SUMMARY

This report presents the 1997 monitoring results from four sites near the Indian Bathtub that contain, or have contained, populations of the Bruneau Hot-spring Springsnail (*Pyrgulopsis bruneauensis*) and compares them with results from previous years. Three of these sites were monitored in 1990 and 1991 by Mladenka (1992), in 1992 by Robinson et al. (1992), in 1993 by Royer and Minshall (1993), and in 1994, 1995, and 1997 by Varricchione and Minshall (1995a, 1996, 1997). An additional seep at Site 3 (New Seep) was included in the 1994, 1995, and 1997 Springsnail monitoring efforts.

Springsnail population fluctuations at Sites 2 and 3 (Original Seep and New Seep) appear to be related to temperature variability. Temperatures at Site 2 were fairly stable. Temperatures at Site 3 (both Original Seep and New Seep) were often below 24°C and may have affected local Springsnail reproductive success. Rockface study Sites 2 and 3 (both Original Seep and New Seep) maintained Springsnail size-distributions and densities within the range of previous monitoring data. Additionally, Springsnail habitat parameters (food resources, water chemistry) appeared to remain fairly consistent with previous monitoring data. Some rockface habitat may become reduced in quality if bacterial-algal complexes expand further into rockface seep habitat.

A relict population of Springsnails was found 1.80 m from Hot Creek (Site 1). Experiments conducted in 1997 indicated that Springsnail movement rate probably was not the factor that has prevented recolonization. Hot Creek *Tilapia zilli* were shown to be capable of eating *P. bruneauensis*, although the Springsnails did not appear to be a preferred food. High water temperatures in the spring outflow where the relict population is found may be preventing migration to Hot Creek. Estimates of Springsnail populations at Site 2 and 3 (including Original Seep and New Seep) in 1997 were approximately 238,000 and 84,000, respectively.

The different temperature regimes and spatial separation of the Bruneau study sites suggest that the sites may harbor unique Springsnail populations. Controlled growth rate studies and population genetics studies are recommended to address this issue. Under present conditions, maintenance of adequate spring-flow appears to be the most important factor for assuring the success of Springsnail populations at the Bruneau study sites. Habitat improvement and a large scale fish-exclosure/Springsnail transplanting experiment at Hot Creek (Site 1) also are recommended.

INTRODUCTION

The snail *Pyrgulopsis bruneauensis* is an endemic species inhabiting a complex of related hot springs near the Bruneau River south of Mountain Home, Idaho. Hershler (1990) provided a complete taxonomic description of *P. bruneauensis*. Mladenka (1992) focused on the life history of *P. bruneauensis*, providing the groundwork on which this monitoring study is based. Mladenka (1992) found only two studies addressing the biology of *P. bruneauensis*: Taylor (1982) described the taxonomy of the snail and Fritchman (1985) studied its reproduction in the laboratory.

Mladenka (1992) found temperature to be the most important factor affecting the distribution of *P. bruneauensis*. Experiments showed the thermal tolerance range for the snails to be 11-35°C. Reproduction occurred between 20° and 35°C. Snail growth and reproduction were retarded at cool temperatures (<24°C). The study also found that under suitable conditions, recruitment and growth may occur at all times of the year, sexual maturity could occur within two months, maximum size could be reached within four months (both under suitable temperature conditions), and the sex ratio of Springsnails was 1:1. In laboratory experiments, Springsnails were found to survive on all types of substrate, although higher numbers were found on gravel and silt than on sand (Mladenka 1992). Rockface seeps had highly variable temperatures, but never exceeded thermal maximum temperatures. Hot Creek maintained temperatures that were less variable, but often above the Springsnail thermal maximum temperature (35°C) (Mladenka 1992).

