

***NORTH
PACIFIC
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TECHNICAL REPORT 3

***Workshop on Salmonid Otolith
Marking***

**Technical Editors: Peter Hagen, David Meerburg, Katherine Myers,
Alexander Rogatnykh, Shigehiko Urawa, and Eric Volk**

Vancouver, Canada

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Seattle, Washington, USA, March 21, 2001

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Foreword

The NPAFC International Workshop on Salmonid Otolith Marking was held in Seattle, WA, U.S.A., on March 21, 2001. The Workshop was organized and sponsored by the North Pacific Anadromous Fish Commission (NPAFC). The NPAFC Working Group on Salmon Marking served as the Workshop Coordinators. All necessary arrangements were made by the NPAFC Secretariat in cooperation with the Workshop Coordinators and Local Coordinators.

Over 70 scientists and fisheries officials attended the Workshop. There were 14 oral and 3 poster presentations followed by general discussion sessions. Extended abstracts of the oral and poster presentations are included in this Technical Report, which also contains opening remarks and a short review of the Workshop by the co-chairpersons of the Workshop. The material presented in this Technical Report has not been peer reviewed, and does not necessarily reflect the views of the NPAFC or Parties. Some work may be preliminary. The material has been edited by the Workshop Coordinators for clarity and publication purposes only.

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Opening Remarks

Welcome to the Workshop on Salmonid Otolith Marking sponsored by the North Pacific Anadromous Fish Commission (NPAFC). I thank everyone for attending this workshop. It appears to be a standing room only this morning, but I encourage everyone to try to make yourselves comfortable and stick with us to the end. While some of you may get tired legs, I think you will nonetheless find it to be a very interesting workshop.

We have a very full schedule today. The morning session contains presentations on the techniques of otolith analysis and marking methods. It also includes a presentation on the role the NPAFC is assuming as repository of otolith salmon marks by member countries. The afternoon session is devoted to case studies on the application of these methods to answer a variety of questions associated with salmon biology and salmon management. The poster session is out in the hall, and is available for viewing during the breaks. We will end with an open discussion, and everyone is invited to stay with us and participate.

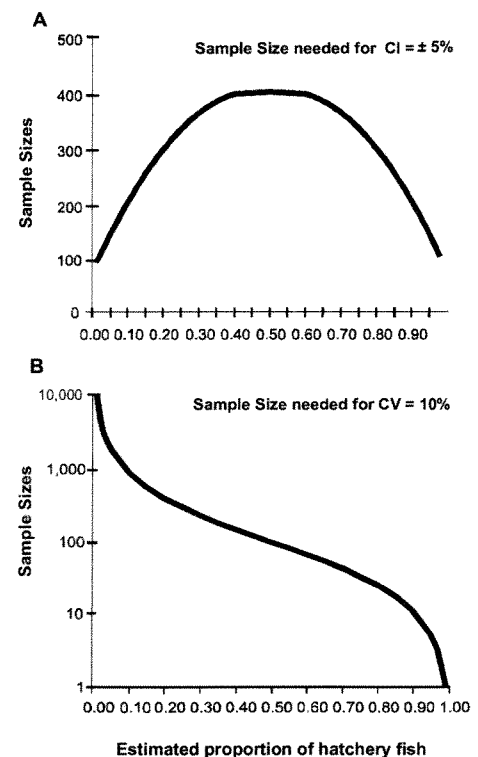
Given the very tight schedule, I have only a few minutes to present some opening remarks. I will attempt to briefly run through some basic concepts from a sampling perspective on why otolith marking works. In particular I will discuss how we can leverage the ability to mark 100% of a group of fish through otolith marking to produce precise estimates when sampling a population of fish for those marks. I will also propose that with some of the techniques presented in this workshop it is within reach to mark all hatchery production of salmon released into the North Pacific. I believe that will give us a much greater understanding of salmon biology and accountability for hatchery production, and should be a long-term goal.

My observation is that there tends to be a hesitancy to embrace otolith marking because of a misconception regarding the amount of work involved in detecting the marks. Certainly detecting an otolith mark requires sacrificing the fish: it involves dissecting the head and in some cases processing and preparing the otolith for viewing. The prospect of individually handling each sample can seem daunting, and at first may not seem a worthwhile effort. However, as you will learn from the talks and the poster session today, the methodology is now available to mark large numbers of fish at very little cost. With millions of fish available for

sampling, the sample requirements for recovering the marks are greatly reduced. And with a few caveats, it is fairly straightforward to draw inferences about the population with relatively high precision from small sample sizes. This is possible because with 100% marking the underlying distribution for recovering the marks can be approximated as binomial (marked, unmarked) or multinomial (mark A, mark B, etc.).

To consider the sample size requirements one first needs to address the question of what level of precision is necessary when estimating the proportion (P^{\wedge}) of the marked fish in a sampled population. Precision is typically expressed as the standard error (SE) of the estimate, and there are two ways in which a target level is determined. Precision based on the absolute standard error of the estimate is typically cast as the confidence interval (CI) (e.g., $P^{\wedge} \pm 1.96 * SE$), and precision defined as relative standard error is referred to as coefficient of variation (CV) (SE / P^{\wedge}). In practice these can result in very different sampling goals. Figure 1A illustrates the number of samples required to ensure the 95% CI is $\pm 5\%$ of the estimated proportion of hatchery fish in a population. With this goal the worst case scenario is when 50% of the population is hatchery fish. In that situation 400 samples must be examined to ensure that 95% of the time a similar sample size will produce an estimate between 45% and 55% hatchery fish. If the actual percentage is greater or less than 50%, a sample size of 400 will produce even better precision. In contrast to the dome-shaped CI, Figure 1B shows a different shape that is based on the sample size requirements to achieve a target CV of 10% as a function of mark proportion. Using a CV-based goal, the sample size requirements are burdensome when the population of interest is uncommon, and not very rigorous when the population is abundant.

Fig. 1. (A) Sample sizes necessary to achieve a confidence interval of $\pm 5\%$ around the estimate of the hatchery proportion in the population. (B) Sample sizes necessary to achieve a coefficient of variation of 10% on the estimate of the hatchery proportion.



In practice, CV goals are used more often when the interest is in the actual numbers and not proportions of a group within a sample population. It is perhaps more applicable in situations in which the population of interest needs to be protected such as an endangered species, but this is not the case for hatchery fish.

Sampling goals based upon CIs are familiar to most people, and are well established. In most situations they provide sufficient information to characterize the population sampled. In a mark recovery program, trying to achieve a target CI can provide several advantages. For example, Figure 2 illustrates how the confidence bounds (the upper and lower confidence limits) are largely invariant of size of the population being sampled. The exception is very small populations, in which sampling without replacement may have an influence. Assuming the samples obtained are representative of the population, 500 otoliths will provide the same precision regardless of whether the population of interest is 10,000 or 10,000,000.

Another attribute of using the CI approach for setting sampling goals is illustrated in Figure 3. The graph shows how most of the precision, in terms of 95% CI range, is captured in the first 100 samples. After that it appears to be a case of diminishing returns, and there is little to be gained by processing large numbers of additional samples. This has particular advantage for programs in which the timing of decisions is critical, such as fishery management applications. Using a multi-stage processing schedule, it is possible to optimize the processing effort from multiple strata to reduce costs in meeting precision goals.

What happens, however, if the marking rate is not 100%? In those situations the sampling requirements will also increase depending on the underlying percentage of hatchery fish in the population. This is illustrated in Figure 4, which shows the sample size requirements necessary to achieve a precision goal of 95% CI \pm 5% as a function of the marking rate and the proportion of hatchery fish in the population. With higher levels of hatchery fish it is necessary to have a high marking rate to keep the sample sizes manageable. If the marking rate is too low then external, visible marks such as fin clips or tags become more appropriate.

I have shown here that given the ability to otolith mark fish at rates close to 100%, concerns about the need to process the otoliths can be addressed through consideration of the target precision goals and the attendant sample size requirements. As will be presented in a number of papers today, valuable information and inferences on salmon production can be drawn from relatively few samples, and programs can be developed to take advantage of

Fig. 2. The 95% confidence bounds as a function of the size of the population, based on sample sizes of 100, 500, and 1000 for a sampled population containing a marked hatchery proportion of 0.20.

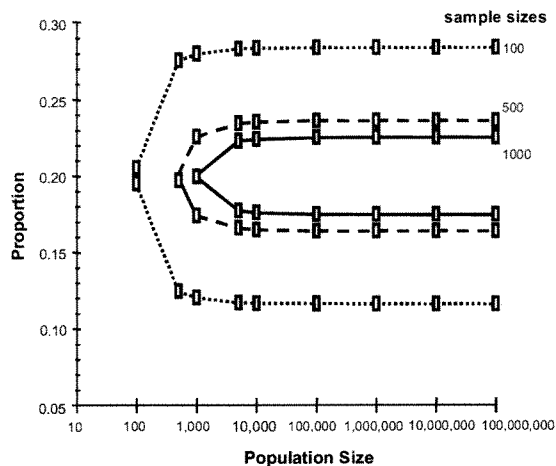


Fig. 3. Changes in the 95% confidence interval range (upper bounds-lower bound) for the case of 50% hatchery fish as a function of increasing sample sizes. Graph illustrates how little there is to gain in precision by processing additional otoliths.

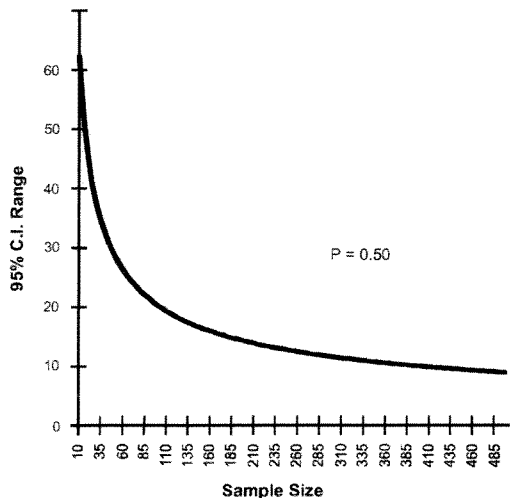
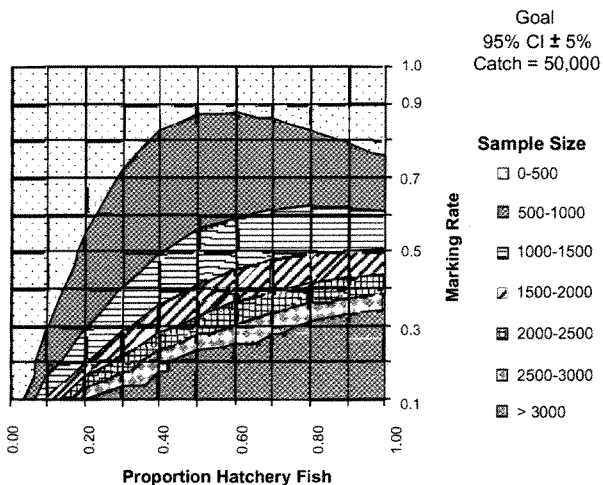


Fig. 4. Sample size requirements to achieve a 95% CI that is \pm 5% of the point estimate as a function of the marking rate and the proportion of hatchery fish in the population. The graph illustrates how the sample size requirements increase as the marking rate decreases at moderate levels of hatchery production.



the marked fish released into the North Pacific. The caveat, of course, is that when relying on small numbers to draw inferences it is also relatively easy to be fooled by samples that are not collected randomly or may not be considered representative. Therefore it is also necessary to give very careful consideration to the sample design being used.

Because of the increasing number of otolith marked fish released into the North Pacific and the increasing overall marking rate, we have the capability to gain even more knowledge about salmon biology, distribution, and abundance. I think that a good case can be made that all hatchery production should be marked by some method. As will be shown in many of the talks today, otolith marks are now a valuable tool in a variety of applications. The number of potential applications will certainly increase as the overall marking rate of hatchery fish increases throughout the North Pacific.

Once again welcome to the workshop.

Peter Hagen
Alaska Department of Fish and Game, U.S.A.
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An Overview of Otolith Thermal Marking

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Keywords: Salmon, otolith, thermal marking

Otoliths have been a primary focus of salmonid mass-marking efforts as they are the first calcified elements to form, are well-known recorders of environmental fluctuations, and induced structural or chemical marks are permanent. In particular, dramatic effects on otolith increment width and optical density have been tied to temperature changes (Campana and Neilson 1985; Neilson and Geen 1985; Mosegaard et al. 1987; Brothers 1990; Volk et al. 1990, 1994; Munk et al. 1993), and investigators have utilized this fundamental connection to place internal otolith marks on large numbers of hatchery salmonids including lake trout (Brothers 1985, 1990; Bergstedt et al. 1990), Arctic charr (Mosegaard et al. 1987), and all species of Pacific salmon (Volk et al. 1990; 1999; Munk et al. 1993).

Hatchery-incubated salmonids are particularly well suited for thermal marking because incubation and yolk absorption stages are protracted, large numbers of fish are concentrated in hatchery incubators, and otoliths begin growing in embryos. As a result, there is a lengthy period during which multiple marks may be administered, and this includes both the pre- and post-hatch otolith regions. Nearly all salmon otolith-marking endeavors mark fish between the eyed-egg and fry stages, and there are few marking alternatives if embryos must be marked, such as when eyed-eggs are placed in remote site incubators.

The basis of otolith thermal marking is that short-term temperature manipulations alter the appearance of one or more otolith increments, producing an obvious pattern of events that could be recognized at any life stage. Depending upon the mean ambient temperature, water may be heated or chilled to produce the desired effect, and exposure times have varied from as little as two hours (chilled water) to heated water exposures lasting several days. The magnitude of the temperature change typically is governed by capacity for producing heated or chilled water, and generally ranges from 2°C to 5°C with larger changes producing more apparent effects (Volk et al. 1999).

Since there is a close relationship between accumulated temperature units and the spacing between induced thermal events, it is easy to create patterns based upon the number and relative spacing of induced events. The two coding schemes currently employed are the bar code (Volk et al. 1994) and the RBr code (Munk and Geiger 1998). Both of these systems utilize band number as well as the spacing between bands or groups of bands induced in different otolith regions to describe a pattern. While large numbers of unique patterns are theoretically possible using either of the coding methods, practical limitations associated with hatchery operations, fish development and visual recognition place important limits on the number of possible patterns (Hagen 1999). Though many marking objectives can be achieved with no organized rules for pattern assignment, a standardized system of organizing pattern information on otoliths potentially offers a larger number of patterns, and also provides the opportunity for coordinating marks between agencies and countries so that duplicates may be avoided in mixed-stock recovery areas (Urawa et al. this volume).

Because we depend upon skilled human readers to detect marks errors are inevitable, and may arise from poor otolith marks, natural mimics of induced patterns, and poor preparations. Patterns of dark increments are easily induced into growing otoliths using temperature changes, but otolith mark recognition is dependent not only upon the effect of planned thermal events on the otolith, but also on the background or ambient increment characteristics against which they must be recognized. Otolith mark recognition is a signal and noise problem where the induced mark signal must be distinguished against the background incremental "noise" created by the ambient temperature regime and other physical disturbances. Thus, the ultimate quality of the otolith mark, and the likelihood of misclassifications, depends not only on the pattern induced, but also on how easily that pattern is discriminated from background increment characteristics resulting from ambient temperature conditions.

The potential for errors in recognizing otolith marks has been studied using known mark status specimens. Bergstedt et al. (1990) reported that among juvenile lake trout held six months after marking, marked and unmarked controls were accurately recognized in 85–98% of the samples (N = 41). Most errors misclassified non-marked fish. Hagen et al. (1995) accurately identified 64–100% of known marked and non-marked adult pink salmon otoliths (N = 36). Among 1,852 chinook and coho salmon otoliths that were known to be marked or non-marked, Volk et al.

(1999) reported an overall mean error rate for known marked fish of 2%. However, for non-otolith marked control fish, there was a discrepancy rate of 6–11% between mark determinations and their true status. Both Volk et al. (1999) and Bergstedt et al. (1990) suggested that the presence of a pattern is more easily recognized than its absence, highlighting the importance of the background otolith pattern in the ultimate success of discerning an induced pattern.

Unfortunately, evaluating the magnitude of these discrepancies in most cases is difficult because there is usually no “gold standard” of known mark status specimens with which to compare mark determinations, and findings are usually reached using several readers to examine each otolith. Recently, Blick and Hagen (in press) discussed the use of agreement measures and latent class models for evaluating the precision of otolith mark determinations. Agreement measures, such as *kappa*, can be useful as a relative measure of the reliability of determinations with two independent readings, but the proportion of marked fish can influence its magnitude. With a third reader, latent class models can provide estimates of the of the error rates for each reader. If key assumptions are met, these methods provide a means to evaluate the precision of mark group composition where no error free standards exist.

Currently some 5 billion juvenile salmon are released by hatcheries into the North Pacific annually, with some 20% carrying thermally induced otolith marks (Urawa et al. this volume). Management of hatchery stocks for harvest coupled with increasing concerns over their impacts on wild fish have placed a growing premium on identification of hatchery-reared fish. A variety of large-scale otolith marking efforts have demonstrated that it is practically feasible and economically viable to mark all hatchery production in the North Pacific with thermal codes. However, the utility of thermal marking for discerning the origins of fish captured in high seas, mixed-stock fisheries may be limited by the number of possible codes available, requiring the use of secondary marks or tags to maintain the necessary number of unique marks.

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The Dry Method of Otolith Mass Marking

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Keywords: Salmon mass marking, otolith, dry method of otolith marking, hatchery, incubator, salmon embryos.

Marking has been traditionally used in fish hatcheries to identify fishes of artificial origin. There are many methods of marking such as fin amputation, opercula punching, branding, application of substances to dye the tissues of the organism, external and implanted radiotelemetry and magnetic tags, marking with radioactive isotopes, etc. All these marking methods have their advantages and flaws, but few are appropriate for mass marking of small fishes such as chum and pink salmon fry raised at the hatcheries.

Mass marking methods based on marking otoliths have been intensively developed in recent years. The countries primarily using these marking technologies are the United States, Canada, Japan and Russia. The most widely used is the method of thermal marking of otoliths developed by American scientists (Volk et al. 1990, 1999; Munk and Geiger 1998).

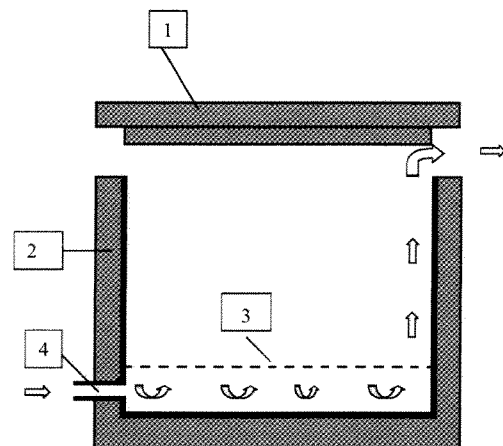
In Russia, we use the dry method for marking salmon otoliths in addition to thermal marking. The idea arose several years ago while studying the reasons for the development of pseudo-rings (additional rings) on embryonic salmonid otoliths. It was noticed that such pseudo-rings appear on the otoliths of embryos lying inside the packed clusters of eggs where the water flow is minimum or null. We hypothesized that there was a relation between the water flow intensity and formation of the otolith rings, and proposed to use the artificial cessation of water flow in incubators to induce recognizable rings on the otoliths of salmon embryos. The principle of this method, as well as that of the thermal marking, is based on the fact that environmental changes cause adequate physiological reactions in fishes to change the thickness of the daily calcium and protein increments in otoliths. Should the influence of this factor be periodical in nature, its periodicity manifests itself as a sequence of dark and light rings of certain width. In our opinion, in the process of dry marking the abrupt changes of ambient conditions put the organism under stress resulting in physiological changes. We feel that the same thing happens when thermal marking is used, where changes in increment characteristics are the result of physiological reactions to changes in ambient parameters.

The dry method of otolith marking is based on periodical changes of the water regime for incubation of eggs. The eggs are dried in incubators according to pre-determined schedules, usually at 24 hours intervals. During one marking cycle, when one dark and one light ring are formed, the eggs are kept dry for 24 hours (without water, in a humid atmosphere), and washed with water during the next 24 hours (with the incubator working in a normal operating mode). To retain a humid atmosphere and prevent abrupt changes in temperatures, the incubators should be covered with a plastic film or any heat-insulating material. When using Atkins and NOPAD incubators, circulation of water should be provided around the bottom and the side panels. The eggs should be thoroughly stirred prior to water drainage to prevent the egg clusters from packing and improve ventilation. The period of embryonic development, during which it may be possible to use the dry marking method, begins from the embryo's eye pigmentation (eyed-egg) stage and lasts until hatching. The length of this "marking window" depends on thermal conditions of the incubation process and may vary at different hatcheries and regions. At the hatcheries of the Magadan Region, this period usually lasts from 20 to 35 days.

The first tests of the dry marking method were conducted in 1996 on chum salmon embryos, and showed that periodic drying of eggs stimulates the formation of otolith rings with the same periodicity. However, in these experiments during interruption of the water flow, the temperature of the incubated eggs gradually equilibrated with the ambient air temperature, and the otolith rings could form not only due to drying of eggs, but also due to the effect caused by the temperature difference in accordance with the mechanisms of the thermal marking.

We conducted another series of experiments using temperature-controlled incubators (Fig. 1) that allowed us to

Fig. 1. Isothermal incubator working in a normal operating mode. 1 – cover, 2 – styrofoam, 3 – mesh tray with eggs, 4 – water inlet.



keep the temperature unchanged when drying the eggs. Over the six days experimental period, the water temperature fluctuated only 0.6°C. Three drying cycles of 24 hours followed by 24 hours of normal water flow resulted in the formation of three rings on the otoliths of pink salmon embryos (Fig. 2). The results indicated that the dry method of salmon marking caused the formation of an otolith mark that does not depend on temperature changes, but occurs due to some other factors related to the egg's drying.

