Imprinting of Hatchery-Reared Salmon to Targeted Spawning Locations: A New Embryonic Imprinting Paradigm for Hatchery Programs

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Imprinting of Hatchery-Reared Salmon to Targeted Spawning Locations: A New Embryonic Imprinting Paradigm for Hatchery Programs

Straying by hatchery-reared salmon is a major concern for conservation and recovery of many salmon populations. Fisheries managers have attempted to minimize negative ecological and genetic interactions between hatchery and wild fish by using parr-smolt acclimation facilities to ensure successful olfactory imprinting and homing fidelity. However, the effectiveness of offsite acclimation for returning adults to targeted locations has been mixed. Since laboratory and field studies indicate that the period of hatching and emergence from the natal gravel is a sensitive period for olfactory imprinting, we propose an alternative imprinting approach wherein salmon are exposed as embryos to targeted waters transferred to their rearing hatchery. To test the feasibility of this approach, we conducted a series of electrophysiological and behavioral experiments to determine whether water can be successfully transferred, stored, and treated for pathogens without jeopardizing its chemical integrity. Stream water could be frozen or stored for one week at 4°C or 10°C without affecting the olfactory signature. Ultraviolet light treatment altered the responses of the olfactory epithelium to stream water; however, behavioral studies suggested that this treatment did not alter the attractiveness of this water. Finally, we describe several alternative approaches to embryonic imprinting using artificial odors.

Impronta en salmones cultivados para incidencia en sitios de desove: un nuevo paradigma embrionario de impronta en programas de cultivo

La fuga de salmones cultivados es un asunto considerable para la conservación y recuperación de muchas poblaciones naturales de salmón. Los manejar de pescaderas han intentado minimizar las interacciones negativas de orden ecológico y genético entre los peces cultivados y los silvestres mediante el uso de instalaciones en las que se asegure una impronta olfatoria y una filopatría exitosas. Sin embargo, la efectividad de la aclimatación remota para que los adultos regresen a los sitios de desove, no ha sido contundente. En virtud de que los estudios de laboratorio y de campo indican que el periodo de cultivo y emergencia en el sitio de nacimiento es un lapso sensible para que se establezca la impronta olfatoria, en este trabajo se propone un enfoque alternativo de impronta en el que el salmón, siendo embrión, es expuesto a sitios seleccionados a los que se les traslada desde las áreas de cultivo. Con el fin de probar la efectividad de este enfoque, se realizaron una serie de experimentos electrofisiológicos y etológicos para determinar si el agua puede ser exitosamente transferida, almacenada y tratada contra patógenos sin comprometer su integridad química. El agua de río puede ser congelada y almacenada por una semana a 4°C o 10°C C sin afectar su firma olfatoria. El tratamiento con rayos UV alteró las respuestas del epitelio olfatorio al agua de río; sin embargo, los estudios etológicos sugieren que este tratamiento no altera la atracción hacia este tipo de agua. Finalmente, se describen diversos enfoques alternativos a la impronta embrionaria utilizando olores artificiales.

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INTRODUCTION

Hundreds of millions of hatchery-reared salmon are released into waters of the United States annually (Rand et al. 2012). The hatchery programs that produce these fish are designed primarily to increase commercial, recreational, and tribal fishing opportunities, but increasingly they have become integral to recovery efforts designed to conserve native populations. The magnitude of these hatchery releases has raised concerns about potentially deleterious ecological and genetic interactions that may occur between wild and hatchery-reared salmon (Rand et al. 2012). One area of particular concern is that rearing and release practices used by many hatcheries may increase straying, the term for dispersal of individuals to nonnatal areas for reproduction, which can further increase undesirable interactions (Brenner et al. 2012). These concerns have led to calls for strict guidelines for hatchery programs to minimize straying to levels that will not impact native salmon populations. For example, a common guideline is that straying outside of the targeted area for a hatchery program should not exceed 5% or 10% (Paquet et al. 2011). Salmon are well known for their extraordinary homing migrations from the ocean to their natal stream for reproduction (Quinn 2005). Though some low level of dispersal from the natal site is normal in both wild and hatchery populations, some hatchery practices can dramatically increase the rate of straying (Pascual et al. 1995). Many hatchery rearing and release practices have been developed to increase survival and to optimize imprinting, but straying by hatchery fish remains a major concern for salmon managers. In particular, one of the most common approaches for imprinting fish to a specific location is to transfer and hold fish at sometimes expensive and logistically challenging acclimation facilities on the river or specific stream reaches that are targeted for homing. Here, we propose a new embryonic imprinting approach to improve successful imprinting and reduce straying by exposing embryonic salmon to waters collected from their targeted return location.

