

NPAFC

Doc. No. 394

Rev. No. _____

On using an adaptive approach to minimize thermal mark duplication and using strontium marking as a means to increase the variety of marking options

By

Peter Hagen

Alaska Department of Fish and Game
P.O. Box 25526
Juneau, Alaska 99821-5526

and

Eric Volk

Washington Department of Fish and Wildlife
MS. 43200—600 Capital Way North
Olympia, Washington 98501-1091

Submitted to the

NORTH PACIFIC ANADROMOUS FISH COMMISSION

By

The United States Party

March 1998

This paper may be cited in the following manner:

Volk, E. and P. Hagen. 1999. On using an adaptive approach to minimize thermal mark duplication and using strontium marking as a means to increase the variety of marking options. (NPAFC Doc. 394). 5p. Alaska Dept. Fish and Game, Juneau Alaska. 99801-5526

Using an Adaptive Approach to Minimize Thermal Mark duplication

Without belaboring the point too much, in a practical sense there is a limited number of distinct thermal mark codes available for use. Base mark codes using a small number of rings are the simplest to produce, but provide few options for creating variable patterns. Unfortunately these base codes are the only option for marking in many locations and it is not realistic to expect that any one facility releasing fish into the North Pacific should have sole rights to those patterns.

This leaves accessory marks, which generally consists of several rings induced after hatching, as another means of trying to distinguish thermal mark releases by each country. However in many locations accessory marks, in conjunction with base marks, are already being used for specific purposes, such as identifying release locations or brood years. In those situations, applying additional accessory codes as country identifier would complicate the mark patterns and potentially create new conflicts. Many hatchery facilities have narrow marking windows prior to hatching and are not able to mark fish after hatching. In these cases applying accessory marks is simply not a possibility. These exceptions would immediately undermine the idea of using a single pattern to represent country of origin.

Despite these cautionary statements, with careful planning and communication it should be possible to minimize the probability of encountering duplicate marks in ocean sampling. This can be done by using an adaptive approach to slightly modifying existing marks with secondary characteristics. For instance, if it is known in advance that two sites are planning to release similar mark patterns, it may be possible for either or both sites to institute minor modifications on ring spacing or apply subtle variations in temperature changes. Such changes could result in similar appearing marks that nonetheless could be separated from each other with measurements and careful observations. These secondary characteristics would be more time consuming to resolve, but should be sufficient for separating otoliths collected from high seas sampling in which there is no direct management need for quick analysis.

The prerequisite for such an adaptive approach is that all parties be kept informed of the marks being planned. In addition, key personnel in each country would need to be notified immediately if there are any disruptions in marking schedules that would result in unintended duplicate patterns. This may allow time for other hatcheries to adjust their schedules in response. The difficulties of coordination would be increased as thermal marking is expanded in other areas, but with careful planning such growth could be accommodated by the full participation of each country in a common database and reporting system which tracks the releases of thermally marked salmon.

Sponsorship of such a system by the Pacific States Marine Fisheries Commission would go along ways towards developing an accounting system for high seas

recoveries of thermal marked salmon. Developing the system will require further workshops and discussions, and support for the PSMFC to secure funding to take on that role. The prototype webpage on thermal marks release information put together by the PSMFC is an example of how that information can be shared. The webpage can be located at:

<http://www.psmfc.org/rmpc/iatmi/>

The data shown on that web site is not real data, but simply used to illustrate how the thermal mark information can be shared as digital images.

Strontium marking as a secondary code to thermal marks

In addition to using an adaptive approach for adjusting the appearance of thermal patterns as a means to avoid duplicate marks, otoliths could also be distinguished by the application of strontium marking (Schroder et al. 1994). Strontium marking is conducted by exposing salmon alevin or fry to dilute solutions (1000 to 5000 ppm) of strontium chloride for 4 to 24 hour periods. Strontium chloride is a harmless salt. When fish are exposed to strontium in solution, the strontium is quickly absorbed by the tissues and over a period of several days it is incorporated into the otolith microstructure as strontium carbonate. This period of brief exposure period produces a highly concentrated band of strontium. Repeated exposures, with a sufficient elapse of time between periods will produce multiple bands. These strontium bands are analogous to thermal rings, but unlike thermal rings, they could not be misinterpreted as natural patterns. Strontium is found in extremely low concentrations in most freshwater systems, and in saltwater the concentration is only in the range of 10 to 30 ppm, thus it is unlikely that fish could naturally be exposed to sufficient amounts of strontium to produce a mark.

