Technical Review of Utility and Costs of the Atlantic Salmon and Atlantic Whitefish Live Gene Banks at the Maritimes Region Biodiversity Facilities

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Acknowledgements

This review of the utility and costs of Maritimes Region live gene banks at the biodiversity facilities was completed for the Director General Fisheries, Environment and Biodiversity Science, Fisheries and Oceans, Ottawa, and based on a Terms of Reference provided in by that office. Review of the document during its completion was provided by D. Meerburg, Senior Advisor, Diadromous Fish, Science Branch, Ottawa.

Author's note: 2016:

This manuscript was prepared in 2004 to satisfy a request by the then Director General of Fisheries, Environment and Biodiversity Science. The information was provided to that office in the form of a draft manuscript and via accompanying teleconferences and emails. The document was readied for publication in 2016 because of a perceived need to make the information more available to serve as a record of the approach to establish the live gene bank and for those involved in the recovery process. Since the original draft was prepared, four of the authors, T.L. Marshall, P.G. Amiro, S.F. O'Neil, and T.R Goff, retired from DFO; as has D. Meerburg, who was the headquarters (Ottawa) based Science representative who participated in the working group that structured the form of the review and provided editorial comments on the manuscript.

Live gene banking of salmon is ongoing and at the time of this writing is planned to continue, subject to periodic reviews of programs as are common in Science programs conducted by government. A key component of the ongoing live gene banking has been confirmation that the genetic diversity can be retained. Documentation of that aspect of the LGB occurred after this technical review was conducted in 2004 and is available as a research document: O'Reilly, P.T., and C.A. Harvie. 2009. *Conservation of genetic variation in the inner Bay of Fundy Atlantic salmon captive breeding and rearing program.* Can. Sci. Adv. Sec. Res. Doc. 2009/95. 61p. (http://www.dfo-mpo.gc.ca/csas-sccs/publications/resdocs-docrech/2009/2009_095-eng.htm).

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ABSTRACT

Marshall, T.L., P.G. Amiro, P.T. O'Reilly, S.F. O'Neil, and T.R. Goff. 2016. Technical Review of Utility and Costs of the Atlantic Salmon and Atlantic Whitefish Live Gene Banks at the Maritimes Region Biodiversity Facilities. Can. MS Rep. Fish. Aquat. Sci. No. 3104, 40 p.+App.

Living Gene Banks (LGBs) are extensions of supportive breeding and rearing operations (classical enhancement) that seek to prolong the existence of a species or distinct population segment while minimizing the loss of genetic diversity and fitness. Application of this approach, a first in Canada for an endangered fish population, was initiated for the endangered inner Bay of Fundy Atlantic salmon population, listed as endangered by the Committee on the Status of Endangered Wildlife in Canada; and was being considered for other salmon populations considered at risk but not yet listed, and for the COSEWIC Schedule 1 listed Atlantic whitefish. Use of the Maritimes Region fish culture facilities (biodiversity facilities) for a live gene bank came at a time that classical enhancement of Atlantic salmon had ceased as a policy decision of Fisheries and Oceans. A technical review of live gene banking as a component of population maintenance was asked for by senior management of Fisheries and Oceans and a terms of reference for the review provided. It was to include consideration of the scientific merit, options, and costs. This document addresses the terms of reference and summarizes the origins of the inner Bay of Fundy salmon live gene bank.

RÉSUMÉ

Marshall, T.L., P.G. Amiro, P.T. O'Reilly, S.F. O'Neil, and T.R. Goff. 2016. Examen technique de l'utilité et des coûts des banques de gènes vivants pour le saumon de l'Atlantique et le corégone de l'Atlantique dans les installations de biodiversité de la région des Maritimes. Can. MS Rep. Fish. Aquat. Sci. No. 3104, 40 p.+App.

Les banques de gènes vivants sont une extension des activités de reproduction sélective et d'élevage (mise en valeur traditionnelle des stocks) visant à prolonger l'existence d'une espèce ou d'une population distincte tout en réduisant au minimum la perte de diversité génétique et du succès reproducteur. Cette approche, une première au Canada pour une population de poissons en voie de disparition, a été mise en place pour la population du saumon de l'Atlantique de l'intérieur de la baie de Fundy, qui est considérée comme en voie de disparition par le Comité sur la situation des espèces en péril au Canada (COSEPAC) et inscrite à l'annexe 1 de la *Loi sur les espèces en péril* (LEP). Elle a aussi été envisagée pour d'autres populations de saumons considérées comme en péril, mais pas encore inscrites, ainsi que pour le corégone de l'Atlantique, aussi considéré comme en voie de disparition et inscrit à l'annexe 1 de la LEP. L'utilisation des installations de pisciculture (installations de biodiversité) de la région des Maritimes pour une banque de gènes vivants a commencé à un moment où la mise en valeur traditionnelle des stocks du saumon de l'Atlantique avait cessé en raison d'une décision stratégique de Pêches et Océans Canada (MPO). La haute direction du MPO a demandé la réalisation d'un examen technique de la banque de gènes vivants comme composante de la stabilisation des populations, et un cadre de référence pour l'examen a été fourni. Cet examen devait inclure la prise en compte du mérite scientifique, des options et des coûts. Le présent document porte sur le cadre de référence et résume les origines de la banque de gènes vivants pour le saumon de l'intérieur de la baie de Fundy.

INTRODUCTION

Living Gene Banks (LGBs) are extensions of supportive breeding and rearing operations (classical enhancement) that seek to prolong the existence of a species or distinct population segment while minimizing the loss of genetic diversity and fitness. Live gene banking of anadromous species such as Atlantic salmon (*Salmo salar*) coincides with alarming decreases in populations and their abundance and follows a century or more of fish culture targeted at enhancement for economic gain, as well as supportive breeding and rearing for economic advantage.

The change from enhancement to live gene banking of Atlantic salmon was precautionary and dictated by stark scientific realities, i.e., some wild populations are at very low abundance and progeny resulting from the leveraging of a few individuals through supportive breeding and rearing tend to dominate the population and risk deleterious genetic, dynamic and demographic effects impacting their persistence. The transition to live gene banking was facilitated by the arrival of critically important genetic tools to detect and effect timely pedigree-based matings and the existence of appropriate physical facilities. These facilities were formerly called 'hatcheries', but are now referred to as 'biodiversity facilities', a term reflecting their purpose rather than a part of their operation. However, modifications to these physical plants were required in order to suit the necessary changes in husbandry that focuses on rearing juvenile fish to adult maturity rather than from eggs to juvenile life stages.

This document addresses *Terms of Reference* (Appendix A) developed by Senior Management to provide background, a technical review, and costs of live gene banking initiatives ongoing in the Maritimes Region into 2004. It therefore focuses on measures to sustain the Region's endangered anadromous fish populations (inner Bay of Fundy (iBoF) Atlantic salmon *Salmo salar* and the Atlantic whitefish, *Coregonus huntsmani*); see Appendices B, C and D for details of these listings). A summary "Business Case" later prepared for Senior Management is appended, i.e., Appendix K.

TERMS OF REFERENCE

A. With Respect to the Practice of Live Gene Banking, Indicate:

(A1) Premise That Would Necessitate Live Gene Banking

In the Maritimes Region, iBoF Atlantic salmon and Atlantic whitefish have been designated as "endangered" by the Committee on the Status of Endangered Wildlife in Canada (COSEWIC), (Appendices C and D) and listed on Schedule 1 of the Species at Risk Act (SARA). Outer Bay of Fundy and Southern Upland Atlantic salmon are regionally considered to be equally at risk (Appendix E) and are incorporated in the live gene banking or supportive rearing and captive breeding programs at the Region's three biodiversity facilities: Mactaquac in N.B. and Mersey and Coldbrook in N.S. (Figure 1). Under the Species at Risk Act (SARA), listed species that have received Governor in Council review require specified schedules of actions to be taken by the competent Minister. In the cases of Atlantic whitefish and iBoF Atlantic salmon, recovery teams have been formed that include all interested parties (provincial, federal, First Nations and other stakeholders) and formal plans have been drafted and followed. In the case of both listed species, biodiversity facilities have been indicated as the only viable tool to maintain residual populations, to provide the opportunity for research that is required to determine if recovery is possible and to effect recovery if possible. Technical groups on the recovery teams believe that recovery (conservation efforts that improve species viability in the wild) is technically and biologically feasible.

The concept of using LGBs within the Department was not original to the east coast. LGBs were, at about the same time as the inauguration of the iBoF Salmon Recovery Team, being penned into earlier drafts of Pacific Region's "Wild Salmon Policy" (Anon. 2000). "Principle Five" in that document suggested that "salmon cultivation techniques may be used in strategic intervention to preserve populations at greatest risk of extirpation" i.e.,

> *"Genetic diversity and fitness are threatened by chance events whenever local population abundance declines to critically low levels. Under these circumstances, short-term intervention to increase abundance will be beneficial if the genetic changes that result from the intervention are less detrimental than the genetic changes that occur from continued low abundance. Technologies such as fish culture, broodstock rearing (aquaculture), and gene banking may be used strategically to reduce loss of genetic diversity at critically low abundance. The same technologies can also be effective in re-introducing salmon species to habitat where they formerly occurred. However, strict guidelines are required to ensure that these technologies do not adversely and irreversibly affect the long-term productivity of the conservation unit."*

The purpose of the SARA is to prevent wildlife species from being extirpated or becoming extinct, encourage their persistence, and to provide for the recovery of wildlife species that are extirpated, endangered or threatened as a result of human activity. Therefore, the first objective that the competent Minister must attempt is to *prevent* listed wildlife species from being extirpated or becoming extinct; the second is to provide for the *recovery* of species at risk (where technically, biologically, and perhaps economically feasible).

Failure to address either objective could invoke extirpation, lingering or replacement by an expanding population or habitat occupation by a similar trophic competitor. In the case of iBoF salmon, no such competitor has risen to fill the many niches vacated by salmon some ten years or more ago. While infrequent occurrences of outer Bay of Fundy salmon and aquaculture escapes have been detected in iBoF rivers, no sustaining population of Atlantic salmon has been detected in comprehensive juvenile fish surveys in 2000 and 2002 (Gibson et al. 2003a). Based on observations, recruitment models and current low sea survival, expert opinion associated with genetic and colonization patterns suggest that it is unlikely that a sporadic population of Atlantic salmon will establish in iBoF rivers in the near future. Additionally, iBoF salmon are genetically unique and their replacement via colonization by outer Bay of Fundy or aquaculture salmon would result in the loss of genetic diversity within the species. Therefore, maintenance of the population through live gene banking and researching the cause of their decline remains a worthy objective.

Live Gene Banking of iBoF salmon has to-date provided:

- collections of the last naturally recruited representatives of the iBoF gene pool; and
- valuable animals with which to research the cause of the decline. Because LGB fish have been DNA fingerprinted, they and their offspring, can be tracked, and are hence useful in carrying out scientific ecological experiments.

(A2) Objectives of Live Gene Banking

The goal of the LGB program is to harbour and protect and to provide for the re-establishment and recovery of threatened or endangered populations through maintenance of genetic diversity. Within the Maritime Provinces, representative populations of the iBoF Minas Basin and Chignecto Bay salmon lineages, Atlantic whitefish, and the extremely low Southern Upland and outer Bay of Fundy salmon populations (Appendix E) are being maintained with LGBs. Survival and recovery of such populations can only be achieved by meeting two objectives: (1) maintaining genetic diversity, and (2) minimizing loss of fitness (ability to survive and reproduce) in the wild. The maintenance of genetic diversity is essential to minimize the accumulation of inbreeding over time, and to ensure that future generations of LGB salmon are capable of adapting to the environment upon their release. Minimizing loss of "wild" fitness is necessary to allow released salmon to persist in great enough numbers, and for sufficient generations, to escape an extinction vortex, and to maintain the opportunity for adaptation to changing environmental conditions.

(A3) Level of Diversity Sought

Since the iBoF LGB program includes several populations and at least three different lineages of salmon (also true for Southern Upland and outer Bay of Fundy salmon) among and within population diversity needs to be considered.

Among Population Genetic Diversity:

Small (fewer than 30) to moderate (several hundred) numbers of juvenile salmon have been retrieved from several rivers that were still harbouring declining populations. The physiography and hydrology of these rivers vary in many regards including, gradient, geology, and location within the Bay (Table 1 and Figure 2). In addition to possibly exhibiting local morphological adaptations to these different environments, salmon from these rivers also exhibit different life history strategies. The unique inner Bay of Fundy distinct mitochondrial DNA haplotype indicates a unique evolutionary lineage of origin (Table 1, Verspoor et al. 2002), that is also suggestive of local adaptation. Within the iBoF LGB, many of the unique characteristics can be captured by inclusion of the relatively genetically diverse Big Salmon and Stewiacke river populations. Inclusion of the Gaspereau component ensures that an alternate life history strategy is also represented. It is unknown, but unlikely that any of the other inner Bay populations now being maintained in captivity represent important evolutionarily lineages of salmon. This is because their small adult population sizes and their limited genetic diversities suggest populations that have experienced genetic bottlenecks which have resulted in small effective population sizes.

Within Population Diversity:

On the basis of an individual population, maintaining a large effective population size and genetic diversity is important in order to minimize the accumulation of inbreeding and deleterious recessive alleles thereby retaining the future adaptability of populations in response to changing conditions. Calculations based on fundamental genetic principles indicate that in a randomly mating population consisting of 25 males and 25 females, where each parent contributes an equal number of offspring, inbreeding will accumulate at a modest rate of 1% per generation (Hallerman 2003). Reflecting on this and the observation that the effective population size will probably be somewhat smaller than the census population size, conservation geneticists generally recommend that small captive population sizes be maintained at a minimum of 100-200 individuals, providing that certain breeding recommendations are adhered to (Kincaid 1983; Allendorf and Ryman 1987). Recent large scale, replicated empirical studies involving invertebrates by Woodworth et al. (2002) further indicate that fitness in small populations may be maximized when random mating populations are on the order of 100-200 individuals; the authors of this study suggest that larger populations may actually experience reductions in fitness due to domestication selection.

Another important consideration is the retention of genetic variation and the ability to adapt to future environmental challenges. Genetic variation is lost at rates inversely proportional to the effective population size. Low frequency alleles are likely to be lost first, followed by more common alleles (Table 2).

Recommended Population Size:

From the perspective of maintaining genetic variation, the population size depends on: (1) the frequency of alleles targeted to be retained (0.5, 0.2, 0.1, etc.), (2) the number of generations the LGB program is expected to run, and (3) the acceptable probability of allele loss. For example, low frequency alleles (0.1) may be important in enabling a portion of a population to persist when exposed to novel virulent pathogens. Alleles at a frequency of 0.01 were not likely to be present in more than a couple of individuals in even the largest iBoF populations, so a reasonable frequency of alleles to target for retention may be 0.1. From Table 2, an effective population size of 50 would likely retain rare alleles present in the founding inner Bay population for at least 10 generations. Therefore, retention of 100-200 unrelated individuals, would appear to be adequate to mitigate small population effects and to retain most of the original genetic diversity of iBoF LGB founder populations.

Currently, the number of individuals collected from the wild (founders), in a given year, from each of the two primary LGB populations (Big Salmon and Stewiacke) are similar to, or slightly more than, that recommended for minimizing small population effects and loss of variation (Table 3). In fact, because many crosses are conducted between salmon collected in different years and between year classes, collection years can be effectively combined, bringing the total number of founders for each of the two primary LGB populations to at least 1,000 individuals, considerably more than the minimum recommended by geneticists. However, extensive genetic analyses using molecular markers and the kinship reconstruction method of Smith et al. (2001) indicate that many of the iBoF founders (within and between years) are related at the full- or half-sib level. Under these conditions, the minimum numbers of recommended breeders would be higher than the value given above for unrelated founders.

After correcting for kinship, it was determined that greater than 200 unrelated founders were collected from each of the two primary LGB rivers (Stewiacke and Big Salmon). However, through the early stages of the LGB program, as approaches were being finalized, there was a loss of some of the original iBoF LGB founder lineages. It is expected that there will continue to be a loss of lineages in the future due to sampling events and early mortality, though procedures have been put in place to limit such loss.

Typically, LGB programs involve large animals that produce few offspring. In such instances, all offspring are tagged at birth, and pedigrees are maintained by keeping records of male and female parents. As long as individuals successfully reproduce, few family lineages are lost over time and as much as 90% of the original genetic diversity can be retained over many generations. Since salmon fry can neither be tagged nor reared separately, pedigrees need to be maintained by sampling a small number (5) of eggs from each cross and reared communally. At a later date, multi-locus genotype data is used to determine parentage thereby maintaining the complete pedigree (discussed further below). Early in the iBoF salmon program, some families were lost due to sampling effects necessitated by the combining of egg lots before embryos were collected (embryos are now obtained prior to combining). Additional losses could occur if none of the five embryos survive through to first spawning.

Detailed analyses that consider the number of founders, the number of unrelated founders, initial loss due to sampling events, ongoing loss due to high early mortality, and the overall efficacy of the mean kinship broodstock management program employed, are being carried out that may allow reduction in the size of the primary LGBs.

Recent observations of populations in the wild indicate that a small amount of gene flow (one migrant per generation) can be very effective at decreasing small population effects and increasing population viability in the wild (Bryant et al. 1999, Westemeier et al. 1998; Madsen et al. 1999; and Vila et al. 2003). However, large amounts of cross breeding could result in outbreeding depression and loss of local adaptation and is therefore not prescribed. In light of this fact and if safe reductions in the size of the two primary LGB populations are required then an option would be to carry out small amounts of prescribed migration between the two primary LGBs or from the other small LGB populations into the primary LGB populations

(A4) Steps Involved in Live Gene Banking and Variations thereof

The LGB concept for maintenance of the Inner Bay of Fundy Atlantic salmon population is based on a combined captive and in-river program. Fish from the two larger rivers in the inner Bay of Fundy, the Stewiacke and Big Salmon rivers, that were known to have the largest remaining Atlantic salmon populations, were originally chosen as representatives of the population based on DNA analysis (Verspoor et al. 2002; Amiro 1987). Salmon from the Gaspereau River, NS, which also genetically typed as iBoF salmon, were also included in the LGB because of their unique phenotypic character and low stock status (Amiro and Jefferson 1996). Subsequently, collections were also made from several other iBoF rivers to contribute to the founder broodstock.

In general the LGB has four key components (Figure 3), with minor exceptions, depending on the stock.

1. Wild juvenile Atlantic salmon are captured and transported to a holding location. In Nova Scotia, parr are taken to the Coldbrook Biodiversity Facility, and in New Brunswick to the Mactaquac Biodiversity Facility. Capture of parr late in the autumn permits collection of the wild fish after a lengthy exposure to the natural environment (the parr have been in the river for a minimum of 18 months after hatching) but bypasses the high marine mortality that is apparent in iBoF stocks.

2. Parr are reared to sexual maturity in freshwater to minimize the complexity and costs in the program and to avoid the diseases that could be a risk rearing salmon in sea cages. Individuals are tissue sampled for genetic analysis and marked for identification purposes to aid in the mating strategy.

3. Sexually mature fish are mated according to a prescribed plan which limits inbreeding depression and is designed to maximize preservation of the genes of separate families (described below). Exposure to the natural environment will affect priority in any mating scheme so that: wild caught individuals have a higher priority than progeny of captive bred fish that have been released for wild exposure as unfed fry and captured at a later stage or progeny of adults released to spawn in the wild which in turn have higher priority than "wild-selected" salmon that were released at later stages (age-0 parr released in the autumn or age-1 smolt) which in turn have a greater priority than animals that have been held in captivity. The maximum possible number of families is reared, in numerical balance, so that common families will not displace rare ones. Once re-introduced into the wild, the natural freshwater and marine habitats utilized by iBoF salmon will determine which families survive.

4. Most fish (>99%) are released into the natural environment, at various stages, with a small portion being retained in the biodiversity facility to protect against the loss of families. The LGB is sustained by capture of fish from the wild, most of which would be LGB products, and in the instance where families are not represented in recaptured fish, their genetic complement is maintained in the program through use of the animals that were retained in captivity.

The "in-river" LGB, which is developed by stocking juveniles or adults from the captive LGBs, is used to minimize non-natural influences in the biodiversity facilities while evaluating survival depending on release stage.

River Release Strategy:

A large portion of fish is being released at an early stage to maximize natural selection. Fish are divided into one of three categories, based on proportionate family representation. Fish with few siblings or those that have not spawned more than once are identified as high or medium priority. Fish are considered lower priority if their genetics are widely represented. The Stewiacke River (Table 4) and Big Salmon River (Table 5) are the principal recipient rivers for high priority fish. In Nova Scotia, low priority fish are released into north Minas Basin rivers (Chiganois, Debert, Folly or Salmon). In New Brunswick, low priority fish have been released into the Petitcodiac or Demoiselle rivers.

Mating of Salmon According to a Mean Kinship Based Program:

Captive broodstock are genotyped at nine microsatellite loci following steps outlined in Figure 4. Resulting genotype information is then used to reconstruct population specific pedigrees (Figure 5). In the case of the founder broodstock, kinship is estimated in the absence of parental genotype data using the approach of Smith et al. (2001); this analysis allows the partial reconstruction of the previous (G-1) generation. For subsequent generation salmon (G1, etc.), parentage is determined by using simple compatibility analyses as described in O'Reilly et al. (1998). The resulting pedigree information and the history of previous spawnings are then used to create mating lists that minimize inbreeding and loss of genetic variation. Starting in 2003, prescribed pair-wise matings were performed following a Mean Kinship broodstock management program (see Figure 5; Ballou and Lacy 1995). This method is acknowledged as being very efficient at minimizing loss of variation and reducing evolutionary change (domestication selection). In order to further reduce domestication selection, the program also includes the "in river" or wild component (see operation 4a, Figure 3), as previously described. Here, the majority of siblings from each family are released into the wild, and spawning preference within families is given to siblings with the most (largest number of accumulated months) exposure to natural selection.

(A5) Precedents for Salmonids and Other Species, in Other Jurisdictions

In the United States, the National fish hatchery system is involved in the recovery of 33 listed species, including the Colorado pikeminnow *(Ptychocheilus lucius)*, Gila topminnow *(Poecitiopsis occidentalis)*, Apache trout *(Oncorhynchus apache)*, Gila trout *(Oncorhynchus gilae)*, pallid sturgeon *(Scaphirhynchus albus)* and shortnose sturgeon *(Acipenser brevirostrum)* (Andreasen and Springer, 2000). In the National Marine Fisheries Service's and the U.S. Fish and Wildlife Service's Draft Recovery Plan for the Gulf of Maine Distinct Population Segment of Atlantic Salmon, *Salmo salar*, captive rearing and gene banking are integral to restoration programs for six of eight listed river populations.

One of the longest running captive breeding programs in the U.S. involves Redfish Lake Sockeye. Because of a large number of barriers to upstream and downstream fish passage on the Snake and Columbia rivers, the number of returning adults to Redfish Lake (located at the head of the Snake River) steadily declined throughout the 1980's, and in 1991, this distinct population segment was listed federally as endangered. This same year, the Idaho Department of Fish and Game began live gene banking remnants of this population in two separate facilities, until fish passage could be improved. In 2002, 257 hatchery-produced adults returned to the system to spawn naturally (Frost et al. 2002). In Norway, declining salmon populations, due to both acid precipitation and introduction of the parasite *Gyrodactylus salaris*, prompted the government to initiated a very large scale gene banking program, involving the establishment of several LGBs and the cryopreservation of milt from dozens of salmon from each of approximately 100 stocks (Gausen 1993).