During the mass marking of coho and chum salmon from brood year 2000 at the Yana Hatchery, the temperature also remained relatively constant. The water temperature in the incubator water-supply system ranged from 2.9 to 3.1°C during the 13 days of marking. The air temperature at the hatchery varied from 2.8 to 3.0°C. Distinct marks were found on the otoliths upon completion of the marking process (Fig. 3). One hundred embryonic coho salmon otoliths were inspected to evaluate the quality of the marks and similar marks were formed in all otoliths. American NOPAD incubators were used at the Yana Hatchery for dry marking. Japanese Atkins incubators were used to conduct the other experiments. During marking, the layer of eggs in incubators varied from 2–3 cm up to 25–30 cm (from 1 to 12 inches). The marking was conducted with alternating dry and wet intervals of either 24 hours or 12 hours. In the year 2000, the hatcheries of the Russian Far East released almost 4.5 million salmon marked by the dry method.

The dry method of otolith mass marking is based on the ability of the salmon eggs to normally develop in a humid atmosphere. We discovered that the drying of eggs for 12 or 24 hours does not lead to an increase in mortality rate. The quantities of dead eggs during the marking process and at other stages of ontogenesis (larva incubation and fry raising) remained the same in both experimental and control samples. In some of our experiments, we allowed the eggs to be incubated in a humid atmosphere for 440 hours (18 days). Figure 4 shows the mark consisting of 12 rings. Mortality rate of marked eggs remained the same as the eggs hatching under normal conditions. We also found no difference between drying and wet eggs with regard to either hydro fraction content or in a water flow resistance test applied to the fry.

People working at the hatcheries know that fish farmers often use the drying of eggs in practice. For instance, the eggs are dried for a short period of time for sorting. We can also give examples of more continuous incubation of eggs in a humid atmosphere. In the Russian Far East, such incubation has been used at the Ushki Hatchery in Kamchatka since 1934. The eggs of sockeye, chum, coho and chinook salmon were sometimes incubated in a humid atmosphere for 50–60 days. The released fry demonstrated good vitality (Rassokhina 1998).

Fig. 2. Dry mark on the otolith of a pink salmon embryo (above) and control otolith of a pink salmon embryo (below) in the Kulkuty River, 1998.

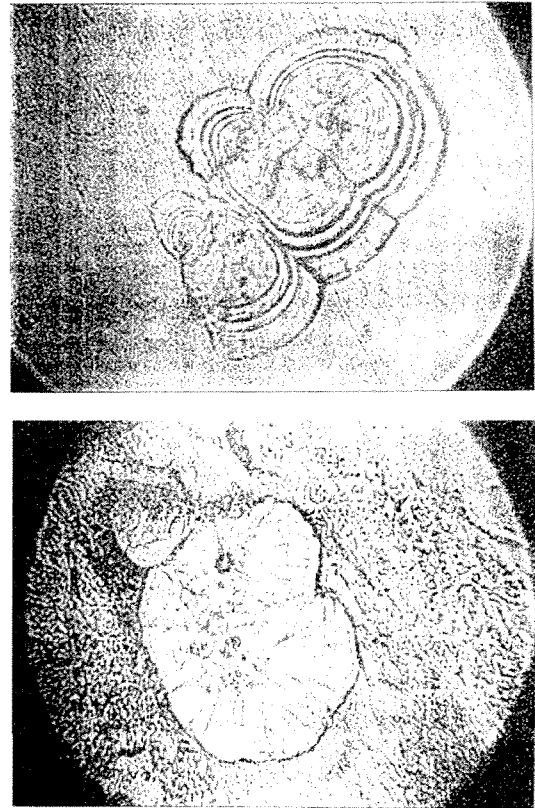


Fig. 3. Dry mark on the otolith of a coho salmon embryo in Yana Hatchery, 2001.

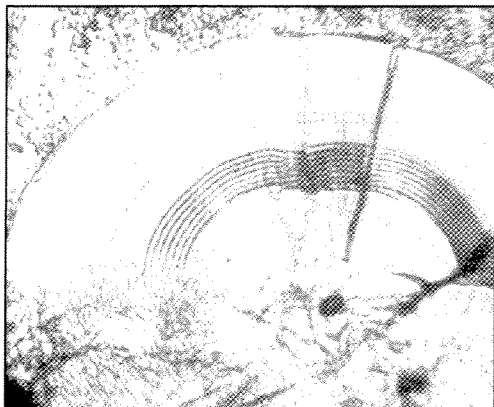


Fig. 4. Dry mark on the otolith of a chum salmon embryo in Ola Hatchery, 1998.



The drying of eggs was officially accepted in fish farming for salmon from the genus *Oncorhynchus* (Smirnov 1958) and *Salmo salar* (Yandovskaya *et al.* 1996). At the present time, drying of the salmon eggs at the “eyed” stage is being used for the transportation of eggs and for incubation in a humid atmosphere (Mikhailenko and Sokhnov 1997; Tiaptirganov *et al.* 1997). It should be noted that during either marking or transportation, the eggs should be kept in a humid atmosphere and the upper layer of eggs should not be allowed to dry.

The flaw of the dry method of marking is that it cannot be used for marking of the salmon larvae and fry. In general, the dry marking method is simple, convenient and requires no special (including electrical) equipment. The marking can be conducted separately in each incubator within the optimum time period. The dry marking method can be applied at any hatchery and can also be used for salmon farming in field conditions.

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Otolith Marking with Fluorescent Substances at the Eyed-egg Stage of Chum Salmon

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Keywords: Otolith; alizarin complexone; chum salmon; eyed-egg; marking

Otolith marks have some advantages for mass marking of salmon because of their simplicity and short marking time. Moreover we can mark otoliths at the embryonic stages. However, to find the marked fish we have to extract the otoliths from the heads and process the specimens by grinding. We have found that otolith marking with fluorescent substances at the embryonic stage may be a better method than thermal marking (Tsukamoto 1985, 1988; Tsukamoto et al. 1989). A fluorescent mark in an otolith can be found without any treatment, and we seldom mistake the mark due to the clear coloration of fluorescent mark. We used an alizarin complexone (ALC) for mass marking chum salmon, *Oncorhynchus keta*. In order to improve salmon propagation technology for the Sea of Japan stocks in Hokkaido we have examined the early coastal ecology of juvenile chum salmon by means of mass marking of otolith with ALC from 1994 to 1999.

We used eggs from the Syokanbetsu River on the northern coast of the Sea of Japan in Mashike of Hokkaido, Japan. The eggs were moved into the Mashike Branch of the Hokkaido Fish Hatchery. We marked otolith of eggs at the eyed stage with ALC. The eggs were put into two box type incubators, whose capacity was six hundred thousand eggs, and the refrigerator and pump were set in another incubator to maintain a water temperature at 7°C to 9°C. The eggs were immersed for 24 hours in a solution of ALC (200mg/l) with 1N KOH before dilution (Tsukamoto 1988). Because the otolith continues to grow with the cumulative temperature changes, we were able to use a single or double treatments for marking. The cumulative temperatures of the embryo ranged from 242°C to 427°C in November to December. After treatment the eggs were reared in a vertical type incubator with other eggs. The juveniles with marked otolith were stocked, ranging from 795,000 fish to 2,274,000 fish, in the Syokanbetsu River in April from 1995 to 1999 (Table 1). After releasing we recaptured the marked juvenile and adult chum salmon in coastal and offshore waters, and the Syokanbetsu River from 1995 to 2000 (Kawamura et al. 1998, 2000). We observed the fluorescent marks in otoliths using a UV light microscope.

We recaptured 15 juvenile fish (1995), 397 fish (1996), 447 fish (1997), 645 fish (1998) and 1616 fish (1999) whose otoliths were marked in the inshore waters off Mashike with Sayori tounet (Table 2). One otolith marked chum salmon (22.9cm, 118g) was recaptured in offshore waters off the west Kamchatka in the Sea of Okhotsk in September 1996 (Ueno et al. 1998). As a result of our observations of adult chum salmon which returned to the Syokanbetsu River from 1997 to 2000, we found some marked otoliths among adult salmon ranging from 3 to 5 years old (Table 3). The otolith marked chum salmon were as follows; 4 fish at 3 years old (1997), 21 fish at 3 years old and 70 fish at 4 years old (1998), 92 fish at 3 years old, 243 fish at 4 years old and 27 fish at 5 years old (1999), and 18 fish at 3 years old, 59 fish at 4 years old and one fish at 5 years old (2000). We could easily locate the marked otoliths among different aged fish under UV microscope.

Five otolith marked juveniles were found in the stomachs of arabesque greenling, *Pleurogrammus azonus* captured by set net located 90 km north of Mashike in 1999, and eleven otoliths with ALC were observed in the fecal materials of gulls (*Larus schistisagus*, *L. crassirostris*) around the mouth of the Syokanbetsu River in 1999 (Table 4).

We confirmed that a fluorescent mark in otolith with ALC was retained for more than five years. We concluded that otolith marking with ALC was beneficial to the study of chum salmon on distributions, migration and prey-predator interaction.

Table 1. Otolith marking with ALC and releases of juvenile chum salmon.

Date	Stage	Cumulative temperatures	Treatment	No. juveniles released	Body weight (g)
1994.12.8-12.8	eyed-egg	419-427	single	795,000	1.08
1995.11.23-11.24	eyed-egg	242-249	single	1,123,000	0.95
1995.12.13-12.14	eyed-egg	391-399	double	1,138,000	1.32
1996.11.21-11.22	eyed-egg	245-253	small single	1,282,000	0.77
1996.12.10-12.11	eyed-egg	391-398	large single	1,264,000	1.09
1997.12.15-12.16	eyed-egg	405-412	single	2,274,000	0.93
1998.11.24-11.25	eyed-egg	284-301	small single	1,726,000	0.75
1998.12.7-12.8	eyed-egg	380-400	large single	1,902,000	0.85
Total				11,504,000	

Table 2. Recapture of juvenile chum salmon with marked otoliths.

Year	Location	Total of sample fish	No. marked fish	Treatment	% of marked fish
1995	coastal waters off Mashike	1,624	15	single	0.92
1996	coastal waters off Mashike		199	single	
	coastal waters off Mashike		198	double	
	Subtotal	5,446	397		7.29
	offshore waters in the Okhotsk Sea	84	1	double	1.19
1997	coastal waters off Mashike		249	small single	
	coastal waters off Mashike		198	large single	
	Subtotal	5,473	447		8.17
1998		7,723	654	single	8.47
1999	coastal waters off Mashike		698	small single	
	coastal waters off Mashike		918	large single	
	Subtotal	4,140	1,616		39.03
Total		24,490	3,130		12.78

Table 3. Recapture of adult chum salmon with marked otoliths.

Year	Location	Total of fish samples	Age	No. marked fish	Treatment	Ratio (%)
1997	Syokanbetsu River	872	3	4	single	0.46
1998	Syokanbetsu River		3	21	single or double	
	Syokanbetsu River		4	70	single	
	Sub-total	2,089		91		4.36
1999	Syokanbetsu River		3	92	small or large single	
	Syokanbetsu River		4	243	single or double	
	Syokanbetsu River		5	27	single	
	Sub-total	2,979		362		12.15
2000	Syokanbetsu River		3	18	single	
	Syokanbetsu River		4	59	small or large single	
	Syokanbetsu River		5	1	single or double	
	Sub-total	1,011		78		7.72
Total		6,951		535		7.70

Table 4. Predators feeding on juvenile chum salmon based on marked otolith with ALC.

Year	Predator	Sample	No. sample	No. marked juveniles or marked otolith
1999	fish (arabesque greenling)	stomach contents	72	5
1999	sea birds (gulls)	fecal materials	36	11

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Marking Salmonids with Strontium Chloride at Various Life History Stages

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Keywords: Salmonids, fish marking, strontium chloride, transgenerational marks, marking at fertilization

A long-standing challenge faced by fishery researchers and managers has been how to tag or mark small salmonids without injuring them. Numerous methods have been developed. Some like fin clipping and half-length CWTs (Thrower and Smoker 1984) require that each fish be individually handled. Others, for example, fluorescent spray marking, can be applied simultaneously to many fish but will leave some unmarked, are inherently stressful, and become more difficult to detect over time. Thermal marking of otoliths, on the other hand, appears to be a benign and universal way to mark embryonic salmonids. It does, however, require that fish be cultured or held for several days or longer. In some instances, it is desirable to quickly mark wild fish or those produced from spawning channels or other large incubation settings where thermal marking is not practical.

About forty years ago, Trefethen and Novotny (1963) recommended that stable isotopes be introduced into fishes to create recognizable marks. Since then investigators have introduced bone-seeking cations into fishes by feeding, injections, and immersion baths. For example, Behrens Yamada and Mulligan (1982, 1987) exposed salmonids to strontium chloride by holding them in dilute baths (1 ppm) or using strontium enriched diets. Their methods produced recognizable marks but took weeks to complete. We modified their approach by exposing salmonid fry to strontium baths containing up to 9000 ppm for 24 hrs (Schroder et al. 1995).

Calcified tissues from these fish were analyzed with inductively coupled mass spectrometry (ICPMS), and clear marks were discerned. ICPMS is a bulk analytical tool that requires examined specimens to be dissolved in ultra pure nitric acid before analyses can take place. Because entire structures, e.g., otoliths, centra, and so on, are analyzed, the relative concentration of introduced Sr becomes diluted as a fish grows. One way to circumvent such dilution is to use micro-probe analytical procedures. These techniques do not destroy a sample; instead they examine the elemental composition of a specimen in discrete and relatively small locations. Wave-length dispersive spectrometry (WDS) is one such method. In this case, a specimen is bombarded with primary electrons from an artificial source. These electrons interact with the specimen to produce backscattered electrons, secondary electrons, and x-rays. X-rays are detected and used to identify elemental composition, and backscattered electrons can be used to create backscattered electron images (BEIs). BEIs taken from otoliths collected from fish we marked clearly showed where strontium had been deposited at the time of marking. Such deposits will last throughout a fish's lifetime.

The value of any marking technique will be increased if multiple marks can be produced. Experiments showed that varying marking bath concentrations and exposure times creates distinct strontium marks. In addition, if the fish being marked can be held for several days it is possible to use multiple immersion events to create bar codes much like those employed in thermal marking.

Some work by Kalish (1990) suggested that it might be possible to create trans-generational marks in fishes by using strontium. He showed that female salmonids maturing in marine waters passively absorb strontium and incorporate it into their eggs. During early ontogeny, their offspring deposit this strontium into their otoliths. We speculated that artificial trans-generational marks could be induced in fishes by injecting high levels of dissolved strontium into gravid females (Buckley et al., personal communication). This marking approach was tried on several different species of marine fishes, where gravid females were injected with 9000, 30000 ppm Sr or a control saline solution. Otoliths collected from the offspring of these fish showed that those coming from mothers injected with Sr had elevated levels of this element in their otoliths.

A review of ion regulation in teleost eggs by Alderdice (1988) also suggested that salmonids could be marked with strontium at fertilization. He recounted that in salmon eggs, negatively charged proteins associated with the plasma membrane are inverted into the space between the chorion and plasma membrane immediately after activation. The proteins absorb water and also attract cations that are then bound to the proteins in the perivitelline space or moved across the plasma membrane into the yolk material. Absorbed cations mainly Ca, Na, and K may be essential for further embryonic development, particularly in waters that have low hydro-mineral content. The

capacity to absorb cations in this fashion continues for about three hours post fertilization, and after that the chorion becomes relatively impervious to ion exchange (Alderdice 1988). Given the proclivity to absorb cations, we felt that recently activated eggs would readily absorb strontium. To test this idea, we exposed newly fertilized chum salmon eggs to 5000, 2500, 500, and 50 ppm strontium chloride solutions for three hours. ICPMS analyses showed that an unambiguous increase in Sr occurred in otoliths collected from fry treated with the 5000 ppm Sr bath at fertilization. Moreover, recent WDS scans made on otoliths obtained from fry exposed to our marking baths disclosed that these fish possessed elevated levels of Sr in the inner portions of their otoliths. Hence, clear Sr marks can be induced on salmonids at fertilization by simply activating their gametes in solutions containing this element.

Salmonid alevins also appear to be an ideal life-history stage to mark with strontium because they are able to absorb ions through their gills, intestinal epithelia, and yolk sac (Behrens Yamada and Mulligan 1987). We exposed brown trout alevins to 1000 and 100 ppm marking solutions for 24 hrs or 4 hrs. All the alevins were subject to four separate marking episodes that were conducted at either two or five day intervals. BEI images of otoliths collected from these fish illustrated that all the treatments produced visible marks.

In summary, salmonid fishes can be marked in mass by using strontium solutions at diverse life history stages. Trans-generational marks also appear to be possible. The widespread use of this method will depend on its acceptance by federal regulatory agencies. Canadian researchers using our methods have marked large numbers of sockeye salmon. One of us (Pete Hagen) is working closely with the U.S. Food and Drug Administration (FDA) and has gained approval to mark 26 million sockeye fry. With continued FDA support we anticipate that strontium marking will be used in diverse research and management settings.

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Development of a New Stock Discrimination Tool for Naturally Spawning Sockeye Salmon (*Oncorhynchus nerka*) within Alberni Inlet from Stable Isotopic Composition of Otoliths

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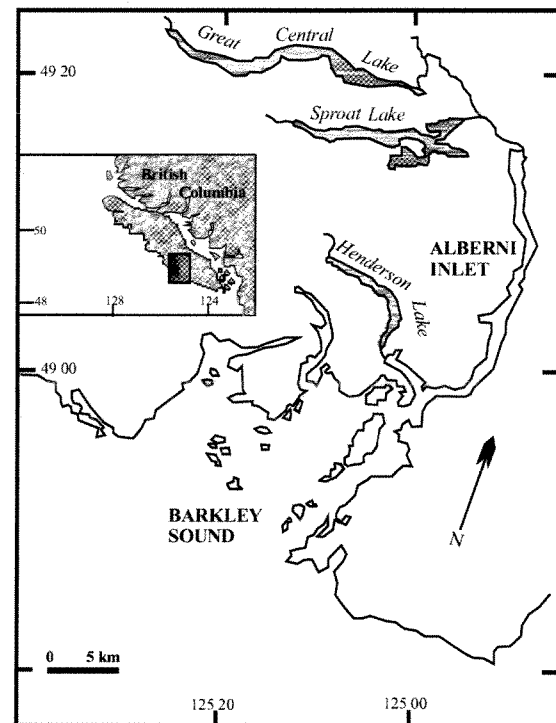
Keywords: Stock discrimination, natural otolith mark, sockeye salmon, lakes, $\delta^{18}\text{O}$ and $\delta^{13}\text{C}$

Great Central Lake, Sproat Lake, and Henderson Lake are tributaries to Alberni Inlet on the west coast of the Vancouver Island, British Columbia (Fig. 1). Two of these lakes, the Great Central and Sproat lakes support sockeye salmon (*Oncorhynchus nerka*) populations of magnitudes that support considerable commercial, recreational, and First Nations' harvest. The third lake, Henderson Lake, has a much smaller population of sockeye salmon which has not been able to support fisheries other than limited First Nations' harvest for food. With limited information on which to base decisions, managers attempt to limit harvest of Henderson Lake stock, during fisheries targeting Great Central and Sproat Lake sockeye stocks, through time and area restrictions. In this study we evaluate the use of stable isotopic composition of otoliths as a stock discrimination tool for improved management of Alberni Inlet sockeye fisheries.

Stable isotope ratio analysis ($^{18}\text{O}/^{16}\text{O}$ or $\delta^{18}\text{O}$, and $^{13}\text{C}/^{12}\text{C}$ or $\delta^{13}\text{C}$) of otoliths provides a new chemical tool for stock discrimination of sockeye salmon populations from different lakes. This is based on the hypothesis that otoliths are deposited in, or very close to oxygen isotopic equilibrium between the mineral aragonite and the ambient water where a fish lived, and that the isotopic fractionation of $^{18}\text{O}/^{16}\text{O}$ is temperature dependent (e.g., Urey 1947; Epstein et al. 1953; Devereux 1967; Grossman and Ku 1986; Kalish 1991). Carbon isotope ratios are generally precipitated in isotopic disequilibrium with the ambient water, but are influenced by metabolic sources of the fish and dietary shifts (e.g., Mulcahy et al. 1979; Schwarcz et al. 1998). Sockeye salmon life history usually includes spending the first one to two years in a nursery lake before smoltification. If the nursery lakes have different $\delta^{18}\text{O}$ and $\delta^{13}\text{C}$ values, these isotope signatures would constitute a natural otolith mark that can be used to identify the natal sources of different tributary stocks of the fish (Nelson et al. 1989; Gao 1997; Gao and Beamish 1999). Consequently, otoliths of sockeye salmon appear to be ideal as a proxy for isotope analysis.

For the first phase of a project in April–May 2000, we collected 77 otolith samples of smolts from the three neighbouring lakes (Great Central, Sproat, and Henderson) draining Alberni Inlet (Fig. 1). Microsampling was conducted by using the Dremel method (Gao 1999). The aragonite powder samples were taken from the surface of sockeye smolt otoliths from annual increments, and were analysed for their oxygen and carbon isotope ratios. The powder samples were then reacted with 100% phosphoric acid to release CO_2 gas into a "Kiel" carbonate preparation system that coupled directly with a Finnigan MAT 251 mass spectrometer. All the measurements were reported in the standard δ notation (‰): $\delta^{18}\text{O} = \{[(^{18}\text{O}/^{16}\text{O})_X / (^{18}\text{O}/^{16}\text{O})_S] - 1\} \times 1000$, for instance, where X is sample and S is standard (VPDB via NBS-19). Precision of the analysis is better than 0.1 ‰ for both $\delta^{18}\text{O}$ and $\delta^{13}\text{C}$ values.

Fig. 1. The location of the three neighbouring lakes in the Alberni Inlet of the Western Vancouver Island, British Columbia.



Among the three lakes studied, the data showed there were significant isotopic differences between Henderson Lake (-10.0 to -9.2 ‰ VPDB in $\delta^{18}\text{O}$ and -16.1 to -14.5 ‰ VPDB in $\delta^{13}\text{C}$) and either Great Central Lake or Sproat Lake (Fig. 2 and Table 1). There were no significant differences in the isotopic composition between Great Central Lake (-13.6 to -9.5 ‰ VPDB in $\delta^{18}\text{O}$ and -18.4 to -15.9 ‰ VPDB in $\delta^{13}\text{C}$) and Sproat Lake (-11.0 to -9.6 ‰ VPDB in $\delta^{18}\text{O}$ and -18.0 to -16.3 ‰ VPDB in $\delta^{13}\text{C}$). The maximum 3.5 ‰ VPDB differences in $\delta^{18}\text{O}$ of otoliths among the three lakes might be related to water temperature that could be in turn related to fish growth (Gao 1997). Henderson Lake may be cooler than other two lakes, which would cause the slower growth rate of smolts. Based on these results, the isotopic technique will provide a new tool for sockeye stock discrimination to meet management requirements for the Henderson Lake sockeye stock.

