</td> <td>0</td> </tr> </tbody> </table>	Dieldrin (ppm)	pH of solution at 25° C	No. of frogs dead at 30 days	0.10	5.77	10	0.08	5.61	0	0.05	5.59	0	Frogs placed in 200 cc of test solution. Neuro-muscular changes produced; excessive thrashing and abnormal reactivity to stimulation were observed.	Kaplan and Overpeck, 1964		
Dieldrin (ppm)	pH of solution at 25° C	No. of frogs dead at 30 days																		
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Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE	
Dieldrin (cont'd)	- <u>Rana esculenta</u> <u>R. pipiens</u> <u>R. temporaria</u>	Approx. 150 eggs per treatment	motor end-plate	$10^{-5}$ to $10^{-4}$ M caused no significant effects on frequency and amplitude of miniature end-plate potentials, or on end-plate membrane potential.	Temp. $10 \pm 2^\circ\text{C}$ .	Akkermans et al., 1976	
	- <u>Xenopus laevis</u>		spinal chord	Application of up to $5 \times 10^{-5}$ M <u>in vitro</u> failed to produce any significant effects.		Akkermans et al., 1975c	
	- <u>Rana temporaria</u>		tadpoles	Treatment with 0.5 mg/ml ppa produced 5% mortality and frantic/resigned behaviour after 24 hr, 47% mortality after 48 hr. Snout abnormalities noted. 0.02 ppm had no effect.		Cooke, 1972	
	<u>Bufo bufo</u>		tadpoles	Exposure to 0.5 ppm for 48 hr caused no mortality; resigned behaviour and snout abnormalities noted.			
	- <u>Limnodynastes tasochensis</u>		embryos	Mortality, otic capsule and cephalic pigmentation abnormalities after exposure to 0.1 ppm for 7 hours; accelerated growth relative to controls. No effects with exposure to 0.01 ppm.		Observed abnormalities resemble those described for tadpoles exposed to chlorzhal.	Brooks, 1981
	- <u>Rana pipiens</u>		abdominal skin	$2 \times 10^{-4}$ M caused significant increases in the short circuit current while decreasing the resistance.		Suggests altered the sodium permeability of the outer barrier.	Webb et al., 1979
	- <u>Xenopus laevis</u>		lateral-line organ	Treatment with $5 \times 10^{-4}$ M for several hr produced no significant effect. Preparations taken from animals showing severe symptoms of poisoning after exposure to 5-5 ppm dieldrin did not show any sign of repetitivity.			Akkermans et al., 1975a
		1974	cutaneous touch receptors	No sign of repetitive activity. No effects of temperature changes noted.			

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE																				
Dieldrin (cont'd)	- <u>Bufo arenarum</u>	3 per treatment	embryos	Embryos developed in 0.2 and 2.0 mg/L died between 20-25 days; 25-30 % of those developed in 0.02 mg/L had front legs earlier than controls.	All determinations made on 11 day embryos. Values are mean $\pm$ SE.	de Lijnes et al., 1985																				
				<table border="1"> <thead> <tr> <th rowspan="2">Treatment</th> <th rowspan="2">Time Exposed</th> <th colspan="2">% Inhibition</th> </tr> <tr> <th>ADChE</th> <th>ChE</th> </tr> </thead> <tbody> <tr> <td rowspan="2">Unfertilized oocyte + 2 mg/L</td> <td>1 hr</td> <td>19.7 <math>\pm</math> 1.8</td> <td>17.2 <math>\pm</math> 1.7</td> </tr> <tr> <td>4 hr</td> <td>22.3 <math>\pm</math> 1.4</td> <td></td> </tr> <tr> <td rowspan="2">Recently fertilized oocyte + 2 mg/L</td> <td>4 hr</td> <td>22.7 <math>\pm</math> 1.2</td> <td>25.9 <math>\pm</math> 1.3</td> </tr> <tr> <td>continuous</td> <td>26.7 <math>\pm</math> 1.4</td> <td>25.5 <math>\pm</math> 1.8</td> </tr> </tbody> </table>			Treatment	Time Exposed	% Inhibition		ADChE	ChE	Unfertilized oocyte + 2 mg/L	1 hr	19.7 $\pm$ 1.8	17.2 $\pm$ 1.7	4 hr	22.3 $\pm$ 1.4		Recently fertilized oocyte + 2 mg/L	4 hr	22.7 $\pm$ 1.2	25.9 $\pm$ 1.3	continuous	26.7 $\pm$ 1.4	25.5 $\pm$ 1.8
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Dimefox (DF)	- <u>Bufo viridis</u>	3	adults	Residual cholinesterase at time of death following injection of 2 LD50 into dorsal lymphatic sac: 0% 51%	LD50 - 1410 ppm.	Eder and Schatzberg-Poreth, 1960																				

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT*	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS						REMARKS	REFERENCE
				Conc. (%)	Development of Eggs			Dvpt. of Tadpoles			
			Time to metamorphosis (days)		Av. length at metamorphosis (mm)	No. Died	Time to metamorphosis (days)	Av. length at metamorphosis (mm)			
methoate (OP)	- <u>Rana tigrina</u>	60	eggs tadpoles pre-limb, 29 mm	Ctrl	35	16.8±0.96	0	20	16.0±0.74	Temp. 30-38°C.  Recommended dosage for spraying is .04%, higher than concentrations causing development delays.	Dutta and Mohanty - Hejmadi, 1978
	0.0001		46	16.0±0.63	0	61	16.0±0.74				
				0.0002	54	16.0±0.02	0	64	16.0±0.54		
				0.0003	62	16.0±0.71	3	64	16.0±0.74		
				0.0004	62	16.0±0.74	3	64	15.0±0.54		
				0.0005	60	15.0±0.52	4	64	15.0±0.74		
				0.0006	65	14.5±0.52	4	64	15.0±0.54		
				0.0007	60	14.0±0.54	5	64	14.0±0.54		
				0.0008	63	13.5±0.57	6	67	14.0±0.74		
				0.0009	54	13.5±0.57	6	70	14.0±0.54		
				No eggs treated with 0.001 - 0.009% reached metamorphosis.							
	- <u>Bufo melanostictus</u>	60	tadpole tails	After 7 days in 0.05 ppm melanophore size increased significantly, puncta-stellata melanophores were visible and developed dendritic processes and branching.							Pandey and Jaiswal, 1985

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT*	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS				REMARKS	REFERENCE						
				Conc. %	% metamorphosed		Mean time for metamorphosis (days)								
			egg		limb bud	egg	limb bud								
methoate (cont'd)	- <u>Rana tigrina</u>	60	eggs, limb bud stage tadpoles	0.001	--	80	--	59	Temp. 30-38°C.  Mean length at metamorphosis reduced at 0.0008 - 0.0009% (eggs) and 0.007 - 0.001% (tadpoles).	Mohanty-Hejmadi and Dutta, 1981					
				0.0009	40	100	54	70							
				0.0008	40	100	63	67							
				0.0007	50	100	60	64							
				0.0006	60	100	65	64							
				0.0005	60	100	60	64							
				0.0004	70	100	62	64							
				0.0003	70	100	62	64							
				0.0002	100	100	54	64							
				0.0001	100	100	46	64							
				Control	100	100	35	18							
				dimethylhydrazine compound used in jet and rocket fuel)	- <u>Xenopus laevis</u>	100 100 100 100 100 100	embryo				conc. (mg/L)	% malformed	Kinked tail was most frequent malformation. Most larvae with kinked tails metamorphosed into normal frogs. Other malformations included microcephaly, failure of elongation and edema.	Greenhouse 1976b	
								1,2-dimethylhydrazine			control	3			
		10	4												
		20	4												
		40	5												
		50	100												
		80	100												
1,1-dimethylhydrazine		control	3												
		1	3												
		2	10												
		5	50												
		10	67												
		20	100												
Susceptible during neurulation.															

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE																								
Dimethylhydrazine (cont'd)	- <u>Xenopus laevis</u>	5-69 per treatment	larvae	1 mg/L 1,2 - DMH lethal to 100% larvae within 7 days; continuous contact with 0.1 mg/L allowed normal metamorphosis. Exposure to $\leq 1$ mg/L 1,1 - DMH did not affect survival or metamorphosis; higher concentrations lethal.		Greenhouse 1976a																								
		50-100 per treatment	embryos	$\leq 40$ mg/L 1,2 - DMH not toxic if embryos transferred to uncontaminated water prior to hatching; $\geq 50$ mg/L teratogenic. $\geq 5$ mg/L 1,1 - DMH teratogenic.	Failure to elongate, tail kinks most common abnormalities.																									
2,4-Dinitrophenol	- <u>Rana temporaria</u>		cutaneous pectoris nerve-muscle	$10^{-4}$ M caused rise in miniature endplate potential frequency 30 min after application following reduction in first 20 min.		Stethem et al., 1978																								
Diacyldiphenylamine (component of jet fuel)	- <u>Rana pipiens</u> - <u>Xenopus laevis</u>	200	embryo larvae	Up to 1 g/L not toxic to either species.	No deleterious effects on development. Controls had no malformed embryos.	Greenhouse, 1976a																								
Diquat (M)	- <u>Rana pipiens</u>	500 per treatment	eggs	After exposure to 100 ppm for 21 days eggs exhibited reduced viability and increased exogastrulation. None survived beyond 14 days post-hatch.	Temp. 18°C.	Bieber and Mitchell, 1978																								
	- <u>Rana pipiens</u>	200 per dose group	embryos; early gastrula and 15 days of ego	5% alive 16 days post-treatment after exposure at gastrula stage (G) or 15 days of ego (15): Diquat (mg/L)	No abnormalities observed in embryos.	Dial and Dial, 1987																								
				<table border="1"> <thead> <tr> <th colspan="2">0</th> <th colspan="2">2</th> <th colspan="2">5</th> <th colspan="2">10</th> </tr> <tr> <th>G</th> <th>15</th> <th>G</th> <th>15</th> <th>G</th> <th>15</th> <th>G</th> <th>15</th> </tr> </thead> <tbody> <tr> <td>81.8</td> <td>90.0</td> <td>79.5</td> <td></td> <td>70.9*</td> <td></td> <td>30.1*</td> <td>78.3</td> </tr> </tbody> </table>	0		2		5		10		G	15	G	15	G	15	G	15	81.8	90.0	79.5		70.9*		30.1*	78.3		
0		2		5		10																								
G	15	G	15	G	15	G	15																							
81.8	90.0	79.5		70.9*		30.1*	78.3																							
				* significantly different from controls.																										

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE																
Diquat (cont'd)	- <u>Xenopus laevis</u>	50	embryo tail-bud stage	<table border="1"> <thead> <tr> <th>Concentration (ppm)</th> <th>48 hr Survival*</th> </tr> </thead> <tbody> <tr> <td>control</td> <td>100%</td> </tr> <tr> <td>0.5</td> <td>95%</td> </tr> <tr> <td>0.75</td> <td>90-95%</td> </tr> <tr> <td>1.0</td> <td>70%</td> </tr> <tr> <td>1.25</td> <td>75%</td> </tr> <tr> <td>1.5</td> <td>70%</td> </tr> <tr> <td>2.0</td> <td>20%</td> </tr> </tbody> </table>	Concentration (ppm)	48 hr Survival*	control	100%	0.5	95%	0.75	90-95%	1.0	70%	1.25	75%	1.5	70%	2.0	20%	0.75 - 2.0 ppm reduced body size and pigmentation; 1.5 ppm produced some distortion in body shape.	Anderson and Prahled, 1976
Concentration (ppm)	48 hr Survival*																					
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1.5	70%																					
2.0	20%																					
				* taken from mortality curve.																		
Diquat/Nabam (M)	- <u>Xenopus laevis</u>	50	embryo tail-bud stage	0.75 - 1.25 ppm diquat/2.0 ppm nabam produced 100% mortality within 6 days.	Size of larvae reduced, melanin synthesis retarded, muscle fibres less developed.	Anderson and Prahled, 1976																
Distillery Effluent	- <u>Rana sylvatica</u>	5 per treatment	tadpoles pre-hind limb emergence	Increase in concentration (0.03, 0.06, 0.12%) reduced period of limb bud emergence and tail resorption; elevated values for length of limbs, tail and body wt. of adults.		Haniffa et al., 1985																
Edrophonium	- <u>Rana pipiens</u>		nerve-muscle	Neuromuscular transmission suppressed at $5 \times 10^{-4}$ and $1 \times 10^{-3}$ M. Contraction evoked by muscle stimulation also suppressed. No effect at $1 \times 10^{-4}$ M.	Temp. 22°C.	Iakono et al., 1977																

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS			REMARKS	REFERENCE
				Endrin (ppm)	pH of solution at 25°C	No of frogs dead at 30 days		
Endrin (OC)	- <u>Rana catesbeiana</u>	10	adulte	Frogs force-fed mosquitofish which had been exposed to 2 ppm endrin solution for 7 days: - 100% mortality - Survival time 15.6 ± 5.03 (x̄ ± SD) - Ratio predator: prey mean weight 418:1.			Resistant mosquitofish tolerate endrin levels that will kill predators many times their own weight.	Rosato and Ferguson, 1968
	- <u>Rana pipiens</u>		abdominal skin	2 × 10 <sup>-4</sup> M produced no significant effects on the short circuit current or resistance.				Webb et al., 1979
	- <u>Rana pipiens</u>		adulte 65 g, 8.9 cm long	Endrin (ppm)	pH of solution at 25°C	No of frogs dead at 30 days	Frogs placed in 200 cc of test solution. Neurovascular changes produced; excessive thrashing and abnormal reactivity to stimulation were observed.	Kaplan and Overpeck, 1964
		20		0.01	5.86	6		
		10		0.02	5.80	0		
	10		0.015	5.78	0			
Ethanol	- <u>Rana temporaria</u>	10 per series	tadpoles 8 days old	At 1000 ppm, all died within 10 min; at 900 ppm, tadpoles survived for up to 1 day; ~800 ppm had no effect.				Paulov, 1987b
		100	tadpoles	The body weight of tadpoles kept in 500 ppm from day 7 to day 22 increased compared to controls; onset of metamorphosis accelerated by up to 8 days; all died.				
		10	adulte blood, muscles	6 day exposure to 1000 ppm led to enhanced enzymatic activities of aspartate aminotransferase and alanine aminotransferase.				

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS			REMARKS	REFERENCE
				Conc. (ppm)	% dead at 96 hr	% abnormal at 96 hr		
Ethylenethiourea (degradation product of ethylenebis-dithiocarbamate fungicides)	- <u>Microhyla ornata</u>	50 per treatment	embryos gastrula stage	0.0	0.0	0	Abnormalities included wavy notochord, abnormal pigmentation and movement.	Chate, 1986
				5.0	0.0	22		
				10.0	2.0	100	Effects similar to those induced by dithiocarbamates.	
				20.0	10.0	100		
				30.0	10.0	100		
				40.0	30.0	100		
				50.0	80.0	100		
Ethylenethane-sulfonate	- <u>Rana pipiens</u>		narrow cells	7-fold increase in sister chromatid exchanges at 0.1 mg/ml in ambient water. Mitotic index reduced and cell cycling time lengthened in dose-dependent manner.			Geard and Soutter, 1986	
	- Frog		eye lens epithelium	Increase in pre-mitotic, G2, population and decrease in mitosis; micronucleated cells present in frogs immersed in 25-200 ug/L for 1 week.			Kung et al., 1987	
Eulan Ma New (mothproofing agent)	- <u>Rana temporaria</u>	20 per concentration (x 4)	tadpoles weight 21.0 ± 1.0 mg	Tadpoles exposed to 1.0 mg/L exhibited decreased movement for first 48 hr, slower development, abnormalities, decreased feeding rate and weight, 92.5% mortality. Tadpoles exposed to 0.1 mg/L showed slower development and no mortality.			Temp. range 17.5 - 15°C. Animals not fed during first 48 hr. Osborn and French, 1981	

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE		
Fenitrothion (OP)	<u>Rana ligorina</u>		tadpoles	Effects on feeding at age (F) and limb bud at age (L):	Temp. 30-38°C.	Mahony-Hajjaji and Dutta, 1981		
				Conc. % metamorphosed			Mean time for metamorphosis (days)	
					Mean length at metamorphosis reduced in limb bud tadpoles at 0.00004%.			
				F		L	F	L
				0.00004		20		53.0
				0.00005		60		54.3
				0.00002		60	51.0	54.6
				0.00001	40	100	59.6	
				Control	80	100	31.5	15.4
			eggs	Complete mortality at 0.00001%.				
	<u>Rana catesbeiana</u>	7 per treatment	larvae	Significant decrease in mean activity time recorded at 2 ppm but not at 1 ppm.	Temp. 21 ± 1°C. 24 hr acclimation.	Layne et al., 1976		
Fenprop (H)	<u>Litoria georgi</u> <u>Litoria denticulata</u> <u>Litoria tasmaniana</u>	20 per conc.	adults	No deaths observed in either species following exposure to 80-400 ppm for 96 hr.		Johnson, 1976		
Fenprothrin (PY)	<u>Xenopus laevis</u>		lateral-line sense organ	After 1 - 1.5 hr <i>in vitro</i> exposure to $5 \times 10^{-6}$ M trains of impulses recorded with mean of 530 impulses per train.		Vijverberg et al., 1982		
			sciatic nerves	Exposure to up to $10^{-5}$ M for more than 24 hr did not induce repetitive activity after a single stimulus.				
Fenthion (OP)	<u>Bufo boreas</u>	20	juveniles ( $\bar{x}$ snout-vent length = 19.8 cm)	Exposure of hydrated toads to 60 ppb significantly lowered temperature tolerance.	23-24°C; 24 hr exposure.	Johnson and Price, 1976		

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE		
Fenthion (cont'd)	<u>Hyla regilla</u>		tadpoles 3 weeks old	Thermal tolerance significantly lowered:	24 hr exposure. Recommended dosage rate for mosquito control in California - 100 ppb.	Johnson, 1980		
				Conc. (ppb)			Onset of Spasms (°C)	
							Mean	Range
				Control			37.5	36.9-37.8
				25			36.9	36.3-37.7
50	36.7	36.2-37.9						
100	35.7	31.9-37.2						
	<u>Rana catesbeiana</u>		tadpoles	All tadpoles exposed to 5 ppm survived. Bioconcentration factor: $\bar{x}$ 62.	Continuous flow apparatus for 96 hr. Brain cholinesterase activity of mallards fed dosed tadpoles was 16.9% of that of controls. All ducks died within 5 hr.	Hall and Kolbe, 1980		
Fenvalerate (PY)	<u>Xenopus laevis</u>		lateral-line sense organ	Acted slowly; frogs had to be treated <i>in vivo</i> with $5 \times 10^{-6}$ M for up to 23 hr before repetitive activity induced.		Vijverberg et al., 1982		
			sciatic nerves	Up to $10^{-5}$ M for more than 24 hr did not induce repetitive activity after a single stimulus.				
Furanace (fish bactericide)	<u>Rana pipiens</u>		larvae	Exposure to 0.2 - 20 ppm for 96 hr produced no mortality after 7 days. Many in 10 and 20 ppm were immobilized during exposure but recovered.		Marking et al., 1977		



Table 2 - Laboratory Studies (cont'd)

CONTAMINANT*	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE
DE (OC) ieldrin analogue)	- <u>Rana pipiens</u>		liver microsomes (adults)	Microsomal metabolism (apparent V <sub>max</sub> and K <sub>m</sub> values ± SE): App V <sub>max</sub> = 0.013 ± 0.001 nmol/min/mg App K <sub>m</sub> = 29 ± 8 nmol/mg.		Ronis and Walker, 1985
optachlor (OC)	- <u>Rana pipiens</u>		abdominal skin	2 × 10 <sup>-4</sup> M produced no significant effects on short circuit current or resistance.		Webb et al., 1979
	- <u>Bufo arenarum</u>		embryos	5 and 15 ppm produced 100% mortality on day 15 and 15, respectively. 1 ppm reduced time to metamorphosis.	Exposed from fertiliza- tion, medium replaced daily.	Juarez and Guzman, 1984b
exachlorophene (F) nd other biphenols	- Grass Frogs		blood	1.2 × 10 <sup>-6</sup> M produced 50% hemolysis in washed, nucleated erythrocytes; 1.60 × 10 <sup>-6</sup> produced 50% hemolysis in nucleated erythrocytes in whole blood.	Temp. 22 ± 1°C.	Flores and Buhler, 1974
-Hexane	- <u>Bufo arenarum</u>		embryos	5 and 15 ppm produced 100 % mortality on day 29 and 17, respectively. 1 ppm was not toxic.	Exposed from fertiliza- tion, medium replaced daily.	Juarez and Guzman, 1984b

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT*	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS		REMARKS	REFERENCE	
Hydrazine (component of jet fuel)	- <u>Xenopus laevis</u>	10-86 per treatment	larvae	Concentrations ≥ 1 mg/L produced irreversible toxic effects and death within 24-28 hr. 0.1 mg/L was not toxic to larvae.		Kinked tail was most frequent malformation. Most larvae with kinked tails metamorphosed into normal frogs. Other malformations included tail duplica- tion, microcephaly, failure of elongation and edema.	Greenhouse, 1976a	
		50-100 per treatment	embryos	Normal development in 5 mg/L but did not survive as larvae if not transferred to <1 mg/L at hatching. Malformations in some batches of eggs at 10 mg/L.				
		embryos	Concentration (mg/L)	% malformed	Greenhouse, 1976b			
	100 100 150 100 50 150		Control 1 10 25 50 100	2 3 55 100 100 100				
			Susceptible during neurulation.					
Hydrazine Sulfate (compound used in jet and rocket fuel)	- <u>Xenopus laevis</u>		embryos	Concentration (mg/L)	% malformed	Kinked tail was most frequent malformation. Most larvae with kinked tails metamorphosed into normal frogs. Other malformations included microcephaly, failure of elongation and edema.	Greenhouse, 1976b	
		100 100 200		Control 10 40	0 3 100			
				Susceptible during neurulation.				

Table 1 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE
Ionizing Radiation	- Assorted species/review		cells, tissues, organs, whole body	Effects include: suppressed mitotic activity of cells; formation of giant cells; formation of vacuoles; damage to nucleus; changes in skin epithelium and connective tissue; damage to spleen, CNS, liver, intestine, eyes; developmental abnormalities.		Brunat, 1965
	- <u>Caudivertora caudivertora</u>		larvae	Dose-effect relationship demonstrated between level of radiation (50-150 rads) and presence of micronucleated blood cells.	Exposure for 10 days.	Herasilla and Corrasco, 1985
Iron	- <u>Bufo boreas</u>		larvae	144 µg/L - all died within 24 hr. 30 µg/L - all died within 20 days. 20 µg/L - all metamorphosed.	Greater than 39 days exposure to Fe <sup>2+</sup> .	Porter and Hakanson, 1976
LAS (Linear chain alkylbenzenesulphonate)	- <u>Xenopus laevis</u>		embryo	Caused malformations.	No levels given.	Cotta Rebusino, 1980
Lead	- <u>Rana catesbeiana</u>		eye	5.0 and 12.5 µM Pb <sup>2+</sup> produced 9% and 20% decrease, respectively, in amplitude of rod response. No effect on cone potential.	Effect reversible.	Fox and Sillinen, 1979

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS							REMARKS	REFERENCE
				Hg (ppm)	Pb (ppm)							
Lead (cont'd)	- <u>Xenopus laevis</u>		embryos	10	1	.5	.1	.01	.001	Exposure for 6 days at 18°C.	Miller and Landeman, 1978	
				200	D	M	SL-M	N	N			
					0	99	88	93	88			
				20	D	M-S	M	N	N			N
					0	81	100	78	96			96
				2	D	M-S	M	N	N			N
					0	59	92	95	95			97
				.2	D	S	M-S	M-S	SL			SL
					0	73	98	98	95			93
					0	D	S	S	S			S
		0	39	100	74	96	82					
			D - Death; S - Severely deformed; M - Moderately deformed; SL - Slight effects; N - No effects; numbers = % survival from blastula to feeding stage.									
	- <u>Rana nigromaculata</u>		eggs, tadpoles	Exposure of eggs to 70 ppm resulted in partial reduction of primordial germ cells at the 9-12 mm body length stage. 70 ppm was lethal to tadpoles.							Treatment prior to the first cleavage.	Moh, 1978
Lead Acetate	- <u>Rana catesbeiana</u>	143	blood of tadpoles	25 ppm lead acetate or lead nitrate produced destruction of erythrocytes and necrosis of liver, spleen and intestinal mucosa; high mortality. Action of lead nitrate more rapid than action of lead acetate.								Barrett, 1987
	- <u>Xenopus laevis</u>		embryos	Exposure to ≥1 mg/L always produced lethality and/or abnormalities. Main anomaly in embryos exposed before neurula stage was unfused neural tube; in those exposed after neurula stage main anomaly was edema in thorax and kinky tail.								Kamimura and Iamsuro 1985

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE
Lead Acetate (cont'd)	- <u>Rana hexadactyla</u>		heart	Perfusion by $10^{-7}$ M for 20 min augmented digoxin-induced cardiotoxicity; reduction in total digoxin exposure and time taken for systolic arrest.	Exposure to $6.5 \text{ mM Ca}^{+2}$ attenuated this cardiotoxicity when prior to or simultaneously with Pb exposure.	Krishnamoorthy et al., 1987
	- <u>Necturus</u>	13	blood	Newts maintained in bath of 20 cc of 1% lead acetate and 4 L water suffered destruction of mature erythrocytes which stimulated differentiation and proliferation of erythrocytes in blood.		Dawson, 1955
	- Frogs		eggs, tadpoles, adults	1-5 ppm can partly or fully inhibit germination of eggs. Adults killed at 100 ppm. 2 ppm was toxic to tadpoles.	Tadpoles with external gills more sensitive than those with internal gills.	Dilling and Healey, 1926
Lead Nitrate	- Frogs		eggs, tadpoles, adults	1-5 ppm can partly or fully inhibit germination of eggs. Adults killed at 100 ppm. 2 ppm was toxic to tadpoles.	Tadpoles with external gills more sensitive than those with internal gills.  Lead salts had higher inhibitory effects on germination of frog ovum than zinc, thorium, copper, beryllium or thallium.	Dilling and Healey, 1926
	- <u>Rana esculenta</u>		leg skin	$10^{-5}$ M inhibited isometric, active transport-coupled volume flow.		Colentano et al., 1979

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS			REMARKS	REFERENCE
				Total Concentration Pb (ppm)	No. of frogs dead at 30 days			
Lead Nitrate (cont'd)	- <u>Rana pipiens</u>	24 per group	adults 9 cm, 65 g	Control 25 50 100 150 200 300	0 1 3 7 17 22 24		Temp. 8°C -sloughing of the integument -loss of postural tone -sluggishness -red and white cell counts decreased progressively in increasing lead concentrations -lead deposits in liver at 25 ppm.	Kaplan et al., 1967
	- <u>Rana catesbeiana</u>	143	blood of tadpoles	25 ppm lead acetate or lead nitrate produced destruction of erythrocytes and necrosis of liver, spleen and intestinal mucosa; high mortality. Action of lead nitrate more rapid than action of lead acetate.				
Leptophos (OP)	- <u>Rana pipiens</u>		nerve-muscle	No effect on muscle contraction evoked by indirect or direct stimulation at up to $2.5 \times 10^{-4}$ M.			Temp. 22°C.	Iakeno et al., 1977
Leptophosoxon (OP)	- <u>Hyla chrysoscelis</u>	embryos		Conc. (ppm)	Mortality %	Abnormal Embryos (%)	Dermally exposed for 24 hr, no. survivors determined 120 hr after exposure.	Fullon and Chambers, 1985
			76	0	1	0		
			75	1.0	0	0		
			76	2.5	34	0		
			80	5.0	85	0		
80	7.5	100	0					

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE																																																																	
Lindane (UC)	- <u>Rana pipiens</u>		ventral abdominal muscle	Initial rate of movement of lindane (10 ppm) through live or dead chain was 0.680 ppb/hr; at 8 hr, rates were 0.691 and 0.666 ppb/hr, respectively.	At 2 ppm 6 week old tadpoles showed up to 100% mortality.  Development from hatch to metamorphosis at 0.5 ppm was 4 weeks longer than controls; average weight lower in treated groups. Exposed tadpoles frequently had morphological abnormalities.	Kosow and Dunham, 1972-73																																																																	
	- <u>Xenopus laevis</u>		eggs	<table border="1"> <thead> <tr> <th rowspan="2">Trial</th> <th colspan="2">Control</th> <th colspan="3">Lindane</th> </tr> <tr> <th>Water</th> <th>Acetone</th> <th>0.5 ppm</th> <th>1 ppm</th> <th>2 ppm</th> </tr> </thead> <tbody> <tr> <td>I No. eggs</td> <td>30</td> <td>30</td> <td>30</td> <td>30</td> <td>30</td> </tr> <tr> <td>No. eggs hatched</td> <td>17</td> <td>19</td> <td>20</td> <td>17</td> <td>8</td> </tr> <tr> <td>II No. eggs</td> <td>30</td> <td>15</td> <td>45</td> <td>45</td> <td>45</td> </tr> <tr> <td>No. eggs hatched</td> <td>16</td> <td>11</td> <td>20</td> <td>24</td> <td>11</td> </tr> <tr> <td>III No. eggs</td> <td>75</td> <td>75</td> <td>75</td> <td>75</td> <td>75</td> </tr> <tr> <td>No. eggs hatched</td> <td>37</td> <td>32</td> <td>38</td> <td>35</td> <td>28</td> </tr> <tr> <td>Total eggs</td> <td>155</td> <td>120</td> <td>150</td> <td>150</td> <td>150</td> </tr> <tr> <td>Total eggs hatched</td> <td>70</td> <td>62</td> <td>78</td> <td>76</td> <td>47</td> </tr> <tr> <td>Hatching rate (%)</td> <td>52</td> <td>52</td> <td>52</td> <td>51</td> <td>31</td> </tr> </tbody> </table>			Trial	Control		Lindane			Water	Acetone	0.5 ppm	1 ppm	2 ppm	I No. eggs	30	30	30	30	30	No. eggs hatched	17	19	20	17	8	II No. eggs	30	15	45	45	45	No. eggs hatched	16	11	20	24	11	III No. eggs	75	75	75	75	75	No. eggs hatched	37	32	38	35	28	Total eggs	155	120	150	150	150	Total eggs hatched	70	62	78	76	47	Hatching rate (%)	52	52	52	51	31
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- <u>Rana pipiens</u>	cutaneous pectoris nerve-muscle	At 100 uM frequency of miniature end-plate potentials increased by $327 \pm 26\%$ ( $\bar{x} \pm S.E.$ ) and amplitude reduced to $21 \pm 14\%$ of controls after 30 min.	Suggest decreased amplitude were reflection of decreased sensitivity of end-plate to ACh.	Jay et al., 1987																																																																			
- <u>Rana temporaria</u>	cutaneous pectoris nerve-muscle	$5 \times 10^{-5}$ M caused marked increase in miniature end plate potential frequency.	Results suggest $Ca^{+2}$ permeability and $Ca^{+2}$ entry increased.	Publicover and Duncan, 1979																																																																			

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE			
Lindane (cont'd)	- <u>Rana temporaria</u>		cutaneous pectoris muscle	$5 \times 10^{-5}$ M for 40 min caused extensive damage to myofilaments; 20 min exposure had little effect.	In the presence of $Ca^{+2}$ , damage greater at 23°C than at 18°C.	Publicover et al., 1979			
	- <u>Rana pipiens</u>		abdominal skin	$2 \times 10^{-6}$ M increased both the short circuit current and the resistance.	Likely that the sodium permeability of the outer barrier was altered.	Webb et al., 1979			
Lithium Salts (Chloride and Sulphate)	- <u>Rana pipiens</u> and other species		eggs	Severity of effects increased with increased duration of exposure, concentration or temperature.	Microcephalic defects observed.	Hall, 1942			
Magnesium (M) (see also lead, cadmium, mercury, manganese)	- <u>Xenopus laevis</u>	50 per group	embryos	Magnesium ions moderated the toxicity of lead, cadmium, manganese and mercury ions.	Tests run for 6 days at 18°C.	Miller and Lendeman, 1978			
Malathion (UP)	- <u>Hyla regilla</u>		tadpoles 3 weeks old	Thermal tolerance significantly lowered:	24 hr exposure.  Recommended dosage rate for mosquito control in California-500 ppb.	Johnson, 1980			
				Conc. (ppb)			Onset of Spasms (°C)		
							Mean	Range	
				54			Control	37.5	36.9-37.8
				28			25	36.9	35.9-37.9
31	50	36.4	35.2-37.2						
35	100	36.4	35.1-37.5						
27	500	36.1	35.2-37.7						