A flood in the summer of 1991 contributed much silt, sand, and gravel to Hot Creek. In particular, Indian Bathtub was reduced to less than one-half its size before the flood because of sediment addition. Available habitat in the immediate vicinity of Indian Bathtub was reduced because of this and other sedimentation events (Mladenka 1992). The Springsnail's habitat has diminished considerably in recent years because of agricultural-related groundwater mining in the area (Berenbrock 1993). The Indian Bathtub population has apparently been reduced to zero (Mladenka 1992). Springsnail populations were reduced drastically in Hot Creek (Site 1) by a major runoff event in July 1992 (Royer and Minshall 1993) and have since failed to recover. As of November 1997, there is no evidence to suggest that Springsnails have recolonized Hot Creek since July 1992. Gut analyses performed on two Hot Creek fish taxa, *Gambusia sp.* and *Tilapia sp.*, showed that their diets consisted of organic matter and insects, but not of *P. bruneauensis*. However, these analyses were performed in 1995, a year when Springsnails were apparently absent from Hot Creek (Varricchione and Minshall 1995b).

This report presents the continued biomonitoring of Mladenka's (1992) study sites through November 1997. Additionally, at the suggestion of Varricchione and Minshall (1997), more detailed descriptions of Springsnail populations and habitat at the rockface seep sites (2 and 3 (both Original Seep and New Seep)), along with results from experiments conducted in Hot Creek aimed at determining reasons for the lack of Springsnail recolonization at Site 1, are presented in this report.

METHODS

Site Description

Mladenka (1992) described in detail the three original Springsnail study sites (1, 2, and 3 Original Seep). Figure 1 shows the locations of the three study sites with respect to the Bruneau River. Figure 2a shows a map view of Site 1 at Hot Creek and an adjacent rockface seep. Figures 2b and 2c show front views of the hot-spring study areas (Sites 2 and 3 respectively). Royer and Minshall (1993) recommended that the Site 3 location be divided into two sub-sites: the Original Seep (right side) and a New Seep (left side) (Fig. 2c). These two seeps are approximately 4 m apart from each other and each "seep" has a distinct spring-flow. Their populations were monitored separately during 1994, 1995, 1996, and 1997. Site 2 is also comprised of two "seeps", but their population data have been combined since the first monitoring year. The purpose of the division of Site 3 was to allow the 1994, 1995, 1996, and 1997, Original Seep data to remain consistent with data from previous years and to allow for the inclusion of a recently discovered Springsnail population and habitat into monitoring efforts. [NOTE: The remainder of this report will refer to Site 3 (Original Seep) as Site 3-OS and Site 3 (New Seep) as Site 3-NS.]

Both spring-rockface and stream habitats were examined for *P. bruneauensis* at Site 1. Spring-rockface habitats were monitored at Sites 2, 3-OS and 3-NS. "Spring-flow-covered rockface", or "SFC rockface", was defined as madicolous habitat (rockface covered by a thin layer of running water). "Rockface wetted but lacking flow", or "rockface W/LF", was defined as moist rockface adjacent to spring-flow-covered rockface. Springsnails occur in both types of habitats.

Study quadrats (Appendix A) were established at each site for monitoring purposes. To estimate *P. bruneauensis* size-distribution and density-fluctuation inside a study quadrat, a meter stick (baseline) was positioned flush against the rockface and parallel to the direction of spring-flow. Ten transects,