The benefits of the isotopic technique include the ability to identify the origin and life history of individual fish, the relatively high precision and accuracy compared to either DNA analysis or parasite analysis, and the relatively low cost. The next phase of our project will examine the interannual variability of isotopic composition from otoliths of spawning adults from the mixed stocks or areas to verify and complete the development of this stock discrimination tool.

Fig. 2. The oxygen and carbon isotopic composition of sockeye salmon smolt otoliths from the three lakes, showing the isotopic differences for Henderson Lake and the two northern lakes.

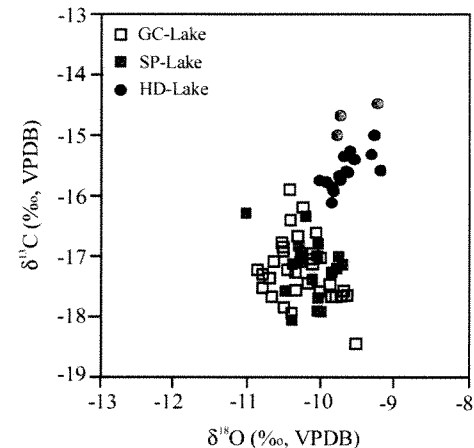


Table 1. ANOVA (one-way) of sockeye salmon smolt otoliths from three lakes in Alberni Inlet.

Isotope	Source	*DF	SS	MS	Fs	F. _{.05[2, 74]}
$\delta^{13}\text{C}$	Factor	2	46.847	23.423	95.11	3.15
	Error	74	18.225	0.246		
	Total	76	65.072			
$\delta^{18}\text{O}$	Factor	2	5.430	2.715	11.06	3.15
	Error	74	18.173	0.246		
	Total	76	23.604			

*Column headings: DF -degrees of freedom, SS -sums of squares, MS -mean squares, Fs -sample variance ratio, and F._{.05[2, 74]} -critical F-values.

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Compiling and Coordinating Salmon Otolith Marks in the North Pacific

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Keywords: Salmon, otolith mark, stock identification, coordination, database

Otolith marking is an effective tool to determine the hatchery origin of individual salmon in high seas and coastal waters. The North Pacific Rim countries (Canada, Japan, Russia, and USA) are employing this mass marking technique for anadromous salmon study and management. Although the annual total release of hatchery salmon is almost stable around five billion fish, the number of otolith marked salmon released from hatcheries has increased year by year, reaching one billion fish in 2000, which makes up 20% of the total releases (Fig. 1). The number of mark groups is also increasing every year, with 134 mark groups released in 2000 (Fig. 2). This rapid increase promises a high possibility of finding otolith marked fish in ocean samplings.

The otolith marking technology has performed well for salmon management programs in coastal fisheries (Hagen et al. 1995). Recently high seas researchers are focusing on the use of otolith marks for salmon population studies in offshore waters (Ignell et al. 1997; Carlson et al. 2000; Urawa et al. 2000). As the number of mark releases increases, however, it becomes a concern that duplicate otolith marks of salmon originating from different hatcheries will be encountered in ocean samples (Table 1).

There are practical limits on the number of mark patterns available for use, due to the narrow marking window at hatcheries (Hagen 1999). Complex patterns can increase marking costs for hatcheries and preclude a quick analysis of the patterns for timely stock management. Otolith marking programs usually have highest priority in near-shore management. As a result there has been little coordination within or between countries. In addition, there is no common database for otolith marks of salmon released from hatcheries in the Pacific Rim countries.

These circumstances led to the establishment of the North Pacific Anadromous Fish Commission (NPAFC) *Ad Hoc* Working Group on Salmon Marking in 1998. This group was soon turned into a permanent working group in 1999.

The roles of this working group are:

- (1) coordinating otolith mark patterns among member countries to minimize duplications,
- (2) creating an international database of otolith mark releases,
- (3) exchanging information on the development and standards of otolith mark techniques, and
- (4) exchanging information on the applications of otolith marks for salmon biology and stock management.

Fig. 1. Number of salmon released from hatcheries in the North Pacific Rim, 1995–2000. Number of total releases in 2000 is unknown.

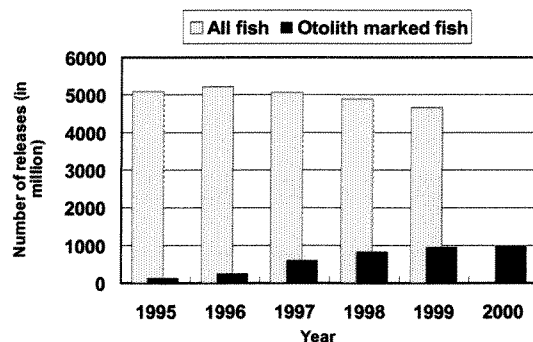


Fig. 2. Hatchery releases of otolith marked salmon by species, and number of mark groups in the North Pacific Rim countries, 1995–2000.

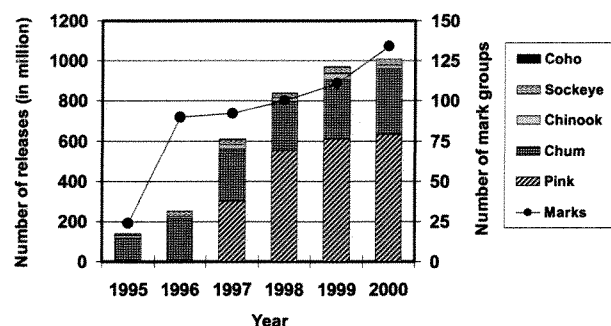


Table 1. Examples of duplicate otolith marks.

Species	Year Brood	RBr code	Stock	Region/Country
Chum	1995	1:1.3	Nitinat River	BC, Canada
Chum	1995	1:1.3	Ola River	Magadan, Russia
Chum	1996	1:1.6	Gastineau	Southeast Alaska, USA
Chum	1996	1:1.6	Wells River	Southcentral Alaska, USA
Chum	1999	1:1.5	Nitinat River	BC, Canada
Chum	1999	1:1.5	Ola River	Magadan, Russia
Pink	1999	1:1.4	A. F. Koernig	Southcentral Alaska, USA
Pink	1999	1:1.4	Gastineau	Southeast Alaska, USA
Sockeye	1998	1:1.4	Tahltan Lake	Southeast Alaska, USA
Sockeye	1998	1:1.4	Hidden Lake	Southcentral Alaska, USA

Base mark codes using a small number of rings are the simplest way to distinguish regions or countries. However, the assignment of country codes is difficult, because of the limited number of distinct codes (Hagen 1999; Munk 1999). To increase mark patterns, we need to develop other marking techniques such as strontium marking (Hagen and Volk 1998; Schroder et al. this volume).

At the present time, careful planning and communication among or within countries are important to minimize the probability of encountering duplicate marks. The working group members agreed on a practical process for mark coordination:

- (1) An otolith mark coordinator should be identified for each country or region. Initially the working group members would serve in this role.
- (2) Each country should submit to NPAFC an otolith mark plan applied for current brood year stocks by the end of July.
- (3) The mark coordinators should identify duplicate marks among the national plans.
- (4) Duplicate marks should be avoided by using modified codes or secondary characters.

In the case of system failure during marking operations, the mark coordinator must immediately notify the other coordinators and mediate the compromised code with other countries or regions.

An Internet-accessible database of otolith mark releases is indispensable for the efficient use of otolith marks in field surveys. We are planning to place an otolith mark database on the NPAFC web site (<http://www.npafc.org>) in cooperation with member countries that furnish information on otolith mark releases at the NPAFC annual meeting (Fig. 3). The database includes: ID#, brood year, date of release, species, country, state/province, region, agency, facility, stock, final release site, stage/size at release, number of releases, RBr code (Munk and Geiger 1998), hatch code (Hagen et al. 2000), and a graphic image of mark patterns (Fig. 4). Digital photo images of otolith marks may be also available through the Internet to facilitate the mark identification.

Fig. 3. The outline of Internet-accessible database for otolith mark releases.

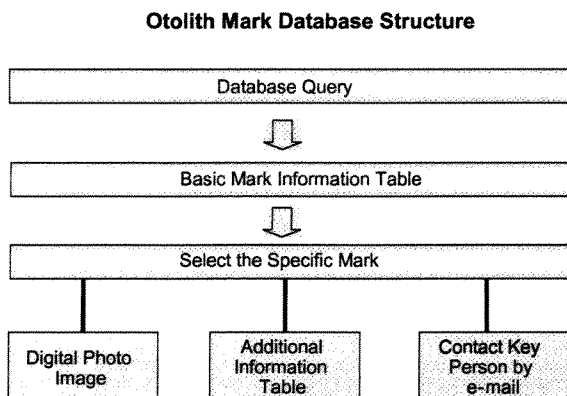


Fig. 4. An example of a database search on the Internet.

Otolith Mark Basic Information		Additional Information	
ID #	J98-01	ID #	J98-01
Mark Type	TM	Date Last Released	4/18/99
Year Brood	1998	State/ Province	Hokkaido
Year Released	1999	Region Released	Japan Sea coast
Species	CHUM	Agency	NASREC
Country	JAPAN	Facility	Chitose Hatchery
Stock	Chitose River	Release Site	Chitose River
RBr Code	1:1.4	Stage	early fed fry
Hatch Code	4H	Weight (g)	1.04
Prehatch Graphic		Length (mm)	52.8
Posthatch Graphic		Total Released	1,227,500
Digital Photo Image	yes	OM ID	chitose98chum -e
Additional Information	yes	Temp. Shift Direct.	down
Contact Person	M. Kawana	Comments	excellent mark

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Early Marine Growth and Habitat Utilization of Two Major Southeastern Alaska Chum Salmon Stocks, Based on Thermally Marked Otoliths Recovered 1997–2000

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Keywords: Salmon, juvenile, chum, marine, growth, habitat, southeastern Alaska

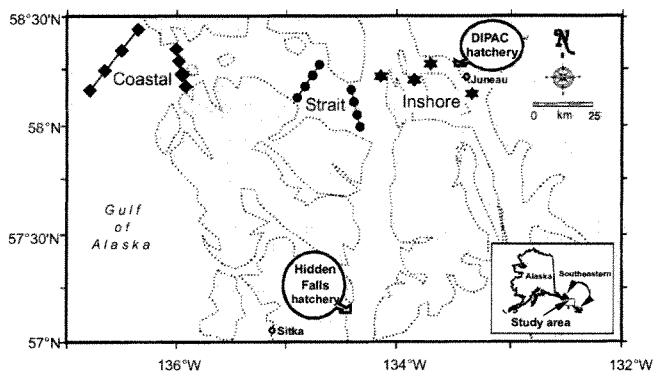
Identifying stock-specific life history characteristics of Pacific salmon (*Oncorhynchus* spp.) during their early marine residence period is critical to adequately assess interactions among biophysical parameters, marine survival, inter- and intra-specific competition, and coastal habitat utilization. The marine migration patterns and growth rates of juvenile salmon are not well documented (Walters et al. 1978). Such parameters must be derived from representative field data to establish accurate inputs for bioenergetics models (Ney 1993). These models can then be used to estimate prey consumption and growth potential of juvenile salmon and production capacity of their marine habitats.

Over the past century, chum (*O. keta*) and pink salmon (*O. gorbuscha*) have been the numerically dominant species in the commercial salmon fisheries in southeastern Alaska. Record annual harvests of chum (16 million fish) and pink salmon (78 million fish) occurred in the 1990s (ADFG 2000). From 1996 to 2000, the average annual commercial chum salmon ex-vessel value was the highest (\$30 million) of any salmon species. Record abundance of salmon in the region resulted from both increased hatchery production and favorable survival conditions for wild stocks. Over 77% of the chum salmon catch in the region is now produced by hatcheries, raising questions about the maximum production capacity of the marine environment of sustain both wild and hatchery juveniles. Accurate information on stock-specific life history characteristics can contribute to a better understanding of the factors limiting production.

The development and implementation of otolith-marking technology within the last decade have enabled scientists to track the migration and growth, and hatchery managers to evaluate ocean survival and harvest rates, of specific stocks of chum salmon in southeastern Alaska (Tersteeg and Focht 1998). Douglas Island Pink and Chum (DIPAC) Hatchery, a private non-profit (PNP), began production-scale otolith thermal marking in 1990 when salmon were marked at the eyed stage, and released as fed fry the following spring (Munk et al. 1993). Since then, up to 100% of all species of salmon produced by DIPAC have been otolith thermal marked. At another PNP, Hidden Falls Hatchery, about 60% of chum salmon are released thermally marked. Between the two major hatcheries, >130 million marked juvenile chum salmon are released in the region annually.

We initiated a study in the northern region of southeastern Alaska in 1997 to determine growth and habitat utilization of seaward migrating juvenile salmon (Orsi et al. 1997). Juvenile salmon were sampled with a surface trawl 1.5–65.0 km offshore during four periods each year from June to September 1997–2000 in three geographic habitats: “inshore” waters far inside the Alexander Archipelago, “strait” waters encompassing the major inside migration corridor of the region, and “coastal” waters adjacent to and in the Gulf of Alaska (Fig.1). The catch was identified and measured at sea, and juvenile salmon were frozen. About one third of the frozen chum were later weighed, and the right sagittal otolith was removed and mounted for thermal mark identification. Two “readers” independently examined each otolith to ensure accuracy; a third reader resolved any disagreements in identification.

Fig. 1. Sampling conducted in inshore, strait, and coastal habitats of southeastern Alaska, June–September, 1997–2000. Localities of the two hatcheries (DIPAC and Hidden Falls) releasing thermally marked chum salmon are shown.



Seasonal habitat utilization of chum salmon was described by catch per haul, and the percent composition of DIPAC, Hidden Falls, and unmarked stocks each month was pooled by years. The number of Hidden Falls fish was estimated by adjusting for the fraction marked; the estimate of unmarked stocks was the unmarked fish less the Hidden Falls adjustment. Size of chum salmon and time of release from the two facilities was derived from hatchery release information and voucher samples. Each year, a weighted mean size and time of release was computed by release group for each facility. To determine migration rates, distances were measured from seawater net pen localities to central points within each habitat. Migration rates for the two stocks were determined only for the strait and coastal habitats in July of each year, as insufficient numbers of both stocks were captured in inshore habitats.

Habitat utilization of the two major chum salmon stocks varied seasonally; most juvenile chum salmon migrated seaward through the habitats in June and July, with annual densities highest in strait habitat and lowest in inshore habitat (Fig. 2). Of the 3,823 chum salmon processed in the four-year period, 2% were from inshore habitat, 75% were from strait habitat, and 23% were from coastal habitat. Low catches in inshore habitat resulted from fish being abundant closer to shore than our gear sampled. The two hatchery stocks represented about 77%, 62%, and 28% of the juvenile chum salmon catch composition in inshore, strait, and coastal habitats, respectively. Migration rates of the two stocks, from the time of hatchery release in May until synoptic recoveries in strait and coastal habitats in July, averaged 0.9–3.6 km.d⁻¹ over 60–240 km distances (Fig. 3). These recoveries indicated slower migration rates and higher relative growth rates in strait compared to coastal habitat. Condition factors were also higher for both stocks in strait ($K = 9.3–10.3$, $\bar{x} = 9.8$) compared to coastal ($K = 8.8–9.6$, $\bar{x} = 9.2$) habitat. These differences were attributed to higher temperatures and zooplankton biomass in strait habitat, and to the shorter distance to strait habitat, which allowed energy to be allocated to growth rather than migration.

We examined stock-specific growth rates in strait habitat, where adequate numbers of thermal marks from both stocks were recovered each year. Instantaneous growth rates (% body wt.d⁻¹) were generally highest in the May–June period ($\bar{x} = 4.1$), intermediate in the June–July period ($\bar{x} = 3.7$), and lowest in the July–August period ($\bar{x} = 2.3$) (Fig. 4). Annual growth rates of fish in the strait habitat during the June–July period were highest in 1997, an El Niño year coinciding with high annual temperature and zooplankton biomass. Thermal mass marking of the two major hatchery chum stocks in the region has enabled us to determine stock-specific growth and habitat utilization patterns. We plan to use this information to estimate relationships among biophysical parameters, inter- and intra-specific competition, and marine survival, and as inputs for bioenergetics models to better define the salmon production capacity of southeastern Alaska.

Fig. 2. Seasonal stock composition and abundance of juvenile chum salmon in inshore, strait, and coastal habitats of southeastern Alaska, June–September 1997–2000.

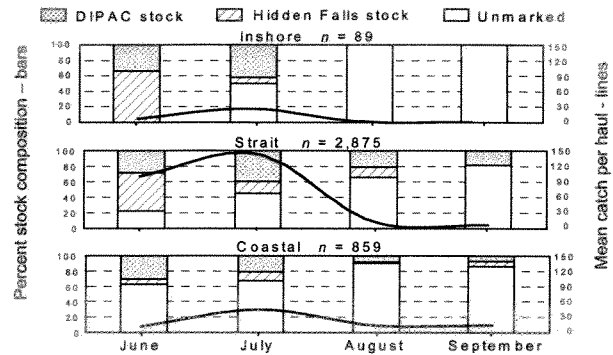


Fig. 3. Growth and migration rates of two juvenile chum salmon stocks released in May and recovered concurrently in strait and coastal habitats of southeastern Alaska in July, 1997–2000. Lines about growth rates are one standard deviation.

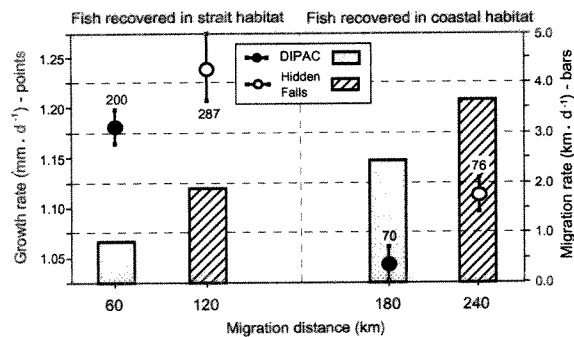
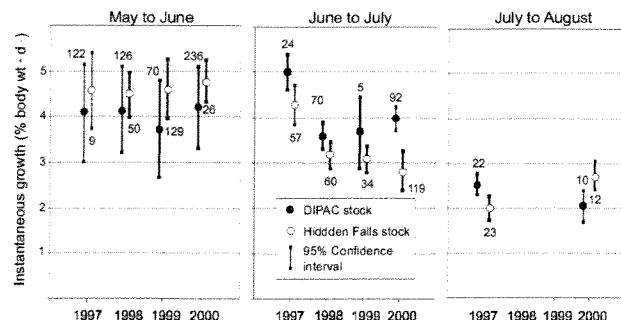


Fig. 4. Instantaneous growth rates of two juvenile chum salmon stocks at different time periods in strait habitat of southeastern Alaska, May–June, June–July, and July–August, 1997–2000.



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Application of Otolith Thermal Mass Marking in British Columbia, Canada

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Keywords: Pacific salmon, otolith, thermal mass marking applications, British Columbia, research, fisheries management

The first experiments in British Columbia, Canada, with otolith thermal mass marking of salmon were conducted at Robertson Creek Hatchery in 1989–1990. Large-scale (total production) otolith thermal marking of chinook was subsequently implemented at this same hatchery for the 1992 brood year. Since then thermal mass marking has gradually expanded to include chinook from many other hatcheries in B.C. including Nitinat Hatchery in 1992, Conuma Hatchery in 1994, Chilliwack Hatchery in 1995, and Quinsam Hatchery in 1996. The releases of otolith thermal mass marked chinook salmon from B.C. hatcheries increased from about 9 million in 1993 to more than 20 million by 1997 (Table 1). Releases of otolith thermal marked chum salmon (all from Nitinat River Hatchery) increased to about 35 million during the same period.

The number of hatcheries in B.C. that have implemented thermal otolith mass marking programs has continued to increase since the mid-1990s. Some B.C. hatcheries have also expanded marking in recent years to include routine thermal mass marking of coho and chum salmon, in addition to chinook salmon. However, the total number of thermal otolith marked fish released each year from all B.C. hatcheries has increased more slowly, mainly due to reduced spawner escapements of many B.C. salmon stocks that occurred during the 1995–2000 period. Additional details of the early implementation of thermal otolith mass marking in B.C., including the numerous thermal mark patterns that were applied to each stock each year, were provided in a previous report (Hargreaves et al. 1998).

Otolith thermal mass marking of salmon has been applied for several different purposes in B.C. Most of the early applications of the otolith mass marking technique were in support of scientific research activities and objectives. In recent years there has been increasing application of the otolith thermal mass marking for routine stock assessment and fisheries management purposes.

The research applications of otolith thermal mass marking have focused mainly on using this technique to confirm the hatchery origin of salmon smolts in situations where both wild and hatchery salmon co-migrate. This method has also been used to distinguish wild and hatchery smolts during downstream migrations (e.g., Somass River downstream trapping program; Wood et al. 1992), to identify juvenile hatchery and wild fish in estuaries (e.g., Campbell River and Somass Rivers), during the early sea life period (e.g., Nitinat Lake, Alberni Inlet and Barkley Sound), and in the open ocean (e.g., along the B.C. continental shelf; Perry et al. 1996). In recent years otolith thermal mass mark patterns have also been used to confirm the B.C. origin of maturing and adult chum salmon that have been captured in fisheries conducted in the Gulf of Alaska and Bering Sea.

Table 1. Number of thermal otolith marked salmon released each year from major British Columbia production facilities.