Homing is governed by olfactory discrimination of home-stream water, and exposure to the home stream during appropriate juvenile stages is critical for olfactory learning (imprinting) and successful completion of the adult homing migration (Dittman and Quinn 1996). Ensuring that juvenile salmon experience specific water sources during appropriate periods for imprinting can be a challenging problem for artificial production programs because logistical realities (e.g., access to ground water, ability to obtain construction permits, and financial cost) often require that salmon are incubated and reared at large centralized hatcheries that use water sources that are different than target waters. Furthermore, salmon are often transported between facilities and released off-site to supplement specific populations or fisheries. While most salmon will typically return as adults to their juvenile release site after transfer (Donaldson and Allen 1957), such transfers and off-site releases tend to increase the rate of straying from the targeted return site (Pascual et al. 1995; Hard and Heard 1999). To address this concern, many hatchery programs have developed specific acclimation and release facilities designed to optimize the imprinting process by allowing salmon to experience imprinting cues for an extended period prior to release during the parr-smolt transformation (PST), the developmental period characterized by endocrine, physiological, and behavioral changes that prepare salmon for life in the ocean (Hoar 1976).

PARR-SMOLT IMPRINTING AND ACCLIMATION

The PST acclimation strategy has been employed because the PST has been identified as a critical period for successful olfactory imprinting in both Pacific Oncorhynchus spp. (Hasler and Scholz 1983) and Atlantic Salmo salar (Morin et al. 1989) salmon. A long history of transport studies (Lister et al. 1981) and a series of experimental assessments of imprinting using artificial odors (Hasler and Scholz 1983; Morin et al. 1989; Dittman et al. 1996) have pointed to the PST as a sensitive period during which imprinting occurs. Subsequent laboratory studies have also demonstrated that the peripheral olfactory system is sensitized to imprinted odorants (Nevitt et al. 1994) and olfactory sensitivity increases during the PST (Morin and Doving 1992). Among the many endocrine changes that are associated with the PST is a distinct surge in the plasma levels of the hormone thyroxine (Dickhoff et al. 1978) that has been linked to successful olfactory imprinting (Hasler and Scholz 1983). This was demonstrated most clearly in experiments wherein Coho Salmon O. kisutch exposed to odors prior to the PST did not demonstrate long-term imprinting memories for these odors unless their thyroxine levels were also experimentally elevated (Hasler and Scholz 1983). Elevated thyroxine levels also stimulated proliferation of olfactory sensory neurons (Lema and Nevitt 2004) and have been linked to imprinting in other vertebrate species (Yamaguchi et al. 2012).

