FINAL REPORT

To

The Kootenai Indian Tribe of Idaho
P.O. Box 1269
Bonners Ferry, Idaho 83805

TITLE: Behavior and Habitat of Young Kootenai River White Sturgeon− 2008

by

Boyd Kynard[1] and Erika Parker[2] (Co-PIs)

Brian Kynard[1] (Proj. Ldr.) and Tim Parker[1] (Technician)

[1] BK-Riverfish, LLC, 28 Echo Hill Rd., Amherst, Mass. 01002

[2] U.S. Geological Survey, Leetown Science Center
S.O. Conte Anadromous Fish Research Center
Turners Falls, Mass. 01376

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Summary

We studied behavior of Kootenai River white sturgeon *Acipenser transmontanus* free embryos, larvae, and year-0 juveniles during studies in 2008. Experiments found density of juveniles (single fish vs. groups of eight fish) during foraging and wintering had no significant effect on substrate preference. Thus, fish can be tested for habitat preference in groups.

During tests to determine the effect of four combinations of small, medium, and large rocks on downstream movement and selection of rocks by free embryos (days 0−11 fish), fish selected small rocks (3−6 cm diameter pebble) significantly more than medium size rocks (7−12 cm diameter small rubble), or large rocks (13−20 cm diameter medium-large rubble). However, ≤ 10% of the free embryos were moving downstream in any of the four rock combinations, so any composition provided habitat for the 15 free embryos in tests. The data on selection of rock size suggested restored spawning−rearing habitat for eggs and free embryos should have between 30−50% small pebbles for free embryos. Larger rubble should suffice for spawning and egg attachment−rearing. Although the lowest number of fish passes by early larvae (days 12−16 fish) occurred when the highest percent of large rubble (60%) was present, rock size did not have a significant effect on downstream movement. Also, most early larvae using rocks continued to use small rocks, like free embryos. With increasing larval age, an increasing percent moved downstream and selected open habitat.

Location and behavior of days 17−40 larvae moving downstream one-half loop around the stream tanks with 60% small vs. 60% large rocks follow: (1) in all tanks, most fish were at the water surface (30 cm above the bottom) or in the water column, and (2) most were in the fastest flow near the outside wall. Mean movement speed (ground speed) of larvae in the 17 cm/s (mean water velocity) was 14.4 cm/s (range, 1.85−31.9 cm/s). Thus, fish were moving at about 84% of the mean water speed. There was a significant trend for fish speed to be slower over small substrate ($P = 0.03$), an unexpected result. Modeling the dispersal of wild foraging larvae that encounter and respond to diverse physical factors (velocities and bottom substrates) and biological factors (in particular, forage habitat and prey abundance) will be difficult because of the lack of data on behavior of larvae in the river.

Winter underwater video surveys in the meandering reach of the Kootenai River found sand was the dominant substrate and small woody debris was the dominant cover. In the river delta, mud was the dominate substrate in shallow water (0.9−1.8 m deep) and sand dominated the deep water samples (15.2−27.4 m deep). Small woody debris and logs were the dominant cover types in the lower river sites; sparse vegetation and detritus were the dominant cover types in the delta. Previous studies found wintering year-0 juveniles preferred sand not rock substrate, and sand is abundant in both the lower river and delta.

Summer underwater video surveys in the meandering reach found sand dunes, clay terraces, and mud were the dominant substrate types; whereas in the littoral zone of the lake, mud dominated shallow water sites (0.9−1.8 m deep) and sand dominated all other sample sites in mid-depth (7.6−15.1 m deep) or deep water (15.2−27.4 m deep). In the river, sparse vegetation, detritus, small rocks, and clay bits were the dominant cover types, whereas in the littoral zone of the lake, cover was mainly detritus or sparse vegetation at all depths.

Introduction

We tested habitat preference of single vs. groups of Kootenai white sturgeon (hereafter, Kootenai sturgeon) year-0 juveniles during the fall and winter to determine if fish density affects preference. These data are needed before proceeding with summer and winter habitat tests.

Some of the present studies continue the study of innate habitat preferences of young life stages, particularly preference for bottom habitat. Sturgeons are benthic foragers, so the relationship of larvae and juveniles to substrate type where food is located is probably strong. Laboratory observations on young Kootenai sturgeons found that fish foraged on the bottom and vertical walls of tanks.

The spawning and rearing habitat of eggs and free embryos of shortnose sturgeon *A. brevirostrum* has rocks from large rubble to small gravel (Kieffer and Kynard unpubl. data). The high variability of rock sizes creates variability for surface area, interstitial spaces, etc. Hatchling free embryos of Kootenai sturgeon seek hiding places and few move downstream (Kynard and Parker 2006, Kynard et al. 2007). Fraser River white sturgeon select small substrate (S. McAdam personal communication), but in most Kootenai sturgeon spawning areas, the rocky habitat is likely to be variable, not uniformly small. We conducted tests in artificial streams to evaluate the effect of different rock-size compositions on downstream movement and selection of rock size by free embryos to better understand the effect of rock composition on behavior of free embryos to provide guidance to river habitat restoration efforts.

During 2005–2007, our studies on young Kootenai sturgeon behavior in artificial streams repeatedly showed that larvae have a long dispersal style that likely takes them to the lower part of the meandering reach of the Kootenai River or even to the river delta or to Kootenay Lake (Kynard and Parker 2006, Kynard et al. 2007, Kynard et. al. unpubl. data). In 2008, we conducted preliminary tests to investigate movement speed and behavior of larvae relative to the percent of two dominant rock sizes: 60% small pebble vs. 60% large rubble.

We continued a survey of bottom type and cover type of the Kootenai River and Kootenay Lake using an underwater video camera system. These survey data and the data from summer 2007 (Kynard et al. 2008) provide the basis for selecting the bottom types to test in habitat preference tests with larvae and year-0 juveniles.

One of the unknowns about life history of sturgeons is the importance of imprinting by early-life intervals during life history. This information has great significance for all restoration programs that involve rearing early life stages away from the natal reach, or rearing and stocking juvenile sturgeons, which are not allowed to complete natural movements. Studies on shortnose sturgeon indicate interruption of natural movement patterns causes dysfunction later in life when fish are mature (Kynard et. al. unpubl. data).

Shortnose sturgeon, like other sturgeons yet studied in North America, spawn at small discrete sites and the offspring do not return until they return to spawn (like Kootenai sturgeon). This life history migration pattern suggests imprinting of the life stage(s) at or near the spawning area may occur to provide fish information on their birth reach. In shortnose sturgeon, like Kootenai sturgeon, free embryos remain at the spawning site after hatching until they develop into larvae, and then disperse downstream (Kynard and Parker 2006, Kynard et al. 2007). Do the free embryos imprint to the olfactory characteristics of the water in their rearing area? If free embryos imprint to their natal site, spawning adults should show fidelity to their natal reach in at least two ways. First, they should search for and show a behavioral preference for natal water. Second, their olfactory system should show enhanced sensitivity to natal water.

To test this hypothesis with Kootenai sturgeon is not time-efficient, but the most time-consuming work to test these hypotheses for shortnose sturgeon has already been done by B. Kynard and E. Parker, who 11 years ago (1998) assembled several live boxes containing free embryos from the eggs of one female. Some live boxes were placed at a known spawning site with rocky bottom in the Connecticut River and others were placed in similar habitat in a nearby tributary (Deerfield R.) where sturgeon do not spawn. Boxes were removed after fish developed into larvae, taken to the Conte AFRC, and fish from both groups were reared on city (well water). The two groups were kept separate for 6 years, then PIT-tagged, and combined into a common tank supplied with well water in 2006. Dr. Li, of Michigan State University, an expert in olfactory studies on fish (Siefkes and Li 2004), lead this part of the research. Andrew Wildbill, Dr. Li's student technician, helped us design and construct the systems needed to conduct tests with sturgeons at Conte AFRC.

Objectives for 2008:

1. Determine the effect of fish density on foraging and wintering habitat selection of year-0 juveniles. (Continues research begun in 2007 to determine if we can test groups instead of single fish.)
2. Determine the effect of four rock-size compositions on (1) downstream movement of free embryos and early larvae and, (2) the selection of rock size by free embryos and early larvae.
3. Determine the short-term movement speed and swimming height above the bottom of larval over rock compositions with 60% small vs. 60% large rocks.
4. Survey bottom and cover types in the meandering reach of the lower Kootenai River, the delta, and the littoral zone of Kootenay Lake.
5. Construct EOG system and conduct preliminary tests with olfactory sensitivity of shortnose sturgeon**.**

Methods by Objective

General rearing procedures for young sturgeons.—Fertilized eggs of Kootenai sturgeon were sent to us by the Tribal hatchery (see Acknowledgements) and reared in a McDonald hatching jar. We transferred about 2,000 hatchling embryos (day-0 fish) into an outdoor stream tank for rearing all summer and fall. We moved fish into indoor 1.5 m diameter tanks for rearing during wintering studies.

We used temperature-controlled dechlorinated city water (Montague, MA) for rearing and experiments during summer–fall, and then switched to Connecticut River water during winter rearing and experiments. Temperatures during rearing and experiments were the same (±1°C).

We maintained the natural photoperiod for the Turners Falls, MA, latitude (42.6°N).

We used the number of days post-hatching to characterize age of fish, not the number of days post-fertilization, because we did not know how early egg-rearing (particularly water temperature) varied during shipping before we received eggs.

Objective 1. Determine the effect of fish density on habitat preference during foraging and wintering.— During December 2007 to February 2008 (wintering) and during October 2008 (foraging), we determined the effect of fish density of year-0 juveniles on habitat preference. These studies will guide later habitat preference studies (i.e., the need to test single fish vs. small groups of fish).

Tests used a rectangular artificial stream tank that created the same velocity across the entire arena (Kynard et. al 2007). The tank was divided equally lengthwise into two small test tanks. Tests gave fish a choice of a 50:50 area of two substrates: sand vs. small rubble.

We tested a single fish during 15 replicates, observing each fish overnight from afternoon to mid-day. Afterward, we similarly tested groups of 8 year-0 juveniles during four replicates (N = 32 fish).

Fish were monitored by two video cameras and IR lights for 10 min/h from about 1400 h on day 1 to about 1200 h on day 2 to determine the time spent (min) on each substrate type. We used the mean time fish spent on sand to compare differences among single fish, among fish groups, and among single and fish groups using a paired t-test.

Objective 2. Determine the effect of four rock-size compositions on (1) downstream movement of free embryos and early larvae and (2) rock size selection by free embryos and early larvae.— We used eight identical circular 180-cm diameter artificial stream tanks (570 cm circumference; Fig. 1). Each tank had a circular channel 35 cm wide x 330 cm long with a mostly uniform width and velocity, although in the widest part velocity near the inside wall was slow. Water depth in tanks was 30 cm, except in the dispersal video camera viewing area, where a ramp reduced depth to 13.5 cm for a short distance.

Tanks had a mean bottom velocity of about 17 cm/s in the channel as determined in one tank of each substrate composition by measuring the velocity along 19 transects, three stations per transect, 2.5 cm above the bottom as in Kynard et al. (2007).

The water system for the artificial stream tanks and the rearing stream for other fish was a constant circulating system. Water constantly drained from a head tank, which regulated water flow through the system, to each tank and overflowed into the common drain, where the water was pumped through a chiller-heat pump back to the head tank. The chiller-heat pump within the water system maintained the same water temperature in all tanks. Individual pumps at each of the tanks provided the water velocity regime by pumping water through the flow outlet in the ramp. Each tank was covered with a fine-mesh netting to exclude insects and aerial debris from falling into the water. The entire system was outside on a platform and protected from weather by a tent cover.