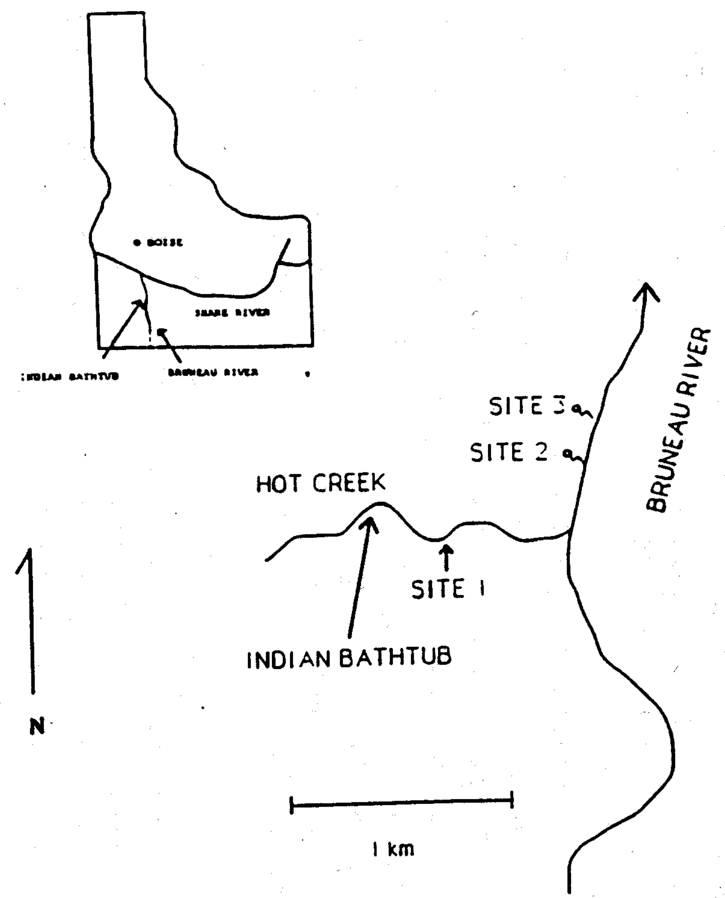

Figure 1. Map showing the locations of the Bruneau hot-spring springsnail study sites. The flow of water between Indian Bathtub and about 100 m upstream of Site 1 is primarily subsurface flow (Reprinted from Mladenka 1992).

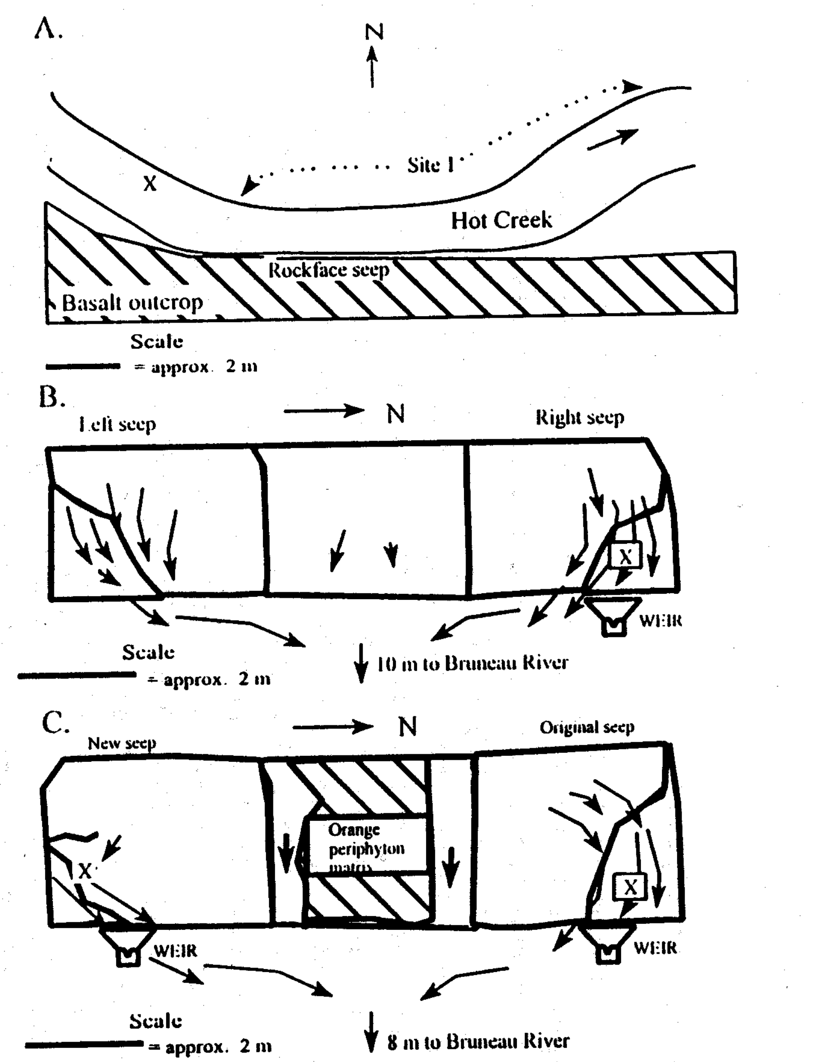

Figure 2. Temperature data logger locations for each of the study sites. Data loggers are represented by "x". A. Map view of Site 1 (Hot Creek). B. Front view of Site 2 rockface. C. Front view of Site 3 rockface (Original and New Seeps).

each perpendicular to the meter stick, were established at 10-cm intervals along the baseline. Random number lists were used to determine random rockface-sampling locations for Springsnail size and density monitoring. The random numbers were used to determine the distance across a transect each sample would be taken or monitored.