Though the PST is an important developmental period for imprinting, freshwater migratory patterns of wild juvenile salmon suggest that the process and timing of imprinting may be much more complex (Quinn 2005). The best example of this is Sockeye Salmon O. nerka, which typically spawn in streams flowing into lakes, and then, upon emergence from their natal sites, their offspring migrate to a nursery lake and rear 1–3 years before the PST and seaward migration. Upon returning from the ocean as adults, these fish spawn in their natal streams rather than the nursery lake they experienced during the PST. Complex and extensive migrations away from the natal site before PST are common for many salmon species (e.g., Daum and Flannery 2011), particularly in association with changing seasons, temperatures, flows, densities, and other ecological factors (Beckman et al. 2000), yet adults almost invariably return to their natal location to spawn. For example, Chinook Salmon O. tshawytyscha populations can migrate away from their natal site as either fry, parr, or smolts, and different populations have different proportions of migrants at different life stages (Healey 1991; Figure 1). These observations led us to hypothesize that the process of imprinting involves a complex interaction between developmentally regulated periods for imprinting, environmental stimuli (e.g., flow and temperature), and migration (Dittman and Quinn 1996). The diversity of juvenile migratory patterns coupled with extensive transport studies (reviewed in Lister et al. 1981) led Harden Jones (1968) and Brannon (1982) to propose a sequential imprinting hypothesis for salmon homing: salmon learn a series of olfactory waypoints, beginning at the natal site, as they migrate downstream to the ocean, and later retrace their path as returning adults using these waypoints to guide them (Figure 1). Under this scenario, returning salmon would be expected to return to their site of release and then, if available or detectable, seek an earlier imprinting signal until they reach their natal site (Figure 1).
The complexity of the imprinting process, combined with logistical realities of salmon artificial production programs, makes the management of salmon populations extremely challenging. The infrastructure required for large-scale artificial production (hatcheries, personnel, pumps, wells, etc.) essentially requires that fish are reared at large central facilities, whereas the population dynamics of these species require fine-scale outplants to ensure appropriate spatial and genetic segregation or integration of hatchery and wild fish, depending upon the program goals (Paquet et al. 2011). For segregated hatchery programs, designed to enhance harvest, the goal is typically to outplant salmon that will be captured in fisheries and also to ensure that those fish that avoid capture return to locations where broodstock can be collected or spawn when and where they will not interact with wild populations. On the other hand, the goal of integrated hatchery programs is to return hatchery-produced salmon to the same locations where wild fish spawn to enhance the wild population (Paquet et al. 2011). Finally, conservation hatchery programs are designed to reintroduce fish into historical or recovered habitat with the strategy of releasing fish that will imprint and ultimately return to these locations as adults.

All of these programs share a common dilemma: releasing salmon into the wild at earlier life stages provides a better opportunity for successful imprinting and homing, but releasing salmon at later life stages (i.e., larger sizes) provides a better opportunity for survival (Zabel and Achord 2004) and may reduce deleterious ecological interactions with other species (Pearsons and Temple 2010). These two competing concerns force managers of hatchery programs to weigh the likely tradeoffs of managing for natal homing versus managing for survival. In most cases, hatchery programs have adopted the smolt release strategy, taking advantage of the PST sensitive window for imprinting and the increased survival of larger fish reared through the PST in the hatchery. In many cases this strategy requires dedicated acclimation facilities, ranging from natural ponds to complete small-scale hatcheries, near the targeted site for returning adults (Figure 2). Most acclimation facilities are only operated during the spring prior to release, but some (e.g., Clarke et al. 2012) acclimate fish beginning in the winter prior to release. Parr-smolt acclimation and imprinting facilities have been developed or proposed as part of most hatchery supplementation programs in the Pacific Northwest, and hundreds of millions of dollars have been spent or proposed for construction, operation, and maintenance of these facilities.

For the most part, acclimation prior to release improves survival (e.g., Clarke et al. 2010, although see Kenaston et al. 2001), and most salmon tend to return to the vicinity of their release site (Garcia et al. 2004). However, offsite acclimation (i.e., moving parr from a central rearing hatchery to a smaller facility on a different stream prior to release) has not always been successful in providing adult returns to targeted locations (Dittman et al. 2010; Williamson et al. 2010). The major problem with acclimation sites is their locations relative to...
desired spawning locations for returning adults (Dittman et al. 2010; Williamson et al. 2010). If acclimation sites are located too close to initial rearing hatcheries, adults tend to return to hatchery locations rather than juvenile release sites (Lister et al. 1981; Dittman et al. 2010). Many acclimation sites were developed years ago before improvements in our understanding of the imprinting process and for different programmatic needs. Furthermore, siting of acclimation facilities is often driven by cost, site availability, environmental permitting, and physical access (e.g., roads and snow) issues rather than biology. This means that acclimation and release sites frequently must be located away from, and often downstream of, appropriate spawning habitat. It was hypothesized that salmon would return to their acclimation sites and then seek appropriate spawning habitat upstream, but in most cases studied, spawning was observed closer to acclimation sites rather than at locations farther upstream typically used by wild spawners (Dittman et al. 2010; Williamson et al. 2010). Thus, for parr-smolt acclimation and release strategies to successfully meet the needs of salmon management programs seeking to supplement spawning populations in specific tributaries or at even finer spatial scales, multiple expensive acclimation sites may be needed within each drainage system.