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE																																										
Malathion (cont'd)	- <u>Microhylis ornata</u>	100 per concentration	embryos yolk-plug stage	Percent abnormal:  Duration of treatment  <table border="1"> <thead> <tr> <th>conc. (ppm)</th> <th>24 hr</th> <th>48 hr</th> <th>72 hr</th> <th>96 hr</th> <th>% mortality after 96 hr</th> </tr> </thead> <tbody> <tr> <td>0</td> <td>0</td> <td>0</td> <td>0</td> <td>0</td> <td>0</td> </tr> <tr> <td>1</td> <td>0</td> <td>5</td> <td>13</td> <td>23</td> <td>0</td> </tr> <tr> <td>5</td> <td>100</td> <td>100</td> <td>100</td> <td>100</td> <td>0</td> </tr> <tr> <td>10</td> <td>100</td> <td>100</td> <td>100</td> <td>100</td> <td>35</td> </tr> <tr> <td>15</td> <td>100</td> <td>100</td> <td>*</td> <td></td> <td>100</td> </tr> <tr> <td>20</td> <td>100</td> <td>*</td> <td></td> <td></td> <td>100</td> </tr> </tbody> </table>	conc. (ppm)	24 hr	48 hr	72 hr	96 hr	% mortality after 96 hr	0	0	0	0	0	0	1	0	5	13	23	0	5	100	100	100	100	0	10	100	100	100	100	35	15	100	100	*		100	20	100	*			100	Abnormalities include head, trunk and tail defects, behaviour, loss of balance, poor pigmentation and retarded growth.  Temp. - 25°C.	Pawar et al., 1983
	conc. (ppm)	24 hr	48 hr	72 hr	96 hr	% mortality after 96 hr																																										
0	0	0	0	0	0																																											
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5	100	100	100	100	0																																											
10	100	100	100	100	35																																											
15	100	100	*		100																																											
20	100	*			100																																											
	- <u>Plethodon cinereus</u> <u>P. glutinosus</u>	10 per treatment	adults	Brain cholinesterase was significantly inhibited at 5.6 kg/ha in <u>P. glutinosus</u> but not in <u>P. cinereus</u> . No changes in feeding, endurance or coordination.	Salamanders exposed to substrate treated with malathion.	Baker, 1985																																										

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE																																	
Malathion (cont'd)	- <u>Rana hexadactyla</u>	6 per treatment	gastrocnemius muscle	<table border="1"> <thead> <tr> <th>Treatment</th> <th>Twitch Amplitude (cm)</th> <th>Period of Latency (s)</th> </tr> </thead> <tbody> <tr> <td>I - Control</td> <td>2.6 ± .1</td> <td>.390 ± .021</td> </tr> <tr> <td>A</td> <td>3.2 ± .3 *</td> <td>.205 ± .022 *</td> </tr> <tr> <td>10 ppm M</td> <td>2.2 ± .2</td> <td>.164 ± .018 *</td> </tr> <tr> <td>20 ppm M</td> <td>1.0 ± .1 *</td> <td>1.025 ± .016 *</td> </tr> <tr> <td>30 ppm M</td> <td>0.7 ± .2 *</td> <td>.984 ± .020 *</td> </tr> <tr> <td>20 ppm M + A</td> <td>2.4 ± .2</td> <td>1.025 ± .016 *</td> </tr> <tr> <td>II - Control</td> <td>1.9 ± .2</td> <td>.943 ± .025</td> </tr> <tr> <td>A</td> <td>2.6 ± .3 *</td> <td>.738 ± .036 *</td> </tr> <tr> <td>20 ppm M</td> <td>0.4 ± .2 *</td> <td>.287 ± .042 *</td> </tr> <tr> <td>20 ppm M + A</td> <td>0.9 ± .2 *</td> <td>.246 ± .032 *</td> </tr> </tbody> </table> <p>A = Atropine, M = Malathion; values are mean ± SD. * significantly different from controls p &lt; 0.05.</p>	Treatment	Twitch Amplitude (cm)	Period of Latency (s)	I - Control	2.6 ± .1	.390 ± .021	A	3.2 ± .3 *	.205 ± .022 *	10 ppm M	2.2 ± .2	.164 ± .018 *	20 ppm M	1.0 ± .1 *	1.025 ± .016 *	30 ppm M	0.7 ± .2 *	.984 ± .020 *	20 ppm M + A	2.4 ± .2	1.025 ± .016 *	II - Control	1.9 ± .2	.943 ± .025	A	2.6 ± .3 *	.738 ± .036 *	20 ppm M	0.4 ± .2 *	.287 ± .042 *	20 ppm M + A	0.9 ± .2 *	.246 ± .032 *	Results indicate that action of malathion is similar to that of a pharmacologically active myo- and neuro-toxic agent.	Kowsalya et al., 1987
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	- <u>Rana pipiens</u>	24 per concentration	adults approx. 9 cm length	Following exposure to concentrations of 50, 75, 100, 125, 150, 175 ppm for 15 days: - frequency of deaths rose with concentrations; 20 dead after 15 days at 175 ppm - progressive anemia and leucopenia occurred with successively higher concentrations - white blood cell count showed progressive neutropenia and lymphocytosis with increasing concentrations.		Kaplan and Glaczenski, 1965																																	

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS				REMARKS	REFERENCE
				Conc. %	Percent metamorphosed	Mean length of metamorphosis	Mean time for metamorphosis in days		
Methionin (cont'd)	- <u>Rana tigrina</u>		tadpole limb bud stage	0.0007	20	16.2*	30.0	Temp 30-38°C.	Mohanty-Hajmadi and Dutta, 1981
				0.0006	40	16.2	28.0		
				0.0005	60	16.2	27.0		
				0.0004	60	16.2	30.0		
				0.0003	60	16.3	27.3		
				0.0002	100	16.4	25.8		
				0.0001	100	16.4	27.4		
				Control	100	16.7	19.6		
				* significantly different than controls.					
			tadpole feeding stage	None metamorphosed at concentrations 0.00001% to 0.005%.					
			eggs	No tadpoles from eggs treated with 0.00001% to 0.004% metamorphosed.					
	- <u>Rana catesbeiana</u>		tadpoles	40% of tadpoles exposed to 5 ppm died.				Continuous flow apparatus for 96 hr. Brain cholinesterase activity of mollusks fed dosed tadpoles was 90.6% of that of controls. None died.	Holl and Kolbe, 1980
	- <u>Bufo arenarum</u>	3 per treatment	embryos recently fertilized	47.3 mg/L produced 100% mortality within 5 days of exposure. 0.47 mg/L allowed larval development and metamorphosis. At 0.47 and 47.3 mg/L, as immersion time increased, inhibition of AChE augmented.				No behavioural effects.	de Lencas et al., 1985

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS				REMARKS	REFERENCE
				Treatment Group	No. of limbs examined	No. of limbs w/abnormalities			
						Slight	Severe		
Mareb 80 (F) (Manganese Ethylenedithiocarbamate)	- <u>Ixalurus cristatus carnifex</u>	36 Expt., 36-Ctrl.	adults	5 ppm caused delayed growth, reduced melanogenesis and malformations in regenerated limbs.				Temp 24 ± 1°C. Exposed for 4 days/week.	Artes and Zavanella, 1979
	- <u>Xenopus laevis</u>	30-60 per treatment	embryos yolk plug stage	Exposure (1-5 ppm) produced growth retardation, absence or reduction in melanogenesis in eyes, shortened tail, distinct notochord waviness. At higher concentrations organisms were unable to swim as well as control specimens.				Temp. 22 ± 1° C. Exposed for 1-10 days.	Bancroft and Prahled, 1973
	- <u>Ixalurus cristatus carnifex</u>	150		No tumours were found in any animals at a concentration of 5 ppm. Some edema of the soft tissues and enlargement of spleen, vascular congestion in gut walls found; glycogen content increased in treated sales.				Exposure for 19-23 weeks. Temp. 17 ± 1° C for 3 mo then 20 ± 1° C for following 2 mo, pH - 7.4.	Zavanella et al., 1979
	- <u>Ixalurus cristatus carnifex</u>	30 per treatment	whills	Effect of Mareb 80 on regeneration of limbs:				Female newts only, exposed 5 days/week percutaneously. Temp. 24 ± 1° C.	Zavanella et al., 1984
								Growth delayed in Mareb-exposed newts.	
				Tap water	10	6	1		
				Dispersant and wetting agent	10	3	2		Severe proximal and distal limb abnormalities in all animals exposed to Mareb 80.
				5 ppm Mareb 80	10	0	10		

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT*	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE																																																																																	
Manganese (M)	- <u>Xenopus laevis</u>		myelinated nerve fibres	Decreased permeability constant in K and Na systems; reversibly shifted Na activation curve in positive direction along potential axis.	Exposure for 6 days at 18°C.	Achew, 1980																																																																																	
	- <u>Xenopus laevis</u>		embryos	<table border="1"> <thead> <tr> <th rowspan="2">Hg (ppm)</th> <th colspan="6">Mn (ppm)</th> </tr> <tr> <th>10</th> <th>5</th> <th>1</th> <th>.1</th> <th>.01</th> <th>.001</th> </tr> </thead> <tbody> <tr> <td>200</td> <td>N</td> <td>N</td> <td>N</td> <td>N</td> <td>N</td> <td>N</td> </tr> <tr> <td></td> <td>94</td> <td>98</td> <td>92</td> <td>94</td> <td>84</td> <td>86</td> </tr> <tr> <td>20</td> <td>SL</td> <td>N</td> <td>N</td> <td>N</td> <td>N</td> <td>N</td> </tr> <tr> <td></td> <td>94</td> <td>90</td> <td>91</td> <td>100</td> <td>90</td> <td>94</td> </tr> <tr> <td>2</td> <td>H-S</td> <td>SL-M</td> <td>N</td> <td>N</td> <td>N</td> <td>N</td> </tr> <tr> <td></td> <td>96</td> <td>96</td> <td>88</td> <td>93</td> <td>96</td> <td>87</td> </tr> <tr> <td>.2</td> <td>S</td> <td>S</td> <td>H-S</td> <td>SL</td> <td>SL</td> <td>SL</td> </tr> <tr> <td></td> <td>72</td> <td>50</td> <td>92</td> <td>74</td> <td>96</td> <td>93</td> </tr> <tr> <td>0</td> <td>S</td> <td>0</td> <td>S</td> <td>S</td> <td>S</td> <td>S</td> </tr> <tr> <td></td> <td>54</td> <td>0</td> <td>79</td> <td>87</td> <td>87</td> <td>92</td> </tr> </tbody> </table> <p>D - Death; S - Severely deformed; M - Moderately deformed; SL - Slight effects; N - No effects; numbers = % survival from blastula to feeding stage.</p>		Hg (ppm)	Mn (ppm)						10	5	1	.1	.01	.001	200	N	N	N	N	N	N		94	98	92	94	84	86	20	SL	N	N	N	N	N		94	90	91	100	90	94	2	H-S	SL-M	N	N	N	N		96	96	88	93	96	87	.2	S	S	H-S	SL	SL	SL		72	50	92	74	96	93	0	S	0	S	S	S	S		54	0	79	87	87
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Mercury (M)	- <u>Bufo regularis</u>	8-10 per group	adults females, 25-30 g	Single i.m. injection of 5.60 mg Hg <sup>2+</sup> /kg produced significant decrease in serum alkaline phosphase, lactic dehydrogenase activities. Total bilirubin, serum total protein, aspartate aminotransferase, alanine aminotransferase increased.	5.60 mg/kg = 96 hr LD50. 96 hr exposure. EDTA not a significant treatment.	Hilmy et al., 1986b																																																																																	

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT*	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE																					
Mercury (cont'd)	- <u>Gastrophryne carolinensis</u>	100	eggs	Percent mortality and teratogenesis at hatching (H) and 4 days posthatching (PH) of eggs exposed to sediment-bound mercury:	Hardness - 200 ppm as CaCO <sub>3</sub> . pH - 7.5 - 8.0. Percent mortality expressed as frequency in experimental population/controls. * Percentages of survivors bearing gross congenital deformities at hatching.	Birge et al., 1977																					
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- <u>Rana catesbeiana</u>	eye	Hg <sup>2+</sup> caused irreversible decrease in rod response amplitude; did not affect cones.	Fox and Sillman, 1979																								
- <u>Xenopus laevis</u>	myelinated nerve fibres	Hg <sup>2+</sup> irreversibly shifted Na activation curve in positive direction along the potential axis.	Achew, 1980																								
- <u>Rana nigromaculata</u>	eggs, tadpoles	Exposure of eggs to 0.8 ppm Hg <sup>2+</sup> caused damage to primordial germ cells and their proliferation rate thereafter seemed to be lower. Lethal to tadpoles. Abnormal tadpoles at 0.4 and 0.8 ppm.	Treatment prior to first cleavage. Heh, 1978																								

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS						REMARKS	REFERENCE	
				Mg (ppa)	Mg (ppa)							
			1		.1	.05	.01	.001				
Mercury (cont'd)	<u>Xenopus laevis</u>		embryos	200	0	M-5	M-5	N	N	Exposure for 6 days at 18°C.	Miller and Lordean, 1978	
				20	0	M-5	M-5	N	N			
Mercury (inorganic)	<u>Xenopus laevis</u>		embryo-larval	2	0	M-5	M-5	N	N	Survival at 4 days post-hatch for embryos maintained in 0.16 and 0.34 ug/L averaged 46 and 28%, respectively. Adults had been maintained in 0.2 ug/L for 11 months prior to spawning.	Flow-through system, pH-7.5, water hardness - 102 ppm CaCO <sub>3</sub> .	Birge et al., 1979b
				.2	0	M-5	M-5	SL	SL			
Mercury (Mercuric Chloride)	<u>Rana pipiens</u>	100 per treatment	embryos	Percent survival following 4 days of continuous exposure:						Flow through system.	Birge and Just, 1975a	
				Concentration (ppm)								
				10	1	0.1	0.01	0.001	0.0001	Control		
				Cleavage	0	0	0	0	93	94	95	
				Blastula	0	0	0	0	82	78	84	
				Gastrula	0	0	0	80	85	95	95	
				Neurula	0	0	0	93	88	96	100	
				Tail Bud	0	0	20	80	93	95	95	

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS												REMARKS	REFERENCE					
				Percent survival of adults (A) and larvae (L):																		
				Exposure (days)	Concentration (ppm)																	
					50		25		10		7.5		5.0		2.5				1.0		0.5	
					L	A	L	A	L	A	L	A	L	A	L	A			L	A	L	A
Mercury (Mercuric Chloride) (cont'd)	<u>Rana pipiens</u>	100 per treatment	larvae adults	1	0	0	0	0	70	0	80	0	100	100	100	100	100	100	Flow through assay.	Birge and Just, 1975a (cont'd)		
				2	0	0	0	60	80	100	100	100	100	100	100	100	100	100				
				3				30	80	100	100	100	100	100	100	100	100	100				
				4				30	80	100	100	100	100	100	100	100	100	100				
				5				20	80	100	100	100	100	50	100	100	100	100				
				6				20	60	100	100	100	100	50	100	100	100	100				
				7				10	60	100	100	100	100	50	100	100	100	100				
				8				0	60	100	100	100	100	50	100	100	100	100				
				9				0	60	100	100	100	100	50	100	100	100	100				
				10				0	60	100	100	100	100	50	100	100	100	100				
	<u>Microhyla ornata</u>	50 per concentration	embryos	300 ug/L produced almost total mortality within 24 hr. 200-250 ug/L caused body blisters; hatched tadpoles were abnormal showing curved body axis, retarded growth, underdeveloped eyes. Most died within 72 hr. 100-150 ug/L produced low mortality, abnormalities were less severe. No significant changes noted at 50 ug/L.												pH 7.1. Temp. 21-25°C. 51 mg/l CaCO <sub>3</sub> alkalinity. Exposure 96 hr.	Ghate and Mulherker, 1980b					
				tadpoles (8 - 10 days)	50 ug/L caused total mortality within 24 hr. Survivors at 100-200 ug/L had distended body cavities and were sluggish after 24-48 hr.																	



Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE
Mercury (Methylmercury)	- <u>Rana pipiens</u>	5 per compound  8	ventral surface skin  adulte	Dimethylmercury did not alter the P.D. or the short circuit current across the skin at a concentration of $10^{-3}$ M. Methylmercuric chloride at $10^{-4}$ M reduced both the P.D. and S.C.C. Electrical resistance rose initially and then declined. The effects were irreversible. Methylmercuric chloride antagonized the action of vasotocin on osmotic water transfer and the $O_2$ consumption was depressed.  Injection into dorsal lymph sac of 20 mg/kg methylmercuric chloride or 18.4 mg/kg dimethylmercury caused no detectable toxic effects.  Immersion in 23 mg/L dimethylmercury for 7 days caused no detectable toxic effects.		Yorio and Bentley, 1973

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS			REMARKS	REFERENCE	
				Exposure	Concentration (ppm)				
Mercury (Methylmercury) (cont'd)	- <u>Rana pipiens</u>	200	tadpoles		0.001-0.01	0.05-0.01	0.5-1.0	Tadpoles raised in methylmercuric chloride water.  Tadpoles injected with 0.025 ml of 0.1% solution methylmercury chloride on alternate days for 10 days showed extensive swelling and distension of their legs.	Chang et al., 1974
				1 hour	No observable effect	Irritative movements	Irritative movements, difficulties in breathing, escape attempts		
				8 hours	"	Irritative movements, abnormal swimming postures.	As above, 50% mortality		
				12 hours	"	Abnormal swimming postures, difficulties breathing.	75% mortality		
				24 hours	Appeared lethargic	40% mortality	100% mortality		
				48 hours	Lethargic, no other changes after 4 months of observation and exposure	100% mortality			

Table 2 - Laboratory Studies (cont'd)

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Mercury (Methylmercury) (cont'd)	- <u>Rana pipiens</u>	N=45 per concentration	embryos	Effects of methylmercury on development of embryos treated for 3 days:  <table border="1"> <thead> <tr> <th rowspan="2">Conc. (ppb)</th> <th colspan="2">3 days post treatment</th> <th rowspan="2">% dead on 5th day</th> </tr> <tr> <th>Abnormal/Surviving</th> <th>% abnormal</th> </tr> </thead> <tbody> <tr> <td colspan="4"><b>Stages: Blastula</b></td> </tr> <tr> <td>0</td> <td>8/82</td> <td>10</td> <td>12</td> </tr> <tr> <td>5</td> <td>13/38</td> <td>34</td> <td>19</td> </tr> <tr> <td>10</td> <td>13/45</td> <td>29</td> <td>23</td> </tr> <tr> <td>15</td> <td>27/41</td> <td>66</td> <td>9</td> </tr> <tr> <td>20</td> <td>34/34</td> <td>100</td> <td>54</td> </tr> <tr> <td>30</td> <td>40/0</td> <td>---</td> <td>100</td> </tr> <tr> <td colspan="4"><b>Stages: Gastrula</b></td> </tr> <tr> <td>0</td> <td>8/81</td> <td>10</td> <td>20</td> </tr> <tr> <td>5</td> <td>3/39</td> <td>8</td> <td>20</td> </tr> <tr> <td>10</td> <td>8/41</td> <td>20</td> <td>31</td> </tr> <tr> <td>15</td> <td>30/36</td> <td>83</td> <td>60</td> </tr> <tr> <td>20</td> <td>18/18</td> <td>100</td> <td>91</td> </tr> <tr> <td>30</td> <td>2/2</td> <td>100</td> <td>100</td> </tr> <tr> <td colspan="4"><b>Stages: Neural Plate</b></td> </tr> <tr> <td>0</td> <td>6/83</td> <td>7</td> <td>13</td> </tr> <tr> <td>5</td> <td>10/45</td> <td>22</td> <td>29</td> </tr> <tr> <td>10</td> <td>18/42</td> <td>43</td> <td>20</td> </tr> <tr> <td>15</td> <td>13/45</td> <td>29</td> <td>22</td> </tr> <tr> <td>20</td> <td>11/39</td> <td>28</td> <td>60</td> </tr> <tr> <td>30</td> <td>31/0</td> <td>---</td> <td>100</td> </tr> </tbody> </table>	Conc. (ppb)	3 days post treatment		% dead on 5th day	Abnormal/Surviving	% abnormal	<b>Stages: Blastula</b>				0	8/82	10	12	5	13/38	34	19	10	13/45	29	23	15	27/41	66	9	20	34/34	100	54	30	40/0	---	100	<b>Stages: Gastrula</b>				0	8/81	10	20	5	3/39	8	20	10	8/41	20	31	15	30/36	83	60	20	18/18	100	91	30	2/2	100	100	<b>Stages: Neural Plate</b>				0	6/83	7	13	5	10/45	22	29	10	18/42	43	20	15	13/45	29	22	20	11/39	28	60	30	31/0	---	100	Temp. 21 ± 1°C.  Solutions changed daily. Abnormalities observed were tail defects, exogastric, stunting and poor general development.  Death and severe defects occurred over a narrow range of concentrations and increased with exposure time and increased concentrations.	Dial, 1976
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Mercury (Methylmercury) (cont'd)	- <u>Iriturus viridescens</u>	25 per group	adults	Initiation of regeneration of amputated hindlimbs was delayed in newts raised in 0.01-0.07 ppm. No delay in initiation observed in newts raised in 0.1-1.0 ppm.  Rate of regeneration markedly increased in all concentrations. All newts raised in 0.3 and 1.0 ppm died after 17 and 8 days, respectively.		Chang et al., 1976																																																																										
	Metacid (DDT + methyl parathion)	- <u>Rana tigrina</u>		tadpoles	Effects on feeding stage (F) and limb bud stage (L):  <table border="1"> <thead> <tr> <th rowspan="2">Conc. %</th> <th colspan="2">% metamorphosis</th> <th colspan="2">Mean time for metamorphosis (days)</th> </tr> <tr> <th>F</th> <th>L</th> <th>F</th> <th>L</th> </tr> </thead> <tbody> <tr> <td>0.0003</td> <td>-</td> <td>60</td> <td>-</td> <td>34.6</td> </tr> <tr> <td>0.0002</td> <td>-</td> <td>80</td> <td>-</td> <td>37.5</td> </tr> <tr> <td>0.0001</td> <td>20</td> <td>100</td> <td>55.0</td> <td>33.6</td> </tr> <tr> <td>0.00009</td> <td>20</td> <td>no data</td> <td>59.0</td> <td>no data</td> </tr> <tr> <td>0.00008</td> <td>40</td> <td>"</td> <td>60.0</td> <td>"</td> </tr> <tr> <td>0.00007</td> <td>40</td> <td>"</td> <td>59.5</td> <td>"</td> </tr> <tr> <td>0.00006</td> <td>40</td> <td>"</td> <td>60.0</td> <td>"</td> </tr> <tr> <td>0.00005</td> <td>60</td> <td>"</td> <td>64.6</td> <td>"</td> </tr> <tr> <td>0.00004</td> <td>80</td> <td>"</td> <td>64.5</td> <td>"</td> </tr> <tr> <td>0.00003</td> <td>100</td> <td>"</td> <td>66.6</td> <td>"</td> </tr> <tr> <td>0.00002</td> <td>100</td> <td>"</td> <td>66.8</td> <td>"</td> </tr> <tr> <td>0.00001</td> <td>100</td> <td>"</td> <td>67.2</td> <td>"</td> </tr> <tr> <td>Control</td> <td>100</td> <td>100</td> <td>35.6</td> <td>21.8</td> </tr> </tbody> </table>	Conc. %	% metamorphosis		Mean time for metamorphosis (days)		F	L	F	L	0.0003	-	60	-	34.6	0.0002	-	80	-	37.5	0.0001	20	100	55.0	33.6	0.00009	20	no data	59.0	no data	0.00008	40	"	60.0	"	0.00007	40	"	59.5	"	0.00006	40	"	60.0	"	0.00005	60	"	64.6	"	0.00004	80	"	64.5	"	0.00003	100	"	66.6	"	0.00002	100	"	66.8	"	0.00001	100	"	67.2	"	Control	100	100	35.6	21.8	Temp. 30-38°C.  Reduced length at metamorphosis at 0.0001% for feeding stage tadpoles.
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			eggs	<table border="1"> <thead> <tr> <th>Conc. %</th> <th>% metamorphosis</th> <th>Mean time for metamorphosis</th> </tr> </thead> <tbody> <tr> <td>0.00005</td> <td>20</td> <td>62.5</td> </tr> <tr> <td>0.00004</td> <td>50</td> <td>64.2</td> </tr> <tr> <td>0.00003</td> <td>80</td> <td>62.5</td> </tr> <tr> <td>0.00002</td> <td>90</td> <td>59.2</td> </tr> <tr> <td>0.00001</td> <td>100</td> <td>57.6</td> </tr> <tr> <td>Control</td> <td>100</td> <td>34.9</td> </tr> </tbody> </table>	Conc. %	% metamorphosis	Mean time for metamorphosis	0.00005	20	62.5	0.00004	50	64.2	0.00003	80	62.5	0.00002	90	59.2	0.00001	100	57.6	Control	100	34.9																																																							
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Table 2 - Laboratory Studies (cont'd)

CONTAMINANT*	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS			REMARKS	REFERENCE
Methoxychlor (OC)	<u>Rana pipiens</u>		abdominal skin	2 x 10 <sup>-4</sup> M produced no significant effect on the short circuit current or resistance.			Frogs placed in 200 cc of test solution. Frogs which died had exhibited neuromuscular changes, excessive thrashing and abnormal reactivity to stimulation. Survivors were free from disorders.	Webb et al., 1979
			adults 65 g, 8.9 cm long	Methoxychlor (ppm)	pH of solution at 25° C	No. of frogs dead at 30 days		
			20	0.80	6.31	5		
			10	0.60	6.10	0		
			10	0.40	5.95	0		
Methyldeeton (OP)	<u>Rana tigrina</u>		eggs	Conc. %	% Metamorphosis	Mean time for metamorphosis (days)	Temp-30-38°C. Doses used were lower than suggested field applications. Reduced size at metamorphosis at 0.00002%.	Mohanty-Hajmadi and Dutta, 1981
				0.00003	20	42.0		
				0.00002	30	46.6		
				0.00001	50	47.2		
				Control	70	33.7		
tadpoles feeding stage	Conc. %	% Metamorphosis	Mean time for metamorphosis (days)					
	0.00004	40	51.0					
	0.00003	60	51.6					
	0.00002	60	55.6					
	0.00001	80	56.5					
Control	100	33.2						

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT*	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS			REMARKS	REFERENCE
Methyldeeton (cont'd)	<u>Rana tigrina</u>		tadpoles limb bud stage	Conc. %	% Metamorphosis	Mean time for metamorphosis (days)	Reduced size at metamorphosis at 0.0002%	Mohanty-Hajmadi and Dutta, 1981 (cont'd)
				0.0008	60	29.3		
				0.0007	60	28.6		
				0.0006	40	28.5		
				0.0005	80	22.5		
				0.0004	100	19.4		
				0.0003	100	18.4		
				0.0002	100	16.8		
				0.0001	100	18.8		
				0.00004	no data	no data		
				0.00003	no data	no data		
				0.00002	no data	no data		
				0.00001	no data	no data		
Control	100	18.6						
Methyl Fluoroacetate	Green Frog		skeletal nerves	5 mM blocks nerve in 3-4 hr; 50 mM blocks within 1.5 hr. .005 and .001 M produced 80 and 25%, respectively, inhibition of respiration after 2 hr.				Boyaraky et al., 1969

Table 1 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE		
Methylhydrazine (Component of Jet Fuel)	- <u>Xenopus laevis</u>	50-110 per treatment	larvae	1 mg/L produced irreversible toxic effects and death within 48 hr. Necrotic metamorphosis occurred in larvae in continuous contact with 0.1 mg/L.	Most common abnormality was tail kinks.	Greenhouse, 1976a		
		50-100 per treatment	embryo	≤5 mg/L harmless if embryo transferred to uncontaminated water by completion of neurulation. Teratogenic at ≥10 mg/L.				
	- <u>Xenopus laevis</u>		embryo	Concentration (mg/L)		% Malformed	Kinked tail has most frequent malformation. Most larvae with kinked tails metamorphosed into necrotic frogs. Other malformations included microcephaly, failure of elongation and edema.	Greenhouse, 1976b
		100 100 200 150 50		control 3 5 10 15		3 1 52 93 100		
Methyl isothiocyanate (F)	- <u>Xenopus laevis</u>	60 per concentration	embryo yolk plug stage	Susceptible during neurulation.				Birch and Prahalad, 1986
				Conc. (ppb)	% Mortality at day 10	% Abnormal at day 10		
				Control 0.001 0.005 0.01 0.05 0.1 0.5 1.0	5 5 5 10 50 70 90 100	0 0 0 0 0 0 0		
				Embryo exposed to 100-500 ppb did not survive after day 5; those exposed to 600 ppb did not complete neurulation.				

