Technical Report

An Investigation into the Homing Patterns and Water Chemistry of Crawfish and West Crawfish Inlets Related to Hatchery Chum Production

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Introduction

At the inception of NSRAA's Crawfish Inlet fisheries enhancement program in 2014, utilization of a fall stock chum was intentional to avoid spatial overlap of returning hatchery adults with the wild summer chum run present in West Crawfish Inlet, originating from unnamed stream AWC #113-32-10050. This stock serves as an ADFG NSEO summer-run chum index stream and is currently being considered by ADFG for removal as an indicator stock. Scrutiny over hatchery chum straying into the West Crawfish headstream has prompted this study beginning in 2023 to investigate why adult NSRAA-produced chum are attracted to this location.

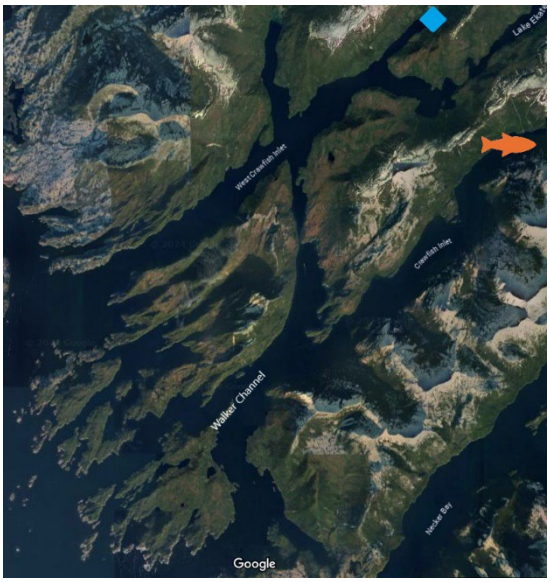

Figure 1. Crawfish Inlet fish rearing/release site location (fish), and unnamed stream AWC #113-32-10050 (diamond).

The anticipated adult migration route for hatchery fish returning to the Crawfish Inlet release site was through the mouth of the Inlet at Walker Channel (Figure 1). However, an unknown proportion of returning adult chum primarily enter West Crawfish Inlet and traverse through Cedar Pass before turning north to the release site in Crawfish Inlet proper. Adult chum have been observed in tidewaters near the index stream in August when NSRAA-produced fish are moving through West Crawfish Inlet en route to the release site, and wild summer chum are staging to spawn.

By examining the outmigration behavior of fry post release, conducting carcass surveys in the West Crawfish headstream to evaluate the proportion of hatchery spawners, and comprehensive water quality testing of streams that may interfere with imprinting within the fry outmigration route, we hope to gain an understanding of why this production of fish is behaving so and explore mitigation strategies.

Hatchery Chum Fry Outmigration Monitoring

One hypothesis as to why NSRAA-produced adult chum are annually attracted to this unnamed stream is that once released, fry undertake the reverse migration route as the adults prior to fully completing their olfactory imprinting phase. In this case, any fry that reside in West Crawfish Inlet at or near the unnamed stream may imprint on this water source and potentially stray from the release site as an adult. By intercepting fry post-release and then reading the otolith to determine origin, we hope to gain a rudimentary insight into hatchery fry migration behavior. However, it must be stated that the organization understands that simple interception will not tell us two things. First, interception of fry will not denote residency time in a given location without the implementation of acoustic tagging equipment or mark-recapture methodology. Second, it's unclear whether the hatchery-origin spawner carcasses recovered in the unnamed stream actually imprinted and successfully homed to the unnamed stream, or did they imprint appropriately to the release site and simply stray into the stream and capitalize on suitable

spawning habitat. Regardless, an objective of this study is to gain some baseline information about fry behavior post-release.