Environmental conditions were measured or monitored at the study quadrat (± 1 m) of each site on a monthly basis. These parameters included discharge and stream habitat at Hot Creek (Site 1), amount of flow-covered- and wetted-rockface (Sites 2, 3-OS, and 3-NS), water chemistry, water temperature, and food availability (periphyton abundance). Stream substrate size (particle diameter) was measured at 100 random locations along a 50-m reach of Hot Creek (Site 1 ± 25 m) beginning in June 1995 and continuing on an annual basis.

Springsnail Size Distribution

To determine if the Site 1 Springsnail population was recovering from previous flood events, arbitrary creek substrate and spring-rockface locations within a 50-m reach of Hot Creek (Site 1 ± 25 m) were examined, without magnification, for the presence of *P. bruneauensis*.

Within the sampling quadrats at Sites 2, 3-OS, 3-NS, Springsnails were washed from random locations into a standard petri dish using streams of water from a squirt bottle. The sizes of the snails were determined on site using a Bausch and Lomb dissecting microscope. The microscope ocular was marked with 0.14-mm units (under 7x magnification). Snail lengths were rounded to the nearest 0.14-mm unit (i.e. a snail whose length was 8.8 units long was noted as being in the 9-unit, or 1.26-mm, size class). Sample size was 100 for both sites 2 and 3. Beginning in 1994, population censusing at Site 3 was partitioned between the Original Seep (n=50) and the New Seep (n=50).

Springsnail Population Fluctuations

Density was not measured at Site 1 because Springsnails have not been found there since flooding that occurred in July 1992. Springsnail density was measured at the rockface sites (Sites 2, 3-OS, and 3-NS). Densities were estimated as the number of Springsnails present within the circumference of a petri dish (8.5 cm diameter) at 10 random locations within the sampling quadrat. Densities were reported as the number of snails per m^2. A small Garrity flashlight (2 AA batteries, PR 104 bulb) was used to help distinguish the snails from the dark rockface.

Discharge, Temperature, and Water Chemistry Fluctuations

Stream water velocities were measured across a permanent transect at Site 1 (Hot Creek) using a small Ott C-2 current meter. This transect was moved slightly upstream or downstream (1 or 2 m) if instream vegetation was too thick to allow proper operation of the current meter. Stream discharge (calculated from the measured velocities) was determined using the methods described in Platts et al. (1983). Spring-flow and wetted-rockface area estimates at the rockface study quadrats adjacent to Site 1 were not possible, in general, because of the large amount of vegetation (primarily sedges) obscuring the rockface.

The amount of potential snail habitat at Sites 2 and 3 was estimated by establishing a horizontal transect across each quadrat (Appendix A). The length of the transect which passed over spring-flow-covered or wetted habitat was measured. These values were compared with the width of the transect to obtain estimates of the percentage of the quadrat area covered by spring-flow and the percentage of the quadrat rockface that was moist.

Because of the frequent breakage or loss associated with using maximum/minimum thermometers in earlier monitoring years, miniature temperature data loggers have been used at all sites beginning in 1994. Internal sensor loggers (Onset Hobo-Temp HTI-05+37) were used from 18 February 1994 to 26 September 1994, and then replaced with external sensor data loggers (Onset StowAway-Temp STEB02-05+37) on 26 September 1994 at Sites 1, 2, and 3-OS. Beginning in November 1996, an additional logger was installed at Site 3-NS. Data loggers were downloaded and relaunched approximately every two months, in the laboratory, using LogBook for Windows v.2.03 software (Onset Instrument Corp.).

