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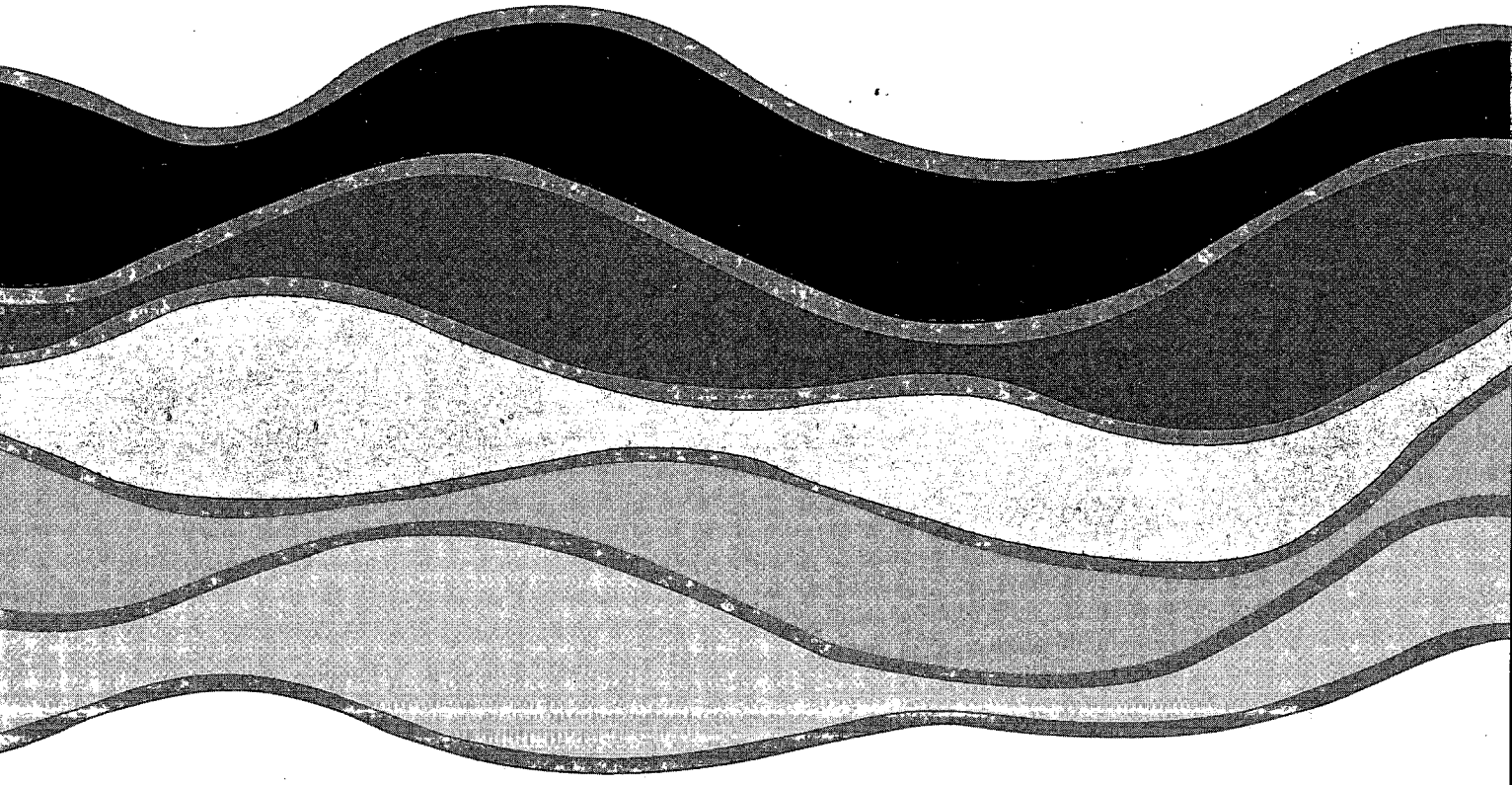

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NWRI Contribution No. 91-70A

DEGRADATION OF CHLOROFLUOROCARBON-113 UNDER ANAEROBIC CONDITIONS

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MANAGEMENT PERSPECTIVE

Chlorofluorocarbon-113 (CFC-113; 1,1,2-trichloro-1,2,2-trifluoroethane)is a solvent which is used as a degreasing agent in the electronic industry and as a refrigerant in commercial and industrial air conditioning. It is denser than water and thus can migrate downwards through soils and below the water table where it can form pools of dense non-aqueous phase liquids (DNAPLs) which contaminate groundwater. This study was done to further elucidate the discovery of metabolites of CFC-113 in groundwater at the Gloucester landfill near Ottawa. One of these compounds, CTFE (chlorotrifluoroethene; CFC-1113), is a known toxic compound. Laboratory studies were conducted to measure the rate of degradation of the various metabolites of CFC-113 under the anaerobic conditions which prevail in the landfill. CFC-113 was found to be degraded rapidly in landfill leachate (half-life of 5.3 days). The major product of transformation was HCFC-123a, the toxicity of which is not known. CTFE was formed in the absence of microbes, in water containing only buffers. It was in turn found to be unstable in water containing sulfide ions, such as is the case in the Gloucester landfill, but degraded only slowly in leachate from another source. Groundwater contamination by CFC-113 is therefore of concern, because this compound is decomposed in groundwater to compounds which are more toxic than the starting material. This study was done within the groundwater contamination project which is mandated to conduct research on the fate of organic contaminants in groundwater.

SOMMAIRE À L'INTENTION DE LA DIRECTION

Le chlorofluorocarbure-113 (1,1,2-trichloro-1,2,2-trifluoroéthane) est un solvant utilisé comme agent dégraissant dans l'industrie electronique et comme réfrigérant pour la climatisation commerciale et industrielle. Il est plus dense que l'eau et peut donc migrer vers le bas à travers le sol et s'introduire dans la nappe phréatique où il peut former des flaques de liquide dense non-aqueux qui peuvent contaminer l'eau souterraine. Cette étude a été effectuée pour élucider la découverte de produits du métabolisme du CFC-113 dans l'eau contaminée du site d'enfouissement de Gloucester près d'Ottawa. Un de ces composés, la chlorotrifluoroéthène (CTFE, CFC-1113) est un produit toxique connu. Des études en laboratoire on été effectuées pour mesurer la vitesse de décomposition des divers produits du métabolisme du CFC-113 dans les conditions anaérobies qui prévalent dans le site d'enfouissement. Le CFC-113 se décompose rapidement dans le lexiviat (demi-vie de 5.3 jours). Le produit principal de la transformation est le HCFC-123a dont la toxicité est inconnue. La CTFE est formée en absence de microorganismes, dans l'eau contenant seulement des sels tampons. La CTFE est instable dans l'eau contenant des ions sulfures, comme l'eau du site de Gloucester, mais est décomposée lentement dans le lexiviat d'une autre source. La contamination des eaux souterraines par le CFC-113 est donc problématique parce que ce produit peut se décomposer en des produits plus toxiques que lui. Cette étude a été effectuée dans le cadre du project de la contamination des eaux souterraines qui a pour mandat de faire de la recherche sur le devenir des contaminants organiques dans les eaux souterraines.

ABSTRACT

A series of microcosms were set up to verify the degradation pathway of CFC-113 in water under anaerobic conditions and to measure its half-life and that of its major degradation products HCFC-123a and chlorotrifluoroethene (CTFE). Anaerobic landfill leachate was used as a source of bacteria acclimated to chlorinated solvents. The rate of reaction in methanogenic leachate was also compared to that obtained in a buffer containing reduced hematin. In methanogenic landfill leachate, CFC-113 was transformed to HCFC-123a with a half-life of 5 days at 20°C. The same reaction occurred in sodium sulfide/cysteine buffers containing hematin, but at a much slower rate. The production of CTFE was independent of the presence of HCFC-123a and occurred abiotically. Under methanogenic conditions, HCFC-123a was found to be further dechlorinated to HCFC-133 and HCFC-133b. CTFE was found to be relatively stable in methanogenic landfill leachate but was decomposed rapidly in a buffer containing sulfide.

RÉSUMÉ

Suite à la découverte des produits HCFC-123a et chlorotrifluoroethène (CTFE) dans des eaux souterraines anoxiques contaminées par le CFC-113, une chaîne métabolique reliant ces produits par une suite de déchloration réductrices et d'éliminations, avait été proposée. Une série de microcosmes furent construit afin de vérifier cette hypothèse et de mesurer les demi-vies des produits. Du lexiviat anaérobie de site d'enfouissement a été utilisé comme source de bactéries acclimatées aux solvants chlorés. La vitesse de réaction a aussi été comparée à celle obtenues dans des solutions tampons contenant de l'hématine réduite. Dans le lexiviat, le CFC-113 a été transformé au HCFC-123a dans une demi-vie de 5.3 jours à 21°C. La réaction était plus lente dans l'hématine et la vitesse de réaction était dépendante de la concentration de l'hématine. On a trouvé que la production de la CTFE était indépendante de la présence du HCFC-123a et se produisait abiotiquement. Dans un milieu méthanogène, le HCFC-123a est déchlorée pour produire le HCFC-133 et le HCFC-133b. La CTFE est relativement stable dans le lexiviat d'eau souterraine, mais est hydrolysée rapidement dans une solution tampon contenant des ions sulfures.

INTRODUCTION

The presence of CFC-113 (1,1,2-trichloro-1,2,2-trifluoroethane) in groundwater has become of concern since this chemical has largely replaced trichloroethylene as a degreasing solvent. It is also used as a refrigerant in industrial and commercial air conditioning. It has a density of 1.56 g/cm^3 (1) and thus can rapidly penetrate through soil and water and form pools on top of low permeability layers in aquifers. It has been found in many groundwater investigations in California, where it is used extensively as a degreasing agent by the electronic industry in the manufacture of semi-conductor chips (2).

A possible pathway for the reductive dechlorination of CFC-113 in anaerobic groundwater (Figure 1) was postulated by Lesage et al.(3) on the basis of other fluorocarbons found in anaerobic groundwater samples from the Gloucester landfill (Ottawa, Ontario). Since then, other field evidence of the degradation of CFC-113 has been presented (4,5,6). Lesage et al.(3) had drawn a parallel to the well documented reductive dechlorination of chlorinated hydrocarbons under anaerobic conditions (7). In addition, the cytochrome P-450 system was postulated as the enzyme responsible for this transformation, as is the case for many chlorinated hydrocarbons (8,9). The cytochrome P-450 enzyme system is one of the enzymes known to be capable of catalysing the reductive dechlorination reaction in mammalian as well as microbial systems. Reductive dehalogenation of 1,1,1-trichloro-2,2,2-trifluoroethane (CFC-113a) by cytochrome P-450 was demonstrated in liver microsomes (10) and there is every reason to believe that CFC-113, its isomer, would be transformed similarly.This reaction has been observed for highly chlorinated methanes and ethanes, and the rate of the reaction generally decreases with the number of halogens on the molecule (11). In methanogenic bacteria, corrinoids and coenzyme F_{430} may be involved as catalysts (12). Similarly, reduced corrinoids have been found to reductively dechlorinate tetrachloromethane and CFC-11, -12 and -13 (13). The dehalogenation seems to be common to metallated porphyrins and corrins, although the rate of reaction may vary, as was found with lindane (14).

Early studies had shown that CFC-113 was relatively non-toxic to humans, and that most of the exposure was through inhalation (15). A more recent report concluded that less than 5% of the inhalation dose was recovered unchanged and that CFC-113 could be metabolized (16). Very few published studies

concern its presence in water. It was the finding of metabolites in landfill leachate which prompted this study. Indeed, CTFE (chlorotrifluoroethene), which was found to be formed from CFC-113 in anaerobic landfill leachate, is much more toxic than its parent. It has an inhalation LC_{50} of 1000 ppm (17) and an oral LD_{50} of 268 mg/kg (18). At sublethal levels, it causes kidney dysfunction (17).