YEAR	SPECIES	B.C. SALMON STOCK					
		Robertson Creek	Nitinat River	Sarita River	Conuma River	Chilliwack River	Quinsam River
1993	Chinook	8,400,429	500,000	156,632	0	0	0
	Chum	0	0	0	0	0	0
1994	Chinook	6,939,205	6,195,122	210,776	0	0	0
	Chum	0	28,363,894	0	0	0	0
1995	Chinook	7,272,539	6,353,525	237,979	663,691	0	0
	Chum	0	30,831,080	0	0	0	0
1996	Chinook	8,273,553	4,073,259	7,086	390,040	813,089	0
	Chum	0	24,649,925	0	0	0	0
1997	Chinook	8,451,699	7,474,233	58,469	507,047	2,055,821	3,628,008
	Chum	0	31,941,437	0	0	0	0
1998	Chinook	8,927,415	6,341,195	307,914	176,496	1,921,522	2,712,900
	Chum	0	34,830,668	0	0	0	0

The reason that thermal otolith mass marking was first initiated in B.C. was to provide a new research tool for distinguishing between wild and hatchery chinook during the early sea life period. The main interest of this research was to examine differences in size and behaviour between wild and hatchery chinook that might provide insight into factors affecting survival rates, predation rates, marine growth, and possible competition for food between wild and hatchery chinook during the early sea life period. The Somass River chinook salmon stock, located on the west coast of Vancouver Island, B.C., was selected as the focus for this study. Robertson Creek Hatchery produced about 8–10 million chinook smolts each year, which represented 50–70% of the total (wild plus hatchery) chinook smolt production from the Somass River. Historical data indicated that very large interannual variations in marine survival rates had occurred since 1972 when Robertson Creek Hatchery first began production. The lowest survival rates tended to coincide with strong El Niño years, and it was speculated that increased predation or possibly increased competition for food between hatchery and wild chinook might explain the very poor marine survival rates that occurred in years with strong El Niño events. An extensive sampling program was conducted in Alberni Inlet and Barkley Sound from 1987 to 1993 to investigate these hypotheses. However, progress was initially hampered by the difficulty in distinguishing wild from hatchery chinook during the early sea life period. To overcome this problem experiments were initiated in 1989–1990 to test the practicality and utility of using the otolith thermal mass marking technique to mark all of the chinook released from Robertson Creek Hatchery. These first experiments in B.C. were based on the pioneering work that had previously been done on this technique in other locations and on other fish species (e.g., Volk et al. 1990). The immediate success of the initial experiments at Robertson Creek Hatchery led to the subsequent otolith thermal mass marking of all chinook salmon released from Robertson Creek Hatchery since 1992. This allowed all hatchery chinook to be easily distinguished from wild chinook, even during the early sea life period when variations in growth rates and migration rates quickly obscured the initial differences (e.g., size) between hatchery and wild chinook. Results obtained using the otolith thermal mass mark information showed clear differences in the distribution, abundance, growth rates, migration rates and predation rates of wild and hatchery chinook in Alberni Inlet and Barkley Sound. For example, the numbers of wild and hatchery chinook were approximately equal in the near-shore areas (sampled with beach seines) in 1993, but virtually all of the juvenile chinook that were caught simultaneously in the same time periods in open water areas (using a purse seine) were hatchery fish (Table 2).

Otolith thermal mass marking of salmon has also recently been gaining increasing importance in B.C. as a powerful new tool for stock assessment and fisheries management. Current applications in B.C. include: (1) assessment of the contribution of fish from various hatcheries to local mixed-stock fisheries, (2) allowing independent evaluation of potential biases in alternate marking methods (coded-wire tag, multiple fin-clip, etc.), (3) assessing the hatchery contribution to mixed hatchery and wild spawning populations, (4) determining the straying rates of salmon released from major hatcheries, (5) estimating exploitation rates for key salmon stocks, and (6) evaluating the effectiveness of various fisheries management actions. An example of the latter application is provided in more detail below.

In 2000 the return of chinook to both wild and hatchery stocks along the west coast of Vancouver Island (WCVI), B.C., was forecasted to be extremely low. In order to provide adequate protection to conserve these stocks and achieve even minimal escapement levels, conventional fisheries management would have required complete closure of the recreational fishery along the entire WCVI. This would have been devastating to local recreational fishermen and also to the local communities and businesses, which are heavily dependent on the fishing tourist industry. In consultation with industry representatives, Fisheries and Oceans Canada (DFO) agreed to examine possible alternative management approaches. DFO staff examined historical data from recoveries of chinook with coded-wire tags (CWTs) caught by all fishing gear sectors in previous years when chinook stocks were more abundant. This analysis confirmed that the recreational fishery typically caught substantial numbers of chinook along the entire WCVI. However, this analyses also indicated that very few of the chinook captured by recreational fishermen at locations farther than one nautical mile from shore originated from local chinook stocks spawning

Table 2. Origin of chinook salmon captured in near-shore and open water locations during the early sea-life period in 1993 in Alberni Inlet and Barkley Sound, B.C., hatchery fish were identified using otolith thermal mass mark patterns. Chinook that could not be identified clearly as either hatchery or wild based on the otolith thermal mass mark alone are classified as "not certain".

Time Period	Beach Seine			Purse Seine		
	Hatchery	Wild	Not Certain	Hatchery	Wild	Not Certain
26–27 May	0	62	0	0	1	2
02–13 June	100	85	17	52	3	0
14–25 June	109	118	8	27	5	3
Totals:	209	265	25	79	9	5

along the WCVI. Based on this information DFO implemented a new chinook “conservation corridor” along most areas of the WCVI, which extended from the surf line to approximately one nautical mile offshore (Fig. 1). Within this conservation corridor recreational fishing for all species salmon was prohibited. However, outside (farther offshore) of this corridor recreational fishing and retention of chinook were permitted. The inner “surf line” boundary generally corresponds to the region very close to the shore line where surf (breaking waves) typically occur. The “surf line” boundary is more formally defined and legally described in DFO regulations for the Pacific Region.

Results from otolith thermal mass marks provided the only data available for assessing the effectiveness of this new chinook conservation corridor. Two major concerns about the new conservation corridor management approach in 2000 were that the chinook returning in 2000 might migrate farther offshore in 2000, or that recreational fishermen might fish illegally inside the corridor, and catch and retain chinook from local WCVI stocks. However, all of the chinook released from the three major hatcheries along the WCVI (Robertson Creek, Nitinat, and Conuma) have been otolith thermal mass marked since 1994. Thus all of the adult chinook that returned from these three stocks in 2000 to spawn were otolith thermal mass marked, with distinctive patterns for each hatchery. Earlier work had confirmed that the chinook from these three main hatchery stocks are also representative (marine survival rates, migration routes and timing, etc.) of the wild chinook stocks along the WCVI. The DFO implemented an appropriate sampling program to obtain otoliths from chinook that were caught during August and September 2000 by recreational fishermen along the WCVI. A total of 485 chinook otoliths were examined for the thermal mass mark patterns that had been applied by the three major WCVI hatcheries. Only one of these chinook was from Robertson Creek Hatchery. These results confirmed that the conservation corridor was in fact very effective, and allowed the continuation of an important recreational fishery while also providing adequate protection for local WCVI chinook stocks. It should be noted that evaluation of this new management strategy could not be done using the conventional CWT method, due to the very small number of chinook that were caught with CWTs.

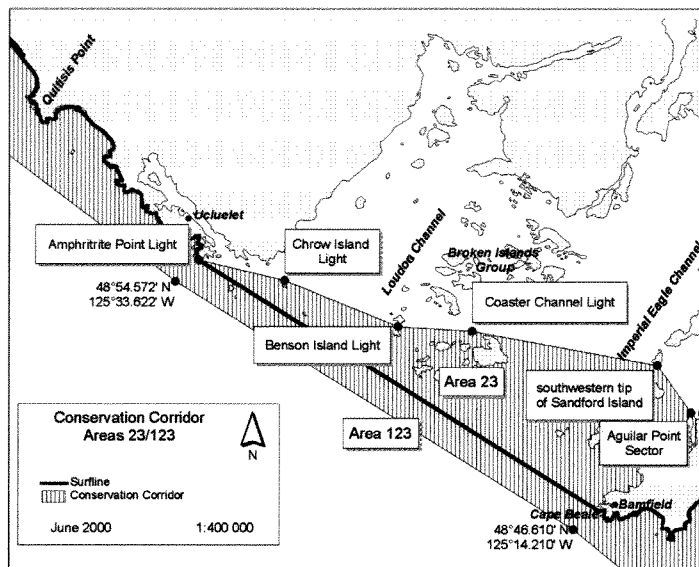
The implementation and application of otolith thermal mass marking will likely continue to grow in B.C. The use of this method has changed from predominantly a research focus in the early years to mainly stock assessment and fisheries management in recent years.

To a large degree this transition has been made possible by the consistent long-term thermal marking programs that have been implemented at many B.C. hatcheries. Maintaining routine thermal marking programs at these hatcheries for many years has resulted in thermal mass marking of most or all of the adult production that is now returning each year. This in turn provides the opportunity to use these thermal marked salmon in ways that were not even anticipated when these marking programs were initiated, e.g., the use of otolith thermal mark patterns to verify the effectiveness of the conservation corridor for chinook along the WCVI in 2000 (described above). This application was not even anticipated when the thermal marks were originally applied. The conservation corridor would not have been considered by DFO, and the recreational fishery likely would have simply been closed if the otolith thermal mark information was not available to assess the effectiveness of this management approach.

Otolith thermal mass marking in B.C. will likely continue to expand to include other species and more hatcheries. Originally only chinook were thermally marked in B.C. hatcheries. The thermal marking program subsequently expanded to include chum salmon (e.g., Nitinat Hatchery) and more recently coho salmon (e.g., Robertson Creek Hatchery). Additional hatcheries are also currently planning to implement otolith thermal marking programs (e.g., Fraser River system hatcheries in the B.C. interior). There is also steadily growing interest in many small hatcheries throughout B.C. that have low annual production (e.g., Public Involvement Program hatcheries) to use otolith thermal mass marking to evaluate their contribution to local fisheries and spawner escapements.

Continued expansion of otolith thermal mass marking in B.C. and other countries poses some significant challenges. The number of “useable” thermal marks that can be applied is quite limited, and there is already

Fig. 1. Map showing the boundaries of the new conservation corridor (in statistical areas 23 and 123 only) that was implemented by DFO in 2000 to protect adult chinook returning to spawn along the entire west coast of Vancouver Island.



competition for the “best” mark patterns even among B.C. hatcheries. This problem is further amplified if there is any concern in a particular application that thermally marked fish may be encountered (e.g., in mixed-stock fisheries along the B.C. coast) that originate from hatcheries outside B.C. Within Canada there is also inadequate coast-wide coordination of thermal marking programs. In many cases this method is used to address only local questions and problems (e.g., assessing hatchery contribution to spawning populations), and so there is little interest or concern about thermal marks that might be applied by other hatcheries or encountered in more distant locations. However, this potentially can diminish the utility of the otolith mass method in other locations (mixed stock ocean fisheries) if the mark patterns used by all hatcheries along the Pacific Coast are not adequately coordinated. The authors are encouraged by and support the recent initiative through the North Pacific Anadromous Fish Commission for the international coordination of otolith thermal marking patterns and the international sharing of relevant thermal mark data.

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Variations in Catch Per Unit Effort of Thermally Marked Pink and Chum Salmon Juveniles in the Gulf of Alaska during 1996 and 1998 in Relation to Adult Hatchery Salmon Returns

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Keywords: Thermal otolith mark, pink salmon, chum salmon, Gulf of Alaska, CPUE

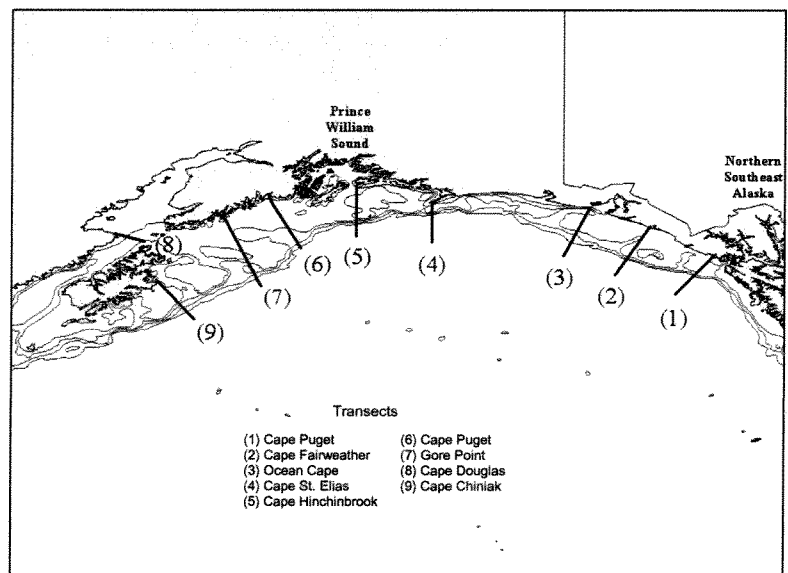
Thermal marking of salmonid otoliths has become an important, cost-effective tool to identify hatchery salmon at sea. In recent years, releases of thermally marked salmon into the North Pacific Ocean from hatcheries in Washington, British Columbia, and Alaska have numbered in the billions. The large numbers of thermally marked salmon released into the North Pacific Ocean have greatly increased the probability of recovering marked salmon during high-seas sampling thus potentially providing information on the biology and management of Pacific salmon (Ignell et al. 1997; Farley and Munk 1997; 1998; Farley et al. 1999; Kawana et al. 1999; Carlson et al. 2000; Farley and Carlson 2000; Kawana et al. 2000; Urawa et al. 2000).

In 1996, the Ocean Carrying Capacity (OCC) program at the Auke Bay Laboratory, National Marine Fisheries Service initiated a comprehensive program to study the distribution, migration, origin, size, growth, and diet of juvenile, immature, and maturing salmonids in the Gulf of Alaska (GOA; Carlson et al. 1996). One objective of this ongoing program is to collect and analyze otoliths from salmonids to determine hatchery origin of these fish and to partition hatchery from wild stocks in our samples. In this paper we summarize information on catch per unit effort (CPUE) of thermally marked juvenile pink and chum salmon caught in the GOA during July and August 1996 and 1998 in relation to adult hatchery pink and chum salmon returns one and three years later, respectively.

Juvenile salmon were captured along transects within the coastal waters of the GOA during July and August 1996 and 1998 (see Carlson et al. 1996 and 1998 for details; Fig. 1). The salmon were frozen whole and brought back to the laboratory where sagittal otoliths were removed from the juvenile salmon and examined for thermal marks. Otolith thermal mark patterns from juvenile chum salmon were compared to voucher specimens collected from Gastineau and Hidden Falls hatcheries located in northern Southeast Alaska (NSEAK; Fig. 2A); otolith thermal mark patterns from juvenile pink salmon were compared with voucher specimens collected from Armin F. Koernig, Cannery Creek, Solomon Gulch, and Wally H. Noerenberg hatcheries located in Prince William Sound (PWS; Fig. 2B).

Yearly CPUE of juvenile PWS and NSEAK hatchery pink and chum salmon was estimated by summing the numbers of these salmon caught in each haul over the entire survey then dividing by the total number of trawl hours. The CPUE data was then compared to returns of adult PWS hatchery pink salmon 1 year later and adult NSEAK hatchery chum salmon 3 years later. To make qualitative comparisons, we assumed that our 1996 and 1998 surveys sampled through the peak of the juvenile salmon migration leaving PWS and NSEAK and entering the continental shelf of the GOA.

Fig. 1. Transects sampled by the OCC program during July and August 1996 and 1998.



Plots of hatchery pink and chum salmon CPUE by transect and year resembled a bell-shaped curve indicating our surveys sampled through the peak migration of juvenile PWS hatchery pink (Figs. 3A and B) and NSEAK hatchery chum salmon (Figs. 4A and B). Annual differences in CPUE of juvenile hatchery pink salmon was lower during 1996 than 1998 matching the pattern of returning hatchery pink salmon to PWS during 1997 (lower) and 1999 (higher) (Fig. 5A). CPUE of juvenile hatchery chum salmon was also lower during 1996 than 1998 (Fig. 5B), suggesting that the 2001 return of NSEAK hatchery chum salmon may be higher than the 1999 return of 8 million.

Our analysis of juvenile PWS hatchery pink salmon catch suggests that CPUE, as a measure of relative abundance of juvenile salmon along the continental shelf of the GOA, may prove useful in predicting adult PWS pink salmon returns one year later. The large CPUE of juvenile PWS hatchery pink salmon found during 1998 corresponds to a record return of pink salmon to PWS hatcheries during 1999. Additional years of sampling will be needed, however, to establish the significance of the association. We note that the large CPUE of PWS pink salmon during our 1998 survey does not appear to be related to the number of hatchery pink salmon released. Hatchery releases of pink salmon in PWS were larger during 1996 (642 million) than during 1998 (542 million) (McNair 1997; 1999), indicating annual differences in early marine survival of juvenile pink salmon within PWS.

Fig. 2. Locations of Southeast Alaska hatcheries (A) and Prince William Sound hatcheries (B). Figures courtesy of the Alaska Department of Fish and Game.

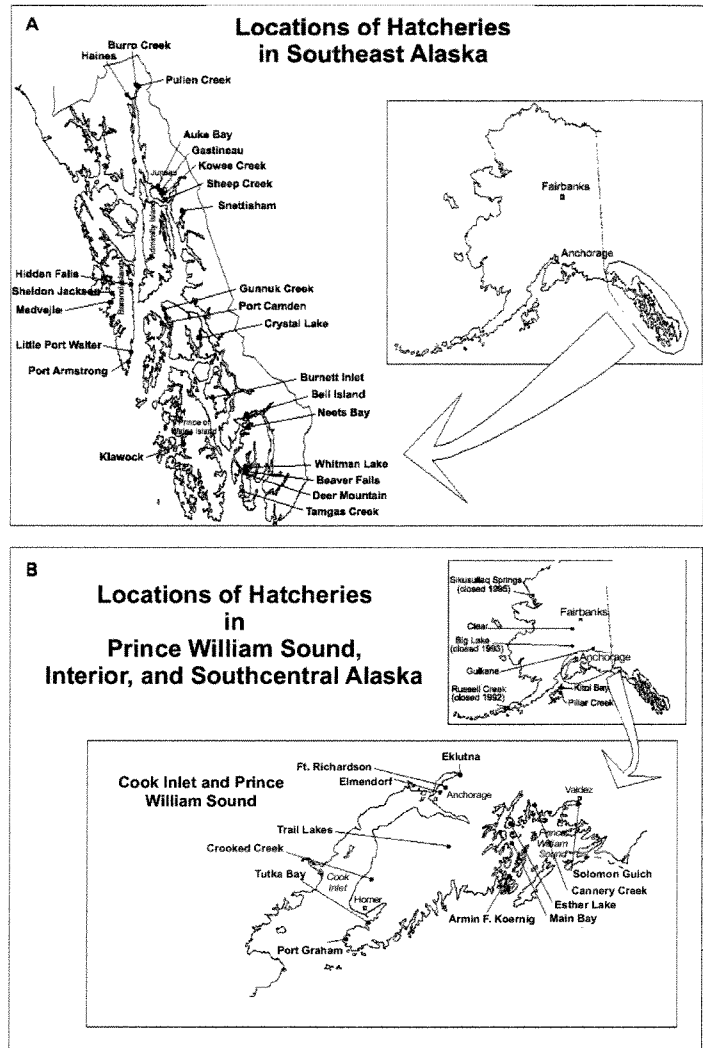


Fig. 3. Catch per unit effort of juvenile Prince William Sound hatchery pink salmon along transects sampled by the OCC program during 1996 (A) and 1998 (B).

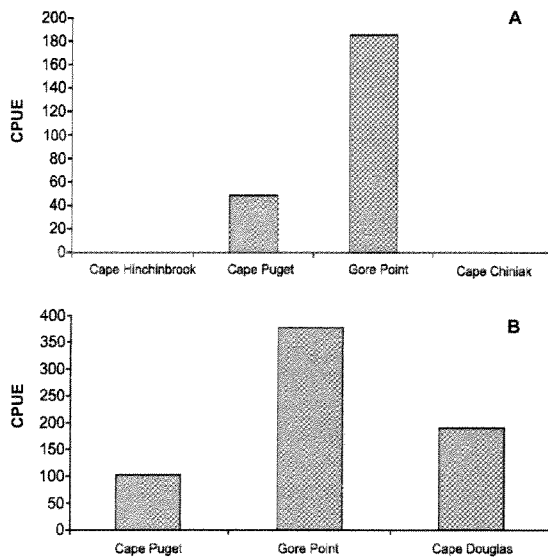


Fig. 4. Catch per unit effort of juvenile northern Southeast Alaska hatchery chum salmon along transects sampled by the OCC program during 1996 (A) and 1998 (B).

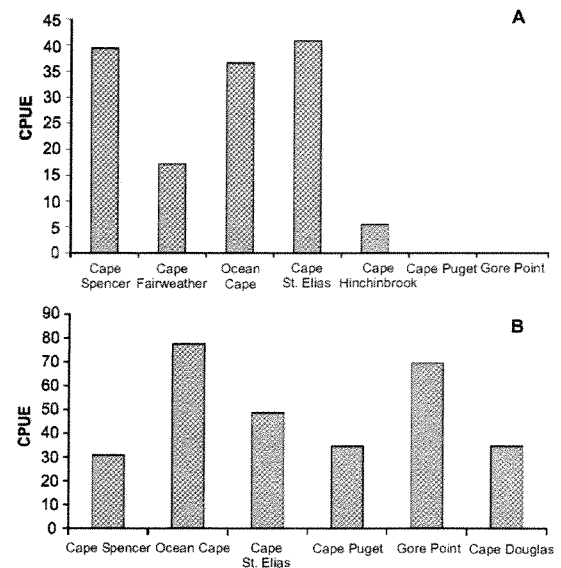
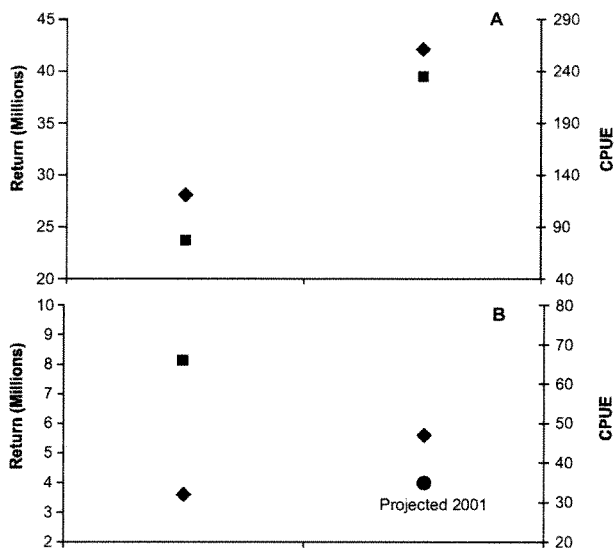


Fig. 5. Catch per unit effort (CPUE) of juvenile Prince William Sound hatchery pink (A) and northern Southeast Alaska hatchery chum (B) salmon (diamonds; first year) in relation to adult hatchery returns (squares; second year and three years, respectively). The circle in Fig. 5B represents the projected return of northern Southeast Alaska hatchery chum salmon during 2001.



