

Mechanisms of Salmon Homing Migration from Molecules to Behavior

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Salmon have an amazing ability to migrate thousands kilometers from the open ocean to their natal stream for reproduction after several years of oceanic feeding migration. It is now widely accepted that some specific factors of the natal stream are imprinted to particular nervous systems of juvenile salmon during downstream migration, and that adult salmon evoke these factors to recognize the natal stream during homing migration. A number of studies have investigated the amazing ability of salmon to migrate long distances from the ocean to their natal river for spawning (Ueda and Shoji 2002). For a better understanding of the mechanisms of salmon homing migration, three different analyses have recently been applied using Japanese chum salmon (*Oncorhynchus keta*) homing migration in Hokkaido, Japan.

The first is behavioral analysis of upstream tactics of chum salmon through a rehabilitated segment of the Shibetsu River by electromyogram (EMG) telemetry and micro-data logger tracking in collaboration with Yuya Makiguchi, Masumi Akita (Hokkaido University), Hisaya Nii, and Katsuya Nakao (Hokkaido Farming Fisheries Promotion Corporation). To restore the ecological condition of a previously channelized river to its more natural, meandering state, a segment of the Shibetsu River was reconstructed. We investigated the upstream tactics of chum salmon as they migrated up the channelized segment, reached the confluence point with the restructured segment, and then entered the rehabilitated area. Fish were radio tracked with EMG telemetry and micro-data loggers. The monitored salmon preferred to hold in deep, slow moving areas of the channelized river, and EMG telemetry data revealed that these extended holding times provided the fish with efficient rest. In the reconstructed segment, the fish swam at more shallow depths and against stronger currents. Although all fish chose to enter the reconstructed segment, few fish remained in the segment for long. This implies that the reconstructed area may not provide suitable holding sites for migrating salmon.

The second is endocrinological analysis of hormone profiles in the brain-pituitary-gonadal axis in collaboration with Tomoko Kitani (Hokkaido University), Masafumi Amano (Kitazato University), and Masatoshi Ban (National Salmon Resources Center). Gonadotropin-releasing hormone (GnRH) molecules produced in the various brain regions are considered to be involved in many physiological functions of teleost life cycle. In order to clarify GnRH roles on salmon homing migration, two molecular types of GnRH, salmon GnRH (sGnRH) and chicken GnRH-II (cGnRH-II) in different brain regions, as well as gonadotropin (GTH) and steroid hormones, were measured using specific time-resolved fluoroimmunoassay (TR-FIA) systems (Yamada et al. 2002; Leonard et al. 2002). Maturing chum salmon were caught at nine points from the Bering Sea to the Chitose River. After decapitation, the brain was divided into six regions: olfactory bulb (OB), telencephalon (TC), diencephalon (DC), optic tectum (OT), cerebellum (CB), and medulla oblongata (MO). During the spawning migration of chum salmon, sGnRH levels in OB, TC, and pituitary of both sexes increased from the coastal sea to the point where the Chitose River branches from the Ishikari River. Moreover, sGnRH levels in the pituitary tended to increase at the same time as the elevation in female pituitary GTH II and ovarian GTH I levels. The cGnRH-II level in MO was increased at the pre-spawning ground in both sexes, and levels of OT were also increased in males. Both GnRH levels in DC showed no significant changes during spawning migration. The GTH II levels in gonads were not detected though the sampling period. Serum steroid hormone levels showed profiles similar to previous observations (Ueda 1999); estradiol-17 β in females and 11-ketotestosterone in males increased during vitellogenesis and spermatogenesis, respectively, and 17 α ,20 β -dihydroxy-4-pregnen-3-one increased dramatically at the time of final gonadal maturation in both sexes. It is quite interesting to note that both sGnRH content in TC and serum testosterone level showed coincident peaks at the branch point of the Chitose River from the Ishikari River. These results confirm the previous findings that sGnRH plays a role in GTH secretion in the pituitary of chum salmon, and sGnRH and cGnRH-II might be involved in brain region-dependent roles of sexual maturation and behavior in salmonid fishes.

The third is olfactory analysis of the ability to discriminate the natal river in collaboration with Yuzo Yamamoto (Hokkaido University) and Takayuki Shoji (Tokai University). For upstream homing migration from the coastal area to the natal stream, the olfactory hypothesis proposed by Hasler and Wisby (1951) has been discussed in many behavioral and electrophysiological studies, but the odor substances of the home stream are still unknown. We found that the response to artificial stream water based on the compositions of amino acids and salts closely

resembled the response to the corresponding natural water (Shoji et al. 2000), and we carried out behavior experiments to test whether amino acid mixtures of the home stream have attractive effects on the upstream movement of chum salmon. Mature male chum salmon (mainly four year olds) were captured at the weir in the Osaru River, Hokkaido, Japan, in the late spawning season of 2002, transferred to the Toya Lake Station, Hokkaido University, and reared for several days before experiments. Behavior experiments were conducted in the two-choice test tank. Artificial home stream water was prepared by dissolving the amino acid and related-substance composition of the Osaru River in artificial freshwater. A total of 44 chum salmon was tested, and 28 fish (63.6%) showed upstream movement to one choice arm of the tank. Among those that moved, 24 fish (85.7%) were found in the arm with artificial home stream water, and four fish (14.3%) were observed in the arm with natural lake water. These results demonstrate clearly that artificial home stream water, reconstituted by the amino acid composition of the home stream, has attractive effects on the upstream-selective movement of chum salmon. We concluded that amino acids dissolved in the home stream water are home stream odorants, and the hypothesis that amino acids dissolved in stream waters are home stream substances for salmon homing is strongly supported by these results.

By means of these different new approaches, the following three topics will be future research projects on chum salmon homing migration in my laboratory:

- 1) What kinds of environmental factors might influence the endocrinological changes of salmon that initiate homing migration?
- 2) When, where, and why might salmon cease foraging behavior during the homing migration?
- 3) What kinds of sensory cues might salmon use to navigate for a migration of several thousands of kilometers, and how would endocrinological changes influence sensory cues during the homing migration?

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