This paper presents the results of a controlled microcosm study aimed at determining the rate of degradation of CFC-113 under anaerobic conditions. Municipal landfill leachate was used as a source of an anaerobic consortium of bacteria. Since it contained other chlorinated hydrocarbons, a portion of the leachate was purged with N_2 to find out whether or not the presence of other chlorinated organic compounds was necessary for the cometabolism to occur, or if their presence was, on the contrary, inhibitory (19). No sterilized leachate was used because residual activity has often been observed which such controls, where it is impossible to tell whether this activity is from incomplete sterilization, extracellular or lysed cell enzyme activity, abiotic transformations (20), or sorption (21).

Hematin was used as a model of the porphyrin system present in cytochrome P-450, which catalyses the electron transfer reactions in the reductive dechlorination reaction of chlorinated ethanes and ethenes (22). The mechanism of the reaction, as postulated by Castro et al. (23), involves a 3-step mechanism where the reduced iron porphyrin first reacts with the chlorinated alkane to produce a complex, the carbon-halogen bond is then cleaved producing an alkyl radical, which then is scavenged by the heme. Hydrolysis of this alkyl-heme produces the final reduced species. The reduced porphyrin provides an electron in each reductive step, the net result being a two electron transfer. The thus oxidized iron porphyrin is quickly reduced back in the presence of reducing agents such as sulfide and cysteine. The experiment was conducted at two different concentrations of hematin to verify its enzymatic role. In enzyme-mediated reactions, the rate of the reaction is directly proportional to the concentration of the enzyme substrate complex (24). The hematin concentrations were thus chosen such as to bracket the initial substrate concentration.

Two buffer systems were used as controls, a simple phosphate buffer at pH 7, to measure possible hydrolysis and other losses, and a reducing buffer containing sodium sulfide and cysteine. The reducing buffer was used to reproduce sulfate reducing conditions present in the Gloucester landfill where the degradation had been observed (3), and as a control for the hematin-containing microcosms, since they also contained phosphate, sodium sulfide and cysteine. Its purpose was to verify whether the observed

transformation can occur abiotically under the redox conditions prevalent in anaerobic groundwater or whether enzymatic catalysis is essential.

Similar experiments were also conducted with the observed metabolites HCFC-123a (1,2-dichloro-1,2,2-trifluoroethane) and CTFE in order to assess their stability and fate, and to verify whether CTFE was produced by the dehydrohalogenation of HCFC-123a.

METHODS

Each microcosm experiment was performed using the following series of treatments:

1. control buffer: K_2HPO_4
2. redox buffer: Na_2S, cysteine
3. redox buffer + hematin- level 1 mg/L
4. redox buffer + hematin- level 2 mg/L
5. purged leachate
6. leachate

MATERIALS

All chemicals were of the purest available grade and were used as received. Potassium phosphate dibasic and CFC-113 were obtained from Caledon Labs, HCFC-123a and CTFE from PCR Incorp., hematin (ferriprotoporphyrin IX hydrochloride) and sodium sulfate nonahydrate from Sigma Chemical Co., sodium hydroxide, sodium carbonate anhydrous and phosphoric acid from B.D.H., J.T. Baker, and Aldrich Chemical Companies respectively.

In the CFC-113 degradation experiment, the landfill leachate was collected from the Guelph municipal landfill, in Guelph Ontario on April 16, 1990. For the experiments with HCFC-123a and CTFE the landfill leachate was from the same site but was collected July 26, 1990. This latter sample was found to contain much less volatile compounds than the first leachate collected. The leachates were stored in tightly capped glass containers in the dark at 8°C.

All references to water refer to reverse osmosis water passed through an organic carbon filter (activated charcoal and ion exchange resin, Barnstead/Thermolyne Corporation, Dubuque, Iowa).

Glassware was washed successively with phosphate-free soap, water, methanol and then water once again. Glassware coming in contact with the treatment solutions after they were filtered was sterilized by autoclaving. The microcosms were 43 mL clear glass screw cap sample vials with a Teflon-lined silicone septum, designed for sampling for volatiles for EPA method 624 and purchased from Chromatographic Specialites, (Brockville Ont).

In the experiments with CTFE and HCFC-123a, approximately 0.5 cm of crushed glass was put into the bottom of the microcosm vials to facilitate microbial growth. After the crushed glass was added, the vials were sterilized by autoclaving.

SOLUTION PREPARATION

The "purged leachate" treatment involved purging the leachate with nitrogen gas for three hours. The unpurged leachate solution was the landfill leachate used as received. The control buffer, 0.1 M K_2HPO_4, was prepared by dissolving 17.42 g of potassium phosphate in water in a 1 L glass volumetric flask. The sulfide redox buffer was prepared by dissolving 24.0 g of $Na_2S.9H_20$ in 1 L of control buffer. A hematin solution of 100 mg/L hematin was prepared by dissolving 100 mg of hematin and 500 mg of sodium carbonate in 100 mL of control buffer solution, the sodium carbonate being added to help the hematin dissolve. This hematin solution was used to prepare two concentrations of redox-hematin buffers (1 mg/L and 2 mg/L), made by adding 10 mL or 20 mL of 100 mg/L hematin to sulfide redox buffer. The buffers were adjusted to pH 7.0 with concentrated phosphoric acid, and filtered through a 0.45 µm membrane into a sterile glass suction flask, transferred to sterile glass amber bottles, tightly capped and stored in the dark at 8°C. Before adding the chlorofluorocarbons, the four buffers were purged for one hour with nitrogen gas to eliminate oxygen, the pH was verified and adjusted if necessary.

CFC STANDARDS

The three chlorofluorocarbon spiking solutions (CFC-113, HCFC-123a and CTFE) were prepared in methanol. A cold 43 mL volatile vial was filled with prepurged methanol at -20°C. Using a gas tight syringe, 25 µL of pure CFC was injected through the vial septum into the vial of cold methanol. The resulting concentrations were calculated using the respective densities of CFC-113 and HCFC-123a.

The CTFE standard was prepared differently, since CTFE is a gas at room temperature. A 43 mL vial was half-filled with cold purged methanol, capped, left to sit until it warmed enough to cease sweating (approximately 5 minutes), wiped dry and then tared. CTFE was then bubbled into the methanol for 30 seconds. The vial was immediately capped and weighed and then filled to the top with more cold purged methanol. These standards, in methanol, were stored at -20°C and kept for no longer than 24 hours.

CFC ADDITION

To ensure optimal uniformity between vials of the same solution, the CFC was added to the buffers and landfill leachates in a batch method. The nominal amount of CFC added to experiments CFC-113 and HCFC-123a was 1.5 mg/L. In the CTFE experiment, the nominal concentration of CTFE added was 18 mg/L. The CFC standard in methanol (1.8 mL) was added using a glass syringe while the solution was being slowly stirred with a magnetic stir bar. After one minute, the bottle was returned to the cold room until the contents were transferred to the microcosm vials (one-half hour later). The microcosm vials were filled with the CFC spiked buffer and leachate solutions within a nitrogen purged glove box. The microcosms were stored in randomly selected sets of four, upside down in a 4 L capacity paint can which had been purged with N_2 to prevent diffusion of O_2 into the microcosms. The cans were labelled with scheduled sampling times and then stored in an environmental chamber at 20°C.

The following are the sampling times for each experiment:

CFC-113 Experiment: 0, 2, 6, 14, 28, 42, 66, 90 days
HCFC-123a Experiment: 0, 4, 8, 14, 28, 63, 113 days
CTFE Experiment: 0, 2, 8, 14, 28 days

In the experiments with HCFC-123a and CTFE, the vials for time zero were stored at 8°C for 20 hours before being analyzed. This was done to ensure that complete mixing of the CFC had occurred.

DECHLORINATION WITH Zn

The vicinal dechlorination of CFC-113 by zinc was verified in a separate experiment conducted in a 43 mL vial as described above. Zinc granules (0.2 g) were added to 35 mL of a 3 mg/L solution of CFC-

113 in water which was acidified with a drop of 3M HCl. The headspace was sampled every hour and the formation of CTFE was verified using a Photovac portable gas chromatograph.

ANALYTICAL METHODS

The analysis for the chlorofluorocarbons was conducted using an automated system consisting of a Dynatech PTA-30 autosampler (Chromatographic Specialties, Brockville Ont.) interfaced with an Envirochem Unacon 810 purge and trap concentrator, in turn interfaced directly with a Hewlett-Packard 5890-5790 GC-MSD. This automated system eliminated any manual handling of the samples and thus reduced possible losses of analytes to the atmosphere. The gas chromatograph temperature was programmed from -15°C to 135°C at 10°C/min. Mass acquisition was in the scan mode from 45 to 250 a.m.u. The column was a J&W Scientific fused silica capillary column (30m X 0.32 mm) coated with DB-624, 1.0 µm thick. The carrier gas was helium. Extracted ions 85, 117, and 116 peaks were used to quantify CFC-113, HCFC-123a and CTFE respectively. Chlorobenzene d-5 was used as the internal standard.

In order to minimize cross contamination, samples were analyzed in the order of the treatment as listed above. A blank sample was run between each set of solutions (every three samples) and after the standards. A standard was run at the beginning and at the end of analyses each day. The system was flushed and the samples diluted as required, with 2.5% methanol in water. This concentration was found to be sufficient to reduce carry-over without affecting the purging efficiency of the analyte.

To neutralize noxious hydrogen sulfide fumes released during analysis, sodium hydroxide pellets were added to the waste bottle, and the autosampler was covered with a large clear plastic bag with the edges sealed to the table top with tape. One open vial containing aqueous sodium hydroxide was placed in the autosampler tray.

After volatile analysis, pH and dissolved oxygen measurements were performed. The pH was measured with an Orion Research Model 231 Ph/mV temperature meter and dissolved oxygen was measured with a YSI Model 54A oxygen meter and probe. Fluoride concentration in the CTFE experiment was measured using a Waters liquid chromatograph with a conductivity detector.

RESULTS AND DISCUSSION

CFC-113

The pseudo first order rate constant was calculated using the equation:

$$\ln C = -kt + \ln C_0$$

where C_0 was the nominal initial concentration. This was done to compensate for the apparent slow dissolution rate of CFC-113; the observed concentration at T_0 was always lower than at T_2. In a separate experiment it was found that, even in the presence of an excess of CFC-113, it took up to two weeks for the aqueous phase to reach saturation.

The half-life for CFC-113 under the various conditions was calculated using the following equation:

$$T_{1/2} = \frac{\ln 2}{k}$$

The results of these calculations with the standard deviations (n = 24) are presented in Table 1.

Table 1. CFC-113 Degradation Rate in Microcosms at 20 °C

Treatment	k ± SD ($days^{-1}$)	Half-life ± SD (days)
1. Control buffer	$7.6 \pm 2.2 \times 10^{-3}$	90 ± 27
2. Redox buffer	$9.1 \pm 2.4 \times 10^{-3}$	75 ± 20
3. Redox-hematin 1 mg/L	$1.2 \pm 0.2 \times 10^{-2}$	57 ± 11
4. Redox-hematin 2 mg/L	$1.6 \pm 0.3 \times 10^{-2}$	43 ± 8
5. Purged leachate	$9.9 \pm 1.4 \times 10^{-2}$	7.0 ± 1.0
6. Leachate	$1.4 \pm 0.3 \times 10^{-1}$	5.0 ± 1.2

The pH remained constant at 7 in all the buffered systems, but decreased most significantly from 7.2 to 6.5 in the purged leachate after 40 days of incubation. The leachate was at a pH of 6.7 at the onset and decreased only slightly to 6.5 during the 90 day period. Dissolved oxygen remained below 0.5 ppm for

all treatments throughout the study period, except for the phosphate buffer which gave measurements of 2-5 ppm.

Control buffer

The concentration of CFC-113 in phosphate buffer decreased slowly during the 90 day incubation period, with almost half of the material remaining at the end (Figure 2a). Because of possible losses through the teflon-lined septa, the appearance of metabolites, in addition to the rate of loss of the parent compound, was used as unequivocal evidence of degradation.