If the CPUE-adult return relationship shown in the pink salmon data holds true and is applicable to chum salmon, then the higher CPUE of juvenile NSEAK hatchery chum salmon observed during 1998 could indicate a larger adult return of hatchery chum salmon to NSEAK during 2001 than observed during 1999. The forecast for adult hatchery chum salmon returns to NSEAK in 2001, however, predicts a nearly 50% reduction over 1999 returns (McNair 2001). We note that NSEAK hatcheries only mark a portion of their chum salmon during incubation, increasing the variation in and covariance between juvenile NSEAK hatchery CPUE from our survey and adult hatchery returns to this region 3 years later.

The forecast could be artificially low due to possible changes in maturation of chum salmon during ocean residence. The forecast includes a sibling model based on the number of returning 3-year old chum salmon in the prior year. Three year old chum salmon were noticeably absent in both the wild and hatchery chum salmon returns to southeast Alaska during 2000. Absence of 3-year old chum salmon is often a good indicator of brood strength, but not always. In some years, either due to competition for food resources or reduced food

abundance, chum salmon mature at older ages. Could the high densities of pink and chum salmon juveniles seen in the summer of 1998 and the small size of adult pink salmon returning in 1999 be an indicator that chum salmon from the 1997 brood are going to mature later? The age and abundance of chum salmon returns to southeast Alaska in 2001 should be very interesting to observe.

Our results demonstrate that sufficient numbers of thermally marked hatchery salmon can be recovered during coastal salmon surveys to provide significant new stock-specific information on distribution, migration, and CPUE. We plan to continue analyzing stock specific CPUE during future July–August (2001–2004) coastal GOA surveys and relating CPUE to adult returns. We encourage all agencies to mark 100% of their hatchery releases, so that future marine sampling efforts will yield a better picture of hatchery and wild salmon interactions.

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High-Seas Ocean Distribution of Alaskan Hatchery Pink Salmon Estimated by Otolith Marks

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To investigate the ocean distribution of Alaskan hatchery pink salmon (*Oncorhynchus gorbuscha*), we examined otoliths for thermal marks from maturing pink salmon caught in the Gulf of Alaska and central North Pacific Ocean in the summers of 1998–2000. Pink salmon captured in the Gulf of Alaska were identified as originating from hatchery locations in Prince William Sound (PWS), Alaska. Large numbers of Alaskan hatchery pink salmon have been thermally marked and released (Geiger and Munk 1998). Thermal marks are an effective tool for stock identification in high-seas and coastal waters (Farley and Munk 1997; Kawana et al. 1999a; Carlson et al. 2000; Urawa et al. 2000a). Salmon otoliths are thermally marked by exposing fish to alternating (relatively lower and higher) temperatures (Volk et al. 1990), whereby a "thermal ring" (dark ring) is induced by exposure to the lower temperature (Munk et al. 1993). Different thermal marks are made by varying the number and spacing of thermal rings in the RBr notation in Alaskan hatcheries (Munk and Geiger 1998).

Thermally-marked pink salmon were released from four PWS hatcheries: Armin F. Koernig Hatchery (AFK), Wally H. Noerenberg Hatchery (WHN), Cannery Creek Hatchery (CCH), Solomon Gulch Hatchery (SGH), and one southeast Alaskan hatchery: Gastineau Hatchery (GH). For pink salmon brood years 1996, 1997, and 1998, the numbers of thermally-marked fry released were 489.60, 551.08, and 607.80 million, respectively. More than 98% of thermally-marked pink salmon were released from PWS hatcheries, where 100% of all hatchery pink salmon released were thermally marked. Each hatchery stock had unique thermal mark patterns coded in RBr notation, with the exception of brood year 1996 where RBr code (1:1.4) was used for both GH and one of two stocks of AFK (Geiger and Munk 1998; Hagen et al. 1999). Pink salmon were caught by gillnets (non-selective varied research mesh, traditional commercial mesh, and experimental mesh) during June and July along two offshore transects (145°W and 165°W) in the Gulf of Alaska by the T/S *Oshoro maru* and one offshore transect (180°) in the central North Pacific Ocean by the R/V *Wakatake maru*. The total catches were 813, 1297, and 574 maturing pink salmon in 1998, 1999, and 2000, respectively (Walker et al. 1998; Kawana et al. 1999b; Yamaguchi et al. 1999; Urawa et al. 2000b; Yamaguchi et al. 2000). Fork length (mm), body weight (g), sex, and gonad weight (g) were recorded and sagittal otoliths were collected from 383, 778, and 349 pink salmon in 1998, 1999, and 2000, respectively. Scales were also collected for age determination. The catch per unit effort (CPUE) was calculated as total catch (number of fish) per one set of the non-selective gillnet (30 tans, 1 tan = 50 m long and approximately 6 m depth; Takagi 1975; Walker et al. 1998; Kawana et al. 1999b; Yamaguchi et al. 1999; Urawa et al. 2000b; Yamaguchi et al. 2000). A gonadosomatic index (GSI) was calculated as $100 \times \text{gonad weight (g)} / \text{body weight (g)}$ to examine maturity. Fork length, body weight, gonad weight, and GSI were compared by Kruskal-Wallis test and Scheffe's test. The left sagittal otoliths were mounted on individually labeled glass slides using thermoplastic cement. If the left otolith was missing or ground through the primordia, then the right otolith was used. Otoliths were ground to expose the primordia, and examined under a microscope. Thermal marks were recorded in the RBr notation. If the same RBr code was used for a brood year class at different hatcheries, then microstructural patterns were compared with voucher specimens that were collected from the hatcheries before release.

One hundred and fifty-one thermal marks were found among 1,510 maturing pink salmon examined (Table 1). Along the 145°W transect, 25 thermally-marked fish were found (8.1%, $n = 307$) in 1998, 86 marked fish were found (15.1%, $n = 568$) in 1999, and 34 marked fish were found (12.0%, $n = 284$) in 2000. The thermally-marked salmon were from three of four pink salmon hatcheries in PWS: AFK ($n = 57$), CCH ($n = 39$), and WHN ($n = 49$). Along the 165°W transect, only four thermally-marked fish were found (5.3%, $n = 76$) in 1998, and two marked fish were found (2.6%, $n = 78$) in 1999. Their origins were AFK ($n = 3$), CCH ($n = 2$), and WHN ($n = 1$). Along the 180° transect, no thermally-marked fish were found in 1999 ($n = 132$) and 2000 ($n = 65$). Marked pink salmon released from SGH and GH were not found along either transect. PWS hatchery pink salmon were widely distributed from 49°N to 56°N along the 145°W transect and from 47°N to 50°N along the 165°W transect, but not

Table 1. Composition of pink salmon hatchery origins caught along the 145°W, 165°W, and 180° transects during June and July 1998–2000. PWS, Prince William Sound; A, Armin F. Koernig Hatchery; C, Cannery Creek Hatchery; W, Wally H. Noerenberg Hatchery.

Latitude (N)	1998			1999			2000								
	PWS			PWS			PWS								
	A	C	W	A	C	W	A	C	W	unmarked	%				
145°W transect															
56°00'	3	3	4	62	12.5	10	6	11	117	18.8	3	3	0	20	23.1
55°00'	3	4	3	50	16.7	10	4	7	74	22.1	3	5	4	38	24.0
54°00'	-	-	-	-	-	12	2	6	76	20.8	1	3	4	49	14.0
53°00'	-	-	-	-	-	1	2	3	56	9.7	1	0	1	27	6.9
52°00'	1	0	0	27	3.6	2	3	2	47	13.0	1	0	1	33	5.7
51°00'	1	0	1	58	3.3	2	0	1	56	5.1	0	0	0	42	0
50°00'	0	2	0	16	11.1	0	1	1	56	3.4	2	1	0	40	7.0
49°00'	0	0	0	69	0	-	-	-	-	-	1	0	0	9	10.0
48°00'	-	-	-	-	-	-	-	-	-	-	0	0	0	1	0
Total	8	9	8	282	8.1	37	18	31	482	15.1	12	12	10	250	12.0
165°W transect															
50°00'	1	0	0	17	5.6	2	0	0	42	4.5	-	-	-	-	-
48°30'	0	2	0	4	33.3	-	-	-	-	-	-	-	-	-	-
47°00'	0	0	1	29	3.3	0	0	0	19	0	-	-	-	-	-
45°30'	0	0	0	22	0	0	0	0	12	0	-	-	-	-	-
44°00'	-	-	-	-	-	0	0	0	3	0	-	-	-	-	-
Total	1	2	1	72	5.3	2	0	0	76	2.6	-	-	-	-	-
180° transect															
47°30'	-	-	-	-	-	0	0	0	83	0	0	0	0	11	0
47°00'	-	-	-	-	-	0	0	0	36	0	0	0	0	17	0
46°00'	-	-	-	-	-	0	0	0	8	0	0	0	0	21	0
45°00'	-	-	-	-	-	0	0	0	3	0	0	0	0	7	0
44°00'	-	-	-	-	-	0	0	0	1	0	0	0	0	3	0
43°00'	-	-	-	-	-	0	0	0	0	0	0	0	0	4	0
42°00'	-	-	-	-	-	0	0	0	1	0	0	0	0	1	0
41°00'	-	-	-	-	-	-	-	-	-	-	0	0	0	1	0
Total	-	-	-	-	-	0	0	0	132	0	0	0	0	65	0

along the 180° transect. However, 83% of total CPUEs of PWS hatchery pink salmon along the three transects caught by non-selective gillnets were represented by catches in northern waters (54–56°N) of the 145°W transect (Fig. 1). Among the three hatchery stocks caught along the 145°W transect, differences in the body and gonad measurements were not significant ($p > 0.05$; Kruskal-Wallis test, by year and sex). Among three sampling years along the 145°W transect, fork lengths and body weights of PWS hatchery females in 1998 were significantly different from 1999 and 2000 ($p < 0.001$, Scheffe's test), but no significant difference was observed among males ($p > 0.05$, Table 2). There was also a significant difference in GSI of PWS hatchery males between 1998 and 2000 ($p < 0.05$, Table 2).

All thermally-marked pink salmon detected in the present study were from PWS. In April and May, PWS hatchery pink salmon were distributed in southern (43–48°N, 145°W and 42–46°N, 165°W) and northern (50–55°N, 145°W) waters in 1998 and 1999 (Carlson *et al.* 2000). But in June and July, they were absent from the southern waters and became increasingly abundant in the northern part (54–56°N) of the 145°W transect corresponding to

Fig. 1. Catch per unit effort (CPUE) of thermally-marked pink salmon released from Prince William Sound hatchery caught along the 145°W (left) and 165°W (right) transects in the Gulf of Alaska during June and July 1998–2000. The CPUE values are based on the catch (number of fish) per one set of a non-selective varied research mesh gillnets (30 tans).

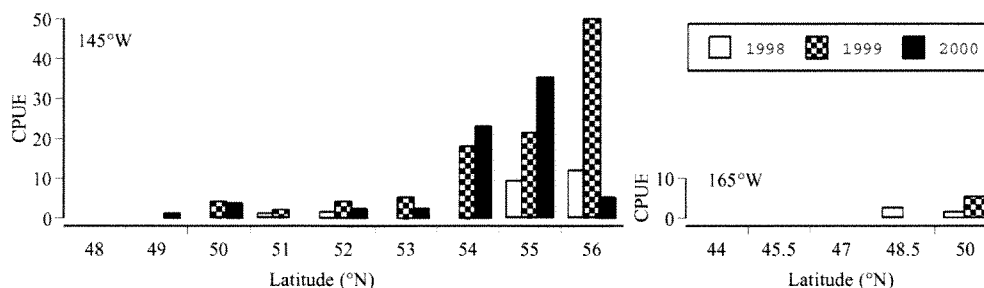


Table 2. A comparison of body and gonad measurements of maturing thermally-marked Prince William Sound hatchery pink salmon caught along the 145°W transect in the Gulf of Alaska during July 1998–2000. Values are given as the mean \pm SD. Numbers in parentheses are sample sizes. FL, fork length (mm); BW, body weight (g); GW, gonad weight (g); GSI, gonadosomatic index = 100 \times gonad weight (g) / body weight (g).

Sex	Measurement	1998	1999	2000	Probability		
					1998–1999	1998–2000	1999–2000
Female	FL	483 \pm 24 (12)	448 \pm 15 (48)	439 \pm 35 (15)	p < 0.001	p < 0.001	p > 0.05
	BW	1328 \pm 227 (12)	1066 \pm 122 (47)	979 \pm 169 (15)	p < 0.001	p < 0.001	p > 0.05
	GW	47 \pm 21 (12)	44 \pm 12 (48)	35 \pm 10 (15)	p > 0.05	p > 0.05	p > 0.05
	GSI	3.54 \pm 1.41 (12)	4.08 \pm 0.78 (47)	3.62 \pm 0.87 (15)	p > 0.05	p > 0.05	p > 0.05
Male	FL	460 \pm 16 (13)	442 \pm 22 (38)	449 \pm 27 (19)	p > 0.05	p > 0.05	p > 0.05
	BW	1111 \pm 123 (13)	1036 \pm 140 (38)	1013 \pm 218 (19)	p > 0.05	p > 0.05	p > 0.05
	GW	14 \pm 6 (13)	19 \pm 8 (38)	21 \pm 13 (19)	p > 0.05	p > 0.05	p > 0.05
	GSI	1.26 \pm 0.53 (13)	1.82 \pm 0.72 (38)	2.01 \pm 0.97 (19)	p > 0.05	p < 0.05	p > 0.05

their northward homeward migration. Year-to-year differences were observed in the body size of PWS hatchery females because half of the sampled fish in 1998 were larger in fork length or body weight than any fish in 1999 and 2000. This may be a result of differences in sample sizes or characters of PWS hatchery pink salmon caught in 1998. In this study, maturing pink salmon were sampled in the Gulf of Alaska during their homeward migration, and incidence of otolith thermal marks was used to identify their hatchery origin. Our results show that high-seas stock identification, abundance, and migration timing data are available prior to commencement of commercial harvest, and therefore may aid the management of PWS hatchery pink salmon.

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Estimating the Abundance and Distribution of Locally Hatchery-Produced Chinook Salmon Throughout a Large River System Using Thermal Mass-Marking of Otoliths

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The role of hatchery production in the management of Pacific salmon (*Oncorhynchus spp.*) has undergone extensive review and scrutiny in recent years (for example, Schramm and Piper 1995). Among the potential hazards to natural production caused by the presence of hatchery fish are deterioration of wild stock gene pools due to incorporation of genes from hatchery strays into wild populations (Campton 1995), overharvest of wild fish by fisheries harvesting at the higher rates appropriate to the hatchery fish mixed with them (Lichatowich and MacIntyre 1987), and masking of the true abundance status of natural populations due to the misidentification of hatchery strays on natural spawning grounds as natural production.

To assess the risks posed by these hazards to naturally produced fish in a large river basin, we thermally mass-marked chinook salmon (*O. tshawytscha*) released at the two hatcheries in the area, and sampled returning adults in a local terminal fishery and comprehensively throughout the natural spawning grounds and hatchery return facilities. The study area is the Snohomish basin (4,800 km²), which produces a significant fraction of the naturally spawning Pacific salmon returning to Puget Sound in northwest Washington State, USA. Chinook salmon in the basin are managed for natural spawning objectives, but approximately one-half the return to the river each year is production from the Wallace River hatchery, located approximately 60 km upstream of the Snohomish River mouth. In addition, significant numbers of chinook salmon produced at the Bernie Kai-Kai Gobin Hatchery return to Tulalip Bay, located in the Snohomish River estuary.

From brood year 1993 through 1997 all chinook salmon produced at the Wallace and Gobin hatcheries were thermally mass-marked, and can be identified to brood year and hatchery of origin by microscopic examination of otoliths. Otolith marking followed the method described by Volk et al. (1990). Water chillers lowered incubation water 3–5°C below ambient temperature on prescribed schedules to induce unique otolith banding patterns on all fish in a production group. Each combination of hatchery and brood year received a unique banding pattern. Two broodstocks are used as sources of eggs: an early returning group (“summer”) originally derived from fish within the Snohomish system and a late returning group (“fall”) originally derived from outside the system. Beginning with the 1994 brood year, the summer and fall fish at Wallace hatchery received distinguishable marks. Releases of thermally marked chinook salmon from the two facilities are summarized in Table 1.

Table 1. Numbers of thermally marked chinook salmon released at the Gobin and Wallace River hatcheries, brood years 1993–1997. Each combination of hatchery, brood year, and stock (summer or fall) received distinguishable marks. Yearling and fingerling release stages are distinguishable from scale patterns.

Brood Yr.	Hatchery, stock, and release stage				
	Gobin	Wallace			
	Fall	Summer	Fall	Summer	Fall
	Fingerling	Yearling	Fingerling	Yearling	Fingerling
1993	1,280,000	281,000	642,700	268,000	519,200
1994	1,265,000	278,000	0	280,000	1,200,000
1995	1,860,000	270,000	918,000	265,000	975,000
1996	1,900,000	530,000	1,120,000	0	1,110,000
1997	1,700,000	394,000	920,000	0	0

Sampling of the terminal marine fishery in and near Tulalip Bay, adjacent to the Gobin Hatchery return facility, commenced in 1997. Otoliths were extracted from 100 chinook salmon per week and preserved in 95% ethanol for later examination for mark presence. Details of weekly catch and sampling for 1998 are shown in Table 2. The estimates of the contribution rate of Gobin hatchery chinook salmon to the Tulalip Bay fishery over the three years ranged from 92.5% (1998) to 98.0% (1999) (Table 3).

Sampling of hatchery rack returns began at Gobin Hatchery in 1998 and at Wallace Hatchery in 1997. At Gobin Hatchery, 100% of fall chinook salmon sampled at the hatchery rack were of local origin (Table 4). At Wallace Hatchery the local contribution to hatchery escapement ranged from 73% to 96% (Table 4).

Sampling of the natural escapement involved extraction of otoliths from spawned-out carcasses throughout all areas in the Snohomish system where chinook salmon spawn. Over the three years, otoliths were extracted from 9% of the total estimated naturally spawning population. An example of the detailed distribution of otolith marks by stratum for 1999 is shown in Table 5. Clearly, the stratification is necessary, since the estimated contribution rate of marked hatchery fish to the natural spawning populations ranged from 6% to 94% among the six strata over the three years (Table 6).

There is a large variance in marked fish contribution rate among both years and strata, and the pattern of variation appears to be consistent, indicating that year and stratum factors could be estimated separately (Fig. 1). It would also be possible to determine if factors that vary annually, such as streamflow, are related to the stray rates of hatchery fish. We have not yet completed the necessary statistical analysis to estimate these factors.

Table 2. Tulalip Bay terminal fishery weekly catch, otolith samples, and number of Gobin Hatchery (GH) marks in the sample, 1998.

Stat. Week	Catch	Samp.	GH Marks	95% c.i. ^a	
				lower	upper
32	1446	100	83	75%	89%
33	1904	100	93	87%	97%
34	1286	100	96	91%	98%
35	1102	100	96	91%	98%
36	522	100	94	88%	97%
37	672	100	93	87%	97%
≥38	169	0			
Total	7101	600	555	89%	94%

^aConfidence intervals for proportions computed using the method described by Fleiss (1981), eqns. 1.26 and 1.27.

Table 3. Annual estimates of the contribution of Gobin hatchery (GH) chinook salmon to the Tulalip Bay fishery, 1997–1999.

Year	Catch	Samp.	GH Contrib.	95% c. i.	
				Lower	upper
1997	8,295	514	95.3% ^a	93%	97%
1998	7,101	600	92.5%	89%	94%
1999	15,076	507	98.0%	97%	99%

^aThis estimate is for age 3 and 4 fish only since the 1993 brood year was the first year marked.

Table 4. Annual estimates of the fraction of the hatchery return that was from local hatchery production for the Gobin and Wallace hatcheries.

Hatchery	Year	Total Sample	Otolith Mark ^a				Fraction Local ^b
			UNM	GH	WRH	UNR	
Gobin	1998	50	0	50	0	0	100%
Gobin	1999	28	0	28	0	0	100%
Wallace	1997	195	48	4	142	1	73%
Wallace	1998	250	27	2	217	4	88%
Wallace	1999	200	7	0	191	2	96%

^aUNM: unmarked, GH: Gobin hatchery mark, WRH: Wallace hatchery mark, UNR: otolith mark unreadable.

^bNumber of otoliths with local hatchery mark divided by the number of readable samples (Total sample – UNR).

Table 5. Example of results of stratified sampling of natural escapement, 1999.

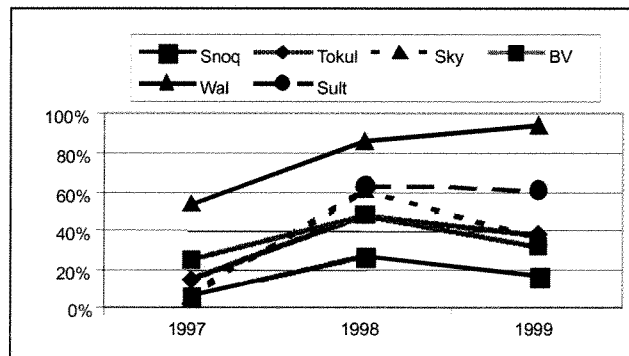
Stratum	Total Sample	Otolith Mark				CWT ^a	Proportion marked ^b
		UNM	GH	WH	UNR		
Snoqualmie	119	96	17	2	3	1	17%
Tokul Creek	98	57	26	9	6	0	38%
Skykomish	92	58	1	33	0	0	37%
Bridal Veil	41	28	0	13	0	0	32%
Wallace	119	7	0	104	8	0	94%
Sultan	64	25	0	39	0	0	61%
Total	533	271	44	200	17	1	

^aCWT: otolith was not marked, but the fish contained a coded-wire tag indicating a stray from outside the system.