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE	
Methylparathion (OP) (see also metacid)	- <u>Bufo boreas</u>	20	Juveniles (7 snout-vent length - 19.8 mm)	Exposure of hydrated toads to 25 ppb significantly lowered temperature tolerance.	Temp. 23-24°C; 24 hr exposure.	Johnson and Prine, 1976	
	- <u>Rana cyanophictis</u>	6 per treatment	tadpoles approx. 4 weeks old		Exposure for 24 hr.	Yasmeen and Haqueunnisa, 1985	
				Brain Glucose Level (mg/g ww)	Oxygen Consumption (ul/g/h)		
				control 5 ppm	4.76 ± 0.30 * 2.4 ± 0.91 p > 0.05	205.81 ± 8.1 305.24 ± 65.42 p = 0.01	
				* values are mean ± S.D.			
	- <u>Hyla regilla</u>		tadpoles 3 weeks old	Thermal tolerance significantly lowered:	24 hr exposure.	Johnson, 1980	
			Conc. (ppb)	Onset of Spases (°C)		Recommended dosage rate for mosquito control in California - 100 ppb.	
				Mean	Range		
		54	Control	37.5	36.9-37.8		
		27	25	35.7	31.7-37.0		
		34	50	34.3	29.1-36.8		
		36	100	33.6	30.7-36.3		
	- <u>Rana cyanophictis</u>		tadpoles approx. 4 weeks old	Exposure for 24 hr to 2.5 ppm affected the qualitative nature of the brain lactate dehydrogenase isozymes.		Yasmeen and Haqueunnisa, 1986	
Mine Drainage (Iron, Zinc, Copper)	- <u>Bufo boreas</u>	200 - test, 200 - control	zygotes	All zygotes placed in mine drainage (pH 2.79, 260 mg/L Fe, 39 mg/L Zn, 5.7 mg/L Cu) died within 12 hours and before they reached the second cleavage stage.	Zygotes cultured in lake water (pH 7.1, negligible quantities of metals) developed normally through metamorphosis.	Porter and Hakenson, 1976	

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS		REMARKS	REFERENCE
				Conc. (ppb)	No. surviving at day 10		
sodium Methyl sonate (M) (MSA)	- <u>Litoria ewingi</u> <u>Litorodactylus</u> <u>tasmaniensis</u>	20 per conc.	adults	No deaths observed following exposure to 300-400 ppm for 96 hr. No deaths observed following exposure to 130-520 ppm for 96 hr.			Johnson, 1976
near 592 HC)	- <u>Scaphiopus couchi</u>	5 adults per group, 15 juveniles per group	adults 50-61 mm snout-vent length, juveniles 18-19 mm snout-vent length	100 ppm (47.74 ppm MSHA) was not toxic to juveniles. 1,000 ppm (477.4 ppm MSHA) was toxic to juveniles but not to adults. Cumulative % mortality for juveniles: 120 hr - 6.6% 168 hr - 53.3% 192 hr - 86.6% 10,000 ppm (4,774 ppm MSHA) was toxic to both adults and juveniles; adults were less sensitive. Cumulative % mortality for juveniles: 48 hr - 60.0% 72 hr - 100.0% 100,000 ppm (47,740 ppm MSHA) was toxic to all adults and juveniles within 2 hr of exposure.		Possible reasons that juveniles more susceptible than adults: 1) surface area: volume ratio 2) skin permeability 3) metabolic rate 4) prior exposure	Judd, 1977
abam (F) (Ethy- nabis dithiocar- bamic acid sodium salt)	- <u>Xenopus laevis</u>	30-60 per treatment	embryos yolk-plus stage	1-3 ppm induced waviness of notochord and punctate melanophores.		Temp. 22 ± 1°C. Exposure - 1-10 days.	Bancroft and Prahlad, 1973

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS			REMARKS	REFERENCE
				Conc. (ppb)	No. surviving at day 10	% Abnormal at day 10		
abam (F) (Ethy- nabis dithiocar- bamic acid sodium salt) (cont'd)	- <u>Xenopus laevis</u>	60 per concentration	embryos yolk plus stage	Control	57	0	Abnormalities involved swollen and kinked notochords. Storage of dilute abam solutions for 1 week greatly enhanced its teratogenic and lethal potential.	Birch and Prahlad, 1986
				10	58	0		
				20	56	0		
				30	57	5		
				40	60	62		
				50	58	79		
				100	57	90		
				1000	56	96		
	- <u>Xenopus laevis</u>	30	embryos stage 10-11	Exposure to 4 ppm for 7 days caused alterations in pigmental retina, notochord and skin at ultrastructural level.				Prahlad et al., 1974
nickel (Ni <sup>2+</sup> ) (N)	- <u>Iriturus pyrrogastrer</u>		stomach mucous epithelial cells	Lower concentrations did not affect the electrical properties of the cells. 10 <sup>-6</sup> M decreased membrane potential to 82% of control value.				Kanno et al., 1978
	- <u>Xenopus laevis</u>		myelinated nerve fibres	Slowed down kinetics of K system, decreased permeability constant in K and Na systems, reversibly shifted Na activation curve in positive direction along potential axis.				Arhee, 1980
diclofenac sulfonamide sulfonamide sulfonamide (Sayer 73)	- <u>Bufo regularis</u>	100	adults 50 g	Force feeding 0.3 mg daily during non-breeding season induced formation of kidney tumours in 2 toads.			Other researchers induced lesions in 23% of animals treated during breeding season using same dose and methodology.	Sabry and El-Mofty, 1986
amprolium (OC)	- <u>Rana pipiens</u>		abdominal skin	Produced a decrease in the short circuit current and increased the resistance.				Hobb et al., 1979

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE
Noniphenoethoxy- late (surfactant)	- <u>Rana esculenta</u>		leg skin	$5 \times 10^{-5}$ M inhibited the isotonic, active transport-coupled volume flow.		Colantoni et al., 1979
NRDC 119 (PY)	- <u>Rana temporaria</u>		arterius nervo-muscle	Multiple end-plate potentials appeared after exposure to $>2$ mg/L for 18-45 min. 1-100 mg/L had no effect on muscle resting potential or amplitude or time course of the initial evoked end-plate potential.		Evans, 1976
Octyl-phenyl- $\alpha$ - naphthylsulfonic (component of jet and rocket fuel)	- <u>Rana pipiens</u> <u>Xenopus laevis</u>	200	embryo larva	Up to 1 g/L not toxic to either species.		Greenhouse, 1976a
	- <u>Rana pipiens</u> <u>Xenopus laevis</u>	100 per treatment	embryo	200 and 1 mg/L had no deleterious effects on development of <u>R. pipiens</u> and <u>X. laevis</u> , respectively.		Greenhouse, 1976c
DMPA (DP) (Dimethyl Pyrophosphoride)	- Frog		arterius muscle	10 mM eliminated response to carbacetylcholine almost completely. Amount of inhibition was strongly dependent upon concentration of DP and temperature.		Oekin et al., 1978
	- <u>Rana pipiens</u>	24 in each concentration	adult approx. 9 cm long	Following exposure to concentrations of 2500, 2800, 3100 ppm for 15 days: - Anemia and leucopenia produced, worsened with increasing concentration - Progressive neutropenia and lymphocytosis evident with increasing concentration - Posture drooping, activity decreased, flaccid paralysis.		Kaplan and Glazzenaki, 1965
	- Frog		arterius muscle	Reduced amplitude of response without significantly shifting the dose response curve along the abscissa.	Results suggest that DMPA may bind to at least 3 sites on receptor.	Guy et al., 1977

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE																	
Oxamyl (C)	- <u>Rana temporaria</u>		tadpoles	Exposure to 100 ppm oxamyl for 1 hour resulted in 90% of tadpoles with vertical curvature deformities.		Cooke, 1981																	
Paraoxon	- <u>Hyla chrysoeclia</u>		embryo	Not toxic or teratogenic at 100 ppm.	Orally exposed for 24 hr, no survivors counted 120 hr post-exposure.	Fulton and Chambers, 1985																	
Paraquat (H)	- <u>Rana pipiens</u>	110 per dose group	embryo	<table border="1" style="width: 100%; border-collapse: collapse;"> <thead> <tr> <th rowspan="2"></th> <th colspan="5">Concentration (ppm)</th> </tr> <tr> <th>0</th> <th>0.1</th> <th>0.5</th> <th>2</th> <th>10</th> </tr> </thead> <tbody> <tr> <td>No. alive on day 12 post-treatment</td> <td>83</td> <td>76</td> <td>6</td> <td>0</td> <td>0</td> </tr> </tbody> </table>		Concentration (ppm)					0	0.1	0.5	2	10	No. alive on day 12 post-treatment	83	76	6	0	0	Temp. $21 \pm 1^\circ\text{C}$ . Paraquat rapidly disappears from water thus timing of application is critical.	Dial and Bauer, 1984
		Concentration (ppm)																					
		0	0.1	0.5	2	10																	
No. alive on day 12 post-treatment	83	76	6	0	0																		
- <u>Rana pipiens</u>	80 per dose group	embryo: early gastrula and 15 days of age	% alive 16 days post-treatment after exposure at gastrula stage (G) or 15 days of age (15): <table border="1" style="width: 100%; border-collapse: collapse;"> <thead> <tr> <th colspan="4">Paraquat (mg/L)</th> </tr> <tr> <th colspan="2">0.5</th> <th colspan="2">2</th> </tr> <tr> <th>G</th> <th>15</th> <th>G</th> <th>15</th> </tr> </thead> <tbody> <tr> <td>6.3</td> <td>66.7</td> <td>0.0</td> <td>5.0</td> </tr> </tbody> </table>	Paraquat (mg/L)				0.5		2		G	15	G	15	6.3	66.7	0.0	5.0	All figures significantly lower than controls. Retardation of growth, tail malformations, poor head development observed.	Dial and Dial, 1987		
Paraquat (mg/L)																							
0.5		2																					
G	15	G	15																				
6.3	66.7	0.0	5.0																				
- <u>Rana esculenta</u>	10 per treatment	lung and liver	Activities of antioxidant enzymes were decreased or not affected by $10_{50}$ injected into abdominal lymph sac at $4^\circ\text{C}$ ; $10_{100}$ resulted in significant increase in activity for most enzymes. Lipid peroxidation of liver decreased in response to paraquat at $4^\circ\text{C}$ , increased at $20^\circ\text{C}$ ; in lung lipid peroxidation increased at both temperatures.	$260$ mg/kg = $10_{50}$ at $20^\circ\text{C}$ ; $360$ mg/kg = $10_{100}$ at $20^\circ\text{C}$ . Lower temperature increased survival.	Barabas et al., 1985																		

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT*	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE
Parathion (OP)	- <u>Acris crepitans</u>	72 per treatment	adults	Mortality at 96 hr exposure was dose-related (0, 0.1, 1.0, 10 ppm).	pH - 7.6; CaCo <sub>3</sub> - 80 mg/L Temp - 23-26°C. 1 bird ( <u>Falco sparverius</u> ) died within 3 hr after consuming 5 frogs exposed to 10 ppm for 96 hr.	Fleming et al., 1982
	- <u>Bufo arenarum</u>		tadpoles	95% of tadpoles exposed to 5 ppm died.  Bioconcentration factor: $\bar{x}$ 64	Continuous flow apparatus for 96 hr. Brain cholinesterase activity of mallards fed dosed tadpoles was 0.8% of that of controls. All ducks died within $\frac{1}{2}$ hr.	Hall and Kolbe, 1980
	- <u>Bufo arenarum</u>	32 per treatment	adults brain, liver, blood	Toads exposed to 0.1 ml/m <sup>2</sup> fumigant for 45 days or 2 hr experienced a plasma cholinesteratic activity decrease of 86% the normal value. Activity quickly recovered when toads replaced into normal conditions.	Temp. 16 ± 2°C during hibernation, 22 ± 3°C during mating season.	Guzzen and Guardia, 1978
	- <u>Rana pipiens</u>	24 in each concentration	adults approx. 9 cm long	Following exposure to concentrations of 5, 10, 15, 20, 25 ppm for 15 days: - Anemia and leucopenia which worsened with increasing concentration produced - Progressive neutropenia and lymphocytosis as concentration increased - Marked decrease in activity, decreased muscle tone, generalized edema		Keplen and Glaczenaki, 1965

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT*	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE									
Parathion (cont'd)	- <u>Bufo viridis</u>	3 per compound	adults	Residual cholinesterase at time of death following injection of 2 LD50 into dorsal lymphatic sac:	LD50: parathion - 967 ppm. paraoxon - 188 ppm.	Edery and Schatzberg-Porath, 1960									
			blood mixed nervous tissue	Parathion			Paraoxon								
				57%			5%								
	livers	Metabolism of parathion incubated with liver slices for 30 min (values in $\mu$ mol ± S.E.M.):	Paraoxon readily degraded thus levels given are difference between production and destruction.												
	- <u>Rana pipiens</u> - <u>Bufo sp.</u>	4 4		<table border="1"> <thead> <tr> <th>No. replicates</th> <th>Parathion remaining</th> <th>Paraoxon recovered</th> </tr> </thead> <tbody> <tr> <td>6</td> <td>52.2 ± 5.34</td> <td>2.20 ± 0.47</td> </tr> <tr> <td>4</td> <td>37.6 ± 4.24</td> <td>2.81 ± 0.45</td> </tr> </tbody> </table>	No. replicates	Parathion remaining	Paraoxon recovered	6	52.2 ± 5.34	2.20 ± 0.47	4	37.6 ± 4.24	2.81 ± 0.45		Potter and O'Brien, 1964
No. replicates	Parathion remaining	Paraoxon recovered													
6	52.2 ± 5.34	2.20 ± 0.47													
4	37.6 ± 4.24	2.81 ± 0.45													
Permethrin (PY)	- <u>Xenopus laevis</u>		lateral-line sense organ  peripheral nerves	<p><math>5 \times 10^{-6}</math> M <u>in vitro</u> for 3 hr or <math>10^{-5}</math> M <u>in vivo</u> for 5 hr induced short trains of nerve impulses.</p> <p><math>5 \times 10^{-6}</math> M caused repetitive activity within first 2 hr of exposure.</p>	Temp. 8-22°C. Number of impulses per train and train duration increased with cooling.	Vijverberg et al., 1982									
pH (see also aluminum)	- <u>Xenopus laevis</u>	10-20	embryos stages 10-13	Fatal inhibition of hatching occurred at pH 3.9 in sulfuric acid and pH 4.3 in bog water. In both types, at low pH embryos underwent a tight coiling associated with shrinkage of the perivitelline space; when jelly layer removed at pH 4.3 the embryos developed normally.	Temp - about 25°C. Possible cause - block in functioning of hatching enzyme.	Dunson and Conwell, 1982									
	- <u>Rana catesbeiana</u>	26	adults 100-300 g	Brain AChE activity altered in pH-dependent fashion. Animals raised at pH 4.6 died within 7-10 days.	Brain AChE levels not affected by presence of Al <sup>3+</sup> .	Merquis 1982									

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE
pH (cont'd)	- <u>Rana sylvatica</u>	30 per treatment	embryo 4 hr post-fertilization	Differences in acid tolerance among populations of embryo was not related to levels of acidity in ponds.	Temp. 15°C, medium changed daily.	Pierce and Harvey, 1987
	larvae stages 25-27		Tadpoles produced by adult from acidic ponds were more acid-tolerant.	Temp. 15°C.		
	- <u>Ambystoma macrodactylum</u>		larvae	pH 3 - all died within 12 hr. pH 4 and 5 - significantly slower rate of development and growth than those reared at pH >5.		Ling et al., 1986
	<u>Rana sylvatica</u>		larvae	pH 3 - all died within 2 hr. No differences in development noted at any other treatment.		
	- <u>Myia crucifera</u>		larvae 4 weeks old	96 hr survival was over 90% at pH 7.0 and about 70% at pH 5.0. No tadpoles survived 24 hr at pH 4.0.	Static bioassay.	Correll et al., 1987
- <u>Ambystoma jeffersonianum</u> <u>Rana sylvatica</u>	larvae	Acute exposure to pH 2.5 - 4.0 depressed sodium influx and markedly accelerated sodium efflux - resulting net loss of 50% body Na was fatal. Chronic exposure caused 21-62% reduction in body Na level, K content did not change.			Fredo and Dunson, 1985	
embryo	<u>A. jeffersonianum</u> did not hatch below pH 4.50; <u>R. sylvatica</u> hatched at pH 4.25.					

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE																																	
pH (cont'd)	- <u>Rana temporaria</u>	4 per treatment	eggs early cleavage	Mortality Rate (mean $\pm$ SD):	Most hatchlings at pH 4.0 were deformed; at pH 4.5 or 5.0 few deformities noted. Mortality figures for eggs of <u>R. arvalis</u> , <u>R. esculenta</u> , <u>B. bufo</u> similar.	Leuven et al., 1986																																	
	<u>Rana arvalis</u> <u>Rana esculenta</u> <u>Bufo bufo</u>		larvae stage 29-33	<table border="1"> <thead> <tr> <th colspan="3">% Mortality</th> </tr> <tr> <th>pH</th> <th>Basic medium</th> <th>Al medium</th> </tr> </thead> <tbody> <tr> <td>4.0</td> <td>0.0</td> <td>0.0</td> </tr> <tr> <td>4.5</td> <td>0.0</td> <td>75.0</td> </tr> <tr> <td>5.0</td> <td>0.0</td> <td>83.3</td> </tr> <tr> <td>4.0</td> <td>6.3</td> <td>100.0</td> </tr> <tr> <td>4.5</td> <td>6.7</td> <td>100.0</td> </tr> <tr> <td>5.0</td> <td>0.0</td> <td>100.0</td> </tr> <tr> <td>4.0</td> <td>23.8</td> <td>14.3</td> </tr> <tr> <td>4.5</td> <td>0.0</td> <td>20.6</td> </tr> <tr> <td>5.0</td> <td>0.0</td> <td>47.6</td> </tr> </tbody> </table>	% Mortality			pH	Basic medium	Al medium	4.0	0.0	0.0	4.5	0.0	75.0	5.0	0.0	83.3	4.0	6.3	100.0	4.5	6.7	100.0	5.0	0.0	100.0	4.0	23.8	14.3	4.5	0.0	20.6	5.0	0.0	47.6	Al medium: [Al <sup>3+</sup> ] = 185 $\mu$ mol/L	
% Mortality																																							
pH	Basic medium	Al medium																																					
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5.0	0.0	47.6																																					
	- <u>Plethodon cinereus</u>	30	adults	Animals given substrate choice were found 50% of time on pH 6-6.3, 31% on pH 4.5-5, 19% on pH 3-3.5.		Wyman and Hawkesley - Lowcull, 1987																																	
		10 per pH		Animals held on substrates of pH 2-2.5 died within 1 week; those on pH 3-6 survived.	Temp. 8-10°C. 1 month exposure.																																		
		10 per pH		40% held on substrates of pH 3 died within 4 mo, all others survived. Growth reduced 60% and 45% in salamanders living on pH 3 and 4, respectively. O <sub>2</sub> consumption rate reduced by 32% in animals living on pH 3.	8 month exposure.																																		



Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE		
pH (cont'd)	-		adults	Selection of substrate pH:		* = significant difference.	Mushinsky and Brodie, 1975	
				Field pH	% selecting pH 5.5			% selecting pH 7.7
				6.8-7.0	27.5			72.5*
				4.5-5.5	43.2			56.8*
				5.5	37.5			62.5*
				-	40.0			60.0*
				5.5	47.0			53.0
				-	32.9			67.1*
				-	50.0			50.0
				-	53.3			46.7
- Assorted species/review		adults, larvae, embryos	Inter- and intraspecific variation and ontogenetic change in acid tolerance; mechanisms of tolerance reviewed.			Pierce, 1985		
- <u>Bufo bufo</u>	4 per test	Ladpoles 30-35 mm	pH of solution	Survival time	Temp. 18°C.	Jones, 1939		
			2.0	55 min				
			2.8	125 min				
			3.4	6.5 hr				
			3.8	18 hr				
			4.0	over 24 hr				
			4.2	no apparent effect in 3 days				
- <u>Rana clamitans</u>	21-26	premetamorphic tadpoles, juveniles	Exposure to pH 4.0 (nominal) caused substantially increased transepithelial net ion loss and net acid uptake, and a slight inhibition of active ion transport. Disturbances disappeared by 7 hr exposure in tadpoles but persisted in juveniles.		Soft water (Ca <sup>2+</sup> = 300 µequiv/L). Prior acclimation to pH 5 did not reduce disturbances.	McDonald et al., 1984		

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE			
pH (cont'd)	-		embryos	Mean % of hatch or coil (±SE):		total number of eggs tested in parentheses.	Karna, 1984		
				Boq (pH 4.2)				Fen (pH 7.5)	
				Hatch	Coil			Hatch	Coil
				11.0 ± 1.15 (1758)	77.2 ± 2.12 (164)			96.3 ± 0.61 (2121)	0.4 ± 0.21 (152)
				0	59.1 ± 13.51 (164)			97.3 ± 0.29 (1921)	0
				0	14.6 ± 2.76 (3038)			93.0 ± 2.21 (1921)	0.2 ± 0.16 (1921)
				0	0.9 ± 0.66 (435)			98.5 ± 0.66 (987)	0
				0	0 (285)			64.5 ± 10.38 (232)	0
				0	0 (325)			71.5 ± 7.13 (419)	0.3 ± 0.30 (419)
									Fertilization of <u>R. sylvatica</u> , <u>B. americanus</u> , <u>R. pipiens</u> eggs in boq water not significantly different than controls.
- <u>Rana pipiens</u>		tadpoles	Linear increase in survival time and thus pH tolerance during first 8 weeks of development. Tadpoles at pH 4.4 and 5.8 grew at rates of 5.9 and 36.0 µg/day, respectively.		24 day test period.	Frede and Dunson, 1985c			
		embryos	"Lethal" pH (100% mortality)	"Critical" pH (50% mortality)	Sensitivity of <u>X. laevis</u> , <u>Ambystoma jeffersonianum</u> greater in late stages of development than during initial cleavage.	Tome and Pough, 1982			
			3.0	3.5					
			-	3.5					

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS					REMARKS	REFERENCE						
				pH	Ca (ppm)	% hatched	% of hatchlings deformed	% of embryo killed								
								Early			Late					
pH (cont'd)	<u>Ambystoma jeffersonianum</u>	20 per treatment	embryo	5.0	1	100	0	0	0	Addition of Ca, Mg and >10 ppm Na prevented early mortality of embryos. Increasing concentrations of ions caused curling defect. <u>A. jeffersonianum</u> could usually hatch even though curled but <u>R. sylvaticus</u> usually died.	Freda and Dunson, 1985b					
					20	95	0	5	0							
40					100	0	0	0								
4.5				1	70	100	0	5	0							
				20	100	5	0	0								
				40	100	0	0	0								
4.0				1	0	0	100	0	0							
				20	0	0	0	100	0							
				40	20	0	5	75	0							
				80	40	50	5	55	0							
pH (cont'd)				<u>Rana sylvatica</u>	20-40	embryo	5.8	1	95			0	0	5		
								20	100			25	0	0		
								40	95			16	0	5		
							5.0	1	100			0	0	0		
	20	80	0					20	0	0						
	40	100	10					0	0	0						
	4.5	1	90				5	10	0	0						
		20	100				0	0	0	0						
		40	55				51	5	62	0						
	4.0	1	10				75	58	32	0						
		20	15				20	7	80	0						
		40	8				0	15	77	0						
		80	0				0	10	90	0						

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS		REMARKS	REFERENCE				
				pH	Mean % hatching ± S.E.M.						
pH (cont'd)	<u>Rana pipiens</u> , <u>R. catesbeiana</u> , <u>R. clamitans</u>		larvae	Acute exposure to pH 2.5 - 4.0 depressed Na influx and accelerated Na efflux. Increased external Ca slowed loss of Na. Initial body Na content was inversely correlated with acid tolerance.		Net loss of 50% body Na was fatal. Species differed in Na content under control conditions.	Freda and Dunson, 1984				
				<u>Rana sibilans</u>	abdominal skin			Increase in influx and backflux of Cl and ammonia and in backflux of Na at pH 2.5 in short-circuited skin. Total conductance and short-circuit current increased at pH 2.5.		Control = pH 7.4.	Ferreiro and Hill, 1982
								<u>Ambystoma opacum</u>	4-12		
	pH		Mean % hatching ± S.E.M.								
	7.2 - 7.6	71.7 ± 6.3									
	6.0	77.3 ± 9.1									
	5.0	82.6 ± 7.6									
	4.0	72.1 ± 6.8									
	3.0	0 ± 0									
	2.0	0 ± 0									
<u>Rana sylvatica</u>	442 131 145 416 375 125	embryo	pH		% survival		Pierce et al., 1984				
			≥4.0	100.0							
			3.75	93.3							
			3.5	60.0							
			≤3.25	0							
10	larvae (early feeding)	pH		% survival							
		≥3.75	100.0								
		3.5	90.0								
≥3.25	0										

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE							
H (cont'd)	- <u>Ambystoma texanum</u>	100 per treatment	embryos	Hatching success:	Temp. - 17° C in pond water from collection sites, pH range 6.4 - 6.7.	Punzo, 1983							
				Percent Survival pH									
	Temp. °C		4	5			6	7	8	9	10	11	
	5	0	0	0	0	0	0	0	0				
	10	13	21	75	83	81	71	69	19				
	15	17	30	95	92	90	78	75	29				
	20	9	20	64	74	66	59	50	2				
	25	6	14	30	24	27	19	7	0				
	30	0	0	0	0	0	0	0	0				
				Highest oxygen consumption rates at pH 6.6; significant decrease at pH 4 and 11.									
	- <u>Rana pipiens</u>		eggs	Sperm motility decreased with decreasing pH below 6.5 in solutions acidified with H <sub>2</sub> SO <sub>4</sub> or HNO <sub>3</sub> ; below 5.5 in solutions acidified with HCL. No eggs developed at ≤ pH 4.8. Fertilization and formation of healthy embryos decreased below pH 6.3. Lower limit for optimal fertilization and early development was pH 6.0.									
	- <u>Bufo boreas</u>		larvae	pH 3.1 - all died within 24 hr. pH 4.0 - all metamorphosed.									
				Buffered solutions. Up to 59 days exposure.									
				Porter and Hakanson, 1976									

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE			
pH (cont'd)	- <u>Ambystoma maculatum</u>		embryos at or prior to 4th cleavage division	12°C		21°C		At low temperature egg masses at pH ≤ 3.5 turned milky-opaque within 2 hr of exposure; at pH 4.5 embryos tightly coiled, those that hatched were deformed.	Dale et al., 1985
				pH	n	% hatch	n		
				7.0	48	23	54	43	
					52	52	47	51	
				6.0	86	6	62	40	
					85	26	42	81	
				5.0	67	40	75	32	
					57	11	76	46	
				4.5	56	2	118	2	
					111	9	56	11	
				4.0	62	0	72	0	
					42	0	83	1	
				3.5	> 50	0	> 50	0	
					> 50	0	> 50	0	
	- <u>Hyla crucifer</u>	35 per treatment	embryos mid-blastula stage	pH	% hatch		% hatch, later died		Temp. 20°C. All hatched larvae at pH 4.0 were deformed.
				8.0	85		0		
				7.0	80		0		
				6.0	80		0		
				5.0	77		11		
				4.0	54		100		
				3.0	0		-		

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS					REMARKS	REFERENCE
				12°C			21°C			
pH (cont'd)	<u>Rana sylvatica</u>		embryo at or prior to 4th cleavage division	pH	n	% hatch	n	% hatch	At pH 4.0, those which hatched at low temperature died.	Dole et al., 1985 (cont'd)
				7.0	451	92	473	92		
					409	95	213	98		
				6.0	338	51	378	77		
					318	46	346	80		
				5.0	473	51	262	97		
					357	39	319	90		
				4.5	340	38	337	95		
					296	40	323	82		
				4.0	320	17	119	8		
	313	6	276	< 1						
3.5	> 200	0	> 200	0						
	> 200	0	> 200	0						

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS			REMARKS	REFERENCE
				pH	% survival after 38 days at 5°C	% survival after 14 days at 21°C		
pH (cont'd)	<u>Rana clamitans</u>	10 per replicate	larvae stage 25-29	6.0	40	100		Dole et al., 1985 (cont'd)
					20	100		
					60	100		
				5.0	80	100		
					80	100		
					80	100		
				4.5	80	100		
					90	100		
					60	100		
				4.0	50	100		
	90	100						
	80	100						
3.5	30	100						
	60	100						
	60	100						
3.3	-	50						
		0						
		0						

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS			REMARKS	REFERENCE	
				pH	% hatch	% hatched, later died			
pH (cont'd)	<u>Rana palustris</u>	43 37 41 53 49 22 40 44 39 48	embryos at or before 32 cell stage	7.0	100	0	Temp. 12°C.	Dale et al., 1985 (cont'd)	
				6.0	100	0			
				5.0	41	50			
				4.5	5	50			
				4.3	0	-			
				4.3	0	-			
				4.3	0	-			
				4.3	0	-			
				4.3	0	-			
				4.3	0	-			
	<u>Bufo americanus</u>	46-51 per replicate	embryos early gastrula	pH		% hatched	Temp. 21°C.		
				6.0		100			
				4.5		100			
				4.3		100			
				4.0		82			
				3.8		84			
	<u>Notophthalmus viridescens</u>	10 per replicate	adults	pH		% survival after 38 days at 5°C	% survival after 14 days at 21°C		
				5.0		100	100		
				5.0		100	100		
				5.0		100	100		
				5.0		100	100		
5.0					100	100			
3.5					10	75			
3.5					20	87			
3.5		30	50						

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS			REMARKS	REFERENCE
				pH	A. jeffersonianus	R. sylvatica		
pH (cont'd)	<u>Ambystoma maculatum</u>	Approx. 150 per treatment	eggs at grey crescent stage	Initial perivitelline fluid pH's decreased significantly 7 to 96 hr after initial immersion of eggs in water altered to pH 4.0 and 5.0. Rate of H <sup>+</sup> flux into the fluid is greater in eggs in pH 4 than in those in pH 5 or 6.				Robb and Jones, 1987
				At pH 3.0, 100% mortality occurred after 1.25 hr larval immersion; at pH 3.5, 75% mortality occurred after 2.5 hr; at pH 4, 50% had mortality not occurred after 6 days.				
				Trends observed for pH 3.6 - 6.5: - maximum size attained was positively correlated with pH - time to foreleg emergence was negatively correlated with pH.				
	<u>Rana temporaria</u>	12 per treatment	larvae newly hatched				Tadpoles raised in given pH to metamorphosis.	Cummins, 1986
	<u>Ambystoma jeffersonianum</u> <u>Rana sylvatica</u>	20 per pH	embryos	Mean % hatchings:			In artificial soft water.	Froda and Dunson, 1986
				pH	A. jeffersonianus	R. sylvatica		
				5.80	100	95		
				4.75	100			
				4.50	25	100		
			4.25	0	80			
			4.00	0	0			
* significant difference between the two species.								

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE																																																	
pH (cont'd)	<u>Hyla andersoni</u> <u>Bufo woodhousei</u> <u>Rana clematis</u>	40 40 20	embryo	Percent hatching: <table border="1"> <tr> <td></td> <td colspan="2"><u>B. woodhousei</u></td> <td colspan="2"><u>H. andersoni</u></td> <td colspan="2"><u>R. clematis</u></td> </tr> <tr> <td>pH</td> <td>ASD</td> <td>BD</td> <td>ASD</td> <td>BD</td> <td>ASD</td> <td>BD</td> </tr> <tr> <td>5.8</td> <td>100</td> <td></td> <td>100</td> <td></td> <td></td> <td></td> </tr> <tr> <td>4.25</td> <td>100</td> <td></td> <td>90</td> <td></td> <td>100</td> <td>35</td> </tr> <tr> <td>4.10</td> <td>85</td> <td>45</td> <td></td> <td>93</td> <td></td> <td></td> </tr> <tr> <td>4.00</td> <td>0*</td> <td>0</td> <td>97**</td> <td>85</td> <td>100</td> <td>10</td> </tr> <tr> <td>3.75</td> <td>0</td> <td>0</td> <td>25</td> <td>20</td> <td>0</td> <td></td> </tr> </table>		<u>B. woodhousei</u>		<u>H. andersoni</u>		<u>R. clematis</u>		pH	ASD	BD	ASD	BD	ASD	BD	5.8	100		100				4.25	100		90		100	35	4.10	85	45		93			4.00	0*	0	97**	85	100	10	3.75	0	0	25	20	0		Foto done in artificial soft water (ASW) or naturally acidic bog water (BW). Growth significantly reduced at lower pH's during first 10 days.	Freda and Duncan, 1986 (cont'd)
		<u>B. woodhousei</u>		<u>H. andersoni</u>		<u>R. clematis</u>																																																	
	pH	ASD	BD	ASD	BD	ASD	BD																																																
	5.8	100		100																																																			
4.25	100		90		100	35																																																	
4.10	85	45		93																																																			
4.00	0*	0	97**	85	100	10																																																	
3.75	0	0	25	20	0																																																		
- <u>Rana sylvatica</u>	20-30 per treatment	eggs	larvae	At pH 7.2 - 7.6 hatching success was 95.3%, success dropped significantly to 86.6% at pH 4 and to 41.9% at pH 3.75. Time required for hatching increased significantly as pH decreased.  All survived for 24 hours at pH 7.2 - 7.6; at pH 3.5 only 37% survived and average survival time was 18.4 ± 0.41 hr.	At pH 4 only slight differences observed in embryo tolerance among progeny of males mated to same females; progeny from different females differed significantly in acid tolerance.	Pierce and Sikard, 1985																																																	
- <u>Rana catesbeiana</u>	30 per pH	tadpoles stages 25-30		Percent mortality increased from near 0% at pH 4.3 to 78% at pH 4.2; mortality was 100% at pH 3.9.	Exposure for 4 days.	Gascon and Bider, 1985																																																	
<u>Rana clematis</u>	30 per pH	tadpoles stages 25-30		Mortality (estimated from graph) was 10% at pH 4.5 and 90% at 4.1.																																																			
- <u>Rana pipiens</u> <u>Xenopus laevis</u>	10 9	adult/skino		Lowering of pH caused a decrease in short circuit current generated by the skins of both species.	Skin of <u>X. laevis</u> was more sensitive than that of <u>R. pipiens</u> to acid stress.	Frasco, 1981																																																	

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE																																		
pH (cont'd)	- <u>Ambystoma maculatum</u> <u>A. jeffersonianum</u>		embryo	<u>A. maculatum</u> - tolerated pHs from 6-10 - greatest hatching success at 7-9.  <u>A. jeffersonianum</u> - tolerated pHs from 4-8 - greatest hatching success at 5-6.	pH optimum shifted upward with increasing temperature for <u>A. jeffersonianum</u> and downward for <u>A. maculatum</u>	Pough and Wilson, 1977																																		
	- <u>Acris gryllus</u> <u>Pseudacris nigrita</u> <u>Hyla andersoni</u> <u>H. crucifer</u> <u>H. versicolor</u> <u>Rana catesbeiana</u> <u>R. clematis</u> <u>R. palustris</u> <u>R. pipiens</u> <u>R. sylvatica</u> <u>R. virgatipes</u>			Effects of acidity on development of 11 species: <table border="1"> <tr> <td></td> <td>A*</td> <td>B**</td> </tr> <tr> <td></td> <td>4.6</td> <td>4.1</td> </tr> <tr> <td></td> <td>4.1</td> <td>3.8</td> </tr> <tr> <td></td> <td>3.8</td> <td>-</td> </tr> <tr> <td></td> <td>4.2</td> <td>3.8</td> </tr> <tr> <td></td> <td>4.3</td> <td>-</td> </tr> <tr> <td></td> <td>4.3</td> <td>-</td> </tr> <tr> <td></td> <td>4.1</td> <td>-</td> </tr> <tr> <td></td> <td>4.3</td> <td>4.0</td> </tr> <tr> <td></td> <td>4.1</td> <td>3.7</td> </tr> <tr> <td></td> <td>3.9</td> <td>-</td> </tr> <tr> <td></td> <td>3.8</td> <td>-</td> </tr> </table>		A*	B**		4.6	4.1		4.1	3.8		3.8	-		4.2	3.8		4.3	-		4.3	-		4.1	-		4.3	4.0		4.1	3.7		3.9	-		3.8	-
	A*	B**																																						
	4.6	4.1																																						
	4.1	3.8																																						
	3.8	-																																						
	4.2	3.8																																						
	4.3	-																																						
	4.3	-																																						
	4.1	-																																						
	4.3	4.0																																						
	4.1	3.7																																						
	3.9	-																																						
	3.8	-																																						
Phenol	- <u>Xenopus laevis</u>		embryo	No teratogenic effect. No effect on embryos at 50 ppm, but animals died within 6 days to 3 weeks of completing embryonal development.		Dumert, 1987																																		
Phenols	- Assorted species/review			Cellular neurophysiological effects reviewed.		Kasla, 1982																																		