Redox buffer

The results with the redox buffer were virtually identical to those obtained with the phosphate buffer, but small amounts of CTFE (0-35 µg/L; Figure 2f) were formed throughout the incubation period. Only trace amounts of HCFC-123a were observed. The presence of sulfide seems to promote the formation of CTFE. However, sulfide also enhances the rate of degradation of CTFE, which is probably why it doesn't seem to accumulate with time (*vide infra*). The sulfide ion can act as an electron donor to produce the elimination of Cl^-. The reaction is probably a nucleophilic substitution followed by an elimination. CTFE was also formed from CFC-113 in a slightly acidic medium in the presence of Zn metal with an estimated half-life of 3 days. The mechanism of the two reactions are different because in the case of zinc, the initial attack is on the chlorine, not the carbon.

Redox-Hematin

The addition of hematin resulted in the additional formation of HCFC-123a (Figure 2e). Although hematin is initially in the oxidized state (Fe^{+3}), its addition to a strongly reducing buffer causes its immediate reduction to the active (Fe^{+2}) state, as can be seen by a persistent red colour, and the excess buffer ensures its immediate reconversion. It is the iron porphyrin in the reduced state which is deemed responsible for the reductive dechlorination reaction, and, as had been observed for carbon tetrachloride and 1,1,1-trichloroethane (22), the reaction rate seemed to be dependent on the concentration of hematin (Table 1; the data is not quite statistically significant at the 95% confidence level, but the trend is present). It is interesting to note that in the case of redox-hematin buffers, while some HCFC-123a was formed, substantial quantities of the primary substrate remained throughout the experiment. The amount of metabolite formed was much lower than in the methanogenic leachates. These results parallel results found by Bouwer and Lew who found that sulfate reducers converted only 60% of CFC-113 to

HCFC-123a (25). Iron sulfide, seen as a black precipitate, and H_2S, were formed in these microcosms, but not in the leachate. Indeed the landfill leachate used for this experiment did not contain any appreciable quantity of sulfide. The iron within the hematin molecule could be the source of this precipitate.

Leachate and purged leachate

The results for the degradation of CFC-113 are depicted on Figure 2 a-c. As can be seen from the data in Table 1, CFC-113 was degraded with an average half-life of 5 and 7 days for leachate and purged leachate respectively. For both the leachate and the purged leachate the disappearance of the primary substrate was accompanied by an increase of the primary metabolite, HCFC-123a (Figure 2b). The calculated rate constants indicate that the reaction was slightly faster in leachate than in purged leachate, but the difference was not statistically significant. The other chlorinated solvents present in the leachate did not exhibit any inhibitory effect (dichloromethane; trichloroethene; 1,1 and 1,2-dichloroethane; 1,1-dichloroethene; cis and trans-1,2-dichloroethene; vinyl chloride ; total concentration 3 mg/L) contrary to what had been observed in the case of the dechlorination of dichloromethane (19).

Analysis by gas chromatography of the headspace formed in the leachate microcosms confirmed that they were methanogenic. The leachates were not amended with any other carbon source, since they contained in excess of 3,000 mg/L of volatile fatty acids and the CFC-113 solution was in methanol, which contributed approximately 800 mg/L of carbon. The addition of methanol probably stimulated the methanogenic population already active within the landfill.

The production of CTFE did not seem to be tied to the formation of HCFC-123a. As discussed above, its formation in the redox buffer and by the addition of zinc demonstrates that this reaction occurs abiotically. The maximum concentration of HCFC-123a was reached at 64 days in leachate and purged leachate and represented over 85% of the initial CFC-113 concentration. Therefore, reductive dechlorination to HCFC-123a is the preferred degradation pathway in anaerobic landfill leachate. It was then important to be able to assess the fate of HCFC-123a.

HCFC-123a

The fate of HCFC-123a, the primary metabolite of CFC-113, was studied under identical conditions. The results are presented in Table 2 and on Figure 3. The previously postulated elimination of HCl (Figure 1) did not seem to occur as no CTFE was found in any of the treatments. No degradation products were identified in any of the buffers or redox-hematin treatments. However, after two weeks of incubation in the leachate, two early eluting peaks were observed. The total ion chromatogram and the two spectra are shown on Figure 4 . The structure of the compound eluting at 4.8 min was assigned to 1-chloro-1,2,2-trifluoroethane (HCFC-133) on the basis of the mass spectral fragmentation shown on Figure 5a, whereas the compound eluting at 3.9 min was deduced to be 1-chloro-1,1,2-trifluoroethane (HCFC-133b) (Figure 5b). Trace amounts of these compounds were confirmed in retrospect in the CFC-113 microcosms. The initial rate of formation of HCFC-133b was faster than that of HCFC-133, but their concentrations eventually became equal after 67 days of incubation (Figure 6). Since there were no standards available for these compounds, it wasn't possible to quantify them exactly. However assuming an equal response to the mass spectrometer it was estimated that there was approximately thirty times more dechlorination in the leachate than in the purged leachate. These products were not observed in the redox-hematin treatments and the buffers. No further reductive dechlorination products were observed during the 115 days incubation period.

Table 2. HCFC-123a Degradation Rate in Microcosms at 20 °C

Treatment	k ($days^{-1}$)	Half-life (days)
1. Control buffer	$8.0 \pm 4.2 \times 10^{-3}$	127 ± 47
2. Redox buffer	$4.6 \pm 2.0 \times 10^{-3}$	150 ± 64
3. Redox-hematin 1 mg/L	$4.4 \pm 2.2 \times 10^{-3}$	157 ± 80
4. Redox-hematin 2 mg/L	$4.2 \pm 1.9 \times 10^{-3}$	166 ± 77
5. Purged leachate	$1.2 \pm 0.1 \times 10^{-1}$	5.6 ± 0.6
6. Leachate	$1.2 \pm 0.1 \times 10^{-1}$	5.6 ± 0.4

Dissolved oxygen remained below 0.2 ppm for all treatments except in the phosphate buffer which varied between 0.2 and 1 ppm. In this experiment, the pH of the purged leachate started at 8.3, decreased to

7.3 after 25 days and slowly rose again to 8 after 115 days of incubation. Leachate pH remained around 7.2 for most of the experiment, but increased to 8.5 at 115 days. These changes in an unbuffered system could be attributed to the formation followed by the depletion of organic acids.

Although no CTFE was formed from HCFC-123a in any of the microcosms, its formation could be induced in a buffer solution at high pH (>10). In this case, the formation of CTFE is a base-catalysed elimination of HCl. Although the final product is the same, the direct formation of CTFE from CFC-113 probably occurs via a different mechanism. The formation of CTFE from CFC-113, in buffers at neutral pH, was not correlated with the presence of HCFC-123a.

CTFE

The stability of CTFE was assessed under the same conditions as the CFC-113 and HCFC-123a microcosms described above, primarily to ensure that its wasn't lost through volatilisation and diffusion through the septa, but also to determine whether its rate of degradation could in fact equal its rate of formation and thus possibly explain why it wasn't observed in the HCFC-123a microcosms and did not accumulate in the CFC-113 microcosms.

The results are presented in Table 3 and on Figure 7. The concentration of CTFE in the control buffer remained stable during the 30 day incubation period. Similarly it was found to be stable in both purged and unpurged leachates. In the redox buffer and the redox-hematin treatments, it was found to be rapidly degraded with a half-life of 0.3 day. Because of the rapid rate of disappearance in sulfide containing buffer, it wasn't possible to fully assess the effect of the addition of hematin. CTFE is most probably degraded by nucleophilic attack of the double bond by HS^-. Indeed fluorinated olefins, unlike their other halogenated analogs, are very susceptible to nucleophilic addition because of the strong electron-withdrawing effect of the fluorine atom (26). The difference in rate between the phosphate buffer and the redox buffer can be explained by the much higher reactivity of sulfide versus hydroxyl as nucleophiles.

Table 3. CTFE Degradation Rate in Microcosms at 20 °C		
Treatment	**k (days^{-1})**	**Half-life (days)**
1. Control buffer	$4.1 \pm 0.2 \times 10^{-2}$	17 ± 1
2. Redox buffer	2.4 ± 0.1	0.3 ± 0.01
3. Redox-hematin 1 mg/L	1.4 ± 0.1	0.5 ± 0.03
4. Redox-hematin 2 mg/L	2.9 ± 0.6	0.2 ± 0.05
5. Purged leachate	$1.8 \pm 1.1 \times 10^{-2}$	38 ± 24
6. Leachate	$1.6 \pm 1.0 \times 10^{-2}$	43 ± 26

The degradation product of CTFE was not isolated, but the formation of nearly two mole equivalent of fluoride was measured by ion chromatography. It is therefore most likely that complete hydrolysis occurs. CTFE would not be expected to accumulate in the sulfate reducing area of a contaminated aquifer, yet this is exactly where it was found originally (3). This implies that it was constantly being replenished by a slowly dissolving pool of CFC-113. Although CTFE is not persistent in sulfide buffers, it did persist in methanogenic landfill leachate. Of the degradation products of CFC-113, it is CTFE that is of the most concern because of its toxicity.

SUMMARY

The revised pathway for the degradation of CFC-113 is depicted in Figure 8. It now appears that the elimination reactions are abiotic whereas the reductive dechlorination reactions can either be carried out by methanogenic bacteria or by reduced hematin. Hematin is analogous to the porphyrin portion of cytochrome P-450, one of the enzymes capable of catalyzing this transformation in microbial and mammalian systems.

This experiment confirmed that CFC-113 could be degraded by a consortium of bacteria acclimated to chlorinated organic solvents. The reductive dechlorination of CFC-113 in landfill leachate occurred rapidly (half-life of 5 days). The presence of other chlorinated solvents did not reduce the rate of the reaction. HCFC-123a was confirmed as the primary metabolite of CFC-113 and was found to be further reductively

dechlorinated to HCFC-133 and HCFC-133b. CTFE, unlike what had been postulated previously , appears to be formed directly from CFC-113. CTFE was relatively stable in leachate and phosphate buffer, but was rapidly decomposed in the presence of sulfide.