We tested four rock-size compositions using eight stream tanks (two replicates per composition). We tested combinations of three rock sizes represented by A, B, and C: A (small) = 3–6 cm diameter pebble, B (medium) = 7–12 cm diameter small rubble, and C (large) = 13–20 cm diameter medium-large rubble. Rock-size classification was from the modified Wentworth classification system. There were four rock compositions (I–IV) according to the percent of each of the three rock-sizes: composition I (A = 60%, B = 30%, C = 10%); composition II (A = 10%, B = 30%, C = 60%); composition III (A = 30%, B = 50%, C = 20%); and composition IV (A = 50%, B = 10%, C = 40%). Percent of rocks in each composition was determined by volume using a 19 L bucket, and then, rocks of each size class for that replicate were counted and duplicated for the second replicate. Position of rock patches (about one bucket of rocks per patch) are shown in Fig. 1; a bare tank bottom was between rock patches.

To view downstream movement of fish at the ramp area of each tank (number of fish passes/4 min viewing period per hour), we placed a color video camera with two 60-watt yellow lights (to see fish at night) over the viewing area in each tank. Also to assist seeing small fish at night, the walls in the viewing area had a silver reflective tape covering and the ramp was painted white.

We placed 15 hatchling free embryos in each tank, and after 1 h acclimation, began to monitor downstream movement and rock use. Data collection continued until a few days after fish completed development into dispersing larvae (day 16). We used a video camera (Fig. 1) to record downstream movement for 4 min each hour for 24 h/d during the 16 d.

We reviewed movement tapes and determined the net number of downstream fish passes (mean number of downstream fish passes – mean number of upstream fish passes = net mean number of downstream fish passes) for every 2 h of each day. We used a repeated measures ANOVA to compare the mean number of daily fish passes within and between tank treatments (replicate tanks combined if they were not different). A daily time series of the net mean number of downstream fish passes showed the pattern of downstream movement of free embryos and early larvae in the different rock combinations.

In addition, once daily during the daytime on days 0−16, we moved rocks and visually observed fish in all tanks for their use of small, medium, or large rocks. (We only observed fish once per day because of the time and labor involved and to minimize the disturbance to fish.) During each observation period, we recorded the number (and later calculated the percent of total fish) in each of the following categories: moving downstream, in the open, under rocks of each size, and contacting rocks of each size. We compared replicates for each rock composition, the test compositions for the percent of fish moving downstream, and the percent of fish using small, medium, and large rocks in each rock composition using repeated measures ANOVA.

Objective 3. Determine the short-term movement speed and vertical swimming height of larvae over a substrate dominated by small vs. large rocks.—After observations on free embryos and early larvae were finished in Obj. 2, we observed behavior and dispersal speed of days 17−40 larvae in the same tanks. General methods follow those of Kynard et al. (2002). Observations were done in four streams: two tanks with the greatest percent (60%) of small rocks and two tanks with the greatest percent (60%) of large rocks.

Each day we randomly captured 20 larvae from the rearing tank and placed five fish into each the four test tanks. After 1 h acclimation, we observed individual larvae in each tank for the time to move one-half loop (one-half circumference = 285 cm). We could not observe fish for a full loop because of physical constraints. After observing a fish pass the downstream limit (leading edge of ramp), we captured it, and observed another fish. This procedure continued until all five fish were observed in each tank. Each day we alternated which group of tanks was observed first. In addition, we observed downstream movement on five nights using the same methods and number of fish per tank ($N = 20$/observation day). In all these tests the probability that a fish tested on one day could be recaptured and retested later was small due to the large number of potential test fish (about 1,000).

We collected data on the vertical distribution of fish (1 − in the water column, 2 − at the surface, or 3− on the bottom) and their location relative to flow (1− in fast flow = near the outside wall, 2− in slow flow = near the inside wall, or 3− in moderate flow = between the two walls). The time for a fish to swim one-half loop was converted to the rate of movement (m/s) or ground speed. We also measured water velocity in the fast flow used by most fish and calculated the mean speed of fish relative to water speed. The velocity regime in the fast flow along the wall of all tanks was similar (mean = 17.0 cm/s).

We used a time series of movement rate to show if the rate changed with fish age. We compared the mean speed of fish in small vs. large substrate tanks using paired *t*-tests. We used ANOVA to compare the mean speed of fish at night during early, mid, and late dispersal periods.

Objective 4. Characterize substrate and cover types in the meandering reach of the lower Kootenai River, the delta, and the littoral zone of Kootenay Lake.— We used an underwater video system (Kynard et al. 2007) to conduct surveys during winter (22 January 2008) and summer (1-4 August 2008). Table 1 shows the GPS locations of sampling stations.

Sampling was done by suspending the video camera from a boat and continuously recording the view of the bottom. The camera, which was equipped with two fixed lasers for determining size of bottom materials, was suspended about 1−1.5 m above the bottom to record an area of about 0.6 m^2. To collect a video sample, we pulled the boat (delta and lake samples) or drifted the boat (river samples) along an anchored line for a fixed distance. This controlled survey method allowed the camera to slowly view bottom habitats. At most river locations, we sampled three sites: each bank and the channel. At curves in the river, we sampled three sites: inside, outside, and center of curve. In the river delta during the winter, we sampled five stations: West shallow, Center deep, East shallow and East deep (two sites), and during the summer, we sampled five stations, some similar to the winter stations: Center shallow, Center mid-depth, Center deep, East shallow, and East deep. In the delta, deep is 27.4−15.2 m, mid-depth is 15.1−7.6, and shallow is 1.8−0.9 m.

We reviewed transect tapes and sub-sampled the video frames recording point samples at 5 sec intervals for the total duration of each transect. The percent frequency of each substrate type or sub-type in each transect and for all transects was determined for river macro-habitat (channel or riverbank in straight sections, inside, outside, or center of curve, and Shorty's Island sites) and for the lake-delta transects (# of samples of each habitat type ÷ the total number of samples in the transect). During this initial survey, we calculated the percent of each substrate and cover type that was present in the lake-delta, river channel, and river shoreline, and ranked the substrate and cover types for relative abundance.

In January 2008, we observed substrate and cover type in river and lake stations (Figs. 2, 3). We classified bottom substrate as sand, sand dune, mud, and mud dune. We classified bottom cover as small wood (< 2.4 cm diameter and > 2.5 cm long), medium wood (2.5−5.0 cm diameter and > 2.5 cm long), large wood (5.1−15.0 cm diameter and > 2.5 cm long), log (> 15.0 cm diameter and > 2.5 cm long), or detritus (wood pieces and organic debris < 25 mm long).

In August 2008, we sampled sites in the lower river, including Shorty's Island, and sites in Kootenay Lake (Figs. 2, 3). In the river, we sampled curves (inside bank, center channel, and outside bank) and straight reaches (bank and channel). Locations sampled at Shorty's Island follow: downstream end of island, channel near the rockpile, bank upstream from the rockpile, and the rockpile). In Kootenay Lake, we sampled several sites in the littoral zone: shallow, mid-depth, and deep (characterized previously). Substrates were classified as sand (level with bottom), sand dunes, mud, clay (smooth clay level with bottom), clay terrace (clay with stepped vertical levels), clay outcropping (a raised hump of clay), clay dunes (a series of clay outcroppings), and rock (all sizes). We classified bottom cover using the following categories: sparse vegetation (can see bottom through vegetation), dense vegetation (bottom not visible through vegetation), wood (small− ≤ 2.4 cm diameter and > 2.5 cm long , medium− 2.5 to 5.0 cm diameter and > 2.5 cm long , large− 5.1 to 15.0 cm diameter and > 2.5 cm long), log (> 15.0 cm diameter and > 2.5 cm long), detritus (wood and leaf pieces, and other organic debris < 25 mm long), leaves (number of whole or almost whole leaves), rock (small− ≤ 2.4 cm diameter , medium− 2.5 to 5.0 cm diameter, large− 5.1 to 15.0 cm diameter), or clay bits (small bits of clay from clay terraces or outcroppings).

Objective 5. Construct EOG system and conduct preliminary tests with shortnose sturgeon on olfactory sensitivity.—During 2 weeks in May 2008, we obtained equipment and constructed the various systems needed to create an electrical-free field in which to hold test fish, a water supply system for testing fish, and a water supply system for holding odorants. Dr. Li supplied the EOG equipment and Andrew Wildbill (Dr. Li's student) assisted us with building the systems and with preliminary tests.

Results & Discussion

I. **Density Effect on Habitat Selection.**—Tests found no effect of fish density on habitat preference of year-0 juveniles. For year-0 foraging juveniles tested in October 2008, there was no difference (*P* = 0.08) in the percent (52.1%) of single juveniles vs. percent (57.8%) of eight fish selecting sand vs. rock at night. Also, during tests with wintering fish in February 2008, there was also no difference between single year-0 juveniles (61.8%) vs. groups of eight year-0 juveniles (57.1%) for the percent selecting sand vs. rock in the day (*P* = 0.17). We conclude we can use eight fish in our tanks for habitat preference tests and do not have to test fish singly.

II. Effect of substrate composition on downstream movement and substrate selection of free embryos and early larvae.—Video observations on the scaled number of fish passes show almost zero downstream movement by days 0−10 free embryos in any substrate combination and only a few free embryos in one replicate (30% small rock) moved downstream on day 11 (Fig. 4).

Visual daily observations on free embryos in all tanks found ≤ 10% of the days 0−11 fish were moving downstream in any of the substrate combinations and only seven fish out of a total of 120 (all replicates all compositions) were in open habitat away from rocks (Fig. 5). Thus, all observations on fish passes indicated any of the combinations of small, medium, and large diameter substrates provided adequate cover for the 15 free embryos. Visual observations also found most free embryos did not use open habitat, but were under rocks, not just in contact with rocks (Fig. 5).

During daily visual observations on the size of rocks free embryos selected, each rock was carefully lifted or moved to determine the size of rocks fish selected. This proved difficult and involved two people, but the data on percent of fish under small, medium, and large rocks show a clear trend for free embryos to select small substrate more than medium or large substrates (Fig. 6). This result is consistent with observations on Frasier River white sturgeon by Steve McAdam (personal communication). Also, when the percent of small substrate in our tests was the greatest (50 or 60%), the percent of free embryos using small substrate was also the highest 60−80% (Fig. 7). There was a significant relationship between the percent of free embryos under small rocks and the percent of small rocks, i.e., the 60% small rock composition had significantly more free embryos under small rocks than the 30% small or 10% small rock compositions, but 60% small and 50% small were not significantly different from each other for number under small rocks (repeated measures ANOVA, $P<0.05$). These data suggest that when creating habitat for spawning and egg-free embryo rearing, the rock composition should contain between 30−50% small pebbles, with the remaining rock size being various sizes of rubble.

Free embryos did not just contact rocks, they swam under them (Fig. 7). Less than 1% of the free embryos (all observations, all rock compositions) were only contacting rocks.

As free embryos developed into larvae on days 12−13, video observations found a few left cover and moved downstream in all substrate types, with the fewest moving in the 60% large substrate composition (Fig. 4). The downstream movements of larvae in the four rock compositions were more alike than they were different ($P > 0.05$ except for the replicates of two rock compositions, which ANOVA found were significantly different. The replicates that were different both had greater downstream movement than the other replicate of the pair. Because these two replicates were so different from all the other replicates of all rock compositions, which were not different, we classified the two different replicates as anomalies. With increasing age of larvae there was an increasing number of fish passes (Fig. 4), and visual observations established that this indicated an increasing number of fish moving downstream, not just a few fish moving more rapidly around the tank with increasing age (Fig. 5).

Visual observations on larvae moving downstream found a similar percent of days 14−16 fish were moving downstream in all rock compositions (*P* > 0.05; Figs. 5, 8). Thus, rock composition had no significant effect on the number of larvae moving downstream among rock compositions. This supports our interpretation on the anomalous behavior of larvae in the two replicates in Fig. 4.

There was no significant trend for fewer day 14 and older larvae to move downstream in the tanks with the greatest percent of large substrate (Figs. 5, 8). These data do not support the 2007 test results of Kynard et al. (2008), where fewer downstream fish passes were observed in tanks with 100% large-rubble patches. However, the percent of rubble in the present patches was only 60% (maximum), whereas, in the 2007 tests, the rubble patch was 100% rubble, so the percent of large rubble could still affect larval movement.