Examples of genetic conservation programs involving captive rearing of salmonids for the purpose of ongoing and/or future restoration of wild self-sustaining populations.

Species	**Component**	**General comments**	**Reference**
Atlantic salmon	inner Bay of Fundy assemblage	Several river stocks conserved in combination of captive and in river Living Gene Banks, ongoing attempts at cryopreservation of sperm; possible threats are many, much uncertainty, primary cause of decline is known to be high marine mortality although the source of the mortality.	DFO (National Recovery Team for inner Bay of Fundy Atlantic Salmon Populations). (2002). DFO. 2010. Recovery Strategy for the Atlantic salmon (Salmo salar), inner Bay of Fundy populations [Final]. Recovery of Nationally Endangered Wildlife (RENEW). Ottawa, Ontario. 57p
Atlantic salmon	Gulf of Maine distinct population segment	Six river stocks conserved in captive programs in part to "provide a reservoir of diverse genetic material from the DPS to protect from catastrophic losses in the wild"; threats include aquaculture practices, acidified water and associated aluminium toxicity, endocrine disrupting chemicals, poaching, incidental capture, competition with native and non-native species.	National Marine Fisheries Service and U.S. Fish and Wildlife Service. 2004. Draft Recovery Plan for the Gulf of Maine District Population Segment of Atlantic Salmon (*Salmo salar*). National Marine Fisheries Service, Silver Spring, MD.
Atlantic salmon	River populations of Norway, un-assessed evolutionary significance	Over 100 river stocks conserved in a system of cryopreserved Sperm and Living Gene Banks; threats include acid precipitation, *Gyrodactylus salaris*, ecological and genetic effects of escaped farm salmon.	Gausen, D. 1993. The Norwegian Gene Bank Programme for Atlantic salmon (*Salmo salar*). In Genetic Conservation of Salmonid Fishes, edited by J. G. Cloud and H.G. Thorgaard, Plenum Press, New York.
Sockeye salmon	Redfish Lake Sockeye salmon	Redfish Lake salmon conserved in long-term captive breeding program; primary threat is multiple barriers to fish passage on Columbia and Snake rivers.	https://pisces.bpa.gov/release/documents/documentviewer.aspx?pub=a00004664-1.pdf

In the United States, 33 of the 74 national fish hatcheries are involved, in some capacity or another, with conservation efforts for 28 federally and 11 state-listed species. Most of these programs include captive rearing and supplementation of declining populations, or re-introduction of future populations of salmonids. Many other non-salmonid fishes, including numerous species of African Cichlids, several species of North American desert fishes, the razorback sucker, and others, are also being reared in captivity for re-introduction into the wild.

B. With Respect to Live gene Banking in the Maritimes region Indicate:

(B1) Events That Led to its Inauguration

Retrospective analysis of the data indicate that the salmon population in the Stewiacke River, a major contributor to the iBoF population, has been in decline since at least as early as 1983 (Gibson and Amiro 2003). This is consistent with earlier assessments that led to closures in 1984 of the commercial fishery and in 1989 of the recreational fishery. Early assessments interpreted the downturn as a normal aberration up to 1994 (Amiro and Longard 1995); the 1995 assessment noted the lack of effectiveness of smolt stocking to mitigate the downturn (Amiro and Jefferson 1996). The ineffectiveness of hatchery stocking, the dwindling numbers of returns, and concern about the genetic impacts of mating small numbers of adults resulted in the termination of broodstock collections in 1995. The 1996 assessment recommended a review of gene banking based on collections of juveniles and grow out to mature adults (Amiro and Jefferson 1997).

At the June 22, 1998, meeting of the combined Salmon Fishing Area 22 and 23 Zone Management Advisory Committee, an *ad hoc* committee was appointed to develop criteria and proposals for cultured broodstock as a remedial action. The committee consisted of Science personnel (three from DFO, one from the Province of New Brunswick and one from the Atlantic Salmon Federation) and met on October 5, 1998. Based on successful trial releases of cage-reared adults to the Big Salmon River, 1994 to 1996, a proposal was made to adopt a program of supportive breeding based on grow-out of live-captured wild parr to mature adult salmon and associated research. A proposal for supportive breeding and limited gene banking was drafted and circulated within the *ad hoc* committee and the Zone Management Advisory Committee (Appendix F).

In light of the severity of the situation, the New Brunswick provincial representative asked whether more immediate action was necessary. Based on the available juvenile population data, the *ad hoc* committee decided that immediate action was required and parr collections began on October 8, 1998.

The rearing program was integrated into the existing and diminishing enhancement programs now being conducted by Salmon Care in NS, and first at the Canadian Sturgeon Conservation Society operated Saint John Hatchery in NB and later at DFO's Mactaquac facility also in NB, in 1998 and 1999. Collections were initially made in the Stewiacke and Big Salmon rivers and searches in other rivers of the iBoF were made in 1999, 2000 and 2002. Based on the paucity of juvenile salmon in those searches and the implications of small-population-size genetics, the supportive breeding program moved to live gene banking in 2000, i.e., pedigree-based, genetic-diversity driven and domestication-selection-reduced.

This change was formally supported at an inaugural meeting of the iBoF Salmon Genetics Advisory Committee on May 26, 2000 (Appendix G). The Committee consisted of three members from Universities, two who specialized in genetics and one in fisheries management, one post-doctoral fellow in genetics, one graduate student in genetics, two visiting scientists, two invited DFO experts, and a DFO chairperson. The Minutes of the meeting reflect the opinion that a LGB was appropriate for the iBoF salmon case (Appendix G) and commitments were made by the non-DFO members to provide research and operational products to conduct a fully pedigree-based LGB. These products were developed and provided to DFO *gratis*. These methods and programs have been used by the LGB, published by the scientists and offered to other LGB operations worldwide. Another action item from that meeting called for a memo from the Committee to DFO supporting the actions taken to date and confirming that, in this case, live gene banking was an appropriate action for DFO to follow (Appendix H). That Committee became the Genetics and Fish Culture Technical Committee within the Planning Group of the iBoF Salmon Recovery Team.

(B2) Context of Their Compliance with Accepted International Guidelines

In 1990, the North Atlantic Salmon Conservation Organization (NASCO) agreed on "Guidelines for the Establishment and Operation of Salmon Gene Banks CNL (90) 56." These guidelines were primarily developed for cryopreservation of sperm, rather than for a LGB, as is being considered in this document. Some of the considerations are relevant however:

1.1 In establishing a gene bank priority, priority should be given to those stocks which are considered to be particularly vulnerable or vulnerable to loss in order to preserve those genotypes.

1.4 In the event that results of genetic mapping are available, the optimal sampling strategy should be determined on the basis of this information. In particular, the occurrence of sympatric genetically isolated populations may require additional sampling.

5.3 In the event of loss of a natural population, the LGB samples could be used for re-establishment of the population. In the case of total loss, female gametes could be obtained from the nearest neighbouring river with similar ecological conditions. Alternatively, the recently developed techniques of androgenesis in which the nuclear DNA in the egg is inactivated by irradiation could provide a method of producing progeny with paternal genes only. The techniques are still experimental however, and will not be applicable to gene banks established to protect wild stocks until high survival rates are possible.

The iBoF LGB program is closely aligned with these considerations.

(B3) The External/NGO Support for That Specific Activity

Several external (non-DFO) groups have contributed funding or other support to assist with genetic analyses associated with the live gene banking of salmon from the Bay of Fundy. The Big Salmon River Angling Association contributed $5K to assist with genotyping costs of founders of the Big Salmon River LGB population. Parks Canada is contributing $20-$30K a year in support of rearing and $5-10K in support of genetic analyses for live gene banking and associated research for Upper Salmon and Point Wolfe iBoF rivers on an ongoing basis. Considerably greater "in kind" support, in terms of field staff, helicopter time, equipment, etc., associated with the collection of founders and research into alternate LGB management practices, has also been contributed by Parks Canada. Finally, Parks Canada has also provided funding and personnel to assist DFO's early efforts to cryopreserve milt from inner Bay of Fundy salmon. The Atlantic

Salmon Federation has provided over $24K during the 2001-2004 period to assist with genotyping costs associated with either live gene banking directly, or research into the efficacy of alternate LGB practices, on declining outer and inner Bay of Fundy salmon. The Atlantic Salmon Federation has also assisted with "in kind" contributions in terms of labour, equipment, laboratory materials, etc., and prompted a letter of support from the World Wildlife Fund. Local community and aboriginal groups (e,g. Fort Folly First Nation) have provided support in terms of labour, particularly with regard to collections and the in-river component of the program. Supporting letters were also written by the Maritimes Aboriginal Peoples Council and by Parks Canada.

(B4) Facilities Involved, Production Capacity Devoted to Gene Banking or Variations Thereof, General Costs based on 2004 information, Stocks on Hand in and Rationale for Each

Live gene banking is supported by three biodiversity facilities: Mactaquac in New Brunswick, and Coldbrook and Mersey in Nova Scotia (Figure 1).

Mactaquac:

Mactaquac has a large number of production ponds and abundant surface and well water. The Mactaquac facility was constructed and operated under an agreement with New Brunswick Power to support the Atlantic salmon population of the Saint John River system as a result of the loss to production by construction of the Mactaquac Dam. The program delivery at this site has undergone a significant change towards LGB support of the diminished stocks in southwestern New Brunswick. About 25% of this facility's capacity is devoted to the live gene banking of iBoF Atlantic salmon. The remainder is or has been used for supportive breeding of the outer Bay of Fundy Saint John and St. Croix River populations and live gene banking of the Magaguadavic River population. Well water is used to provide biosecurity against diseases not endemic to the inner Bay of Fundy watersheds.

The production capacity devoted to live gene banking of iBoF salmon at Mactaquac can be summarized as follows:

- Seventeen of twenty-seven 7.6m Swede ponds, each with carrying capacity for 300 grilse or 200 large salmon;
- Three of the 27 ponds have been permanently modified for small tanks to accommodate LGB wild parr or smolts on arrival and for spawning; two of the 27 have been modified for research; the remaining five of 27 are allocated for the Saint John River program;
- Currently all LGB fish are spawned twice, requiring five ponds per stock, i.e., capacity would be three stocks in five ponds each, unfed fry or adults only, with two ponds remaining for possible parr or smolt production;
- If acceptable to spawn each brood fish only once, only four ponds per stock would be required for unfed fry or adult production (one each for age-0 parr, smolts, grilse and salmon) i.e., capacity would be four stocks with one pond remaining for parr or smolt production. Depending upon the stock and the proportions maturing as grilse and large salmon, there may be some capacity to retain fish from rare families for a second spawning in the salmon pond;

- Ponds are individually supplied with well water to provide isolation from the endemic furunculosis and parasite threats of the Saint John River water supply and between populations originating in different rivers, and
- The LGB has been free of ‘certifiable’ diseases since 1998. Furunculosis has not been detected recently but in spite of the best precautions, has a moderate probability of being detected in the future. This is because of the proximity of Saint John River water supplied ponds in the same facility. Such an outbreak would likely only have a minor one-year impact on programs and not necessarily involve all stocks, due to pond isolation on groundwater.

Program delivery costs at Mactaquac total $558K Salary (incl. fishway operation) and $165K O&M (incl. fishway operation and $75K external funding) plus Property Management office costs (electrical and maintenance). LGB operating expenses per pond (independent of labour or electrical cost) are as follows:

- $25K per five pond or stock program; i.e. three stocks = $75K total
- $20K per four pond or stock program; i.e. four stocks = $80K total

Facility capacity and costs for the Big Salmon, Black, Upper Salmon, Point Wolfe, outer Bay of Fundy programs and their estimated costs appear in Table 6. Additional details, including program rationale is provided in Appendix I; river distributions appear in Table 5.

Mersey and Coldbrook (as of 2004):

The Mersey and Coldbrook operations are complementary, i.e., their program delivery capacities are distinct and separate. Mersey is a large facility built on land leased from Nova Scotia Power Inc. It has abundant surface water of seasonally variable temperature supplying separately a hatching building where 2 million eggs can be incubated, twenty 7.6m and sixteen 11m ponds capable of producing upwards of 350,000 fall fingerlings and up to 350,000 1-year smolts and several fiberglass tanks put in service as required. The water supply has also proven appropriate for rearing and holding all stages of Atlantic whitefish. Recently improved biosecurity, water supply and other features are expected to permit increased capacity for both rearing of eggs and holding of salmon broodstock.

Coldbrook, by contrast is entirely owned by the Department, has a small footprint and is supplied by cool artesian wells and surface waters of a relatively more constant temperature than that at Mersey. These temperatures have proven ideal for holding and natural rearing of small numbers of adults from wild captured parr, i.e., the basis of a LGB operation. Unlike Mersey however, the capacity and colder summer water temperatures are not suited to the production of juvenile salmon; about 1 million eggs can be incubated, but given the cool water temperatures, fry will not initiate feeding and fish are transferred 1-2 weeks after hatching or released as unfed fry (partial yolk-sac absorption). The facility has 48 fiberglass tanks ranging in size from 2 to 4m in diameter, two 7.6 m ponds, and numerous smaller tanks that can be put to use as needs dictate. The tanks are capable of being supplied with well or surface water.

Approximately 50% of the capacity of the two NS facilities is directed at the LGB for iBoF Atlantic salmon; the remainder is used for live gene banking and supportive breeding of Southern

Upland populations and Atlantic whitefish. Program delivery costs for the combined facilities in 2004-05 are $300K salary; $80K O&M plus perhaps $18K Property Management costs. A breakdown of the facility capacity devoted to each program element, and associated costs is provided in Table 6. The rationale and production numbers for the Stewiacke and seven other stocks appear in Appendix H; river distributions appear in Table 4.

(B5) Understanding the Time Frame over Which the Activity would be maintained

From the Recovery Strategy: "The success of the program should be evaluated annually and the first major assessment of progress towards establishing self-sustaining populations is proposed for 2015, i.e., approximately 15 years (3 generations) since intensive efforts toward population recovery were initiated. Even in the event that the cause of the current decline in iBoF salmon is resolved or ceases to exist over the next few, significant progress towards population self-sustainability should not be expected before 2015, i.e., at least three salmon generations."

In practical terms this would require operation of a supportive effort through the LGBs until 2015 to effect a change in the population in the natural environment.

The proposed major assessment in 2015 will be an opportunity to use the available data to assess the utility of LGBs and any new knowledge about the cause of the decline to predict the likelihood of achieving recovery, with or without the LGBs.

(B6) Benefits and Total Costs

Maintaining NB and NS inner Bay of Fundy salmon in semi-wild LGBs is preventing the imminent extinction of what COSEWIC and others (Verspoor et al. 2002) have identified as an evolutionarily distinct and potentially important component of Atlantic salmon. From a broad species perspective, maintenance of individual components is recognized as important in maintaining the health and likelihood of persistence of the species as a whole. From a more regional perspective, maintenance of original iBoF populations is beneficial because future reintroduction measures are more likely to succeed both initially, and in the long term (e.g., restored salmon are more likely to persist over time) if locally adapted salmon are used over salmon obtained from the nearest viable population, hundreds of coastal kilometres distant. Furthermore, if remaining iBoF salmon are retained, production from distant populations will not have to be commandeered to assist with conservation efforts elsewhere.

From a simplistic economic perspective, the above benefits can be ascribed total costs, based on 2004 information, of approximately $400K (394K) per year (Table 6) comprised of:

- $281K annual salary [$140K (NB) plus $141K (NS)]
- $113K annual O&M [$75K (NB) plus $38K (NS)].

About $40K of the NB costs are contributed by Parks Canada. Salary expenditures comprise about 70% of the total, are principally for full time continuing staff and could be viewed as a core commitment to the entire LGB and supportive rearing programs.

C. With Respect to Live Gene Banking in Nova Scotia, Indicate

(C1) Options for Cut Back and Impact on Conservation

Options for cut-backs and impact on the fledgling LGBs were first considered in March 2000 when lease agreements for the Mersey, Coldbrook and Cobequid facilities were terminated by 'Salmon Care'. Options for DFO were at that time i) operate the three facilities, ii) operate Mersey and Coldbrook; dispose of Cobequid, iii) operate Coldbrook and mothball Mersey, and iv) close all three facilities. Option ii) was chosen in anticipation that DFO species-at-risk (SARCEP) would bear a large portion of the costs (Appendix J). LGB operations and supportive breeding programs expanded in 2000-2002, both with the identified 'need' for greater effort on behalf of the endangered iBoF (listed), and Southern Upland (unlisted) salmon and Atlantic whitefish (listed) populations and access to SARCEP funding. (The iBoF salmon, SU salmon and whitefish had high ranked priorities and each qualified for nearly $250K for research monitoring and LGB facilities). Continuation of efforts in integrated fisheries management and "mitigation" or restoration of acid impacted populations, activities embodied in the 'Salmon Care' program, were gradually dropped in spite of opposition expressed in a public consultation (Marshall et al. 2001) so that Science could focus on the most basic of conservation initiatives, i.e., LGBs and captive broodstock production.

Re-profiling of the Department's species priorities for SARECP funding in 2003-2004, i.e., a drop of approximately 20 "ranks" in priority for each of iBoF salmon and Atlantic whitefish, and complete exclusion of priority for SU salmon dramatically reduced opportunity for SARCEP funding. This resulted in correspondingly increasing pressures on regional Science to realign gene banking operations with precious few risk-managed resources. These moves were in stark contrast to expenditures at the Coldbrook and Mersey biodiversity facilities by DFO Real Property and Assets Management 2001-2004 of nearly $1M in major capital funding for Rust-Out and Occupational Safety and Health issues.

For 2004-2005, Mersey and Coldbrook required $214.3K salary for two full time continuing and one seasonal regular salaried employees (2.75 FTEs) at Mersey and 2 FTEs at Coldbrook. Together there was a projected requirement for the facilities of $90K casual salary (total $304K; Table 6) and $80K O&M. This program was sharply reduced from that of 2003-2004 by concentrating on the captive production of broodstock for natural spawning; live gene banking and pedigreed matings of only the highest priority populations within the inner Bay of Fundy area (similar reductions in SU salmon), elimination of unfunded holdings, and restriction of research to the endangered iBoF population and Atlantic whitefish. Program delivery costs in 2003-04, as a result of re-profiling, was $289K total salary and $74K O&M, i.e., only $78K salary and $74K O&M being unsourced prior to late contributions from DFO SARCEP and the Regional Director General's Office.

Options, for further reducing costs (and programs) in **2005-2006** and beyond, impacts and savings are as follows:

A) Reduce Mersey to a seasonal operation:
- terminate the production of all smolts for release;

- terminate the Atlantic whitefish program; attempt to transfer a small number of broodfish to Coldbrook for holding;
- reduce iBoF salmon fish production;
- multi-task 2 FTE positions during winter shut down;
- shorten the term of the seasonal FTE ,

> save approximately $20K casual salary and $10K O&M.

B) Close and mothball Mersey:
- end all production of juvenile salmon beyond the unfed fry stage;
- limit live gene banking to captive rearing of adults at Coldbrook for release (assumes that freshwater captive-reared adults will successfully naturally spawn; only 0.25 million eggs could be taken for release as unfed fry);
- retain Mersey caretaker (~ $15K per annum) and abide by conditions of lease arrangement with NSPI,

> save approximately $50K casual salary and $45K O&M with 2.75 FTEs ($131K salary) being "affected" and requiring redeployment.

C) Close and mothball Mersey and Coldbrook:
- terminate all salmon live gene banking and whitefish efforts in Nova Scotia;
- invite application to the Minister for special permission to freely move iBoF gene bank salmon (incl. stream reared within the Bay of Fundy eco-region to Mactaquac (disease regulations/provincial boundary currently inhibits such a move), and as a consequence
- likely eliminate the potential of live gene banking Southern Upland stocks because of (i) difficulty in moving fish from an Atlantic eco-region across provincial boundary to Mactaquac and (ii) the fact that Mactaquac is intended to mitigate lost salmon production on the Saint John River system,

> save $60K casual salary and $80K O&M (retain caretaker at Mersey and Coldbrook at cost of approx. $30K salary; n/c potential cost of $10-15K for transporting Nova Scotia populations to Mactaquac) with 4.75 FTEs ($214K salary) being "affected" and requiring redeployment.

(C2) Live Gene Bank Options Available with Closure of One or Both Facilities

The potential for live gene banking of iBoF and Southern Upland salmon as well as Atlantic whitefish are increasingly limited with the closure of Mersey or Mersey and Coldbrook. They can be summarized as follows:

A. Closure of Mersey:
    - continue captive rearing of broodstock at Coldbrook; use existing capacity of Coldbrook to take 0.25 million eggs from captive reared brood fish for hatching and release (necessary) as unfed fry. Captive reared brood fish would need to be released for natural spawning. (does not affect existing captive adult production of iBoF or Southern Upland salmon);
    - hold small inventory of Atlantic whitefish at Coldbrook (production capacity would be minimal) or inquire as to the interest/capacity of NS Agriculture and

Fisheries McGowan Lake Hatchery to assume the role of custodian of Atlantic whitefish (previously not an option);
- expand egg holding capacity of Coldbrook, and
- investigate the possibility of transferring eggs surplus to the 0.25 million to the private sector for rearing to later stage juveniles (highly unlikely because the NS facilities, unlike those in the private sector, are not certified disease-free).

B. Closure of Mersey and Coldbrook:
- seek Ministerial Approval to move iBoF salmon from Nova Scotia to Mactaquac and back again, and
- reduce the Big Salmon River program at Mactaquac (currently lacking any regular funding) to accommodate Stewiacke River (or other). [New Brunswick Department of Natural Resources and Energy has just closed Minto Hatchery where a back-up of pedigreed iBoF salmon were being held.]

Fish Health Concerns Related to Moving Wild Salmon Across the NB-NS Boundary:

A Ministerial approval to move Atlantic salmon within what might be termed an NB-NS inner Bay of Fundy marine eco-region poses disease related concerns. To consider the movement of wild Atlantic salmon from an arguably different eco-region such as the Southern Uplands to the iBoF region presents even greater concerns. Background to those concerns is summarized as follows:

- furunculosis is present in the three major NB watersheds (Saint John, Miramichi, Restigouche) but has not been identified in any iBoF river in either NB or NS and is a relatively rare pathogen in NS watersheds, and
- furunculosis is considered to be a pure salmonid pathogen (hence "*salmonicida*") and only a few cells are needed to transmit the infection (A. MacKinnon, vaccine development biologist, Aquahealth Ltd, pers. comm.).

At Mactaquac:
- all fish are dip-vaccinated (limited preliminary coverage) on entry to the LGB and later injected with vaccine that provides near-full protection for two years. A booster is required for adult broodstock held longer than two years. This is precautionary because even though the LGB ponds are supplied with well water, thereby enabling watershed isolation, these ponds are adjacent to the Saint John program river water supplied ponds;
- immuno-competence follows vaccination after about 400 degree-days (approx 1-2 months depending on water temperature), and
- there has been no outbreak of a notifiable[2] disease (furunculosis included) since the beginning of the LGB in 1998. However, in spite of the best precautions, a positive

[2] Updated footnote: As a member of the OIE (Office International des Epizooties) and the World Trade Organization, Canada is obliged to implement OIE standards for trade purposes, including trade in aquatic and terrestrial animals. Canada may be required to attest, for export purposes, that aquatic animals and their products originate from regions, farms or sites that are free of

furunculosis detection is probable over time because of the proximity of Saint John River surface water supplied ponds. It may occur only once every 10 years or more, would likely only have a minor one-year impact on programs, and not necessarily involve destruction of all ponds or stocks due to pond isolation on groundwater.