List of figures

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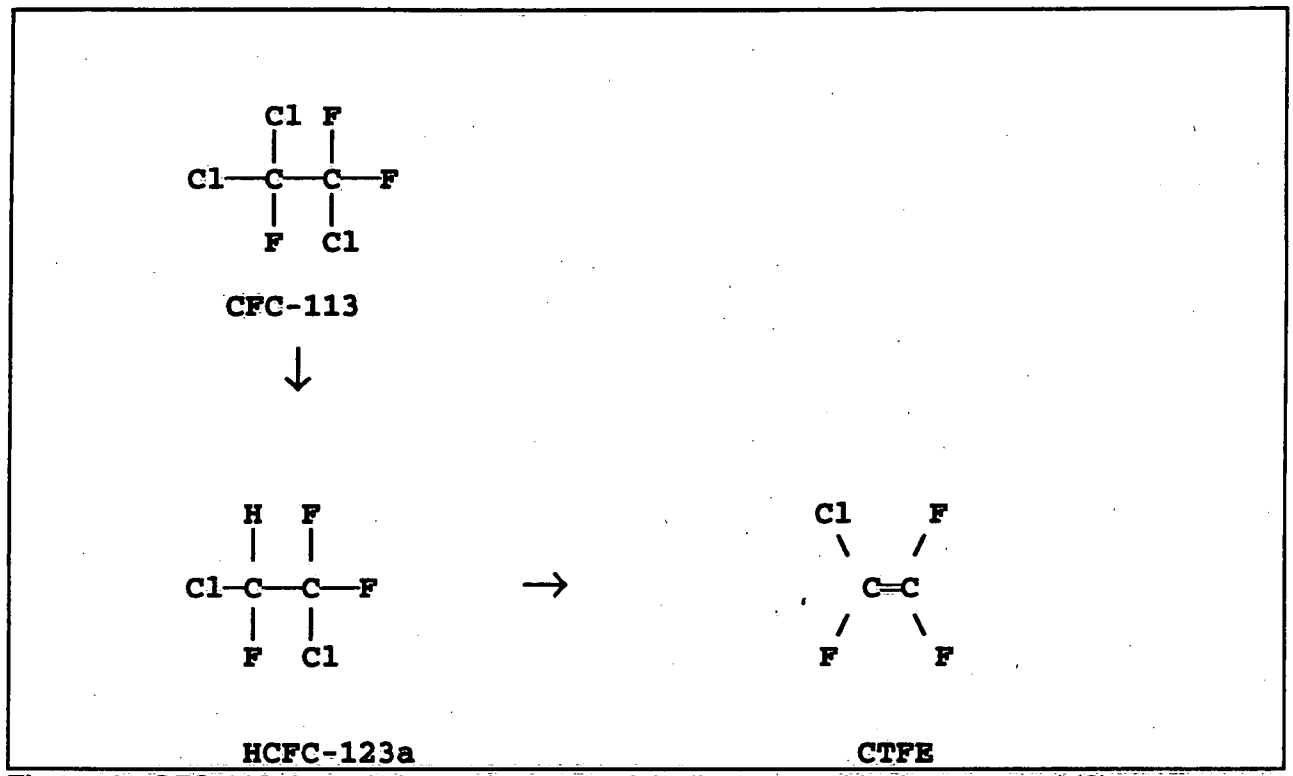
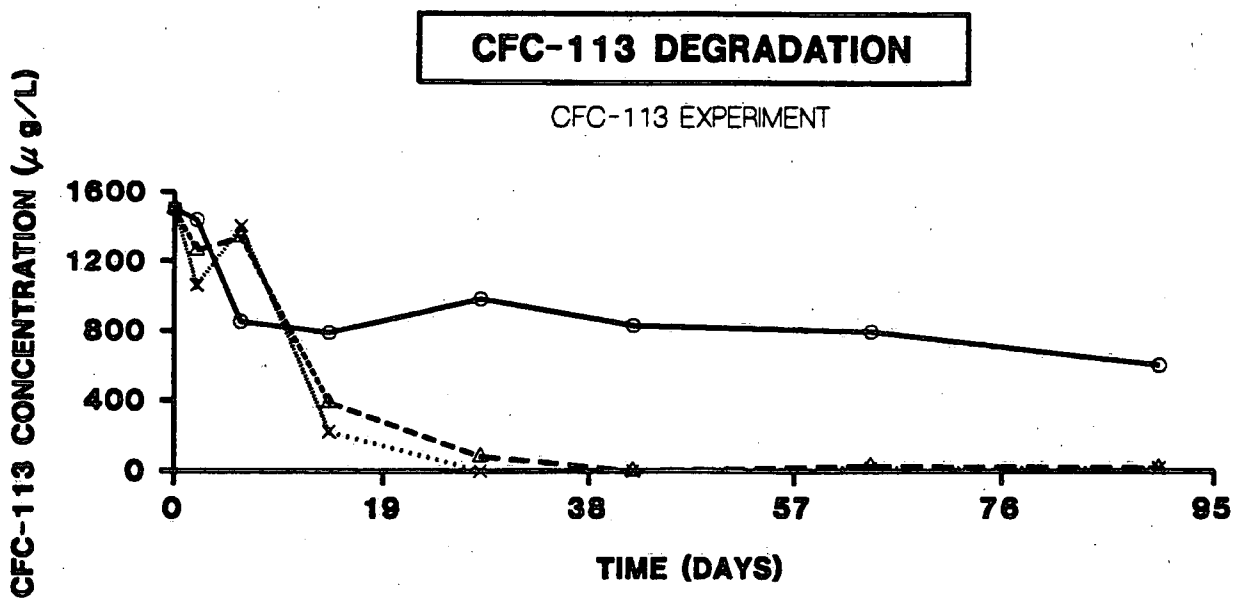
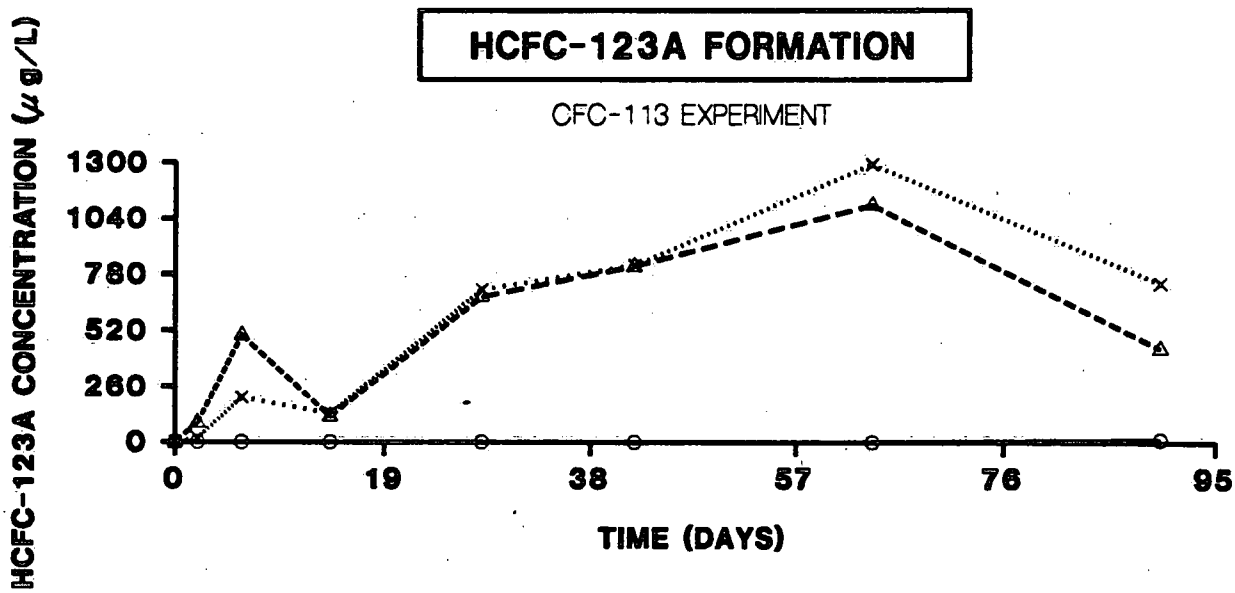


Figure 1. CFC-113 degradation pathway as originally proposed by Lesage et al (3).

a)


CFC-113 DEGRADATION
CFC-113 EXPERIMENT
CFC-113 CONCENTRATION (µg/L)
1600
1200
800
400
0
0
19
38
57
76
95
TIME (DAYS)

b)


HCFC-123A FORMATION
CFC-113 EXPERIMENT
HCFC-123A CONCENTRATION (µg/L)
1300
1040
780
520
260
0
0
19
38
57
76
95
TIME (DAYS)

c)

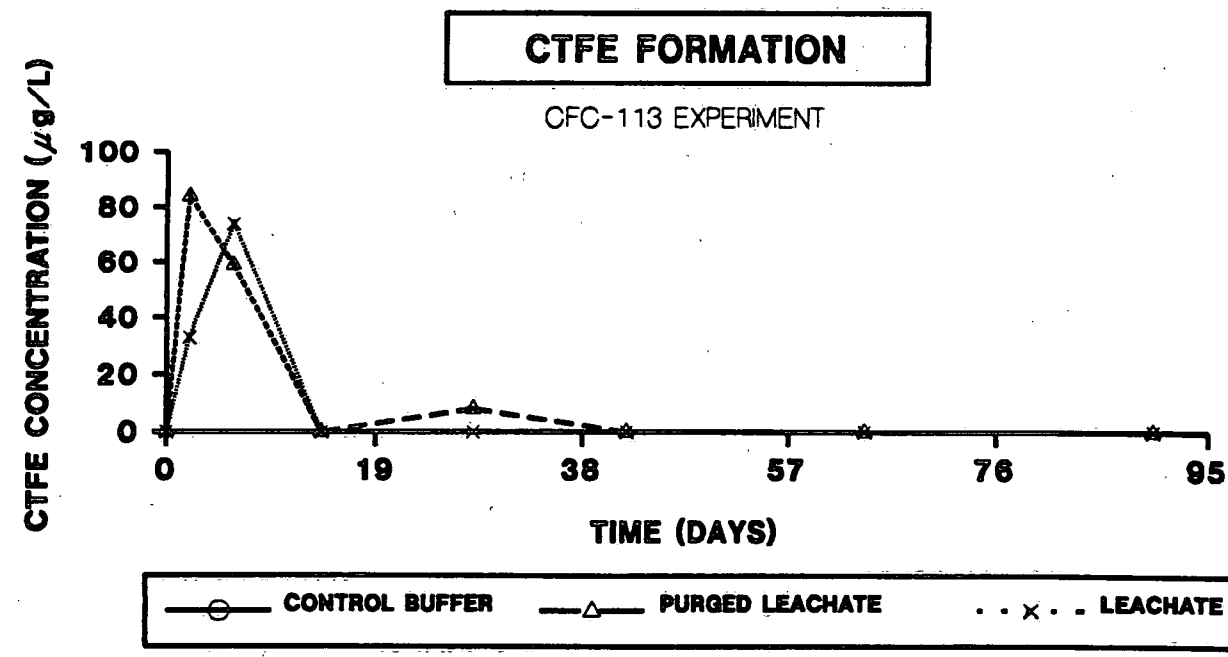
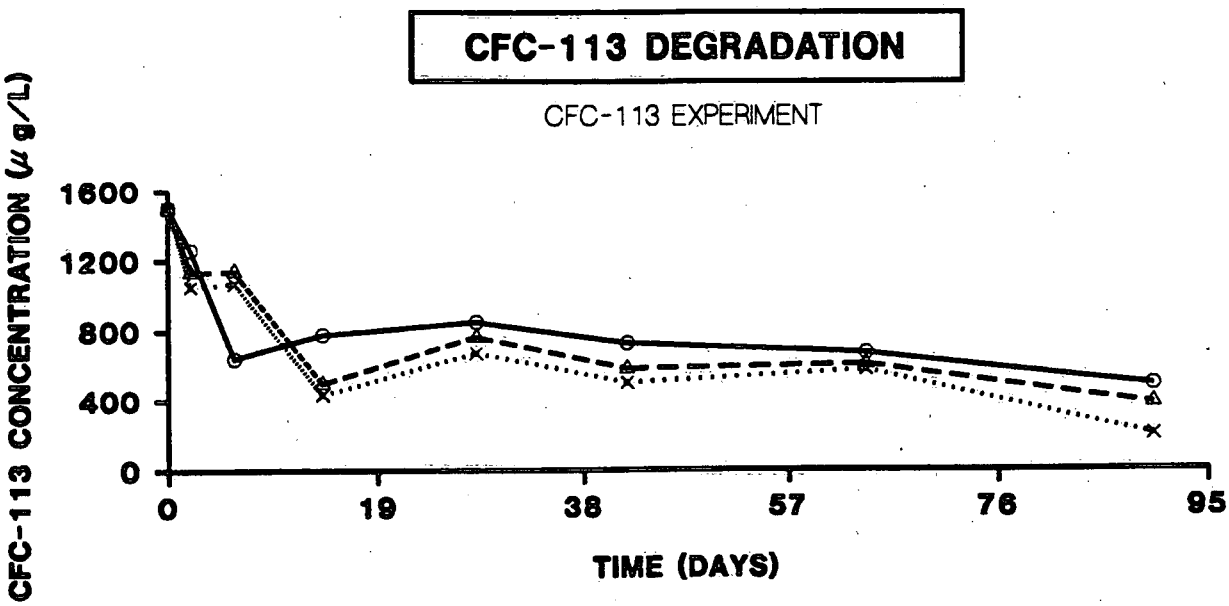
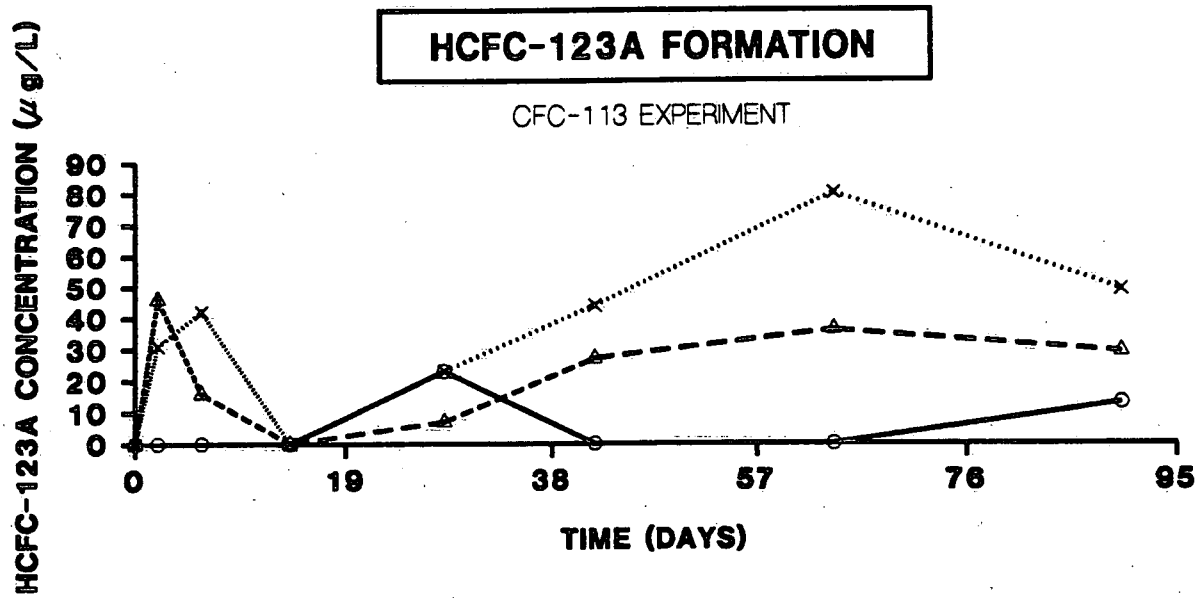