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE		
-Phenyl-s- aphthylamine component of jet and rocket fuel)	- <u>Rana pipiens</u> <u>Xenopus laevis</u>	100-176 per treatment	embryo	All <u>R. pipiens</u> embryos exposed to 20 or 200 mg/L developed malformations and all were dead by heart beat stage; 96.8% of <u>X. laevis</u> exposed to 6 mg/L developed malformations while exposure to 15.2 mg/L produced no observable effects.	Estimated ED <sub>50</sub> = 4-5 mg/L. Malformations included shortened trunk and intestinal tract and edema.	Greenhouse, 1976a,c		
			embryo	Conc. (ppb)	Mortality (%)	Abnormal Embryos (%)	Exposure for 24 hr. no. survivors determined 120 hr after exposure. Edema was most common defect. 500 ppb induced similar deformities in <u>Castrophyne carolinensis</u> .	Fulton and Chambers, 1985
	<u>Rana sphaerocephala</u>	120 160 210 140		0 100 500 1000	0 0 6 99	0 0 53 100		
	<u>Hyla chrysaecelis</u>	161 86 84 80 160 40		0 100 250 500 750 1000	2 7 10 32 52 92	0 0 0 10 58 100		
thodran (OP)	- <u>Rana pipiens</u>	24 in each concentration	adults	Following exposure to concentrations of 6, 9, 12, 16, 20 ppm for 15 days: - Produced anemia and leucopenia which worsened with increasing concentration - Differential white cell count showed progressive lymphocytosis and neutropenia with successively higher concentrations - Decrease in activity, flaccid paralysis.		Keplan and Giaczenski, 1965		

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE	
Pollution (Micropollutants in drinking water)	- <u>Pleurodeles waltl</u>		Larvae approx. 40 mm	Larvae reared in untreated tap water had higher levels of micronucleated erythrocytes than those reared in filtered treated water.	Used to evaluate mutagenic activity in drinking water.	Jeylet et al., 1987	
(Textile Plant Effluent)	- <u>Rana cyanophlyctis</u>	10	adults	25% mortality after 96 hr, 50% after 144 hr.	Frogs maintained in effluent.	Shrinivas et al., 1982	
Potassium Chromate	- <u>Rana hexadactyla</u>		gastrocnemius muscle	Percent change in succinate dehydrogenase (SDH) and Hg <sup>+2</sup> ATPase activity levels over control levels in muscle soaked in potassium chromate (A), well water contaminated by factory effluent (B) or factory effluent (C):	Exposure - 10 min. Contamination of effluent from a chromate and chemical factory examined.	Rajendrababu and Nandakumar, 1987	
				Conc. (ug/ml)	% change SDH activity	% change Hg <sup>+2</sup> ATPase activity	
				A 10	-8.50 ± 0.41	-7.12 ± 0.35	
				50	-24.45 ± 1.2	-9.0 ± 0.45	
				100	-32.28 ± 1.36	-17.11 ± 0.71	
				200	-47.17 ± 2.31	-36.31 ± 1.51	
				500	-59.24 ± 2.71	-46.43 ± 2.31	
				B 15%	-54.23 ± 2.71	-43.62 ± 2.12	
				C 15%	-50.00 ± 2.50	-27.64 ± 1.36	
Potassium Dichromate	- <u>Xenopus laevis</u>	50 per treatment 60 per treatment	tadpoles stage 38 eggs	Only those kept in concentrations up to 10 ppm developed into toads. In 5 and 7.5 ppm, 15 ± 5% and 30 ± 15%, respectively, of embryos died; weaker pigmentation in those at 2.5 ppm.	Exposure for 90 days.	Ompert, 1987	

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE
Procteryna (H) (Gosgard 50)	- <u>Rana temporaria</u>	500 per treatment	tadpoles just after hatching or after completion of oporevica	All younger tadpoles treated with 0.05% suspension for 3 days died within a few days. All older tadpoles exposed to 0.01 or 0.05% suspension died after 24-72 hr.	Static toxic.	Jordan et al., 1977
Pyrazophos (F) (Aflugon)	- <u>Rana temporaria</u>		tadpoles	1 mg/L - lethal dose. .1 mg/L - attenuation of growth, total inhibition of metamorphosis. - marked change of transaminase enzymatic activity.		Poulav, 1981
Pyrethroids	- <u>Xenopus laevis</u>		myelinated nerve fibres	Closing of the activation gate in a fraction of the sodium channels that open during depolarization delayed, resulting in a prolonged sodium tail current after repolarization of membrane.	Rate of dissociation decreased gradually between 25° - 0°C.	Vijverberg and Ruigt, 1981
	- Assorted species/ frogs		nerve system	Principal effect is to induce repetitive activity, particularly in sensory nervous system.		Vijverberg and van den Bercken, 1982
Selenium (sodium selenite)	- <u>Xenopus laevis</u>	50-100 per treatment	tadpoles	Tadpoles surviving continuous exposure to 2, 5, 10 ppm had cellular damage, including disorganization and degeneration, in epithelial and muscle cells. Damage more extensive at higher doses.		Brooks and Dumont, 1980
	- <u>Xenopus laevis</u>		embryo tadpoles	Concentrations of 2 ppm and above resulted in severe chromatid and increased mortality. Toxicity increased with increasing concentration up to 20 ppm.		Brooks and Dumont, 1979

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE														
Sodium Diethyldithiocarbamate (F) (NaDEDC)	- <u>Microphyla ornata</u>		embryo	5 mg/L - highly embryotoxic, growth and development retardation, death in "about 24 hr". 3 mg/L - survivors were "highly retarded" and malformed. After 72 hr - general oedema, abnormal bending of body axis, kinky notochord. 2 and 1 mg/L same effects, less severe. At 0.5 mg/L tadpoles more or less normal - tail slightly shortened and broader.	NaDEDC is potent teratogen. Mainly abnormalities in notochord.	Ghata and Mulherker, 1980a														
Sodium Dodecylbenzenesulfonic Acid	- <u>Xenopus laevis</u>	60 per treatment	eggs	No effect at 0.1, 1, 5 and 10 ppm. At 50 ppm, eggs did not develop past 2 cell stage.		Dampert, 1987														
Sodium Fluoroacetate	- Green Frog		sciatic nerves	No action potential changes produced by up to 100 mM. 0.01 and 0.10 M produced 3 and 40% respectively, inhibition of respiration after 3 hr.		Boysarsky et al., 1989														
Styrene	- <u>Rana pipiens</u> <u>R. temporaria</u>	7	adults	Extensive ultrastructural alterations in olfactory epithelium following exposure to 665 ± 30 ppm for 60 min: - increased secretion from sustentacular cells - membrane fusion of cilia. Reduction in EOG.		Eklow et al., 1984														
Temephos (OP)	- <u>Hyale regilla</u>		tadpoles 3 weeks old	Thermal tolerance significantly lowered:	24 hr exposure.	Johnson, 1980														
				<table border="1"> <thead> <tr> <th rowspan="2">Conc. (ppb)</th> <th colspan="2">Onset of Spoons (°C)</th> </tr> <tr> <th>Mean</th> <th>Range</th> </tr> </thead> <tbody> <tr> <td>Control</td> <td>37.5</td> <td>36.9-37.8</td> </tr> <tr> <td>25</td> <td>36.2</td> <td>34.6-37.1</td> </tr> <tr> <td>50</td> <td>35.5</td> <td>34.2-36.8</td> </tr> </tbody> </table>	Conc. (ppb)	Onset of Spoons (°C)		Mean	Range	Control	37.5	36.9-37.8	25	36.2	34.6-37.1	50	35.5	34.2-36.8	Recommended dosage rate for mosquito control in California - 50 ppb.	
Conc. (ppb)	Onset of Spoons (°C)																			
	Mean	Range																		
Control	37.5	36.9-37.8																		
25	36.2	34.6-37.1																		
50	35.5	34.2-36.8																		
		54																		
		39																		
		37																		



Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE	
EPP (OP) tetraethyl pyrophosphate)	- <u>Rana pipiens</u>	24 in each con- centration	adults 9 cm	Following exposure to concentrations of 10, 20, 40, 60, 80 ppm for 15 days: - With increasing concentrations, anorexia and leucopenia became progressively more marked - Differential white cell count showed neutropenia and lymphocytosis - Red blood cells distorted in shape - Visceral organs desiccated, spasticity in hindlimbs.		Kaplan and Glaczenski, 1965	
	- <u>Bufo varidis</u>	3	adults  blood and nervous tissue	Residual cholinesterase at time of death following injection of 2 LD50 into dorsal lymphatic sac:  15%  23%	LD50 - 540 ppm.	Ebery and Schatzberg-Porath, 1960	
2,3,7,8 tetrachlorodibenzo- dioxin (TCDD)	- <u>Rana catesbeiana</u>	15 per group	tadpoles	Dose (ug/kg)	% survival on day 50 post-injection (i.p.)	All surviving tadpoles successfully completed metamorphosis with no morphological abnormalities.	Batty et al., 1976
				0	80		
				25	87		
				50	73		
				100	93		
				200	80		
				1000	80		
		5 per group	adults 150-250 g	No mortality during 35 day observation at same doses as above. Some lessened food intake in group injected with 500 ug/kg in early phase. No histopathologic lesions were found at any dose level.			

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE
Challium (M)	- Frogs		embryos	In N/500,000 tadpoles killed on emergence.		Dilling and Healey, 1926
Chorus (M)	- Frogs		embryos	Development to tadpole stage in N/100,000 was 50%.		Dilling and Healey, 1926
MBE (tert.-butyl methyl ether) anti-detonation preparation)	- <u>Rana temporaria</u>		tadpoles	≤2000 ppm had no lethal effect. 100 ppm in water led to increased weight, stimulated course of metamorphosis.		Paulov, 1987c
Chloroform	- <u>Triturus cristatus</u>		hematopoietic system	Exposure via immersion 5 times/week at 5 ppm or 10 ppm caused macrocytic hypochromic anemia which was more severe at higher concentrations.		Garavini and Seren, 1979
	- <u>Rana pipiens</u> - <u>R. temporaria</u>	17	adults	Structural alterations in olfactory mucosa following exposure to 585 ppm for 1 hr: - increased secretion from sustentacular cells. Reduction in EDG in frogs exposed to 2000 ppm for 1 hr; no reduction in those exposed to 585 ppm.		Eklom et al., 1984
Chloroform Disso- cyanate	- <u>Rana pipiens</u>		erythrocytes	Inhibited isoproterenol- and fluoride ion - stimulated adenylyate cyclase activity in a dose dependent manner.	Suggests non-specific inhibition of activity.	McKay and Brooks, 1983

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS			REMARKS	REFERENCE
				Ionophore (ppm)	pH of solution at 25° C	No. of frogs dead at 30 days		
Toxaphene (OC)	<u>Rana pipiens</u>		adults 65 g, 8.9 cm				Frogs placed in 200 cc of test solution.	Kaplan and Overpeck, 1960
		20		0.60	5.83	5	Neuro-muscular changes were produced; excessive thrashing and abnormal reactivity to stimulation were observed.	
		10 10		0.45 0.30	5.76 5.75	0 0		
Tributyltin (oxide and fluoride)	<u>Rana temporaria</u>	10 per treatment	embryo post-gastrula	Survival not affected at 0.3 or 3 ppb; mortality was 40% and 50% at 30 ppb TBFO and TBTF, respectively. Rights discarded only at 30 ppb.			Exposure for 5 days.	Laughlin and Linden, 1982
Trichlorophen (OP)	<u>Rana temporaria</u>	30 per group	blood	Erythrocyte count, haemoglobin level and hematocrit value were decreased following administration of 50, 100, 300 or 3 x 100 mg/kg via dermal lymphatic sac. 50 and 100 mg/kg decreased leucocyte count; 300 mg/kg caused no change; 3 x 100 mg/kg caused leucocytosis.				Szubartowska, 1979
	<u>Rana temporaria</u>	30 per treatment	adults	3 hr following injection of 50, 100, 300 or 3 x 100 mg/kg b.w. into dermal lymphatic sac: - Significant reduction in no. erythrocytes and erythroblasts - Reduction in haemoglobin level and hematocrit value - Changes in no. and % age composition of leucocytes and increased thrombocyte content.				Graczyk - Kalkowska and Szubartowska, 1986
Tri-n-tolyl Phosphate	<u>Hylo chrysocelis</u>		embryo	Not toxic or teratogenic at 10 ppm.			Derally exposed for 24 hr, no. survivors counted 120 hr post-exposure.	Fulton and Chambers, 1985

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS			REMARKS	REFERENCE
Trisodium Nitratotriacetate (Na <sub>3</sub> NTA)	Leopard frog		tadpoles	No significant mortality occurred in buffered solutions of 600 mg/L or less.			With pH 9.7 or greater, animals died within an hour irrespective of NTA concentration.	Flannagan, 1971
	Salamander sp.		larvae	Survived up to 350 mg/L over 96 hr in buffered solutions.				
Trithion (OP)	<u>Rana pipiens</u>	24 per concentration	adults	Following exposure to concentrations of 120, 140, 160, 180, 200 ppm for 15 days: - Progressive anaemia occurred with increasing concentration - White cell count dropped progressively - Neutropenia, lymphocytosis - Flaccid paralysis, excessive skin shedding.				Kaplan and Giacconski, 1965
Uranyl Ion (UO <sub>2</sub> <sup>+2</sup> )	<u>Triturus pyrrhogaster</u>		stomach mucous epithelial cells	Almost no effect on the electrical properties of the cell at 10 <sup>-6</sup> M to 10 <sup>-4</sup> M.				Kanno et al., 1978
S-cis-Verbenol	<u>Rana temporaria</u>		tadpoles	≥15 ppm was lethal.				Paulov et al., 1985

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE			
Zinc (M)	<u>Gastrophryne carolinensis</u>	100	eggs	Percent mortality and teratogenesis at hatching (H) and 4 days posthatching (PH) of eggs exposed to sediment-bound zinc:	Hardness - 200 ppm as CaCO <sub>3</sub> . pH - 7.5 - 8.0. Percent mortality expressed as frequency in experimental population/controls. * Percentages of survivors bearing gross congenital deformities at hatching.	Birge et al., 1977			
				Zn added to sediment (ppm)			Concentration in sediment (ppm)	% mortality	
								H	PH
				0.1			104.6	6 (3)	14
1.0	112.6	3 (0)	5						
10.0	124.5	7 (1)	14						
100.0	222.7	7 (2)	8						
	- Frogs		tadpoles	Few survived long in N/20,000; those which survived in N/50,000 for >3 months were stunted and had no limb buds.		Dilling and Healey, 1926			
	- <u>Triturus pyrrhogaster</u>		stomach mucous epithelial cells	10 <sup>-4</sup> M Zn <sup>+2</sup> decreased membrane potential to 50% of the control value. Effect increased with increasing concentration from 10 <sup>-7</sup> to 10 <sup>-4</sup> M.	Action antagonized by cysteine and acetylpenicillamine.	Karno et al., 1978			
	- <u>Bufo boreas</u>		larvae	39 mg/L Zn <sup>+2</sup> - all died within 24 hr. 0.1 mg/L - all metamorphosed.	Up to 61 days exposure.	Porter and Hakanson, 1976			
	- <u>Xenopus laevis</u>		myelinated nerve fibres	3.4 mM slowed down kinetics of K systems; decreased the permeability constant, increased the time constant, shifted the K activation curve along the potential axis.		Arhem, 1980			

Table 2 - Laboratory Studies (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES	N	STAGE/TISSUE	LEVELS USED/EFFECTS	REMARKS	REFERENCE				
Zinc Sulphate (see also cadmium chloride)	<u>Xenopus laevis</u>	20 per test	tadpoles stage 54-58	Mortality (%) of pre-treated (P) and non pre-treated (C) tadpoles:	Pretreatment with 5.0 mg/l Zinc sulphate or 2.5 mg/l cadmium chloride for 96 hr.	Woodell et al., 1988				
				Pre-treat./ Exposure Metal			Cd conc. (mg/l)	Time (hr)		
								15	45	75
				Zn/Zn			10 P	0	0	0
							C	37	80	80
							15 P	0	0	0
							C	14	33	45
				Cd/Zn			20 P	4	15	15
							C	44	50	50
							10 P	0	0	0
							C	0	10	18
							15 P	0	0	0
C	8	15	18							
20 P	4	5	5							
C	12	26	40							

<sup>a</sup> OC - organochlorine insecticide; OP - organophosphate insecticide; C - carbamate; IGR - insect growth regulator; PY - pyrethroid; A - acaricide; H - herbicide; F - fungicide; M - metal

TABLE 3  
FIELD STUDIES

Contaminant <sup>a</sup>	Location/Habitat Species	Rate and Method of Application	Reported Effects on Amphibians	Water Temperature, Residue Levels, Miscellaneous	Reference																		
Acephata (OP) (Orthene)	- Larose Forest, Ontario Mixed wood stands <u>Rana sylvatica</u> , <u>Hyla crucifer</u> , <u>Ambystoma laterale</u>	0.56 kg active ingredient applied by fixed-wing aircraft flying just above canopy; Micronair AV-3000 delivery system.	Caged <u>R. sylvatica</u> tadpoles not affected. No mortality noted in natural populations of 3 species.	- $\bar{x}$ in ponds after spray - 0.3 L/ha. - $\bar{x}$ around ponds - 0.2 L/ha.	Buckner and Macleod, 1975																		
Aldrin (OC)	- 0.025 ha pond in California <u>Rana catesbeiana</u>	Sprays prepared from emulsifiable concentrate formulations and applied at rate of 89.9 L/ha from 3 gallon (12.45 L) sprayers.	Application Rate (Active ingredient) (kg/ha)	Water depth 20.32 - 30.48 cm. pH 7.5 - 8.0.  Tests conducted from June to October 1962. Maximum daily water temp. 26.6° - 35°C. One pond per treatment; N = 20 caged tadpoles (age unspecified) per pond.	Mulla, 1963																		
			% 24 hour or cumulative mortality days after treatment																				
			<table border="1"> <tr> <td></td> <td>1</td> <td>2</td> <td>5</td> <td>6</td> <td>7</td> </tr> <tr> <td>0.11</td> <td>10</td> <td>30</td> <td>30</td> <td>-</td> <td>-</td> </tr> <tr> <td>0.56</td> <td>80</td> <td>100</td> <td>*</td> <td>0</td> <td>-</td> </tr> </table>		1	2	5	6	7	0.11	10	30	30	-	-	0.56	80	100	*	0	-		
	1	2	5	6	7																		
0.11	10	30	30	-	-																		
0.56	80	100	*	0	-																		
	- Field trials in California <u>Rana catesbeiana</u>	.11 kg/ha. .56 kg/ha.	Deemed 'toxic' to tadpoles.		Mulla, 1962																		
Aluminum	- Temporary woodland ponds in coastal Maryland <u>Ambystoma maculatum</u>		Embryonic survival negatively correlated with aluminum concentration (range 0.11-0.51 ppm) in water.		Albers and Prouty, 1976																		
Ambithion (OP) (malathion + fenitrothion)	- Indian rice fields Frog species unspecified.	Sprayed by helicopter from height of 2-3 m over fields at 1.0 kg/ha between 7:30 and 9:00 a.m.	90% mortality.	Population estimates made 48 hrs. after spraying.	Thirumurthi et al., 1973																		

Table 3 - Field Studies (cont'd)

Contaminant <sup>a</sup>	Location/Habitat Species	Rate and Method of Application	Reported Effects on Amphibians	Water Temperature, Residue Levels, Miscellaneous	Reference
Azinocarb (C) (Matacil)	- Laurentians, Quebec Balsam fir, black spruce, hazel, cherry, aspen, maple. Grasses: rubus, honeysuckle, wildlily, blueberry <u>Rana pipiens</u> , <u>R. sylvatica</u> , <u>Bufo americana</u>	175 g active ingredient in 2.24 L/ha. 150 ha sprayed June 28, 1979 by fixed-wing aircraft.	<u>R. pipiens</u> and <u>R. sylvatica</u> - no significant change in activity over short term. <u>B. americana</u> - activity decreased during 2 months post-spray possibly related to reduced prey availability.	Date collected July 12 - Aug. 31, 1978 and June 1 - Aug. 31, 1979. 2 sites - 25 km apart.	Bracher and Bider, 1979
	- Richibucto, New Brunswick <u>Rana clamitans</u> assorted other species	Aerially applied at 0.11 kg/ha.	Mortality in caged <u>R. sylvatica</u> tadpoles; no other toxic effects noted.		Rick and Price, 1974
	- Unspecified	0.04 kg/ha.	Adult and larval amphibians were apparently unaffected.		Pearce and Price, 1974
Arco (Larvicide)	- Butte County, California, 2 test ponds - 3.4 x 38.5 m with 4.3 m between them <u>Rana catesbeiana</u>	22.4 L/ha applied using a hand sprayer on June 15, August 3, 24, and September 8, 1971.	No dead frogs were observed after August 24 and September 8 treatments.	Water depth at centre 0.6 m; water temperature 20.5-25.5°C. <u>R. catesbeiana</u> migrated into the ponds prior to the 3rd treatment.	Hagen et al., 1973
Azinphos-methyl (OP) (Guthion)	- 0.025 ha pond Location unknown <u>Bufo boreas</u> <u>Scaphiopus hammondi</u>	Applied as aqueous spray at rate of 89.9 L/ha. Active ingredient = 0.11 and 0.45 kg/ha.	No mortality after 24 hours.	Water depth 20.32 - 30.48 cm. 20 tadpoles caged in pond.	Mulla et al., 1963
	- Field tests <u>Rana catesbeiana</u>	.45 kg/ha.	"Safe" to tadpoles.		Mulla, 1962

3 - Field Studies (cont'd)

Pesticide	Location/Habitat Species	Rate and Method of Application	Reported Effects on Amphibians	Water Temperature, Residue Levels, Miscellaneous	Reference
er 22408 )	- 0.025 ha pond Location unknown <u>Rana catesbeiana</u>	Applied as aqueous spray at rate of 89.9 L/ha. Active ingredient = 0.56 and 2.24 kg/ha.	No mortality after 24 hours.	Water depth 20.32 - 30.48 cm. 20 tadpoles caged in pond.	Mulla et al., 1963
er 29952 )	- 0.025 ha pond Location unknown <u>Rana catesbeiana</u>	Applied as aqueous spray at rate of 89.9 L/ha. Active ingredient = 0.11 and 0.45 kg/ha.	Application Rate    % 24 hr mortality .11                    0 .45                    5	Water depth 20.32 - 30.48 cm. 20 tadpoles caged in pond.	Mulla et al., 1963
er 34042 )	- 0.025 ha pond Location unknown <u>Rana catesbeiana</u>	Applied as aqueous spray at rate of 89.9 L/ha. Active ingredient = 0.11 and 0.45 kg/ha.	No mortality after 24 hours.	Water depth 20.32 - 30.48 cm. 10 tadpoles caged in pond.	Mulla et al., 1963
er 37289 )	- 0.025 ha pond Location unknown <u>Bufo boreas</u> <u>Scaphiopus hammondi</u>	Applied as aqueous spray at rate of 89.9 L/ha. Active ingredient = 0.11 and 0.45 kg/ha.	No mortality after 24 hours.	Water depth 20.32 - 30.48 cm. 20 tadpoles caged in pond.	Mulla et al., 1963
	- Field tests in California <u>Bufo boreas</u> <u>Scaphiopus hammondi</u>	.11 kg/ha. .45 kg/ha.	Deemed 'safe' at these application rates for tadpoles of both species.		Mulla, 1962
er 38920 )	- 0.025 ha pond Location unspecified <u>Rana catesbeiana</u>	Applied as aqueous spray at rate of 89.9 L/ha. Active ingredient = 0.56 and 2.24 kg/ha.	Application Rate    % 24 hr Mortality 0.56                    100 2.24                    100	Water depth 20.32 - 30.48 cm. 10 tadpoles caged in pond.	Mulla et al., 1963
	- Field tests in California <u>Rana catesbeiana</u>	0.56 kg/ha.	Caused 100% mortality to tadpoles.		Mulla, 1962

3 - Field Studies (cont'd)

Pesticide	Location/Habitat Species	Rate and Method of Application	Reported Effects on Amphibians	Water Temperature, Residue Levels, Miscellaneous	Reference
er 44831 )	- 0.025 ha pond Location unspecified <u>Rana catesbeiana</u>	Applied as aqueous spray at rate of 89.9 L/ha. Active ingredient = 0.22 and 0.90 kg/ha.	No mortality after 24 hours.	Water depth 20.32 - 30.48 cm. 20 tadpoles caged in pond.	Mulla et al., 1963
bofuran (C) radan 3)	- Rice fields, Texas <u>Acris crepitans blanchardi</u>	.56 kg/ha carbofuran applied from fixed-wing aircraft 5 to 8 weeks after rice planted. 6 applications.	Frogs found paralyzed or exhibited abnormal behaviour 15 minutes after treatment. Only 1 death recorded.	Study dates - May 2 and July 7, 1970 and 1973-75. Searches conducted 15 mins. - 11 days post-treatment. Rice was 30-50 cm high and in 20-30 cm of water.	Flickinger et al., 1980
bopheno- on (OP) ithion)	- 0.025 ha pond Location unknown <u>Rana catesbeiana</u>	Applied as aqueous spray at rate of 89.9 L/ha. Active ingredient = 0.45 and 1.79 kg/ha.	100% mortality after 24 hours at both rates.	Water depth 20.32 - 30.48 cm. 10 tadpoles caged in pond.	Mulla et al., 1963
	- Field test in California <u>Rana catesbeiana</u>	.45 kg/ha.	100% mortality of tadpoles.		Mulla, 1962
ordana (OC)	- 0.025 ha pond in California <u>Rana catesbeiana</u>	Sprays prepared from emulsifiable concentrate formulations and applied at a rate of 89.9 L/ha from 3 gallon (12.45 L) sprayers.	Application Rate (Active ingredient) (kg/ha)	Water depth 20.32 - 30.48 cm. pH 7.5 - 8.0.	Mulla, 1963
			% 24 hour or cumulative mortality days after treatment	Tests conducted from June to October 1962. Maximum daily water temp. 26.6°-35°C. One pond per treatment; N = 20 caged tadpoles (age unspecified) per pond.	
		0.11	1 2 3 4 5 6 7		
		0.56	0 30 30 30 -		

Table 3 - Field Studies (cont'd)

Contaminant <sup>a</sup>	Location/Habitat Species	Rate and Method of Application	Reported Effects on Amphibians	Water Temperature, Residue Levels, Miscellaneous	Reference
Chlordane (OC) (cont'd)	- Field trials in California <u>Rana catesbeiana</u>	.11 kg/ha. .56 kg/ha.	Deemed 'safe' to tadpoles at these application rates.		Mulla, 1962
Cobalt 60	- Yosemite, California mountain meadow <u>Bufo canorus</u>  <u>B. boreas halophilus</u>	Tablets containing 20-30 uc used as radio-tagging device.  Subcutaneous tag of 40 uc.	3 toads survived 1 year.  Survived.		Keristrom, 1957
DDT (OC)	- Golf course greens <u>Bufo boreas</u>  - Stream in Algonquin Provincial Park, Ontario Bullfrogs, Green Frogs, Mink Frogs, Wood Frog  - Forest in northeastern Ontario Wood Frog  - 64 coastal plain ponds within 20 mile radius of Savannah, Georgia Frog species unspecified	Hand sprayed: 1.12 kg/ha. 2.24 kg/ha.  Aerially applied on July 21: 6.7 kg/ha.  Up to 4.5 kg/ha.  .11-.45 kg/ha DDT applied by rotary duster. 0.6-.22 kg/ha DDT applied by air pressure hand sprayer.	95-98% control of juvenile toads after 1 treatment.  14 of 22 caged frogs killed within 5 days of spray.  "Very slight" effects on amphibians.  A few frogs were killed by treatment of 0.11 DDT as dust or emulsion. DDT emulsions more toxic than solutions and dusts.	Combination DDT and toxaphene.  DDT - oil solution.  Formulations of both oil-water emulsions and standard oil solutions used.  Observations at 24 and/or 48 hours.  Weekly sprays.	Mulla, 1962  Lozier, 1949  Speirs, 1949  Ierzwell, 1950

Table 3 - Field Studies (cont'd)

Contaminant <sup>a</sup>	Location/Habitat Species	Rate and Method of Application	Reported Effects on Amphibians				Water Temperature, Residue Levels, Miscellaneous	Reference	
			Site/Cage	Behaviour of exposed tadpoles					
				Day 1	Day 3	Day 14			
DDT (cont'd)	- Great Britain <u>Rana temporaria</u>  a) Small ditch - 1 m wide Max. depth - 0.4 m  b) Larger ditch - 5-6 m wide Max. depth - 1.0 m  c) Pool - 11 x 16 m Max. depth - 1.3 m  d) 2 marshy pools (20 x 20 m; 4 x 30 m) separated by a bank Max. depth - 0.5 m and 0.1 m respectively  e) Gravel pit - 25-30 x 140 m Max. depth - 2.0 m	0.4-0.5 kg/ha applied on water surface in a 2 m wide band around water edge by hand operator.	a)/ 1)	Frantic*	Resigned	No survivors	a) cage 1 cage 2	Distance of caged tadpoles to bank central central	Cooke, 1975a
			2)	Frantic	Some frantic Some resigned	No survivors	b) cage 1 cage 2	1 m 1 m	
			b)/ 1)	Some normal Some frantic	Few normal Most frantic	Four normal One moribund	c) cage 1 cage 2	2 m 1 m	
			2)	Frantic	Most resigned Most frantic Few resigned	Thirteen normal Three moribund	d) cage 1 cage 2	4 m 1 m	
			c)/ 1)	Frantic	Frantic	Normal	e) cage 1 cage 2	5 m 7 m	
			2)	Frantic	Frantic	Normal			All survivors on day 3 had abnormal snouts at site a.
			* Frantic behaviour is characterized by incoordinated hyperactive activity.						No behavioural or morphological abnormalities noted at site d or e.

Table 3 - Field Studies (cont'd)

Contaminant <sup>a</sup>	Location/Habitat Species	Rate and Method of Application	Reported Effects on Amphibians	Water Temperature, Residue Levels, Miscellaneous	Reference																		
DDT (cont'd)	- Hubbard County, Minnesota Two small woodland pools <u>Rana sylvatica</u>	Aerial application.	2 and 33 hours after spray - frogs were as numerous and activity was normal. 21 days after spray - dead frogs found on each pool. On June 1 and July 2 no live frogs were found.	May 21 date of spraying.  Fent caterpillar larvae dropped into ponds, covering the surface. All but two frog stomach samples (n=36) contained caterpillars; not known if mortality caused by contact or accumulation of contaminated prey.	Fashingbauer, 1957																		
	- Four open ponds in Florida <u>Rana pipiens</u> <u>R. grylio</u> <u>Hyla c. cinerea</u>	Aerial application: 1 - 5% DDT at .34 kg/ha; 2 - 5% DDT at .67 kg/ha; 3 - 20% DDT at .34 kg/ha; 4 - 20% DDT at .67 kg/ha.	2 of 3 frogs from #4 returned to lab for observation ( <u>R. pipiens</u> , <u>R. grylio</u> ) showed lack of coordination symptoms. The third ( <u>H. cinerea</u> ) showed no symptoms. <u>R. grylio</u> died after 4 days; <u>R. pipiens</u> recovered in 10 days.	Canopy cover dispersed the spray.	Herald, 1949																		
	- 0.025 ha pond in California <u>Rana catesbeiana</u>	Sprays prepared from emulsifiable concentrate formulations and applied at a rate of 89.9 L/ha from 3 gallon (12.45 L) sprayers.	Application Rate (Active ingredient) (kg/ha)	% 24 hour or cumulative mortality days after treatment	Water depth 20.32 - 30.48 cm. pH 7.5 - 8.0.  Tests conducted from June to October 1962. Maximum daily water temp. 26.6° - 35°C. One pond per treatment; N = 20 caged tadpoles (age unspecified) per pond.	Mulla, 1963																	
		0.11 1.121	<table border="1"> <tr> <td></td> <td>1</td> <td>2</td> <td>5</td> <td>6</td> <td>7</td> </tr> <tr> <td>0.11</td> <td>0</td> <td>0</td> <td>0</td> <td>-</td> <td>-</td> </tr> <tr> <td>1.121</td> <td>30</td> <td>80</td> <td>*</td> <td>0</td> <td>-</td> </tr> </table>		1	2	5	6	7	0.11	0	0	0	-	-	1.121	30	80	*	0	-		
	1	2	5	6	7																		
0.11	0	0	0	-	-																		
1.121	30	80	*	0	-																		
		* 10 fresh tadpoles added.																					