Notwithstanding the above, and given that Fish Health Lab testing capability is only at the 90 or 95% confidence limits, there is the possibility of introducing furunculosis into NS and NB watersheds that are currently negative. However, it is more probable that furunculosis will find its way to iBoF rivers from sources other than Mactaquac, e.g., the widespread furunculosis distribution in NB watersheds; occurrence in the aquaculture industry (numerous facility depopulations in 2003 to deal with a drug-resistant strain); documented escapement of aquaculture adults in iBoF rivers and previous and continuing (not just DFO) stocking of NB iBoF watersheds. While introduction of this pathogen to a negative river already struggling against extirpation would possibly have short term effects, experience has shown that in the long term, furunculosis appears to have minor impacts on the Restigouche and Miramichi populations where the disease is ubiquitous.

A greater concern would surround the entry of Atlantic coast stocks into New Brunswick and their return to Nova Scotia. Wild Atlantic coast parr for live gene banking at Mactaquac would require holding in disease free water for several weeks in Nova Scotia while a portion of them or surrogates (frequently brook trout from the same watershed) were tested for notifiables at DFO's Moncton Fish Health Lab. The same testing would again be required on fish destined for return to Nova Scotia, a process which in total has the potential to be "consuming" of small stocks. Sampling for disease typically requires 30 animals per group (60 in total).

Facilities in Nova Scotia that are physically capable of handling some aspects of the Mersey-Coldbrook program are retained by the provincial government and private sector. The Province has two facilities that focus on the production of brook trout and are themselves strapped for resources or ability and expertise to conduct Live Gene Banking. They as well are prone to disease which is transmittable to Atlantic salmon and as such would curtail the potential for the reintroduction of reared fish to the wild. Private hatcheries are usually certified disease-free operations and as such would be unwilling to risk the acceptance of fish from the wild or an uncertified hatchery.

Stakeholder partnering to operate Mersey and Coldbrook facilities has not been fully explored, in part because clients habitually point to the maintenance of hatcheries, stocking and the conductance of live gene banking as being a federal responsibility.

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reportable or **notifiable** diseases. Managed by the Canadian Food Inspection Agency: http://www.inspection.gc.ca/english/anima/aqua/queste.shtml

(C3) Alternative Approaches to Conserving Near-Extirpated Stocks and Costing Estimates

The goal of the LGB program is to prevent wild salmon populations of the iBoF from becoming extinct while maintaining their genetic integrity. Other alternatives for achieving these goals include:

- *cryopreservation of gametes;*
- *transplantation of salmon from distant stocks into inner Bay of Fundy rivers;*
- *maintenance in landlocked environments*;
- *translocation to distant rivers, and*
- *introduction of other species.*

Cryopreservation of gametes:

Much of the remaining genetic variation in small threatened populations can be conserved by sampling and cryopreserving sperm or milt from an adequate number of males (Allendorf and Phelps 1981). Genetic material stored in this way can be preserved for an almost unlimited period, but new individuals cannot of course, be created using salmon sperm alone. Thorgaard and Cloud (1993) review two approaches to reconstituting original native populations from cryopreserved sperm.

Firstly, cryopreserved sperm from an extirpated population can be used to fertilise eggs from the nearest extant population. The disadvantages of this approach are (1) considerable effort and time are required to reconstitute an approximation of the original native gene pool; two generations of backcrossing F1 to the cryopreserved sperm (three generations in total) are required to produce salmon that are 87.5 % native; (2) some introgression of genetic material from the host population will be unavoidable; (3) maternal genetic material (mitochondrial DNA and possibly, sex linked nuclear DNA) from the original cryopreserved population will be irrevocably lost, and (4) genetic change associated with multiple (at least four) generations of captive rearing (three listed earlier plus a forth required to produce a large number of fertilized eggs) will be incurred in the production of the final generation of juveniles intended for release into wild river habitat.

A second approach discussed by Thorgaard and Cloud (1993) involves producing embryos with all paternal inheritance (androgenesis). Unfertilized eggs obtained from females from a nearby extant donor population would first be irradiated to inactivate the genetic material, and then fertilised using normal cryopreserved sperm from the original native population. Androgenic diploids consisting of DNA solely derived from the original native population would be produced by repressing the first cleavage division. Because such individuals would be homozygous at all loci (all pairs of alleles would be identical), survival and reproduction would likely be greatly reduced. Additional crosses would be necessary to restore heterozygosity and wild fitness. The disadvantages of this approach are (1) considerable effort and time are required to produce outbred populations of individuals (2) genetic change associated with multiple generations of captive rearing will be incurred in the production of a final generation of heterozygous juveniles intended for release into wild river habitat, (3) maternal genetic material (mitochondrial DNA and sex linked nuclear DNA) is lost, and (4) reduced survival of embryos associated with the treatment used to block cleavage.

The ability to preserve salmon embryos for long periods of time would greatly simplify the future recovery of populations, but more importantly, could also mitigate some of the potential risks associated with the restoration of wild salmon runs from cryopreserved sperm, or from multigenerational LGB populations. Unfortunately, no reports of successful cryopreservation of salmon embryos (or eggs) were found. Although salmon embryos do not appear to survive cryopreservation, isolated cells from blastoderms often do; viable offspring could be reconstituted by transplanting such cells into host embryos obtained from an extant salmon population (of course, the transplanted cells would have to contribute to the germ-cell lineage), or by transplanting diploid nuclei to enucleated host eggs (Thorgaard and Cloud 1993). While promising, these techniques are not presently feasible for reconstituting populations, and it remains to be seen whether they will be developed sufficiently in the foreseeable future. However, cryopreserved sperm can also be used to add genetic variation to future depauperate populations, and to minimize genetic changes, such as domestication selection, in captive populations. Much of the genetic variation of the original founder males can be conserved by cryopreserving the milt of males recovered from the wild, and approximately half that of the original founder females by cryopreserving sperm from the first generation (G1) of LGB males. It must be noted that, to date, all attempts to preserve wild Atlantic salmon sperm or embryos in Atlantic Canada by these methods have fallen short of expectations and therefore are extremely risky solutions to maintenance and survival of iBoF salmon or Atlantic whitefish at this time. However, cryopreservation is considered an important research component of these LGB programs that could provide valuable alternatives to future endangered populations.

Transplantation:

Transplantation of distant salmon may be a possible option, but its efficacy, particularly in the inner Bay of Fundy is, at best, uncertain. Although successful instances of seemingly wild self-sustaining populations of salmonids becoming established through intentional introductions of non-local individuals exist e.g., Medway River to LaHave River upstream of Morgans' Falls (Gray 1986), they are rare. It is likely that most instances of failed translocations go unreported, thus distorting the perception of the utility of this management action. In a recent review of patterns of sub-specific anthropogenic introgression in salmon, Utter (2001) reported that "Many studies noted the relative ease of translocating freshwater over anadromous salmonids, and this difference has been related to the more complex adaptations of anadromous populations (e. g., freshwater and marine residence, smoltification, juvenile and adult migration) that obstruct their translocation to conspecifically colonized areas".

More direct information has been provided by tagging studies reported by Ritter (1975). Here he reported on hatchery-reared smolts descended from parents obtained from Northern New Brunswick rivers that were released into native and increasingly distant rivers, including those of the inner Bay of Fundy. Adult return rates were estimated from tag returns from river fisheries and spawning escapements. Not only did Ritter find that return rates declined with increasing coastal distance between recipient and native streams (which he interpreted as evidence for the existence of heritable differences in ocean migration routes among stocks), but recoveries to inner Bay of Fundy Rivers in particular were very low. Similar findings, that is decreased recovery of hatchery reared individuals with increased distance between natal and recipient rivers, were reported by Reisenbichler (1988) for Coho salmon. Reisenbichler (*op cit*) also interpreted these findings to indicate the presence and importance of adaptive differences among stocks.

Importation of an alternative strain of Atlantic salmon into rivers of the inner Bay of Fundy has been attempted at least three times over the past sixty years. No undisputed evidence exists to suggest that success was achieved. Transplantation of Restigouche salmon stock to the Avon River, Maccan River and Apple River occurred in the 1930's to 1950's, Miramichi stock in 1960's and 1970's, and Saint John River (SJR) stock in the 1980's (Gibson et al. 2003). Returns were minimal at best. Earlier reports that these stocks contributed substantially to returns to these rivers (Huntsman 1942) were unquantified and assessment of tagging effects and unmarked introductions have not been analysed. The detection of SJR salmon among iBoF salmon is not currently possible.

Other potential local salmon population sources such as the Annapolis River or Tusket River salmon are believed extirpated; residual fish in those rivers are likely the result of strays or transplantations from Atlantic coast rivers during the 1970's and 1980's. These residual populations are themselves not likely sustainable. This is because recent marine survival rates for distant migrating salmon stocks along the southern range of the Atlantic salmon in both North America and Europe are at record lows. In the Maritimes Region in general, the productivity rates observed for these rivers are below that required for replacement and many are either impacted by dams or acidification due to acid rain and local geology, or both.

Further explanation for this prognosis can be found in the freshwater survival of North American Atlantic salmon of 0.21% (Jessop 1975) to 3.2% (Chadwick 1982) which is substantially less than survival in the marine environment which is generally 1% to 8% for Maritime salmon stocks (Anon. 2004). Therefore, freshwater productivity is generally the limiting factor to population size i.e., population size is a function of freshwater production area and productivity (Chadwick 1985). The reciprocal of marine survival is the required lifetime recruit per spawner for population sustainability e.g., at 2% marine survival 50 smolts per adult lifetime. Because marine survival for iBoF salmon is currently less than 1% at best (Gibson et al. 2003b) and the estimated iBoF recruits (smolt) per spawner is 15 smolts lifetime at 0.25% survival from egg to smolt in the Big Salmon River 1964 to 1968, there is no possibility for sustainability of transplantation to an iBoF river with similar rates. In addition, few repeat-spawning salmon are now found in the population, further reducing the lifetime smolts per spawner and counteracting any compensatory effects on egg to smolt survival.

These data and observations indicate that transplantation of extant populations of Atlantic salmon into rivers of the iBoF under current marine survival conditions will not likely be successful.

Maintenance in landlocked environments:

No reports of the use of intentionally landlocked anadromous populations in restoring wild self-sustaining anadromous runs were observed in the available literature. Potential benefits of such an approach include (1) reduction or elimination of selection for captive conditions, (2) continuation of some form of natural selection to minimize the accumulation of recessive alleles and to maintain predator avoidance behaviour, etc., and (3) continuation of mate choice and associated benefits on the fitness of progeny. Risks include (1) potential loss of the original population due to the possible inability of anadromous iBoF salmon to complete their life cycle (without intervention) in the selected lake environment, (2) introduction of a non-native species into a natural environment, (3) possible selection for (and adaptation to) freshwater conditions in late-life-cycle stages, and either loss of anadromy or some level of reduced marine fitness in later generations.

Translocation:

Exporting iBoF fish to another river for recapture and inclusion in the escapement or LGB program is a variation of transplantation. It has a higher viability than transplantation because it does not rely on freshwater productivity but rather on marine survival. However, current knowledge and thinking suggests that return rates are inversely proportional to distance of transplantation (Ritter 1975). Nonetheless, there are few controlled studies to demonstrate this widely accepted fact. Regardless, any survival back to an export river would provide the opportunity to test the hypothesis concerning migration and provide marine exposed fish for the LGB. Because the LGB is not dependent on the success of this strategy, but success of this strategy would greatly expand the possibilities for population maintenance at reduced costs, it could be considered experimentally.

Translocation of unmarked fish would require a river devoid of natural salmon or a river where virtually all returns are processed and genetic scanning of all adults would be required to select iBoF returns. Translocation of marked fish into an existing population for future removal, where recapture was almost assured may have little impact on the existing population if the host population was stable and the numbers exported were relatively low. However, there are no known rivers that currently meet these criteria.

However, marine survival rates, smolt to adult returns to Atlantic coast rivers, are assumed to be similar to that on the LaHave River, Southern Upland, Nova Scotia, where they have recently been observed to vary between 2 to 4%. Maximum productivity is about 15 smolts per spawner (DFO 2003). Survival to repeat spawning in the LaHave ranged to 24% in the 1980's but is now ranging from 0 to 5%. Hence there is no reason to expect sustainability and more extirpations are expected in the Southern Upland rivers of Nova Scotia (Appendix E). Therefore, although some of these Atlantic coast rivers are now void or soon will be void of Atlantic salmon, even though some have productive salmon habitat, they do not represent viable alternatives for translocation of iBoF salmon.

While salmon survival rates are unknown for the highland rivers of Cape Breton, higher parr densities and proportions of quality habitat suggest that 50 smolts per adult spawning salmon are possible. This may be particularly true for rivers of the Gulf of St. Lawrence where increasing numbers of larger repeat-spawning salmon are now observed. However, there are no known suitable rivers void of salmon and competitors in close proximity to the iBoF donor stock and thus the issue of reciprocal transplantation and or translocation and the research that it might yield is moot.

Introduction of Other Species:

Both rainbow (*Oncorhynchus mykiss*) and brown (*Salmo trutta*) trout have been introduced into iBoF salmon rivers. Transfers of wild, native origin, rainbow trout have not been permitted into the Maritimes Region for over 25 years. In almost all cases rainbow trout introductions to iBoF rivers were escapes from fish farming and therefore likely of highly domesticated strains. Consequently, the probability of establishing a self-sustaining population of rainbow trout based on farmed escapes is low and few juvenile rainbow trout are encountered in fish surveys in the iBoF. Incidences of rainbow trout are closely associated with fish farms or planned fishery enhancements above impoundments and are common in iBoF rivers of New Brunswick.
The timing of the introduction of brown trout is uncertain but likely dates to the turn of the 19^{th} century. Established populations of brown trout are present in iBoF rivers of Nova Scotia and are particularly noted in Stewiacke and Cornwallis rivers. In almost all cases the size frequency in the population indicates that few are truly anadromous i.e. >2.0kg as found in many anadromous brown trout populations. Fish surveys indicate that slow, deep freshwater in these rivers provide the habitats for these fish and populations are relatively small.

These data and observations indicate that estuarial and local migration, a strategy utilised by many other populations of these species, may not be favourable in the highly turbid conditions found in most iBoF river estuaries and these species are not likely to flourish.

(C4) Potential Alternate Service Delivery

One option to offset a portion of the costs of the NS facilities and other approaches to population preservation is the utilization of surplus production space at the Mersey Biodiversity Facility for the rearing of smolts for sale to the aquaculture industry. Proceeds from the sale have the potential to significantly offset the costs of the LGB and captive rearing conservation initiatives. The approach is consistent with recent Branch discussions re: the promotion of government revenue generation to help offset the cost of government products and services such as is currently championed in Ireland.

The Case (in Brief):

Current rearing regimes for Atlantic salmon strive to reduce the effect of domestication by introducing most of the product to the wild at a very young stage. Consequently about one-half of the Mersey pond capacity could be available for alternate use (e.g., the sixteen 11m ponds). These ponds could yield between 250,000 and possibly 750,000 parr of which 60-75% could make 1-year smolt. The quality is expected to be high. On a sale of 250,000 smolts at $1.30/smolt, (current price approximates $1.75 -$2.00 but is expected to fall), the revenue income would be about $325K. To produce these fish additional to the ongoing program, it is anticipated that costs of the Mersey/Coldbrook operation might increase from $380K to $450K, i.e., the net cost to Canadians of the Nova Scotia portion of the Live Gene Banking and supportive breeding conservation initiatives could be as low as $125K per annum.

The contractual agreements to undertake such a venture may have to be creative, i.e., there is need of insurance, upfront resources to purchase eggs and assurances with industry and the public that smolts for sale were unsubsidized. Scenarios for outsourcing these kinds of difficulties include the initiation of a JPA with an NGO, such as the Atlantic Salmon Federation, which in turn would make contractual arrangements for the provision of smolts to the industry, establish an endowment fund (depository for revenue from smolt sales), purchase insurance and on paper receive LGB fish for release etc. The intent of the most creative arrangement would be to stimulate increased cooperation between the aquaculture industry and conservation interests.

D. With Respect to the Preceding, Provide Analysis, Including Costs, and Recommendations

The operation of the Maritimes Region biodiversity facilities are and will continue to be important to the preservation and potential recovery of threatened and endangered anadromous fish species. Listed or not, all regional salmon populations of mainland Nova Scotia are endangered, as are Atlantic whitefish. Striped bass *Morone saxatilis* are threatened, as may be shortnose sturgeon, Atlantic sturgeon, and the American eel. Each of these species has the potential to benefit from knowledge gained through experimentation in fish culture facilities e.g., partnering with academia, provision of experimental animals or production of progeny.

The Nova Scotia facilities have had about $1M improvements to infrastructure in the past decade. New pipelines, bio-security improvements including two buildings to house brood fish, tanks and oxygenation system, complimented by dedicated full- and part-time staff make them unique in North Atlantic countries and valuable assets with potentially diverse future applications. Complimented by a recognized population geneticist, live gene banking at these facilities is recognized as second to none by North Atlantic salmon producing countries.

- The operation of Mersey and Coldbrook should be continued at the current reduced level ($380K per annum) through 2006-07.
- A business plan should be explored and developed, possibly with the assistance of Aquaculture Management Directorate that, through grow-out and sale of smolts to the aquaculture industry, reduces the overall costs of the NS facilities to less than $200K per annum.
- The plan should be implemented by November of 2005.

The Mactaquac Biodiversity Facility is bound by a nearly 40-year old legal agreement between the Crown and NB Power to provide for the mitigation for salmon lost to NB Power through alteration of river habitat. Infrastructure improvements over the last three years have made it a state-of-the-art facility, well equipped for live gene banking and now, captive production of broodstock.

- Mactaquac culture operations should continue at their current level of about $625K.
- Client, esp. OGD support such as Parks Canada, should be sought at the same ($75K) or greater level.
- Terms of the original agreement with NB Power should be reviewed with a view to transferring more of the costs, e.g., power to operate the facility, from government to the utility.

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TABLES

Table 1. Information being considered in the management of salmon from rivers of the inner Bay of Fundy.

Population and year of sample collection	Bay/ Basin	MtDNA clade 1-3+	Primary life -history charact- eristics	Primary responsible agency	Allele richness (variance)	Gene diversity
Primary Live Gene Bank (sample year)						
Big Salmon R.(2000)	C	no	L	DFO	11.35 (197.04)	0.848
Stewiacke R. (2000)	M	yes	L	DFO	10.61 (187.86)	0.833
Additional Live Gene banks (underway or under consideration)						
Gaspereau R. (1999-2001)	M	yes	D	DFO	8.48 (188.38)	0.756
Great Village .(2000)	M	yes	L	DFO	8.68 (202.35)	0.807
Economy R. (2000)	M	yes	L	DFO	6.81 (135.72)	0.738
Harrington R. (2002)	M	n/a	L	DFO	n/a	n/a
Black R. (2000)	C	no	L	DFO	n/a	n/a
Irish R. (2000)	C	no	L	DFO	n/a	n/a
Upper Salmon R. (2001, 2002)	C	n/a	L	EC	9.27 (140.45)	0.791
Saint John River reference population (2001)		no	D	DFO	11.67	0.850

C-	Chignecto Bay
M-	Minas Basin
MtDNA clade 1-3+	Two related mtDNA haplotypes found at high frequency in mulitple Minas Basin rivers that have not been observed outside the inner Bay of Fundy Verspoor et al. (2002).
L	Local migration, high 1-sea-winter component, high incidence of multiple repeat spawning
D	Distant migration, one and multi-sea-winter components
DFO	Department of Fisheries and Oceans, Canada
EC	Environment Canada
Standardised number of alleles	Estimated by standardising to the smallest sample size observed (N=42) using re-sampling procedures
Gene diversity	Also referred to as effective heterozygosity, was estimated according to Nei (1973).

Table 2. Probabilities of losing an allele due to genetic drift alone for four allelic frequencies, over one, four and ten generations, given different numbers of effective breeders (N_e). Adapted from Tave (1993).

	Probability of loss over a single generation				Probability of loss over four generations				Probability of loss over ten generations			
	Allele frequency				Allele frequency				Allele frequency			
N_e	0.5	0.2	0.1	0.01	0.5	0.2	0.1	0.01	0.5	0.2	0.1	0.01
2	0.0625	0.4096	0.6561	0.9606	0.2275	0.8785	0.9860	0.9999	0.4755	0.9949	0.9999	1
5	0.001	0.1074	0.3487	0.9043	0.0039	0.3651	0.8200	0.9999	0.0098	0.6788	0.9863	1
10	0	0.0115	0.1216	0.8179	0	0.0453	0.4046	0.9989	0.0001	0.1095	0.7265	1
15	0	0.0012	0.0424	0.7397	0	0.0049	0.1591	0.9954	0	0.0123	0.3515	0.9999
20	0	0.0001	0.0148	0.669	0	0.0005	0.0578	0.9880	0	0.0013	0.1383	0.9999
30	0	0	0.0018	0.5471	0	0	0.0072	0.9580	0	0	0.0179	0.9997
50	0	0	0	0.3660	0	0	0.0001	0.8385	0	0	0.0003	0.9895
100	0	0	0	0.1340	0	0	0	0.4375	0	0	0	0.7627
200	0	0	0	0.0180	0	0	0	0.0699	0	0	0	0.1657
400	0	0	0	0.0003	0	0	0	0.0013	0	0	0	0.0032

N_e - effective population size

Table 3. Number and stage of collection of wild salmon (founders) recruited into the primary LGB programs.

Primary Live Gene Bank	Year	Number of juveniles recruited into primary iBoF LGBs	Stage collected	Possible origins of collected juveniles
Big Salmon	1998	268	parr	wild
Big Salmon	1999	216	parr	wild
Big Salmon	2000	313	parr	wild
Big Salmon	2001	304	parr	wild
Big Salmon	2002	454	parr	wild/LGB*
Big Salmon	2003	323	parr/smolt	wild/LGB*
Stewiacke	1998	401	parr	Wild
Stewiacke	1999	189	parr	Wild
Stewiacke	2000	232	parr	Wild
Stewiacke	2001	201	parr	Wild
Stewiacke	2002	4	parr	wild/LGB*
Stewiacke	2003	n/a	n/a	wild/LGB*

* stocked out as unfed fry (BSR) or unmarked fry/0^+ parr (Stewiacke).

Table 4. Numbers of juveniles and adults distributed from the Nova Scotia, iBoF LGB to inner bay rivers in Nova Scotia, 2001-2004. Stock origin is Stewiacke River, except in the case of the Gaspereau River which has its' own stock.

River	Year	Unfed Fry	6-week old fry	Fall parr (0+)	Spring parr (1+)	1yr smolt	2yr smolt	Adult Spawners
Stewiacke	2001	12722	29484	34083				
	2002	24000	42000	88328		6040		
	2003	34750		27000		17613		
	2004*	23314*	20876*			8400*		
	2005*						2500*	
Chiganois	2002	24000	27000	37081				
	2003	42605	46500	32920				
	2004*					8154*		
Debert	2002	10000	27000	45510				
	2003	49806	34000	47805				
	2004*			13000*				
Folly	2002	32000	27000	24570				
	2003	9690	35000	43773				
	2004*			10000*		5054*		
Great Village	2004*					13512*	vaccinated	
	2004*					12300*	un-vaccinated	
Economy	2004*	7000*						
Salmon River, Col.	2002							190
	2003							132
	2004*							
Gaspereau								
	2001			42694		10860		
	2002		7394			16508		
	2003			21726	18600	27422		
	2004*			12000*		11267*		
	2005*					11000*		
Totals	2001	12722	29484	34083				
	2002	90000	130394	195489		22548		190
	2003	136851	115500	173224	18600	45035		132
	2004*	30314*	20876*	35000*		58702*		
	2005*					11000*	2500*	

*preliminary

Table 5. Numbers of juveniles and adults distributed from the Mactaquac, NB LGB to inner bay rivers in New Brunswick, 2001-2004. Stock origin is Big Salmon River unless noted otherwise.