CTFE FORMATION
CFC-113 EXPERIMENT
CTFE CONCENTRATION (µg/L)
100
80
60
40
20
0
0
19
38
57
76
95
TIME (DAYS)
CONTROL BUFFER
PURGED LEACHATE
LEACHATE

Figure 2

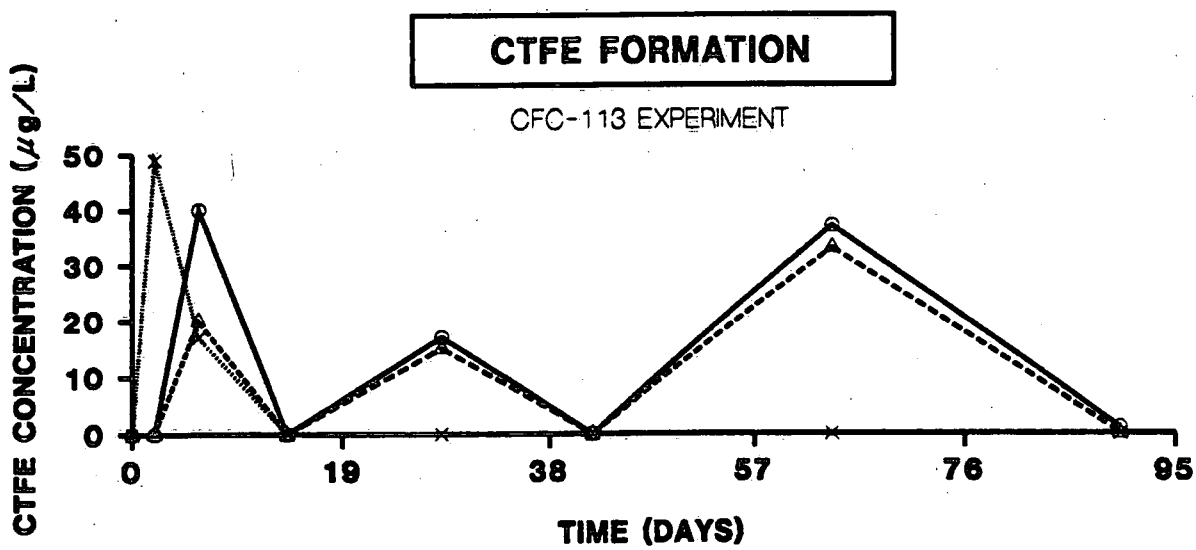
d)


CFC-113 DEGRADATION
CFC-113 EXPERIMENT
CFC-113 CONCENTRATION (µg/L)
1600
1200
800
400
0
0
19
38
57
76
95
TIME (DAYS)

e)


HCFC-123A FORMATION
CFC-113 EXPERIMENT
HCFC-123A CONCENTRATION (µg/L)
90
80
70
60
50
40
30
20
10
0
0
19
38
57
76
95
TIME (DAYS)

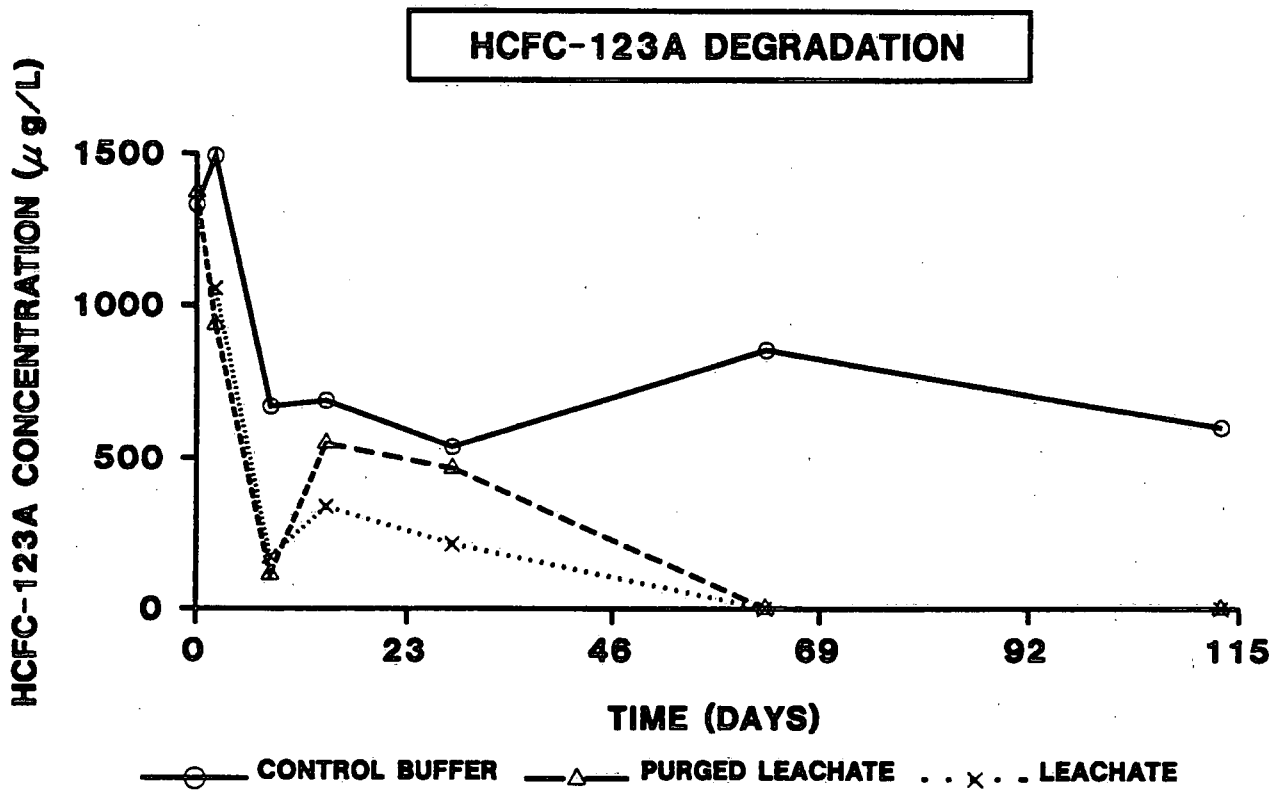
f)


CTFE FORMATION
CFC-113 EXPERIMENT
CTFE CONCENTRATION (µg/L)
50
40
30
20
10
0
0
19
38
57
76
95
TIME (DAYS)


REDOX BUFFER
REDOX HEMATIN 1 mg/L
REDOX HEMATIN 2 mg/L

Figure 2

a)


HCFC-123A DEGRADATION
HCFC-123A CONCENTRATION (μg/L)
1500
1000
500
0
0
23
46
69
92
115
TIME (DAYS)
CONTROL BUFFER
PURGED LEACHATE
LEACHATE

b)