^bNumber of marked otoliths divided by number of readable samples (Total sample - UNR).

Table 6. Summary of estimated marked fish contribution rates by stratum and year, 1997–1999.

Stratum	Year					
	1997		1998		1999	
	Sample	% marked	Sample	% marked	Sample	% marked
Snoqualmie	70	6%	116	27%	119	17%
Tokul	41	15%	39	48%	98	38%
Skykomish	33	6%	99	61%	92	37%
Bridal Veil	67	25%	75	48%	41	32%
Wallace	110	54%	105	86%	119	94%
Sultan	0		93	63%	64	61%
Total	321		527		533	

Fig. 1. Contribution rates of marked hatchery fish to natural escapement by stratum and year, 1997–1999.

To estimate the contribution of hatchery fish to the overall natural spawning escapement, we multiplied the stratum-specific contribution rates reported above by the estimated number of natural spawners, as determined from a combination of aerial and foot surveys using standard expansion methods employed by the Washington Department of Fish and Wildlife (Smith and Castle 1994). Summing these stratum-specific estimates for each year allows us to report escapement to the river in four categories according to both the origin (natural or hatchery production) and destination (natural or hatchery escapement areas) of the fish (Table 7). Over the three years the system-wide contribution rate of hatchery fish to natural spawning escapement ranged

from 19% to 55%, and the bias in the estimate of natural escapement ranged from -2% to 95%.

Our results show that by marking all hatchery production in the area and sampling the fishery, hatchery escapement, and natural escapement with appropriate stratification we can develop the information necessary to improve management of the chinook salmon resource. Because this species is now listed under the US Endangered Species Act, we are required to maintain harvest rates on wild stocks below strict guidelines and to make necessary adjustments to hatchery programs to minimize the impacts of this production on wild populations. The precise estimates of hatchery contribution to the fishery and escapement from our study is the first step implementing these management mandates. The high variability in hatchery contribution to natural escapement among years and among strata shows that hatchery straying is a complex problem. The regular pattern of variation observed gives us optimism that we may be able to discover relationships with independent variables that allow us to predict and ultimately to adjust our programs to minimize stray rates.

Table 7. Summary of stratified estimates of the escapement of natural and hatchery origin fish to natural and hatchery escapement areas. A small number of non-local hatchery fish (fish from outside the Snohomish system, identified from coded-wire tags) are included with the hatchery fish in this table.

Year	Escapement areas and origin				Overall HCR ^a	Bias in NEE ^b
	Natural areas		Hatchery facility			
	Natural	Hatchery	Natural	Hatchery		
1997	3,525	770	868	2,639	0.18	-2.2%
1998	2,856	3,450	375	4,377	0.55	95.1%
1999	2,436	2,354	240	6,089	0.49	78.9%

^aOverall hatchery contribution rate (HCR) is hatchery origin fish escaping to natural areas divided by total escapement to natural areas.

^bBias in natural escapement estimate (NEE) is (total escapement to natural areas less total natural origin escapement to natural and hatchery areas) divided by total natural origin escapement to natural and hatchery areas.

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Using Thermally-Marked Otoliths to Aid the Management of Prince William Sound Pink Salmon

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Keywords: Thermal otolith mark, hatchery, pink salmon, Prince William Sound

Starting in the fall of 1995, thermal marks were applied to the otoliths of all hatchery pink salmon, *Oncorhynchus gorbuscha*, in Prince William Sound, Alaska. Prior to that time, coded wire tags were used as a means of stock separation, but several assumptions used to generate stock estimates, such as those concerning tag shedding, differential mortality, and tag induced straying, were contentious and may have been flawed (Sharr et al. 1995; Habicht et al. 1998). Mosegaard et al. (1987), Volk et al. (1990), and Munk et al. (1993) found that carefully controlled incubation water temperature changes would mark otoliths of Pacific salmon *Oncorhynchus* spp. and Atlantic salmon *Salmo salar*. The first otolith marks were applied to pink salmon embryos in the fall of 1995 in Prince William Sound, and were highly visible on voucher samples taken from hatchery fry in the spring of the following year. For pink salmon brood years 1996 and 1997, accessory thermal marks, applied after the fry hatched, allowed identification of within-hatchery treatment groups (Fig. 1).

Double-blind tests were conducted on otoliths taken from emergent fry from brood years 1995–1997 to assess the ability of laboratory personnel to correctly identify hatchery otolith marks. The tests indicated that the probability of a successful identification between hatchery and wild pink salmon was 99.6%, 99.7%, and 99.3% for brood years 1995–1997, respectively.

Starting in 1997, thermal otolith marks were recovered from the commercial catch of pink salmon in Prince William Sound and used to estimate hatchery contributions. Because every otolith of a hatchery cohort is marked, precise estimates of hatchery contributions can be obtained with relatively few otolith recoveries. A very important prerequisite for such an estimate is that a representative sample is taken from the fishery. With this in mind, catch-sampling and estimation protocols were developed in 1995 and 1996 to ensure that estimates were usefully precise and accurate. A proportional sampling scheme was developed such that otoliths were sampled from all tenders in a manner proportional to their load. Such a sample is self-weighting and leads to many simplifications in sample size calculations and data analysis. To determine how a sample should be taken from a tender, we examined the degree of mixing of fish within the hold of a travelling tender. No significant mixing was found to occur (Table 1), and a systematic sample was therefore taken from each tender to ensure a representative sample was achieved. The systematic sample was taken by removal of a salmon from a processor belt at set intervals throughout the unloading process. The sampling interval was adjusted according to the number and speed that pink salmon were being processed. The sagittae otolith bones were extracted and placed in order of selection in numbered trays. The sample collection technique was somewhat self-weighting (larger loads generated more otoliths). After all tenders had been sampled, otoliths were subsampled from each tender collection to fine-tune the proportional sample. Ultimately, 96 otolith pairs formed the weighted systematic sample.

Fig. 1. Thermally-marked brood year 1997 (BY97) pink salmon otoliths sampled from Prince William Sound hatcheries.

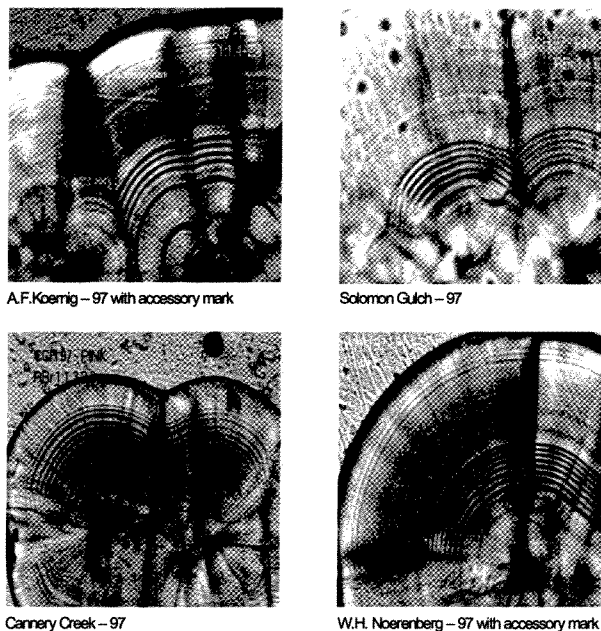


Table 1. Tender mixing data.

Loading Location of Fin Clipped Fish on Tender	Percentage Marked by Stage in Off Loading Samples		
	Start	Middle	End
Bottom	3.3	6.8	11.7
Middle	4.0	7.4	3.3
Top	16.1	10.8	0.9

The thermal mark program has led to a number of significant improvements in estimation of hatchery contributions over the coded wire tag recovery program. In the latter, large numbers of fish had to be sampled to provide useful estimates, while the otolith program can provide more precise hatchery contribution estimates from far smaller samples. This development contributed to better management of some important fishery strata. Analysis of otolith samples from test fisheries that routinely harvest small numbers of salmon provided information that resulted in fishery openings that would not have occurred under the sample-intensive coded wire tag program. Another improvement afforded by the otolith-marking program is that hatchery contribution estimates are available much sooner. Timely information is extremely important for decisions regarding harvest. In Prince William Sound, preliminary estimates of the stock composition of an area-time specific catch were available within 24 hours after a fishery closure.

Perhaps the best asset of the Prince William Sound thermal mark program is that fishery managers and the commercial fleet believe the generated stock contribution estimates. The high degree of confidence associated with otolith-derived estimates originates in large part from the assumption-free nature of the estimation procedure, and the effort made to ensure representative sampling. The highly efficient data-tracking and data management mechanisms built into our system also contributed to this confidence. Data summaries and updates can be executed within minutes after data entry, giving managers more time to make decisions.

Otolith marks have also allowed a number of *ad hoc* studies that depend on knowledge of hatchery contributions. For example, they have allowed study of the proportion of hatchery pink salmon straying into selected wild stock streams. Many streams close to the large production hatcheries and along the migration corridors were inundated by stray hatchery-released fish. The number of stray hatchery pink salmon increased as the spawning season progressed with the highest percentage of stray hatchery salmon occurring in the last sampling strata of the spawning season. The most obvious explanation for the large contribution of hatchery salmon to these escapements lies in the numerical dominance of hatchery over wild runs. Sharp et al. (2000) estimates that 26.0 and 25.6 million hatchery pink salmon and 2.3 and 5.3 million wild stock pink salmon returned in 1997 and 1998, respectively. These stray pink salmon may also be a function of a large number of unharvested salmon remaining at the hatcheries at the end of the commercial season that were beyond broodstock needs. More information is needed on reproductive success, domestication, and gene flow of these stray pink salmon if we are to assess their effects on the wild salmon populations.

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Use of Thermal Mark Technology for the In-Season Management of Transboundary River Sockeye Fisheries

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Keywords: Thermal mark, otolith, inseason management, sockeye salmon

The transboundary Stikine and Taku Rivers originate in British Columbia and flow through Southeast Alaska before entering the Pacific Ocean. Stikine and Taku salmon are harvested in commercial, recreational, personal use, and subsistence fisheries in the U.S. and Canada. Discrete wild stocks exist in both rivers and are intensively managed based on abundance and on harvest sharing guidelines in the Pacific Salmon Treaty. In 1989 a joint U.S./Canada fry-planting program was implemented to increase the sockeye salmon available to fisheries in both countries. Both countries agreed that some quantifiable method of assessing salmon production from this program was desired and that thermal marking of the planted fish and recovery of marks from catches and escapements would likely provide the best assessment method. Canada collects gametes from spawners in Tahltan Lake in the Stikine system (Fig. 1) and Tatsamenie Lake in the Taku system (Fig. 2) and transports them to a U.S. hatchery at Port Snettisham. The U.S. incubates the eggs, thermally marks the eggs and alevins, and backplants the resultant fry into Tahltan and Tatsamenie lakes. Tahltan origin fry are also planted into Tuya Lake in the Stikine system (Fig. 1) above a migration barrier to adult anadromous salmon. Both countries collect and process otoliths at various times from rearing juveniles, emigrating smolts, fisheries that harvest returning adults, and spawning escapements. This paper focuses on using thermal mark technology for in-season management of transboundary river salmon fisheries.

Fishery managers recognize two groups of wild Stikine sockeye, the Tahltan stock and the Mainstem stock conglomerate in addition to the planted Tahltan and Tuya stocks. The primary U.S. harvest of Stikine stocks along with four other major stock groups occurs in Districts 106 and 108 gillnet fisheries while the Canadian harvest of Stikine fish occurs in inriver gillnet fisheries located near the U.S./Canada border and around Telegraph Creek, B.C. (Fig. 1). Inseason management of fisheries is based on catches, catch per unit effort, stock composition estimates, and migratory timing. Rapid identification of thermal marks from sampled fisheries allows managers to estimate the relative abundance of planted fish in the various fisheries. The U.S. initially uses historical stock composition estimates to estimate catches of Tahltan and Mainstem fish. Canada uses inseason analysis of egg diameters to separate the Tahltan and Tuya fish, which have small eggs from the Mainstem fish, which have large eggs. Otolith samples are collected from U.S. and Canadian sockeye catches; immediately after each fishery closure they are shipped to Juneau, Alaska for analysis. Generally preliminary estimates of thermal mark prevalence in the catches are available to managers with 24 to 48 hours of fishery closures. This information yields the number of planted Tahltan and Tuya fish in the U.S. and Canadian catches. The incidence of thermal marks correlated with the egg diameter measurements from inriver catches are used to estimate the relative abundances of the planted Tahltan and Tuya fish to the

Fig. 1. The Stikine River and U.S. and Canadian Fisheries.

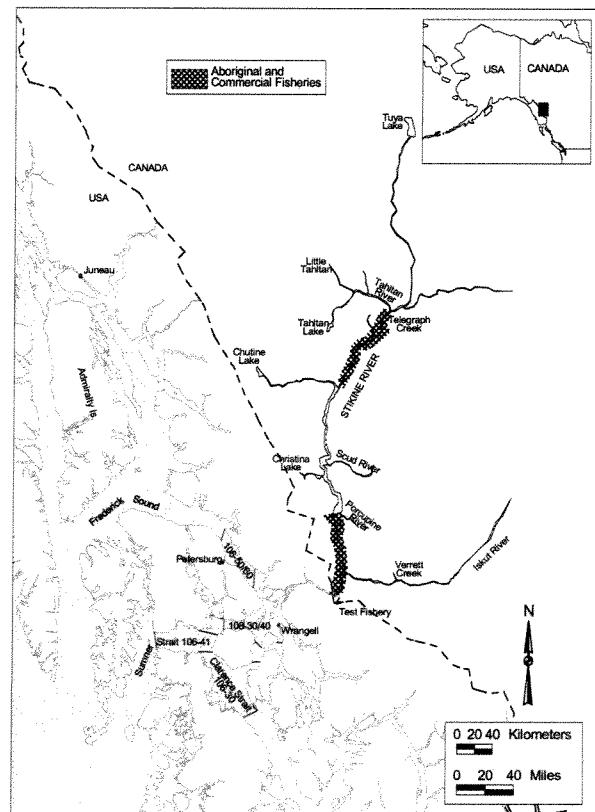
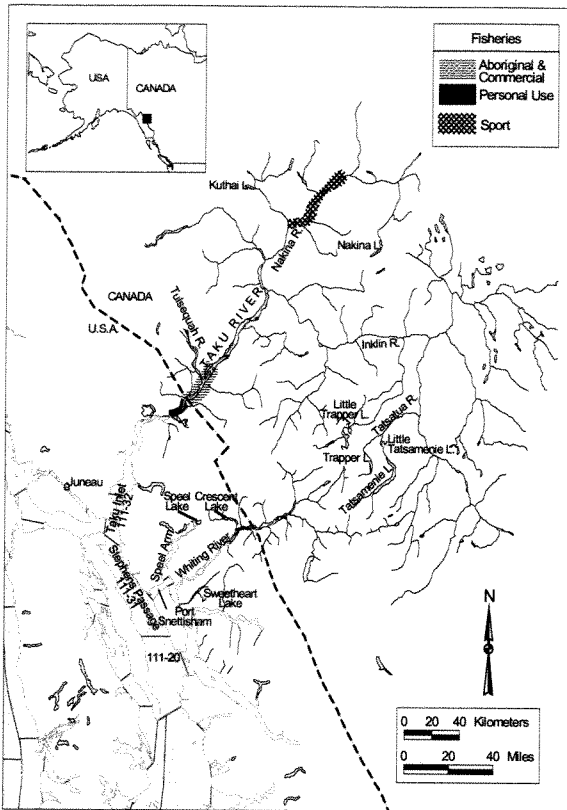


Fig. 2. The Taku River, Port Snettisham, and U.S. and Canadian Fisheries.



wild Tahltan and Mainstem stocks. The inriver ratio of thermally marked Tuya fish to the wild Tahltan and Mainstem stocks is applied to the marine catch of thermally marked Tuya fish to revise the initial weekly marine harvest estimates of these stocks. These inseason stock composition estimates are combined with historical migratory timing information to project the total runs of all three stocks. Scale pattern analysis is combined with thermal marks and egg diameters postseasonally to estimate harvests and reconstruct runs of Tahltan, Tuya, and Mainstem sockeye salmon. Planted Tahltan and Tuya sockeye salmon have contributed an annual average of 25,000 fish to U.S. catches and 23,000 fish to Canadian catches since 1994.

Fishery managers recognize four groups of wild Taku sockeye salmon, the Kuthai, Trapper, Mainstem, and Tatsamenie stocks (Fig. 2). In addition there are two major wild stocks from Crescent and Speel Lakes in Port Snettisham and a stock produced at Port Snettisham hatchery that are also harvested in U.S. marine fisheries. The primary U.S. harvest of Taku and Port Snettisham sockeye stocks occurs in the District 111 gillnet fishery while the primary Canadian harvest of Taku fish occurs in an inriver gillnet fishery above the U.S./Canada border. The abundance of the Taku stocks is estimated inseason from a mark-recapture program in which tags are applied to fish live-captured downstream of the U.S./Canada border and recoveries are made in the Canadian inriver fishery. Contributions of planted Tatsamenie fish and the Port Snettisham hatchery stocks

are estimated inseason from analysis of thermal marks. Matched scale, otolith, and parasite samples are collected from the marine fishery and unmatched otolith and scale samples are collected from the inriver fishery. As with Stikine area fisheries the otoliths from Taku area fisheries are processed and analyzed within a day or two after fishery closures. This information allows fishery managers to estimate the relative contributions of wild and hatchery fish to both U.S. and Canadian catches. Postseason analysis of scale patterns provides stock composition estimates for wild sockeye stocks from inriver catches and is combined with parasite prevalence analysis for the marine stock composition estimates. Planted Tatsamenie and Trapper stocks have contributed an annual average of 2,300 fish to U.S. and 700 fish to Canadian catches since 1995. In addition, Port Snettisham hatchery fish have contributed an average of 12,000 fish to U.S. catches during this period.

Collecting and processing otoliths from commercial fisheries in and near the Taku and Stikine Rivers has become an integral part of stock composition estimation and fisheries management. During the 1994 season the Juneau otolith lab processed 4,700 otoliths inseason and 4,700 otoliths postseason, with 378 marks recovered from 55 strata from these fisheries. This last year the numbers had grown to 7,000 otoliths processed inseason of a total of 14,000 otoliths processed with 3,400 marks recovered from 84 strata. Inseason analysis of thermal marks allows fishery managers to determine the relative abundance of hatchery and wild sockeye salmon. This inseason information is critical to shape fisheries to maximize the harvests of the hatchery fish while minimizing the risk of overharvesting wild stocks.

Use of Otolith Marking for Evaluation of Hatchery Output Efficiency

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Keywords: Hatchery, otolith, dry method of marking otoliths, thermal marking, monitoring of marked fish

This report presents information on the status of otolith marking at Russian fish hatcheries. We also use the results of otolith-mark identification of hatchery fish to evaluate the effectiveness of fish farming in the Magadan Region.

The method of marking salmon otoliths with subsequent identification of marked fish in mixed-stock catches is not new, and is successfully used in salmon hatcheries in Canada and the United States. Mass marking of salmon otoliths has been conducted in Magadan since 1994. In recent years, salmon hatcheries in Kamchatka and Sakhalin began to conduct large-scale marking based upon our recommendations.

The first years of method introduction were used to adapt otolith marking to the operational conditions of Russian salmon hatcheries and to search for methods to increase the number of possible marks and their information content. The dry method of marking otoliths, which was recently invented, allowed us to a great extent to avoid the problem of a short marking "window." As a result, we can use either thermal- or dry-marking methods or both to produce otolith marks. The increase in the number of possible types and content of marks allows us to arrange the marking processes for all hatcheries to avoid mark duplications. This provides an opportunity to recognize salmon from different reproduction areas (Kamchatka, Sakhalin, Magadan) and hatcheries among salmon stocks.

We are now ready to undertake total marking at all hatcheries producing Pacific salmon. However, it is important not only to conduct marking but also to identify marked fish in spawning runs. The problems related to marking can be generally regarded as solved, however, several methodological and organizational problems arose while arranging the monitoring of marked fish. Resolution of these issues is essential, even for the small rivers of Taiu Bay. Correct organization of data collection is especially important for long rivers with numerous tributaries in Kamchatka and Sakhalin. For example, depending on the task assigned, it is necessary to correctly identify the places for collection of spawning individuals, the number of individuals to be taken in order to determine the proportion of the hatchery fish in the stock mixture, and the frequency of data collection.

Hatchery production of fish in the Magadan Region is young. The majority of hatcheries began operations quite recently. The oldest hatchery started up fifteen years ago. Marked fish, which can be recognized by otolith marks, have been identified among spawning salmon returning to hatchery rivers during the past three years (1998–2000). The main subject of our study is chum salmon. Evaluation of hatchery output efficiency, assessment of rearing techniques, and identification of features characterizing the hatchery fish in the mixed stock are the main tasks of our studies.

The spawning runs of the marked fish in 1998–2000 were analyzed, and the following observations were made:

- The identification of otolith-marked fish showed that the main portion of mixed spawning runs of chum salmon is represented by wild fish. The oldest hatchery, which releases up to 20 million juveniles per annum, is located on the Ola River. Inseminated eggs from donor rivers have been delivered to that hatchery since it was founded. Based on the identification of adult salmon that were otolith-marked as eggs, the calculated return coefficient of hatchery salmon was lower than that of the wild chum salmon populations. The portion of hatchery released chum salmon was about 6% of the total catch in 2000 (Fig. 1).

Fig. 1. The percentages of hatchery-released chum salmon in total catches in the Yana and Ola rivers, 1998–2000.