Distributed to:	Year	Unfed Fry	Fall parr (0+)	Spring parr (1+)	1yr smolt	"pre-grilse"	grilse	"MSW" Spawners
Big Salmon R.	2001	185523	77718					
	2002	138682	34062		19725			
	2003	296818	54000	21025	13647			Female 15[2]
	2004	369109	90843	5448	13224			Female 13[2]
Petitcodiac R.								
Pollet R.	2002	56159						
	2003							
Little R.	2002							53
	2003					549[3]		
Demoiselle	2001	16222						
	2002	10080		1078				
	2003							
Weldon Creek	2004	130197						
Point Wolfe R.	2003						286[4]	
	2004						248[4]	32[5]
Upper Salmon R.	2004						nil	
(USR origin)								
Black River	2004	53482						49
(Black R. origin)								
Other								
Minto Hatchery	2001	30000	F1 1000					
	2002		F1 1000					
	2003		F1 1000					
Total	2001	231745	78718					
	2002	204921	35062	1078	19725			53
	2003	296818	55000	21025	13647	549[3]	286[4]	Female 15[2]
	2004	552788	90843	5448	13224		248[4]	94

[2]These females to be priority-selected from wild LGB fish to provide additional females for marine-surviving n`

[3] released in May as 0.5-1.0 kg "pre-grilse"

[4] released in October, 2-3 kg grilse

[5] released in October, 3-4 kg salmon

Table 6. Details of Coldbrook, Mersey and Mactaquac production, and costs[a] based on 2004 information, devoted to each program element.

Facility	**Program**	**Percentage of facility in production**	**Salary ($K)**	**O&M ($K)**
Coldbrook and Mersey	iBoF- Stewiacke	22	66	18**
	iBoF- Gaspereau	15	45	12**
	iBoF- other Minas Basin	10	30	8
	SU-LaHave River	14	42	11
	SU-Medway River	8	24	6
	SU-Gold River	8	24	6
	SU-Tusket	3	9	2
	SU-Eastern shore population	8	30	6
	Eskasoni FN – Indian Brook	1-2	4-5	34
	Atlantic whitefish	10	30	8
TOTAL		**100**	**304**	**80**
Mactaquac[b]	iBoF- Big Salmon	17	95	55
	iBoF- Pointe Wolfe & Upper Salmon	5	28**	15**
	iBoF- other	2	17	5
	Magaguadavic River	3	21	10**
	Aroostook River	8	42**	25**
	Saint John River (Tobique, main river, Nashwaak, Hammond)	65	355	55**
TOTAL		**100**	**558**	**165**

** A portion or all of these costs are recovered from outside agencies.

[a] excludes power and cleaning costs paid by DFO Real Property and Assets Management.

[b] includes approx. $125 K fishway operation total costs including salary and O&M; $165K O&M also includes $35K direct additive cost of LGBs to base cost of $130K for 67 ponds. Total O&M includes $75K in client funds in 2004-2005 of which $20K was for LGB ponds; power costs at Mactaquac exceed $90K of which a large portion is for well water (pumps) to supply LGB ponds.

FIGURES

Figure 1. Maritimes Region biodiversity facilities; clockwise from top, Mactaquac, Coldbrook and Mersey.

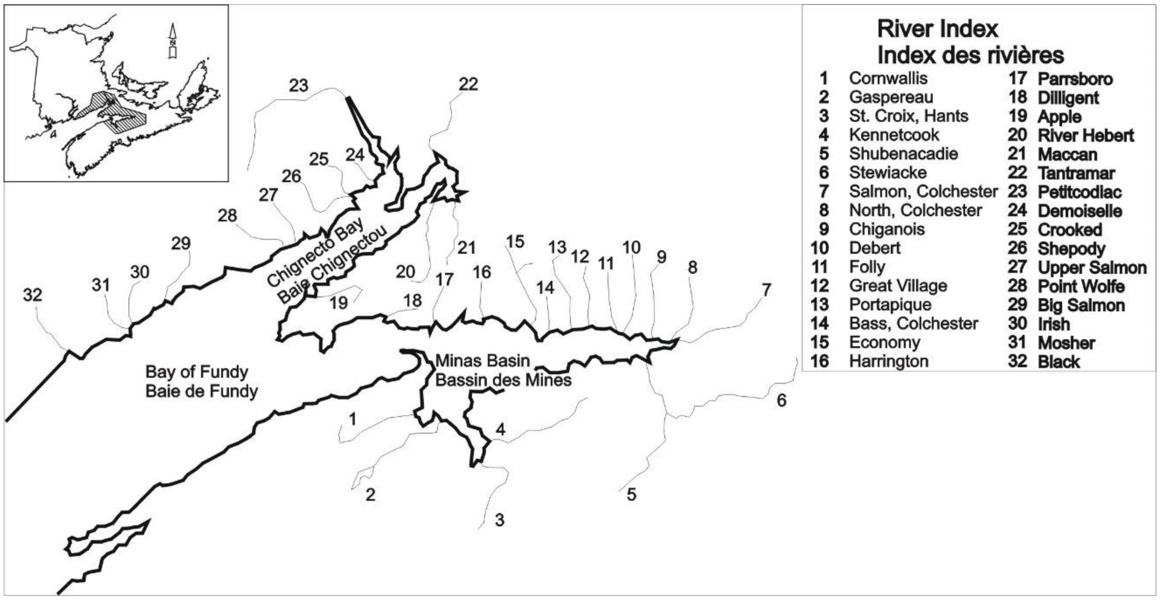

Figure 2. Schematic of 32 inner Bay of Fundy rivers.

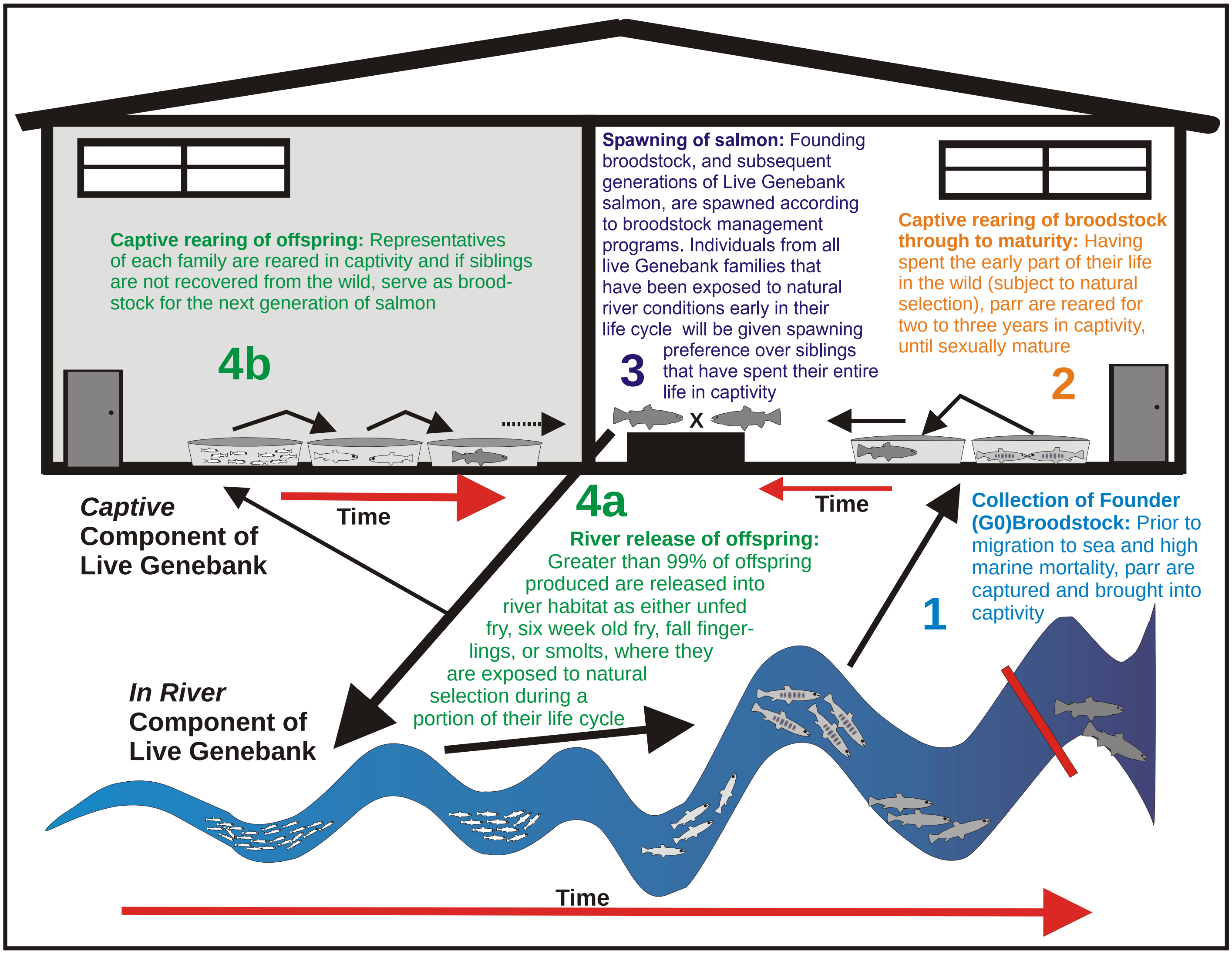

Figure 3. Schematic depicting the inner Bay of Fundy Live Gene Banking program, including "Captive" and "In River" components.

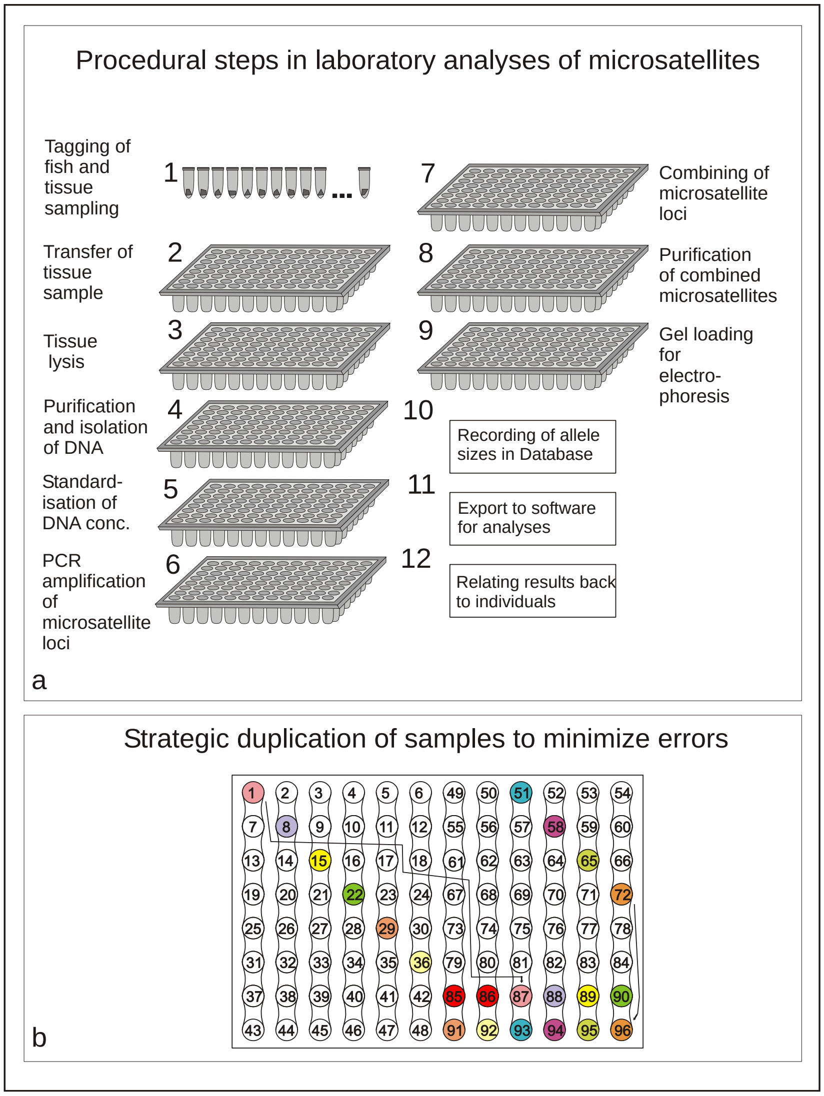

Figure 4. Steps in microsatellite genotyping.
Procedural steps include tissue sampling; tissue lysis; DNA extraction and purification; quantification of DNA concentrations; adjustments of DNA concentrations for down-stream applications; Polymerase Chain Amplification (PCR) of microsatellite loci; combining of PCR products from multiple microsatellite loci; cleanup of PCR products; size separation of alleles via acrylamide electrophoresis; allele size determination; management of allele size information; statistical analyses; and relating results back to relevant samples. Given the number of operations or steps involved, even a very low rate of error per procedural step will result in genotype information incorrectly assigned to a particular sample. Certain errors, including the inadvertent skipping of wells or unintentional duplication of samples into adjacent wells, inversion of strips of tubes (see below), incorrect orientation of PCR plates, etc., are particularly worrisome, as they *could* result in many or all genotype profiles being incorrectly matched to the appropriate sample identifiers. Thus, a single error may result in the incorrect pedigree placement of a large portion of LGB salmon, *and* their subsequent offspring. In this schematic of a typical high-throughput microsatellite analyses utilising a 96 well microplate format, red coloured wells contain cross-gel standards that insure alleles of a particular size, but occurring in different individuals and analysed on different days, are assigned the same values. All other coloured wells represent duplicated samples. Wells 1 and 87, for example, contain tissue from the same individual, as do wells 8 and 88. Most procedural errors, from tissue sampling, through to the uploading of data, that involve multiple consecutive samples of eight or more individuals, will be identified using this approach.

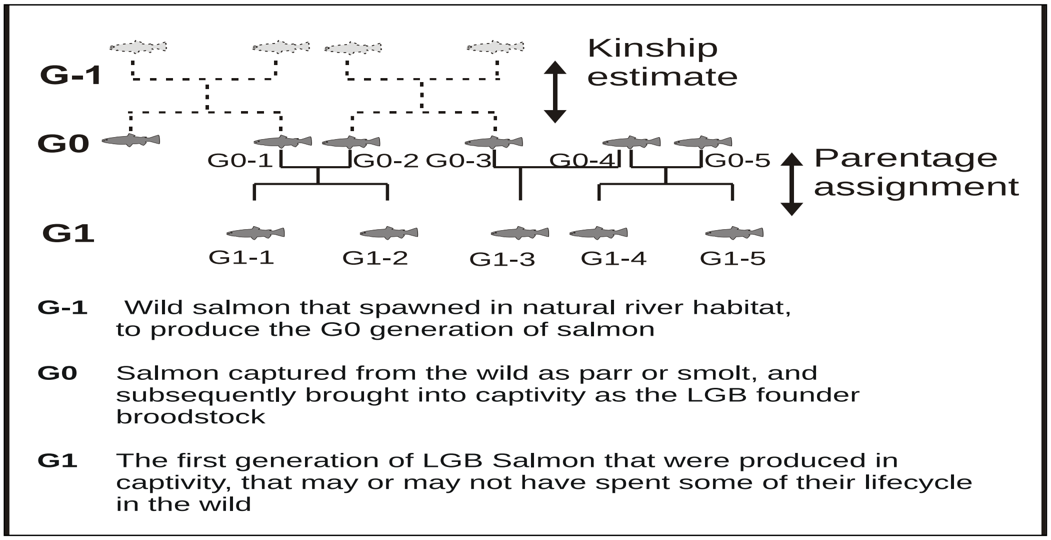

Figure 5. Broodstock management programs that maximise effective population size.

By minimising mean kinship utilise pedigree information from as many generations back in time as possible, to determine average kinship among contributing individuals. Generally, the founding broodstock, when obtained from the wild, are assumed to be unrelated unless there is reason to suspect otherwise. Relationships in the pedigree in all subsequent generations are easily maintained by tagging offspring of prescribed matings between known male and female parents; most such programs typically involve birds or mammals, which are large enough to mark at birth. Atlantic salmon, on the other hand, is a highly fecund species, and collections of dozens, even hundreds of wild juvenile parr, especially from small, declining populations, may be descended from relatively few parents. Genetic analyses of electrofished parr recovered from multiple sites from the Upper Salmon River (an inner Bay drainage), for example, indicate that up to 52% of the 27 juveniles sampled belonged to the five largest full sib families. Assessment of relatedness among the founding populations in such instances is very important in minimising inbreeding in the next and subsequent generations. In the iBoF LGB program, ancestry or relatedness among the founding generation (G0) LGB salmon was estimated using the Kinship method of Smith et al. (2001), which identifies siblings based on their microsatellite genotype profiles without genotype information from their parents, and visual inspection of genotype information of similar full sib families.

Reconstruction of relatedness among the initial, founding broodstock is an ongoing process. Each year, additional parr are obtained from the wild and placed into their respective families by analyses conducted in the context of wild parr recovered in previous years. Kinship assignments of all broodstock are also continuously re-evaluated, as more information becomes available, including data from recently developed microsatellite loci, but also genotype information from new members of full sib family groupings. As additional parr are collected and as the number of clustered full sibs grows, an increasing number of all four "parental" alleles are sampled from a given full sib family at an increasing number of loci surveyed. As a result, a pair of full sibs that by chance inherit mostly different alleles from common parents at multiple loci may be grouped together because of clustering to other sibs with whom they share more alleles.

Implementing pedigree-based broodstock management programs for iBoF Atlantic salmon presents additional challenges- offspring are too small to be physically tagged, and the number of crosses too numerous to keep families separate by rearing in different tanks. Therefore, in subsequent generations, five to ten offspring from each family will be reared communally in captivity, and when large enough to tag, they will be genotyped and pedigreed using parentage analysis.

Juveniles recovered from the wild in 2002, and in all subsequent years, will first be compared to the founder generation (G0) parents using parentage analysis; those matching specified crosses will be placed into the G1 tier of their respective population specific pedigrees. Individuals not matching any sets of parents will be identified as new founders, analysed for kinship in the context of all previously collected founder broodstock according to Smith et al.. 2001, and placed into their respective population specific pedigrees.

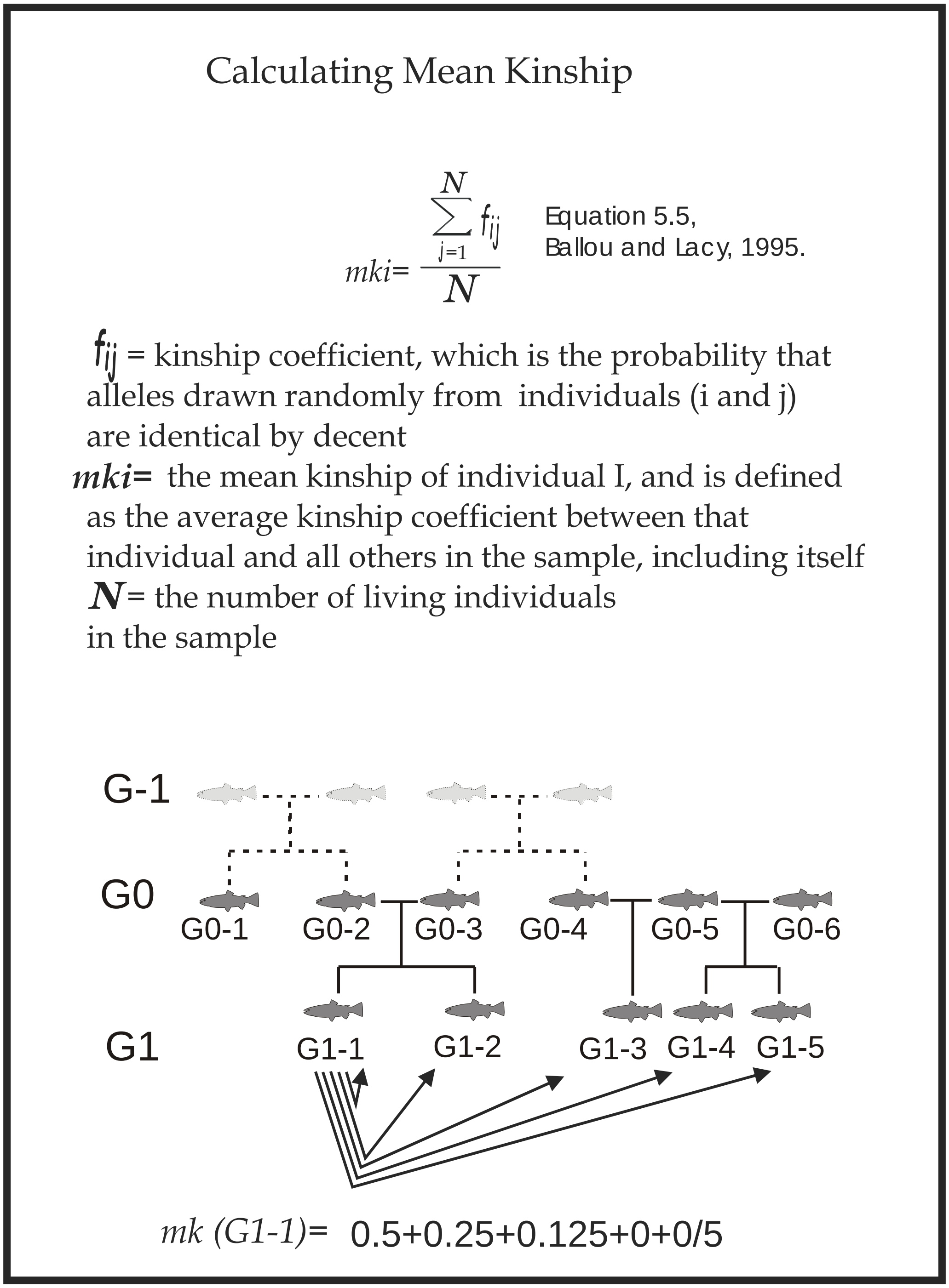

Figure 6. Mean kinship and other similar broodstock management programs.

In the production of one generation from an earlier generation, the gene pool of the first is incompletely sampled (as not all alleles are passed onto offspring, and not all individuals successfully breed), resulting in a change in allele frequencies over time, a process called random genetic drift. The smaller the effective population size, the greater the expected changes in allele frequencies due to drift, and the greater the likelihood of losing alleles, particularly rare alleles. In small captive populations, drift is expected to be the primary cause of loss of genetic variation. Effective population size, and retention of genetic variation, can be maximised by increasing number of breeders, equalising sex ratios, equalising family size, and by minimising variation in population size over time.

When pedigree information is available, however, more efficient methods for conserving genetic variation can be implemented (Ballou and Lacy, 1995). Individuals that are less related to all others in the population are more likely to carry low frequency alleles. By preferentially breeding such individuals (see below), the likelihood of losing rare or lower frequency alleles is reduced, and gene diversity is maximised. Several mating regimes have been developed that put this principle into practise by first determining the relatedness or co-ancestry of an individual to all others in the population (basically, the number of relatives an individual has in a population and the degree of relatedness to each) and then minimising the mean kinship of the breeding population by ranking potential contributors to the next generation by average relatedness (individuals with low average relatedness are assigned higher mating priority).