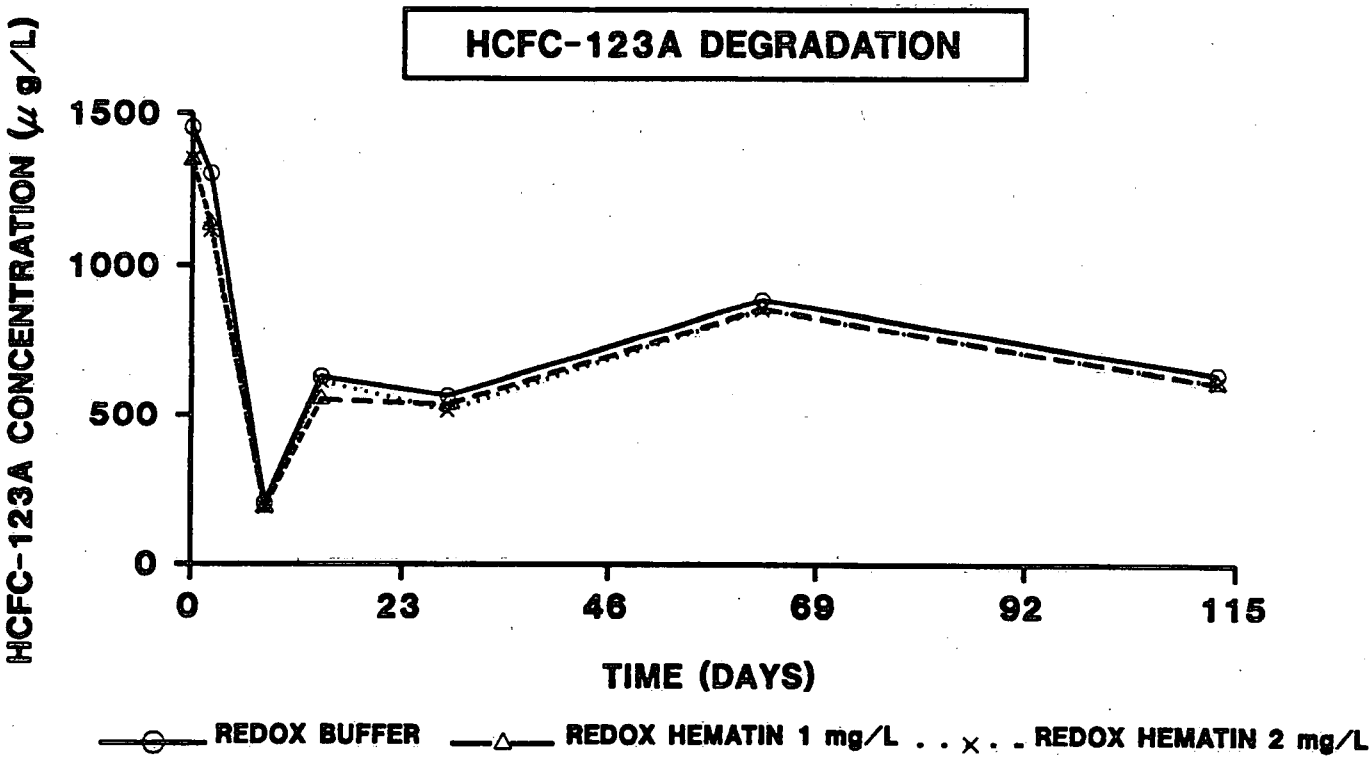
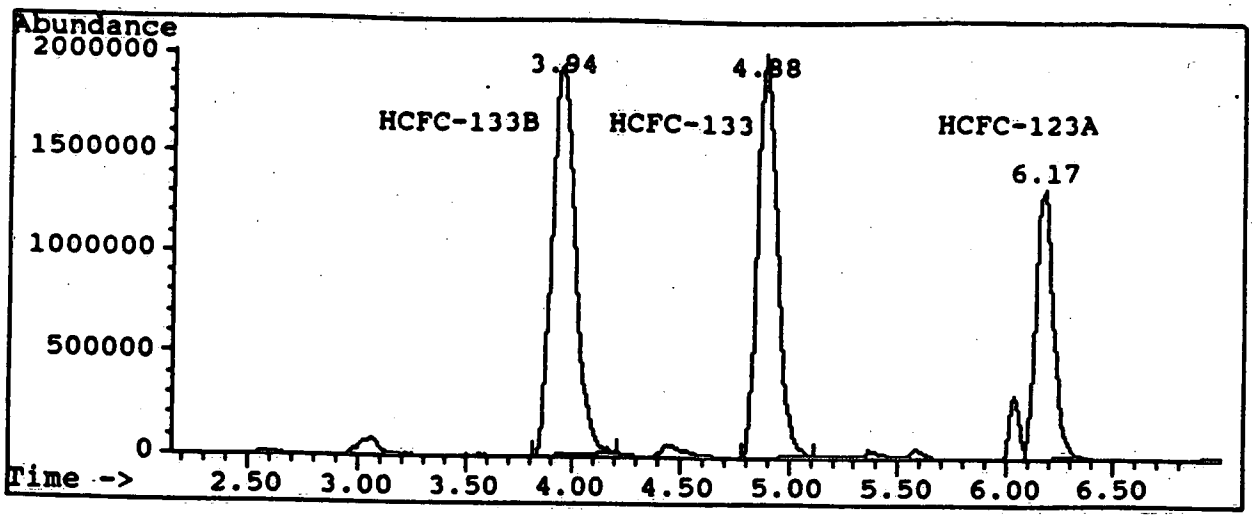
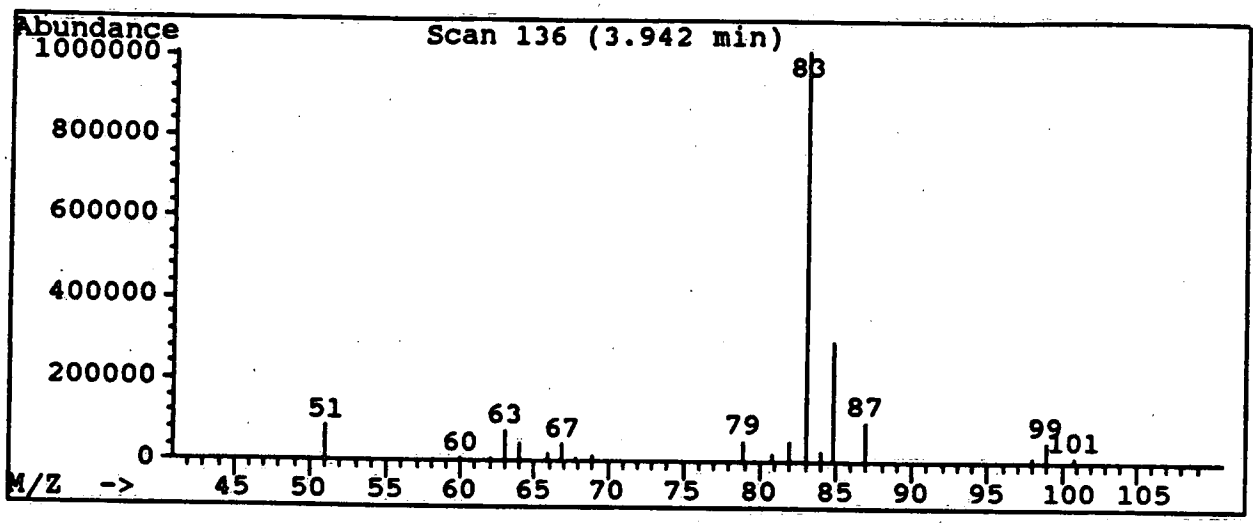

HCFC-123A DEGRADATION
HCFC-123A CONCENTRATION (μg/L)
1500
1000
500
0
0
23
46
69
92
115
TIME (DAYS)
REDOX BUFFER
REDOX HEMATIN 1 mg/L
REDOX HEMATIN 2 mg/L

Figure 3

a)


TOTAL ION CHROMATOGRAM
Abundance
2000000
1500000
1000000
500000
0
3.94
4.88
HCFC-133B
HCFC-133
HCFC-123A
6.17
Time ->
2.50 3.00 3.50 4.00 4.50 5.00 5.50 6.00 6.50

b)


HCFC-133B SPECTRUM
Abundance
Scan 136 (3.942 min)
1000000
800000
600000
400000
200000
0
83
51
60
63
67
79
87
99
101
M/Z ->
45 50 55 60 65 70 75 80 85 90 95 100 105

c)

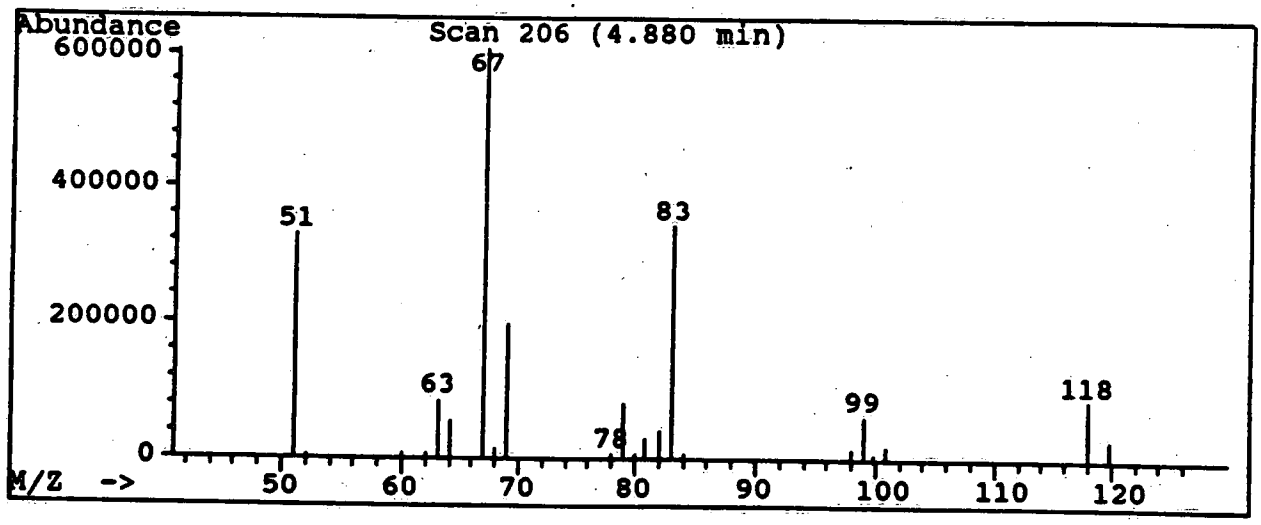

HCFC-133 SPECTRUM
Abundance
Scan 206 (4.880 min)
600000
400000
200000
0
67
51
83
63
78
99
118
M/Z ->
50 60 70 80 90 100 110 120