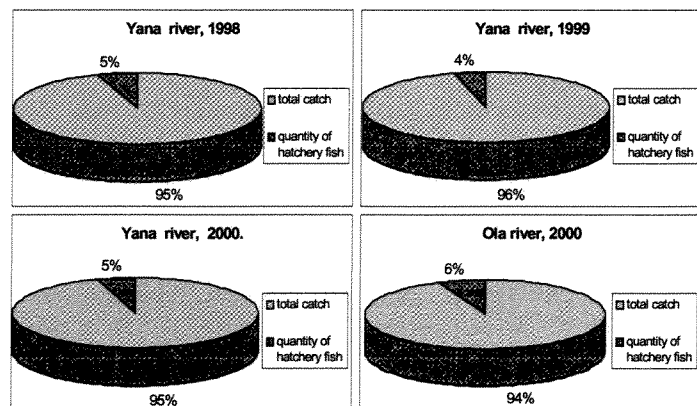


Table 1. The Yana River salmon hatchery return coefficients (based on results of marking).

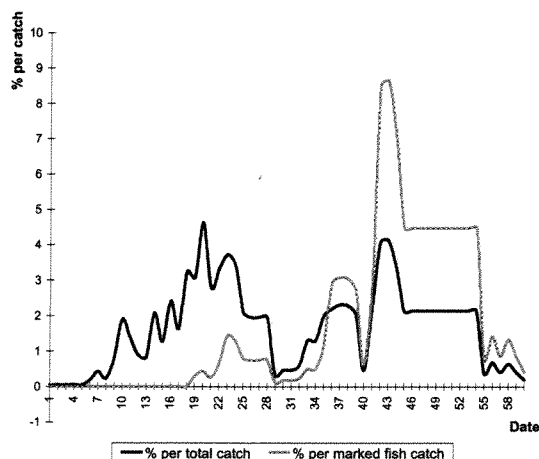
Generation	Fry released next year in thousand	Hatchery fish return in thousand			Total return in thousand	Return coefficient (%)
		1998	1999	2000		
1994	800	3+ 2.233	4+ 0.398	5+ -	2.631	0.32
1995	7759	2+ -	3+ 0.214	4+ 1.187	1.401	0.02
1996	2432	-	2+ -	3+ 0.222		
1997	669	-	-	2+ -		
Total:		2.233	0.612	1.409		

Previous indirect assessments, however, led us to expect a much higher percentage of hatchery fish in the catch. An analysis of the Ola River Hatchery output efficiency showed that the use of inseminated eggs delivered from donor rivers is an ineffective practice. Despite large quantities of eggs incubated each year, the number of chum salmon in spawning runs in 1998–2000 did not increase. We are facing a situation in which the inefficiency of the donor-river eggs is evident, but at the same time home-river spawning individuals are too scarce to increase the quantities of salmon. At present, we do not have any high-yield hatchery fish stocks.

Monitoring of chum salmon on the Yana River, where hatchery activities have been conducted since 1994, showed that the wild chum salmon comprise the main portion of the spawning runs. The analysis of marked-fish spawning runs at the Yana Salmon Hatchery also confirms the low efficiency of the practice of delivering inseminated eggs from donor rivers. The return of 1994-generation chum salmon was higher for home-river spawning fish than for salmon produced from eggs brought from other rivers in 1995 and 1996. The 1994-generation return coefficient was 0.32% (Table 1). We consider this to be the most productive release group in the past few years. The return coefficient for the 1995 generation was considerably lower.

The low returns of hatchery salmon, evaluated by the recovery of otolith-marked fish, demonstrated the inefficiency of delivering eggs from donor rivers. This is why the Magadan fish hatcheries have gradually discontinued this practice.

- It is extremely important for the hatcheries to be able to evaluate salmon returns in order to select the most effective biotechnical techniques. The results obtained by identification of fish marked at the hatcheries confirm the increased returns of fry raised in marine net pens prior to release and released in the most productive coastal areas of Tauu Bay. The return of the chum salmon raised by this method was estimated to be from 1.5 to 1.8% of the total number released. This result demonstrates the potential of marine rearing of chum salmon fry, which would make it feasible to create artificial industrial populations, one hundred percent of which could be used for commercial purposes.
- It was found that introduced salmon retain the ability to spawn during the period typical for their home rivers. The monitoring of the chum salmon spawning runs belonging to artificially produced commercial populations in the Kul'kuty River shows that the hatchery fish retain the features characterizing the parent stock. The population is represented and formed mostly by the late-run (fall) chum salmon. The returning hatchery salmon fully retain the ability to spawn during the period of the natural spawning migration, as well as the size and weight characteristics typical for the parent stock.
- The analysis of otolith-marked salmon spawning runs allows us to develop rational strategies for exploitation of salmon stocks. We found that hatchery salmon tend to migrate at the end of the spawning run (Fig. 2). Thus, the commercial fishery targets wild

Fig. 2. Chum salmon spawning runs dynamics, Yana River, 2000.

salmon, while the hatchery fish reach the spawning grounds. This fact results in directed selection, which may, over the course of time, have a negative impact on the population structure and its reproduction efficiency. In consideration of this situation, it seems that the intensity of commercial fishing should be increased at the end of the spawning migration and decreased at the beginning and the middle of the spawning season in order to preserve the most valuable natural subpopulations.

The impact of hatchery production on natural chum salmon populations in the rivers of the Magadan Region is generally insignificant except in the Ola River. In conclusion, we wish to emphasize that our results are based on a preliminary analysis of data that were collected during only three years. The subsequent use of otolith marking and further collection of data on identification of hatchery salmon will provide a means for solving all kinds of fish hatchery problems.

Otolith Marking at Kamchatka Salmon Hatcheries

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Keywords: Otolith, thermal mark, dry mark, salmon hatchery, Kamchatka

Rational use of fish stocks is impossible without their reliable differentiation. In Kamchatka most of the hatcheries are oriented to release small juveniles after a short rearing period. After a detailed analysis of all present methods of fish marking, we concluded that the most acceptable method for the hatcheries in Kamchatka should be otolith marking (Chebanov and Kudzina 1999). The purpose of this study was to provide background history and estimate the results of thermal and "dry" otolith marking at Kamchatka salmon hatcheries in 1999 and 2000.

At the Kamchatka hatcheries, otolith marked fish are regionally identified by 3 stripes in the first marking block with additional blocks used to identify where the fish was released, the year of marking, or, in some cases, the rearing techniques or transportation. Rogatnykh et al. (2000) suggested several marks for Kamchatka hatcheries according to maximum age of salmon. Since some age classes are uncommon in returning fish it is possible to reduce the number of marks and reuse them later when there are no conflicts between brood years. Based on these considerations we have worked out a scheme of salmon marking at Kamchatka hatcheries providing identification of species, generation, and hatchery origin for individuals in mixed catches (Table 1).

Thermal marking in Kamchatka was first applied in 1995 and 1996 at Malkinsky hatchery (Vasilkov 1995, 1996). Marking has continued since then with juvenile sockeye marked in March and chinook salmon in January (Table 2). The examination of the standards collected after marking showed that only the marks of sockeye salmon

Table 1. The scheme of salmon marking in Kamchatka salmon hatcheries.

№	Year of marking (release)	Chinook			Sockeye			Chum		
		Rbr-coding	Marking time (days)	Calendar period	Rbr-coding	Marking time (days)	Calendar period	Rbr-coding	Marking time (days)	Calendar period
Malkinsky hatchery										
1	1999 (2000)	2:1.3,2.5	16	14.10-	1:1.3,2.3	12	14.10-			
2	2000 (2001)	2:1.3,2.1	8	09.11	1:1.3,2.1	8	09.11			
3	2001 (2002)	2:1.3,2.2	10		1:1.3,2.4	14				
4	2002 (2003)	2:1.3,2.3	12		1:1.3,2.1,3.1	12				
5	2003 (2004)	2:1.3,2.4	14		1:1.3	6				
Ozerky hatchery										
1	1999 (2000)				1:1.3,2.2	10	20.10-	1:1.3,2.2	10	20.10-
2	2000 (2001)				1:1.3,2.2n+2n	12	22.11	1:1.3,2.2n+2n	12	15.11
3	2001 (2002)				1:1.3,2.5	16		1:1.3,2.5	16	and
4	2002 (2003)				1:1.3,2.1,3.2n	12		1:1.3,2.1,3.2n	12	15.11-
5	2003 (2004)				1:1.3,2.2n, 3.1	12				10.12
Ketkinsky hatchery										
1	1999 (2000)							1:1.3,2.4	14	17.10-
2	2000 (2001)							1:1.3,2.1	8	06.11
3	2001 (2002)							1:1.3,2.3	12	18.11-
4	2002 (2003)							1:1.3,2.1,3.1	12	13.12
Paratunsky hatchery										
1	2000 (2001)							1:1.3	6	16-28.10
2	2001 (2002)							1:1.3,2.2n	8,5	and
3	2002 (2003)							1:1.3,2.2n,3.1	12	10-22.11
4	2003 (2004)							1:1.3,2.3n	9,5	

Table 2. The scheme and approximate number of juvenile chinook and sockeye salmon marked in Malkinsky hatchery from the egg incubations of 1994–1998.

Year of incubation	Year, month of marking	Approximate number of juveniles marked	Graphic image of the mark
Sockeye salmon			
1994	1995, March	370.100	
1995	1996, March	669.500	
1996	1997, March	331.700	
1997	1998, March	716.700*	
1998	1999, March	592.300	
Chinook salmon			
1994	1995, January ?	228.800	
1995	1996, January ?	530.900	
1996	1997, January ?	757.500	
1997	1998, January ?	336.900	
1998	1999, January ?	601.500	

* Note: - additionally to the mentioned number 17.100 juvenile chinook salmon were left at the hatchery for one more year; released in 1999.

released in 1997 and 1999 were readable. Chinook salmon marks for the same years were poor. One of the reasons for poor quality marks might be that they took place late, during a period when unexpected stripes formed in the otolith, making the process of detection more complicated. Another reason for poor marks is that fish were held in big pools, and it took a rather long time to change water temperatures. That led to a “smearing” of the marks, which also made identification difficult.

Akinicheva and Rogatnykh (1997) determined that the optimal stage for sockeye salmon otolith marking is the eye pigmentation stage and for chinook salmon the pro-larva stage. Based on that study and the poor quality of earlier marks, we began to mark fish during the year of incubation (October–December). In the fall of 1999, research on marking started in three Kamchatka hatcheries – Malkinsky, Ozerki and Ketkino. In Malkinsky hatchery, 770 thousand sockeye salmon were thermally marked at about 422–394 degree-days in an expanded-type Atkins incubator. Marking consisted of 2–3°C temperature change with periodicity of 24 hours at a starting ambient temperature of about 7.6°C. Two blocks of equidistant rings were created by using a 3-day break during marking. This produced the Rbr code 1:1.3,2.3 (Table 1.) A photo of the mark is shown in Fig. 1. Chinook salmon marking at Malkinsky hatchery began at about 600–706 degree-days, using a temperature drop of 4.6°C. Pro-larvae were allocated on a tubular substrate in the plastic pools of the “bath-spring” type. Water circulation period in the pools and incubators was 60 minutes. Two blocks of equidistant rings 3 and 5 were produced (Table 1, Fig 2).

Ozerki hatchery is equipped with cold-water supply, therefore an experiment using the dry marking method was conducted on 422 thousand chum salmon embryos and 92 thousand juveniles sockeye salmon. For chum salmon marking began 379–354 degree-days, and consisted of periodically draining the water for 24 hour periods. Water temperature was approximately 5.3°C, and air temperature in the incubation chamber was 5.3–5.6°C. The dry mark did not differ fundamentally from thermal mark in appearance, and similar code structure was used (Table 1, Fig. 3 and Fig 4).

Fig. 1. Otolith mark in juvenile sockeye salmon released from Malkinsky hatchery in 2000.

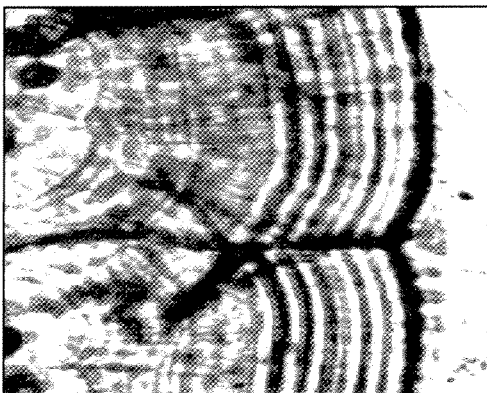


Fig. 2. Otolith mark of juvenile chinook salmon released from Malkinsky hatchery in 2000.

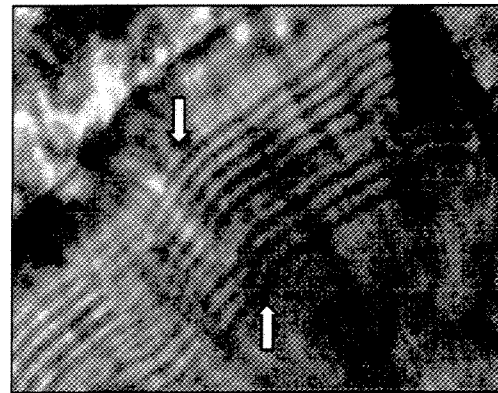


Fig. 3. Otolith mark in juvenile chum salmon released from the Ozerki hatchery in 2000.

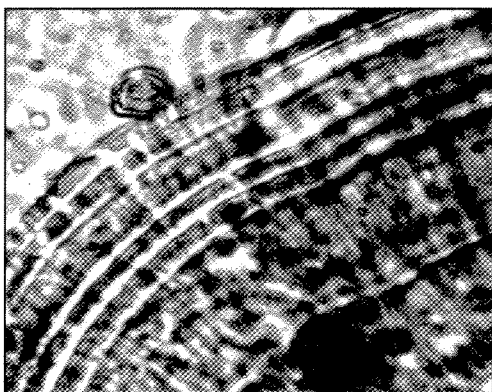
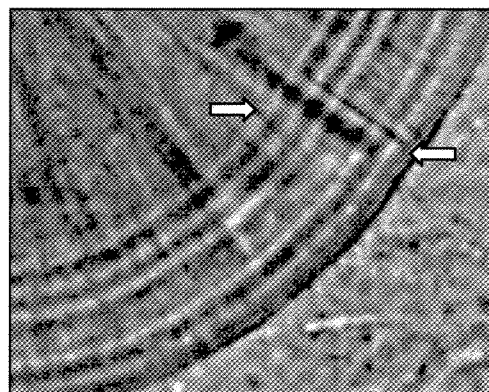


Fig. 4. Otolith mark of juvenile sockeye salmon released from Ozerki hatchery in 2000.

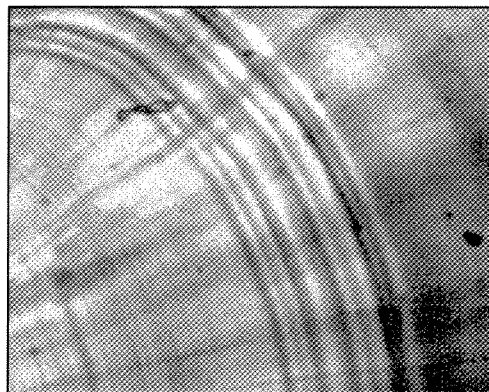


At Ketkino hatchery 621 thousand chum salmon were also experimentally marked using the “dry” method. Marking began 101 days after fertilization and also consisted of two blocks of equidistant rings (Table 1., Fig. 5)

In November–December 2000, marking continued at the three Kamchatka hatcheries using the same methods as in the previous year. About 591 thousand chinook salmon pro-larvae and 785 thousand of sockeye salmon embryos were marked at Malkinsky hatchery; 5.508 thousand sockeye salmon embryos and 2.627 thousand of chum salmon embryos marked at Ozerki hatchery; and about 3.283 thousand chum salmon embryos marked at Ketkino hatchery. The patterns are shown in Table 1. The marks formed on embryos and eggs in all cases were “readable” and conformed to international standards.

Otolith analysis of sockeye salmon from the return to Malkinsky hatchery in 2000 produced an estimated total of 2375 adults from individuals with the otolith mark introduced in 1995–1998. The analysis of otoliths in 299 individuals showed that the percent of adult fish from the incubation of 1994, 1995, 1996 and 1997 in the return of 2000 was respectively 0.7%, 35.1%, 36.5% and 14%; the percent of undetermined fish was 13.7%. The return of hatchery fish by the generations released was 0.004% in 1995, 0.12% in 1996, 0.26% in 1997, and 0.05% in 1998.

Fig. 5. Otolith mark of juvenile chum salmon released from Ketkino hatchery in 2000.



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Alaska Department of Fish and Game Otolith Marking and Recovery Program

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Keywords: Otolith, thermal mark, latent class models

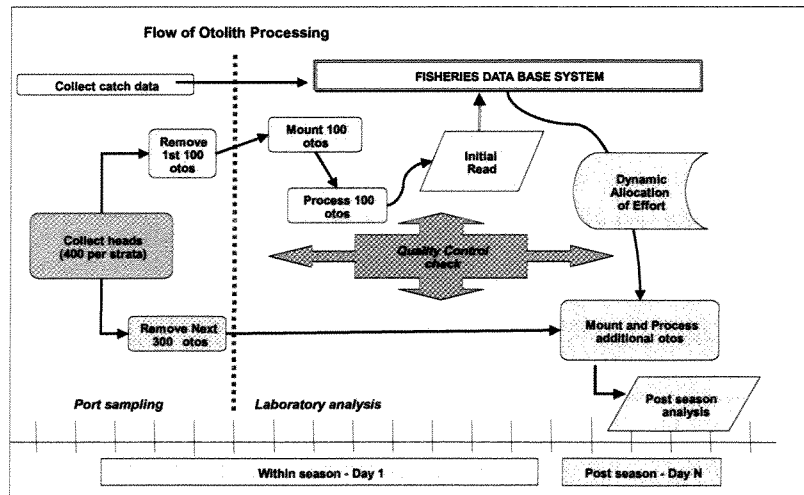
Since the inception of otolith marking in Alaska over ten years ago, approximately 5 billion hatchery salmon have been marked and released. Last year, 37 release groups representing over 900 million salmon (60% of the statewide hatchery production) were thermally otolith marked. The volume and number of marked groups is expected to increase. This paper highlights the role of the Alaska Department of Fish and Game’s Mark, Tag and Age laboratory in coordinating the assignment of thermal mark patterns in Alaska, the procedures and methods used in recovering marked fish from commercial fisheries, and the effort made to ensure standardization of methods and reading accuracy by other otolith reading laboratories in Alaska.

When coordinating the assignment of thermal patterns, the Mark Tag and Age lab works closely with hatchery operators, fishery managers, and affected parties to match marking capabilities of each facility with the availability of unique mark patterns. Because there are limitations to the number of unique marking codes (Hagen 1999), and because the facilities can vary in their ability to control water temperatures, assigning unique patterns can be difficult. To help mitigate this problem, the lab looks for ways to ensure that secondary characteristics, such as ring spacing and ring position, can be used to distinguish among marking groups when distinct patterns are not possible. To help design marks, a modeling approach is used which incorporates the anticipated temperature regimes at the hatchery sites and the relationship between temperature and otolith growth. To identify when marking can begin, staff examine the developing otolith from eyed egg stages. Marking can typically begin once the otolith primordia have coalesced or fused. This usually occurs during the eyed-egg stage at approximately 300 cumulated temperature units (CTUs), though stock specific differences have been observed and must be considered. Upon completion of marking and prior to release of the fish, hatcheries provide representative specimens and associated data for each mark group. These samples constitute the voucher collection. The voucher otoliths are removed from the fish, mounted, and ground to a thin-section. They are examined for mark quality based on the appearance of the thermal rings. Measurements on ring spacing and mark location are taken, and the variability of the mark pattern within a voucher group is identified. The voucher collection provides otolith lab staff with references to identify marks in returning fish and allows feedback on the mark quality to hatchery personnel. The otolith lab is currently developing a digital image library of voucher specimens that enables the transmission of digital images to

hatchery operators, other labs and interested parties. Voucher otoliths are maintained according to returning brood years expected in the management season.

Within Alaska, approximately 30,000 otoliths are collected and examined annually by two ADF&G otolith labs: one in Cordova, which provides mark information for Prince William Sound fisheries (Joyce and Evans this volume) as well as the Juneau based Mark, Tag and Age lab which provides mark information in support of the Stikine and Taku sockeye fisheries in Southeast Alaska (Jensen and Milligan this volume). In both laboratories, a two-tiered processing schedule is used to provide in-season management information (Fig. 1) while prioritizing

Fig. 1. Illustration of the two-tiered processing system used by the Alaska Department of Fish and Game otolith processing lab. The first one hundred otoliths from a sample are processed upon receipt of the sample while additional specimens are processed post-season to reduce uncertainty in hatchery contribution estimates.



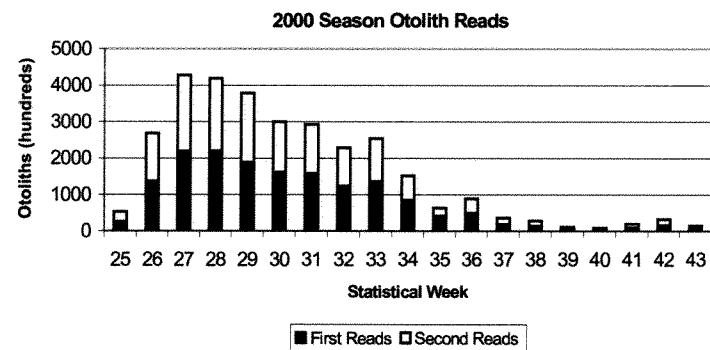
effort between multiple fisheries and maintaining production rates (Hagen et al. 1995). A subsample of otoliths from each stratum is processed within hours of receipt while a portion of the remaining otoliths are processed post-season using an optimizing algorithm designed to minimize the overall uncertainty in the hatchery contribution estimate (Geiger 1994). A bar code label is used to track specimens in the lab. Each reading station is equipped with dissecting and compound microscopes, a grinding wheel, a touch screen data entry computer, and a bar code scanner. The sample and specimen data, as well as the reader observations, are stored in a relational database to enable the rapid reporting of results.