Strontium bands in otoliths can be detected with several different instruments, each of which represent an increasing level of detection power as well as expense. The same preparation used in thermal mark recovery - grinding the otolith to expose the central core - is used as a first step in all the detection methods. An additional preparation step is to insure there is a smooth polish on the otolith surface. The least expensive instrument to use is a Scanning Electron Microscope (SEM) equipped with backscatter imaging. Strontium is a heavier element than the more abundant calcium and thus has more valence electrons which are emitted when it is subject to the bombardment of an electron beam. These secondary electrons are known as backscatter electrons and detectors in SEM's use them to form real-time images on the view screen. Under this viewing mode, the bright band of strontium stands out in contrast to the calcium background in the otolith. With a properly outfitted SEM this detection process can be done relatively rapidly. The most time consuming process is waiting for the vacuum pump to clear the chamber of air.

The difficulty with SEM backscatter imaging is that it is only a very coarse method of detecting the presence of heavier elements. If the strontium band is not of

sufficient concentration then it may not be observed as a ring. In addition there appears to be differences in the quality of backscatter detectors on different models of SEM's.

More quantitative methods for detecting strontium includes WDS microprobe analysis which involves analyzing the x-rays emitted by strontium atoms when subject to an electron beam. The result is precise measure of strontium in the otolith at a particular location. Variants of this approach include elemental mapping, in which a digital picture of the otolith is created by converting the concentration of elements at each x-y coordinate into a gray value. This method however is quite time consuming and would likely be too expensive to use on a routine basis. These more sophisticated instruments, including PIXE, and LA-ICPMS, are available at many universities and research facilities. They are able to measure the concentrations of strontium at extremely low levels, much lower than would be necessary to detect the marks.

One of the primary challenges in using strontium as a mass mark will be in developing efficient methods to recover the marks with little additional expense. Backscatter imaging with an SEM is likely to be the most cost effective approach.

Another challenge in using strontium has been convincing the regulatory agencies to allow its use. However that appears to be changing. In Canada, mass marking sockeye with strontium has been taking place for the last three years (Doug Lofthouse, Department of Fish and Oceans, personnel communication). In 1998 approximately 1 million sockeye fry were marked at the Upper Point River north of Vancouver and 2 million at Henderson Lake on Vancouver Island. This was done as part of program to evaluate release strategies. The marking was carried out under an Experimental Studies Certificate with the Bureau of Veterinary Drugs, Health Canada. In the United States, Washington and Alaska have submitted a New Animal Drug Request form to the Food and Drug Administration (FDA) for approval of strontium as a marking agent. That request is currently under review. In the mean time, the Prince William Sound Aquaculture Corporation along with Alaska Dept. Fish and Game is seeking an Investigation of New Animal Drug (INAD) permit to strontium mark up to 30 million sockeye at the Gulkana incubation facility in Prince William Sound this spring. Three release groups would be marked using a one and two ring patterns.

While there is still much work to be done with the application of strontium marking, it may prove to be a useful as a secondary mark in association with thermal marking. Under this process, a base thermal mark would first be applied and some time after the fish hatch they would be exposed to strontium one or several times. To recover the marks, the otolith is examined with a light microscope to see if the thermal mark is present, if so it is then examined to determine the presence or absence of strontium rings. In that manner strontium marks can serve to compliment thermal mark patterns.

Strontium marking however will likely be used in many application where thermal marking is impractical or in possible. The Gulkana incubation site is one such example as there appears to be no other means to mark these fish at that location. In addition strontium marking can be done without the capital costs associated with heating or chilling equipment. This may be suitable for studies in which only one or two years of marking is needed for distribution studies. While it is likely premature to talk about the specifics of a strontium code structure, it would be important to start documenting information about their application in marking in a common database, similar to thermal marks.

References

Schroder, S. L., C.M. Knudsen, and E. C. Volk 1995. Marking salmon fry with strontium chloride solutions. *Can. J. Fish. Aquat. Sci.* 52:1141-1149.