Perhaps the most well known and often used approach in the captive rearing of endangered zoological populations of animals is that developed by Ballou and Lacy (1995), in which both the selection of individuals that will contribute to the next generation, and the prescribed crosses (which specific male will be crossed with which specific female) are decided in a single step. An illustration of how this procedure is carried out, utilising the salmon pedigree described in Figure 4, is given above.

First, mean kinship, or the average degree of relatedness between a specific potential breeder and all other potential breeders, including itself, is determined using the co-ancestry coefficient, F(ij), which is defined as the probability that alleles drawn randomly between individuals i and j are identical by descent. The Fij for individual G1-1 and itself is 0.5. To estimate the Fij for G1-1 and G1-2, all common ancestors in the pedigree must be identified. In this example, the only ancestors in common are the immediate parents, G0-2 and G0-3, so the Fij between these two individuals is that which one would expect for full sibs, 0.25. When additional common ancestors exist (for example, the sharing of a great grandparent by G0-2 and G0-3), Fij can be easily calculated from pedigree information using path analysis. Next, Fij is computed for G1-1 and G1-3, G1-1 and G1-4 and G1-1 and G1-5. All Fij values are then summed, and divided by N. This is the Mean Kinship for individual G1-1. The same procedure is then carried out for all other individuals in the population. Males and females are then ranked by Mean Kinship, and the male and female with the lowest Mean Kinship selected to be mated, producing a single offspring for the next generation. Mean Kinship values for all breeding individuals are then recalculated, incorporating the new single offspring produced, and the highest ranked male and female selected once again. This could be the same male and female selected earlier, in which case the breeding pair would be assigned to produce a second offspring. By prioritising matings based on the Mean Kinship, the Mean Kinship of the population is minimised. Once the capacity of the rearing facility is met, the process is terminated. The result of the procedure is (1) a list of which males and females are to be mated, (2) designation of which female is to be mated with which male, and (3) a list of how many offspring to produce from each paired mating.

APPENDICES

APPENDIX A. TECHNICAL REVIEW OF UTILITY AND COSTS OF MARITIMES REGION BIODIVERSITY FACILITIES – TERMS OF REFERENCE

As a result of a telecom chaired by Serge Labonte, Director General, Fisheries, Environment and Biodiversity Science, Ottawa, on June 16, 2004, a Working Group (Denis Rivard, Advisor; D. Meerburg and L. Marshall co-chairs) was appointed to provide background, a technical review and costs of Live Gene banking initiatives ongoing in the Maritimes Region. The Working Group is to complete its task by telecom, email and internal report by August 30, 2004.

TERMS of REFERENCE

A. With respect to the practice of live gene banking indicate the

1. premise that would necessitate live gene banking
2. objectives of live gene banking
3. level of diversity sought
4. steps involved in live gene banking and variations thereof
5. precedents for salmonids, and other species, in other jurisdictions.

B. With respect to live gene banking in the Maritimes Region indicate the

1. events that led to its inauguration
2. context of their compliance with accepted international guidelines
3. the external/NGO support for that specific activity)
4. facilities involved, production capacity devoted to gene banking or variations thereof, general costs, stocks on hand in and rationale for each
5. understanding of the time frame over which the activity would be maintained
6. benefits and total costs.

C. With respect to live gene banking in Nova Scotia, indicate the

1. options for cut back and impact on conservation
2. LGB options available with closure of one or both facilities
3. alternative approaches to conserving near-extirpated stocks and costing estimates
4. potential alternate service delivery.

D. With respect to the preceding, provide analysis, including costs, and recommendations.

Rev DJM June 30/04

APPENDIX B. INNER BAY OF FUNDY ATLANTIC SALMON

The assemblage of Atlantic salmon (*Salmo salar* L.) once found in rivers northeast of the Saint John River in New Brunswick and northeast of the Annapolis River in Nova Scotia (See Figure 2) may have been as high as 40,000 adult fish and has declined 90% or more in abundance since 1989. Salmon populations in 32 of these rivers, termed **inner Bay of Fundy** (iBoF) salmon by Perley (1852), have been noted for their instability (Huntsman 1931a, b). Although the stock historically varied in abundance, the decline since 1989 is more severe and the population is at a lower abundance than previously documented. A mean generation time of 3.65 years is the result of young smolt age, 2.58 years, and early maturity, 1.07 years, relative to adjacent salmon stocks. The once high survival between successive annual spawning that is now rare contributed substantially to population stability. Local migration and distinct genetic profiles distinguishes two iBoF sub-populations separate from adjacent salmon populations.

The phylogenetic origin of iBoF salmon, as interpreted by mitochondrial DNA analysis, suggests a divergent evolutionary history that departs from a simple isolation by distance model. The occurrence of a rare haplotype shared by salmon from Minas Basin and Chignecto Bay rivers, and of a genetic haplotype unique to the Minas Basin, probably reflects the pattern of post-glacial re-colonization of the area c. 18,000 years ago. The Caledonia and Kent hills, where the modern day Big Salmon River is found, was the first deglaciated area (Shaw et al. 2002) and thus probably colonized first. Colonists most likely came from refugial populations on the now submerged George's Bank, opposite the mouth of the Bay of Fundy, rather than from isolated and more distant refugial populations which probably existed in non-glacial rivers south of modern day New York. As the ice retreated further, rivers in the Chignecto Bay and Minas Basis were probably gradually colonized by strays from the Caledonia rivers and, to a lesser extent also the George's Bank refugia, with catchments to the south remaining submerged or glacial until more recently (Shaw et al. 2002).

The divergence of the Minas Basin within the inner Fundy area most probably reflects the fact that, starting at Cape Split, the area appears to have been a single river system before the post-glacial rise in sea level (Shaw et al. 2002). The initial population would have evolved a unique genetic identity within the Fundy salmon group which is reflected in the unique haplotype possessed by the area's populations today. This would account for the mitochondrial data and points to the Minas Basin and Chignecto Bay salmon stocks, also known as iBoF salmon, being two distinct but related "evolutionary significant units" or ESU's, as defined by Waples (1991, 1995). These data, analyses and hypotheses, unpublished at the time, were made available as a supplemental submission (E. Verspoor pers. comm.) to a status report prepared for COSEWIC in June 2000 (Amiro 2003) and reviewed in May 2001 and were subsequently published (Verspoor et al. 2002).

The status of iBoF salmon is indexed from the two largest residual populations, the Stewiacke River in Nova Scotia tributary to the Minas Basin, and Big Salmon River in New Brunswick tributary to Chignecto Bay. Prior to the closing of the fisheries in 1990, commercial and recreational catches in addition to scientific fish counts, were used to estimate spawning escapements. In support of these data, scientific and research monitoring of juvenile salmon in the two largest rivers, as well as in six other rivers of the iBoF, up to the year 1999 confirmed that the decline was extensive within iBoF rivers. A multi-indexing approach recently developed for estimating spawning escapements to the Stewiacke and Big Salmon rivers, which incorporates historic fisheries and more recent widespread scientific surveys, indicates an even greater decline than previously estimated (Gibson and Amiro 2003 and Gibson et al. 2003). These analyses indicated a 90% probability that the population in the Stewiacke River has declined by more than 99.8% in the last 30 years and by more than 95% since the early 1990s. Estimates of the percent decline for the Big Salmon River from the early 1990s ranged between 69.0 and 81.6%, and between 93.4 and 97.6% over the last 30 years.

Data and analyses indicate that population growth of iBoF salmon was more often limited by marine survival than by freshwater production capacity and that recent marine survivals of 0.1% to 1.0% are below that required for replacement of the population. These data point to loss of environmental stability in the marine habitat for iBoF salmon or a catastrophic collapse of the marine habitat and its ability to carry iBoF salmon. There is no evidence for the exact cause for the collapse in marine survival of iBoF salmon in recent years or in previous years. Direct and incidental harvest of salmon in the Bay of Fundy has been legislatively eliminated since 1985 for commercial fisheries and since 1990 for recreational fisheries, before populations were critically low. Impacts of alterations to the freshwater and marine environments associated with forestry, farming, urban development and industrial development may have contributed to chronic loss in productivity. However, an acute loss of recruitment from the marine environment such as that experienced by iBoF salmon has not been coincidentally noted for other anadromous species such as gaspereau (*Alosa pseudoharengus*). Changes in the biological communities and alternative uses of lower summer sea temperature in rare marine habitats known to have been important as marine habitat for local Atlantic salmon have occurred in the recent past and continue as potential threats to recovery of the stock.

Based on the information up to 1999 which was available in June 2000 and reviewed in May 2001, COSEWIC concluded that salmon of the iBoF constituted a nationally significant population that had undergone a decline of over 90% and was in danger of extirpation. This was the first salmon population reviewed by COSEWIC and, despite some data deficiencies and uncertainties associated with causes and threats, the Committee made the decision and asked for a more rapid than usual re-submission of the case. Much of the ancillary data has since been collected and made available for a follow-up review in June 2004.

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APPENDIX C. EXECUTIVE SUMMARY FOR LISTING OF INNER BAY OF FUNDY ATLANTIC SALMON

(Information may be found at:
http://www.sararegistry.gc.ca/species/speciesDetails_e.cfm?sid=672)

INTRODUCTION

Wild anadromous Atlantic salmon (*Salmo salar* L.) of the inner Bay of Fundy (iBoF) have declined 90% or more in abundance to a level that places them at risk of extinction. Although the stock has historically varied in abundance, the decline since 1989 is more severe and the population is at a lower abundance than previously documented. Early maturity, successive annual spawning, local migration and distinct genetic profiles characterize two distinct stocks within the iBoF. Annual recruitment to spawning of iBoF salmon stocks is not correlated with other Atlantic coast salmon populations. Monitoring of juvenile salmon in the two largest rivers, Stewiacke River and Big Salmon River, as well as in six other rivers of the iBoF, has confirmed that the decline is extensive within iBoF rivers. River-specific population extinction has been noted.

DISTRIBUTION

Rivers of the Minas Basin in Nova Scotia and of the Chignecto Bay in Nova Scotia and New Brunswick, as far south as the Black River, New Brunswick, supported at least two distinct stocks. Marine distribution is known to be local to the Bay of Fundy at least until late autumn and unknown for the winter season.

POPULATION

Identity - Based on genetic markers, salmon of the iBoF are comprised of at least two distinct but related populations that represent distinct lineages of Atlantic salmon. Recognition of the distinct phenotypic features of these stocks dates to the 19^{th} century. Collectively these stocks have been treated as a separate management unit since 1989.
Trend –Commercial and recreational catch data provide an indicator of the population trend that is variable. The populations have fallen markedly since the higher catches of the 1970s. Juvenile abundance data confirm the recent population declines.
Size – The population was estimated to have been as many as 40,000 adult salmon in some years, likely less than 500 adult salmon in 1998 and less than 250 in 1999.

HABITAT

Freshwater - Atlantic salmon require clean, cool water free from chemical or organic pollution. While chronic loss of habitat may have occurred over the longer term, there is no evidence of a substantial habitat loss since 1989. Brown trout (*Salmo trutta*), a species with similar habitat requirements, persist and have increased in many locations now vacant of juvenile salmon. These observations support the contention that productive capacity for juvenile salmon remains essentially unchanged since 1989.

Marine – The Bay of Fundy, a highly dynamic ecosystem, has provided essential habitat to support this population of Atlantic salmon. Disturbance to the ecology caused by tidal barriers and their effects on salmon are unknown. Extensive cage rearing of Atlantic salmon has developed in the marine habitat of iBoF salmon since the 1980s. There is some evidence of ecological change in the Bay of Fundy.

BIOLOGY

Inner Bay of Fundy salmon differ from most anadromous Atlantic salmon in their genetic lineage and marine distribution. Variable survival to first spawning places a dependence on the repeat-spawning component for population stability. A reduction in repeat-spawning survival and survival to first spawning placed the stock in a steep decline. Uncertainty of the cause of recruitment failure and for population growth makes predictions concerning the persistence or recovery of the stock unreliable.

LIMITING FACTORS

Population growth of inner Bay of Fundy Atlantic salmon is and has historically been more often limited by marine survival than freshwater productive capacity.

Protection

In Canada, Atlantic salmon are directly protected under the Fisheries Act and because they require clean water they are indirectly protected under the Environment Act. Internationally, Canada is signatory to the North Atlantic Salmon Conservation Organization (NASCO). Since 1990 no fisheries have been permitted to harvest iBoF salmon.

SUMMARY OF TRAITS IMPORTANT IN STATUS LISTING

- phenotypic and genetic distinctiveness,
- small number of reproductive adults (less than 500 adults),
- steep decline in population abundance in index rivers (≈90% in ten years),
- under-distribution of juvenile salmon in freshwater habitat (79% of sampling locations in the Stewiacke River were void of age-0^+ parr in 1999, up from 0% in 1984),
- persistent low survival from smolt to first spawning compared to historic measures,
- decreased longevity (Big Salmon River aging data 1964 - 1972 compared to the Stewiacke aging data 1992 – 1995),
- absence of any river population greater than the conservation requirement from which re-colonization of salmon populations from extirpated iBoF rivers might occur naturally.

APPENDIX D. ATLANTIC WHITEFISH LISTING

(Atlantic whitefish information may be found at:
http://www.sararegistry.gc.ca/species/speciesDetails_e.cfm?sid=64)

Atlantic Whitefish

Previous names: Acadian Whitefish

Scientific name:	*Coregonus huntsmani*
Taxonomic group:	Fishes (freshwater)
Range:	NS
Status under SARA*:	Endangered, on Schedule 1
Last COSEWIC designation:**	Endangered (November 2000)

*SARA: The Species at Risk Act
**COSEWIC: The Committee on the Status of Endangered Wildlife in Canada

Quick Links: | Photo | Description | Distribution and Population | Habitat | Biology | Threats | Protection | National Recovery Program |

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Description
The Atlantic Whitefish is a salmon-like fish with a black, dark green or blue back, a white underbelly, and silver coloured sides. This species is anadromous in parts of its Canadian range. This means that it lives in the ocean and spawns in freshwater. It is different from most other salmon-like fish because it has larger scales. Unlike the lake whitefish (*Coregonus clupeaformis*), which is also found in the Maritimes, this species has a mouth at the end of its snout instead of under the head. In addition, it has more vertebrae (> 64) and more lateral line scales (>90) than the lake whitefish.

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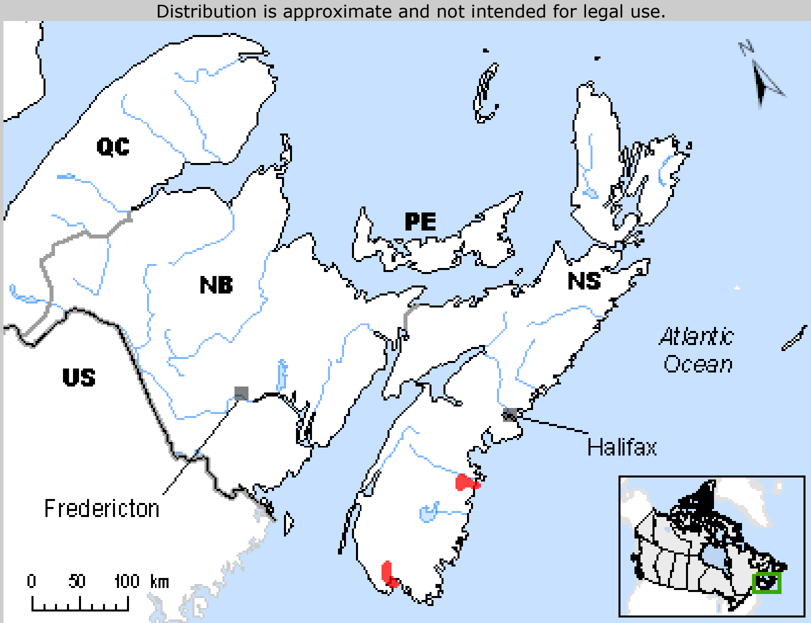
Canadian Distribution of the Atlantic Whitefish (shown in red) [1,2]

Distribution is approximate and not intended for legal use.



[1]Author: Canadian Wildlife Service, 2004

[2]Data Sources: The main source of information and data is the COSEWIC Status Report. In many cases additional data sources were used; a complete list will be available in the future.

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Distribution and Population

The Atlantic Whitefish is an endemic species in Canada, meaning that it is unique to this country. It is found only in the Tusket and Petite Rivière watersheds in southern Nova Scotia.

There are no estimates of population size available for the Atlantic Whitefish. The population in the Tusket river system has declined in recent decades and a small remnant population may or may not remain. The species has been regularly reported in the Petite Rivière watershed since the late 1800's, although its population size is likely small in these lakes and in the lower Petite Rivière.

Top

Habitat

The specific habitat requirements are largely unknown for the Atlantic Whitefish.

Top

Biology

Little is known about the biology of the Atlantic Whitefish. In one Canadian watershed, the species is a sea-run population. Here it is found in estuary and seawaters in the summer. It likely migrates into freshwater for the winter months to spawn, and returns to the sea in the spring. In the other Canadian watershed in which it occurs, the Atlantic Whitefish population is landlocked, meaning that it spends its entire life in these freshwater lakes. The fish of the sea-run population appear to be larger than those living in freshwater. Atlantic Whitefish seem to be found more often in the surface waters of lakes. Their diet ranges from flying insects to small fish. There is no information on this species' spawning behaviour or early life history stages because young Atlantic Whitefish have never been reported in Canada.

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Threats
The Tusket River dam and its ineffective fish ladders have posed barriers to the migratory Atlantic Whitefish. Parts of this river are also very acidic, which may have affected the species' reproductive ability. The spread of introduced fish predators to the Tusket river (e.g. chain pickerel) appear to have posed significant threats to Atlantic Whitefish. The Petite Rivière watershed is apparently better buffered against threats of acidification. Nonetheless, this threat requires monitoring. The possible spread of introduced fish (e.g. brook trout, smallmouth bass) in the Petite Rivière watershed poses significant threats. Poaching, incidental fishing, and the restricted distribution of the animals also limit the recovery of the species.

Top

Protection
The Atlantic Whitefish is protected under the federal *Species at Risk Act* (SARA). More information about SARA, including how it protects individual species, is available in the Species at Risk Act: A Guide.

The Nova Scotia Fishery Regulations under the federal Fisheries Act have since 1970 prohibited the taking of Atlantic Whitefish from all waters of the province by any method at any time of the year. The species' habitat is also protected by provincial legislation.

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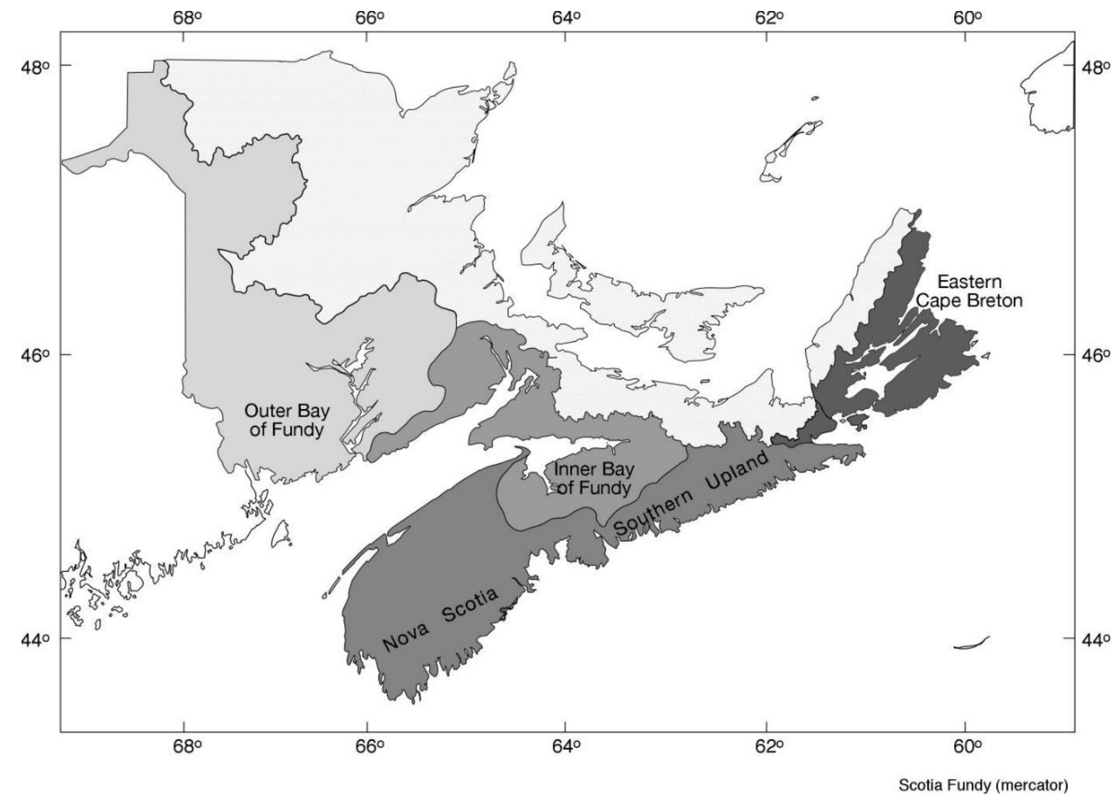
APPENDIX E. CONSERVATION CONCERNS FOR MARITIMES REGION ATLANTIC SALMON (*SALMO SALAR*).

Contact: Summary prepared by T.L. Marshall, now retired but formerly, Manager, Diadromous Fish Division, DFO, Bedford Institute of Oceanography, Dartmouth, NS.

Introduction:

Maritimes Region salmon populations may be considered to consist of four designatable units, those of the inner Bay of Fundy (COSEWIC endangered May 2001), outer Bay of Fundy, Southern Upland and eastern Cape Breton.



Proposed Designatable Units for Atlantic salmon of Maritimes Region

Salmon of the Southern Upland have been most heavily impacted by acid rain and declining marine survival. In 1980, 34 rivers (54%) of the 65 rivers in the Southern Upland fell below pH 5.1 and ranged from unstable to toxic for the production of salmon (DFO 2000). Fourteen of the 34 had mean annual pH <4.7 and were known to have lost their salmon populations; the other 20 rivers had mean annual pH levels of 4.7-5.0, were partially impacted by acidification, but with hatchery intervention maintained low populations of salmon. The continuing impact of acidification and now, decadal downturn in marine survival (DFO 2003; 2004) threaten the persistence of many of the remaining populations, several of which have now been admitted to the Region's living gene bank program. With a few minor exceptions exploitation of these populations is forbidden.

Outer Bay populations are bounded by listed "endangered" populations to both the east (inner Bay) and west (USA-listed Gulf of Maine Distinct Population Segment). Theses populations have been declining over the last 15 years or more (DFO 2003; 2004) in part because of anthropogenic impacts associated with agriculture, urbanization, industrialization, power generation on the main stem of larger rivers and, especially the recent downturn in marine survival. Two likely extirpated populations have been directly impacted by the adjacent "Fundy Isles" aquaculture industry. All fisheries have been closed for several years.

Eastern Cape Breton populations are considerably less affected by anthropogenic impacts but have experienced significant declines as a consequence of reduced marine survival (DFO 2003; 2004). Any closure of continuing recreational hook-and-release fisheries could signal their need for additional protection, possibly under SARA.