Figure 4

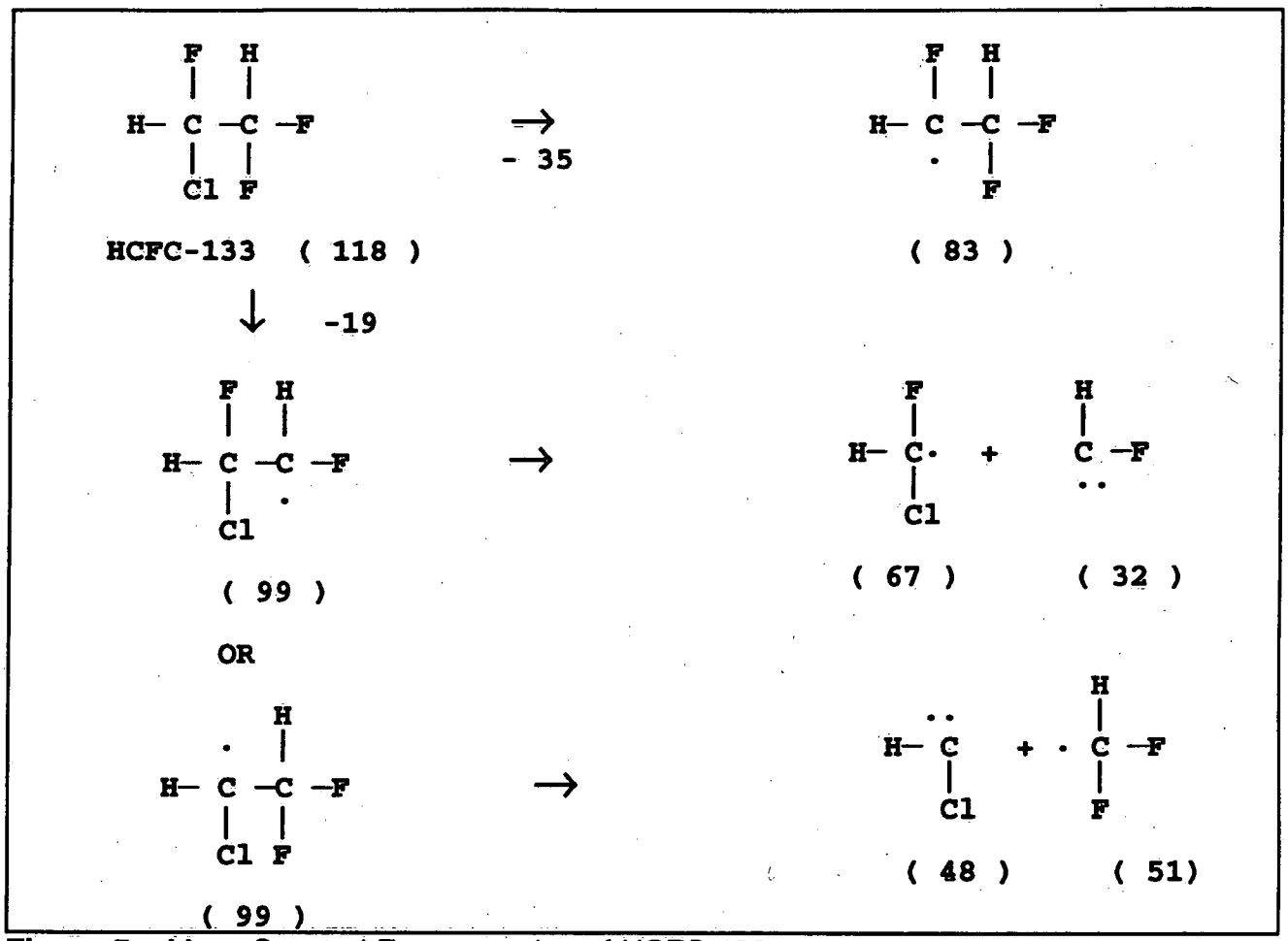


Figure 5a. Mass Spectral Fragmentation of HCFC-133

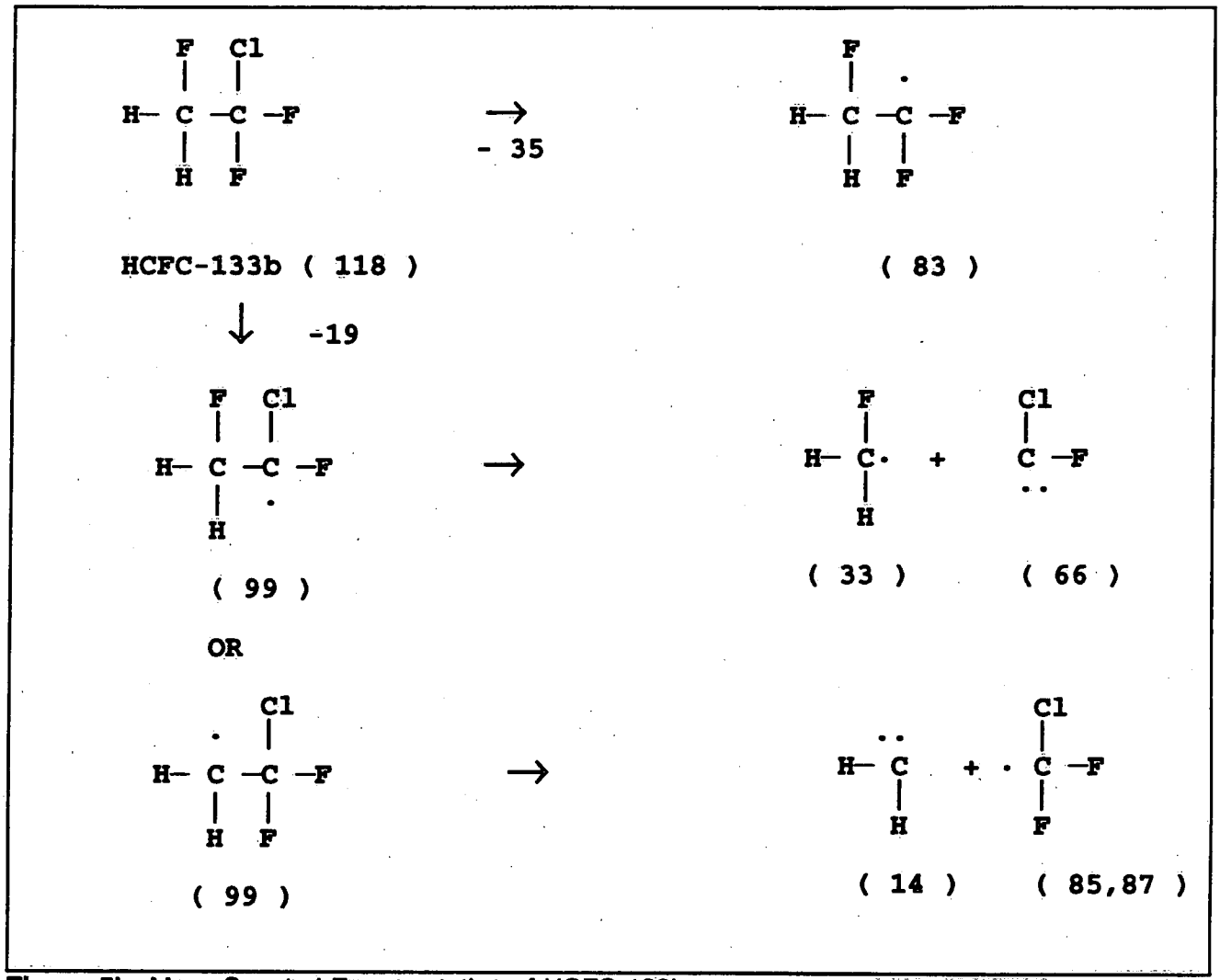
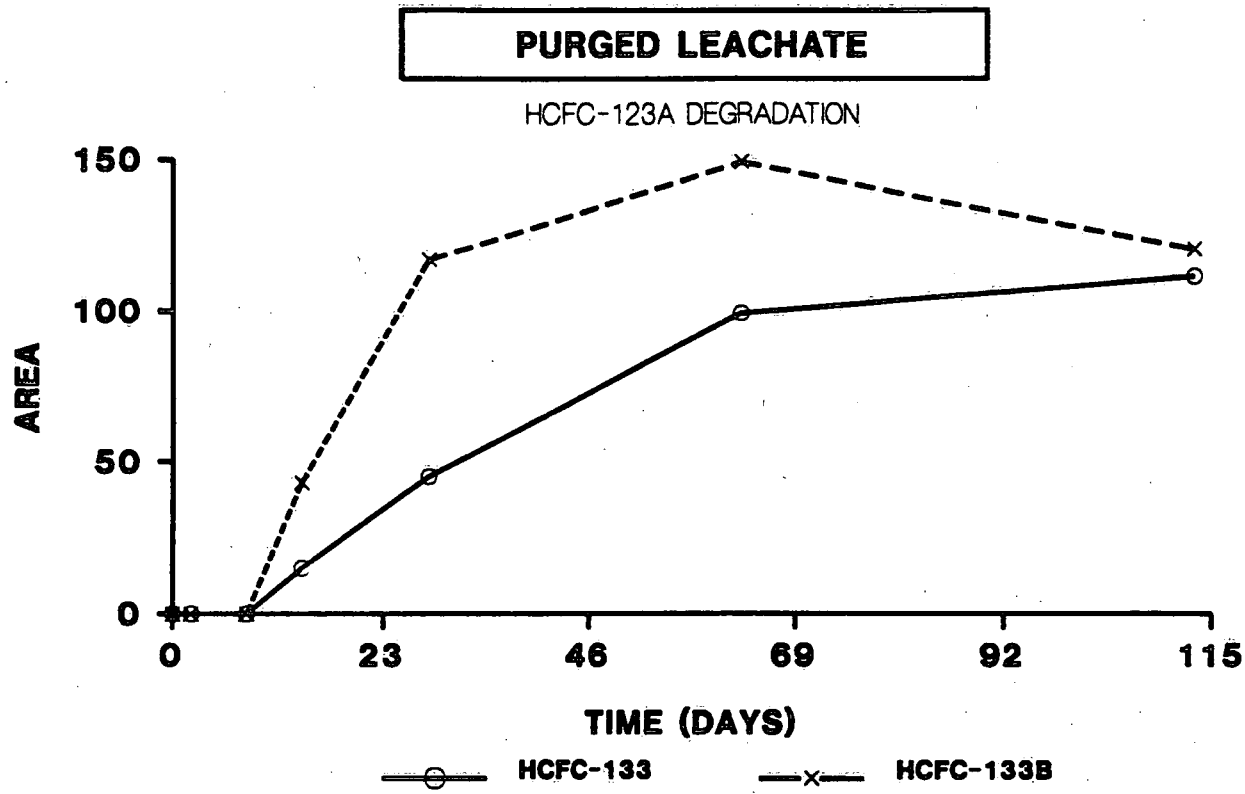


Figure 5b Mass Spectral Fragmentation of HCFC-133b

HCFC-133 AND HCFC-133B METABOLITES

a)



b)

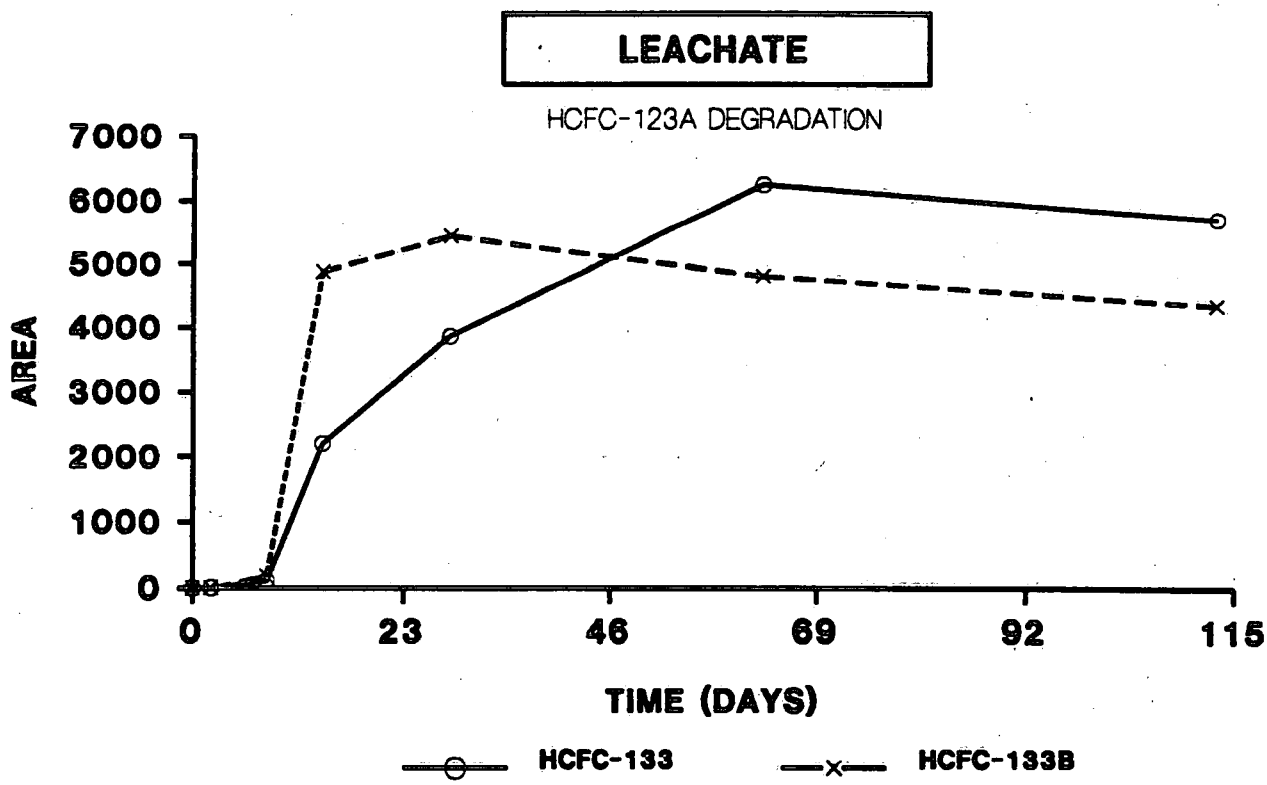
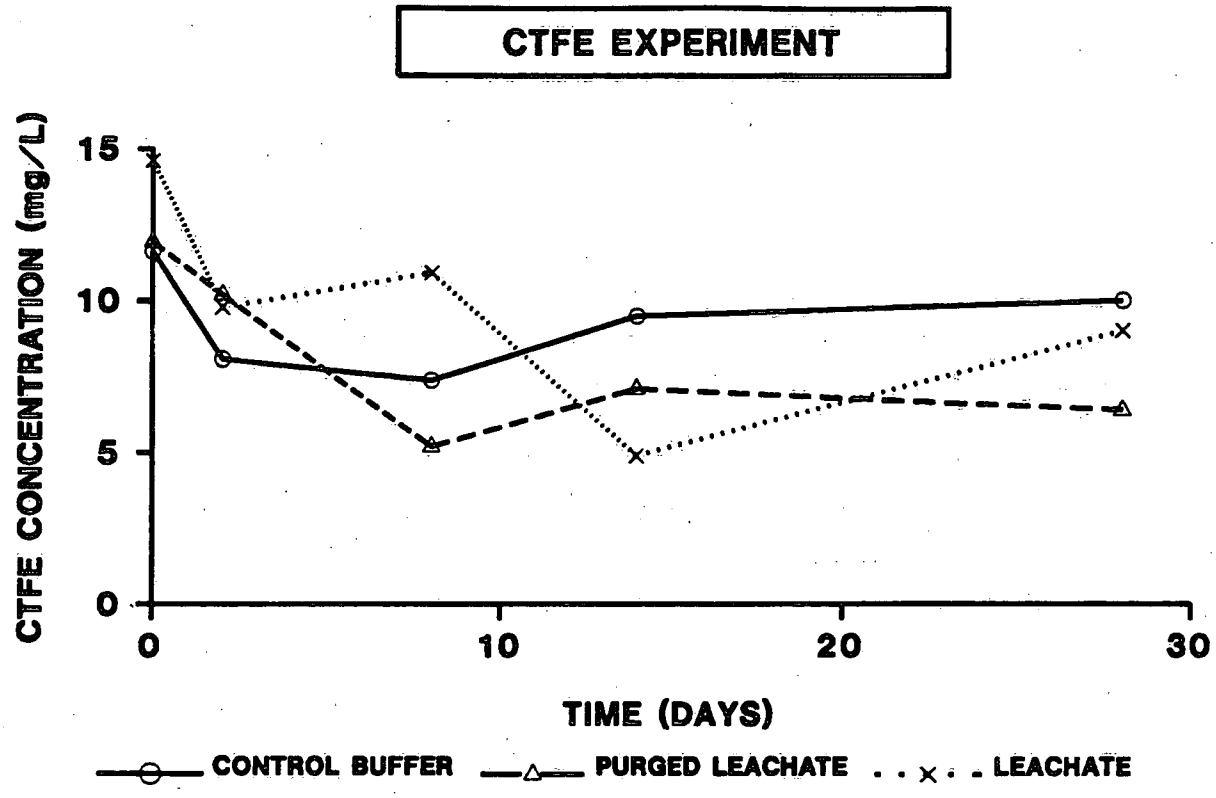