A sub-hypothesis is that the dissolved free amino acid (DFAA) profile in the unnamed stream water is similar to that of the rearing site, or to the hatchery water source in which these fish are incubated. Studies such as Yamamoto et al. (2013) emphasize that DFAA play a role in salmon imprinting to guide nearshore freshwater homing. Dissolved free amino acids come from many sources in streams (Thomas, 1997), such as biofilm on substrate and sediment, as well as organic mucus and metabolic waste of adult spawners (Dittman & Quinn, 2020). Additionally, numerous studies have demonstrated that many bony and cartilaginous fish species, including Pacific salmon, exhibit the characteristic of an embryonic olfactory imprinting window during hatching and gravel emergence (Kimmel, et al. 2023, Havey, et al. 2017).

Finally, a plausible speculation worth noting is that adults are attracted to this stream because it is somewhat of a "geographic trap" given that it is the first freshwater source in a direct straight line from open sea in what we now know to be their nearshore migration route. The turn into Cedar Pass is an acute angle, which can often prove challenging for salmon to navigate (NMFS, 2011). This theory, however, is difficult to fully assess.

2024 Results

Seven surveys were conducted at nine locations to recapture hatchery fry post release to gain insight into their migration route (Figure 2) in May 2024. The primary method of capture was via beach seine; fishing a small beam trawl was also attempted but proved unsuccessful. Otoliths were extracted from 907 subsampled fish and read to determine origin.

Contrary to survey results in 2023 where no hatchery-origin fry were captured west of Cedar Pass, this year they were recaptured at all survey locations. Proportions of hatchery to unmarked fish in a given location, however, were as expected with unmarked fish making up the majority of catch in West Crawfish (Table 1). The "late large" 4-gram release group (compared with the 2-gram group) was detected at higher instances in all four West Crawfish locations, suggesting they are highly mobile, but did not necessarily move offshore more quickly than the 2-gram group. One possible explanation for this difference in detection prevalence between the two size release groups, and more importantly potential influence in catch selection between origins, is the ¼" mesh size of the beach seine

Figure 2. 2024 Fry capture locations in relation to release site. Stars represent beach seine locations, lines for beam trawl transects.

Table 1. Composition of catch by origin (2024).

	Location	No Mark	NSRAA
1	West Crawfish Inlet	87%	13%
2	Shamrock Bay	93%	8%
3	4 Corners Lake Outlet	61%	39%
4	Outer West Crawfish	80%	20%
5	Cedar Pass	39%	61%
6	Lodge Island (North)	31%	69%
7	Lodge Island (South)	8%	92%
8	Walker Channel	52%	48%
9	Jamboree Bay	6%	94%
	Grand Total	**50%**	**50%**

Table 2. Size of fish in grams by origin and location (2024).

Average of Weight (g)		
Location	**No Mark**	**NSRAA**
West Crawfish Inlet	0.69	1.63
Shamrock Bay	1.22	2.90
4 Corners Lake Outlet	1.53	2.52
Outer West Crawfish	2.05	3.76
Cedar Pass	1.41	1.79
Lodge Island (North)	1.45	1.88
Lodge Island (South)	2.00	2.21
Walker Channel	0.92	1.74
Jamboree Bay	2.01	2.32
Grand Total	**1.33**	**2.19**

which was large enough for small fish to escape through. Staff made concerted efforts to land catch quickly but inevitably there was some catch bias towards larger fish which is reflected in Table 2, with hatchery-origin fry measuring almost 40% larger than their wild counterparts (Table 2.) Seine equipment will be modified with smaller mesh for surveys in 2025.

A solution for eliminating catch bias towards larger fish which proportionally overrepresent hatchery fish is to undertake an acoustic telemetry tagging study. Even with very small mesh size, beach seining surveys rudimentarily demonstrate presence and absence of hatchery fry and will not tell us residency time in a specific location. By better understanding their outmigration behavior in combination with water quality analysis data, we hope to discern if fish are departing the release site and then imprinting to other locations with more suitable estuary habitat, such as the West Crawfish Inlet headstream.

2023 Results

Five sites throughout the migration corridor that featured a stream water source providing the potential proclivity for imprinting were selected for hatchery-origin chum fry presence surveys (Figure 3). A beach seine survey regime to evaluate fish detection spatially and temporally in the assumed outmigration corridor relevant to release date. The survey events occurred immediately prior to release, 2 days, 10 days, and 20 days post release.