Table 3 - Field Studies (cont'd)

Contaminant <sup>a</sup>	Location/Habitat Species	Rate and Method of Application	Reported Effects on Amphibians	Water Temperature, Residue Levels, Miscellaneous	Reference																	
Dichlobenil (H)	- 5 m diameter pond in Great Britain <u>Rana temporaria</u> <u>Bufo bufo</u> <u>Triturus vulgaris</u>	1 mg/L granules.	No deaths or changes in activity or development rate within 32 days.  <u>T. vulgaris</u> continued to breed.	Water depth 1-2 m in winter; 1 m in summer.  Frogs and toads were caged; newts were free living.	Cooke, 1977																	
Dieldrin (OC)	- 0.025 ha pond <u>Rana catesbeiana</u>	Sprays prepared from emulsifiable concentrate formulations and applied at a rate of 89.9 L/ha from 3 gallon (12.45 L) sprayers.	Application Rate (Active ingredient) (kg/ha)	% 24 hours or cumulative mortality days after treatment	Water depth 20.32 - 30.48 cm. pH 7.5 - 8.0.  Tests conducted from June to October 1962. Maximum daily water temp. 26.6° - 35°C. One pond per treatment; N = 20 caged tadpoles (age unspecified) per pond.	Mulla, 1963																
		0.11 0.56	<table border="1"> <tr> <td></td> <td>1</td> <td>2</td> <td>5</td> <td>6</td> <td>7</td> </tr> <tr> <td>0.11</td> <td>100*</td> <td>30</td> <td>30</td> <td>30</td> <td>-</td> </tr> <tr> <td>0.56</td> <td>100*</td> <td>100</td> <td>*</td> <td>30</td> <td>30</td> </tr> </table>		1	2	5	6	7	0.11	100*	30	30	30	-	0.56	100*	100	*	30	30	
	1	2	5	6	7																	
0.11	100*	30	30	30	-																	
0.56	100*	100	*	30	30																	
		* 10 fresh tadpoles added.																				
	- Pond in California <u>Rana catesbeiana</u>	.11 - .56 kg/ha.	Complete tadpole kill.		Mulla, 1962																	
Diquat (H)	- 5 m diameter pond in Great Britain <u>Rana temporaria</u> <u>Bufo bufo</u> <u>Triturus vulgaris</u>	1 mg/L sprayed.	No deaths or changes in activity or development rate within 32 days.  <u>T. vulgaris</u> continued to breed.	Water depth 1-2 m in winter; 1 m in summer.  Frogs and toads were caged; newts were free-living.	Cooke, 1977																	
Endosulfan (OC)	- Savanna woodland, Zimbabwe <u>Chiromantis xerampelina</u>	Ultra-low volume aerial applications from a fixed-wing aircraft at 14 g a.i./ha.	No deaths 19 days post-treatment.	N = 14 treatment, 8 control; animals kept in cages in pond.	Cockbill, 1979																	

Table 3 - Field Studies (cont'd)

Contaminant*	Location/Habitat Species	Rate and Method of Application	Reported Effects on Amphibians	Water Temperature, Residue Levels, Miscellaneous	Reference	
Endrin (OC)	- 0.025 ha pond in California <u>Rana catesbeiana</u>	Sprays prepared from emulsified concentrate formulations at a rate of 89.9 L/ha and applied from 3 gallon (12.45 L) sprayers.	Application Rate (Active ingredient) (kg/ha)  0.11 0.36	% 24 hours or cumulative mortality days after treatment  1 2 5 6 7 50 90 90* 0 0 100* 90 100* 80 20	Water depth 20.32 - 30.48 cm. pH 7.5 - 8.0.  Tests conducted from June to October 1962. Maximum daily water temp. 26.6°-35°C. One pond per treatment; N = 20 caged tadpoles (age unspecified) per pond.	Mulla, 1963
	- Field tests in California <u>Rana catesbeiana</u>  - 2 year old artificially excavated pool (10 x 10 m) with clusters of filamentous algae; no inlet <u>Rana temporaria</u> <u>R. esculenta</u>	.11 - .56 kg/ha.  0.5 L of 20% Endrin poured into pool. 0.047 mg/L measured 10 m from point of contamination after 6 hours.	Applied as aqueous spray at rate of 89.9 L/ha. Active ingredient = 0.22 and 0.90 kg/ha.	No mortality after 24 hours.	° 10 fresh tadpoles added.  Complete kill of tadpoles at these application rates.  Adult <u>R. esculenta</u> (wild population) tossed about because of strong cramps and paralysis of limbs and died. Caged <u>R. temporaria</u> tadpoles (without developed limbs) alive only at most distant test stations after 72 hours. 7 weeks after treatment, <u>R. esculenta</u> repopulated pond.	Average water depth 0.44 m. Temperature 26°C, sky 2/3 cloud cover, sunny breaks.
Ethyl Guthion (OP)	- 0.025 ha pond Location unknown <u>Rana catesbeiana</u>	Applied as aqueous spray at rate of 89.9 L/ha. Active ingredient = 0.22 and 0.90 kg/ha.	No mortality after 24 hours.	Water depth 20.32 - 30.48 cm. 10 tadpoles caged in pond.	Mulla et al., 1963	

Table 3 - Field Studies (cont'd)

Contaminant*	Location/Habitat Species	Rate and Method of Application	Reported Effects on Amphibians	Water Temperature, Residue Levels, Miscellaneous	Reference	
Fenitrothion (OP)	- Fundy National Park, New Brunswick <u>Rana clamitans</u> <u>R. sylvatica</u> assorted other species	Aerially applied at 0.14 kg/ha in 2 sprayings 9-17 days apart.	No mortality observed in caged <u>R. sylvatica</u> tadpoles, or in caged or wild adults or tadpoles of other species.		Rick and Price, 1974	
	- Pond in New Brunswick <u>Rana clamitans</u> <u>Bufo americanus</u>	Aerially sprayed with .14 kg/ha on June 1 and 9.	No mortality observed.		Pearce and Ingle, 1969	
	- Indian rice fields Frog species unspecified	Sprayed by helicopter from height of 2-3 m over fields at 0.5 kg/ha between 7:30 and 9:00 a.m.	100% mortality.		Population estimates made 48 hours after spraying.	Thirumurthi et al., 1973
Fenthion (OP)	- Indian rice fields Frog species unspecified	Sprayed by helicopter from height of 2-3 m over fields at 0.5 kg/ha between 7:30 and 9:00 a.m.	100% mortality.		Population estimates made 48 hours after spraying.	Thirumurthi et al., 1973
FLIT MLD (Larvicide)	- Butte County, California 2 test ponds - 3.4 x 38.5 m with 4.5 m between them <u>Rana catesbeiana</u>	2.24 L/ha applied using a hand sprayer on June 15, August 3, 24, and September 8, 1971.	No dead frogs were observed after August 24 and September 8 treatments.	Water temperature 20.5-25.5°C. Water depth at centre 0.6 m. <u>R. catesbeiana</u> migrated into the ponds prior to the 3rd treatment.	Hagen et al., 1973	
Fuel Oil N°2 (Larvicide)	- Butte County, California 2 test ponds - 3.4 x 38.5 m with 4.5 m between them <u>Rana catesbeiana</u>	112.1 L/ha applied using a hand sprayer on June 15, August 3, 24, and September 8, 1971.	No dead frogs were observed after August 24 and September 8 treatments.	Water temperature 20.5-25.5°C. Water depth at centre 0.6 m. <u>R. catesbeiana</u> migrated into the ponds prior to the 3rd treatment.	Hagen et al., 1973	



le 3 - Field Studies (cont'd)

Contaminant <sup>a</sup>	Location/Habitat Species	Rate and Method of Application	Reported Effects on Amphibians	Water Temperature, Residue Levels, Miscellaneous	Reference
7565 (OP)	- 0.025 ha pond Location unknown <u>Rana catesbeiana</u>	Applied as aqueous spray at rate of 89.9 L/ha. Active ingredient = 0.22 and 0.90 kg/ha.	No mortality after 24 hours.	Water depth 20.32 - 30.48 cm. 10 tadpoles caged in pond.	Mulla et al., 1963
8029 (OP)	- 0.025 ha pond Location unknown <u>Rana catesbeiana</u>	Applied as aqueous spray at rate of 89.9 L/ha. Active ingredient = 0.45 and 1.79 kg/ha.	No mortality after 24 hours.	Water depth 20.32 - 30.48 cm. 20 tadpoles caged in pond.	Mulla et al., 1963
10493 (OP)	- 0.025 ha pond location unknown <u>Bufo boreas</u> <u>Scaphiopus hammondi</u>	Applied as aqueous spray at rate of 89.9 L/ha. Active ingredient = 0.22 and 0.90 kg/ha.	No mortality after 24 hours.	Water depth 20.32 - 30.48 cm. 20 tadpoles caged in pond.	Mulla et al., 1963
	- Field tests in California <u>Bufo boreas</u> <u>Scaphiopus hammondi</u>	0.22 kg/ha. 0.90 kg/ha.	Deemed 'safe' to tadpoles at these application rates.		Mulla, 1962
10494 (OP)	- 0.025 ha pond Location unknown <u>Bufo boreas</u> <u>Scaphiopus hammondi</u>	Applied as aqueous spray at rate of 89.9 L/ha. Active ingredient = 0.11 and 0.45 kg/ha.	Application Rate    % 24 hour mortality .11                    0 .45                    5	Water depth 20.32 - 30.48 cm. 20 tadpoles caged in pond.	Mulla et al., 1963
	- Field tests in California <u>Bufo boreas</u> <u>Scaphiopus hammondi</u>	.11 kg/ha. .45 kg/ha.	Deemed 'safe' to tadpoles at these application rates.		Mulla, 1962
13582 (OP)	- 0.025 ha pond Location unknown <u>Rana catesbeiana</u>	Applied as aqueous spray at rate of 89.9 L/ha. Active ingredient = 0.45 and 1.79 kg/ha.	Application Rate    % 24 hour mortality .45                    0 1.79                  100	Water depth 20.32 - 30.48 cm. 20 tadpoles caged in pond.	Mulla et al., 1963

le 3 - Field Studies (cont'd)

Contaminant <sup>a</sup>	Location/Habitat Species	Rate and Method of Application	Reported Effects on Amphibians	Water Temperature, Residue Levels, Miscellaneous	Reference
13582 (cont'd)	- Field tests in California <u>Rana catesbeiana</u>	.45 kg/ha. 1.8 kg/ha.	No mortality in tadpoles. 100% mortality in tadpoles.		Mulla, 1962
ptachlor (C)	- 0.025 ha pond in California <u>Rana catesbeiana</u>	Sprays prepared from emulsifiable concentrate formulations and applied at a rate of 89.9 L/ha from 3 gallon (12.45 L) sprayers.	Application Rate (Active ingredient) (kg/ha)    % 24 hour or cumulative mortality days after treatment  0.11                    1   2   5   6   7 0.56                    0   0   0   0   -	Water depth 20.32 - 30.48 cm. pH 7.5 - 8.0.  tests conducted from June to October 1962. Maximum daily water temp. 26.6°-35°C. One pond per treatment; N = 20 caged tadpoles (age unspecified) per pond.	Mulla, 1963
	- Field tests in California <u>Rana catesbeiana</u>	.11 kg/ha. .56 kg/ha.	Deemed 'toxic' to tadpoles at these application rates.		Mulla, 1962
ptachlor (OC)	- 0.025 ha pond in California <u>Rana catesbeiana</u>	Spray prepared from emulsifiable concentrate formulations at a rate of 89.9 L/ha and applied from 3 gallon (12.45 L) sprayers.	Application Rate (Active ingredient) (kg/ha)    % 24 hour or cumulative mortality days after treatment  0.11                    1   2   5   6   7 0.56                    0   0   0   -   -	Water depth 20.32 - 30.48 cm. pH 7.5 - 8.0.  Tests conducted from June to October 1962. Maximum daily water temp. 26.6°-35°C. One pond per treatment; N = 20 caged tadpoles (age unspecified) per pond.	Mulla, 1963

Table 3 - Field Studies (cont'd)

Contaminant <sup>a</sup>	Location/Habitat Species	Rate and Method of Application	Reported Effects on Amphibians	Water Temperature, Residue Levels, Miscellaneous	Reference	
Kepona (cont'd)	- Field studies in California <u>Rana catesbeiana</u>	.11 kg/ha. .56 kg/ha.	Deemed 'safe' to tadpoles at these two application rates.		Mulla, 1962	
Leptophos (OP)	- Indian rice fields Frog species unspecified	Sprayed by helicopter from height of 2-3 m over fields at 1.0 kg/ha between 7:30 to 9:00 a.m.	100% mortality.	Population estimates made 48 hours after spraying.	Shirumurthi et al., 1973	
Lindane (OC)	- 0.025 ha pond in California <u>Rana catesbeiana</u>	Spray prepared from emulsifiable concentrate formulations and applied at a rate of 89.9 L/ha from 3 gallon (12.45 L) sprayers.	Application Rate (Active ingredient) (kg/ha)	% 24 hour or cumulative mortality days after treatment	Water depth 20.32 - 30.48 cm. pH 7.5 - 8.0. Tests conducted from June to October 1962. Maximum daily water temp. 26.6°-35°C. One pond per treatment; N = 20 caged tadpoles (age unspecified) per pond.	Mulla, 1963
				1 2 5 6 7		
			0.11 0.56	0 0 0 30 - 10 10 10 10 -		
	- Field tests in California <u>Rana catesbeiana</u>	.11 kg/ha. .56 kg/ha.	Deemed 'safe' to tadpoles at these application rates.		Mulla, 1962	
Malathion (OP)	- Oak - poplar forest, North Carolina <u>Plethodon glutinosus</u>	5.6 kg/ha applied with back pack sprayer during 10 weekly applications (27 May to 23 August).	No brain ChE inhibition, changes in abundance, effects on lipid storage patterns noted in adults or juveniles.	ChE inhibition noted at this concentration in lab study.	Baker, 1985	

Table 3 - Field Studies (cont'd)

Contaminant <sup>a</sup>	Location/Habitat Species	Rate and Method of Application	Reported Effects on Amphibians	Water Temperature, Residue Levels, Miscellaneous	Reference
Metals	- Ironwork Katowice region, Poland <u>Rana ridibunda</u>	Emissions from metallurgical work.	Frogs collected from an area close to the factory had lower metabolic rates than those from farther away.		Pytasz et al., 1980
Methy Parathion (OP)	- 0.025 ha pond Location unknown <u>Bufo boreas</u> <u>Scaphiopus hammondi</u>	Applied as aqueous spray at rate of 89.9 L/ha. Active ingredient = 0.11 and 0.45 kg/ha.	No mortality after 24 hours.	Water depth 20.32-30.48 cm. 20 tadpoles caged in pond.	Mulla et al., 1963
	- Field tests in California <u>Bufo boreas</u> <u>Scaphiopus hammondi</u>	.11 kg/ha. .45 kg/ha.	Deemed 'safe' to tadpoles at these application rates.		Mulla, 1962
Hexacarbate (C) (Zectran)	- Acadian Forest Experiment Station, New Brunswick <u>Rana clamitans</u> , <u>R. sylvatica</u> Assorted other species	Aerially applied at 0.007 kg/ha.	Mortality in <u>R. sylvatica</u> tadpoles; no other toxic effects noted.		Rick and Price, 1974
Naled (OP)	- 0.025 ha pond Location unknown <u>Rana catesbeiana</u>	Applied as aqueous spray at rate of 89.9 L/ha. Active ingredient = 0.56 kg/ha.	No mortality after 24 hours.	Water depth 20.32 - 30.48 cm. 10 tadpoles caged in pond.	Mulla et al., 1963

Table 3 - Field Studies (cont'd)

Contaminant <sup>a</sup>	Location/Habitat Species	Rate and Method of Application	Reported Effects on Amphibians	Water Temperature, Residue Levels, Miscellaneous	Reference
Oxamyl (C)	- Ditches beside fields in Great Britain Cage 1 - between barley and potato fields Cage 3 - between barley and sugar beet fields Cage 7 - between sugar beet and potato fields Cage 17 - control pond <u>Rana temporaria</u>	Unspecified.	Cage 1: Development and growth slow, with many deformities, mortality. Cage 3: Development rapid, growth satisfactory, survival good. Late in experiment tadpoles developed 'lateral kinks'. Cage 7: High mortality during first month, growth and development normal. Deformities after 4 weeks. Cage 17: Survival good, development and growth average. 'Lateral' kinks noted late in experiment.	Caged tadpoles maintained near potato fields showed more deformities. No other fields were treated with oxamyl.	Cooke, 1981
Parathion (OP)	- 0.025 ha pond Location unknown <u>Bufo boreas</u> <u>Scaphiopus hammondi</u>  - 2 wetland habitats in farmed areas of California: Habitat 1 - duck pond (4.05 ha) with watergrass, smart weed, rushes. Existing vegetation burned in July before pond flooded. Habitat 2 - borrow pit (10.1 ha) filled with underground seepage water. Vegetation included cattails, watergrass, hornwort. <u>Rana catesbeiana</u>	Applied as aqueous spray at rate of 89.9 L/ha. Active ingredient = 0.11 and 0.45 kg/ha.  Applied by aircraft: Habitat 1 - sprayed once at a rate of 1.12 kg a.i./ha from height of 7.62 m.  Habitat 2 - sprayed 6 times at weekly intervals at a rate of .11 kg/ha from height of 12.2 m.	No mortality after 24 hours.  Habitat 1 - abundance or survival not seriously affected. Habitat 2 - no apparent effect on populations.	Water depth 20.32 - 30.48 cm. 20 tadpoles caged in pond.  Habitat 1 - 25.4 cm in depth. Habitat 2 - 1.83 m in depth. Average amount of parathion which reached water surface: Habitat 1 - .95 kg/ha; Habitat 2 - 0.30-0.08 kg/ha.	Mulla et al., 1963  Mulla et al., 1966

Table 3 - Field Studies (cont'd)

Contaminant <sup>a</sup>	Location/Habitat Species	Rate and Method of Application	Reported Effects on Amphibians	Water Temperature, Residue Levels, Miscellaneous	Reference
Parathion (cont'd)	- Field tests in California <u>Bufo boreas</u> <u>Scaphiopus hammondi</u>	.11 kg/ha. .45 kg/ha.	Deemed 'safe' to tadpoles at these application rates.		Mulla, 1962
Pesticides (miscellaneous)	- Rice field discharge canal and relatively clean pond in the North Caucasus, U.S.S.R. <u>Rana ridibunda</u>  - Meter sources polluted with pesticides <u>Bufo melanostictus</u>	Wide range of pesticides used on fields.	Haemoglobin content increased, leucocytosis observed and number of stab-nuclear neutrophils and monocytes in the leucocytic formula of the blood increased in frogs from rice field.  Higher sister chromatid exchange frequency noted in field specimens than controls exposed identically to BrdU solution.	8-9 times greater organochlorine levels in muscles of frogs from rice fields.  Attributed to high concentration of mutagens/cletogens in tissues of field specimens.	Zhukova, 1987  Dhakrabarti et al., 1984
pH	-Natural breeding ponds <u>Ambystoma maculatum</u>  -Sandy coastal dunes and inland heaths in Great Britain <u>Bufo calamita</u>		Hatching success at pH 4.6 - 0.0%. Hatching success at pH 6.9 - 77.1%.  <u>B. calamita</u> avoided acid heathland ponds (pH > 5.0).	Eggs interchanged between acidic and neutral ponds had intermediate hatching success. Tolerance to acid is built up upon exposure.  Fluctuating temperature, light; no rainfall. Spawn taken and reared in lab; no tadpoles survived to metamorphosis below pH 4.75 and success was minimal below 6.0.	Nielsen et al., 1977  Beebe and Griffin, 1977

Table 3 - Field Studies (cont'd)

Contaminant <sup>a</sup>	Location/Habitat Species	Rate and Method of Application	Reported Effects on Amphibians					Water Temperature, Residue Levels, Miscellaneous	Reference
			pH	4-5	5-6	6-7	7-8		
pH (cont'd)	-Southeast England agricultural land and uncultivated heathland <u><i>Triturus vulgaris</i></u> , <u><i>T. helveticus</i></u> , <u><i>T. cristatus</i></u> , <u><i>Rana temporaria</i></u> , <u><i>Bufo bufo</i></u> , <u><i>B. calamita</i></u>		<u><i>T. vulgaris</i></u>	0*	0	5	1	Survey of amphibian breeding sites.  * No. of ponds in each category containing the various species.	Beebee, 1983
		<u><i>T. helveticus</i></u>	2	2	7	1			
		<u><i>T. cristatus</i></u>	0	0	2	0			
		<u><i>R. temporaria</i></u>	0	1	1	4			
			<u><i>B. bufo</i></u>	0	0	4	1		
			<u><i>B. calamita</i></u>	0	1	1	0		
			TOTAL PONDS	13	4	12	7		
	-Stream draining a construction site in Great Smoky Mountains National Park, North Carolina <u><i>Leurognathus macropus</i></u>		None of the caged salamanders placed upstream ( $\bar{x}$ pH = 7.0) died; most of those placed downstream ( $\bar{x}$ pH = 4.6) died.					Water temperature downstream - 8-10°C.	Huckabee et al., 1975
	-Ephemeral meltwater ponds in central Ontario <u><i>Ambystoma maculatum</i></u>		Percent hatching success was correlated with pH - 88% at pH 6.15, 80% at pH 4.51. Survival of larvae in pond of pH 4.97 was 46%; in enclosures where CaCO <sub>3</sub> added to elevate pH to 5.30, survival was 64%.					Results of transfer experiments suggest that larvae from population breeding in acidic ponds were more tolerant of increased acidity than those from less acidic ponds.	Clark, 1986b
	-Temporary woodland ponds in coastal Maryland <u><i>Ambystoma maculatum</i></u>		Egg mass abundance and survival of embryos not correlated with pond pH. Survival of eggs transferred among ponds of pH 3.66 - 5.18 reduced only at pH 3.66.					Pond longevity, water temperature and perhaps oxygen content seem more important to salamander reproduction than acid precipitation.	Albers and Prouty, 1987

Table 3 - Field Studies (cont'd)

Contaminant <sup>a</sup>	Location/Habitat Species	Rate and Method of Application	Reported Effects on Amphibians					Water Temperature, Residue Levels, Miscellaneous	Reference
			pH	4-5	5-6	6-7	7-8		
pH (cont'd)	-Bog pools at a heathland site in south-east England <u><i>Triturus helveticus</i></u>  <u><i>Bufo calamita</i></u>		Bred in all pools; were rarest in rain-fed ones.					Eutrophicated ponds had pH 5.0-6.5; rain-fed ponds had pH 3.8-4.3.	Beebee, 1987
			Bred only in eutrophicated ponds, as did 4 other unspecified amphibian species.						
	-Ephemeral meltwater ponds in central Ontario <u><i>Bufo americanus</i></u> , <u><i>Rana catesbeiana</i></u> , <u><i>Hyla crucifer</i></u> , <u><i>R. sylvatica</i></u> , <u><i>R. clamitans</i></u>		<u><i>B. americanus</i></u> , <u><i>H. crucifer</i></u> and <u><i>R. catesbeiana</i></u> did not occur in ponds with pH > 5.0. <u><i>R. clamitans</i></u> and <u><i>R. sylvatica</i></u> occurred in all ponds. <u><i>R. sylvatica</i></u> densities increased in more acidic ponds; egg mass density was reduced as pH decreased.					Surveys done in ponds with pH 4.04 - 6.63. Results suggest chronic sublethal effects may be affecting populations at pH's greater than those causing acute toxicity.	Clark, 1986a
			<u><i>T. vulgaris</i></u> rarely encountered in water with pH > 6. <u><i>T. helveticus</i></u> rarely encountered in water with pH > 4.					Breeding site characteristic studied.	Cooke and Frazer, 1976
-Forest and fen sites in Britain <u><i>Triturus helveticus</i></u> <u><i>T. vulgaris</i></u>		<u><i>A. jeffersonianus</i></u> and <u><i>B. woodhousi</i></u> not observed in ponds with mean pH > 4.62 and 4.25, respectively; presence of <u><i>R. sylvatica</i></u> and <u><i>H. andersoni</i></u> unrelated to pond pH. In embryo transfer experiments, mortality of <u><i>A. jeffersonianus</i></u> embryos increased as pH decreased; hatching of <u><i>R. sylvatica</i></u> was not related to pH.						Freds and Dunson, 1986	
-Temporary forest ponds in central Pennsylvania and sphagnum bogs, seeps and ponds in New Jersey <u><i>Ambystoma jeffersonianus</i></u> <u><i>Rana sylvatica</i></u> <u><i>Bufo woodhousi</i></u> <u><i>Hyla andersoni</i></u>									

Table 3 - Field Studies (cont'd)

Contaminant <sup>a</sup>	Location/Habitat Species	Rate and Method of Application	Reported Effects on Amphibians	Water Temperature, Residue Levels, Miscellaneous	Reference																											
pH (cont'd)	-Poorly buffered water bodies in the Netherlands		Relation between water pH and % systems with amphibians:	In acidic waters, egg masses of <i>R. arvalis</i> , <i>R. esculenta</i> , <i>R. temporaria</i> , <i>B. bufo</i> , <i>B. calamita</i> became heavily infested with fungi.	Leuven et al., 198																											
			<table border="1"> <thead> <tr> <th>pH 5 (n=37)</th> <th>4 pH 5 (n=31)</th> <th>pH 4 (n=28)</th> </tr> </thead> <tbody> <tr> <td>84.4</td> <td>57.5</td> <td>11.8</td> </tr> <tr> <td>15.3</td> <td>3.0</td> <td>0.0</td> </tr> <tr> <td>21.9</td> <td>62.5</td> <td>35.3</td> </tr> <tr> <td>76.6</td> <td>76.9</td> <td>63.3</td> </tr> <tr> <td>68.8</td> <td>55.0</td> <td>17.7</td> </tr> <tr> <td>9.1</td> <td>9.7</td> <td>0.0</td> </tr> <tr> <td>8.1</td> <td>6.5</td> <td>0.0</td> </tr> <tr> <td>21.6</td> <td>32.3</td> <td>10.7</td> </tr> <tr> <td>18.9</td> <td>6.5</td> <td>0.0</td> </tr> </tbody> </table>			pH 5 (n=37)	4 pH 5 (n=31)	pH 4 (n=28)	84.4	57.5	11.8	15.3	3.0	0.0	21.9	62.5	35.3	76.6	76.9	63.3	68.8	55.0	17.7	9.1	9.7	0.0	8.1	6.5	0.0	21.6	32.3	10.7
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	<p><i>Bufo bufo</i>  <i>B. calamita</i>  <i>Rana arvalis</i>  <i>R. esculenta</i>  <i>R. temporaria</i>  <i>Triturus alpestris</i>  <i>T. cristatus</i>  <i>T. helveticus</i>  <i>T. vulgaris</i></p>	Snowmelt water.	At many spawning sites of <i>R. arvalis</i> , <i>R. esculenta</i> , <i>R. temporaria</i> and <i>B. bufo</i> no tadpoles found in pH > 5.0.	Egg mass density also correlated with total organic carbon.	Gascon and Pianas,																											
	- Laurentians, Québec <i>Rana sylvatica</i>		Egg mass density was negatively correlated with acidity. Hatching success was inversely correlated with pH. Occurrence of mould on eggs increased in low pH.																													

Table 3 - Field Studies (cont'd)

Contaminant <sup>a</sup>	Location/Habitat Species	Rate and Method of Application	Reported Effects on Amphibians	Water Temperature, Residue Levels, Miscellaneous	Reference																		
pH (cont'd)	-Hardwood-hemlock forests of Delaware Co., New York <i>Plethodon cinereus</i>		8.8% of quadrats with soil pH > 3.7 contained a salamander; 50.8% of quadrats with pH < 3.8 contained a salamander. Juveniles never found on soil with pH > 3.7. Seasonal density correlated with % quadrats with pH > 3.7.	Soil temperature and moisture did not affect density or distribution.	Wyman and Hawksley-Lescault, 1987																		
			Tadpoles from pond with pH 4.5-4.90 had lower body sodium, chloride and water concentrations than those from nearby pond with pH 5.74 - 6.37. Tadpoles from either pond placed in low pH had higher sodium efflux than when placed in high pH.																				
pH (acid precipitation)	-Temporary forest ponds in central Pennsylvania <i>Rana sylvatica</i>			Altitude 460 - 625 m. pH 4.5-7.0 at time eggs laid; by hatching pH had increased by 0.25 to 0.5 units in each pond.	Freda and Dunson, 1985c																		
	-5 ponds in Tompkins Co., New York <i>Ambystoma maculatum</i>	Acid precipitation.	<table border="1"> <thead> <tr> <th>Pond</th> <th>pH</th> <th>Maximum mortality %</th> </tr> </thead> <tbody> <tr> <td>A</td> <td>7</td> <td>0.66</td> </tr> <tr> <td>B</td> <td>6</td> <td>0.91</td> </tr> <tr> <td>C</td> <td>5.5</td> <td>43.7</td> </tr> <tr> <td>D</td> <td>5.0</td> <td>65</td> </tr> <tr> <td>E</td> <td>4.5</td> <td>65</td> </tr> </tbody> </table>	Pond	pH	Maximum mortality %	A	7	0.66	B	6	0.91	C	5.5	43.7	D	5.0	65	E	4.5	65		Pough, 1976
Pond	pH	Maximum mortality %																					
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	-Lake with sphagnum bottom, Sweden <i>Rana temporaria</i>	Lake acidified by acid precipitation.	Mortality low during early embryonic development. Abnormalities observed at low pH.  No embryos survived to become adults.	Water temperature rose from 8-23°C during development.  pH 4.0-4.5.	Hagstrom, 1977																		

Table 3 - Field Studies (cont'd)

Contaminant <sup>a</sup>	Location/Habitat Species	Rate and Method of Application	Reported Effects on Amphibians	Water Temperature, Residue Levels, Miscellaneous	Reference												
pH (acid precipitation) (cont'd)	- Lake Tronovatten, Sweden <u>Triturus vulgaris</u> <u>Bufo bufo</u> , <u>Rana temporaria</u>	acid precipitation.	<u>T. vulgaris</u> increased in acidified lakes; <u>R. temporaria</u> and <u>B. bufo</u> decreased.	Suggests that newts increased as a result of extinction of predatory fish populations.	Hagstrom, 1981												
	- Ponds in Ithaca, NY <u>Ambystoma maculatum</u>	pH range of 4.5-7.0 in ponds.	<table border="1"> <thead> <tr> <th>pH</th> <th>Mortality of embryos</th> </tr> </thead> <tbody> <tr> <td>7</td> <td>0.66%</td> </tr> <tr> <td>6</td> <td>0.91%</td> </tr> <tr> <td>5.5</td> <td>45.7%</td> </tr> <tr> <td>5.0</td> <td>65%</td> </tr> <tr> <td>4.5</td> <td>65%</td> </tr> </tbody> </table>	pH	Mortality of embryos	7	0.66%	6	0.91%	5.5	45.7%	5.0	65%	4.5	65%	Eggs from ponds with pH 4.5, 5.0 failed to retract yolk plug; eggs in pH 5.5 developed chest swellings and stunted gills after gastrulation.	Some and Pough, 1982
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- Connecticut Valley, Massachusetts <u>Ambystoma maculatum</u> <u>A. jeffersonianum</u>	Rainfall: pH = 4.16 in 1976. pH = 4.07 in 1977.	Reciprocal egg transplants indicated no difference in acid tolerance dependent on pond of origin.  No significant correlation between pond pH and % embryonic mortality. <u>A. maculatum</u> 2.1-30.2% mortality. <u>A. jeffersonianum</u> 12.4-40.3% mortality.	Some of this data appeared in Pough 1976.  pH of 6 ponds: 1976 $\bar{x}$ = 5.62. 1977 $\bar{x}$ = 5.10.	Cook, 1983													

Table 3 - Field Studies (cont'd)