Early larvae daily decreased using rocks (Fig. 5). Those few larvae that continued to use rocks used mostly small rocks in all rock compositions, and the only significant difference for fish use among the rock compositions for use of rock size was the number of fish under medium rocks in the 30% small:50% medium:20% large and the rock composition with 60% small (*P* = 0.048; Figs. 6, 8).

III. Downstream movement behavior and speed.—Observations on days 17−40 larvae moving downstream show most were at the surface (57−77%), in the water column (7−13%), or holding position on or near the bottom (5−13%; Fig. 9). There was no difference between the two replicates with 60% small rock (*P* = 0.11), but there was a difference between the two replicates with 60% large (*P* = 0.003), so we show both replicates in Fig. 9. For these tests, where fish were only in the tanks for the observation period and then removed, we did not observe a difference between the two 60% small rock composition tanks like we found in Fig. 4.

Larvae moving downstream were in the fastest flow along the outside tank wall, but did not contact the wall (Fig. 10). Almost zero larvae moving downstream were in the slower flow near the inside wall. This suggests migrants select the fastest velocity and that moving quickly downstream has a high survival value.

Observations on daytime short-term speed found days 17−40 larvae generally moved downstream at similar speeds whether over 60% large or 60% small rocks and speed did not change greatly with fish age (Fig. 11). However, there was a significant trend for mean fish speed to be slower in the tank with 60% small rocks (*P* = 0.03), an unexpected result.

Most fish were moving at or slightly less than the water speed. Fish moved at a mean of 85% of the current speed (fish speed range = 50−110% of water speed).

Speed of fish observed at night was similar to the speed observed in the day (Fig. 12). Speed at night was not different between replicate tanks of each rock composition and mean speed was not different between the small and large rock compositions (*P* = 0.25).

Tests indicated that a dominance of large or small rocks (60% large vs. 60% small rocks) significantly affected the short-term movement speed of larvae in the daytime, but not at night. Fish moved slightly slower in the day in the tank with a dominance of small rocks. It is difficult to interpret these results; perhaps, tests in 2009, which are designed to determine fish speed for longer duration may provide an explanation.

Previous long-term observations on larvae found fish moving downstream in stream tanks with large rock tended to remain longer when they stopped, resulting in fewer downstream fish passes per unit of time (Kynard et al. 2008). However, these observations were made when 100% of the area (same area as in the present tests) was large rubble, not the 60% as in the present tests. Thus, perhaps there is a rock composition effect. Because migrant larvae are above the bottom, the effect of rock size on long-term dispersal speed would not likely be to reduce the speed of fish during a downstream movement bout, but over a long distance, to affect the time fish spend on the bottom, with some size rocks providing a better velocity shelter for drift foraging, so fish remain longer.

IV. Bottom Habitats in the Lower Kootenai River, the River Delta, and the Eastern Littoral Zone of Kootenay Lake

A. January 2008: Kootenai River (Fig. 2).—In river samples, sand dominated the outside of curves, sand dunes dominated the channel, and sand−sand dunes shared dominance in the inside of curves (Fig. 13-A). Thus, sand was the dominant substrate in the sample reaches.

Of the bottom cover observed in the river samples, small and medium pieces of wood were the dominant cover types (Fig. 13-B).

B. January 2008: River Delta & Kootenay Lake (Fig. 3).—The substrate type at the western station was the most diverse of any station, with mostly sand dunes, but small proportions of mud or sand (Fig. 14-A). Sand dominated the Center deep station and mud dominated the Center-East shallow sample. The two East deep samples were both 100% sand.

In the West shallow station and Center-deep, and one East-shallow station bottom cover type was dominated by sparse vegetation (Fig. 14-B). Detritus dominated the other East shallow or deep station. Detritus accumulated in bottom depressions.

In the delta and lake, there was abundant sand. The detritus cover contained mostly dead vegetation, but some leaves and small woody debris were present. Large woody debris and logs were scarce in the delta and lake samples, suggesting that most fallen trees are reduced to small woody debris in the river or are deposited deep in the lake during high flows.

C. August 2008: Kootenai River (Fig. 2).—Sand dunes and sand were the dominate substrates in the channel of both straight and curved river reaches (Fig. 15-A), but clay and rock were present in the straight channel. Mud dominated the inside of curves and clay terraces dominated the outside of curves. Substrate types on the straight river bank were the most diverse (six types) with mud and clay terraces the dominant types.

The dominant cover types in curve channels were detritus and small rock (Fig. 15-B). On the inside of curves, detritus, sparse vegetation, and small rocks were the dominant cover. On the outside of curves, sparse vegetation and small rocks were the dominant cover. In the straight reach, detritus was the dominant cover in the channel and sparse vegetation was the dominant cover along banks. There was a diversity of cover types in curve and straight reaches.

At Shorty's Island, mud was the dominate substrate at the downstream end of the island, sand dunes dominated the channel, mud dominated the outside bank, and rocks dominated the rockpile (Fig. 16-A).

Abundant dense vegetation provided cover on the downstream end of Shorty's Island, the channel had a small amount of cover but it was diverse, the outside bank had abundant dense vegetation, and the rockpile provided rock cover and dense vegetation (Fig. 16-B). It was interesting that vegetation has quickly emerged in the rockpile, likely because rocks were placed over existing vegetation, which is very abundant along the river bank where velocity is slow.

D. **August 2008: River Delta and Kootenay Lake (Fig. 3).—**At all Center stations (shallow, mid-depth, or deep), sand was the dominant substrate type (Fig. 17-A). The shallow East station had only mud substrate. Thus, sampling indicated sand was the dominant substrate in the delta.

Cover type at the Center deep station was evenly split between detritus and clay outcropping (Fig. 17-B). Detritus was the only cover at the Center mid-depth station and dense vegetation was the only cover at the Center shallow station. At the deep East station, detritus was the dominant cover type, and at the shallow East station, the dominant cover was dense vegetation and detritus.

River samples during winter contained few cover types compared to summer (Figs. 13-B and 17-B). At the winter stations, wood and detritus were most abundant, and in the summer stations, small rocks-sparse vegetation-detritus-clay bits were most dominant. Also, abundance of cover seemed reduced in winter compared to summer. If correct, this difference could be due to sampling different stations each season, but some decrease may be due to seasonal changes in cover type or availability, or both.

However, cover types in the delta and lake were mostly sparse vegetation and detritus in the summer and in winter (Figs. 14-B and 17-B). So, cover types did not drastically change seasonally in the delta and lake.

Although no effort was made to quantify abundance of cover at any station, the available cover types in the delta and lake seemed abundant in winter. How this situation affects evolution of the choice of a wintering refuge is not known, but year-0 juveniles going into winter are only an average of about 6 cm TL. It is likely important for survival of fish this small to seek preferred habitat (perhaps, with cover) to survive months of predation by fish. Little is known about wintering habitat preference of year-0 juveniles (Kynard et al. 2007). Because the lower river and the delta−lake areas differ for habitat and cover, information on cover and habitat preference of year-0 juveniles could identify which area has the preferred habitat and cover.

V. **Imprinting.**—We spent the entire 2 weeks building the systems and attempting to create a test area in the main Conte building that was free of low electrical fields. On the last day, we moved the test system into another building outside the Conte Lab and obtained the electrical-free area. We tested a juvenile shortnose sturgeon and verified the new location and systems would enable us to conduct EOG tests with sturgeon olfaction in the future.

Acknowledgments

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Table 1. Sampling sites in the river and lake during winter and summer 2008. Site numbers correspond to those on Fig. 2 and 3. Winter site numbers start with W; summer site numbers start with S

Site	Description	GPS location
WR1	River - outside of curve	49°11'21.36" N, 116° 36'45.84" W
WR2	River - inside of curve	49°11'18.48" N, 116° 36'48.36" W
WR3	River - channel in curve	49°11'03.66" N, 116° 38'14.76" W
WL1	Lake - west - shallow	49°15'46.32" N, 116° 41'30.42" W
WL2	Lake - center - mid-depth	49°15'59.34" N, 116° 41'34.86" W
WL3	Lake - east - shallow	49°16'18.84" N, 116° 41'01.26" W
WL4	Lake - east - deep	49°16'30.60" N, 116° 41'04.08" W
WL5	Lake - east - shallow	49°16'31.44" N, 116° 40'00.48" W
SR1	River - straight reach	48°42'32.64" N, 116°23'07.56" W
SR2	Shorty's Island	48°46'25.98" N, 116°23'17.70" W
SR3	River - straight reach	48°47'38.46" N, 116°22'56.82" W
SR4	River - curve	48°53'32.88" N, 116°25'48.72" W
SR5	River - straight reach	48°53'41.40" N, 116°25'27.78" W
SR6	River - curve	48°52'38.64" N, 116°24'41.94" W
SR7	River - curve	49°10'28.38" N, 116°35'27.72" W
SR8	River - straight reach	49°06'43.02" N, 116°34'54.54" W
SR9	River - straight reach	49°14'31.68" N, 116°41'09.72" W
SR10	River - curve	49°11'38.94" N, 116°39'19.08" W
SL1	Lake - east - shallow	49°17'03.78" N, 116°39'54.66" W
SL2	Lake - east - shallow	49°17'04.68" N, 116°39'53.76" W
SL3	Lake - east - mid-depth	49°17'19.56" N, 116°40'09.84" W
SL4	Lake - east - deep	49°17'19.74" N, 116°40'11.70" W
SL5	Lake - center - deep	49°16'34.92" N, 116°40'46.32" W
SL6	Lake - center - mid-depth	49°16'30.48" N, 116°40'47.04" W
SL7	Lake - center - shallow	49°16'06.12" N, 116°40'42.48" W
SL8	Lake - center -shallow	49°15'57.42" N, 116°41'33.36" W

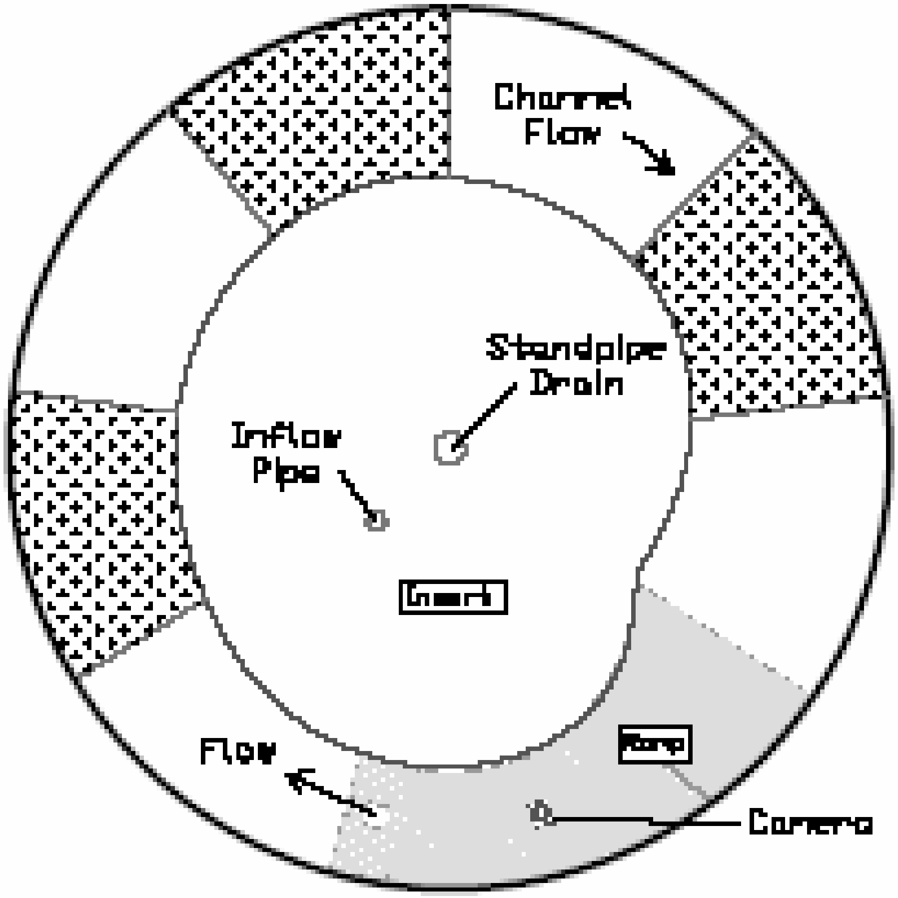

Figure 1. Plan view of the artificial stream tank used to observe downstream movement and habitat use. Dotted areas indicate the three patches of rocky substrate. The video camera monitored downstream movement of fish.