**EMBRYONIC IMPRINTING**

As an alternative, or complementary, approach to the use of parr-smolt acclimation facilities, we hypothesize that embryonic imprinting might be a useful management tool for achieving successful imprinting and homing fidelity to targeted spawning locations without moving fish from their central rearing hatchery prior to release. This new imprinting paradigm is based on the observation that while the PST is an important period for imprinting, salmon also imprint to their natal sites much earlier during development. In the wild, embryonic imprinting is evident from a range of studies that demonstrate very fine-scale homing to the natal site by multiple salmon species (Bentzen et al. 2001; Quinn et al. 2006). Furthermore, laboratory studies have demonstrated that embryonic salmon can distinguish and learn different natural waters based on chemosensory cues (Bodznick 1978), possibly even as early as prehatch eyed embryos (Courtenay 1989). This occurs during a sensitive window for imprinting during hatching and emergence from their natal gravel (Tilson et al. 1994; Figure 3). Using juvenile Sockeye Salmon, Tilson et al. (1994) demonstrated that these imprinting windows coincided with developmentally regulated surges in thyroid hormone levels as evidenced by strong attraction of maturing adult salmon to odors they were exposed to.
to at hatching and emergence (Figure 3). As suggested by the sequential imprinting hypothesis, it appears that wild adult salmon terminate their spawning migration upon reaching the area associated with olfactory cues learned in their natal redd. Therefore, we hypothesize that hatchery-reared salmon returning as adults will seek their earliest detectable imprinted olfactory waypoint as the appropriate location to terminate their spawning migration. Furthermore, if salmon are exposed as embryos to water derived from a targeted location upstream of their release site, they will, as adults, migrate past the release site and spawn at the targeted location.

We suggest that an alternative embryonic imprinting protocol may be useful for many hatchery programs. Using this protocol, hatchery salmon embryos would be exposed to natural waters from locations that managers want them to return to as adults (Figure 4). Rather than transport juvenile salmon from a central hatchery to desired spawning locations, we propose that water from these locations be collected and transported to a central hatchery for use during incubation and early rearing (Figure 4B). At these developmental stages, salmon embryos require relatively small volumes of water for incubation, so large numbers of embryos could be maintained in several small independent single-pass or recirculating systems within the hatchery. Upon emergence and ponding, salmon would be reared under normal hatchery protocols until release. Depending on the goals of the program and availability of parr-smolt acclimation facilities, juveniles would be directly released at locations downstream from the embryonic exposure sites or, ideally, acclimated at existing facilities downstream from the embryo water-exposure sites (Figure 4B). Fish from different upstream embryo-rearing sites could all be acclimated and released from a common site. We predict that returning adults would follow the sequence of odors they experienced as migrating juveniles to home to their release site. At that point, they would continue to migrate upstream to the source of the water they were exposed to as emergent embryos, where they would ultimately spawn (Figure 4C). We designed this protocol to facilitate reestablishment of sustainable natural populations of Pacific salmon in the Columbia River without the need for expensive, potentially environmentally harmful, and logistically challenging acclimation facilities, but we believe that this approach could be effective for all salmon species and locations.
PRACTICAL ISSUES

For embryonic imprinting to be useful and effective, several practical concerns must be addressed before widespread application. First, it is critical that water be collected and maintained in a manner that retains its odor qualities. Though the chemical nature of the odorant profile used by salmon to discern their natal stream is not known, it is hypothesized that these odors are a complex mixture of inorganic and organic chemicals from soil, plants, and aquatic organisms (Hasler and Scholz 1983). Recent work has demonstrated that different combinations of amino acids present in natural stream waters act as chemoattractants for homing salmon, and these compounds may represent part of the chemical signature salmon use to discriminate their homestream water (Shoji et al. 2003). Because organic compounds can be rapidly removed or altered by microbial consumers, care must be taken to ensure that the odor qualities of transported and stored water are retained during embryonic imprinting.