Contaminant <sup>a</sup>	Location/Habitat Species	Rate and Method of Application	Reported Effects on Amphibians	Water Temperature, Residue Levels, Miscellaneous	Reference																												
pH (bogwater)	- Bear Meadows Bog, Pennsylvania sphagnum bog  <u>Rana catesbeiana</u>	Embryos were transferred to a naturally acidic stream draining the bog.	Mortality of transferred embryos:	Maintained in lab at 25°C before exposure. Lab tests using bog water support findings in the field. Observed lethality at pH's of 6.0, thus possibly some other toxic agent is responsible.	Saber and Danson, 1977																												
			<table border="1"> <thead> <tr> <th>pH range</th> <th>Temp. range (°C)</th> <th>N animals (Stage)</th> <th>% mortality</th> </tr> </thead> <tbody> <tr> <td>5.80-3.82</td> <td>14.5-19</td> <td>20</td> <td>100</td> </tr> <tr> <td>3.80-3.82</td> <td>14.5-19</td> <td>20</td> <td>100</td> </tr> <tr> <td>4.00-4.01</td> <td>18</td> <td>30</td> <td>100</td> </tr> <tr> <td></td> <td>16-19</td> <td>30(26)</td> <td>100</td> </tr> <tr> <td>3.70-3.99</td> <td>14-19</td> <td>6(32-38)</td> <td>16.7</td> </tr> <tr> <td>3.70-3.99</td> <td>14-19</td> <td>5(26-30)</td> <td>0</td> </tr> <tr> <td>3.60-4.0</td> <td>14-19</td> <td>6(26-30)</td> <td>83.3</td> </tr> </tbody> </table>			pH range	Temp. range (°C)	N animals (Stage)	% mortality	5.80-3.82	14.5-19	20	100	3.80-3.82	14.5-19	20	100	4.00-4.01	18	30	100		16-19	30(26)	100	3.70-3.99	14-19	6(32-38)	16.7	3.70-3.99	14-19	5(26-30)	0
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Phosphamidon (OP)	- Indian rice fields Frog species unspecified	Sprayed by helicopter from height of 2-3 m over fields at 0.25 kg/ha between 7:30 and 9:00 a.m.	75 % mortality.	Population estimates made 48 hrs. after spraying.	Thirumurthi et al., 1975																												
Pollution (unspecified)	- Minsk and Berezinsky Reserve, U.S.S.R. <u>Rana temporaria</u>	"anthropogenic" pollution.	No differences in chromosome aberration frequencies in frogs in two regions.	Regions subjected to different degrees of anthropogenic impact.	Elisyeyeva et al., 1975																												
	- South Sakhain, U.S.S.R. <u>Rana chensinensis</u>	Site A - polluted with sewage effluent of paper factory. Site B & C - contaminated by municipal gutters.	<table border="1"> <thead> <tr> <th>Site</th> <th>No. with Limb Abnormalities</th> <th>No. with Chondrodysplasia Lesions</th> </tr> </thead> <tbody> <tr> <td>A</td> <td>460 (42%)</td> <td>126 (11.5%)</td> </tr> <tr> <td>B</td> <td>1424 (39%)</td> <td>202 ( 5.5%)</td> </tr> <tr> <td>C</td> <td>500 (31%)</td> <td>0</td> </tr> </tbody> </table>	Site	No. with Limb Abnormalities	No. with Chondrodysplasia Lesions	A	460 (42%)	126 (11.5%)	B	1424 (39%)	202 ( 5.5%)	C	500 (31%)	0	Developmental anomalies and dysplasia also found in <u>R. aureamais</u> , <u>Bufo bufo gargarizans</u>	Mizgirev et al., 1984																
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Table 3 - Field Studies (cont'd)

Contaminant <sup>a</sup>	Location/Habitat Species	Rate and Method of Application	Reported Effects on Amphibians	Water Temperature, Residue Levels, Miscellaneous	Reference					
Pollution (Industrial wastewater products)	- Shanghai, China <u>Bufo bufo garqarizans</u>	Waste water treatment plant and factory oxidation pond.	Sister chromatid exchange frequencies of samples were markedly higher than frequencies of controls.		Wen et al., 1984					
	- <u>Rana temporaria</u>		Level of genetic damage in frogs from polluted regions was higher than in those from protected areas.		Kraskowski et al., 1986					
Pollution (sewage)	- Plays Lake, Texas <u>Ambystoma tigrinum</u>	Polluted with sewage and asphalt wastes.	Hepatic microsomal aryl hydrocarbon hydroxylase levels were elevated. Bladder contents were mutagenic using Ames test.		Buebee et al., 1978					
	- 0.025 ha pond Location unknown <u>Bufo boreas</u> <u>Scaphiopus hammondi</u>	Applied as aqueous spray at rate of 89.9 L/ha. Active ingredient = 0.22 and 0.90 kg/ha.	<table border="1"> <thead> <tr> <th>Application Rate</th> <th>% 24 hr mortality</th> </tr> </thead> <tbody> <tr> <td>.22</td> <td>10</td> </tr> <tr> <td>.90</td> <td>0</td> </tr> </tbody> </table>		Application Rate	% 24 hr mortality	.22	10	.90	0
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.90	0									
	- Field tests in California <u>Bufo boreas</u> <u>Scaphiopus hammondi</u>	.22 kg/ha. .90 kg/ha.	Deemed 'safe' to tadpoles at these application rates.		Mulla, 1962					
Rotenone	- Shallow .05 ha ponds in Georgia <u>Rana pipiens</u>	2 and 5 uL/L of Pro-Noxfish (0.05 and 0.125 uL/L rotenone) applied on Aug. 24 into propeller wash of boat.	Partial mortality of resident population of larval frogs in 5 uL/L treatment.	Water temp. 31-32°C, pH 8.6-9.6 at time of treatment.	Burress, 1982					

Table 3 - Field Studies (cont'd)

Contaminant <sup>a</sup>	Location/Habitat Species	Rate and Method of Application	Reported Effects on Amphibians	Water Temperature, Residue Levels, Miscellaneous	Reference																							
Dieldrin I (DC)	- 0.025 ha pond in California <u>Rana catesbeiana</u>	Sprays prepared from emulsifiable concentrate formulations and applied at a rate of 89.9 L/ha from 3 gallon (12.45 L) sprayers.	<table border="1"> <thead> <tr> <th rowspan="2">Application Rate (Active ingredient) (kg/ha)</th> <th colspan="5">% 24 hour or cumulative mortality days after treatment</th> </tr> <tr> <th>1</th> <th>2</th> <th>5</th> <th>6</th> <th>7</th> </tr> </thead> <tbody> <tr> <td>0.11</td> <td>60*</td> <td>20</td> <td>0</td> <td>-</td> <td>-</td> </tr> <tr> <td>0.56</td> <td>100*</td> <td>80</td> <td>0</td> <td>0</td> <td>-</td> </tr> </tbody> </table>	Application Rate (Active ingredient) (kg/ha)	% 24 hour or cumulative mortality days after treatment					1	2	5	6	7	0.11	60*	20	0	-	-	0.56	100*	80	0	0	-	Water depth 20.32 - 30.48 cm. pH 7.5 - 8.0. Tests conducted from June to October 1962. Maximum daily water temp. 26.6°-35°C. One pond per treatment; N = 20 caged tadpoles (age unspecified) per pond.	Mulla, 1963
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- Pond <u>Rana catesbeiana</u>	.11 kg/ha applied by hand sprayer.	Complete kill of tadpoles.			Mulla, 1962																							
- Residential area <u>Bufo boreas</u> <u>Scaphiopus hammondi</u>	1.12 kg/ha.	98% control of juvenile toads in 24 hrs.			Mulla, 1962																							
Dieldrin II (DC)	- 0.025 ha pond in California <u>Rana catesbeiana</u>	Spray prepared from emulsifiable concentrate formulations and applied at a rate of 89.9 kg/ha from 3 gallon (12.45 L) sprayers.	<table border="1"> <thead> <tr> <th rowspan="2">Application Rate (Active ingredient) (kg/ha)</th> <th colspan="5">% 24 hour or cumulative mortality days after treatment</th> </tr> <tr> <th>1</th> <th>2</th> <th>5</th> <th>6</th> <th>7</th> </tr> </thead> <tbody> <tr> <td>0.11</td> <td>10</td> <td>10</td> <td>10</td> <td>-</td> <td>-</td> </tr> <tr> <td>0.56</td> <td>100*</td> <td>30</td> <td>30</td> <td>-</td> <td>-</td> </tr> </tbody> </table>	Application Rate (Active ingredient) (kg/ha)	% 24 hour or cumulative mortality days after treatment					1	2	5	6	7	0.11	10	10	10	-	-	0.56	100*	30	30	-	-	Water depth 20.32 - 30.48 cm. pH 7.5 - 8.0. Tests conducted from June to October 1962. Maximum daily water temp. 26.6°-35°C. One pond per treatment; N = 20 caged tadpoles (age unspecified) per pond.	Mulla, 1963
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Table 3 - Field Studies (cont'd)

Contaminant <sup>a</sup>	Location/Habitat Species	Rate and Method of Application	Reported Effects on Amphibians						Water Temperature, Residue Levels, Miscellaneous	Reference
			Application Rate (Active Ingre- dient) (kg/ha)	% 24 hour or cumulative mortality days after treatment						
				1	2	5	6	7		
Dioxophene (OC)	- 0.025 ha pond in California <u>Rana catesbeiana</u>	Spray prepared from emulsifiable concentrate formulations and applied at a rate of 89.9 kg/ha from 3 gallon (12.45 L) sprayers.	0.11 0.36	0 100*	0 100	0 0	- 0	- 0	Water depth 20.32 - 30.48 cm. pH 7.5 - 8.0.  Tests conducted from June to October 1962. Maximum daily water temp. 26.6°-35°C. One pond per treatment, N = 20 caged tadpoles (age unspecified) per pond.	Mullis, 1963
	- Golf course greens <u>Bufo boreas</u> <u>Scaphiopus hammondi</u>	1.12 kg/ha. 2.24 kg/ha.						In mixture with DDT.	Mullis, 1962	
	- Field tests in California <u>Rana catesbeiana</u>	.11 kg/ha. .36 kg/ha.						100% kill of tadpoles.	Mullis, 1962	

<sup>a</sup> OC-organochlorine; OP-organophosphate; C-carbamate; H-herbicide; A-accricide.



TABLE 4  
RESIDUES

CONTAMINANT <sup>a</sup>	SPECIES/LOCATION	DATE	N	TISSUE	RESIDUE LEVEL <sup>b</sup>	LEVELS USED/REMARKS	REFERENCE																												
Acephate (OP)	- <u>Rana clamitans</u> /lab	June 1975	9-10 per pool	Tadpoles	Approximately 0.60 after 1 day (identical to concentration in water).	Exposed to 1 ppm.	Lyons et al., 1976																												
	<u>Rana sylvatica</u> /Larose Forest, Ontario			Tadpoles	0.27 after 1 hour; 0.12 at day 1 (similar to water concentration).	Aerially sprayed at rate of 560 g a.i./ha.																													
Acridrine	- <u>Xenopus laevis</u> /lab		5 per 200 ml test solution	Larvae	Maximum concentration in larvae reached 85 ppm wet weight in 60-80 mine.  Elimination, when larvae placed in clean water, was rapid-acridrine was not detectable after 2 hrs.	Larvae exposed to 5.0 mg/L.  Acridrine animal/ Acridrine water = 170.	Davis et al., 1981																												
Aldrin (OC)	- <u>Bufo americanus</u> /Missouri corn fields	1965-67	2 60	Whole body	<table border="1"> <thead> <tr> <th colspan="2">Sample</th> <th colspan="5">Residues</th> </tr> <tr> <th>Wet (g)</th> <th>Dry (g)</th> <th>Lipid (g)</th> <th>Aldrin</th> <th>Dieldrin</th> <th>Range A + D</th> <th>DDI DDC DDD</th> </tr> </thead> <tbody> <tr> <td>10.00</td> <td>2.17</td> <td>0.0930</td> <td>0.03</td> <td>1.37</td> <td>1.25-1.50</td> <td>0.13</td> </tr> <tr> <td>10.01</td> <td>1.75</td> <td>0.2112</td> <td>-</td> <td>4.60</td> <td>2.31-8.30</td> <td>0.07</td> </tr> </tbody> </table>	Sample		Residues					Wet (g)	Dry (g)	Lipid (g)	Aldrin	Dieldrin	Range A + D	DDI DDC DDD	10.00	2.17	0.0930	0.03	1.37	1.25-1.50	0.13	10.01	1.75	0.2112	-	4.60	2.31-8.30	0.07	2 fields treated with 1.121 kg/ha for 15 and 16 of the past 17 years. Young toads (4 pooled samples of 10 tadpoles each) in August contained three times the residues of adults specimens collected in June.	Korschgen, 1970
				Sample		Residues																													
	Wet (g)	Dry (g)	Lipid (g)	Aldrin	Dieldrin	Range A + D	DDI DDC DDD																												
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10.01	1.75	0.2112	-	4.60	2.31-8.30	0.07																													
- <u>Rana pipiens</u> /lab	Ventral abdominal skin	Decreasing rate of accumulation by skin vs. time.	Pithed frogs were suspended in solution of 11 ppb.	Kaiser and Dunham, 1972-73																															

Table 4 - Residues (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES/LOCATION	DATE	N	TISSUE	RESIDUE LEVEL <sup>b</sup>	LEVELS USED/REMARKS	REFERENCE																																								
Aminocarb (C)	- Québec <u>Bufo americanus</u>  <u>Rana sylvatica</u>	1984	3 3 2 1 1 1 1	Larvae	<table border="1"> <thead> <tr> <th colspan="2">Days Post-Spray</th> <th>Stane</th> <th colspan="2">Residue</th> </tr> </thead> <tbody> <tr> <td>0-25</td> <td>25-31</td> <td>.109</td> <td colspan="2">.128</td> </tr> <tr> <td>1-25</td> <td>26-31</td> <td>.023</td> <td colspan="2">.025</td> </tr> <tr> <td>3-0</td> <td>28-35</td> <td>.022</td> <td colspan="2">.026</td> </tr> <tr> <td>9-0</td> <td>28-36</td> <td>.007</td> <td colspan="2"></td> </tr> <tr> <td>14-0</td> <td>31-40</td> <td>&lt;.005</td> <td colspan="2"></td> </tr> <tr> <td>0-25</td> <td>31-32</td> <td>.022</td> <td colspan="2"></td> </tr> <tr> <td>12-0</td> <td>29-37</td> <td>&lt;.005</td> <td colspan="2"></td> </tr> </tbody> </table>	Days Post-Spray		Stane	Residue		0-25	25-31	.109	.128		1-25	26-31	.023	.025		3-0	28-35	.022	.026		9-0	28-36	.007			14-0	31-40	<.005			0-25	31-32	.022			12-0	29-37	<.005			Aerially treated with 87.5 g a.i./ha.  No metabolites detected.	Hamerbach et al., 1987
					Days Post-Spray		Stane	Residue																																							
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12-0	29-37	<.005																																													
Atrazine (H)	- <u>Rana calesabajana</u> /ponds in Kansas	1973-74   1973	1	Tadpoles	Residues (ppm) of atrazine during 1973 and 1974:	Application with 0.3 ppm both summers.	Kilbourne and Kedon, 1979																																								
					<table border="1"> <thead> <tr> <th colspan="5">1973</th> <th colspan="5">1974</th> </tr> <tr> <th colspan="5">Days post treatment</th> <th colspan="5">Days post treatment</th> </tr> </thead> <tbody> <tr> <td>0</td><td>1</td><td>22</td><td>55</td><td>120</td> <td>0</td><td>2</td><td>23</td><td>51</td><td>85</td> </tr> <tr> <td>ND*</td><td>ND*</td><td>-</td><td>-</td><td>-</td> <td>0.289</td><td>0.278</td><td>0.309</td><td>0.235</td><td></td> </tr> </tbody> </table>	1973					1974					Days post treatment					Days post treatment					0	1	22	55	120	0	2	23	51	85	ND*	ND*	-	-	-	0.289	0.278	0.309	0.235		*ND - less than 0.4 ppb.	
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<table border="1"> <thead> <tr> <th colspan="2">Pre-treatment</th> <th colspan="2">3 days</th> <th colspan="2">21 days</th> <th colspan="2">77 days</th> </tr> <tr> <th>Atr.</th><th>Carb.</th> <th>Atr.</th><th>Carb.</th> <th>Atr.</th><th>Carb.</th> <th>Atr.</th><th>Carb.</th> </tr> </thead> <tbody> <tr> <td>ND*</td><td>ND</td> <td>0.276</td><td>ND</td> <td>0.201</td><td>ND</td> <td>0.207</td><td>ND</td> </tr> </tbody> </table>	Pre-treatment		3 days		21 days		77 days		Atr.	Carb.	Atr.	Carb.	Atr.	Carb.	Atr.	Carb.	ND*	ND	0.276	ND	0.201	ND	0.207	ND	Application with 0.3 ppm atrazine and 0.025 ppm carbofuran.																						
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Table 4 - Residues (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES/LOCATION	DATE	N	TISSUE	RESIDUE LEVEL <sup>b</sup>										LEVELS USED/REMARKS	REFERENCE
					Residue (ppb) of atrazine and carbofuran at given intervals post-treatment:											
Atrazine (cont'd)		1973			Pre-treatment	2 days		23 days		54 days		92 days		Application with 0.3 ppm atrazine and 0.050 ppm carbofuran. *ND - less than 0.4 ppb. No bio-magnification of atrazine was observed.	Klaassen and Kedosa, 1979 (cont'd)	
					Atr	Corb	Atr	Corb	Atr	Corb	Atr	Corb	Atr			Corb
					ND	ND	0.277	ND	-	-	-	-	-			-
Benzene Hexachloride (OC) (BHC)	- <u>Rana catesbeiana</u> <u>R. sphenoccephala</u> <u>R. c. clamitana</u> / Oxbow lakes in Louisiana	1980	5 3 3		Not detected in all three species.										Lakes in fertile alluvial cropland. Subjected to heavy pesticide applications. Annual rainfall - 150 cm.	Niethammer et al., 1984
Benzo(a)pyrene	- <u>Pleurodeles waltii</u> /lab			Larvae	Ratio of BaP in larvae to that in surrounding water after 12 hours was approximately 200; not dependent on dose; maximal levels attained after 12 hours. Contaminated larvae placed in uncontaminated water lost 99% contaminant after 100 hrs.										Dose levels between 0.075 - 0.3 x 10 <sup>-3</sup> ppm tested.	Grinfeld et al., 1986
Cadmium (M)	- <u>Rana temporaria</u> /lab			Liver, kidney, skin	Subcutaneous injections of 0.12-0.24 mg/100 g/day for 10 days resulted in significant accumulation in liver and kidney. Within 10 days of exposure to >0.002% CdCl <sub>2</sub> in the aquatic environment, significant amounts Cd found in skin; small amounts found in liver and kidney.											Vesil'eva et al., 1987

Table 4 - Residues (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES/LOCATION	DATE	N	TISSUE	RESIDUE LEVEL <sup>b</sup>		LEVELS USED/REMARKS	REFERENCE
					Concentrations (mean ± SD):			
Cadmium (cont'd)	- lab <u>Rana japonica</u> <u>R. nigromaculata</u>		3-3 per dose	Liver	Cd	Cu	0.225 Cd/kg body wt. injected i.v. 11 times during 15 days; frogs killed 2 days after last injection. No effect on concentrations of 9 other elements.	Suzuki et al., 1986
				Kidney	33.5 ± 21.0*	175 ± 24		
				Liver	65.7 ± 27.1*	9.04 ± 2.99		
				Kidney	33.9 ± 35.6*	48.2 ± 19.9*		
					36.8 ± 22.5*	10.0 ± 3.5		
					* Significantly different than controls.			
	- <u>Plethodon cinereus</u> , <u>P. glutinosus</u> , <u>Bufo americanus</u> , <u>B. woodhousei</u> , <u>Rana sylvatica</u> /oak forest 10 km upwind from 2 zinc smelters in eastern Pennsylvania	June - Oct., 1979	23	Whole body	Mean ± SE (ppm dry weight) for all species: 1.4 ± 0.1.			Beyer et al., 1985
	- Unspecified species/ New Lead Belt, Southwestern Missouri	Sept. 1972	8 pools 1 pool	Tadpoles	Area	Residues (ppm dry wt.)	Dam across Strother Creek provides the principal tailings pond for mine and mill effluents. Samples taken from below dam.	Gale et al., 1973
					Strother Creek	1.4-3.0		
					Control	1.1		

Table 4 - Residues (cont'd)

CONTAMINANT*	SPECIES/LOCATION	DATE	N	TISSUE	RESIDUE LEVEL <sup>b</sup>	LEVELS USED/REMARKS	REFERENCE		
Cadmium (cont'd)	- <u>Rana catesbeiana</u> / lead mining district, southeastern Missouri  - Patuxent Wildlife Research Centre <u>Rana catesbeiana</u> <u>R. clamitans</u> <u>Bufo</u> spp.  <u>Rana</u> spp.  - <u>Xenopus laevis</u> /lab	1981-82	15	Whole body	Residues (geometric mean, range in parentheses):			Site A - upstream from lead belt, sites B and C- downstream from tailings ponds.  Collected from uncontaminated areas. Some of <u>Bufo</u> spp. were captured on or near highways.  Exposed to 29 ug/L cadmium chloride in a dynamic test system.	Niethammer et al., 1985  Hall and Mulhern, 1984  Canton and Slooff, 1982
					Site A	Site B	Site C		
					not detected	0.31* (nd-0.89)	0.26* (nd-0.64)		
					* Significantly higher than Site A.				
					5 pools	Whole body	0.16-0.24		
					2 pools	Whole body	0.10-0.19		
					4	Whole body	0.15-4.0		
					2	Liver	0.08-0.13		
					1 pool	Kidney	1.9		
					12	Carcass	0.19-7.3		
11	Whole body	0.10-0.36							
Cd <sup>2+</sup> content (ug/kg wet wt.) following 100 day exposures:					Control	Cd-exposed			
2 pools of 2	Whole body	0.05 ± .01	3.77 ± .25						
4	Liver	0.26 ± .16	4.02 ± 3.09						
4	Kidney	0.76 ± .21	8.22 ± 1.86						

Table 4 - Residues (cont'd)

CONTAMINANT*	SPECIES/LOCATION	DATE	N	TISSUE	RESIDUE LEVEL <sup>b</sup>	LEVELS USED/REMARKS	REFERENCE				
Cadmium (cont'd)	- <u>Bufo regularis</u> /lab  - <u>Rana perezi</u> / Donana Natl. Park, Spain  - <u>Rana perezi</u> / Donana Natl. Park, Spain  - <u>Iriturus vulgaria</u> <u>Rana esculenta</u> <u>R. temporaria</u> <u>Palombes fuscus</u> <u>Bufo viridis</u> /Poland		260	Adult females 25 ± 4.3 g	Concentrations (estimated from graph, ng/g) following single dose i.m. injection of 6.2 mg Cd <sup>2+</sup> /kg in thigh				Injection of EDIA reduced Cd level in all organs except kidneys.	Hilmy et al., 1986c	
					24 hr	48 hr	72 hr	96 hr			
					Liver	90	127	93			101
					Heart	13	16	21			14
					Kidney	29	31	47			30
					Lung	5	18	10			13
					Spleen	1	3	3			3
					Blood	17	4	2			2
					Muscle	72	36	14			14
					10 composite samples	"A fillet with bones, skin"	Geometric mean: 0.10.				
7	Muscle	0.08 0.07 0.19					Rico et al., 1987				
8	Whole body	Location		Residues (ppm dry weight) range in brackets		Revealed some evidence of food chain accumulation.	Dmowski and Karolowski, 1979				
1		Protected zone near a zinc mill	7.9 (1.3-14.4)								
11		Pine forest near a zinc mill Control area	10.7 0.7 (0.3-1.4)								

Table 4 - Residues (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES/LOCATION	DATE	N	TISSUE	RESIDUE LEVEL <sup>b</sup>	LEVELS USED/REMARKS	REFERENCE		
Cadmium (cont'd)	- <i>Rana pipiens</i> /Icb		100 per treatment	Larvae	Cd Concentration			Exposed to Cd-enriched sediment as embryos to 4 days post-hatch.  <sup>a</sup> mean $\pm$ SD for 3 replicate treatments.  <sup>b</sup> composite from 3 replicate treatments.	Francis et al., 1984
					Sediment <sup>a</sup> (ug/kg)	Water <sup>a</sup> (ug/L)	Tissue <sup>b</sup> (ug/g)		
					1.04 $\pm$ 0.6	1.0 $\pm$ 0.6	-		
					2.28 $\pm$ 0.14	1.1 $\pm$ 0.8	0.08		
					11.48 $\pm$ 0.21	2.1 $\pm$ 4.4	0.14		
					96.8 $\pm$ 2.4	4.4 $\pm$ 1.8	3.08		
					1074 $\pm$ 14	76.5 $\pm$ 17.1	12.55		
Carbaryl (C)	- <i>Rana pipiens</i> /Icb			Skin	Half time rate of dermal penetration (mins.): 6.4.		1 mg/kg total insecticide applied to a 1 cm <sup>2</sup> area on back immediately behind head.	Shah et al., 1983	
Carbofuran (see atrazine)									
Cesium	- <i>Hyla cinerea</i> /contaminated floodplain of Savannah River, South Carolina  /Icb	June - Aug. 1972	141	Whole body	Mean $\pm$ S.E. body burden (pCi/g dry wt.): 204.2 $\pm$ 6.6.		Approximately 261 Ci cesium released from reactors upstream between 1960-1970.	Dapson and Kaplan, 1975	
			33	Whole body	Biological half-life in unfed frogs averaged 30.1 days.		Frogs held at 20°-30°.		

Table 4 - Residues (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES/LOCATION	DATE	N	TISSUE	RESIDUE LEVEL <sup>b</sup>	LEVELS USED/REMARKS	REFERENCE							
Chlordanes (OC)	- <i>Ierichia torosa</i> /British Columbia		N/Days post-treatment		Techn. Chlor-dane						Lake in B.C. sprayed with technical chlordane to a level of 0.010 ppm.  Metabolized and eliminated chlordane more effectively than cutthroat trout.	Albright et al., 1980		
					4/14	Whole body	8.906	1.285	1.132	.857			NA	0.959
						Liver	26.321	3.566	2.883	1.992			NA	3.210
						Stomach	2.233	.301	.232	.161			NA	0.230
					8/279	Whole body	3.425	.452	.231	ND			0.310	1.043
						Liver	6.236	.590	.285	ND			0.373	1.406
						Stomach	.756	.112	.041	ND			0.054	0.222
					6/451	Whole body	2.116	.327	.144	ND			0.153	0.668
						Liver	3.106	.392	.172	ND			0.196	0.925
						Stomach	.322	.035	.013	ND			0.013	0.063
					8/1,036	Whole body	.176	ND	ND	ND			0.010	0.090
	Liver	.289	ND	ND	ND	0.016	0.160							
	Stomach	Trace	ND	ND	ND	ND	Trace							
	- <i>necturus lewisi</i> /Neuse River drainage, North Carolina		50	Whole body	Geometric Mean of residues:				* not detected at 0.01 ppm level of sensitivity.	Hail et al., 1985				
				Cis-chlordane	Trans-nonachlor	Cis-nonachlor	Oxy-chlordane							
				0.02	0.04	ND*	ND							
Chlorophenols	- <i>Rana catesbeiana</i> /Canagogue Creek, Ontario	Nov. 13 1980		Tadpoles	2,6-DCP*	2,4,6-ICP	2,4,5,-ICP	PCP	*DCP - dichlorophenol; ICP - trichlorophenol; PCP - pentachlorophenol. Creek receives domestic and industrial sewage effluent.	Hutchins et al., 1984				
					84,000	10,000	31,000	19,000						
					2,4-DCP; 3,4-DCP; 2,3,4,6-ITCP not detectable.									

Table 4 - Residues (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES/LOCATION	DATE	N	TISSUE	RESIDUE LEVEL <sup>b</sup>	LEVELS USED/REMARKS	REFERENCE
Chromium (M)	- <i>Rana esculenta</i> River Icco, Italy	April- Sept. 1974	150	Tadpoles	Residues (ppm dry wt.): 0.42-9.22.	Standard deviations in brackets.	Baudo, 1976
					Neometamorphosed Frogs		
Copper (M) (see also cadmium)	- <i>Rana esculenta</i> / South Moravia, Czechoslovakia	1982- 1984		Whole body	Mean copper content (ppm dry wt.):	Brno: Suburban gardens, fields, lakes. Musov: large water reservoir. Lednice: abandoned sand pit.	Pavel and Kucera, 1986
					Locality		
					1982 1983 1984		
					Brno 10.5 ± 1.1 (10)* 11.8 ± 0.8 (8) -		
					Musov 10.7 ± 0.7 (12) 12.5 ± 0.4 (12) 14.8 ± 1.2 (10)		
					Lednice 6.2 ± 0.6 (13) 10.6 ± 1.1 (8) -		
					* Number of samples.		

Table 4 - Residues (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES/LOCATION	DATE	N	TISSUE	RESIDUE LEVEL <sup>b</sup>	LEVELS USED/REMARKS	REFERENCE
Copper (cont'd)	- <i>Rana perezi</i> /Donana Natl. Park, Spain		10 Composite samples	"A fillet with bones, skin"	Geometric mean: 1.16.	Standard deviations in brackets.	Hernandez et al., 1987
	- <i>Rana perezi</i> /Donana Natl. Park, Spain	1984 1985 1986	7 4 2	Muscle	0.80 0.28 2.61		Rico et al., 1987
	- <i>Rana esculenta</i> / River Icco, Italy	April- Sept. 1974	150	Tadpoles	Residues (ppm dry wt.): 7.63-32.40.		Baudo, 1976
					Neometamorphosed Frogs	Adults male female	
					Neomet. Liver 50.86 (22.52) 63.08 (24.87) 61.44 (50.54)		
					Frogs = Heart & Lungs 10.22 (5.22) 10.49 (2.97) 7.20 (3.18)		
					43; Kidneys 4.53 (2.07) 10.71 (7.49) 6.69 (3.71)		
					males = Muscles 5.01 (2.64) 2.82 (1.91) 2.53 (1.67)		
					10-22; Skin 9.18 (1.15) 5.95 (2.08) 6.14 (3.00)		
					females = Gonads - 3.71 (1.30) 10.20 (3.88)		
					10-28 Eggs - - 9.54 (0.90)		
	- Unspecified species/ New Lead Belt, southeastern Missouri	Sept. 1972	8 pools 1 pool	Tadpoles	Area Residues (ppm dry wt.)	Dam across Strother Creek provides the principal tailings pond for mine and mill effluents. Samples taken from below dam.	Gale et al., 1973
					Strother Creek Control 17-44 8		

Table 4 - Residues (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES/LOCATION	DATE	N	TISSUE	RESIDUE LEVEL <sup>b</sup>	LEVELS USED/REMARKS	REFERENCE
Copper (cont'd)	- Unspecified species/ New Lead Belt southeastern Missouri		1 pool 2 pools 2 pools 1 pool	Tadpoles Liver & Heart Eviscerated Intestine Whole body	91 ppm. 15-20 ppm. 234-260 ppm. 169 ppm.	Intestine included contents.	Jennett et al., 1979
	- Patuxent Wildlife Research Center						Hall and Mulhern, 1980
	<u>Rana catesbeiana</u> <u>R. clamitans</u> <u>Bufo</u> spp. <u>Rana</u> spp.		5 pools 2 pools 6 11	Tadpoles Tadpoles Whole body Whole body	1.4-3.2 0.93-1.2 2.1-5.0 1.2-3.5		
	- <u>Rana temporaria</u> / northern Finland	1971-1972	106	Liver Male Fecula	Mean ± SE (ppm dry wt.): 156.9 ± 19.3 (entering wintering) - 503.2 ± 33.8 (emerging). 314.0 ± 69.6 (entering wintering) to 845.1 ± 85.6 (spawning).		Pasanen and Koskela, 1974
	- <u>Bufo carinus</u> , <u>Hyla</u> sp., <u>Limnodynastes</u> sp.		21	Liver	10-1640 ppm dry wt.		Beck, 1956
- <u>Bufo marinus</u> /Dominican Republic		6	Liver	367-2091 ppm dry wt.		Goldfischer et al., 1970	

Table 4 - Residues (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES/LOCATION	DATE	N	TISSUE	RESIDUE LEVEL <sup>b</sup>	LEVELS USED/REMARKS	REFERENCE
Copper (cont'd)	- <u>Plethodon cinereus</u> , <u>P. glutinosus</u> , <u>Bufo americanus</u> , <u>B. woodhousei</u> , <u>Rana sylvatica</u> /oak forest 10 km upwind from 2 zinc smelters in eastern Pennsylvania	June-Oct. 1979	23	Whole body	Mean ± SE (ppm dry weight) for all species: 5.8 ± 0.4.		Beyer et al., 1985
	- <u>Xenopus laevis</u> /lab		10 per dose	Liver Kidney	Concentration (mean ± SD):  Control                      Cu 30.0 ± 40.2                      41.5 ± 24.9 9.15 ± 1.73                      28.7 ± 4.4*	1 mg Cu/kg body wt. injected i.m. daily for 5 days; frogs killed 1 day after last injection.	Suzuki et al., 1981
Cypermethrin (PY)	- <u>Rana temporaria</u> /lab		≥8	Brain	Mean ± SE associated with acute toxic signs: 0.08 ± 0.03 ppm.	Value is lower than that for trout, mouse, quail.	Edwards and Milburn, 1985
2,4-D (H)	- <u>Rana temporaria</u> /lab			Tadpoles	Not detected (0.1 ug detection limit).	Treated in 50 ppm for 48 hours.	Cooke, 1972
DDD (OC) (see also aldrin, DDT)	- <u>Rana catesbeiana</u> / Clear Lake, California	May 1958	9	Visceral fat	5 ppm.	DDD sprayed periodically 1949-1958.	Hunt and Bischoff, 1960