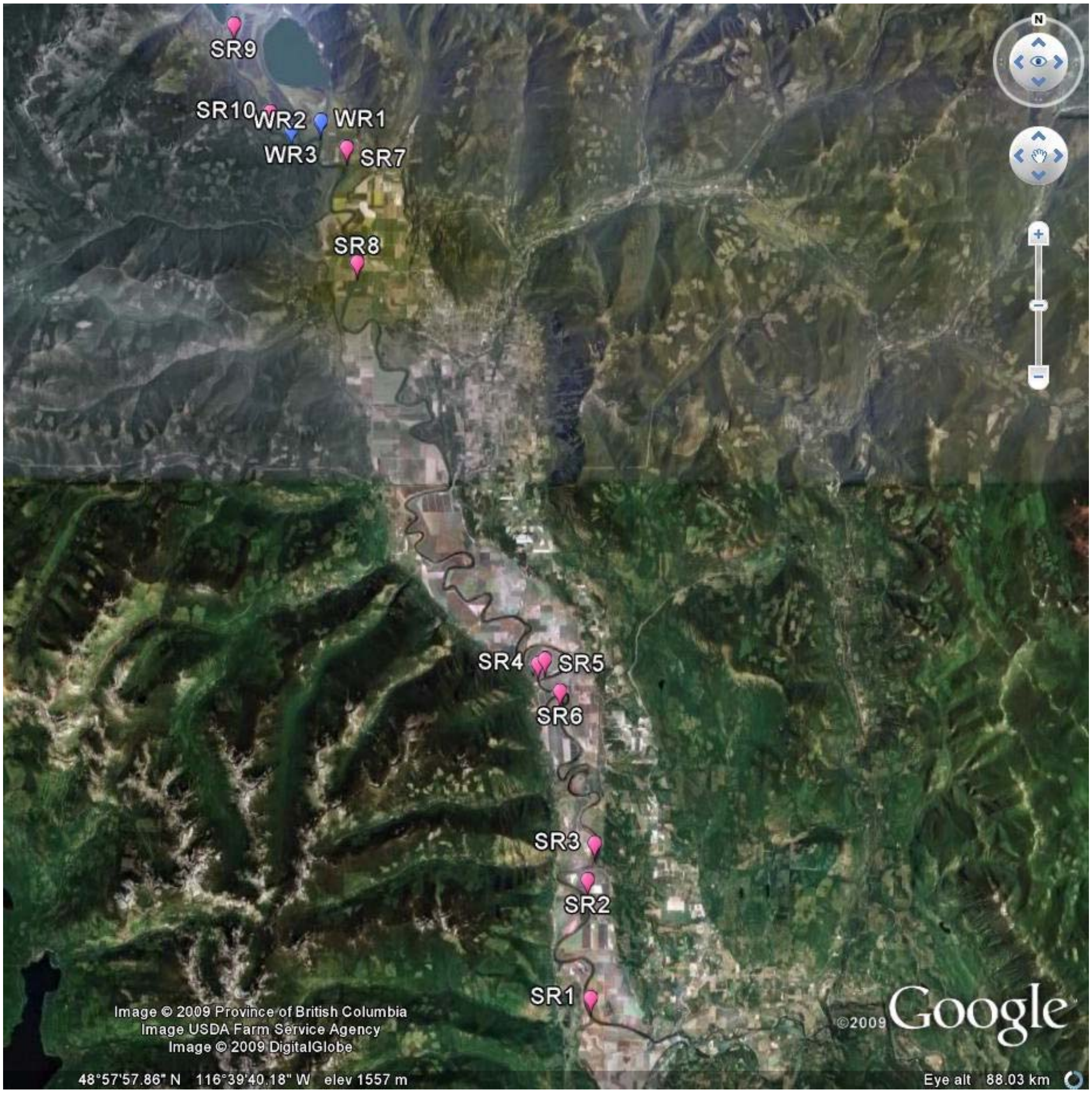

Figure 2. Locations of substrate and cover sampling sites in the Kootenai River. S=Summer, W=Winter, R=River site designations correspond to Table 1 GPS locations.

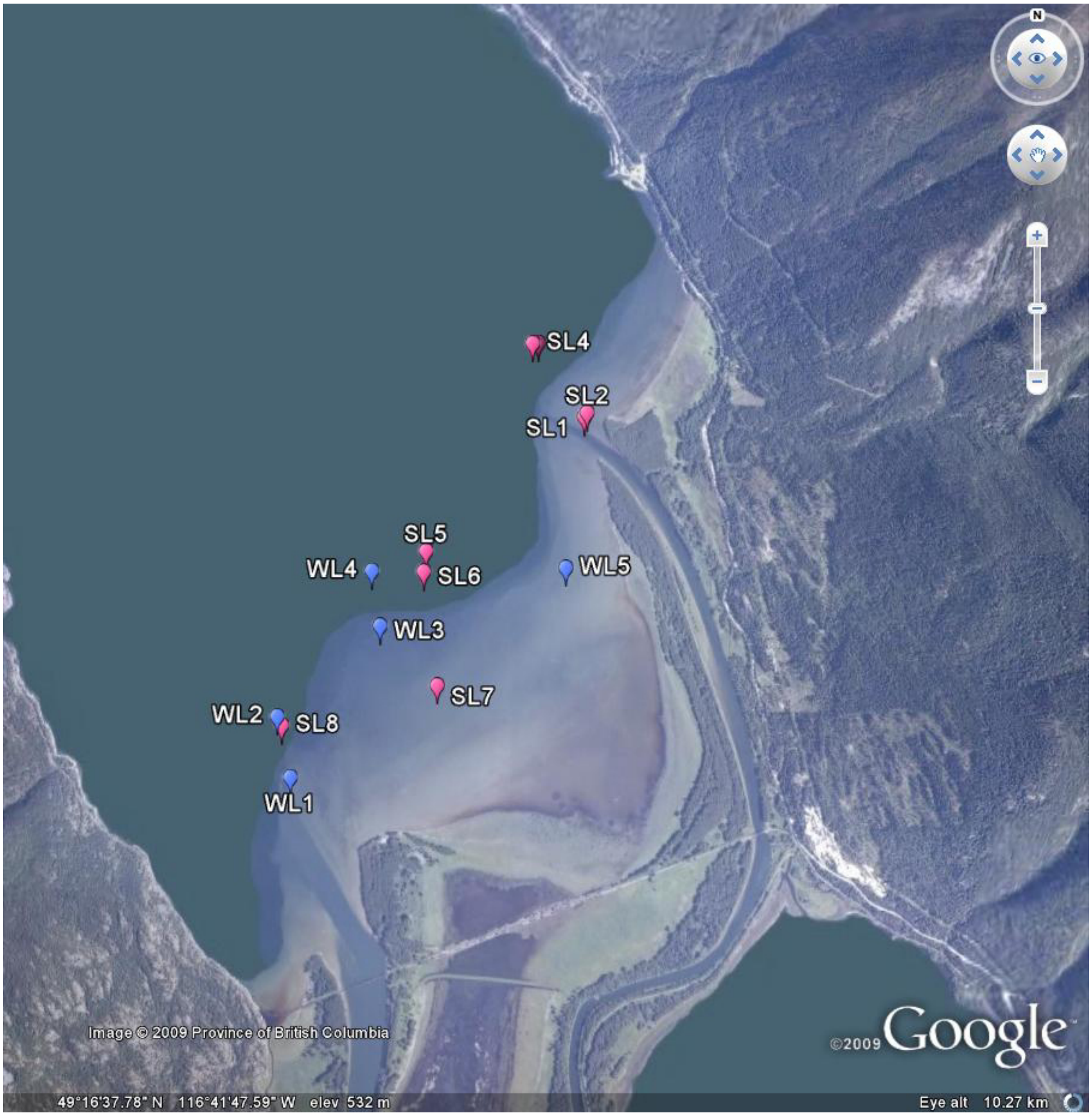

Figure 3. Sampling locations of substrate and cover types in Kootanay Lake. Key: S=Summer, W=Winter, L=Lake. Site locations correspond to Table 1 GPS locations. Note that SL3 and SL4 overlap to an extent that the label for SL3 does not appear.

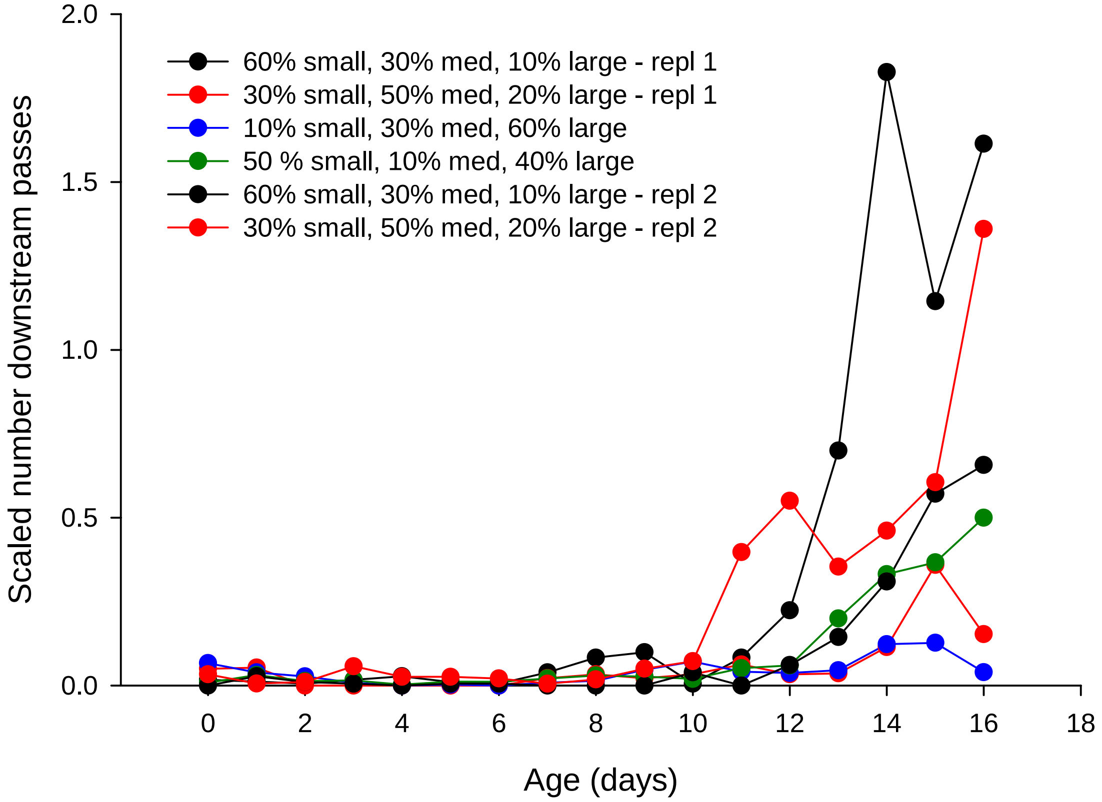

Figure 4. Daily downstream movement of Kootenai white sturgeon free embryos (days 0-11 fish) and early larvae in eight tanks (two replicates of four rock compositions) with each composition made up of small, medium, and large rocks. Diameter of rocks (cm) follows: small - 3 to 6 (large pebble), medium – 7 to 12 (small rubble), and large 13 to 20 cm (medium-large rubble). For each composition, the percent of each rock size is shown. The figure shows the two replicates that were significantly different from their replicate (and other compositions) during the larval interval. These two replicates were considered anomalies.