Otolith mark presence results indicated no hatchery fish were detected at any of the three survey sites in West Crawfish Inlet. The proportion of hatchery-origin fry was far lower than expected given the very large schools of fry numbering in the several hundred thousand to millions observed at the surface in West Crawfish Inlet, specifically in Shamrock Bay and West Crawfish Inlet Head.

Based on these survey results, our hypothesis that hatchery fry are migrating from the rearing site to the unnamed stream AWC #113-32-10050 shortly after release to complete/complement

their olfactory imprinting period appears unsubstantiated.

Table 3. Origin of fry by origin and capture location (2023).

	Pre	2 d Post	10 d Post	20 d Post	
Otolith Mark	22-May	25-May	2-Jun	12-Jun	Grand Total
Crawfish Inlet Head		**29**	**30**	**2**	**61**
CRAWFISH22		15	26	2	43
CRAWFISH22LL		14	4		18
Jamboree			**30**	**1**	**31**
CRAWFISH22				1	1
No Mark			30		30
Cedar Pass	**28**	**20**	**29**		**77**
CRAWFISH22		2	2		4
CRAWFISH22LL		4	1		5
No Mark	28	14	26		68
Shamrock	**30**	**27**	**30**	**1**	**88**
No Mark	30	27	30	1	88
W Crawfish Head	**30**	**29**	**30**		**89**
No Mark	30	29	30		89
W Crawfish N Arm Head	**13**	**5**	**29**		**47**
No Mark	13	5	29		47
Grand Total	**101**	**110**	**178**	**4**	**393**

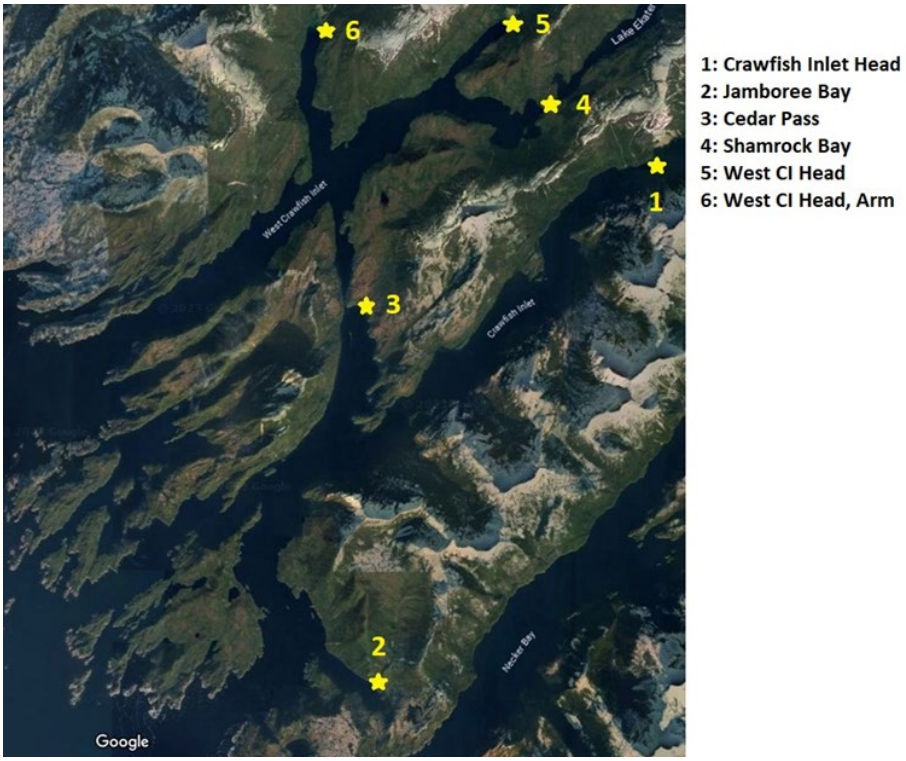

Figure 3. Beach seine survey locations in 2023.