Figure 4. Schematic showing how embryonic imprinting could be applied to a supplementation hatchery program. (A) In a typical integrated hatchery program, wild adults are collected and spawned artificially, reared through the PST at a central hatchery, and then acclimated and released from dedicated acclimation sites. Upon return, adults often return to the vicinity of the release site rather than spawning at a targeted location upstream. (B) Using embryonic imprinting, fertilized embryos are exposed to stream waters collected and transported from targeted spawning sites. In this hypothetical case, water from Tributary A, which no longer has a spawning population, is used to imprint embryos and then to lure returning adults to Tributary A to help recolonize it. Water from Tributary C, which has a small wild spawning population, is used to imprint embryos and then lure returning adults to Tributary C to rehabilitate the wild spawning population. After embryo exposure, fish would be reared under normal protocols through the PST at the central hatchery and then acclimated and released directly or from dedicated acclimation sites. (C) We hypothesize that returning adults would follow the sequence of odors they learned as seaward migrating juveniles until they return to their release site. At that point, fish would seek an earlier imprinting cue, in this case the upstream water source (Tributary A or C) they learned as emergent embryos, and ultimately spawn in the vicinity of this “earliest” imprinting cue. (D) Under an alternative scenario, embryos could also be imprinted to artificial odors chosen by program managers. After normal rearing and release procedures, returning adults could be lured to targeted spawning sites they have never experienced by metering these artificial odorants into waters at the site.
To explore this question, we collected water from a proposed spring Chinook Salmon acclimation site on the White River, Washington, a tributary of the Wenatchee River in the Columbia River Watershed. To test odor stability under different storage regimes, we used an electro-olfactogram (EOG) technique that measures the olfactory responses of the salmon’s olfactory epithelium (Baldwin and Scholz 2005). Specifically, we used a technique termed “cross-adaptation” (Quinn and Hara 1986), wherein the epithelium is continuously exposed to the odors of freshly collected White River water (ambient temperature ~1°C) until the olfactory epithelium adapts and no longer responds to those odors. We then applied stored White River water. If storage alters the chemical nature of the water, then the olfactory epithelium will respond to these different chemicals and a response will be detected. A reciprocal test with each odor pair was also conducted. Using this technique, we found that White River water collected in January could be held for 7 days at either 4°C or 10°C or frozen (~20°C) for 7 days and thawed without altering the olfactory signature (Figure 5). This suggests that under the proper conditions, water can be collected, transferred, and stored for use in embryonic imprinting. However, more research needs to be conducted on different water sources, water collection and storage protocols, and water replacement procedures during imprinting. We also examined effects of using reconstituted White River water samples that had been freeze dried. For freeze drying, a known volume of water was frozen on dry ice–methanol, and then lyophilized under vacuum until all water was removed. The freeze-dried residue was then reconstituted in deionized water to the same volume as the original water sample. The reconstituted water elicited a response from olfactory epithelium that had been adapted to White River water, so this storage method did alter odor qualities of the original water sample (Figure 5). Further study of this method may be warranted to determine whether olfactory cues from the original water source can be preserved.

Additionally, because transferring natural stream water into a central hatchery for embryo imprinting has the potential to introduce pathogens, we were also interested in assessing whether treating the water to kill pathogens altered the water’s olfactory signature. Embryonic salmon are often initially reared in pathogen-free well water, but where stream water is used, it is typically treated with ultraviolet (UV) light or ozone to kill pathogens. In many cases, transferring natural stream water into a hatchery for embryonic imprinting would be prohibited unless that water was treated to remove pathogens. This could alter the water’s chemical composition and, therefore, the olfactory signature. To address this question, we again utilized the cross-adaptation technique using fresh White River water that was either treated with UV light to remove pathogens or left untreated. Interestingly, UV treatment apparently altered the chemical nature of White River water because UV-treated water elicited a different olfactory response than untreated water (Figure 6A). However, we wondered whether the overall odor qualities of the water were conserved enough to provide salmon with the chemical cues necessary to still allow them to distinguish this as White River water. To determine whether salmon could still recognize UV-treated water as equivalent to untreated river water, we conducted behavioral experiments on emergent fry, which tend to be attracted to water in which they were incubated (Bodznick 1978). For these experiments, we were unable to rear embryos in White River water, so we conducted a separate experiment using steelhead *O. mykiss* embryos incubated in Carnes Creek water at the Oregon Hatchery Research Center near Alsea, Oregon, and then tested fish for attraction to different waters at emergence. Emergent fry demonstrated a strong attraction to Carnes Creek water when given a choice of Carnes Creek water vs. well water in a two-choice maze. To assess the effect of UV treatment on the perception of Carnes Creek odor qualities, we tested whether emergent fry would choose untreated Carnes Creek water.
over UV-treated Carnes Creek water in a two-choice maze. We predicted that more fish would choose the untreated arm of the maze, if UV treatment altered the attractive qualities of the water. However, we observed no difference in attraction to treated and untreated water (Figure 6B). Though these results do not show that UV treatment did not alter the odor qualities that allow fish to distinguish Carnes Creek water, they suggest that any changes to treated water that occurred did not influence its attractiveness. Further studies of the effects of UV treatment and other sterilization techniques on odor qualities are needed before embryonic imprinting is accepted for use as a salmon rehabilitation or enhancement tool.