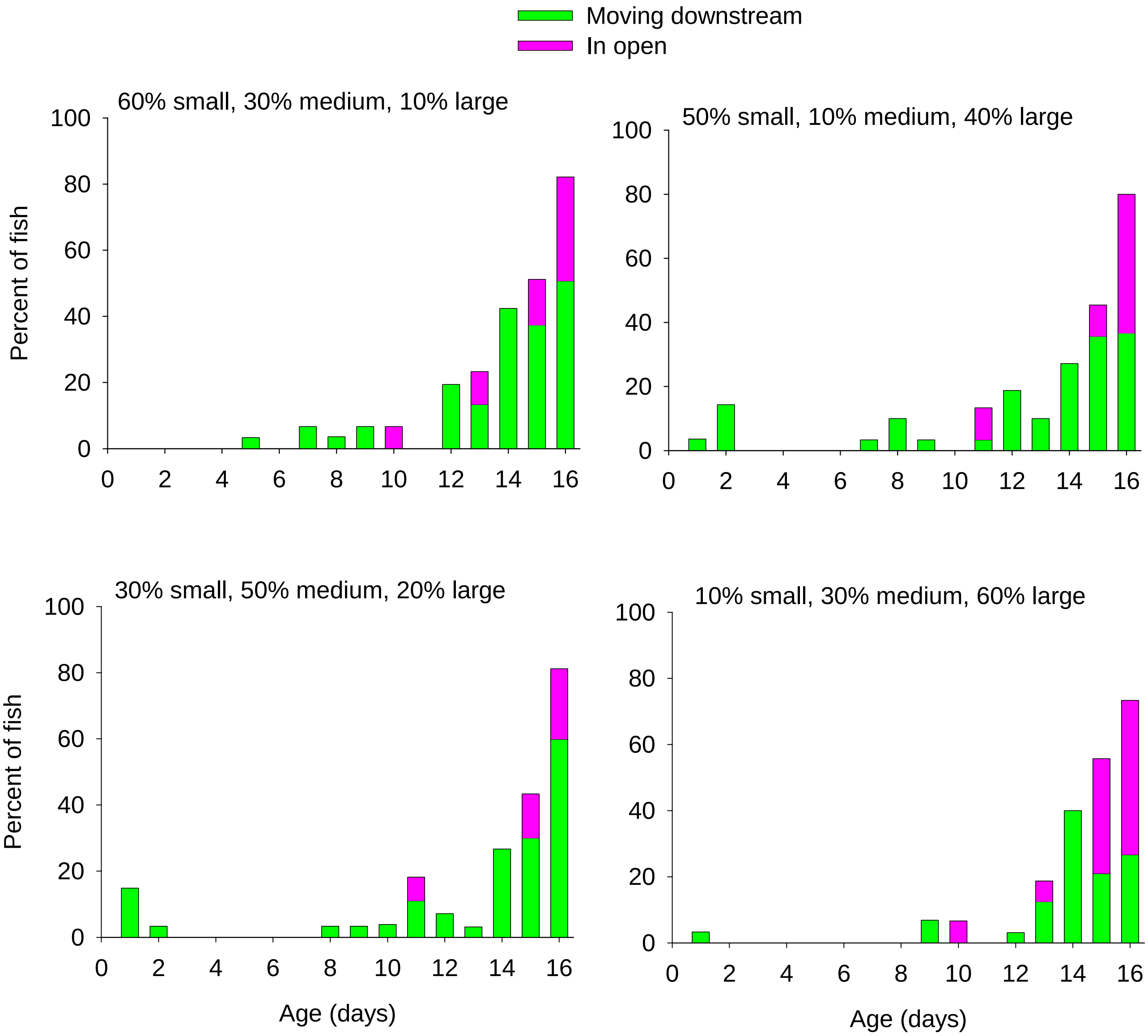

Figure 5. Mean daily percent of free embryos (days 0-11 fish) and early larvae moving downstream or in the open in artificial streams with four combinations of small pebble, small rubble, and medium-large rubble (two replicates for each rock combination). Fish ($N = 15$) were visually observed four times per day.

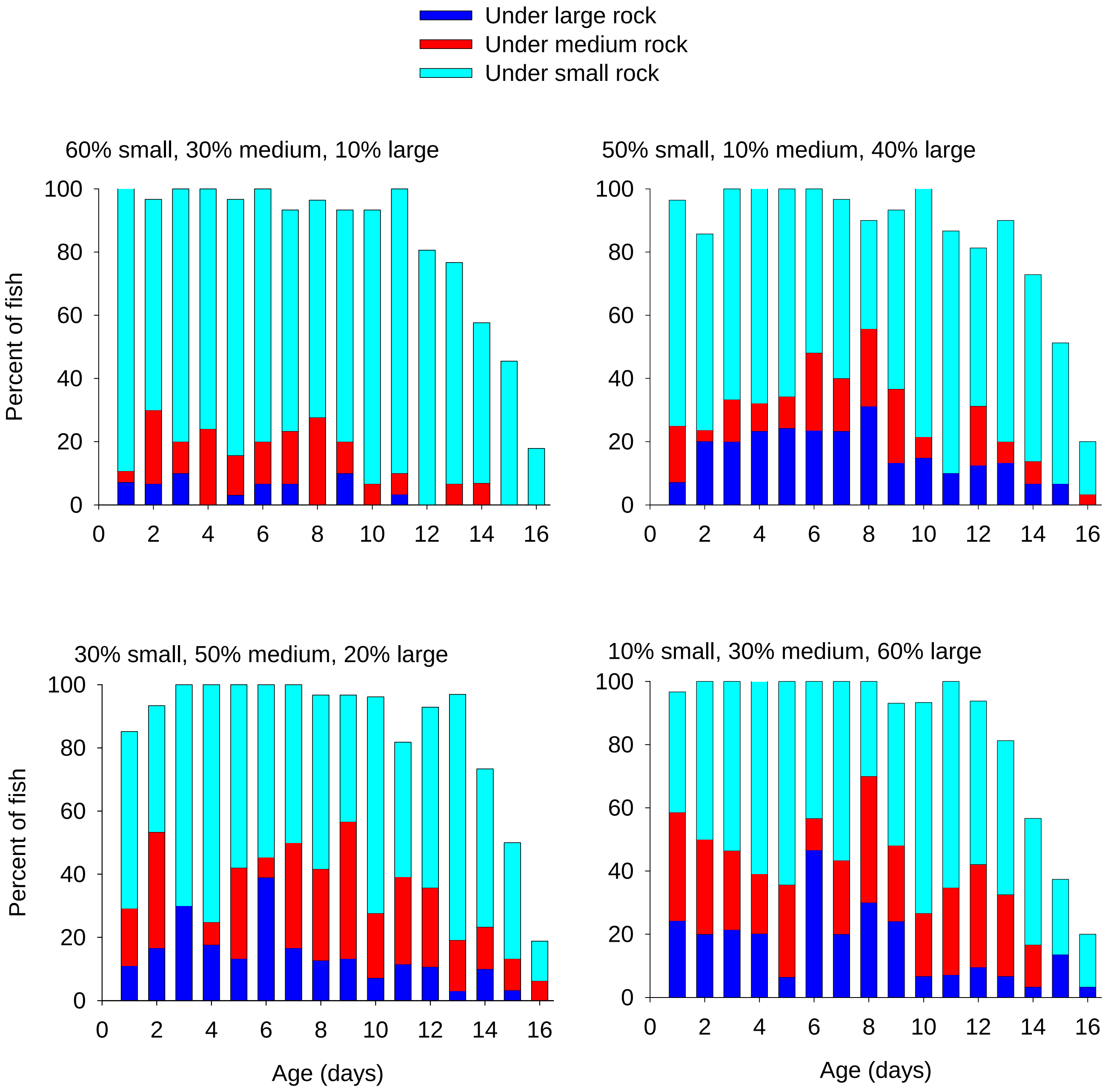

Figure 6. Mean daily percent of free embryos (days 0-11 fish) and early larvae hiding under rocks in four combinations of small pebble, small rubble, and medium-large rubble (two replicates for each rock combination). Fish (*N* = 15) were visually observed four times per day.

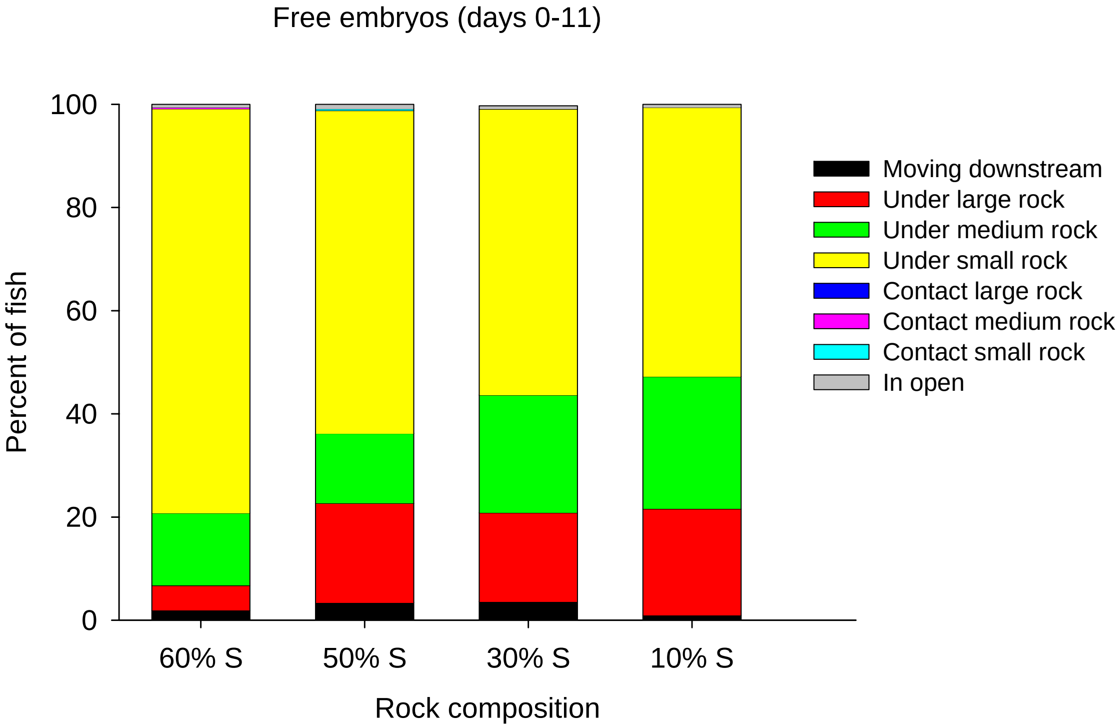

Figure 7. Mean percent of free embryos under rocks, contacting rocks, in the open, and moving downstream in four rock combinations with decreasing percent of small rock (pebbles).