During the 2000 season, over 16,500 otoliths were read in the Mark Tag and Age Lab, providing contribution estimates for 160 strata obtained from commercial fisheries, test fisheries, and escapement surveys. In addition, otoliths were collected on a weekly basis from the Canadian portion of the Stikine and Taku Rivers and processed to identify returning stocks and provide age compositions for returning fish. Additionally, many of the Stikine and Taku River systems otolith specimens are matched to age, weight and length data, scale age data, and brain parasite analysis that further assists in stock and age identification and requires meticulous dissection, data collection and data management.

In addition to the Taku and Stikine fisheries, the Mark Tag and Age Lab examines samples obtained from National Marine Fishery Service's high seas collections (Farley et al., this volume) and conducts second readings on otoliths received from the Cordova laboratory and other otolith reading labs. To monitor the accuracy of the readings, two independent readings were made on most otoliths (Fig. 2) and a third reader resolved reading conflicts. In some cases, otoliths may be independently read three times. Latent class models (Blick and Hagen 1998) are employed postseason to provide an estimate of reading accuracy and agreement. From the 2000 season, specific estimates of reader accuracy for the Taku and Stikine river marks ranged from 0.96 to 0.99 with slightly more accuracy in identifying wild fish than hatchery stocks.

The Alaska Department of Fish and Game's otolith processing lab is involved in otolith research in addition to management and production processing. The lab is developing treatment, processing, and Food and Drug Administration (FDA) protocols to integrate Strontium Chloride marking for remote hatchery marked salmon production. Processing techniques for strontium chloride marking are being investigated for juvenile and adult salmon mark recovery and may be incorporated in elemental analysis applications of other Pacific salmon species and Pacific herring. Salmon straying studies and other investigations continue to challenge the lab to provide vital information on Alaska's salmon resources.

Fig. 2. Tracking graphic of the number of otoliths read per statistical week for the year 2000 management season. The increase in otolith readings between statistical weeks 26 and 33 represent the sockeye otolith contribution, the focus of the lab during the summer season. Following week 33, more attention is devoted to chum and pink salmon otolith reading as well as sockeye salmon escapement survey otolith reading.



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Wandering Pink Salmon: 1999 and 2000 Thermal Mark Recoveries in Southeast Alaska

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**Keywords:** Pink salmon, *Oncorhynchus gorbuscha*, homing, straying, otoliths, thermal marks

Most salmon are believed to return to their natal stream to spawn (Hasler and Scholtz 1983). Although localized movement is known to occur (Habicht et al. 1998), long distance straying has rarely been documented (Quinn et al. 1987, Labelle 1992, Quinn 1993, Sharp et al. 1994). In 1999, pink salmon obtained from the Hawk Inlet fishery in southeast Alaska were examined for thermal-marked otoliths. We expected that most of these pink salmon would be from wild stocks. Based on current hypotheses regarding salmon homing, we expected that any thermal marked otoliths would contain a 5-ring pattern indicating they were released by the Macaulay Salmon Hatchery (formerly DIPAC), Juneau, AK.

From the initial sample, we found evidence that Prince William Sound (PWS) hatchery fish were present. Consequently, additional samples were collected in southeast Alaska, and a study was undertaken to determine if these marks were not PWS marks, but aberrant marks from DIPAC pink salmon or wild pink salmon with a natural pattern that mimicked a PWS mark.

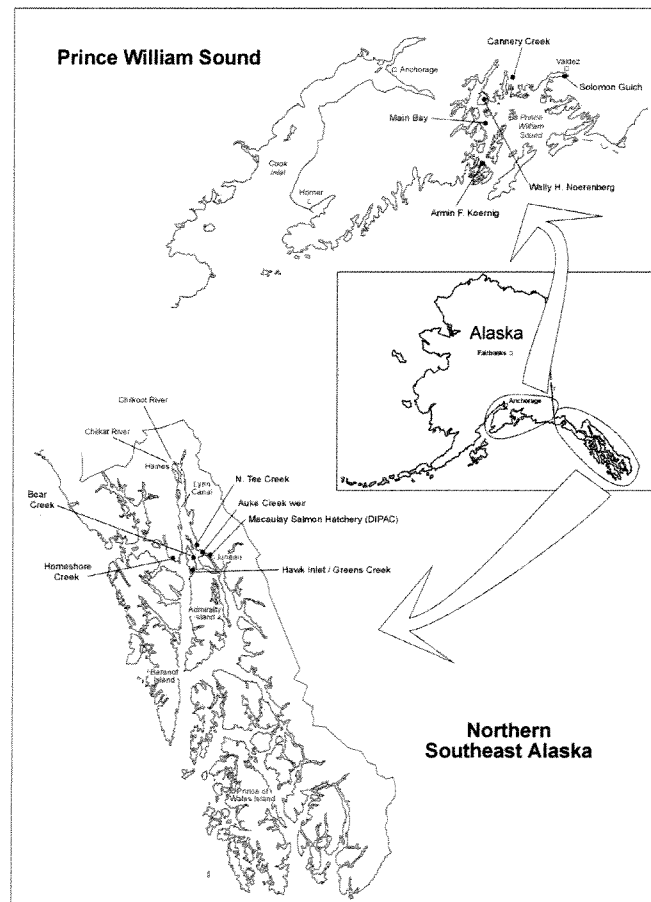
Otoliths were collected in several locations in southeast Alaska in 1999 and in the Haines, Alaska area in 2000 (Fig. 1, Table 1). The first samples were collected during the 1999 test fishery in Hawk Inlet in northern Lynn Canal. Additional samples were collected by Alaska Department of Fish and Game (ADFG) personnel monitoring index streams for escapement counts and ADFG and National Marine Fisheries Service personnel collecting samples from DIPAC hatchery and from Auke Creek weir. Sampling was largely ad-hoc.

ADFG's Mark, Tag and Age Laboratory in Juneau processed the otoliths and examined them for thermal marks (Hagen et al. 1995). Readers made independent determinations on the presence and identification of thermal marks. At least two readings were made of each otolith.

Otoliths were photographed with a video camera attached to a microscope, and the images were measured with imaging software on a personal computer. Because salmon otoliths contain multiple primordia within the core, the location of the thermal mark was defined as the distance from the posterior primordia to the thermal mark along the posterior dorsal quadrant.

Measurements of the thermal marks were compared with measurements from vouchers or known marked samples of DIPAC fry obtained prior to release ( $n = 8$ ). Measurements from known thermal-marked DIPAC otoliths were obtained from returning adult pink salmon found in the DIPAC

**Fig. 1.** Map of the study area.



**Table 1.** Summary of pink salmon samples collected and examined for otolith thermal marks in southeast Alaska in 1999 and 2000.

| Sample Date | Harvest Type | Fishery Location | N   | Marked? |     |       | Pattern |         |       |       |       |
|-------------|--------------|------------------|-----|---------|-----|-------|---------|---------|-------|-------|-------|
|             |              |                  |     | No      | Yes | %     | CCH97   | DIPAC97 | SGH97 | WNH97 | SGH98 |
| 18 Jul 99   | Comm         | Hawk Inlet       | 94  | 91      | 3   | 3.2   |         |         | 3     |       |       |
| 21 Jul 99   | Comm         | Hawk Inlet       | 100 | 96      | 4   | 4.0   |         | 3       | 1     |       |       |
| 20 Aug 99   | Rack         | DIPAC Hatchery   | 25  | 0       | 25  | 100.0 |         | 25      |       |       |       |
| 25 Aug 99   | Escape       | Auke Creek       | 190 | 188     | 2   | 1.1   | 2       |         |       |       |       |
| 16 Sep 99   | Escape       | Bear Creek       | 50  | 50      | 0   | 0.0   |         |         |       |       |       |
| 16 Sep 99   | Escape       | Greens Creek     | 52  | 52      | 0   | 0.0   |         |         |       |       |       |
| 16 Sep 99   | Escape       | Homeshore Creek  | 50  | 49      | 1   | 2.0   | 1       |         |       |       |       |
| 20 Sep 99   | Escape       | Chilkat River    | 45  | 40      | 5   | 11.1  | 4       |         |       | 1     |       |
| 20 Sep 99   | Escape       | Chilkoot River   | 50  | 50      | 0   | 0.0   |         |         |       |       |       |
| 26 Sep 99   | Escape       | N. Tee Creek     | 39  | 39      | 0   | 0.0   |         |         |       |       |       |
| 24 Jul 00   | Escape       | Chilkat River    | 49  | 49      | 0   | 0.0   |         |         |       |       |       |
| 24 Jul 00   | Escape       | Chilkoot River   | 55  | 26      | 29  | 52.7  |         |         |       |       | 29    |
| 28 Jul 00   | Escape       | Hawk Inlet       | 107 | 107     | 0   | 0.0   |         |         |       |       |       |

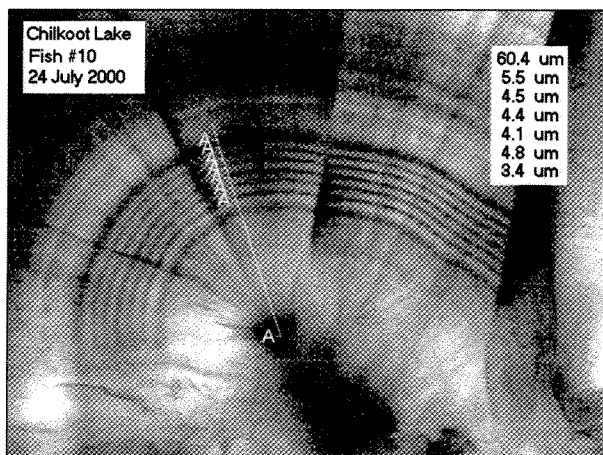
raceway (n = 8). The measurements were not statistically different between the two groups, so the data were pooled. Samples of known Solomon Gulf Hatchery (SGH) salmon were obtained from commercial catches in PWS (n = 12) as well as voucher samples (n = 11). These two collections were not statistically different, so the data were pooled.

The Advanced Instrumentation Laboratory, Department of Geology and Geophysics, University of Alaska Fairbanks, used an electron microprobe to examine the elemental composition of otoliths of known origin and compare them with unknown otoliths. Samples included four otoliths from Chilkat River escapement samples that contained a PWS mark (Cannery Creek Hatchery, CCH97), 25 otoliths from the same sample but identified as wild, and 25 otoliths (CCH97) from PWS commercial fisheries in 1999.

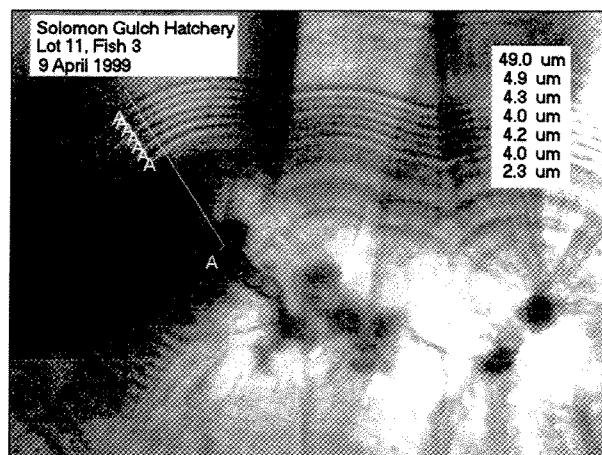
Five points on each otolith, approximately 130–250 microns from the primordia, were randomly selected for analysis. The elements examined included: calcium, potassium, chlorine, phosphorus, sulfur, sodium, magnesium, strontium, and associated oxides. Principal components and discriminant analysis (S-Plus) were used to determine if the elemental concentrations in the otoliths of the unknown pink salmon were similar to that of Chilkat River or PWS salmon.

Of 670 fish sampled in 1999, 15 (2.2%) contained thermal-marked otoliths (Table 1). Three (3; 20%) of the thermal marked otoliths contained the five-ring pattern (DIPAC97) used by Gastineau Hatchery in southeast Alaska. Twelve fish (80%) were marked by a PWS hatchery (SGH, CCH, or Wally H. Noerenberg (WHN)). In 2000, 29 (13.7%) of the sampled otoliths were thermal marked. All of these were marked by SGH (Figs. 2 and 3). These results were quite unexpected. In 1999, 80% of the marked fish were from PWS, and in 2000, 100% of the marked fish were from PWS. These fish were ~950 km from where they should have been.

**Fig. 2.** One of the pink salmon otoliths found in Chilkoot Lake, Haines, AK, 24 July 2000. Appears to have a 6-ring pattern similar to SGH98.



**Fig. 3.** Voucher specimen of the thermal mark applied by from Solomon Gulch Hatchery (SGH98), Prince William Sound in 1998. Also carries a six-ring pattern.



There was no significant difference in inter-ring spacing between the DIPAC97 (n = 16) otoliths and the SGH97 (n = 23) otoliths, but there was a significant difference in thermal mark location ( $p < 0.0001$ ). Measurements from the unknown fish had the same ring spacing as DIPAC97 and SGH97, but the thermal mark location (ANOVA Post hoc test) was similar to that from SGH97. Thus, these results supported the hypothesis that these were PWS fish not DIPAC fish with an aberrant thermal mark.

Analysis of the microprobe data indicated that CCH97 otoliths found in the Chilkat River had trace elements that more closely matched known CCH fish than unmarked pink salmon from the Chilkat River. The elemental concentrations in the otoliths of the unknown fish were most similar to that of the Chilkat River or CCH pink salmon.

Principal components analysis showed that the first two principal components explained all of the variation among the groups. Component one was comprised of Na, Cl, S, Ca, Sr, and K, while component two was comprised of K, Ca, Sr, S, Cl, and P. Based on these components, the data were split into two distinct groups. Two of the unknowns fell well into the CCH97 category, while a third unknown fell marginally into the Chilkat category. Discriminant analysis indicated that replicate readings of elemental compositions of three unknown pinks were classified as CCH97 10 of 15 (66.7%) times, and 24 CCH97 pinks were classified as unknown fish 43 of 120 (35.8%) times. Thus, these results supported the hypothesis that these were PWS fish not wild fish with a natural pattern that mimicked PWS fish.

There is a widely held notion that pink salmon are more prone to stray than other Pacific salmon species (Quinn 1984). A survey of the literature indicated that our observations are the furthest pink salmon have ever been recorded straying (~950 km). Although the extent of this straying is currently unknown, observations from the samples collected in 2000 indicate that straying may be significant at times.

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### **III. Workshop Review**

How to mark small juvenile salmon without injury has long been a concern for salmonid researchers and managers. Thermal otolith marking is a universal way to mark large numbers of hatchery salmon during embryonic and yolk absorption stages, creating distinct mark patterns in the otoliths by water temperature controls. Similar otolith marks are produced by the dry method developed by Russian scientists. This low cost technique can provide high quality otolith marks without special equipment. Chemical otolith marks using strontium or fluorescent substances may be used to supplement mark patterns in hatcheries, because the number of unique thermal or dry marking codes is limited.

In 2000 approximately one billion otolith marked juvenile salmon were released from hatcheries in North Pacific Rim countries. Many marking objectives have been achieved without organized rules for pattern assignment. A standardized system of organizing pattern information on otoliths potentially offers a larger number of patterns, and also provides the opportunity for coordinating marks between countries to avoid mark duplications in mixed-stock fishery analysis. The NPAFC Working Group on Salmon Marking would play an important role by coordinating otolith mark patterns among countries and creating an Internet-accessible database of otolith mark releases.

Application of otolith-marking technologies to the biology and management of salmon is essential. The early applications of otolith marking techniques supported scientific research to distinguish wild and hatchery salmon during early sea life. A recent rapid increase in the number of otolith mark releases has made it possible to track the migration of specific salmon stocks throughout their entire ocean life from coastal waters to the high seas. Current salmon research using otolith marks includes ocean distribution, migration speed, abundance, feeding success, growth, straying of otolith-mark groups, and interactions between wild and hatchery stocks.

Applications of otolith marking for stock assessment and management of terminal fisheries have increased in recent years. In Alaska an otolith marking and recovery program for in-season stock management is well established. Mass otolith marking is an effective tool for estimating the contributions of hatchery fish to overall natural spawning escapement. To minimize the effect of hatchery production on wild salmon populations, this information is critical.

The North Pacific Rim countries (Canada, Japan, Russia, and USA) are successfully conducting mass otolith mark releases under common rules. Otolith mark recovery data will enable us to develop a valuable time series of stock-specific biological information that is indispensable to the sustainable conservation of salmon stocks in North Pacific regions.

Shigehiko Urawa  
National Salmon Resources Center, Japan  
Co-Chairman of the Workshop Coordinators



## APPENDIX 1

### Program of the Workshop

#### OPENING REMARKS

P. HAGEN

#### ORAL PRESENTATIONS

##### I. Otolith Marking Technologies

Moderators: A. ROGATNYKH and E. VOLK

Otolith thermal marking

E. VOLK and P. HAGEN

The dry method of otolith mass marking

A. ROGATNYKH, E. AKINICHEVA, and B. SAFRONENKOV

Otolith marking at the eyed-egg stage of chum salmon with fluorescent substances

H. KAWAMURA, S. KUDO, M. MIYAMOTO, and M. NAGATA

Marking salmonids with strontium chloride at various life-history stages

S.L. SCHRODER, E.C. VOLK, and P. HAGEN

Development of a new stock discrimination tool for naturally spawning sockeye salmon within Alberni Inlet from stable isotopic composition of otoliths

W. LUEDKE and Y.W. GAO

Compiling and coordinating salmon otolith marks in the North Pacific

S. URAWA, P. HAGEN, D. MEERBURG, A. ROGATNYKH, and E. VOLK

##### II. Applications of Otolith Marking

###### *Salmon Biology*

Moderators: D. MEERBURG and S. URAWA

Early marine growth and habitat utilization of two major southeastern Alaska chum salmon stocks, based on thermally marked otoliths recovered 1997-2000

J.A. ORSI, D.G. MORTENSEN, D.L. TERSTEEG, and R. FOCHT

Application of otolith thermal mass marking for examining the biology and interactions of wild and hatchery chinook salmon during early sea life

B. HARGREAVES

Variations in catch per unit effort of thermally marked pink and chum salmon juveniles in the Gulf of Alaska during 1996 and 1998 in relation to adult hatchery salmon returns

E.V. FARLEY Jr., P. HAGEN, and J.H. HELLE

High-seas ocean distribution of Alaskan hatchery pink salmon estimated by otolith marks

M. KAWANA, S. URAWA, P. HAGEN, and K. MUNK



***Salmon Management***

Moderators: P. HAGEN and T. PERRY

Estimating the abundance and distribution of locally hatchery-produced chinook salmon throughout a large river system using thermal mass-marking of otoliths

K. RAWSON, C. KRAEMER, and E. VOLK

Using thermal marked otoliths to aid the management of Prince William Sound pink salmon

T.L. JOYCE and D.G. EVANS

Use of thermal mark technology for the in-season management of transboundary river sockeye fisheries

K.A. JENSEN and P.A. MILLIGAN

The use of otolith marking for evaluation of hatchery output efficiency

E. AKINICHEVA and A. ROGATNYKH

**CLOSING REMARKS**

E. VOLK

**POSTER PRESENTATIONS**

Otolith marking at Kamchatka salmon hatcheries

N.A. CHEBANOV and M.A. KUDZINA

Alaska Department of Fish and Game's Otolith Marking and Recovery Program

J.R. SCOTT, R.P. JOSEPHSON, P.T. HAGEN, B.A. AGLER, and J.W. CASHEN

Wandering pink salmon: 1999 and 2000 thermal mark recoveries in southeast Alaska

B.A. AGLER, P.T. HAGEN, J.R. SCOTT, J.W. CASHEN, and D. MORTENSEN

APPENDIX 2

List of Participants

|                      |                           |                     |
|----------------------|---------------------------|---------------------|
| <i>Canada</i>        | Elisabeth Appleby         | Jack Helle          |
|                      | Greg Bonnell              | Joseph Hinton       |
|                      | Brent Hargreaves          | Kathleen Jensen     |
|                      | Gerry Kristianson         | Ron Josephson       |
|                      | Wilf Luedke               | Tim Joyce           |
|                      | David Meerburg            | Kim Larsen          |
|                      | Jeff Till                 | Barbara McClellan   |
| <i>Japan</i>         | Masa-aki Fukuwaka         | Michael Meeuwig     |
|                      | Yukimasa Ishida           | Jamal Moss          |
|                      | Koichi Ishizuka           | Kate Myers          |
|                      | Hiroshi Kawamura          | Virginia Naef       |
|                      | Morihiko Kawana           | Lang Nguyen         |
|                      | Tetsuichi Nomura          | Ron Olson           |
|                      | Shigehiko Urawa           | Joseph Orsi         |
|                      |                           | Nathanael Overman   |
|                      |                           | Kit Rawson          |
|                      |                           | Robert Rhoads Jr.   |
| <i>Russia</i>        | Elena Akinicheva          | Brenda Rogers       |
|                      | Vladimir Belyaev          | Steve Schroder      |
|                      | Nickolay Chebanov         | Ryan Scott          |
|                      | Vladimir Karpenko         | Johnathan Sivilay   |
|                      | Alexander Rogatnykh       | Bill Smoker         |
|                      | Viatcheslav Vasiliev      | Karl Stenberg       |
| <i>United States</i> | Bev Agler                 | Diana Tersteeg      |
|                      | Marianna Alexandersdottir | Mark Tetrick        |
|                      | Dana Anderson             | Daniel Thompson     |
|                      | Cynthia Bucher            | Eric Volk           |
|                      | Gregory Buck              | Trey Walker         |
|                      | Nancy Davis               | Alex Wertheimer     |
|                      | Andrew Dittman            | Christian Zimmerman |
|                      | Elisabeth Duffy           |                     |
|                      | Valerie Elliott           |                     |
|                      | Rick Focht                |                     |
|                      | Yongwen Gao               |                     |
|                      | Hal Geiger                |                     |
|                      | Jeff Grimm                |                     |
|                      | Peter Hagen               |                     |
|                      | William Heard             |                     |
|                      | <b>NPAFC Secretariat</b>  |                     |
|                      | Vladimir Fedorenko        |                     |
|                      | Yoshikiyo Kondo           |                     |
|                      | Wakako Morris             |                     |
|                      | Tomoko Lumpkin            |                     |
|                      | Almira Safarova-Downey    |                     |