Figure 6

a)


CTFE EXPERIMENT
CTFE CONCENTRATION (mg/L)
15
10
5
0
0
10
20
30
TIME (DAYS)
CONTROL BUFFER
PURGED LEACHATE
LEACHATE

b)

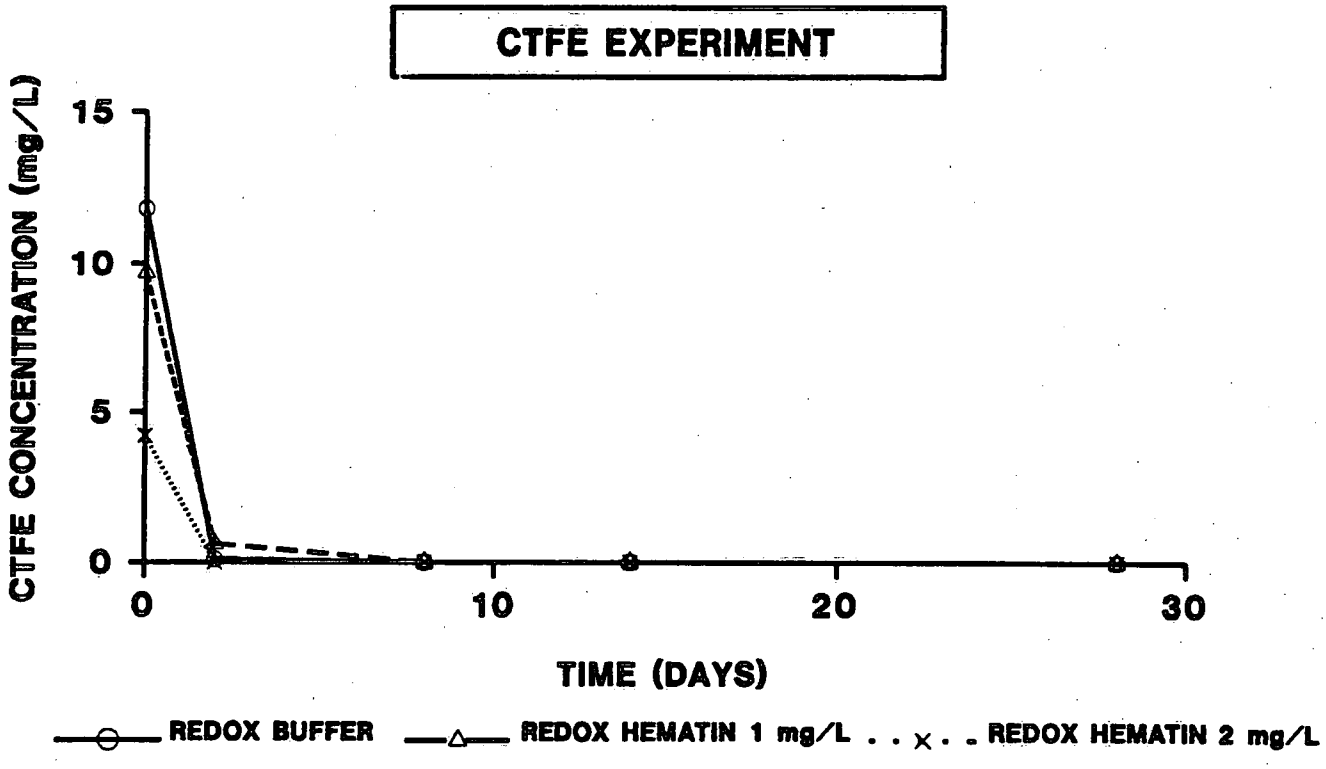

CTFE EXPERIMENT
CTFE CONCENTRATION (mg/L)
15
10
5
0
0
10
20
30
TIME (DAYS)
REDOX BUFFER
REDOX HEMATIN 1 mg/L
REDOX HEMATIN 2 mg/L

Figure 7

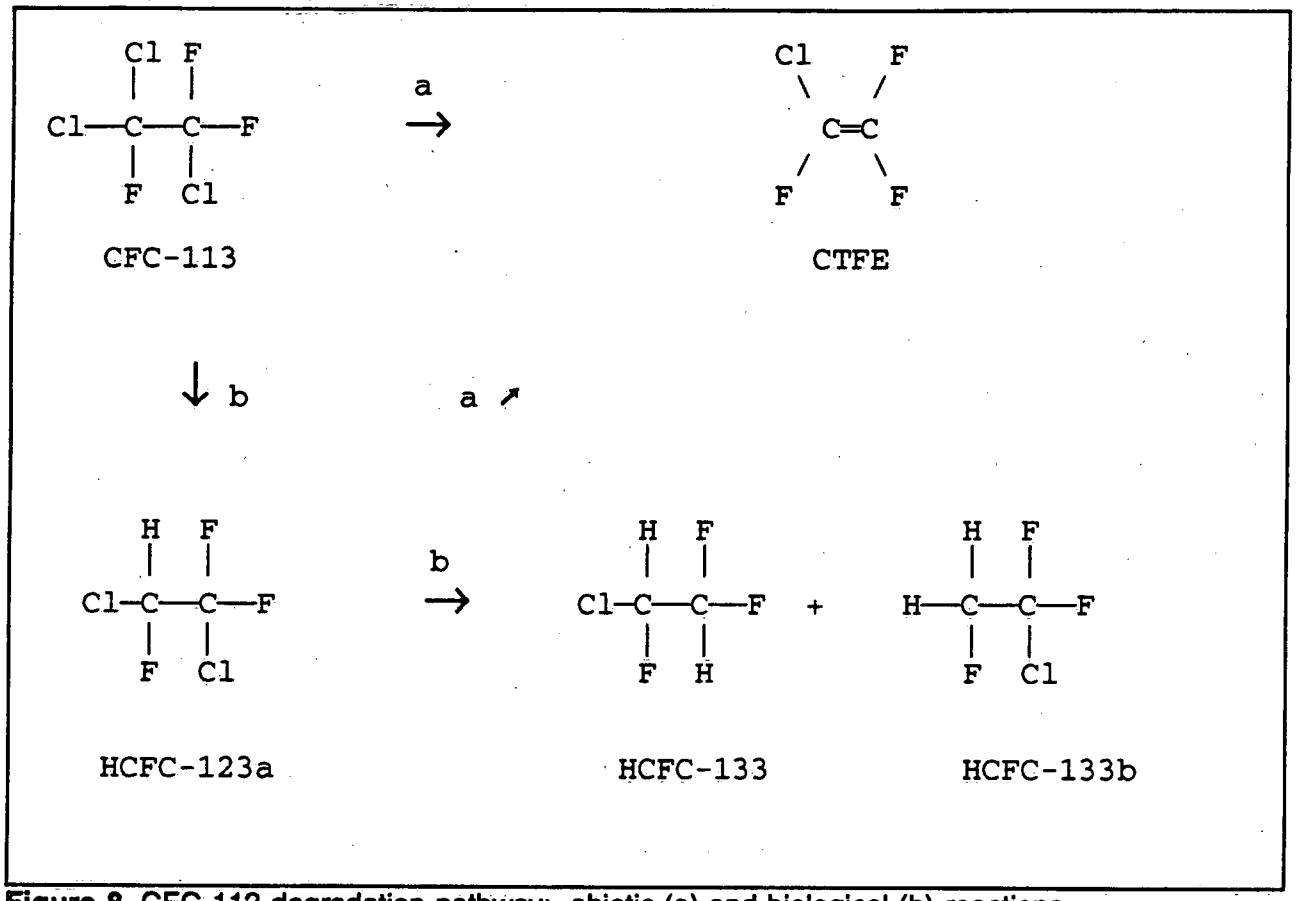
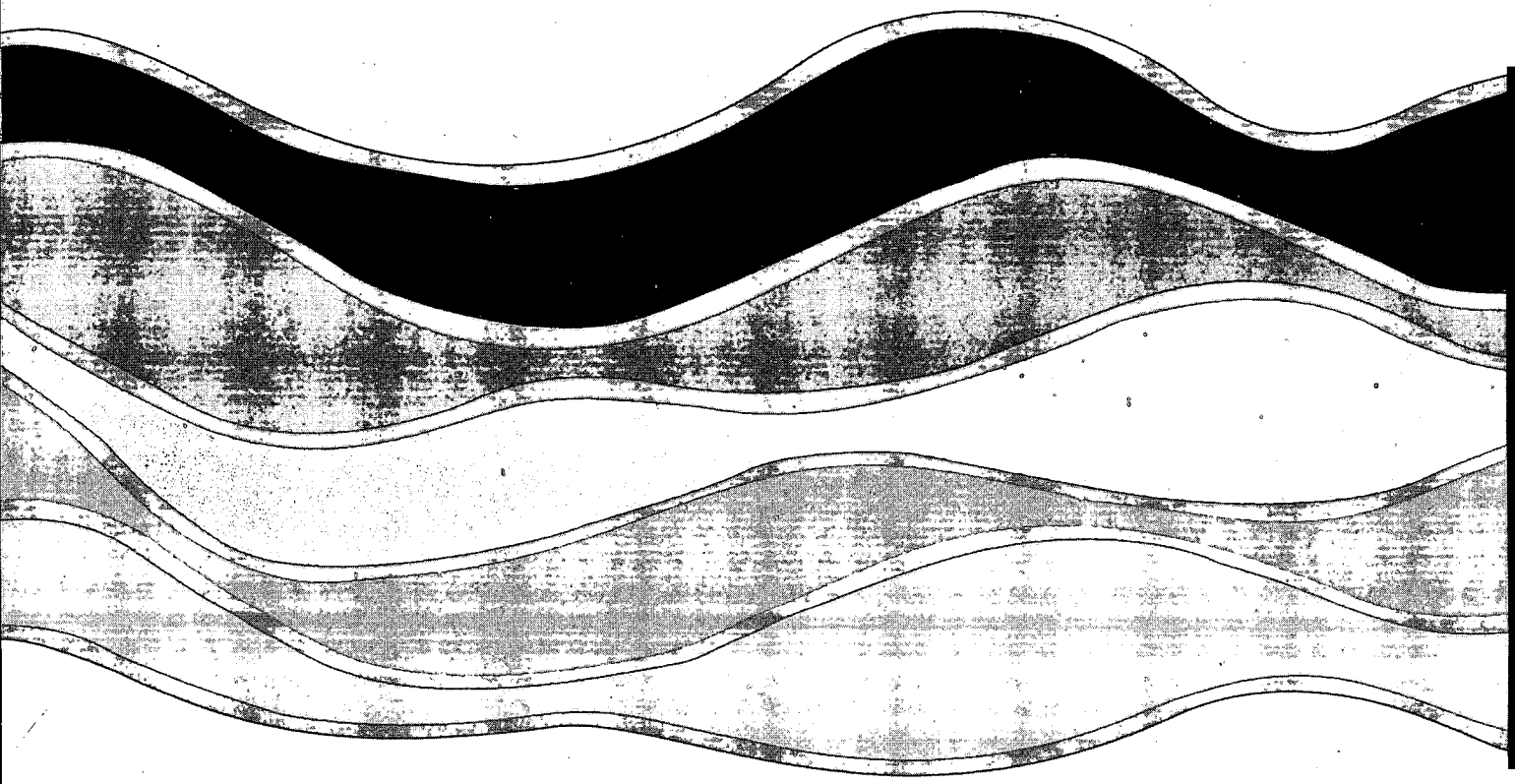


Figure 8. CFC-113 degradation pathway: abiotic (a) and biological (b) reactions.



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