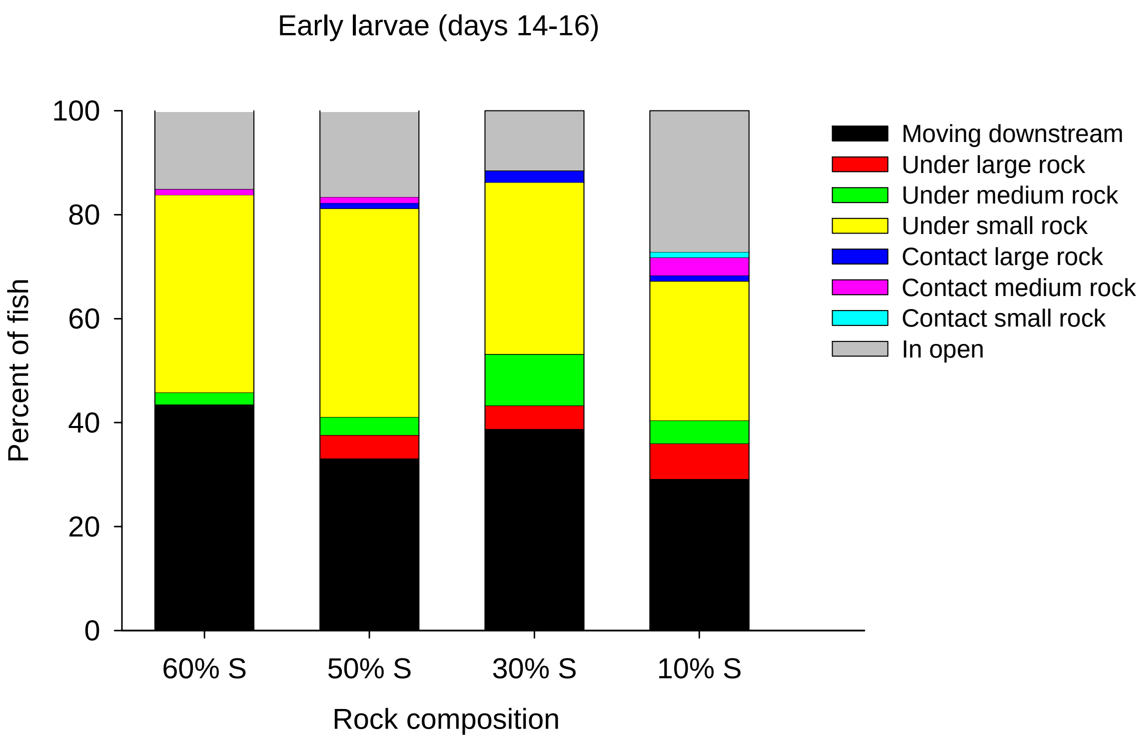

Figure 8. Mean percent of early larvae under rocks, contacting rocks, in the open, and moving downstream in four rock combinations with decreasing percent of small rock (pebbles).

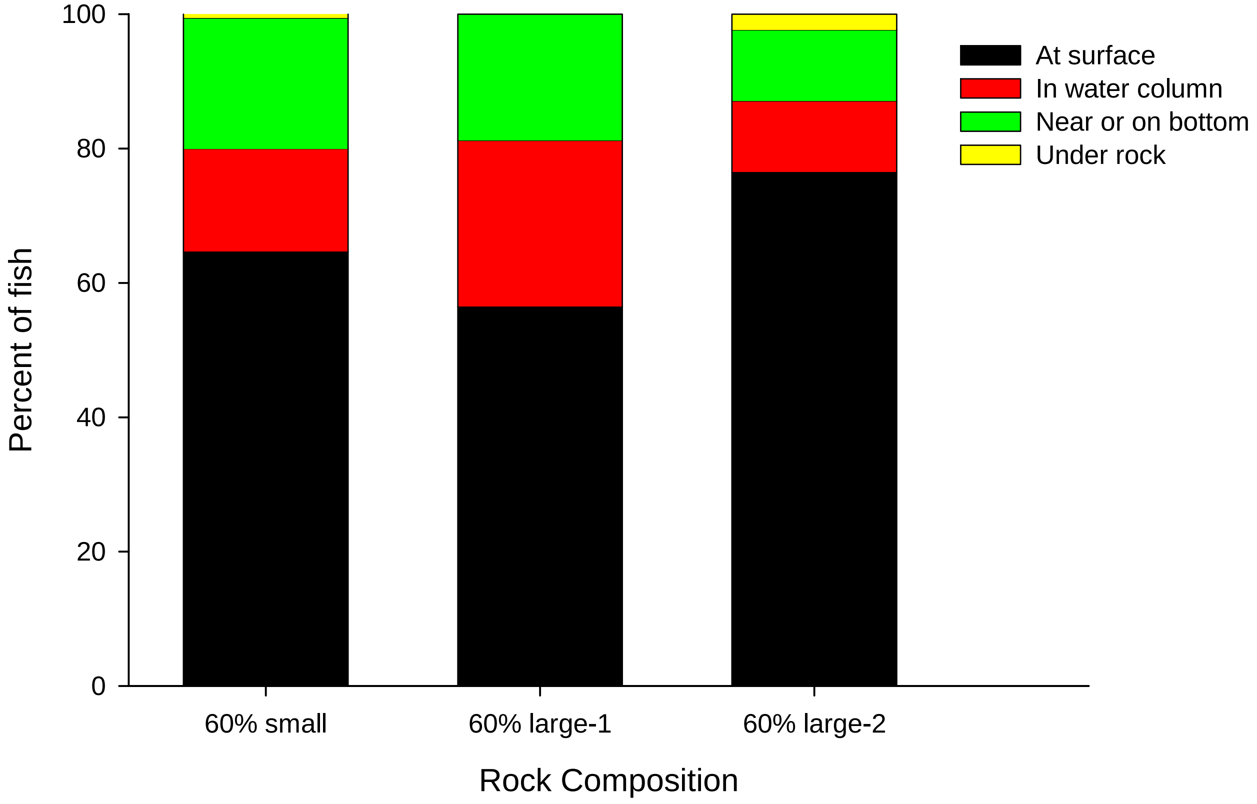

Figure 9. Mean percent of larvae at four locations in the water column when moving downstream in two artificial streams with 60% small pebbles and 10% medium-large rubble vs. two streams with 60% medium-large rubble and 10% small pebbles and two replicate tanks per rock composition. NS difference for percent of fish in the four locations between the two rock treatments.

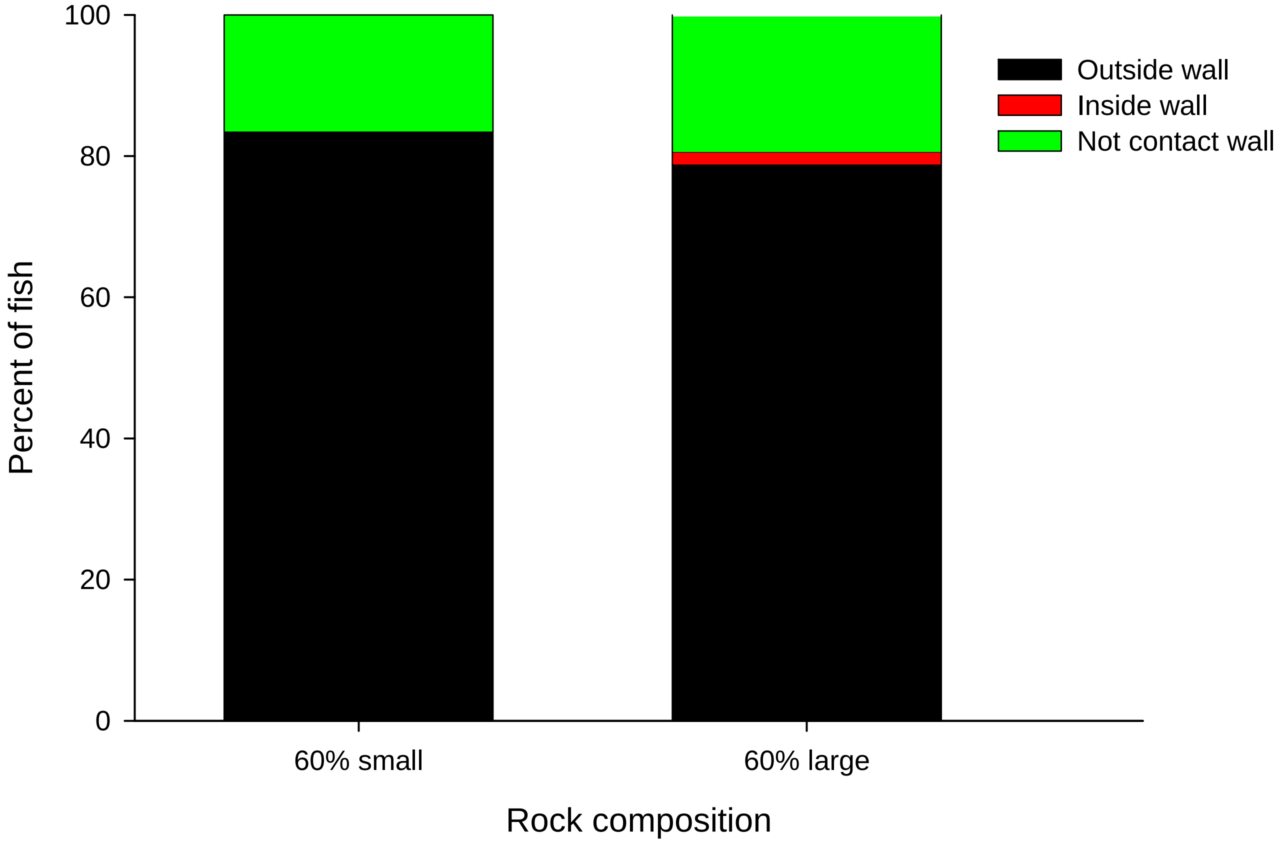

Figure 10. Mean percent of larvae at three wall locations when moving downstream in stream tanks with two rock compositions (60% small pebbles:10% medium-large rubble vs. 60% medium-large rubble:10% small pebbles) and two replicate tanks per rock composition. NS difference for the percent of fish in the three locations between the two rock compositions.

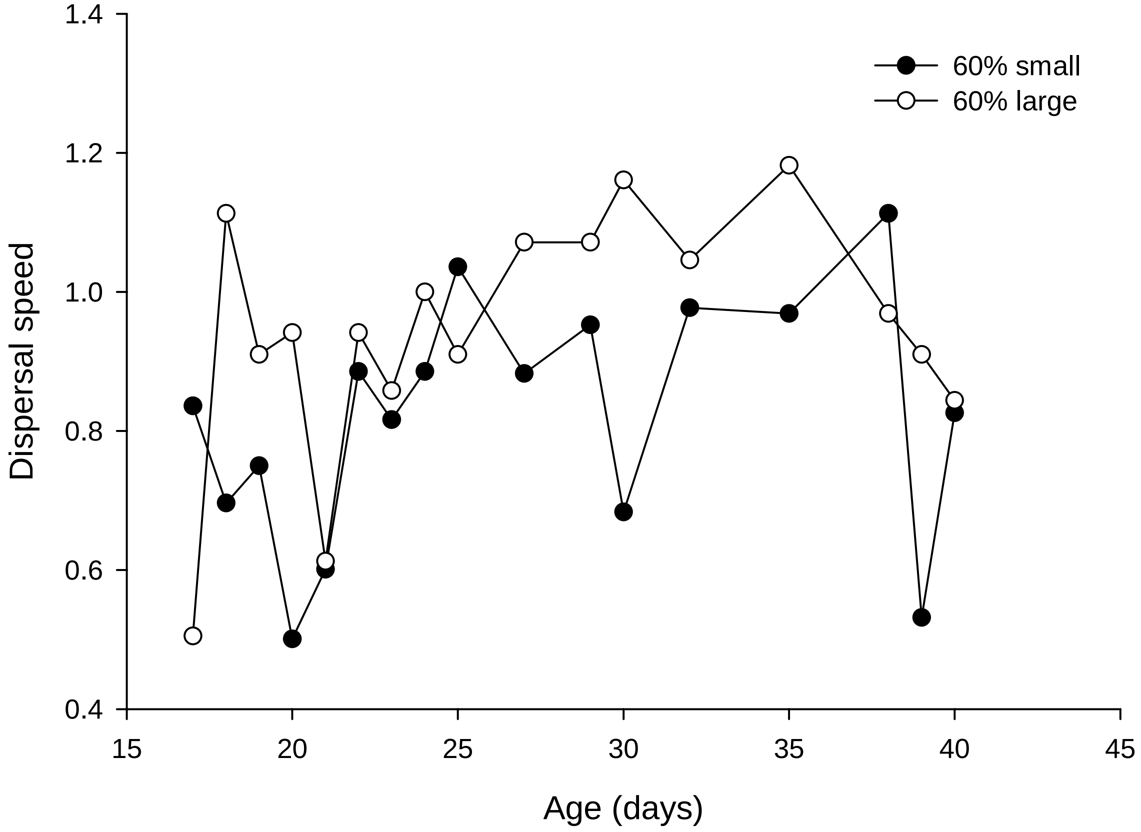

Figure 11. Mean dispersal speed of larvae by daily age observed in the daytime moving 285 cm (one-half loop of a stream tank) in two tanks with the highest percent (60%) of small pebbles vs. two tanks with the highest percent (60%) of medium-large rubble. Observations were done four times per day (five fish per trial) in the daytime. A dispersal speed of 1.0 = water speed; values > 1.0 = fish moving > water speed and values < 1.0 = fish moving < water speed.