Spawner Survey of West Crawfish Inlet NSEO Index Stream

2024 Results

Four foot surveys were conducted weekly beginning August 5th, 2024 for chum carcasses in the West Crawfish index stream. Staff walked ~1.3 miles upstream from the high tide line and opportunistically sampled a total of 118 expired chum spawners for collection of otoliths to determine origin. No expired fish were observed on the first survey (Aug 5th). Sampled fish were GPS marked to evaluate not only entry timing of hatchery-origin chum but also hatchery-spawner range within the watershed. Three independent stream "reaches" were designated based upon changes in spawning habitat and landmarks (Figure 4). It is important to note that these surveys extended farther beyond the range that ADFG traditionally conducts their escapement surveys for this index stream. This raises the question of possible undercounting of wild fish and overrepresentation of hatchery fish, which was exemplified in the data this year in distribution between hatchery and wild spawners by reach.

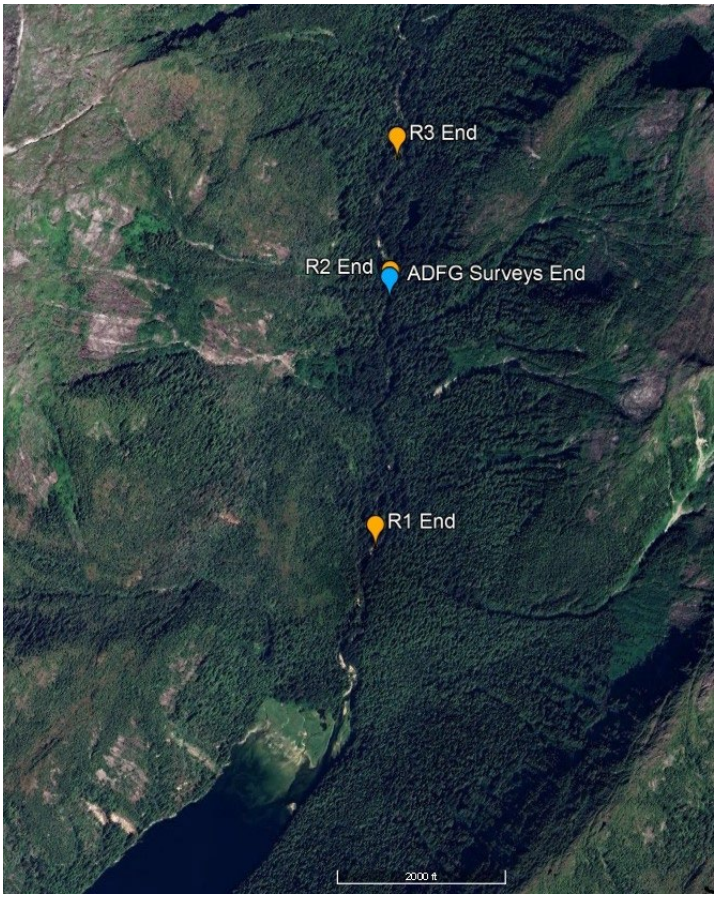

Figure 4. Map of West Crawfish Inlet index stream spawner survey reaches.

The second survey conducted on Aug 13th revealed that in all three reaches the proportion of unmarked chum was greater than that of hatchery origin (Figure 5). In Reach 3, the farthest upstream, no NSRAA-origin fish were detected. This is not surprising given that spatial segregation would be expected given the difference in summer and fall stock run timing.

The subsequent survey conducted on Aug 20th resulted in the largest day overall in terms of number of fish sampled. Reach 1 held a greater proportion of hatchery-origin fish which aligns with peak run timing of this stock in stat week 33. However, as seen in Reach 2 this majority shifts back to unmarked fish as distance from saltwater increases. No fish irrespective of origin were detected in Reach 3.

The fourth and final survey was conducted with assistance from ADFG on Aug 27th. Reaches 1 and 2 held >80% hatchery-origin fish, but interestingly in Reach 3 this majority dramatically shifts back to primarily unmarked fish. This is suggestive that NSRAA-origin fish are not utilizing the full spawning habitat range and primarily residing in lower reaches of the stream. This may consequently result in less

spatial overlap between the end of the summer-stock wild run and the early-arriving fall-stock hatchery spawners, as well as an inaccurate overrepresentation of hatchery fish in ADFG escapement surveys.