**ARTIFICIAL ODORS**

In some circumstances, concerns about disease, water stability, water volume requirements, and other logistical challenges may make transporting stream water to a central hatchery for embryonic imprinting impractical. However, this does not preclude the use of embryonic imprinting as a management tool. One alternative that has been proposed is the use of artificial imprinting odors to lure returning adult salmon to desired locations. Much of our understanding about olfactory imprinting comes from a series of groundbreaking experiments by Arthur Hasler and his colleagues in the 1960–1970s, in which they exposed juvenile salmon to the artificial odors morpholine and phenylethyl alcohol during the PST and then lured these salmon years later as returning adults into unfamiliar streams scented with these chemicals (reviewed in Hasler and Scholz 1983). Based on these studies, it has been suggested that artificial odorants could be used by salmon managers to manipulate migratory patterns and promote increased homing fidelity (Hasler and Scholz 1983). Initial studies indicated that adding artificial odorants to hatchery outlet water had little effect on homing fidelity (e.g., Rehnberg et al. 1985). However, combining artificial odorants with embryonic imprinting may provide a useful tool for integrated hatchery and supplementation programs to direct salmon to specific tributaries or reaches for spawning. Under this scenario, salmon would be exposed to artificial odorants in the central rearing hatchery using the same embryonic exposure system described earlier. We hypothesize that salmon will imprint to these artificial odorants and use them during the final stages of their adult homing migration. Therefore, fish imprinted to artificial odorants and released at a downstream location or acclimation site could be lured to an upstream site they had never experienced by metering the artificial imprinted odorant(s) into the river at the target site (Figure 4D).

One obstacle to utilizing artificial odorants is the lack of safe, inexpensive, and effective odorants for these studies. Early imprinting studies successfully used morpholine and phenylethyl alcohol; however, a more stringent regulatory environment may make these chemicals inappropriate for large-volume releases into natural waters. To be effective as a management tool for homing manipulation, artificial odorants ideally will (1) be safe for release into natural waters, (2) not impact nontarget taxa, (3) be inexpensive and readily available, (4) be stable for storage and after release into natural waters, (5) be detected by the salmon olfactory epithelium at relatively low concentrations, (6) not elicit innate behavioral (attraction or avoidance) or physiological (e.g., endocrine) responses, (7) elicit a learned behavioral response by juvenile salmon, and (8) allow imprinting of juvenile salmon and prove to be an effective cue for adult homing. Further research to identify and test appropriate chemicals will be required before this approach can be utilized.

Finally, another alternative approach to transporting water from a targeted homing location to the central hatchery would be to identify the chemical signature of stream water present at the targeted location and artificially recreate it for use in embryonic imprinting at the hatchery. As indicated earlier, Hasler and Scholz (1983) hypothesized that the odors allowing salmon to discriminate between waters consist of complex mixtures of inorganic chemicals, organic chemicals from soil and plants, and aquatic organisms. Ueda (2012) proposed that the primary chemical cues utilized by homing salmon are amino
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REFERENCES


Lema, S. C., and G. A. Nevitt. 2004. Evidence that thyroid hormone induces olfactory cellular proliferation in salmon during a sen-