Summary analyses (DFO 2004) for the Southern Upland populations (SFA 21 and most of SFA 20), outer Bay populations (western part of SFA 23), and Eastern Cape Breton Island (SFA 19), indicate the following:

- *Adult salmon populations* of the *Southern Upland were assessed on the Tusket, LaHave, East River Sheet Harbour and St. Mary's rivers along the Atlantic Coast of mainland Nova Scotia* ***(SFAs 20 21)*** *in 2003. These rivers are "generally of low productivity" and are negatively impacted by acid precipitation. Of those rivers assessed, only the low-acidified LaHave River met conservation requirements – largely because of an increased return rate of two-sea-winter salmon and hatchery supplementation. At best, 42% of requirements were met on the non-acid impacted St. Mary's River. Returns to the partially acidified Tusket River were 10 wild and 106 hatchery salmon, while 16 hatchery salmon and one wild salmon returned to the partially acidified East River Sheet Harbour. Wild salmon returning to Morgan's Falls on the LaHave River and to St. Mary's River are not expected to achieve conservation requirements in 2004. Analyses of electrofishing surveys suggest that the number of extirpated populations has doubled since 1986 and now includes 50% of 65 rivers in the Southern Upland portion of SFAs 20-21. Wild salmon populations are now critically low and remnant populations require alternative conservation actions to maintain their genetic integrity and extend their persistence.*

- *Adult salmon populations of the outer Bay of Fundy* ***(western part of SFA 23)*** *were assessed on the Saint John River at Mactaquac, Nashwaak, Magaguadavic and St. Croix rivers. Respective attainment of conservation requirements were 12, 7, 2 and 0%. Steady declines in these populations have occurred (the St. Croix and possibly the Magaguadavic have been extirpated) even though a variety of supplementation methods has been applied. To circumvent low marine survival and maintain existing genetic integrity for potential recovery of populations upriver of Mactaquac, wild-captured juvenile salmon are being reared to adults for release and natural spawning. Forecasts suggest zero or near zero probability of exceeding conservation requirements in any of the assessed rivers in 2004.*

- *Adult salmon populations were assessed on the Middle, Baddeck, North and Grand rivers in Eastern Cape Breton* ***(SFA 19)*** *in 2003. Conservation requirements were likely to have been met on the Middle and North rivers, but not on the Baddeck and Grand rivers. No hatchery juveniles have been released since 1997 and hatchery-origin returns would not have been expected in any of these rivers after 1999. The probability of exceeding conservation requirements in 2004 is 45% on the North River, 14% on the Middle River and near zero on the Baddeck and Grand rivers.*

Information follows for only the Southern Upland and outer Bay of Fundy salmon populations.

Taxonomic validity

COSEWIC "Guidelines for Recognizing Designatable Units Below the Species Level" (www.cosewic.gc.ca/eng/sct2/sct2_5_e.cfm) identifies units to which status may be assigned below the species level as follows:

1. named subspecies or varieties, or
2. units identified as genetically distinct, or
3. units separated by major range disjunction, or
4. units identified as biogeographically distinct.

The following synthesis would suggest that the Southern Upland and Outer Bay of Fundy designations are consistent with genetic and possibly, biogeographical distinctness.

In an effort to assist with the identification of potential designable units of Atlantic salmon in the Region, the Department of Fisheries and Oceans commissioned an extensive region-wild survey of phylogenetically informative mitochondrial DNA in anadromous populations along the Atlantic coast of North America. In this study, 700 bp of the ND1 locus was sequenced in over 720 individuals, representing 47 rivers, from the Narraguagus in Maine, though to the Middle and Baddeck rivers of Cape Breton. Inspection of the pattern of mitochondrial DNA types reveals obvious discontinuities between inner and outer Bay of Fundy salmon, outer Bay and Southern Upland salmon, and Southern Upland and the more northern populations of Cape Breton (Verspoor et al. 2002; Verspoor, pers. comm.). These boundaries are concordant with previous groupings which have been based on observed phenotypic differences (life history strategies) and marked differences in freshwater and coastal environments. These allele frequency discontinuities, and the phylogenetic relationship among observed mtDNA haplotypes, indicate (1) reduced gene flow between these assemblages of populations, and (2) possible post-glacial colonization of some assemblages of rivers by unique lineages of Atlantic salmon (Verspoor et al. 2002).

Southern Upland populations

Phenotype/ life history

Salmon of the Southern Upland have persisted in at least 65 rivers that are generally low in base cations, and pH. While subtle morphological differences have been demonstrated between Southern Upland and inner Bay populations, no discriminate functional differences have been demonstrated between their morphometrics. However, Southern Upland populations of salmon have persisted by adapting physiological and life history traits suitable for their environments. These adaptive traits are likely both physiological and behavioural. Many physiological and morphometric differences among populations from the outer and inner Bay and the Southern Upland have been noted in the previous ex-situ culture of these populations.

Historically, salmon returned from the North Atlantic and entered Southern Upland rivers in early spring through to the annual summer drought in July, after which some estuarial stranded salmon entered in the fall. These populations took summer refuge in the many lakes and still waters in these low gradient rivers. This is in stark contrast to many late-summer and fall-run populations of northern Nova Scotia. Population monitoring in at least four rivers of the Southern Upland indicates a shared dependence between age one- and two-sea-winter salmon in annual egg deposition (there are few of either maiden three-sea-winter fish or repeat spawners). This is in contrast to northern Nova Scotia populations that have a high dependence on either a dominate age at maturity or on multiple year spawners.

Genetics

While many salmon from nearly all inner Bay of Fundy rivers exhibit a unique lineage of mitochondrial DNA observed nowhere else in the species' North American distribution, many salmon from nearly all Southern Upland rivers studied are characterised by a different unique lineage of mitochondrial DNA. The observation that these unique inner Bay of Fundy mitochondrial DNA types are completely absent from the hundreds of Southern Upland salmon surveyed, while the unique Southern Upland mitochondrial DNA types are completely absent from the hundreds of inner Bay of Fundy salmon studied, indicates highly restricted gene flow both from inner Bay rivers into the Southern Upland, and from Southern Upland rivers into the inner Bay of Fundy. These results should be interpreted as convincing evidence for the genetic distinctiveness of inner Bay and Southern Upland population assemblages.

Outer Bay of Fundy salmon (Saint John) populations

Phenotype/ life history

With one exception, salmon of the outer Bay of Fundy enter rivers mid-June through July and September through October. A significant proportion of salmon of the outer Bay of Fundy first mature after two winters at sea (three-sea-winter maidens are now rare); a low proportion of one- and two-sea-winter fish have a tendency to repeat spawn. Some populations were dominated by the two-sea-winter component (St. Croix and Magaguadavic) while others that persist have a significant one-sea-winter component. All life history strategies include a marine migration to the Labrador Sea.

Within the various populations the sex ratios range from approximate 10% females for the Saint John River upstream of Mactaquac Dam, to 30-40% among downriver populations. One-sea-winter salmon originating upriver of Mactaquac are the largest such fish reported in North America. The "Serpentine" component of the Saint John River population is unique to North America, but like some large river populations in Scandanavia/ Russia in that one-sea-winter fish return in the late fall, overwinter in the estuary and are the first to ascend the river the following spring.

Genetics

Analyses of mitochondrial DNA (mtDNA) from Saint John River salmon and salmon from the Minas Basin and Chignecto Bay also indicate that outer Bay salmon are genetically distinct from populations to the northeast. A unique mtDNA lineage found in almost all inner Bay of Fundy populations surveyed to date (frequencies up to 50%) are completely absent in analyses of over 100 Saint John River salmon obtained from several tributaries (Verspoor et al. 2002; O'Reilly et al., unpublished data). Nested Clade analyses and molecular analyses of variance of patterns of mtDNA variation in the area are consistent with different patterns of post-glacial colonisation between outer and inner *Bay* of Fundy populations (Verspoor et al. 2002).

Several additional molecular genetic studies, which include at least one Outer Bay of Fundy salmon population and context populations to the southwest and to the northeast, have recently been published. King et al. (2001) surveyed variation at 12 neutral microsatellite markers from wild anadromous Saint John River salmon, and from several remaining salmon populations from Maine. Using multi-locus genotype information and genetic assignment tests, fewer than 6% of Maine salmon were incorrectly assigned to Saint John or Miramichi collections. Conversely, less than 10% of Saint John and Miramichi salmon were incorrectly assigned to Maine collections. Molecular analyses of variation also identified significant differences between Maine and New Brunswick samples. This study was subsequently evaluated by the National Research Council, corroborating the interpretation of both the assignment tests and molecular analyses of variation made by King et al. (2001).

Likely Threats

Southern Upland populations

Marine mortality, acidification/calcium depletion of freshwaters, storage dams and impoundments, hydroelectric generation and invasive species.

Outer Bay of Fundy salmon (Saint John)

Marine mortality, storage dams and impoundments, hydroelectric generation, absence of downstream fish passage, invasive species, industrial and municipal discharges to rivers and marine and freshwater aquaculture activities.

Status

The following status of the proposed designable units is based on the LaHave (Southern Upland) and Saint John River [at Mactaquac] (Outer Bay of Fundy) populations. Both are "best case scenarios" and significant indexes of their respective areas. Analyses are applied to wild salmon returns only even though each is supplemented with hatchery products.

Status of Outer Bay and Southern Upland Salmon on the Basis of COSEWIC Criteria.

COSEWIC Quantitative Criteria	**Outer Bay of Fundy Salmon**		**Southern Upland Salmon**	
	Value	Evidence /Reference	Value	Evidence /Reference
A. Declining Total[a] Population **(**[a] designable unit)				
(1) population size reduction that is observed, estimated, inferred, or suspected in the past 10 years or 3 generations, whichever is longer, where the causes of the reduction are clearly reversible AND understood AND ceased. (%)	NA		NA	
(2) population size reduction that is observed, estimated, inferred, or suspected in the past 10 years or 3 generations, whichever is longer, where the reduction or its causes may not have ceased OR may not be understood OR may not be reversible. (%)	90% (12 yr) marine mortality not understood and ongoing	a, b, c, e 2	78% (12 yr) marine mortality not understood & ongoing; acid rain impacts understood but ongoing	a, b, c, e 1, 2
(3) population size reduction that is projected or suspected to be met within the next 10 years or 3 generations, whichever is longer (up to a maximum of 100 years). (%)				
(4) population size reduction that is observed, estimated, inferred, or suspected in the past 10 years or 3 generations, whichever is longer (up to a maximum of 100 years), where the time period includes both the past and the future, AND where the reduction or its causes may not have ceased OR may not be understood OR may not be reversible. (%)				
B. Small Distribution, and Decline or Fluctuation				
1. Extent of occurrence (km^2) **OR** 2 below	~ 35,000		~22,000	
(a) # extant locations	2/13[+] ([+] = small pop no assess-ment)	a, b, c 2	33/65 in 2000	a, b, c 1, 2, 3
(a) severely fragmented? (Y/N)	N		N	

(b) continuing decline in: (Y/N) i) extent of occurrence ii) area of occupancy iii) area, extent and/or quality of habitat iv) number of locations or populations v) number of mature individuals	 Y Y Y Y Y		 Y Y Y Y Y	
(c) extreme fluctuations in one or more of the following: (Y/N) i) extent of occurrence ii) area of occupancy iii) number of locations or populations iv) number of mature individuals	N		N	
2. Area of occupancy (km^2)				
(a) # extant locations				
(a) severely fragmented? (Y/N)				
(b) continuing decline in: (Y/N) i) extent of occurrence ii) area of occupancy iii) area, extent and/or quality of habitat iv) number of locations or populations v) number of mature individuals				
(c) extreme fluctuations in one or more of the following: (Y/N) i) extent of occurrence ii) area of occupancy iii) number of locations or populations iv) number of mature individuals				
C. Small Total[a] Population Size and Decline (**[a]** assemblage)				
Number of mature individuals (2003)	~ 2,500 wild		~2,500 wild	
(1) estimated continuing decline rate: i) % in 5 years or 2 generations ii) % in 10 years or 3 generations	 17 17		 12 12	
(2) continuing decline, observed, projected, or inferred, in numbers of mature individuals and at least one of the following:				

(a) fragmentation i) maximum # of mature individuals in a population (#) ii) all mature individuals in one population (Y/N) (b) extreme fluctuations in the number of mature individuals (Y/N)	~100 wild within 3 generations N N		~100 wild within 3 generations N N	
E. Quantitative Analysis Indicating the probability of extinction in the wild to be: • 20% in 20 years or 5 generations, whichever is longer (Y/N) • 10% in 10 years (Y/N)	Y		Y	

Evidence:

A. Declining Total Population

a) direct observation

b) an index of abundance appropriate for the taxon

c) a decline in area of occupancy, extent of occurrence and/or quality of habitat

d) actual or potential levels of exploitation

e) the effects of introduced taxa, hybridization, pathogens, pollutants, competitors or parasites.

C. Small Total Population Size and Decline

(1) an estimated continuing decline rate of at least:

a) 20% in 5 years or 2 generations

b) 10% in 10 years or 3 generations

(2) continuing decline, observed, projected, or inferred, in numbers of mature individuals and at least one of the following (a-b):

a) fragmentation – population structure in the form of the following:

- no population estimated

References:

DFO. 2000. The effects of acid rain on Atlantic salmon of the Southern Upland of Nova Scotia. DFO Maritimes Regional Habitat Status Report 2000/2E 19p.

DFO. 2003. Atlantic salmon Maritime Provinces Overview for 2002. Gulf and Maritimes Regions Stock Status Report 2003/026 46p.

DFO 2004. Expert opinion on Atlantic salmon of Salmon Fishing Areas (SFAs) 19-23. DFO Maritimes Region Expert Opinion 2004/01.

King, T.L., S.T. Kalinowski., W.B. Schill, A.P. Spidle, and B.A. Lubinski. 2001. Population structure of Atlantic salmon *(Salmo salar L.)*: a range-wide perspective from microsatellite DNA variation. Molecular Ecology 10: 807-821.

NRC. 2002. Genetic status of Atlantic salmon in Maine: Interim Report from the Committee on Atlantic salmon in Maine. The National Academy Press, Washington, D.C.

Verspoor E., M. O'Sullivan, A.L. Arnold., D. Knox, and P.G. Amiro 2002. Restricted matrilineal gene flow and regional differentiation among Atlantic salmon *(Salmo salar L.)* populations within the Bay of Fundy, Eastern Canada. Heredity 89: 465-472.

APPENDIX F. PROPOSAL FOR SUPPLEMENTATION OF INNER BAY OF FUNDY ATLANTIC SALMON

BACKGROUND (1998):

- Inner Bay of Fundy wild Atlantic salmon occupy about 30 rivers in New Brunswick and Nova Scotia have been shown to be different from other Atlantic salmon stocks.
- The Stewiacke and Big Salmon Rivers contain the largest inner Bay of Fundy populations.
- No fisheries have operated since 1990 and escapements (fish surviving to spawn) to all inner Bay of Fundy rivers have been and continue to be below those required to meet conservation.
- The cause of recruitment failure is known to be marine-based.
- The ratio of recruits/spawner was estimated to be below replacement (0.39) for wild salmon from 1989 to 1994 and above replacement (1.26) for hatchery fish during the same time period.
- There was an estimated 3.23 times survivorship advantage for a hatchery cultured egg over a wild deposited egg from 1989 to 1994.
- Supplementation by hatchery stocking of Stewiacke River origin smolts was stopped in 1997 because of the low adult salmon population available for broodstock.
- Supplementation by hatchery smolt grown-out to mature salmon and stocked to the the Big Salmon River was effective in increasing juvenile salmon populations.
- Recent estimates of wild salmon populations for the Big Salmon River and the Stewiacke River indicate that there are not sufficient adult salmon spawners to ensure genetic diversity for a wild captured adult salmon broodstock based supplementation program.
- Due to recruitment failure, high quality salmon rearing habitat is becoming void of juvenile salmon.
- No improvement in recruitment is anticipated offering opportunity for supplementation to utilise the habitat.
- Extraordinary measures are required to prevent the loss of genetic diversity and fitness of critically low populations.
- The possibility of extirpation of inner Bay of Fundy stocks has increased considerably in 1998.

ACTIONS:

- At the June 22, 1998, meeting of the combined Zone 22 and 23 Salmon Management Advisory Committee, an ad-hoc committee was appointed to develop criteria and proposals for cultured broodstock remedial actions.
- In a follow-up to the June meeting and as a result of the new information on the status of the stock, a steering committee of the Scientific component of the Zone 22 and 23 committee proposed remedial supplementation of the stock as an immediate action.
- In light of the present critically low populations and potential time constraints associated with some actions, the Steering committee considered the relative risks of supplementation programs ranging from:
    - broodstock collection
    - wild smolt collection and grow-out to mature adults
    - on-hand hatchery parr for grow-out to mature adults
    - collection of wild parr for grow-out to mature adults
    - cryopreservation of male gametes.

- The Science steering committee recommended that immediate action be sought for the wild parr collection and grow-out option with the provisos that:
    - At least two stocks be utilised, the Stewiacke River and Big Salmon River

- Up to 500 parr be collected from as many locations within each river as possible
- Parr be cultured separately and over-wintered at one facility, preferably the Mersey Fish Culture Station, in order to maximise the proportion that smoltify in 1999
- Culture smolts to maturity at (at least) two sites, preferably land based and in salt water to minimise the potential for loss and infection and to maximise the size and number of eggs at maturity
- Upon maturation and prior to spawning fish be genetically typed to provide maxim genetic diversity at spawning
- Progeny from these fish be distributed to several grow-out strategies ranging from in-stream rearing to hatchery smolt production
- A cryopreservation project be in place by 1999 for at least mature male parr.

TIME LINE AND ASSIGNMENTS:

- October, 1998
  - ⇒ DFO to arrange parr culture site and permits for transfer. - J. Ritter
  - ⇒ Complete parr collections DFO to co-ordinate with NB DNR. - P. Amiro and P. Cronin
- April, 1999
  - ⇒ Arrange grow-out sites
- May, 1999
  - ⇒ Move parr to grow-out sites
- October 1999
  - ⇒ Collect G2 parr
  - ⇒ Collect male gametes
- May, 2000
  - ⇒ Move G2 smolts to grow out sites.
- September, 2000
  - ⇒ Genetically type all stock
  - ⇒ Collect next parr generation (G3)
- November, 2000
  - ⇒ Spawn G1 fish
  - ⇒ Move out G1 green eggs to interest groups
- April - May, 2001
  - ⇒ Stock out fry G1
  - ⇒ Move G3 parr to grow-out sites
- July-August, 2001
  - ⇒ Monitor fish densities from all methods
- September, 2001
  - ⇒ Program review.

BUDGET:

1998

- Collection of parr 0.0K
- Culture of parr $500/mon@ 2mon = $1.0K

1999

- Contract parr culture $500/mon @ 6mon = $3.0K
- Contract grow-out sites = 2 @ $20K = $40.0K

- Contract to collect male gametes = 1mon @ $2.0K
- Contract to maintain frozen gametes 12mon @$100/mon = $1.2K

2000

- Contract parr culture $3.0K
- Contract grow out sites = 2 @$20K = $40.0K
- Contract genetic work 500 at $10 /sample = $5.0K
- Monitor densities?
- Conduct review?
- Trucking? Oct 5, 1998

APPENDIX G. INNER BAY OF FUNDY SALMON GENETICS ADVISORY COMMITTEE

Friday, May 26, 2000 at 13:30 hrs, Ron Trites Board Room

Attendance:

Mr. Peter Amiro, Recording Secretary
Dr. Roger Doyle
Dr. Christophe Herbinger
Mr. Matt Jones
Dr. Ellen Kenchington, Chair
Dr. Patrick O'Reilly
Dr. John Ritter, Invited Speaker
Dr. Chris Taggart
Dr. Eric Verspoor
Dr. Fred Whoriskey, Observer

Agenda:

13:30 Call to order, introduction of the chair and members.
13:40 Statement of the purpose of the committee.
13:50 Review of the proposed agenda.
14:00 Discussion of the phylogenetic divergence of iBoF salmon.

- Would iBoF salmon qualify as a "distinct population segment" based on the available genetic information?
- Is there a need for more analysis or sampling?

14:30 Regardless of the Species at Risk status of the iBoF salmon stock, DFO has initiated a Recovery Program for iBoF salmon:

- With respect to the live gene bank for iBoF salmon (see attachment for summary) is there a need for a pedigree program in the breeding strategy?
- Is an age-based pedigree breeding strategy sufficient?

15:00 Break.

- Is a molecular-based pedigree required?
- What level of detail is required?
- Is there a need to review all recovery actions or strategies with respect to genetic impacts?

16:00 Protocol for tissue sampling.
16:15 Review of advice to IBoF Salmon Steering Committee resulting from this meeting.

- immediate actions
- future actions
- next meeting

Minutes:

Dr. J. Ritter presented and distributed a proposed management structure for a recovery plan for Inner Bay of Fundy salmon. The structure proposes a Steering Committee, a Planning Committee, an Advisory Committee and three Technical Committees: 1) Genetics, 2) Stock and Habitat Monitoring, and 3) Research. Committee members are expected to commit to an initial five-year term. The various members of these committees have not yet been established. However, each of the Technical Committees, including the Genetics Committee, reports to the Planning Committee. In the future, requests for information and advice will come through the Planning Committee and products of our meetings will be sent back to that committee. Dr. Ritter further refined the immediate concerns of the Planning Committee as follows: 1) What needs to be done further so that we can proceed with breeding plans this year? 2) How would we introduce fish into the system? 3) What should the gene banks constitute?, and 4) What are the numbers of hatchery-produced salmon required and where should they be placed? A brief discussion of funding possibilities followed.

Dr. E. Verspoor presented a seminar on the "Phylogenetic Divergence of Atlantic Salmon stocks in Eastern North America" prior to the meeting. The committee concurred with his conclusion that the mtDNA evidence (ND1-16SrRNA haplotypes and sequence data) suggested that salmon from the Inner Bay of Fundy rivers were genetically different from those from Southern Nova Scotia, Miramichi River and Newfoundland rivers as well as European stocks. Salmon from 18 rivers (nine from inner Bay of Fundy rivers and six from other areas) were examined in more detail using an additional RFLP marker and sequence information. Within the Inner Bay of Fundy rivers, salmon with two distinctive ($P<0.05$) lineages were identified, each with a strong corresponding geographical component. One lineage was found in the rivers of the Minas Basin, and the other in the rivers of adjacent Chignecto Bay, both in the upper reaches of the Bay of Fundy. Economy, Portapique, Great Village, Folly, Stewiacke and Gaspereau rivers possessed a unique variant, which was missing from the Big Salmon, Irish, and Black rivers. The boundaries of the Chignecto Bay lineage were not as well resolved as those of the Minas Basin, especially to the west, and included fish from rivers south and west of the Bay as well as from the Saint John River (salmon traditionally classified as "outer Bay of Fundy"). However, even with the low sample sizes in the analyses the salmon from the Chignecto Bay rivers were significantly different from those of the Saint John River at $P=0.11$

The Canadian Oceans Act, passed in 1997, creates a specific obligation to manage marine resources in accordance with the Precautionary Approach. In keeping with a Precautionary Approach, the Committee recommended treatment of the Inner Bay of Fundy salmon stocks as two regional groups, i.e., the Minas Basin group and the Chignecto Bay group with the later separate from the Saint John River and southern Bay of Fundy stocks. The Committee recommended further sampling and genetic characterization of the salmon from Chignecto Bay and rivers south, including the Saint John River. The specific objective of this further study would be to establish conclusively the status of the Chignecto population group as a genetically distinct entity. Dr. Verspoor agreed to provide the committee with a proposal for this research including a cost estimate for RFLP and sequence analyses.