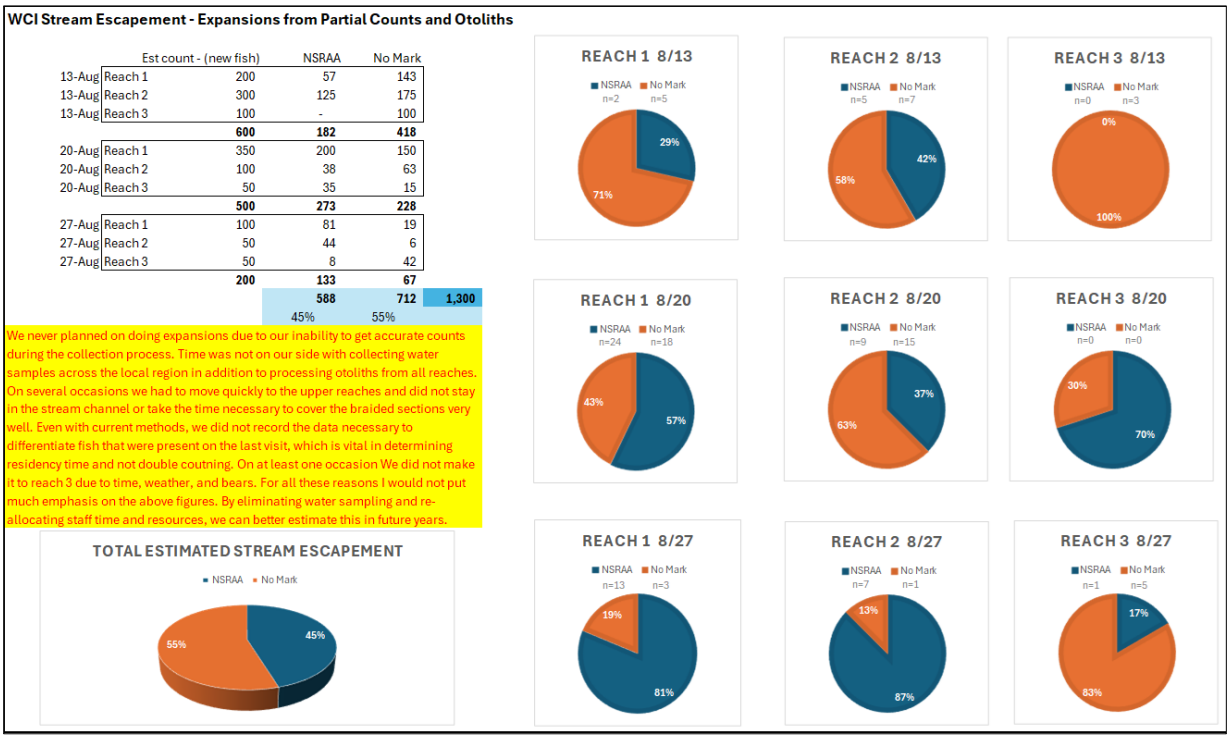

Figure 5. Composition of spawners by origin and stream reach over time (2024).

2023 Results

One survey was conducted on September 6th, 2023 for expired chum spawners in the West Crawfish Inlet index stream. NSRAA staff members walked ~750 ft upstream from the high tide line and opportunistically sampled 94 adult chum spawner carcasses for otolith collection. Given the close proximity that these carcasses were found in relation to the high tide line, where hatchery-origin fish are known to nose into the stream and exit over the following tide cycles, true abundance could not be estimated. The higher proportion of hatchery-origin females to males could be that females had a stronger commitment to the spawning location given the presentation of suitable habitat. Future surveys will encompass a larger range of spawner habitat beyond immediately upstream of the tideline.

Origin	F	M	Grand Total
BEARCOVE18LL	1		1
CRAWFISH18LL	1	2	3
CRAWFISH19	21	8	29
CRAWFISH19LL	14	13	27
CRAWFISH20	3	1	4
CRAWFISH20LL	1	4	5
No Mark	12	11	23
Overground		3	3
Grand Total	**53**	**42**	**95**

Figure 6. Composition of spawners by origin and sex (2023).