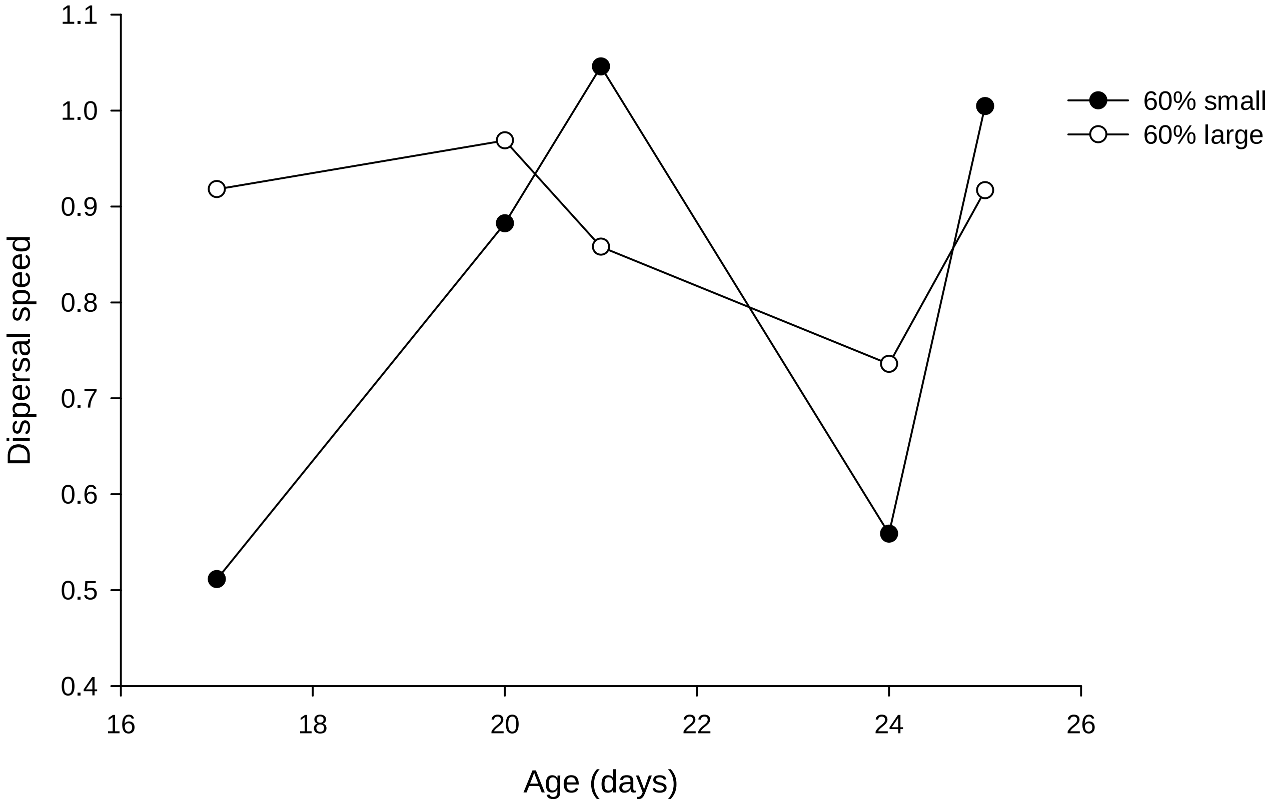

Figure 12. Mean dispersal speed of larvae by age observed moving at night 285 cm (one-half loop of a stream tank) in two tanks with the highest percent (60%) of small pebbles vs. two tanks with the highest percent (60%) of medium-large rubble. Observations were done four times per night (five fish per trial). A dispersal speed of 1.0 = water speed; values > 1.0 = fish moving > water speed and values < 1.0 = fish moving < water speed.

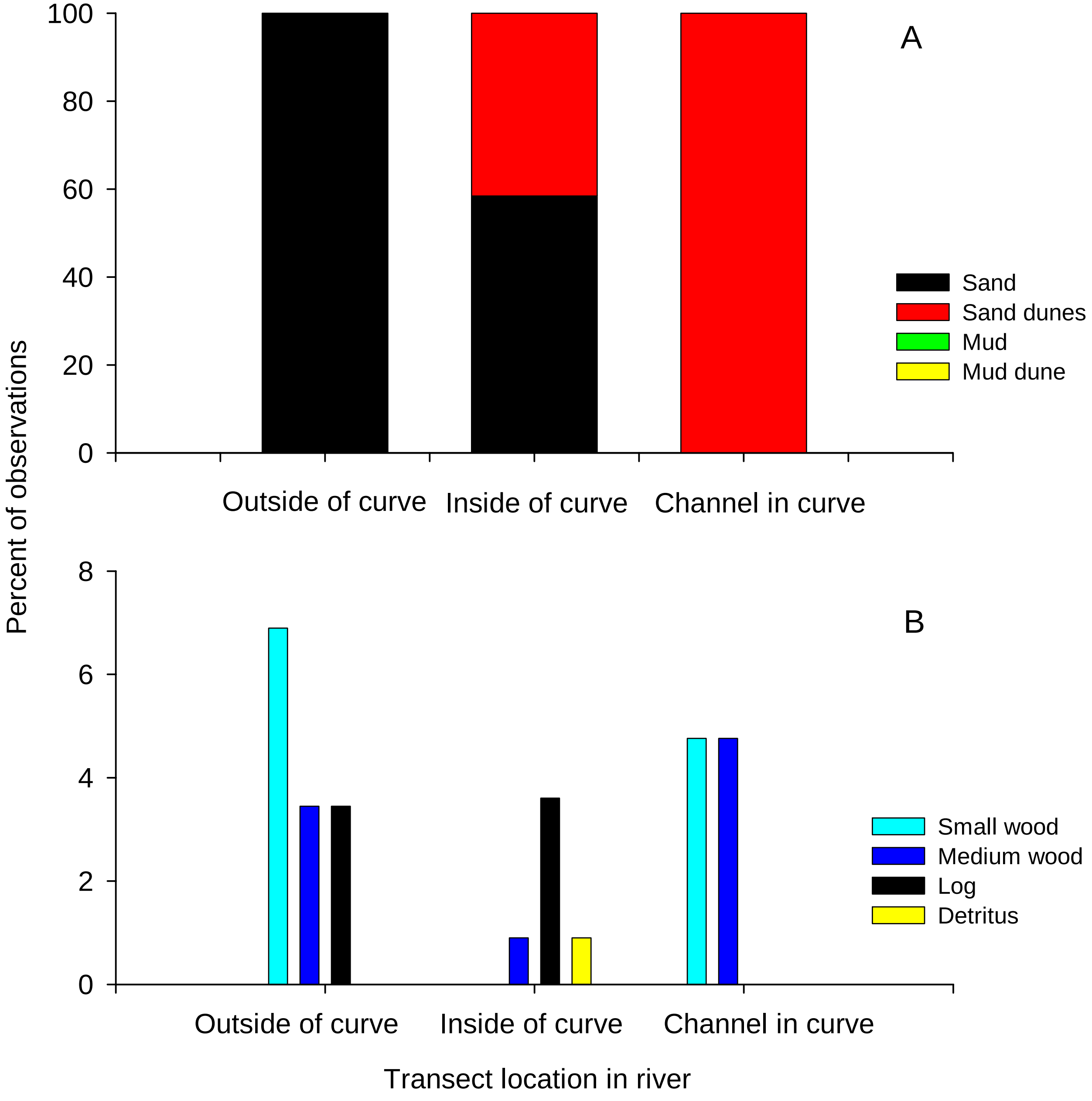

Figure 13. January 2008 surveys of the lower Kootenai River for percent occurrence of bottom types (A) and cover types (B). Characteristics of types are in Methods. Sample locations are shown in Fig. 2.

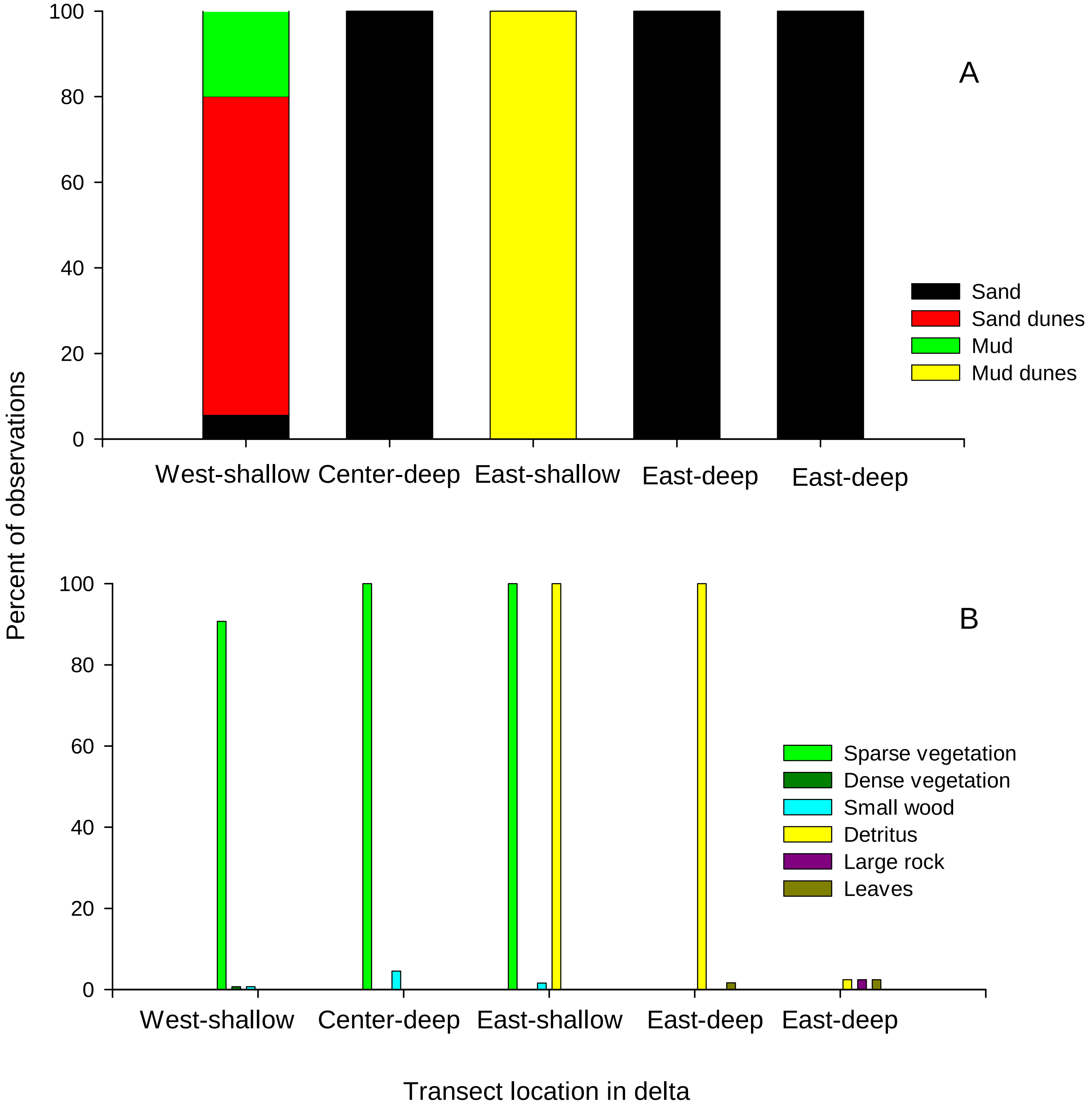

Figure 14. January 2008 surveys of Kootenay Lake for percent occurrence of substrate types (A) and cover types (B). Characteristics of types are explained in Methods. Sample locations are shown in Fig. 3.