The results of a submission by P. Amiro to the Committee on the Status of Endangered Wildlife in Canada (COSEWIC) to list the status of the inner Bay of Fundy salmon as Endangered were reported. The original proposal included salmon from all of the Inner Bay of Fundy rivers as part of an Evolutionary Significant Unit (ESU). COSEWIC, in its response, has requested that the document be revised to list only individual rivers where sufficient quantitative data was available to support listing. However, this Committee recommended that in light of the Precautionary Approach, in which the absence of definitive information shall not be used as an excuse to avoid protective action, decisive recovery actions should be taken immediately as opposed to further delay pending a COSEWIC evaluation and status. The Committee further recommended that the COSEWIC re-submission be framed around the recognition of the two salmon lineages within the larger inner Bay of Fundy complex.

The Genetics Committee recommended that a letter be drafted for signature by the Steering Committee for the Minister of Fisheries and Oceans. The letter should confirm the genetic uniqueness of the inner Bay of Fundy salmon, the critically low population numbers, the requirement to act proactively under the Precautionary Approach and the impending time line before extinction. P. Amiro agreed to draft this letter with assistance from Dr. R. Doyle.

The Genetics Committee supports the initiation of a Live Gene Bank for inner Bay of Fundy salmon. It was agreed that the objectives of the gene bank were to preserve the maximum genetic contribution from the ancestral stock and to minimize the loss of adaptation to the natural environment while rebuilding the wild stocks. The identification of gene bank recruits will form a critical evaluation tool to gauge the success of various recovery techniques e.g., stocking of green eggs, eyed eggs, fall parr and smolts.

A general discussion of the Live Gene Bank design and operation occurred. It became clear that the optimal design might not be achieved for a number of years and that more information on the genetic diversity of the remaining wild fish is required. The vision of the gene bank at present includes a hatchery component with fish retained for a period of three to five years (as opposed to the closed system used by the Americans). These salmon are naturally multi-year spawners and so retention of several cohorts will allow us to mimic nature by crossing different ages of fish. The gene bank will also include fish that have been reared and released into extirpated rivers. These rivers will then be re-sampled and used to assist in the perpetuation of the broodstock.

The hatchery will separately maintain fish from each of the Minas Basin and Chignecto Bay lineages. Within each of these lineages, fish from the largest remaining populations (Stewiacke and Big Salmon respectively) will be maintained. Secondly, two additional composite population gene bank lines, one Minas Basin and one Chignecto Bay, should be developed from the residual populations of the other rivers in those regions. The Committee recommended individual identity of gene bank broodstock and retention of mature adults for possible re-spawning. New broodstock (e.g., parr) from known wild inner Bay of Fundy origins should be added when possible. It is recommended that further sampling of fish from each of these four categories take place this summer.

A discussion concerning the identification of aquaculture escapees in the broodstock highlighted the difficulty of genetic identification of individual aquaculture escapees. The present broodstock was determined to be of wild source fish, however, they may be F1 hybrids of aquaculture escapees. The Committee recommended that visual observation and scale characteristics be first used to identify adult salmon escapees and that research for a discrimination set of molecular markers be pursued. It was suggested that historic scale samples could be used to characterize the ancestral population. Discussion of this research was deferred until the immediate demands of the fall spawning were met.

Discussion of the enhancement of fish from non-extirpated rivers ensued. The Committee recommended deferring decisions on returning reared fish to source populations, or to rivers with low populations, until both hatchery and wild stocks are genetically characterized. It was recognized that the 2000 fall spawning in the hatchery is fast approaching and that the fish must be tagged quickly and samples analyzed in order for the pedigree of the fish to be determined. The Committee agreed to work together to ensure that this was done within the required timeframe.

Towards this goal, P. Amiro stated that he could have the fish sampled within two to four weeks. M. Jones volunteered some PIT tags to accelerate the process and Dr. C. Herbinger agreed to lend his PIT tag reader for this purpose. P. Amiro was to co-ordinate the collection of these and the sampling of the fish. Dr. Verspoor agreed to extract the DNA and use his existing microsatellite markers (8) to characterize the samples. He felt that his lab could do this within two weeks. P. Amiro is to send the tissue samples to Dr. Verspoor as soon as he can process the fish. Dr. Herbinger agreed to perform a pedigree analysis and family categorization on the data, with a time estimate of two weeks. Dr. Doyle agreed to suggest a breeding program based upon that analysis and the available population genetic and ecological data, with a time estimate of three weeks. It was also agreed that the appropriate technology be transferred through Dr. Verspoor from the Fish Genetics Group, FRS Marine Laboratory, Aberdeen, Scotland to the DFO/MGPL genetics lab at Dalhousie University during the coming year so that future analysis of the broodstock can be done locally. All parties involved will submit a cost estimate for this work to the Committee Chair.

A discussion of the stages at which the fish should be released ensued. Dr. Ritter advised that mortality is approximately 30% from eyed egg to fall fingerling (more numbers were discussed here but I don't have a record). It was generally agreed that ultimately eyed eggs should be released (as well as other stages) but that as the potential outlay from breeders for this year is only about 300,000 eyed larvae, it would be prudent to hold the fish to the fingerling stage before release. In general, the Committee recommended recovery techniques that are based on the least retention time in the hatchery.

The need for an archival storage of DNA was noted. This would involve surveying researchers for existing material (tissue, scales, DNA) and arranging for transportation to a common storage site and construction of an appropriate database. The Committee recommended that someone from the Department of Fisheries and Oceans be tasked to outline the needs for this archive.

A sampling protocol for genetic analysis is required to advise the monitoring committee. The Committee recommended that a draft protocol for sampling be drawn for review. Dr. O'Reilly agreed to prepare a draft protocol for revision by the Committee.

The next meeting was suggested for September or when the pedigree data and analyses are available for discussion. Interim meeting(s) may be necessary and these can be done electronically or by conference call.

The Agenda items are summarized as follows:

- The inner Bay of Fundy salmon would qualify as having at least two distinct population segments, the Minas Basin rivers including Gaspereau River, and the Chignecto Bay rivers including Big Salmon River
- More sampling and molecular analyses are required to further differentiate these regional populations including a search for a diagnostic marker
- A pedigree of the broodstock on hand in the live gene bank is absolutely required to evaluate the usefulness of the program, to filter out obvious foreign salmon, and reduce the possibility of inbreeding
- An age-based pedigree is insufficient to ensure maximum genetic diversity in the live gene bank
- Both mitochondria DNA variant analysis and at least four tetra-nucleotide microsatellite loci (nuclear DNA) are required to fully identify the broodstock held in the live gene bank, and for the assessment of the efficacy of recovery approaches

- All recovery strategies need to be evaluated from both the genetic and the numeric population perspective

Respectfully submitted by
P. Amiro, Recording Secretary
and modified by
Dr. E. Kenchington, Chair

Current Status of the Live Gene Bank (May 26, 2000)

Stewiacke River	There are 300 fish from the 1998 Stewiacke River parr collections in the Cobequid hatchery. The fish were collected from as many areas within the river system as feasible. The fish are on average 0.6 kg in weight, with *few expected to spawn* in 2000. **[Amended May 30, 2000 to a high probability of spawning]**
	About 170 large parr and 150 small parr were collected in 1999 and are being moved from the Cobequid Hatchery to the Coldbrook Hatchery.
Big Salmon River	There are 270 fish from 1998 Big Salmon River parr collections in the Mactaquac hatchery. The fish weigh 1.5 kg on average with many expected to spawn in 2000.
	About 150 parr and 143 fry were collected in October 1999, from the Big Salmon River. Some of these fish will mature in 2001.

APPENDIX H. IMPENDING CONSERVATION CRISES IN INNER BAY OF FUNDY ATLANTIC SALMON

MEMORANDUM

To:	Dr. J. Ritter, INTERIM Chair, Inner Bay of Fundy Planning Committee
From:	Dr. E. Kenchington, Chair, Inner Bay of Fundy genetics technical advisory committee
Subject:	Impending conservation crisis in INNER BAY OF FUNDY Atlantic Salmon
Date:	(uncertain)

On the behalf of the inner Bay of Fundy Genetics Technical Advisory Committee, this letter formally urges immediate action regarding the impending conservation crisis for Atlantic salmon in the inner Bay of Fundy.

There is genetic evidence that salmon populations in the rivers of this region are genetically related to each other but are distinct from other populations of the same species anywhere else in the world. Furthermore, the evidence for the contemporary low population levels is sufficient for the Committee to issue this precautionary warning that some populations are in imminent peril of extinction if action is not taken immediately. If some populations become extinct, then the ability of "the stock" to recover may be forever compromised.

The inner Bay of Fundy Genetics Technical Advisory Committee was recently informed that the submission to the Committee on the Status of Endangered Wildlife in Canada (COSEWIC) to list the status of the inner Bay of Fundy Atlantic salmon as “Endangered” was deferred by a Species Specialist Group of COSEWIC to May 2001. The submission was deferred for what the Committee thinks are defendable but primarily technical reasons. A new submission will address those issues. In the meantime, this Committee would like to support the position that there are two or more genetically distinct stocks within the Inner Bay of Fundy that are at critically low abundance, and at risk of extinction. The window of opportunity to take precautionary action to sustain the stock may be very rapidly closing. The ability to sustain, let alone enhance a recovery, is demonstrably impeded by a lack of commitment to take immediate action. The Committee has been informed that adequate resources for action are dependent on listing the status with the COSEWIC. The Committee advises that if action is deferred until a COSEWIC listing is established, it may be too late to take meaningful action.

The Committee endorses and approves the general commitment of COSEWIC to the conservation of Canadian resources in the spirit of the Rio Earth Summit agreements. However, given the overall level of information and concern about the inner Bay of Fundy salmon, the Committee is of the opinion that further delay (for at least one year) in functionally considering the stocks as endangered may impose greater risk.

The Committee highlights the fact that in 1997 Canada passed the Oceans Act and in doing so fully committed itself and its agencies, to the Precautionary Approach for fisheries management as described in the United Nations Agreement on the Responsible Management of Fisheries. The Precautionary Approach is a specific provision of the Act. The most important and distinctive feature of the Precautionary Approach is that the absence of complete information shall not be considered a justifiable rationale to avoid taking preventative measures when a risk is known to exist.

In view of its explicit mandate and obligations to the conservation of marine resources, and its obligation under the Oceans Act to take preventative measures when data are incomplete but there is a strong perception of risk, the Department of Fisheries and Oceans, Canada has initiated a supportive breeding program for the inner Bay of Fundy salmon. It has done so with marginal funding and without a funding future. The Committee acknowledges that there is much room for error in determining the exact level of risk for the various salmon populations and their future. However, the Committee is more than prepared to be wrong. The consequences of being wrong are minuscule relative to the consequences of being right without remedial action.

The Department of Fisheries and Oceans has been forward looking and proactive in its concern about the inner Bay of Fundy salmon. Thus, the Committee strongly recommends that the Department maintain its credibility and its proactive conservation stance and fully fund the Recovery Program. Otherwise, the opportunity to sustain this unique genetic resource may very well be lost.

Respectfully,

Dr. Ellen Kenchington
Chair, Inner Bay of Fundy Genetics Technical Advisory Committee

Committee Membership:
Dr. R. Doyle, Genetic Computation Ltd., Halifax, Nova Scotia
Dr. C. Herbinger, Marine Gene Probe Laboratory, Dalhousie University
M. Jones, Biology Department, Dalhousie University
Dr. P. O'Reilly, Marine Gene Probe Laboratory, Dalhousie University
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.

APPENDIX I. BIODIVERSITY FACILITIES PROGRAMS, 2003, 2004

At Coldbrook and Mersey:

a) Inner Bay of Fundy Atlantic salmon

Stewiacke River

Rationale: COSEWIC-listed endangered iBoF stock; Recovery Team has determined the LGB program is "essential for recovery"

O&M cost: $19K

Program deliverables: 30,000 unfed fry; 24,000 6-week fry; 8,500 smolts

Space requirements: Mersey, four 7.6m ponds for juveniles; two 7.6m ponds for the 2-year old smolt; and three 7.6 m and one 11m ponds for adults; Coldbrook- 8 fibreglass tanks.

Gaspereau River

Rationale: COSEWIC-listed endangered iBoF stock; Recovery Team has determined the LGB program is "essential for recovery".

Program deliverables: 12,000 age-0 parr; 11,000 smolts.

O&M cost: $11K

Space requirements: Mersey, two 6m ponds for juveniles; Coldbrook- 6 fibreglass tanks.

Other iBoF rivers (Portapique, Economy, Great Village, Debert, Folly):

Rationale: COSEWIC-listed endangered iBoF stock; Recovery Team has determined the LGB program is "essential for recovery".

O&M cost: $8K

Program deliverables: Parr reared to adults with some adults spawned (150,000 eggs)

Space requirements: Mersey- one 11m pond; Coldbrook- two tanks.

Inbreeding experiment related to IBOF LGB: one 11m pond at Mersey.

b) Southern Upland Atlantic salmon

Gold River

Rationale: Stock possibly unique among Southern Upland stocks (additional genetic analysis pending) and at critically low levels.

Program deliverables: LGB program involves rearing parr to adults for release into natal stream to spawn; 100 mature adults to be released into the Gold River.

O&M cost: $8K.

Space requirements: Coldbrook, two tanks.

Medway

Rationale: Juveniles and post smolts held in support of avoiding extirpation of this valuable SU stock; additional genetic analysis ongoing.

Program deliverables: Rearing of parr; rearing of post smolts to adults.

O&M cost: $8K

Space requirements: Mersey, one 7.6m pond; Coldbrook –four fibreglass tanks.

Eastern shore Atlantic salmon stock assemblage

Rationale: There are 63 rivers which had Atlantic salmon stocks in the Southern Upland of Nova Scotia with over half of those rivers in the "eastern shore" area. These stocks are critically low and in danger of extirpation. Collections of parr were initiated in 2003 to preserve the genetic diversity by trying to collect a diverse assemblage of fish from several of the rivers for rearing to maturity and later release. This approach will bypass the high marine mortality currently being experienced by these stocks.

Program deliverables: Collecting and rearing parr to maturity for later release (subject to tissue sample and genetic analysis to prescribe an appropriate release strategy to protect the diversity).

O&M cost: $5K including collection.

Space requirements: Coldbrook, two fibreglass tanks.

LaHave River

Rationale: Production of limited juveniles for monitoring sea survival, conduct stock assessment activities, and research.

O&M cost: $14K.

Space requirements: Mersey, two 7.6 m ponds for juveniles.

Tusket River

Rationale: Possibly unique stock among SU stocks; small group taken for morphometric comparison in support of genetics.

Program deliverables: Release of age-0 fish ~5,000; complete morphometric comparison of stocks.

O&M cost: $2K.

Space requirements: Mersey, one 7.6m pond for juveniles.

Atlantic whitefish

Rationale: COSEWIC listed as endangered (endemic to only a single watershed) and Recovery Team directed live gene banking as necessary for recovery.

O&M cost: $10K

Space requirements: Mersey, multiple small tanks kept in the culture building (transfer mid-year of a portion of those fish to Dalhousie University); four fibreglass ponds for brood fish, young of year, and 2-year-olds.

Indian Brook Atlantic salmon

Rationale: Support for First Nation sponsored colonization project of Indian Brook on the Eskasoni First Nation.

O&M cost: $1K; fish are collected by Eskasoni First Nation and delivered to facility. Cost is feed and delivery of juveniles.

Space requirements: Mersey, one 7.6m pond for juveniles.

Sackville River Atlantic salmon

Rationale: Salmon population restoration effort to restore a run of salmon to a river where salmon were extirpated; this program has evolved in the face of low marine survival and poor success from rearing and release of smolts to releasing swim-up fry; also, corporate funded rearing of fish to compensate for a fish kill due to a chemical discharge.

Program deliverables: 15,000 fry; 2,000 smolt corporate funded.

O&M cost: $1K (costs for smolt rearing primarily in previous year)

Space requirements: Mersey, one 7.6m pond.

Fish Friends

Fish Friends is a cooperative program with the Atlantic Salmon Federation. DFO collects hatchery fish and incubates several thousand salmon eggs that are then provided for use in elementary and junior high school programs that are administered by ASF, affiliates or local schools (ongoing).

At Mactaquac:

Big Salmon River

Rationale: COSEWIC-listed endangered iBoF stock; Recovery Team has determined the LGB program is “essential for recovery”.

O&M cost: $30-35K

Program deliverables: 500,000 unfed fry; 60,000 0+ fall parr; 18,000 smolts.

Space requirements: six ponds for LGB including one for reconditioning twice spent broodstock; five ponds for progeny.

Black River

Rationale: Collected as a component of the inner Bay of Fundy program subject to genetic analysis and reared as genetic component of iBoF or outer Bay of Fundy stocks.

O&M cost: $1-2K (food and chemical costs only).

Space requirements: one rearing pond

Upper Salmon River (Fundy National Park)

Rationale: COSEWIC-listed endangered iBoF stock; Recovery Team has determined the LGB program is “essential for recovery”. Program costs for this element are entirely recovered from Parks Canada.

O&M cost: $5K

Space requirements: two rearing tanks for 100 wild smolts.

Point Wolfe River (Fundy National Park)

Rationale: “In-river” LGB for Big Salmon River stock , one of Recovery Team LGB objectives; assessment by Parks Canada of natural spawning by select F1 grilse (from all families)

O&M cost: $5K, paid by Parks Canada

Space requirements: one rearing pond for 286 select F1 grilse (released by helicopter fall ’03).

Balance of 25’ (potential) LGB Ponds used for Client-Funded Programs

Aroostook River

Rationale: Restoration of Atlantic salmon in this major Saint John River headwater tributary – Aroostook River, will benefit the entire river; fully funded by Atlantic Salmon For Northern Maine Inc. and the Maine Atlantic Salmon Commission;

Program deliverables: 2M target egg production

O&M cost: $30K JPA funding is provided to DFO

Space requirements: Four rearing ponds in 2003/04; will mature at five ponds and $40K funding in 2004/05.

Magaguadavic River

Rationale: Client-funded program is captive rearing the last wild Magaguadavic River stock collected from this outer Bay of Fundy river in 1998. The river's wild stock is now near extirpation.

O&M cost: $10K of materials and supplies are purchased by the Magaguadavic Recovery Group.

Space requirements: Two rearing ponds are required; one for adult brood stock and one for young-of-year juveniles.

APPENDIX J. SUMMARY OF OPERATIONAL OPTIONS FOR MAINLAND NOVA SCOTIA HATCHERIES

Summary of Operational Options for Mainland Nova Scotia Hatcheries

March 2000

Science Branch
Maritimes Region

OPERATION OF MAINLAND NOVA SCOTIA HATCHERIES

Proposal

Fisheries and Oceans assume responsibility for the mainland Nova Scotia biodiversity programs which include the gene bank for threatened or endangered salmon stocks, mitigation for losses of genetic diversity due to acid precipitation and stocking fish in support of the integrated fisheries management program. Hatchery facilities divested in 1997 have been returned to DFO by virtue of termination of the lease arrangement by the private group operating the facilities and delivering the program in partnership with DFO. We propose to assume responsibility for the programs and upgrade the facilities to deliver a certain level of program, and allow us an operating period to develop partnerships with the private sector. Program costs in year one would be expected to be borne by DFO. A large portion of the costs in year two and beyond would be expected to be covered by Species-at-Risk Act (SARA) program funds.

Background

Salmon Care Association and operating costs

Fisheries and Oceans operated the three mainland Nova Scotia hatcheries, Mersey, Coldbrook and Cobequid, for a variety of purposes but principally to enhance existing stocks to contribute to recreational and commercial fisheries. In October of 1997, DFO divested hatchery operations to Salmon Care Association, a not-for-profit group dedicated to the conservation and sustainable use of Atlantic salmon. Salmon Care Association consisted of representation from the Native Council of Nova Scotia and a private consultant. The program was directed by the former Executive director for Native Council. Salmon Care terminated the lease for the facilities and responsibility for the program on February 6, 2000, for financial reasons.

Salmon Care's annual operating budget for the three facilities was about $320K. DFO formerly operated the facilities for about $500K annually. Facility maintenance and upgrades have been neglected for the past 5 years. Both the Nova Scotia Department of Fisheries and Aquaculture (NSDFA) and local salmon angling groups persistently opposed the divestiture of the three hatcheries. Provincial opposition blocked funding from government (e.g., $180K in Job Transition funds from HRDC) and private sponsors.

Status of Atlantic salmon stocks

Atlantic salmon stocks are at an all-time low. ICES states that "It is evident from indicators of stock status, including the current and predicted estimates of pre-fishery abundance, that the North American stock complex is in a tenuous condition. If the forecast is accurate then pre-fishery abundance in 1999 will be lower than any other pre-fishery abundance value previously estimated despite nearly complete closures of mixed and single stock fisheries.[3]". Evidence of the decline in salmon stocks in Nova Scotia is provided in greater detail in the Atlantic Salmon Maritime Provinces Overview for 1999, DFO Science Stock Status Report submitted for approval to the ADM Science office.

The status of several stocks or stock complexes is critical because of poor marine survival but also because of issues unique to the Scotia-Fundy coast. **1) Inner Bay of Fundy Atlantic salmon stocks are currently under consideration for endangered status** under COSEWIC and, SARA. These salmon have unique migratory patterns different from other North American salmon. The cause for the sharp decline in numbers is still under investigation. **2)** Populations of salmon have been lost from at least 14 rivers on the Atlantic coast of Nova Scotia due to the **impact of acid precipitation**. A review of those impacts is underway in the region and will be completed by late March, 2000. **3)** Recent declines in returns have also had a negative impact in participation by resource users. Contributions of energy and participation as custodians of the resource have been reduced because of the low numbers of salmon and the management restrictions limiting access. Those affects are being dealt with by lengthy consultations between resource users, and science and fisheries management staff of DFO. The process will produce an **integrated fisheries management plan** which will govern stock conservation as well as resource use and access for the next five years.

Current hatchery programs

The current programs being housed at the three Nova Scotia mainland hatchery facilities are biodiversity support programs which are focused on preservation, conservation and restoration of stocks. The facilities house (1) the Inner Bay of Fundy "gene bank" program, a component of the recovery plan for those stocks; (2) stocks to mitigate the impacts of acid precipitation on several Nova Scotia rivers. Returns of salmon to those rivers cannot sustain populations under the current rate of marine survival; (3) fish in support of the integrated fisheries management program designed to protect and increase support for salmon stocks through involvement of the user groups; and, (4) Aboriginal fisheries strategy support. Aboriginal groups have traditionally taken fish for food on several rivers currently being supported by fish from the hatchery facilities. In addition to these programs, the hatcheries have assisted the Atlantic Salmon Federation's Fish Friends initiative which is aimed at education of youth about Atlantic salmon and getting them directly involved in sustainable use by fostering their hands-on culture of salmon in the classroom for ultimate release in a neighbouring river.

DFO objectives: Restore confidence and credibility

Divestiture of the hatcheries in the Maritime Provinces seriously affected the Departments credibility with diadromous fish resource users. Divestiture occurred at a time when salmon stocks were beginning their precipitous decline on rivers of the Scotia-Fundy coast. The public viewed the divestiture as ill informed and counter to the department's mandate for conservation and sustainable resource use. The mainland Nova Scotia hatcheries were heavily involved in programs where declining salmon returns

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[3] No. 7. Extract of the report of the Advisory Committee on Fishery Management, North Atlantic Salmon Stocks, to the North Atlantic Salmon Conservation Organization

became critical coincident with divestiture. Consequently, the role for the facilities became one of conservation of some stocks about the time the divestiture was exercised. These programs have been maintained by the not-for-profit private group (Salmon Care Association) that managed them after divestiture. Program direction was largely DFO's in consultation with user groups and Salmon Care.