Water Quality Sampling

Influent water samples were collected in May and August 2024 from Medvejie and Sawmill Creek Hatchery' incubation water sources, as well as seven streams within Crawfish Inlet, West Crawfish Inlet, Walker Channel, Lodge Island, and the Four Corners/Cedar Pass area to gather data on freshwater sources that fry may encounter within the outmigration corridor. Samples were analyzed for 11 standard water quality metrics such as pH, alkalinity, dissolved solids, phosphorus, magnesium, etc. Fry beach seine survey locations were used as collection points because of the suggestive nature that if fish are present nearshore, there is presumable exposure to the freshwater runoff source. Fry are continuously imprinting to their outmigration route as they move toward ocean entrances.

In 2023 water samples were also collected from most of the above stated locations and analyzed by NOAA Auke Bay Lab for 17 individual amino acid concentrations known to be present in salmon spawning streams and influence imprinting. Amino acids in stream water are derived from algae, detritus and the mucus from other fish in the system, and act as the "scent" in the water providing cues to aid in nearshore migration navigation for returning adult salmon (Dittman, 2015; Yamamoto, 2009). Juvenile salmon imprint on amino acids in their natal stream (or release site), therefore we are interested to know if there are similar watersheds within their migration corridor, such as the West Crawfish Inlet headstream, that are attractive to returning adults because of the chemical similarity to the release site. Samples were collected again in 2024 and results are pending.

Mitigation Strategies & Future Work

Integrated Multitrophic Aquaculture (IMTA)

Research into the homing behavior of the Crawfish Inlet enhancement production will continue with monitoring hatchery fry outmigration as well as survey the index stream to evaluate proportion of hatchery origin spawners (pHOS). NSRAA plans to continue collaboration with other researchers such as the Sitka Sound Science Center (SSSC), University of Alaska Southeast (UAS), and NOAA to explore methods of improving chum salmon homing accuracy, including testing the feasibility of utilizing a macroalgae (kelp) derived compound for improved imprinting (Machado, 2020). Fisheries research biologists have demonstrated that salmon can be artificially imprinted to synthetic chemicals (Hasler, 1978; Dittman, 2015). This technology can be applied to aid in homing fish to specific locations, such as increasing hatchery fidelity, or targeted return locations for fisheries enhancement.

Implementation of IMTA practices in conjunction with salmon enhancement could impart dissolved free amino acid olfactory cues onto imprinting juvenile salmon and offer ancillary ecological benefit to the marine environment, as IMTA has demonstrated the potential to improve surrounding water quality and benthos of fish rearing sites, provide habitat for released juveniles, as well as provide added economic value to the same footprint (Costa-Pierce, 2010; Troell, 2009).

In 2024, baseline data collection fieldwork was performed by NSRAA staff in collaboration with the UAS Mariculture program at most of our fish release sites to evaluate suitability of IMTA implementation. Water samples for nutrient availability analysis were collected, as well as oceanographic sensors measuring temperature, conductivity, and current patterns. A pilot project examining utilization of a macroalgae-derived compound to imprint hatchery fish onto that will then later serve as a homing cue for returning nearshore adults is currently in development. In collaboration with researchers at the SSSC, UAS and salmon olfactory expert biologists at NOAA, we are undertaking a project to develop a unique amino acid profile to administer to fish during the embryonic imprinting window in pink salmon. Returning adults will then be exposed to this same olfactory cue to assess homing fidelity. The Sheldon Jackson Hatchery (SJH) at the SSSC releases pink salmon for education and enhancement purposes and should funding be secured for an IMTA pilot project, would serve as an excellent location for testing this concept given the short life cycle of pink salmon. NSRAA and co-investigators feel that this technology has implications for remediating hatchery fish homing issues.