Figure 2a shows the location of the temperature data logger submersed in Hot Creek. The logger was located 2 m upstream of the regularly-examined section at Site 1. Figures 2b and 2c show the locations of the temperature data loggers at Site 2 and Site 3, respectively. Water depth at the seep study sites was quite shallow. Therefore, small pits were excavated immediately below the seep outflows in order to submerge the loggers in hot-spring water. The loggers were covered by cobble substrate or hillside talus.

Water chemistry parameters were measured for all the study sites. pH was measured, in the field, using an Orion pH meter (Model 290A). The pH meter was calibrated in the field to standard solutions (Orion pH 7.00 and pH 10.01 buffer solutions) during each monitoring visit. Conductivity (µS/cm) was measured, in the field using an Orion conductivity meter (Model 126). Water samples, for all sites, were collected in 250-ml plastic bottles, kept on ice until returned to the laboratory, and then frozen until processed. In the laboratory, samples were thawed at room temperature and shaken by hand (approximately 5 sec) to resuspend any solids. Alkalinity and hardness were determined using procedures described in Standard Methods for the Examination of Water and Wastewater (APHA, 1992).

Periphyton Levels

Periphyton samples were taken from rock substrata collected within 1 m of the study quadrats. For each sample, a modified syringe tube (3.14 cm^2) was placed on top of the substrate. Closed-cell foam, attached to the base of the modified syringe tube, formed a seal between the tube and the substrate to prevent the loss of periphyton sample. Approximately 5 ml of spring or creek water was added to the tube. A modified toothbrush was used to dislodge periphyton from the rock, and a dropper was used extract the periphyton slurry from the tube. The periphyton slurry was concentrated onto Whatman GF/F glass microfibre filters held in a Nalgene filter holder (Nalge No. 310-4000). A

Nalgene hand vacuum pump (Nalge No. 6131-0010) was used to create the suction necessary to remove the water from the slurry. For each sample, this procedure was repeated 3 times to remove all periphyton from the substrate. Periphyton samples were placed on ice, returned to the laboratory, and kept frozen until processed. In the laboratory, periphyton filters were analyzed for the presence of chlorophyll a (corrected for the presence of phaeophytin a) on a Gilford Instruments spectrophotometer (Model 2600) using procedures described in Standard Methods for the Examination of Water and Wastewater (APHA, 1992). Methanol was substituted for acetone as the solvent used in the analyses (Marker et al. 1980). Chlorophyll a, an indicator of the presence of algal organisms, was expressed as mg chlorophyll a per m^2.

The remaining periphyton material from each sample was used in the determination of algal biomass (expressed as g ash-free dry mass (AFDM) per m^2). The material was dried at 50°C for 24 h, cooled to ambient temperature in a desiccator, weighed on a Sauter balance (Model AR1014) to the nearest 10^{-4}g, combusted in a muffle furnace at 550°C for a minimum of 3 h, rehydrated, redried at 50°C, cooled to ambient temperature in a desiccator, and then reweighed. The difference in weights equaled the AFDM of the sample.

Habitat Assessment at Hot Creek

Beginning in March 1995, stream habitat assessment at Hot Creek (Site 1) was conducted monthly using the Idaho Department of Health and Welfare's Habitat Assessment Field Data Sheet for lowland streams (Appendix B; Robinson and Minshall 1995). The parameters assessed included bottom substrate/instream cover, pool substrate characterization, pool variability, canopy covering, channel alteration, deposition, channel sinuosity, lower bank channel capacity, upper bank stability, bank vegetation protection, streamside cover, and riparian vegetative zone width. Also, 100 random measurements of substrate size were made in Hot Creek on an annual basis within a 50-m reach of Hot Creek (Site 1 ± 25 m). Embeddedness of stream substrate was not measured because <30% of the substrate was composed of materials

$\geq$ 1 cm (Varricchione and Minshall 1997). Future changes in habitat parameters should reflect recovery from prior land use activities (i.e. grazing) and recovery from earlier flooding and sediment deposition events in Hot Creek. Also, changes in these parameters, with time, should reflect any habitat improvements that may be made in the area.