Programs

Preservation: Inner Bay of Fundy Gene Bank

Fisheries and Oceans has been leading development of a long range recovery program for Inner Bay of Fundy Atlantic salmon stocks, a group of stocks currently under consideration for endangered status under COSEWIC and, SARA.

One component of the stock preservation and recovery plan is the "gene bank" currently housed in the facilities. The Cobequid station now houses three year-classes of Stewiacke River salmon. A portion of these fish are expected to spawn in autumn 2000 and provide 50 to 70 thousand eggs and ultimately 25 to 35 thousand parr for release into this impoverished river in 2001. This will be the first planned release of increasing numbers of fish based on fish currently at the facility. This will be a key step in the recovery plan but only one element of the overall plan. Collectively the gene bank for Stewiacke River fish in the hatchery facility will include six year classes of wild fish.

Restoration of stocks negatively impacted by acid precipitation: There are 63 rivers in the Southern uplands of Nova Scotia, the area most impacted by long-range-transport of acid pollutants in the province. Atlantic salmon have been extirpated from 14 of those rivers. Production is heavily impacted on 20 additional rivers (Watt 1997[4]). The consequences of losing additional stocks of salmon is not clear at this time. The risk of loss of more separate stocks is being examined in a regional assessment scheduled for March 1-3, 2000. The results of that review are expected to provide us prospects for the future and some sense of urgency for saving threatened stocks.

During the past two years, approximately 50% of the production from the hatcheries has been directed at mitigating losses due to acidification. Twelve rivers negatively affected by air-borne-acid pollutants have been stocked in recent years. Nine of those rivers have been stocked as a hedge against losses caused by the acidification. Recent program reviews have resulted in plans to alter the stocking in acid impacted rivers to limit it to rivers with some hope of recovery if water chemistry improves. Collectively, those programs would require about 160,000 smolts to offset losses due to low pH.

Conservation: Support for integrated fisheries management plan: A five-year fisheries management plan which governs access to stocks for food or recreational fishing is being developed in consultation with the department and user groups. The five-year plan includes limited access for both principle user groups (Aboriginals and anglers) to stocks along the Atlantic coast of Nova Scotia (Salmon Fishing Areas 19, 20 and 21). Prevailing conditions will dictate access but primarily to adipose-clipped hatchery fish. Production from the facilities to support the plan would have to supplement natural production in 13 rivers. In order to be effective, about 150,000 smolts would be required. Stakeholder participation in the recovery plan for Nova Scotia rivers requiring such action hinges on some form or limited access to the resource which is dependent upon limited hatchery stocking.

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[4] Watt, W.D. 1997. The Atlantic Region Acid Rain Monitoring Program in Acidified Atlantic Salmon Rivers: Trends and Present Status. DFO Can. Stock Assess. Sec. Res. Doc. 97/28. 21p.

Partial list of pending programs.

The following list includes several programs under development which will involve the hatchery facilities. Gene bank for Atlantic whitefish, gene bank for other Atlantic salmon stocks; acid precipitation research; examination of marine mortality issues; interactions of aquaculture fish on wild stocks; possibly stock restoration for striped bass.

OPERATING OPTIONS:

Emergency reduced operating plan for Year 1: Operate Mersey and Coldbrook and close Cobequid

- This plan includes closure of Cobequid Hatchery in June and release of all fish products from there, pushing broodstock collections for some stocks until autumn, and transfer of the Inner Bay of Fundy gene bank from Cobequid to Coldbrook; loss of some river progeny, and loss of ability to grow two-year-old smolts

The costs are:	Operating -	Salary	$200,000
		O&M	$90,000
	Facility repair & modification		$75,000

- Operation of the facilities in a reduced program for the next year would permit us time to minimally fulfil our obligations as described while developing a long-term operating plan.

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- We propose to prepare the long-term plan in consultation with user groups and province for delivery in 6-8 month.

Possible operating options for the long-term:

- Our initial review of program obligations and facility status has resulted in a summary of costs associated with operating the facilities under three separate options or closure.
- A large portion of the facility repair or upgrade costs are expected to be borne by Facilities Management because the funds are required to maintain the facilities in safe working order. No maintenance has occurred at the facilities for the past 4-5 years.

Option 1. Operate 3 hatcheries (expanded program capability).

Maintenance of existing program with expansion of gene bank for other endangered stocks.

- gene bank of up to 4 separate stocks and up to 6 year classes per stock (housed at Cobequid and Coldbrook);
- Cobequid used for production of 2-year old smolt when broodstock collections are not successful;
- Production would include fish for the acid stressed rivers, contributions of fish to rivers associated with the Aboriginal food fisheries and the integrated fisheries management program and from the gene bank.
- Production estimates: 350,000 - 1-year-old smolts; 300,000 fall parr; if necessary, 50,000 2-year-old smolts; and, up to 2 million 6-week-old feeding fry from the gene bank program.

Advantages:

- Continuation and advancement of partnerships with user groups (conservationists, anglers, Aboriginals, universities, private industry);
- Reduced risk and enlargement of gene banking of diadromous fishes;
- Continued mitigation of acid impacts to compensate for years when broodstock are not available (2-year-old smolts);
- Research partnering with local university.

Disadvantages:

- Increased operational and upgrade costs.

Costs Option 1:	2000-2001	Operating	Salary	$315,000
			O&M	$110,000
		Facility repair / upgrade		$95,000
	2001-2002	Operating	Salary	$325,000
			O&M	$110,000
		Facility repair / upgrade		$269,000
	2002-2003	Operating	Salary	$335,000
			O&M	$110,000
		Facility repair / upgrade		$247,000

Option 2. Reduce on-going program to operate at 2 sites

Closure of Cobequid Fish Culture Station to reduce costs and consolidate a portion of the program at the Coldbrook and Mersey facilities.

- Retrofitting facilities to deliver maximum program potential at the two sites.
- Gene bank of 2 separate stocks with up to 6 year classes per stock;
- Production would include fish for the acid stressed rivers, contributions of fish to rivers associated with the integrated fisheries management program and from the gene bank.
- Production estimates: 320,000 - 1-year-old smolts, 300,000 fall parr and 1 million 6-week-old feeding fry from the gene bank program.

Advantages:

- Lower operating costs;
- Lower maintenance and upgrade costs.

Disadvantages:

- Loss of ability to grow two year old smolts;
- Fewer gene banks;
- Greater risk of loss of gene bank because animals cannot be split between two sites.

Costs Option 2:	2000-2001	Operating	Salary	$252,000
			O&M	$100,000
		Facility repair / upgrade		$80,000
	2001-2002	Operating	Salary	$250,000
			O&M	$84,000
		Facility repair / upgrade		$154,000
	2002-2003	Operating	Salary	$260,000
			O&M	$84,000
		Facility repair / upgrade		$90,000

Year 4 would require $30k to $180k for facility work depending on water volume testing.

Option 3. Operate 1 facility to support gene bank only (1 year emergency option only)

Operate Coldbrook for the gene bank and hold Mersey in a dormant state pending review of program. Cobequid hatchery would be closed at the end of June, 2000. This option assumes operation of Coldbrook to hold the existing Stewiacke River gene bank only. No broodstock would be collected in 2000 and hence no smolt production for 2001 and 2001.

Advantages:

- Lower operating costs;
- Lower maintenance and upgrade costs.

Disadvantages:

- Loss of ability to house more than two separate stocks in gene banks;
- No restoration of acid impacted rivers or input of fish for the Aboriginal or recreational fisheries for at least two years;
- Greater risk of loss of gene bank because animals cannot be split between two sites;
- Deterioration of facilities not operated while program is in review;
- Loss of staff and technical knowledge during shut down.

Costs Option 3:	2000-2001	Operating	Salary	$100,000
			O&M	$55,000
		Facility repair / upgrade		$10,000
	Subsequent year costs subject to review.			

Option 4. Closure of all 3 facilities in June 2000

Termination of operation in all 3 facilities by Fisheries and Oceans, removal of fish products, and sale or divestiture for other purposes to private sector.

Advantages:

- Eliminate long term costs.

Disadvantages:

- Further loss of credibility with Aboriginals, the diadromous fisheries community, and the general public.
- Loss of gene banking capability for threatened or endangered stocks of diadromous fishes; no other facility is capable of providing this service in Nova Scotia
- No means of mitigating acid impacts;
- Inability to partner with Aboriginals or salmon angling community, or the general public where diadromous stock conservation is involved;
- Loss of input into Fish Friends program in schools (education) and partnership with associated groups.

Costs Option 4:	2000-2001	Operating	Salary	$50,000
			O&M	$25,000
		Facility repair / upgrade		$5,000[5]

Follow-up

- These facilities have been integral to the ongoing Inner Bay of Fundy Atlantic salmon stock recovery planning process, restoration planning for acid impacted rivers, and maintaining user-group interest in the diminishing Atlantic salmon stocks.
- A long-term (ten-year) plan must be developed which includes planning for all three issues. The role the hatcheries will play in those plans will be laid out in consultation with users, DFO staff, and the province, over the next 8 months.
- Sourcing for funding outside DFO, where applicable, will be identified in the plan.

[5] Divestiture costs may be significant and cannot be estimated at this time.

APPENDIX K. BUSINESS CASE 2004

1. Name of Initiative:

Technical Review of Utility and Costs of Maritimes Region Biodiversity Facilities (A case for designated financial support of the Nova Scotia Biodiversity Facilities)

2. Brief Description of Initiative

The initiative was prompted by the DG, Fisheries, Environment and Biodiversity Science, for purposes of understanding and responding to requests by Maritimes Region for designation of national funding for Science's Mersey and Coldbrook Biodiversity facilities. The facilities are dedicated to the live gene banking and rearing of broodstock for listed and unlisted endangered anadromous fish species in Nova Scotia. The request is for $380K, about the same value as the $340K pre-divestiture funding (1996-97) of Mersey and Coldbrook that was withdrawn from the Region by NHQ in Program Review[6]. Faced with significant budget shortfalls in the last two years, including but not restricted to a low allocation from SARCEP, regional Science reduced the capacity and budget to $380K for both facilities, the amount requested herein for 2005-06 and 2006-07. In 2004-05 approximately $190K of the $380K was risk-managed by Science; the remainder was risk-managed and eventually provided by the RDG and late in the year, SARCEP.

Contact: Michael Sinclair, Regional Director of Science
Maritimes Science
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e-mail: SinclairM@mar.dfo-mpo.gc.ca

3. Decision Requested

Approval in principle of NHQ funding of $380K for NS facilities in each of 2005-06 and 2006-07 (Y/N) ________

Approval of development of a Business Plan to reduce operating costs in 2007-08 (Y/N) ________

More information required (Y/N) ________

4. Benefits and Impacts

National support of Science's Mersey and Coldbrook biodiversity facilities and Live Gene Bank (LGB) programs to hedge against the extirpation of Maritimes Region Atlantic salmon, Atlantic whitefish, and possibly in the future, shortnose sturgeon, striped bass and American eel, would be consistent with return of resources for the Mactaquac Biodiversity Facility. Department support for the facilities would acknowledge for both east and west coasts that LGBs and captive broodstock rearing programs offer opportunities to reduce extirpations, minimize the loss of genetic diversity, position for recovery, and gain the confidence of client groups concerned that the Department is soft on SARA.

Failure to support the initiative will result in the loss (likely extirpation) of representatives of over 100 salmon populations, hamper Atlantic whitefish research and recovery initiatives, deny research and possible recovery initiatives for sturgeon, striped bass and eel and result in the need for reassignment of 4.75 FTE of fish culture technicians.

5. Context

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[6] $400K was initially withdrawn for the Mactaquac Hatchery but later returned to the Region after it was concluded that the Department was legally bound to continue its′ operation.

The NS facilities for which support is sought are two of the three former DFO NS mainland hatcheries which catered to the provision of "access" to fisheries i.e., enhancement. Under "Program Review" in the mid-1990s, their function was deemed to be superfluous to government programs and the facilities surplus to requirements. Thus, in the fall of 1997, the Mersey, Coldbrook and Cobequid facilities were divested to 'Salmon Care', a not-for-profit group with the vision of enhancing fisheries with public support for the public good. This divestiture process involved a 5-year lease period for the facilities and assets. However, the operation went bankrupt in the spring of 2000, the lease was terminated and by default the facilities and their fish stocks were returned to the Region's Science Branch.

Concurrently, in the fall of 1998, an ad hoc committee of concerned conservationists, provincial and federal biologists and researchers including DFO, and volunteer and "retained" expert geneticists, under the umbrella of the Salmon Fishing Areas 22 and part of 23 Salmon Management Advisory Committee devised an emergency plan to save the last remaining populations of iBoF Atlantic salmon, in a LGB. This ad hoc committee, later recognized as a core element of the Planning Group within the inner Bay of Fundy (iBoF) Atlantic salmon Recovery Team, promoted a DFO-led collection of Stewiacke River salmon parr for captive rearing under contract to Salmon Care. Parr were also collected from the Big Salmon River, iBoF NB, and similarly held under contract in the "divested" Saint John Hatchery before eventual transfer to Mactaquac. With Salmon Care's return of the facilities in 2000, along with the 1998 and 1999 endangered Stewiacke River parr collections there was deemed to be no alternatives to the continuation of the LGB by other than DFO. This was because Nova Scotia lacked both mandate and capacity at their trout hatcheries and the aquaculture industry would have been unwilling to risk their largely disease-free status by taking in wild fish.

Options for cut-backs and impact on the fledgling LGBs were first considered in March 2000 and consisted of i) operation of the three facilities, ii) operation of Mersey and Coldbrook; disposal of Cobequid, iii) operation of Coldbrook and mothballing of Mersey, and iv) closure of all three facilities. Option ii) was chosen with the expectation that SARCEP would bear a large portion of the costs. LGB operations and supportive breeding programs expanded in 2000-2002, both with the 'need' for greater effort on behalf of the endangered iBoF (listed), and Southern Upland (unlisted) salmon and Atlantic whitefish (listed) populations and access to relatively abundant DFO species-at-risk (SARCEP) funding. (The three designates had high ranked priorities and qualified for nearly $250K each for research monitoring and facilities). Continuation of efforts in integrated fisheries management (IFM) and "mitigation"/ restoration of acid impacted populations at the facilities and which were embodied in the Salmon Care program were gradually dropped in spite of opposition expressed in a public consultation, so that Science could focus on the most basic of conservation initiatives, i.e., LGBs and captive broodstock production. Delivery of recovery initiatives for Atlantic whitefish through culture and propagation for biological investigations relevant to repatriation of its' former range and re-establishment of anadromy began at the Mersey Biodiversity Facility in 2001 and are available for release in 2005.

Re-profiling of the Department's species priorities for SARECP funding in 2003-2004, i.e., a drop of approximately 20 "ranks" in priority for each of iBoF salmon and Atlantic whitefish, and complete exclusion of priority for SU salmon dramatically reduced opportunity for SARCEP funding. This resulted in correspondingly increasing pressures on regional Science to realign gene banking operations with precious few risk-managed resources. Significant financial support has come from Parks Canada who are totally dependent on the Mactaquac facility to live gene bank populations of two National Park rivers in the inner Bay of Fundy. Encouragement for the continuation of the NS operations has come from the World Wildlife Foundation, the Maritime Aboriginal Peoples' Council and most recently within representations to Minister Regan by the Nova Scotia Salmon Association and Atlantic Salmon

Federation. (In September, the Minister and local MP toured and were familiarized with the Mactaquac facility.)

6. Public Policy Considerations

Support by Ottawa and continued operation of the Mersey and Coldbrook operations would serve to instil some faith in provincial fisheries/natural resource agencies, NGO stakeholder groups and the public in general, that the Department has an interest in carrying out the iBoF Atlantic salmon Recovery Team's short term objectives of harbouring and protecting what remains of the residual populations, positioning for recovery and evaluating progress towards population self-sustainability in 2010.

Closure of the facilities will bring into public question the recent expenditure by Real Property and Assets Management of about $1M on the two facilities and as well challenge the Department to find alternate assignments for five specialized staff in relatively small communities (Liverpool and Coldbrook-Kentville areas). Closure/disposal of the wholly DFO-owned Coldbrook facility would present less of a challenge than the Mersey facility which operates on land leased from Nova Scotia Power and for which there is a relatively new lease agreement and set conditions of abandonment.

7. Options

1) Continue per 2004-2005, with costs at Mersey of $131 K salary for two full time continuing and one seasonal regular salaried employees (2.75 FTEs) and Coldbrook of $83.3K salary (2 FTEs) and together, a requirement of $90K casual salary (total $304K) and $80 K O&M. This program was sharply reduced from that of 2003-2004 by concentrating on the captive production of broodstock for natural spawning; live gene banking/pedigreed matings of only the highest priority populations within the inner Bay of Fundy area (similar reductions in Southern Upland salmon), elimination of unfunded holdings, and restriction of research to the endangered iBoF population and Atlantic whitefish.

2) Reduce Mersey to a seasonal operation, i.e., terminate the production of all smolts for release and the Atlantic whitefish program; attempt to transfer a small number of Atlantic whitefish brood to Coldbrook for holding, reduce iBoF fish production, multi-task 2 FTE positions during winter shut-down and shorten the term of the seasonal FTE. The projected savings are about $20K casual salary and $10K O&M.

3) Close and mothball Mersey, i.e., end all production of juvenile salmon beyond the unfed fry stage, limit live gene banking to captive rearing of adults at Coldbrook for release (assumes that freshwater captive-reared adults will successfully naturally spawn; only 0.25M eggs could be taken for release as unfed fry), retain caretaker at Mersey (~ $15K per annum) and abide by conditions of lease arrangement with Nova Scotia Power. The projected savings approximate $50K casual salary and $45K O&M. 2.75 FTEs ($131 K salary) are "affected" and require redeployment.

4) Close and mothball Mersey and Coldbrook, i.e., terminate all salmon Live Gene Banking and Atlantic whitefish efforts in Nova Scotia, invite application to the Minister for special permission to freely move iBoF LGB salmon (incl. stream-reared fish within the Bay of Fundy eco-region to Mactaquac (disease regulations/provincial boundary currently inhibits such a move and likely precludes live gene banking of Southern Upland stocks because of (i) difficulty in moving fish from an Atlantic eco-region across provincial boundary to Mactaquac (ii) lack of capacity at Mactaquac under the current Agreement with NB Power which prioritizes efforts on the Saint John River system, NB). The projected savings approximate $60K casual salary (caretakers at Mersey and Coldbrook will cost approx. $30 K casual salary) and on the surface, $80K O&M. 4.75 FTEs ($214K salary) are "affected" and would require redeployment. (Costs to truck fish back and forth to Mactaquac would reduce the O&M savings by perhaps $10-15K.)

5) Cryopreservation of gametes, 6) transplantation of salmon from distant stocks into inner Bay of Fundy rivers, 7) maintenance in landlocked environments, 8) translocation to distant rivers and 9) introduction of other species were other options that were considered and dismissed.

In brief, moving from options one through four handicaps the initiatives of two Recovery Teams, DFO's credibility with respect to SARA issues, generates inefficiencies of operation (short of total closure), affects up to 4.75 FTEs and would require, for options 2 through 4, a cleverly crafted Ministerial response to riding constituents (some of which are officials of e.g., Nova Scotia Salmon Association) and conservationists in general as to the Department's lack of support for LGB initiatives and virtual dismissal of an opportunity to prevent extirpation of most of Maritimes Region Nova Scotia mainland salmon populations.

8. Recommended Option(s)

The Nova Scotia facilities have had about $1M improvements to infrastructure. New pipelines, bio-security improvements including two buildings to house brood fish, tanks and oxygenation system, complimented by dedicated full- and part-time staff, make them unique in North Atlantic countries and valuable assets with potentially diverse future applications. Complimented by a recognized population geneticist, live gene banking at these facilities is recognized as second to none by North Atlantic salmon producing countries. Thus the recommendation is to accept a modified option # 1, i.e.:

> Continue the operation of Mersey and Coldbrook through 2005-07 at the current reduced level of about $380K per annum (about the same sum that was withdrawn from the Region at the time of divestiture). By November 2005, however, it is recommended that a business plan be explored and developed that, through grow-out and sale of smolts to the aquaculture industry, would reduce the overall costs of the NS facilities to less than $200K per annum.

The operation of the iBoF LGB at Mactaquac Biodiversity Facility is with the exception of funds from Parks Canada (Fundy National Park) now risk-managed by Science Branch. The action to date has been to reduce the level of gene banking of the Big Salmon River population and other non Saint John River programs. The facility is bound by a nearly 40-year old legal agreement between the Crown and NB Power to provide for the mitigation for salmon lost to NB Power through alteration of river habitat. Infrastructure improvements over the last three years have made Mactaquac a state-of-the-art facility, well equipped for live gene banking and now, captive production of broodstock. Thus Mactaquac should continue culture operations at their current level of about $625K, client, esp. OGD support such as Parks Canada, should be sought at the same ($75K) or greater level and terms of the original agreement with NB Power should be reviewed with a view to transferring more of the costs, e.g., power to operate the facility, from government to the utility.

9. Implementation Strategy/ Work Plan

Operation of Mersey and Coldbrook would continue in the same manner as in 2004-05 but with guaranteed rather than risk-managed funding, i.e., $380K. A two person "team" comprised of the NS "co-ordination and outreach" biologist and "NS facilities supervisor" should explore partnering initiatives with the NS aquaculture industry, possible intermediaries and GO-NGO revenue generation models used in other North Atlantic jurisdictions. The most viable models, if based on other than a Joint Project Agreement (possibly with a non-profit intermediary), should be reviewed by September, 2005 and in the light of "tests for Treasury Board's expenditure and management review" be cognizant of TB guidelines in generating revenues from government products and services. Negotiations by the "team" (with possible assistance from the Aquaculture Management Directorate) with prospective buyers of smolts should be completed by mid-October so that eggs could be acquired for a November 2005 start up and delivery in spring 2007. The financial objective and responsibility of the “team”, and their Division and Branch managers, in any plan, would be to reduce the costs to the public of operating the two facilities for conservation initiatives in NS (and rearing of smolts for the industry) to less than $200K within 2 years.

10. Decision Minute

a) Sr. Management’s Decision

Upon review of Maritimes Region Business Case, supporting ‘Technical Review of Utility and Costs of Maritimes Region Biodiversity Facilities’ and Appendices, it is the view that the two Nova Scotia biodiversity facilities and their Live Gene Banking and adult rearing operations should be financially supported in 2005-06 and 2006-07 with $380K per annum designated funding from Headquarters. These facilities and their science-based programs are internationally respected, have the supported of several headline advocacy groups, two provinces, Parks Canada and are positioned to stave off extirpation of representative populations of SARA-listed and unlisted ‘endangered’/extirpated Atlantic salmon populations and, given time, possibly effect their recovery. As such they are a credit to the Department and Minister Regan. These Science facilities also offer the opportunity to research and propagate the listed ‘endangered’ Atlantic whitefish, and possibly, in the near future, about-to-be-listed populations of shortnose sturgeon, striped bass and American eels.

The Regions proposal to partner with the aquaculture industry should be encouraged and results monitored and reviewed in September 2005. Such a plan offers the opportunity for industry to promote itself as conservationists and supporters of leading edge scientific endeavours to save listed wild endangered populations at the same time as it facilitates the overall reduction in costs to the public of meeting SARA commitments.

b) Instructions for next steps.

Assure the Region of $380K support for the NS facilities in 2005-06 and 2006-07; explore the possibility of securing the resources through SARCEP core funding and, encourage Aquaculture Management Directorate to assist Science Branch, Maritimes Region, in the development of a partnership with industry that in the eyes of the public, benefits both sectors.

c) ______________________________

Larry Murray, Deputy Minister