Table 4 - Residues (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES/LOCATION	DATE	N	TISSUE	RESIDUE LEVEL <sup>b</sup>				LEVELS USED/REMARKS	REFERENCE		
					Lake Providence		Lake Bruin					
					Residue (geometric mean)	% lipid	Residue (geometric mean)	% lipid				
DDE (OC) (see also aldrin, DDT)	- Onbow lakes in Louisiana	1980							Lakes in fertile alluvial cropland. Subjected to heavy pesticide applications. Annual rainfall - 130 cm. Biomagnification through the food chain was evident. Frogs had generally low levels due to increased mobilization and excretion, thus not useful as an indicator.	Niethammer et al., 1984		
	<u>Rana catesbeiana</u>		5		0.25	1.7	0.03	0.4				
	<u>R. sphenoccephala</u>		3		0.64	3.3	0.04	1.1				
	<u>R. c. clamitans</u>		3		0.05	1.1	0.04	0.6				
	- <u>Rana perezi</u> /Donana Natl. Park, Spain		10	Composite samples	Geometric mean: 0.02.							Hernandez et al., 1987
	- <u>Rana perezi</u> /Donana Natl. Park, Spain	1985	4	Muscle	0.02							Hico et al., 1987
		1986	2		0.19							
	- Iowas			Viscera	Sex	Weight (g)	Residue (ppb)				Collections from agricultural areas. Numbers in parentheses show the range of values. Values low due to method of analysis used.	Funzo et al., 1979
	<u>Bufo americanus</u>	1974	4		F	9.45 (9.10-10.27)	Not detected (ND)					
			17		M	3.85 (2.02-6.23)	19 (ND-176)					
	1974	4		F	1.50 (0.19-4.29)	ND						
		3		M	0.25 (0.04-0.71)	ND						

Table 4 - Residues (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES/LOCATION	DATE	N	TISSUE	RESIDUE LEVEL <sup>b</sup>					LEVELS USED/REMARKS	REFERENCE
					Location	Average Altitude (m)	Distance from spray area (km)	N	Residues $\pm$ S.E.		
DDE (cont'd)	- <u>Rana boylei</u> /Sierra Nevada Mountains, California	1970		Fat bodies	Occurrence of p,p'-DDE in fat bodies of frogs:					Concentrations probably represent locally persistent residues of 1953 and 1956 spraying. Concentrations highest in southern and central area due to wind drift.	Cory et al., 1970
					Northern	1993.9	90.0	19	1.32 $\pm$ 0.16		
					Sonora	3027.9	136.7	35	5.38 $\pm$ 0.93		
					Central:						
					west face	1127.3	56.7	12	3.46 $\pm$ 0.25		
					west face	2953.0	128.3	48	3.19 $\pm$ 0.27		
				east of crest	2941.8	153.3	9	0.97 $\pm$ 0.19			
				southern	3300.9	90.0	19	2.07 $\pm$ 0.26			
DDMU (OC)	- Onbow lakes in Louisiana	1980			Not detected in all 3 species.					Lakes in fertile alluvial cropland. Subjected to heavy pesticide applications. Annual rainfall - 130 cm.	Niethammer et al., 1984
<u>Rana catesbeiana</u>		5									
<u>R. sphenoccephala</u>		3									
	<u>R. c. clamitans</u>		3								

Table 4 - Residues (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES/LOCATION	DATE	N	TISSUE	RESIDUE LEVEL <sup>b</sup>				LEVELS USED/REMARKS	REFERENCE		
					Data Found		Residues					
DDT (OC) (see also dieldrin)	- Colorado  Leopard Frog  Garden toad  - Oxbow lakes in Louisiana	1960    1980							Grazed rangeland with a few small stockpounds sprayed with 3.56 kg/ha DDT on June 15, 1960.  No DDD has ever been sprayed in Colorado thus residues must have been converted by animal tissues.  Lakes in fertile alluvial cropland. Subjected to heavy pesticide applications. Annual rainfall - 130 cm.  Biomagnification through the food chain was evident.	Finley and Pillmore, 1963    Niethammer et al., 1984		
							DDT	DDD				
					June 15	0.5						
					June 20	4						
					"	4						
"	1	1										
June 24	0.8	0.5										
June 29	2	0.7										
June 30	8											
		Lake Providence		Lake Bruin								
		Residues (geometric mean)	% lipid	Residues (geometric mean)	% lipid							
		0.03	1.7	ND	0.4							
		ND*	3.3	ND	1.1							
		ND	1.1	ND	0.6							
		* Not detected.										
				Compound		Geometric Mean of Residues						
				DDT	ND*							
				DDC	0.06							
				DDD	0.04							
						* ND - lower limit of reportable residues = 0.01 ppm						
	- <u>Necturus lewisii</u> Neuse River drainage North Carolina		50	Whole body					Hall et al., 1985			

Table 4 - Residues (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES/LOCATION	DATE	N	TISSUE	RESIDUE LEVEL <sup>b</sup>					LEVELS USED/REMARKS	REFERENCE
					Location	DDC	DDD	DDT	TOTAL		
DDT (cont'd)	- <u>Plethodon cinereus</u> / Forest, Northern Maine	July 1970    June 1972    August 1972	N = 10-14 pooled	Whole body	Location	DDC	DDD	DDT	TOTAL	Beaver Brook: sprayed at rate of 1.12 kg/ha in 1958, 1960, 1963.  Sterling Brook: sprayed at rate of 1.12 kg/ha in 1960, 1963, 1964.  Other 2 areas untreated; buffer of 1.5 km.	Banasiak, 1974
					Beaver Br.	-	-	-	.440		
					Sterling Br.	-	-	-	.150		
					Greenlaw Mt.	-	-	-	.020		
					Moaka Br.	-	-	-	.080		
					Beaver Br.	.035	tr	.035	.070		
					Sterling Br.	.050	.015	.055	.120		
					Greenlaw Mt.	tr	tr	.020	.020		
					Moaka Br.	tr	0	.025	.025		
					Beaver Br.	.045	tr	.060	.120		
					Sterling Br.	.140	.025	.150	.270		
					Greenlaw Mt.	.010	0	.045	.055		
					Moaka Br.	tr	0	.020	.020		



Table 4 - Residues (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES/LOCATION	DATE	N	TISSUE	RESIDUE LEVEL <sup>b</sup>			LEVELS USED/REMARKS	REFERENCE		
					Exposure (days)	Residue					
DDT (cont'd)	- <u>Rana temporaria</u> /lab			Tadpoles	Control (39)	pp'-DDE	pp'-DDT	Residues accumulated during chronic dosing did not cause mortality or behavioural abnormalities at body levels of 2-5 ppm. After acute exposure, tissue levels of 2 ppm can cause hyperactivity in tadpoles and death in small frogs.	Cooke, 1973b		
						0.006	0.02				
					0.0001 ppm DDT (55)	not detected	0.82				
						(39)	0.08			0.57	
					0.001 ppm DDT (15)	not detected	2.0				
						(33)	0.25			4.3	
	(39)	0.21	3.3								
	- <u>Rana temporaria</u> /lab		6	Liver Muscle	16.9 ± 4.0 ppm. 1.4 ± 0.5 ppm.					Exposure to 9 subcutaneous injections of 12 mg/kg.	Cooke, 1974
	- <u>Rana perezi</u> /Donana Natl. Park, Spain		10	"A fillet with bones, skin"	Geometric mean: 0.01.						Hernandez et al., 1987
	- <u>Rana perezi</u> /Donana Natl. Park, Spain	1984	7	Muscle	0.05						Rico et al., 1987
		1985	4		0.04						
		1986	2		0.55						

Table 4 - Residues (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES/LOCATION	DATE	N	TISSUE	RESIDUE LEVEL <sup>b</sup>			LEVELS USED/REMARKS	REFERENCE	
					Stage	Nominal conc. DDT (ppm)	Residue (% of DDT originally present in test medium)			
DDT (cont'd)	- <u>Rana pipiens</u> /lab			Ventral abdominal skin	Decreasing rate of accumulation by skin vs. time.			Pitted frogs were suspended in solution of 10 ppb.	Kaiser and Dunham, 1972-73	
	- <u>Rana temporaria</u> /lab		4 large, 4 small	Tadpoles: large - 689 ± 44 ng small - 365 ± 11 ng	Residues (mean ± S.E.): large: 2.5 ± 0.2 small: 7.5 ± 0.6					Tadpoles exposed to 0.1 ppm for 7 days in static system.
	- <u>Rana temporaria</u> /lab							Exposure for 1 hour. No DDE found in samples.	Cooke, 1970	
					I-V	0.01 0.01 1.0 10.0	16.0% 11.0% 2.5% 1.3%			
					IX-XVI	1.0 10.0	3.2% 1.3%			
	- <u>Rana temporaria</u> /lab			pool of 10 per group	Tadpoles younger than stage IV	Treatment Time (h)	Mean content of one tadpole (ug) pp' DDT      pp' DDE		Tadpoles exposed to 0.05 ppm.	Cooke, 1971
						5	0.50	<0.001		
						19	0.61 0.75 1.20	<0.001 0.01 <0.001		

Table 4 - Residues (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES/LOCATION	DATE	N	TISSUE	RESIDUE LEVEL <sup>b</sup>			LEVELS USED/REMARKS	REFERENCE
					Tadpoles used	Mean Residue			
DDT (cont'd)	<u><i>Rhinophrynus dorsalis</i></u>					DDT	DDE	Newts fed on <i>R. temporaria</i> tadpoles exposed as outlined on previous page.	Cooke, 1971 (cont'd)
					Control	0.36	<0.002		
					5 h exposure	0.23	0.62		
				19 h exposure	0.65	1.38			
	- <u><i>Rana sylvatica</i></u> /lab		100 per treatment	Liver, fat, head, lung, heart, gut, rest of body	Only trace residues (<0.01 ppm) found in head, lung, heart, gut and rest of body. High concentrations found in liver and fat. Mean residues in 10 frogs (treated with .003 ppm as tadpoles) sacrificed 14 days after metamorphosis: 37.4 ppm in fat, 9.7 ppm in liver.			Pre-metamorphic, stage 23 tadpoles were exposed to 0.001 or 0.003 ppm DDT for 7 days, then placed in fresh water. DDT content determined in young frogs after metamorphosis. Newly transformed frogs lost DDT more slowly while living a terrestrial life than did tadpoles.	Licht, 1976.
	- <u><i>Rana temporaria</i></u> /lab			Liver, fat body, gall bladder, kidney, stomach, brain, skeletal muscle, ovary, spleen, bone, spinal chord, oviduct, heart	Tissue DDT concentration was correlated with fat concentration of tissues; strong accumulation in fat body, liver, gall bladder, ovary, spleen and bone; minimal accumulation in spinal chord; no activity in kidney, oviduct, heart or skeletal muscle.			About 5 uCi force fed to frogs using gelatin capsules. DDT pre-treatment increased elimination rate.	Harris et al., 1975

Table 4 - Residues (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES/LOCATION	DATE	N	TISSUE	RESIDUE LEVEL <sup>b</sup>					LEVELS USED/REMARKS	REFERENCE		
					Temp. (°C)	Embryonic Stage							
DDT (cont'd)	- <u><i>Rana sylvatica</i></u> /lab		50 per treatment except 30 for stage 20 embryos	Embryos	9	0.188	0.150	0.145	0.956	Treated for 24 hrs. in 0.025 ppm.	95% confidence limits in parentheses.	Licht, 1985	
						(.214-.153)	(.160-.141)	(.151-.131)	(1.080-.832)				
						0.150	0.142	0.147	0.817				Jelly capsules around eggs restrict DDT uptake.
						(.159-.140)	(.153-.133)	(.153-.141)	(.864-.770)				
						0.167	0.158	0.160	1.57				
						(.179-.155)	(.171-.145)	(.173-.147)	(.173-1.37)				
	Jelly	9	0.015	0.017	0.018	-							
			(.016-.14)	(.019-.015)	(.020-.015)	-							
		15	0.015	0.015	0.019	-							
			(.016-.014)	(.017-.013)	(0.022-.016)	-							
21	0.015	0.015	0.017	-									
	(.016-.015)	(.016-.014)	(.019-.015)	-									
	- <u><i>Rana pipiens</i></u> /lab			Skin	Half time rate of dermal penetration (mins): 703.				1 mg/kg applied to 1 cm <sup>2</sup> area on back immediately behind head.	Shah et al., 1983			

Table 4 - Residues (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES/LOCATION	DATE	N	TISSUE	RESIDUE LEVEL <sup>b</sup>				LEVELS USED/REMARKS	REFERENCE
					Nominal Conc. (ppm)	Treatment Time (h)	DDT (ppm)	DDE (ppm)		
DDT (cont'd)	<u>Rana temporaria</u> /lab		10-30	Tadpoles at external gill stage	19.4 ppm detected in tadpoles hatched from fresh spawn exposed to DDT. No DDT detected in tadpoles hatched from spawn treated later in development. DDE not detected in any samples.				Spawn immersed for 1 hour in 0.5 ppm DDT. Detection level 0.01 ug/sample.	Cooke, 1972
				Tadpoles:						
			10 per group	External gills	0.5	24	75.5	ND	ND = not detected (<0.01 ug/sample).	
				Internal gills, no hind limbs	0	24	ND	ND		
					.005	48	ND	ND		
						24	2.4	ND		
						48	3.9	ND		
				.05	24	15.1	ND			
					48	19.4	ND			
				.5	24	55.2	ND			
Hind limb paddles or hind legs	0	24	0.23	ND						
		48	ND	ND						
	.0008	24	0.17	0.01						
		48	0.29	0.03						
	.02	24	2.4	0.12						
		48	5.6	0.19						
	.5	24	36.6	0.67						
Large hind legs	0	24	ND	ND						
	.5	24	20.2	ND						

Table 4 - Residues (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES/LOCATION	DATE	N	TISSUE	RESIDUE LEVEL <sup>b</sup>				LEVELS USED/REMARKS	REFERENCE
					Conc. (ppm)	Treatment Time (h)	DDT (ppm)	DDE (ppm)		
DDT (cont'd)	<u>Bufo bufo</u>		10 per group	Tadpoles:					Tadpoles exposed for 24 or 48 hours. ND = not detected (<0.01 ug/sample).	Cooke, 1972 (cont'd)
				External gills	0	24	ND	ND		
						48	ND	ND		
					.005	24	ND	ND		
						48	ND	ND		
					.05	24	30.0	ND		
						48	ND	ND		
					.5	24	82.0	ND		
						48	306	ND		
				Internal gills, no hind limbs	0	24	ND	ND		
		48	ND		ND					
	.005	24	6.9		ND					
		48	13.5		0.40					
	.05	24	65.2		1.0					
			48	139	ND					
		.5	24	326	ND					
			48	478	ND					
	Hind limb paddle or hind legs	0	24	ND	ND					
			48	ND	ND					
		.0008	24	0.09	ND					
		48	0.54	ND						
.02		24	3.5	0.24						
		48	7.6	0.22						
	.5	24	73.2	3.0						
		48	134	5.1						
<u>Triturus vulgaris</u>	5 per group		Tadpoles with 4 legs and external gills	0	24	ND	ND			
					48	ND	ND			
				.005	24	1.5	ND			
					48	3.0	0.13			
				.5	24	86.3	2.7			
		48	116	2.2						

Table 4 - Residues (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES/LOCATION	DATE	N		TISSUE	RESIDUE LEVEL <sup>b</sup>		LEVELS USED/REMARKS	REFERENCE						
			1966	1967		Years of DDT spray treatment	DDT metabolites (ppm)								
DDT (cont'd)	- Plethodon cinereus/ forests in Maine	1966-67			Whole body		1966	1967	DDT comprised 40 - 60% of total residues; DDD comprised 5-10% of total.  Seasonal variation in levels suggested, with higher levels in spring. Possible explanations include differing age structure of population, seasonal differences in food habits, elimination of DDT in females' eggs.	Diamond et al., 1968					
								22					1967		0.528
								17							0.648
								6							0.168
								7							0.448
								3							2.057
								7					1966		0.237
								2							0.104
								19					1963		0.101
								6			22		1961		0.128
								7			6				0.110
											8				0.058
											3				0.138
											27				0.091
											4	25	1960		0.246
											7	29			0.190
												22			0.037
											16	30	1958		0.044
											5	12			0.079
		6		63-60-58		0.157									
		23	33	64-63-60		0.242									
			12			0.240									

Table 4 - Residues (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES/LOCATION	DATE	N	TISSUE	RESIDUE LEVEL <sup>b</sup>			LEVELS USED/REMARKS	REFERENCE
					Time after Application	Residues (mean ± SE, ppm)	Residues (ppm)		
DDT (cont'd)	- Marsh at southwestern edge of Lake Erie <u>Rana pipiens</u>  <u>Rana catesbeiana</u>  <u>Rana clamitans</u>	July 1964		Whole body	8 hr	0.2 ± 0.0	Treated by helicopter with 0.22 kg/ha technical DDT (chlorine - 36 ring-labeled).  *ND = not detected.	Meeks, 1968	
					1 day	1.3 ± 0.5			
					1 week	1.2 ± 0.5			
					1 month	1.2 ± 1.1			
					2 months	0.3			
					9 months	0.3 ± 0.1			
					12 months	0.2			
					Months after Application	weight (g)			Residues (ppm)
					2	61			1.8
					12	240			0.5
					13	290			0.3
					2	61			1.8
					12	240			0.9
					13	290			ND*
					2	61			-
					12	240			-
					13	290			0.4
					2	61			1.5
					12	240			ND
					13	290			-
2	61	0.2							
12	240	ND							
13	290	0.3							
2	61	1.7							
12	240	0.6							
13	290	0.6							
	Heart, Brain, Eye, Lung, Blood, Femur, Pancreas, Spleen	No detectable residues.							
		No residues detected in tissues.							

Table 4 - Residues (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES/LOCATION	DATE	N	TISSUE	RESIDUE LEVEL <sup>b</sup>	LEVELS USED/REMARKS	REFERENCE					
DDT (cont'd)	<u>Rana sylvatica</u> /lab		5	Liver of tadpoles	Residues (ppm) of DDT in liver following treatment for seven days in different concentrations of DDT:	Figures given are the mean of five animals.  95% confidence limits in brackets.  Tissue levels reached their maximum in 24 hours.	Licht, 1976b					
					Hours after treatment			TREATMENT				
								21°C		15°C		
								0.001 ppm	0.003 ppm	0.001 ppm	0.003 ppm	
					24			9.73 (23.8 to -4.37)	35.84 (61.51 to 10.17)	10.49 (15.13 to 5.84)	27.04 (52.27 to -0.18)	
					48			7.03 (28.06 to 5.30)	5.65 (7.96 to 3.34)	38.85 (64.05 to 13.65)	21.20 (37.20 to 5.18)	
					72			14.86 (29.23 to 0.49)	10.40 (14.80 to 6.00)	20.96 (42.84 to -0.91)	18.26 (28.38 to 8.14)	
96	2.96 (4.86 to 1.06)	6.20 (9.53 to 2.88)	16.51 (34.85 to -1.81)	16.73 (27.92 to 5.54)								
168	2.67 (5.01 to 0.34)	5.27 (11.38 to -0.84)	9.43 (16.15 to 2.71)	9.71 (16.28 to 3.13)								
DEHP (di-2-ethylhexyl-phthalate)	<u>Rana arvalis</u> /lab		100 eggs per treatment	Tadpoles	DEHP accumulated in tadpoles at concentrations ranging from 0.28-246.80 ppm fresh weight.	Exposures for 60 days; DEHP mixed into sediment so levels of 10-800 ppm were obtained.	Larsson and Inurén, 1987					

Table 4 - Residues (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES/LOCATION	DATE	N	TISSUE	RESIDUE LEVEL <sup>b</sup>	LEVELS USED/REMARKS	REFERENCE			
Dieldrin (OC) (see also aldrin)	<u>Rana catesbeiana</u> <u>R. sphenoccephala</u> <u>R. c. clamitans</u>	1980	5		Not detected in all three species.	Lakes in fertile alluvial cropland. Subjected to heavy pesticide applications. Annual rainfall 130 cm.	Niethammer et al., 1984			
			3							
			3							
		<u>Rana</u> sp./Louisiana	1978		Legs Whole body	.010 ppm (mean is exceeded by standard error). Not detected (detection limit .05 ppm).		Dowd et al., 1985		
		<u>Bufo americanus</u>	1974	4	Viscera	Sex	Weight (g)	Residues (ppb)	Collections from agricultural areas. * Not detected. Numbers in parentheses show the range of values.	Punzo et al., 1979
				17		F	9.45 (9.10 - 10.27)	B (ND* - 14)		
						M	3.85 (2.02 - 6.23)	10 (ND - 33)		
		<u>Rana pipiens</u>	1974	4		F	1.50 (0.19 - 4.29)	ND		
				3		M	0.25 (0.04 - 0.71)	ND		
		<u>Rana pipiens</u> /lab			Skin	Half time rate of dermal penetration (mins.): 3766.	1mg/kg applied to 1 cm <sup>2</sup> area on back immediately behind head.	Shah et al., 1983		
	<u>Rana pipiens</u> /lab			Ventral abdominal skin	Decreasing rate of accumulation by skin vs. time.	Pithed frogs were suspended in solution of 110 ppb.	Keiser and Dunham, 1972-73			



Table 4 - Residues (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES/LOCATION	DATE	N	TISSUE	RESIDUE LEVEL <sup>b</sup>	LEVELS USED/REMARKS	REFERENCE
Dinoseb (DP)	- <u>Pseudacris triseriata</u> / hay meadows near Laramie, Wyoming	June	175	Whole body	No residues detected in frogs collected 1-3 days after treatment. Lower limit of detection was 0.01 ppm.	Sprayed at rate of 52 g a.i./ha as either oil-based formulation (2.3 L No. 2 diesel oil/ha) or water-based (emulsifier and 2.3 L water/ha). Concluded frogs not a likely source of secondary poisoning for predatory vertebrates.	Powell et al., 1982
	- <u>Rana catesbeiana</u> /lab			Tadpoles	Significant bioconcentration; average magnification 62 times. Maximum level found in pooled samples was 320 ppm. No fenoxon (metabolite) detected in tissues at sensitivity limit of 0.5 ppm.	Exposed to .01-1 ppm for 96 hr in continuous flow apparatus.	Hall and Kolbe, 1980
Malathion (PY)	- <u>Bufo fowleri</u> , <u>Rana utricularis</u> , <u>R. clamitans</u> , <u>Hyla crucifer</u> /in and around cotton fields near Garland, Arkansas	July 25-26, 1979	1 or 2 (pooled)	Carcass with skin and g.i. tract removed	0.02 (na2) found in <u>B. fowleri</u> . No detectable residues (0.01 ppm detection limit) found in other species.	Aerial application had occurred 5 days prior to collection at rate of 0.112 kg ai/ha. Applications had also been made in previous years.	Bennett et al., 1983

Table 4 - Residues (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES/LOCATION	DATE	N	TISSUE	RESIDUE LEVEL <sup>b</sup>	LEVELS USED/REMARKS	REFERENCE
Dieldrin (DC)	- <u>Rana perazi</u> /Donana Natl. Park, Spain		10 composite samples	"A fillet with bones, skin"	Geometric mean: 0.01.		Hernandez et al., 1987
	- <u>Rana perazi</u> /Donana Natl. Park, Spain	1986	2	Muscle	0.01		Rico et al., 1987
Heptachlor (DC)	- <u>Rana pipiens</u> /lab			Ventral abdominal skin	Decreasing rate of accumulation by skin vs. time.	Pitted frogs were suspended in solution of 6 ppb.	Keiser and Dunham, 1972-73
Heptachlor Epoxide (DC)	- Location not specified	1962			Residues (ppm) from animals found dead one month after spray:	Area sprayed with 2.24 kg/ha.	DeWitt et al., 1962
	Bullfrog		2		13.5		
	Leopard Frog		2		13.0		
	Green Frog		6		1.5		
	Toads		2		19.4		
- Toads				Viscera		Collections from agricultural areas. Values low due to method of analysis used. DDT was used as a general purpose insecticide in this region in past years and aldrin and heptachlor were used for control of soil-insect pests.	Punzo et al., 1979
	<u>Bufo americanus</u>	1974	4		Sex	Weight (range in brackets) (g)	Residues (ppm)
			17		F	9.45 (9.10-10.27)	Not detected (ND)
					M	3.85 (2.02-6.23)	ND
	<u>Rana pipiens</u>	1974	4		F	1.50 (0.19-4.29)	ND
			3		M	0.25 (0.04-0.71)	ND

Table 4 - Residues (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES/LOCATION	DATE	N	TISSUE	RESIDUE LEVEL <sup>b</sup>	LEVELS USED/REMARKS	REFERENCE		
Heptachlor epoxide (cont'd)	- <u>Terichia torosa</u> / British Columbia	1980	N/Dayo post treatment		Residue:	Lako in B.C. sprayed with technical chlordane to a level of 0.010 ppm.  No application rate given.	Albright et al., 1980		
			4/14	Whole body	0.303				
				Liver	0.962				
				Stomach	0.072				
			8/279	Whole body	ND (not detectable)				
	Liver	ND							
	Stomach	ND							
	6/451	Whole body	ND						
	Liver	ND							
	Stomach	ND							
	8/1,036	Whole Body	ND						
	Liver	ND							
	Stomach	ND							
Iron (M)	- <u>Rana esculenta</u> /South Moravia, Czechoslovakia	1982- 1984		Whole body	Mean iron content (ppm dry wt.):	Brno: Suburban gardens, fields, lakes.  Músov: large water reservoir.  Lednice: abandoned sand pit.	Pavel and Kucera 1986		
				Locality	Years				
					1982			1983	1984
			Brno	125.3 ± 7.7 (10)*	119.4 ± 13.6 (8)			-	
			Músov	132.8 ± 13.2 (12)	168.0 ± 25.1 (11)			197.8 ± 83.0 (10)	
Lednice	74.7 ± 6.2 (15)	136.6 ± 12.4 (7)	-						
					* number of samples.				

Table 4 - Residues (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES/LOCATION	DATE	N	TISSUE	RESIDUE LEVEL <sup>b</sup>	LEVELS USED/REMARKS	REFERENCE	
Lead (M)	- <u>Triturus vulgaris</u> <u>Rana esculenta</u> <u>R. temporaria</u> <u>Pelobates fuscus</u> <u>Bufo viridis</u> /Poland	June 1975	8	Whole body	Location	Residues (range in brackets)	Some evidence of food chain accumulation.	Dmowski and Kerolewski, 1979
					Protected zone near a zinc mill	124.8 (46.7-202.9)		
					Pine forest near a zinc mill	461.0		
					Control area	0.0 (0.0-0.0)		
		- <u>Xenopus laevis</u> /lab		3 groups of 6 toads each	Bone Skin Muscle Kidney Liver	Group A    Group B    Group C	Group A - intake of 11.99 ± 1.11 µg/day/toad avail. Pb for 4 wks; 13.63 ± 2.57 avail. Pb for 4 wks. Group B - intake of 12.89 ± 1.19 µg/day/toad for 4 wks.	Ireland, 1977
		- Highway drainage ditches in Maryland and Virginia	July-Dec. 1982		Tadpoles  Tadpoles	Geometric mean (ppm dry wt.)    95% Confidence Intervals	Group C - intake of 0.78 ± 0.10 µg/day/toad for 4 wks; 1.13 ± 0.14 for 4 wks.  Lead concentrations positively correlated with average daily traffic volumes. Concentrations in sedi- ment were usually higher (4-5 times) than those in tadpoles.	Birdsall et al., 1986
		<u>Rana catesbeiana</u>  <u>R. clamitans</u>				14                      2.5-72  14                      7.3-25		



Table 4 - Residues (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES/LOCATION	DATE	N	TISSUE	RESIDUE LEVEL <sup>b</sup>	LEVELS USED/REMARKS	REFERENCE
Lead (cont'd)	- <u>Rana perezi</u> /Donana Natl. Park, Spain		10 Composite samples	"A fillet with bones, skin"	Geometric mean: 0.83.		Hernandez et al., 1987
	- <u>Rana perezi</u> /Donana Natl. Park, Spain	1986 1984 1985	2 7 4	Muscle	1.19 0.48 1.82		Rico et al., 1987
	- <u>Plethodon cinereus</u> , <u>P. glutinosus</u> , <u>Bufo americanus</u> , <u>B. woodhousei</u> , <u>Rana sylvatica</u> / oak forest 10 km upwind from 2 zinc smelters in eastern Pennsylvania	June-Oct., 1979	23	Whole body	Mean ± SE (ppm dry weight) for all species: 12 ± 1.3.		Beyer et al., 1985
	- Unspecified species/ New Lead Belt, southeastern Missouri	Sept. 1972	8 pools 1 pool	Tadpoles	Area Residues (ppm dry wt.) Strother Creek Control 36-1590 28	Dam across Strother Creek provides the principal tailings pond for mine and mill effluents. Samples taken from below dam.	Cole et al., 1973
- Unspecified species/ New Lead Belt, southeastern Missouri		1 pool 2 pools 2 pools 1 pool	Tadpoles: Liver & Heart Eviscerated Intestine Whole Body	22 ppm. 93-213 ppm. 4419-7329 ppm. 4139 ppm.	Intestine included contents.	Jennett et al., 1979	

Table 4 - Residues (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES/LOCATION	DATE	N	TISSUE	RESIDUE LEVEL <sup>b</sup>	LEVELS USED/REMARKS	REFERENCE
Lead (cont'd)	- Patuxent Wildlife Research Centre <u>Rana catesbeiana</u> <u>R. clamitans</u> <u>Rana</u> spp. <u>Bufo</u> spp.		5 pools 2 pools 11 3 pools 1 pool 2	Tadpoles Tadpoles Whole body Carcaass Kidney Liver	2.5-5.2 1.4-1.5 0.88-3.2 21 4.9 2.1-2.6		Hall and Mulhern, 1984
	- Watershed in east-central Illinois <u>Acris crepitans</u> <u>Bufo americanus</u>		14 31	Whole body	Mean residues (ppm dry wt.): 2.7 3.0-3.5	Collected from areas 5 and 50 m from interstate highway.	Rolfe et al., 1977
	- <u>Rana pipiens</u> <u>R. sylvatica</u> / sparsely inhabited areas of Vermont			Liver Kidney	<u>R. pipiens</u> 1.0 - 6.2 <u>R. sylvatica</u> 1.3 - 8.2 14.8 - 31.3 10.2		Schroeder and Ipton, 1968
	- <u>Rana catesbeiana</u> / lead mining district, southeastern Missouri	1981-82	14-15	Whole body	Residues (geometric mean, range in parentheses): Site A 0.97 (.11-6.10) Site B 13.5* (3.50-31.0) Site C 14.0* (2.90-41.0) * significantly higher than Site A.	Site A - upstream from lead belt, Sites B and C - downstream from tailings ponds.	Niethammer et al., 1985
	- <u>Isonopus lewisii</u> /Ieb		12	Dorsal skin	Residues (ppm dry wt.) in frogs pretreated for 2 months on either dark or light backgrounds: Dark-adapted: 187.9 ± 13 Light-adapted: 114.8 ± 17.6	Subjected to 50 ppm lead nitrate for 21 days, medium changed daily. Pb concentrated within melanosomes of melanophores.	Ireland et al., 1979

Table 4 - Residues (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES/LOCATION	DATE	N	TISSUE	RESIDUE LEVEL <sup>b</sup>	LEVELS USED/REMARKS	REFERENCE	
Lead (cont'd)	- <u>Rana esculenta</u> / Rivier Toca, Italy	April- Sept. 1974	150	Tadpoles	Residues (ppm dry wt): 0.58-1.47.	Standard deviations in brackets.	Baudo, 1976	
					Neoterocephalod Frogs			Adults
			Neoterocephalod Frogs = 43; males = 10-22; females = 10-28	Liver Heart & Lungs Kidneys Muscles Skin Gonads Eggs	0.94 (0.65) 1.42 (0.41) 0.94 (0.68) 1.70 (1.40) 0.96 (0.47) - -	1.16 (0.48) 0.40 (0.33) 2.34 (2.21) 0.68 (0.52) 3.66 (4.00) 0.56 (0.29) -	7.56 (10.57) 2.73 (2.83) 4.64 (3.37) 3.69 (5.49) 3.18 (2.39) 4.16 (4.96) 0.89 (0.33)	
Lindane (OC)	- <u>Rana pipiens</u> /Ich		6	Ventral abdominal skin	Frogs were exposed to vaporized lindane for 18, 23 or 38 days	Frogs had higher residues than chickens, rats, pigs, fish or plants. Failure to eat may have lowered their metabolism and thus lowered their excretion of lindane.	Whitacre and Ware, 1967	
					Days following exposure			18
					Brain residues (ppm)	3.7	4.3	55.5
					Liver residues (ppm)	6.5	26.0	53.0
					Fat residues (ppm)	10.0	7.5	None
					Decreasing rate of accumulation by skin vs. time.			Pithed frogs were sus- pended in solution of 10 ppm.
								Kaiser and Dunham, 1972-73

Table 4 - Residues (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES/LOCATION	DATE	N	TISSUE	RESIDUE LEVEL <sup>b</sup>	LEVELS USED/REMARKS	REFERENCE			
Magnesium (M)	- Patuxent Wildlife Research Centre <u>Rana catesbeiana</u> <u>R. clamitans</u>		5 pools 2 pools	Tadpoles	58-160 14-29	Collected from uncon- taminated areas.	Hall and Mulhern, 1984			
	- <u>Rana temporaria</u> / Northern Finland		106	Liver Male Female	Mean ± SE (ppm dry wt): 294.5 ± 30.7 (wintering) -604.2 ± 49.0 (feeding) 335.0 ± 69.1 (pre-emerging) -749.0 ± 43.8 (feeding).	Wintering: Nov. - March Feeding: June - August Pre-emerging: April.	Pasanen and Koskela, 1974			
Manganese (M)	- <u>Rana esculenta</u> /South Moravia, Czechoslovakia	1982- 1984		Whole body	Mean Manganese content (ppm dry wt):	Brno: Suburban gardens, fields, lakes. Muzov: large water reservoir. Lednice: abandoned sand pit.	Pavel and Kucera, 1986			
					Locality			Years		
								1982	1983	1984
					Brno	24.1 ± 2.8 (10)*	25.8 ± 5.9 (8)	-		
					Muzov	24.7 ± 2.0 (12)	35.9 ± 4.2 (12)	51.9 ± 6.3 (10)		
					Lednice	15.6 ± 1.4 (13)	23.4 ± 4.2 (8)	-		
					* Number of samples.					