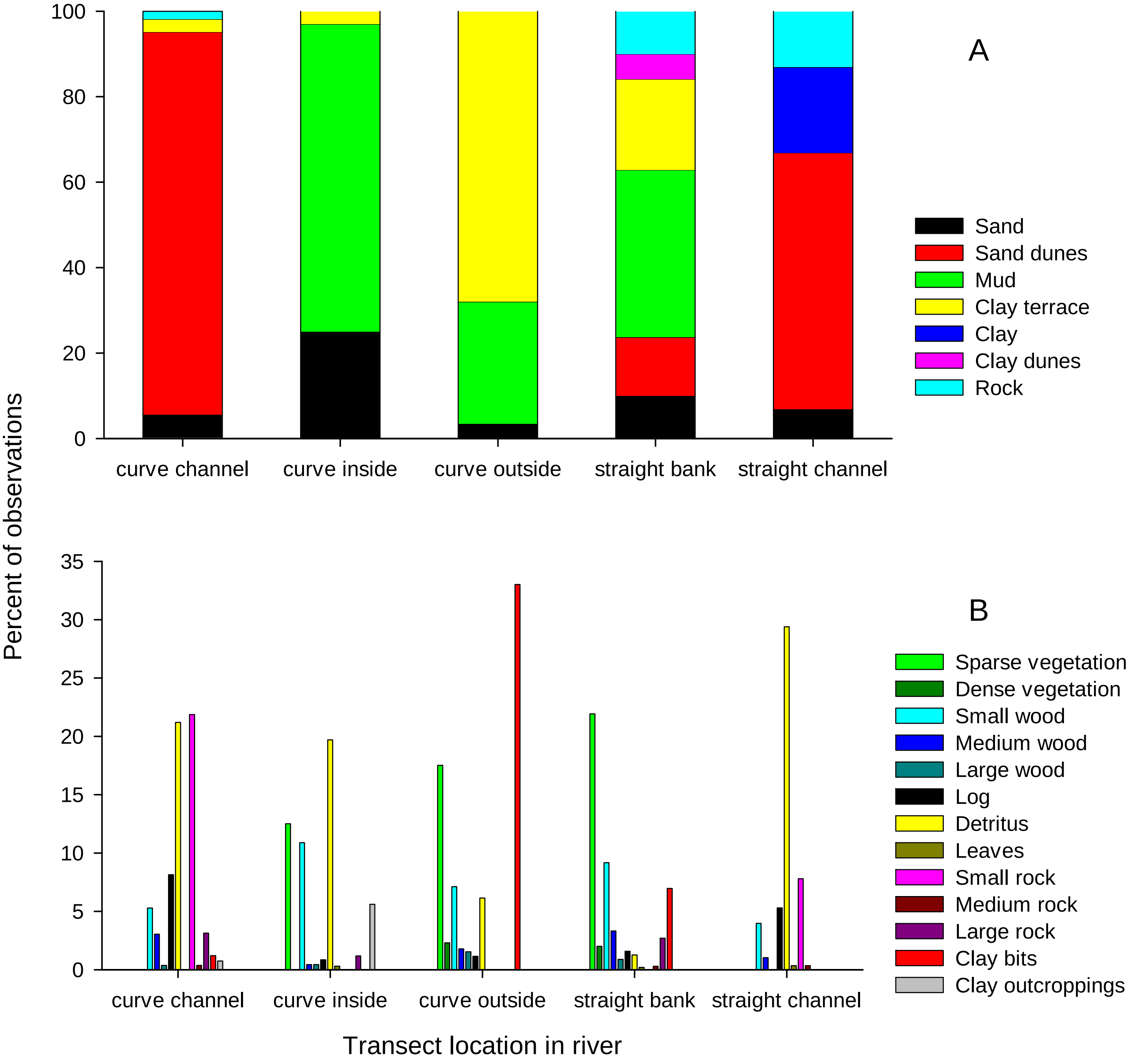

Figure 15. August 2008 surveys of the lower Kootenai River for percent occurrence of substrate types (A) and cover types (B). Characteristics of types are explained in Methods. Sample locations are shown in Fig. 2.

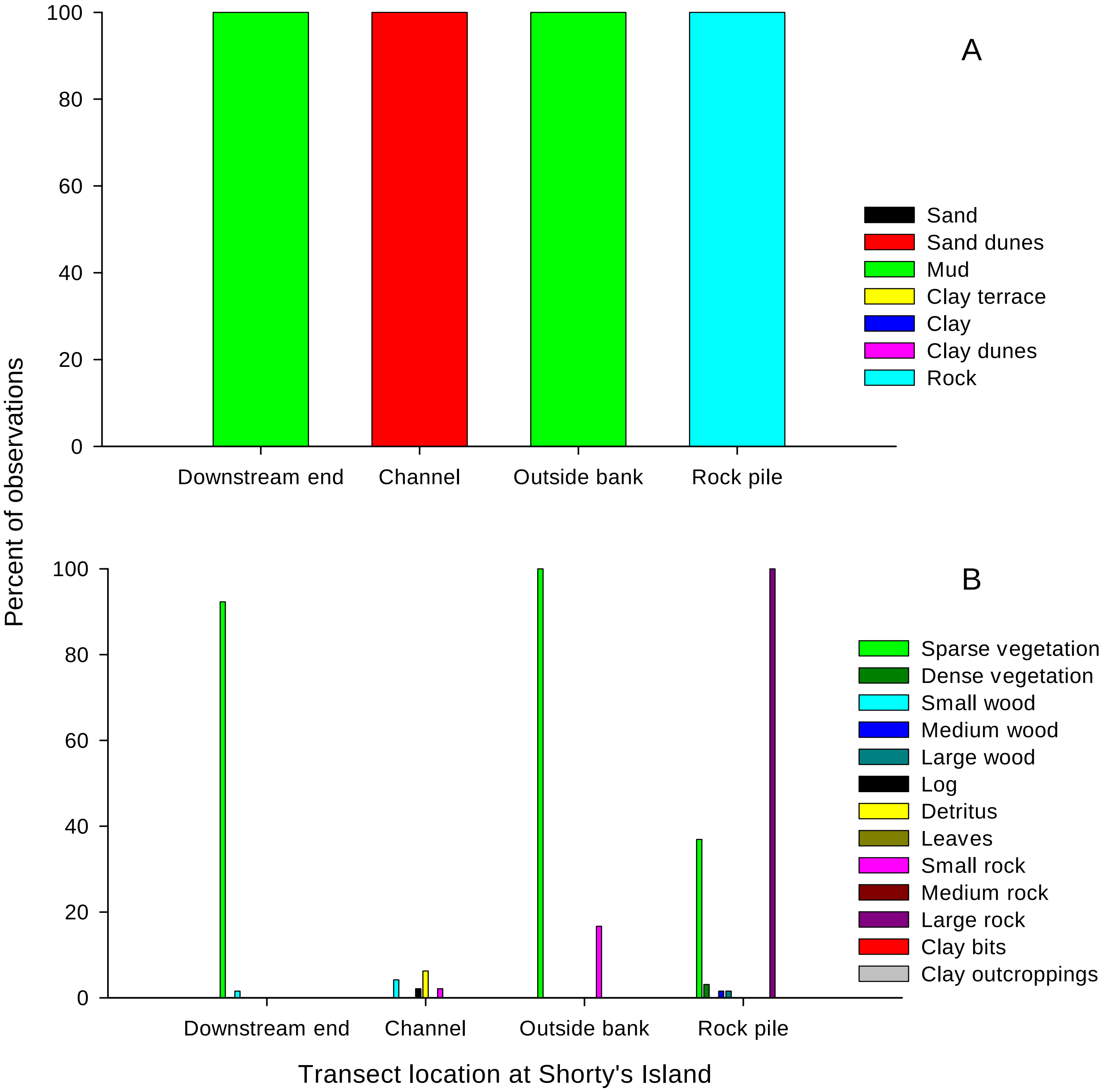

Figure 16. August 2008 surveys of Shorty's Island for percent occurrence of substrate types (A) and cover types (B). Characteristics of types are explained in Methods. Sample locations are shown in Fig. 2.

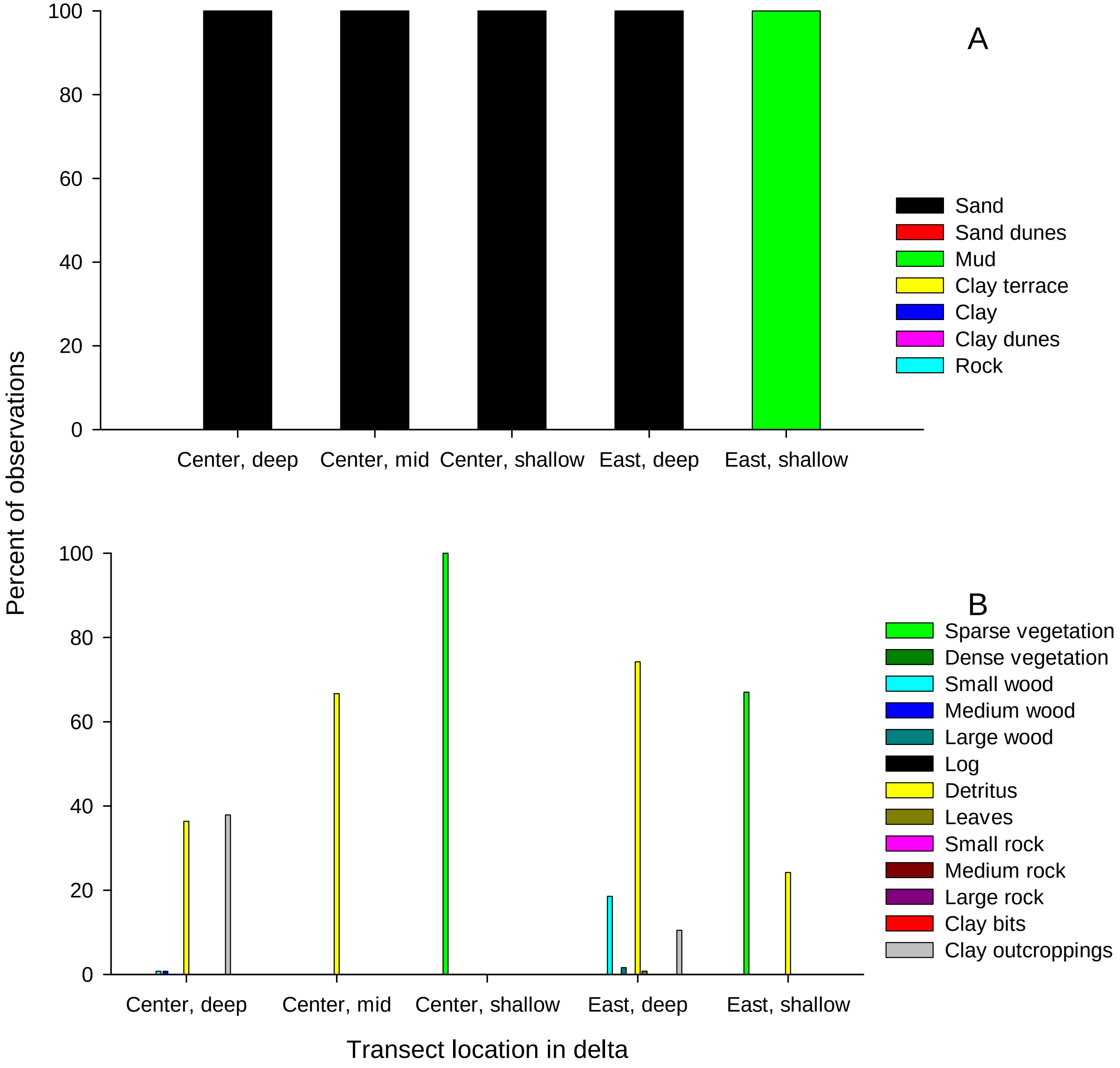

Figure 17. August 2008 surveys of Kootenay Lake and the delta for percent occurrence of substrate types (A) and cover types (B). Characteristics of types are explained in Methods. Sample locations are shown in Fig. 3.