Acoustic Radiotelemetry Tagging

In 2025, NSRAA plans to implement an acoustic telemetry study to investigate the nearshore homing patterns in adult chum returning to districts 113-32 and 113-33. NSRAA has performed similar acoustic telemetry research into chum homing behavior in 2003 and 2004 relative to enhanced adults returning to Deep Inlet in Sitka Sound. By tagging troll-caught adult chum at the entrance of West Crawfish and Crawfish Inlets and performing opportunistic vessel-based otolith reads to confirm the general origin makeup of the school, we intend to monitor migration patterns by stationary and mobile vessel receiver antennas and evaluate residency time and transient migration patterns in the West Crawfish corridor.

References

Costa-Pierce, B. (2010). Responsible use of resources for sustainable aquaculture. Network of Aquaculture Centres in Asia-Pacific.

Dittman, A. H., Pearsons, T. N., May, D., Couture, R. B., & Noakes, D. L. G. (2015). Imprinting of Hatchery-Reared Salmon to Targeted Spawning Locations: A New Embryonic Imprinting Paradigm for Hatchery Programs. *Fisheries*, 40(3), 114–123. https://doi.org/10.1080/03632415.2015.1007206

Dittman, A. H., & Quinn, T. P. (2020). Amino acid cues emanating from Pacific salmon eggs and ovarian fluid. *Journal of Fish Biology*, *97*(5), 1408–1414. https://doi.org/10.1111/jfb.14507

Dittman, A. H., Cunningham, C. J., & Quinn, T. P. (2022). Can unique amino acid profiles guide adult salmon to natal streams? A comparison of streams sampled prior to and after the arrival of adult Pacific salmon. *Hydrobiologia*, *849*(16), 3501–3513. https://doi.org/10.1007/s10750-022-04948-z

Hasler, A., Scholz, A. T., & Horrall, R. M. (1978). Olfactory imprinting and homing in salmon. *American Scientist*, *66*(3), 347–355.

Havey, M. A., Dittman, A. H., Quinn, T. P., Lema, S. C., & May, D. (2017). Experimental Evidence for Olfactory Imprinting by Sockeye Salmon at Embryonic and Smolt Stages. *Transactions of the American Fisheries Society (1900)*, *146*(1), 74–83. https://doi.org/10.1080/00028487.2016.1238409

Kimmel, J. G., Buchinger, T. J., Larson, D. L., Baker, E. A., Zorn, T. G., Scribner, K. T., Li, W., & Cooke, S. (2023). Behavioral evidence of olfactory imprinting during embryonic and larval stages in lake sturgeon. *Conservation Physiology*, *11*(1), coad045-. https://doi.org/10.1093/conphys/coad045

Machado, M., Machado, S., Pimentel, F. B., Freitas, V., Alves, R. C., & Oliveira, M. B. P. P. (2020). Amino Acid Profile and Protein Quality Assessment of Macroalgae Produced in an Integrated Multi-Trophic Aquaculture System. *Foods*, 9(10), 1382-. https://doi.org/10.3390/foods9101382

NMFS (National Marine Fisheries Service). (2011). Anadromous Salmonid Passage Facility Design. NMFS, Northwest Region, Portland, OR.

Thomas, J. D. (1997). The role of dissolved organic matter, particularly free amino acids and humic substances, in freshwater ecosystems. *Freshwater Biology*, *38*(1), 1–36. https://doi.org/10.1046/j.1365-2427.1997.00206.x

Troell, M., Joyce, A., Chopin, T., Neori, A., Buschmann, A. H., & Fang, J.-G. (2009). Ecological engineering in aquaculture — Potential for integrated multi-trophic aquaculture (IMTA) in marine offshore systems. *Aquaculture*, 297(1), 1–9. https://doi.org/10.1016/j.aquaculture.2009.09.010

Yamamoto, Y., & Ueda, H. (2009). Behavioral Responses by Migratory Chum Salmon to Amino Acids in Natal Stream Water. *Zoological Science*, 26(11), 778–782. https://doi.org/10.2108/zsj.26.778

Yamamoto, Y., Shibata, H., & Ueda, H. (2013). Olfactory Homing of Chum Salmon to Stable Compositions of Amino Acids in Natal Stream Water. *Zoological Science*, *30*(8), 607–612. https://doi.org/10.2108/zsj.30.607