Table 4 - Residues (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES/LOCATION	DATE	N	TISSUE	RESIDUE LEVEL <sup>b</sup>	LEVELS USED/REMARKS	REFERENCE																										
Manganese (M) (cont'd)	- <u>Rana esculenta</u> / River Tace, Italy	April- Sept. 1974	150	Tadpoles	Residues (ppm dry wt): 23-958.	Standard deviations in brackets.	Beudo, 1976																										
					<table border="1"> <tr> <th rowspan="2">Necemorphosed Frogs</th> <th colspan="2">Adults</th> </tr> <tr> <th>males</th> <th>females</th> </tr> <tr> <td>15.4 (4.2)</td> <td>20.2 (4.5)</td> <td>22.1 (7.8)</td> </tr> <tr> <td>8.0 (3.3)</td> <td>8.4 (2.0)</td> <td>6.7 (3.7)</td> </tr> <tr> <td>8.4 (6.8)</td> <td>21.3 (14.0)</td> <td>5.1 (3.9)</td> </tr> <tr> <td>5.3 (1.8)</td> <td>4.5 (2.2)</td> <td>5.2 (2.2)</td> </tr> <tr> <td>14.6 (8.8)</td> <td>67.9 (15.8)</td> <td>30.3 (8.2)</td> </tr> <tr> <td>-</td> <td>5.8 (4.1)</td> <td>53.0 (23.1)</td> </tr> <tr> <td>-</td> <td>-</td> <td>8.1 (1.5)</td> </tr> </table>			Necemorphosed Frogs	Adults		males	females	15.4 (4.2)	20.2 (4.5)	22.1 (7.8)	8.0 (3.3)	8.4 (2.0)	6.7 (3.7)	8.4 (6.8)	21.3 (14.0)	5.1 (3.9)	5.3 (1.8)	4.5 (2.2)	5.2 (2.2)	14.6 (8.8)	67.9 (15.8)	30.3 (8.2)	-	5.8 (4.1)	53.0 (23.1)	-	-	8.1 (1.5)
	Necemorphosed Frogs	Adults																															
males		females																															
15.4 (4.2)	20.2 (4.5)	22.1 (7.8)																															
8.0 (3.3)	8.4 (2.0)	6.7 (3.7)																															
8.4 (6.8)	21.3 (14.0)	5.1 (3.9)																															
5.3 (1.8)	4.5 (2.2)	5.2 (2.2)																															
14.6 (8.8)	67.9 (15.8)	30.3 (8.2)																															
-	5.8 (4.1)	53.0 (23.1)																															
-	-	8.1 (1.5)																															
	- Unspecified species/ New Lead Belt, southeastern Missouri	Sept. 1972	8 pools 1 pool	Tadpoles	<table border="1"> <tr> <th>Area</th> <th>Residues (ppm dry wt.)</th> </tr> <tr> <td>Strother Creek Control</td> <td>500-5650 710</td> </tr> </table>	Area	Residues (ppm dry wt.)	Strother Creek Control	500-5650 710	Dam across Strother Creek provides the principal tailings pond for mine and mill effluents. Samples taken from below dam.	Gale et al., 1973																						
Area	Residues (ppm dry wt.)																																
Strother Creek Control	500-5650 710																																
	- Patuxent Wildlife Research Center <u>Rana catesbeiana</u> <u>R. clamitans</u>		5 pools 2 pools	Tadpoles	14-42 1.1		Hell and Mulhern, 1984																										

Table 4 - Residues (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES/LOCATION	DATE	N	TISSUE	RESIDUE LEVEL <sup>b</sup>	LEVELS USED/REMARKS	REFERENCE		
Mercury (M)	- Lake St. Clair, Ontario	1970	1 1 1 1		Sex	Carcass	Liver	Dustman et al., 1972	
					F	0.18	0.61		
					M	<0.10	1.1		
					M	<0.10	0.51		
		<u>R. pipiens</u>							
		<u>R. catesbeiana</u>							
		- <u>Rana tigrina</u> /lab			Liver, Kidney	203Hg accumulated in both high and low MW fractions of kidney 2,4,7 days after administra- tion. In liver, 203 Hg appeared in high MW fractions at 2 days, in both high and low at 4,7 days.	Single i.p. dose of 10 uCi/100 g body weight given.	Mehra et al., 1980	
		- Poljane, Yugoslavia	1975			Hg	MHg (ppm fresh weight)	Sample collected from uncontaminated area.	Byrne et al., 1975
		<u>Bufo bufo</u> male (m)			Liver	1.51	0.37		
		<u>B. bufo</u> female (f)				0.96	0.27		
	<u>Rana dalmatina</u> (m)				0.67	0.83			
	<u>Bombina variegata</u> (m)				2.07	0.35			
	<u>R. arvalis</u> (f)				1.96	1.1			
	<u>B. bufo</u> (m)			Kidney	1.24	-			
	<u>B. bufo</u> (f)				0.60	0.12			
	<u>R. dalmatina</u> (m)				1.01	-			
	<u>B. variegata</u> (m)				0.93	0.08			
	<u>R. arvalis</u> (f)				1.63	0.40			



Table 4 - Residues (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES/LOCATION	DATE	N	TISSUE	RESIDUE LEVEL <sup>b</sup>	LEVELS USED/REMARKS	REFERENCE			
Mercury (cont'd)	Finland (cont'd)				Mercury content (ppm fresh weight) given as mean $\pm$ S.E.:	* Number of samples.	Tehivuo et al., 1984 (cont'd)			
				Tuusula	Porvoo			Helsinki	Hämeenkyrö	
	Liver			.12 $\pm$ .02 (4)*	-			-	-	
	Kidney			.08 $\pm$ .01 (3)	-			-	-	
		<u>Bufo bufo</u>			Liver	.06 $\pm$ .01 (4)	-	-		
		Kidney			.04 $\pm$ .01 (4)	-	-			
		Muscle								
		- <u>Rana</u> sp./Aiken, South Carolina	Aug. 1973 to Jan. 1974	N of analyses 3 3 1	Tadpoles	Total Mercury 2.08 4.36 6.41	Methyl Hg 0.01 0.03 0.00	% Methyl Hg 0.48 0.69 0.00	Low level mercuric ion concentrations of 1.0 and 5.0 ug/l maintained in streams.  Tadpoles eat algae which concentrate Hg.	Cox et al., 1975
		- <u>Rana perezi</u> /Donana Natl. Park, Spain		10 Composite samples	"A fillet with bones, skin"	Geometric mean: 0.20.			Hernandez et al., 1987	
	- <u>Rana perezi</u> /Donana Natl. Park, Spain	1984 1985 1986	7 4 2	Muscle	0.08 0.09 0.15			Rico et al., 1987		

Table 4 - Residues (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES/LOCATION	DATE	N	TISSUE	RESIDUE LEVEL <sup>b</sup>	LEVELS USED/REMARKS	REFERENCE		
Mercury (cont'd)	- Patuxent Wildlife Research Centre <u>Rana catesbeiana</u> <u>R. clamitans</u> <u>Rana</u> spp. <u>Bufo</u> spp.	1972-73	5 pools	Tadpoles	0.05-0.10	Savannah River contained Hg-contaminated fish at levels thought to be hazardous to human health.	Hall and Mulhern, 1984		
	2 pools		Tadpoles	0.04-0.10					
	11		Whole body	<0.01-0.14					
	4		Whole body	0.04-0.14					
	- Savannah River floodplains and tributaries, Georgia <u>Rana pipiens</u> <u>R. catesbeiana</u> <u>Hyla cinerea</u> <u>Bufo terrestris</u>			Adults	Skeletal Muscle 0.07-0.10 0.05-0.26 0.03 ND-0.18	Liver 0.25 0.09-0.44 - -			
Methoxychlor (OC)	- <u>Bufo americanus</u> /lab			Whole body	Days of exposure 1 6 1 1 6 36	Dosage Group (ppm) 0.024 in food " 0.325 in food " 0.069 $\pm$ 0.036 in water " " "	Residue geometric mean (95% confidence limits) 0.013 (0.003-0.043) 0.008 (0.002-0.024) 0.033 (0.012-0.088) 0.145 (0.065-0.323) 0.244 (0.124-0.482) 0.124 (0.048-0.327)	No changes in organ weights, feeding, behaviour or survival.  Residues not correlated with exposures.	Hall and Swineford, 1979

Table 4 - Residues (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES/LOCATION	DATE	N	TISSUE	RESIDUE LEVEL <sup>b</sup>	LEVELS USED/REMARKS	REFERENCE											
Mirex (OC)	- <u>Rana clematis</u> pond in mixed boreal forest, northeastern New Brunswick	June 1985		Tadpole Whole body	None of the samples of caged or wild tadpoles contained detectable levels (>10 ppb, fresh wt.).	Two aerial applications of 70 g a.i./ha on June 2, 8.	Sundaram et al., 1986											
	- <u>Acris gryllus</u> /1.2 ha fish pond, Mississippi	Treatment on Oct. 23, 1970	6 or more individuals in each pooled sample	Whole body	Mirex residues (ppb) at given intervals after treatment:	Application rate (aerial) 1.4 kg/ha.  Pond was part of a larger drainage system.	Collins et al., 1975											
					<table border="1"> <tr> <th>Pre-treatment</th> <th>5 days</th> <th>38 days</th> <th>6 months</th> <th>12 months</th> <th>16 months</th> </tr> <tr> <td>ND</td> <td>0.03</td> <td>2.26</td> <td>2.88</td> <td>0.02</td> <td>0.05</td> </tr> </table>	Pre-treatment	5 days	38 days	6 months	12 months	16 months	ND	0.03	2.26	2.88	0.02	0.05	
Pre-treatment	5 days	38 days	6 months	12 months	16 months													
ND	0.03	2.26	2.88	0.02	0.05													
- <u>Rana gryllus</u> Winston Co., Mississippi	Treatment in May and June, 1972	1	Tadpole	Residue in sample collected less than 6 months after application - 0.12.	Application rate (aerial) 1.4 kg/ha.	Naqvi and de la Cruz, 1975												
	- Orbow lakes in Louisiana <u>Rana catesbeiana</u> <u>R. sphenoccephala</u> <u>R. c. clematis</u>	1980	5 3 3		Not detected in all three species.	Lakes in fertile alluvial cropland. Subjected to heavy pesticide applications. Annual rainfall - 130 cm.	Niethammer et al., 1984											

Table 4 - Residues (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES/LOCATION	DATE	N	TISSUE	RESIDUE LEVEL <sup>b</sup>						LEVELS USED/REMARKS	REFERENCE	
					Application Rate (g/ha)	Pre-treatment	Residues (ppm)						
							Post treatment						
				7 days	1 mo	3 mos	6 mos	12 mos					
Mirex (cont'd)	- Southwest Georgia										Figures in brackets represent number of samples in the pool.  D= discarded cross-contaminated samples. ND= no residues detected at 0.01 ppm level.  Maximum levels detected 1-3 months after sprays gradually declined to lower levels over 1 year.	Wojcik et al., 1975	
	<u>Bufo terrestris</u>	Turner Co.	1971			2.10	D	D	0.94				
									(5)				
		North Co.	1971-72			1.12	ND (3)		0.24				0.02
									(5)				(2)
	<u>Gastrophysyne carolinensis</u>	Tift Co.	1971			1.12	D			0.47			
									(2)				
		Turner Co.	1971			2.10	D	D	2.02	0.41			
									(9)	(14)			
		North Co.	1971-72			1.12	ND (16)	0.12 (5)					0.17 (4)
													0.14 (1)
	<u>Pseudacris ornata</u>	North Co.	1971-72			1.12				0.10			(1)
<u>Acris gryllus</u>	North Co.	1971-72			1.12			9.27	0.14				
								(2)	(3)				
	North Co.	1971-72			1.12			3.01					
								(9)					
<u>B. quercus</u>	North Co.	1971-72			1.12					ND (5)			
										0.08(2)			



Table 4 - Residues (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES/LOCATION	DATE	N	TISSUE	RESIDUE LEVEL <sup>b</sup>	LEVELS USED/REMARKS	REFERENCE			
Organochlorine compounds (DDI, DDD, DDE, g-Hexachlorocyclohexane)	- <u>Rana ridibunda</u> / clean pond and rice field discharge canal, North Caucasus, U.S.S.R.		280	Mussio	Residues (ppm):	Residues in water - 0.00006 in pond, 0.00330 in rice field.	Zhukova, 1987			
								Pond	Rice Field	
					immature	immature				
					male	male				
					female	female				
Dyckhlorodans (OC)	- <u>Tarichia torosa</u> / British Columbia	1980	N/days post treatment		- residues:	Lake in B.C. sprayed with technical chlordane to a level of 0.010 ppm.	Albright et al., 1980			
					ND - not detectable					
					ND					
					ND					
					0.115					
					0.134					
					0.027					
					0.080					
					0.097					
					0.010					
4/14	Whole body	ND				No application rate given.				
								Liver	ND	
								Stomach		ND
								Whole body		
Liver	0.115									
Stomach		0.134								
Whole body			0.027							
Liver				0.080						
Stomach	0.097									
Whole body		0.010								
Liver			0.011							
Stomach				0.011						
Whole body	ND									
Liver		ND								
Stomach			ND							

Table 4 - Residues (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES/LOCATION	DATE	N	TISSUE	RESIDUE LEVEL <sup>b</sup>	LEVELS USED/REMARKS	REFERENCE		
Parathion (OP)	- <u>Acris crepitans</u> /lab		1 pool of 12 per treatment	Whole body	Group	Parathion Conc.	Frogs held individually for 96 hours in static systems.	Fleming et al., 1982	
					Control	40.05			
					0.1 ppm	<0.05			
					1.0 ppm	0.08			
					10 ppm	4.6			
	- <u>Rana pipiens</u> /lab			Skin	Half time rate of dermal penetration (mins.): 198.	1 mg/kg applied to 1 cm <sup>2</sup> area on back immediately behind head.	Shah et al., 1983		
	- <u>Rana catesbeiana</u> /lab			Tadpoles	Significant bioconcentration: average magnification - 64 times. Maximum level found in pooled samples was 96 ppm. No parathion (metabolite) detected in tissues at sensitivity limit of 0.5 ppm.	Exposed to .01-1 ppm for 96 hr in continuous flow apparatus.	Hall and Knino, 1980		
PCBs (polychlorinated biphenyls)	- Oxbow lakes in Louisiana	1980	5		Not detected in all three species.	Lakes in fertile alluvial cropland. Subjected to heavy pesticide applications. Annual rainfall - 130 cm.	Niethammer et al., 1984		
								- <u>Rana catesbeiana</u>	3
								- <u>R. sphenocéphala</u>	
	- <u>R. c. clamitans</u>								
	- <u>Rana sp.</u> /Louisiana		1978	Legs	.017 ppm (mean exceeded by standard error). Not detected (0.10 ppm detection limit).	Site was an area of low insecticide use.	Dowd et al., 1985		
			Whole body						
	- <u>Rana perezi</u> /Donana Natl. Park, Spain	10	Composite samples	"A fillet with bones, skin".	Geometric mean: 0.06.		Hernandez et al., 1987		
	- <u>Rana perezi</u> /Donana Natl. Park, Spain	7		Muscle	0.05		Rico et al., 1987		
		4			0.49				
		2			1.08				



Table 4 - Residues (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES/LOCATION	DATE	N	TISSUE	RESIDUE LEVEL <sup>b</sup>	LEVELS USED/REMARKS	REFERENCE																
PCBs (cont'd)	- <u>Necturus lewisi</u> / Neuse River drainage, North Carolina		50	Whole body	Geometric Mean of residues: 0.39.	Analyzed for PCB 1254.	Hell et al., 1985																
Permethrin (PY)	- <u>Rana pipiens</u> /lab			Skin	Half time rate of dermal penetration (mins): 420.	1 mg/kg applied to 1 cm <sup>2</sup> area on back immediately behind head.	Shah et al., 1983																
Phenols	- <u>Rana temporaria</u> /lab				Distribution of phenols (g/g) in various tissues at given times after injection via lymph sac at 5 mg/kg (PH = phenol, NP = 3-nitrophenol, DP = 3,5-dithylphenol, AP = 4-aminophenol):	Average weight of frogs used was 28 ± 8 g. temperature 20° C.	Nagel and Ulrich, 1981																
								0.5 h				2 h				4 h				15 h			
								Ph	NP	DP	AP	Ph	NP	DP	AP	Ph	NP	DP	AP	Ph	NP	DP	AP
Blood	11.6	8.9	16.5	11.3	5.8	6.4	13.3	10.5	2.1	5.6	15.9	7.1	1.3	0.8	2.7	4.4							
Gut	4.9	2.3	7.2	7.1	3.2	3.5	7.2	7.0	1.5	4.0	7.2	8.7	1.2	1.6	7.8	3.5							
Brain	3.7	3.9	9.4	3.9	3.0	1.5	3.5	3.5	1.1	1.4	1.9	3.5	0.5	0.1	0.5	2.2							
Skin	4.3	16.5	8.8	5.5	3.1	3.9	5.4	4.2	1.4	1.9	3.8	3.8	1.5	0.4	0.9	1.9							
Heart	5.2	6.3	9.6	6.7	3.4	3.0	5.9	3.9	1.2	2.4	4.6	3.2	0.8	0.3	1.1	1.5							
Liver	6.5	5.5	8.3	12.9	2.7	4.4	8.9	6.7	1.4	3.9	7.5	5.6	0.9	1.2	2.2	2.0							
Lung	5.4	7.1	10.7	10.2	2.6	4.2	7.9	5.6	1.7	3.3	5.7	6.0	0.9	0.7	1.9	3.8							
Stomach	4.8	2.7	8.3	6.4	2.3	3.3	7.1	4.8	1.3	2.8	5.3	4.3	0.5	0.4	1.0	1.5							
Spleen	8.0	2.4	9.7	6.9	4.7	4.6	10.3	5.2	1.5	3.4	7.2	4.9	1.1	0.4	2.1	2.5							
Kidney	8.9	16.0	17.9	10.7	12.4	15.1	13.6	6.3	7.8	18.4	9.2	5.8	2.4	8.1	2.9	3.1							
Pancreas	6.2	4.3	10.5	7.2	5.0	4.9	10.4	4.5	1.5	2.5	7.5	4.5	1.1	0.7	1.7	2.0							
Remainder	2.9	4.2	4.6	4.0	1.8	2.9	4.3	3.5	1.1	2.0	3.1	3.6	0.6	0.3	0.8	1.1							
Gall bladder	6.1	4.8	14.3	15.5	2.4	11.9	113	29.4	7.3	73.8	287	69.8	10.0	60.4	126	54.0							
Contents of guta	1.8	8.3	3.8	3.6	3.4	0.9	8.8	9.7	1.4	12.7	7.4	20.9	7.7	10.4	84.3	30.0							

Table 4 - Residues (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES/LOCATION	DATE	N	TISSUE	RESIDUE LEVEL <sup>b</sup>	LEVELS USED/REMARKS	REFERENCE
Selenium (M)	- <u>Lenopus laevis</u> /lab			Embryos at gastrula stage	Uptake rapid and linear until 36 h of exposure. Linear decrease in Se over first 24 h of clearing; rate of depuration similar to rate of uptake.	Gastrulas placed in 5 ppm <sup>75</sup> Se for up to 48 h. After 24 h, some transferred to untreated water.	Browne and Dumont, 1979
				Tadpoles	80% decrease in Se content within 30 min; Se level remained stable for next 3 days.	Embryos exposed for 48 h; hatched tadpoles transferred to untreated water.	
Strontium (M)	- <u>Lariche granulosa</u> /lab			Whole body	Biological half-lives - 136 and 151 days for chronic and acute treatment, respectively. Significant concentrations found in blood, liver, skin, muscle and bone; after 25 days, most <sup>85</sup> Sr in bone.	Retention of <sup>85</sup> Sr following chronic and acute (single i.p. injection) administration at 10°C.	Willie and Yellett, 1971
	- <u>Rana pipiens</u>			Whole body	Biological half-life of 222 days.	Retention of <sup>85</sup> Sr following i.p. injection at 10°C.	
	- <u>Lariche granulosa</u> /lab				Slower loss component had biological half-lives of 130 and 80 days at 10° and 20°C, respectively.	Retention of <sup>85</sup> Sr following chronic or acute treatment at 10° or 20°C.	Willie et al., 1976
	- <u>Rana pipiens</u>				Half-lives longer than those found for newts at the 2 temperatures.	Retention following i.p. injection.	
	- <u>Rana temporaria</u> /lab		100 per tests	Tadpoles 5-7 weeks old	100 tadpoles absorbed approximately 18% (0.02 uc) of the strontium-90 and about 72% of the yttrium-90 in 175 hr.	Treated with 0.125 uc strontium-90 in 500 ml water, giving 2.5 x 10 <sup>5</sup> disintegrations/min strontium-90 and yttrium-90.	Lucas and Pickering, 1958

Table 4 - Residues (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES/LOCATION	DATE	N	TISSUE	RESIDUE LEVEL <sup>b</sup>	LEVELS USED/REMARKS	REFERENCE				
2,3,7,8-TCDD (2,3,7,8-tetrachlorodibenzo-p-dioxin)	- Toad Sp./Italy	1980	1	Whole body	0.0002 ppm found in toad 2 yrs. after exposure.	Explosion of a chemical plant released 2,3,7,8-TCDD.	Fornelli et al., 1980				
	- <u>Rana catesbeiana</u> / Central Arkansas	1984-85	10	Liver	Levels in females (n=7) ranged from 1.2 - 48.0 ppb, wet wt. Levels in males (n=3) ranged from 0.64 - 11.0 ppb, wet wt.	Results suggest frogs tolerate substantial body burden of 2,3,7,8-TCDD.	Korfmacher et al., 1986b				
	- <u>Rana catesbeiana</u> / Central Arkansas	1984-85	6		Residues (ppt ± 95% confidence limits):		Korfmacher et al., 1986a				
				Sex	F		F	F	H	H	
				Muscle	218 ± 60		277 ± 60	87 ± 26	103 ± 17	637 ± 202	566 ± 73
				Skin	364 ± 66		623 ± 105	217 ± 36	637 ± 100	1710 ± 854	-
				Oviduct	988 ± 488		496 ± 57	148 ± 22	2050 ± 197	-	-
				Liver	2390 ± 279		3680 ± 564	1250 ± 174	1260 ± 122	1130 ± 3280	9800 ± 983
				Ovaries	7460 ± 1440		10400 ± 5370	2830 ± 998	2700 ± 340	-	-
				Fat	-		-	-	68000 ± 7120	> 14000	-

Table 4 - Residues (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES/LOCATION	DATE	N	TISSUE	RESIDUE LEVEL <sup>b</sup>	LEVELS USED/REMARKS	REFERENCE			
2,3,7,8-TCDD (cont'd)	- Southern toad/ 3.0 km <sup>2</sup> area on Elgin Air Force Base, Florida	1973-79		Whole body	Residue (ppt): 1,360.	Approx. 2.8 kg applied as contaminant in aerial spraying of 2,4,5-T.	Young and Cockerham, 1982			
TDE (DC)	- Oxbow lakes in Louisiana <u>Rana catesbeiana</u> <u>R. sphenoccephala</u> <u>R. c. clamitans</u>	1980	5		Not detected in all three species.	Lakes in fertile alluvial cropland. Subjected to heavy pesticide applications. Annual rainfall - 130 cm.	Niethammer et al., 1984			
			3							
toxaphene (DC)	- Oxbow lakes in Louisiana	1980			Lake Providence	Lakes in fertile alluvial cropland. Subjected to heavy pesticide applications. Annual rainfall - 130 cm.	Niethammer et al., 1984			
					Lake Bruin					
					Residues (geometric mean)			% lipid	Residues (geometric mean)	% lipid
					<u>Rana catesbeiana</u>			0.09	1.7	ND
		<u>R. sphenoccephala</u>	0.21	3.3	ND	1.1				
		<u>R. c. clamitans</u>	ND*	1.1	ND	0.6				
					* Not detected.					

Table 4 - Residues (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES/LOCATION	DATE	N	TISSUE	RESIDUE LEVEL <sup>b</sup>			LEVELS USED/REMARKS	REFERENCE	
					Location	Residues (ppm dry weight) range in brackets				
Zinc (M)	- <u><i>Triturus vulgaris</i></u> <u><i>Rana esculenta</i></u> <u><i>R. temporaria</i></u> <u><i>Pelobates fuscus</i></u> <u><i>Bufo viridis</i></u> /Poland	June 1975	8 1 11	Whole body	Protected zone near a zinc mill Pine forest near a zinc mill Control area	180.8 (156.0-205.6) 533.5 202.2 (103.7-301.4)		Some evidence of food chain accumulation.	Dzowski and Karolewski, 1979	
						Geometric mean: 18.80.				
						Mean zinc content (ppm dry wt.):				
	- <u><i>Rana perezi</i></u> /Donana Natl. Park, Spain	1984 1985 1986	7 4 2	Muscle	9.46 7.65 31.07	Years		Brno: suburban gardens, fields, lakes.	Hernandez et al., 1987	
						Locality				
	- <u><i>Rana perezi</i></u> /Donana Natl. Park, Spain	1982-84	7 4 2	Muscle	9.46 7.65 31.07	Years		Muscov: large water reservoir.	Pavel and Kucera, 1986	
						Locality				
						Brno	181.7 ± 12.8 (10)*	157.4 ± 11.8 (8)		
						Muscov	151.8 ± 6.0 (12)	173.0 ± 8.1 (12)	142.9 ± 9.1 (10)	
						Lednice	128.1 ± 9.1 (13)	177.8 ± 10.4 (8)		Lednice: abandoned sand pit.
					* Number of samples.					

Table 4 - Residues (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES/LOCATION	DATE	N	TISSUE	RESIDUE LEVEL <sup>b</sup>			LEVELS USED/REMARKS	REFERENCE
					Concentrations of Zn (ug/mg dry wt.):				
Zinc (cont'd)	- <u><i>Triturus cristatus</i></u> /lab		10 exptl. 20 ctrl.; 8 samples analyzed/organ	Anterior brain Posterior brain Liver Intestine Kidney Pancreas Skin Chloanes				Newts left in tank with zinc-plated bottom. Zinc concentration in water of control tank - .20 to .26 ug/L; in water of experimental tank approx. .3 to 2.8 ug/L. Zinc-rich, unusual cells present in primordium hippocampi of poisoned newts.	Taben et al., 1982
					Controls				
					Poisoned				
					.1531 .0505				
					.0579 .1619				
					.4324 .1012				
					.0714 .2381				
					.1135 .0454				
					.0793 .0952				
					.4646 .1190				
.2500 .3710									
.2920 .1523									
.2469 .3608									
3.3918 .7059									
3.4184 .9231									
.6579 .6818									
.8294 1.5975									
5.1282 .2941									
1.0938 1.8142									
.3254 .0785									
.1902 .1950									
.5442 .1020									
.3000 .2848									
.3553 .0829									
.1161 .2389									
.5373 .1681									
.4762 .4564									
3.1405 .2647									
.5072 .6742									
2.6961 3.5593									
3.2368 1.9192									
1.5714 .3968									
.7042 1.2575									
2.1535 .6872									
.9067 1.1644									
					Equilibrium body burden attained by 30 days. Two distinct loss components evident - slower component had half-life of 3 1/3 years and accounted for over 90% of initial body burden. Half-life following injection at room temp. was 1 1/2 years. Highest concentrations in skin, muscle, blood, liver.			Uptake of <sup>65</sup> Zn from water and loss from body after transfer to clean water at 10°C studied. Zn excretion was directly related to ambient temp.	Willis and Valett, 1978
					Whole body				

Table 4 - Residues (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES/LOCATION	DATE	N	TISSUE	RESIDUE LEVEL <sup>b</sup>		LEVELS USED/REMARKS	REFERENCE
					Area	Residues (ppm dry wt.)		
Zinc (cont'd)	- Unspecified species/ New Lead Belt, southeastern Missouri	Sept. 1982	6 pools 1 pool	Tadpoles	Area	Residues (ppm dry wt.)	Oen across Strathair Creek provides the principal tailings pond for zinc and mill effluents. Samples taken from below dam.	Gale et al., 1973
					Strathair Creek Control	160-265 62		
	- Unspecified species/ New Lead Belt, southeastern Missouri		1 pool 2 pools 1 pool 2 pools	Tadpoles: Whole body Eviscerated Liver & Heart Intestine			Intestine included contents.	Jennett et al., 1979
					2808 ppm. 240-236 ppm. 67 ppm. 6926-4696 ppm.			
- Potomac Wildlife Research Center <u>Rana catesbeiana</u> <u>R. clamitans</u> <u>Bufo</u> spp.			5 pools 2 pools 4 pool of 10 pool of 10 11	Tadpoles	9.7-15 3.7-6.0 25-90 84		Hall and Mihlen, 1984	
				Tadpoles	39			
				Whole body	6.2-31			
- <u>Rana temporaria</u> / Northern Finland		1971-1972	106	Liver male female	Mean ± SE (ppm dry wt.): 45.0 ± 3.0 (pre-emerging) - 90.9 ± 7.1 (feeding) 61.0 ± 5.1 (pre-emerging) - 88.8 ± 2.9 (spawning)		Pre-emerging: April; feeding: June - August; spawning: May. Pasanen and Koskela, 1974	

Table 4 - Residues (cont'd)

CONTAMINANT <sup>a</sup>	SPECIES/LOCATION	DATE	N	TISSUE	RESIDUE LEVEL <sup>b</sup>			LEVELS USED/REMARKS	REFERENCE
					Control	Zn			
Zinc (cont'd)	- <u>Xenopus laevis</u> /lab		10 per dose	Liver Kidney	Concentrations (mean ± SD):			5 mg Zn/kg body wt. injected i.m. daily for 5 days; frogs killed 1 day after last injection.	Suzuki et al., 1983
					Control	Zn			
						15.0 ± 1.5 17.3 ± 1.5	35.8 ± 5.5* 53.6 ± 6.3*		
					* Significantly accumulated.				
	- <u>Plethodon cinereus</u> , <u>P. glutinosus</u> , <u>Bufo americanus</u> , <u>B. woodhousei</u> , <u>Rana sylvatica</u> / oak forest 10 km upriver from 2 zinc smelters in eastern Pennsylvania	June - Oct., 1979	23	Whole body	Mean ± SE (ppm dry weight) for all species: 88 ± 8.7.				Bayer et al., 1985
	- <u>Rana catesbeiana</u> / lead smelting district, southeastern Missouri	1981-1982	15	Whole body	Residues (geometric mean, range in parentheses):			Site A - upstream from lead belt, Sites B & C - downstream from tailings ponds.	Niethammer et al., 1985
					Site A	Site B	Site C		
					20.9 (15.9-28.4)	42.7* (29.8-64.5)	29.9* (20.3-37.5)		
					* Significantly higher than site A.				

<sup>a</sup> OC = organochlorine insecticide, OP = organophosphate insecticide, C = carbamate insecticide, PY = pyrethroid, H = herbicide, M = metal.

<sup>b</sup> Residues given in ppm wet weight unless otherwise indicated.