Cibola High Levee Pond Annual Report 2003

By

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REPORT SUMMARY

Bonytail and razorback sucker have once again spawned and produced swim-up larvae in Cibola High Levee Pond (CHLP). CHLP continues to support annual recruitment of bonytail while recent razorback sucker recruitment remains elusive. Thus far, razorbacks have experienced intermittent years of spawning success.

Both native species were observed spawning on, or near, the riprap on the river levee. Razorbacks spawned from late January until mid-March over gravel and large cobble along the levee toe (2-3 m depth) and bonytail spawned along the levee shoreline during mid-April. Razorback suckers rapidly fin during the reproductive act, which flushes fines from the substrate and leaves gravel relatively clean. Bonytail on the other hand, appear to spawn over or on substrate that has been disturbed by beaver activity. Substrate scour or disturbance appears to be an important factor in spawning site selection.

Spawning activity was recorded during approximately 120 hours of underwater videography. These films revealed that expulsion of gametes typically triggered feeding frenzies. Both species would aggressively feed on their own eggs and razorback suckers were frequently found feeding among spawning bonytail. In contrast, bonytail were never seen scavenging razorback sucker eggs although it appears they concentrated around the periphery of spawning sites to feed on emerging larvae. Videos also recorded concentrations (average 0.89 to 3.66 animals/m^2) of bullfrog (*Rana catesbeiana*) tadpoles and red swamp crayfish (*Procambarus clarkii*) on spawning sites. Preliminary tank tests indicate both these nonnatives are effective egg and larvae predators.

Telemetry studies revealed that adult bonytail are nocturnal and occupied the interspaces of large riprap during daylight hours. Study fish remained inside these cavities during daylight hours and did not venture out into open water until after sunset. They showed a high fidelity toward specific areas of the pond and often return to the same cavities just before sunrise. All life stages exhibited schooling behavior; however younger year classes make up larger aggregations.

Gut contents suggest adult bonytail diets consist of algae, vegetative material, small fish, and crayfish. Small bonytail were observed feeding near or from the surface on large zooplankton and invertebrates. At least 5 of 27 adults (18%) were infested with intestinal tapeworm (species unknown).

Some nonnative fish continue to occur in CHLP. Bluegill and largemouth bass were removed in November 2002 and others have been observed with underwater cameras.

Predator/prey tank experiments revealed that most nonnatives were effective predators on early life stages of razorback sucker. Bullfrog tadpoles consumed both eggs and larvae. Literature suggests that at current densities, tadpoles have the potential to consume nearly a million fish larvae per day in CHLP. Crayfish were also an effective predator of sucker fry. Small (< 6cm) sunfish, rainbow trout (5 and 18 cm), red shiner (<7 cm), largemouth

bass (<8 cm), yellow bullhead (<14 cm), channel catfish (<13 cm) and bonytail (<8 cm) were all found to be aggressive predators of 10-50 mm razorback sucker.

INTRODUCTION

This past year (2003) represented the second of a four-year field effort designed to describe the early life ecology of bonytail and razorback sucker in CHLP. Last year we conducted extensive surveys aimed at measuring standing crop of the native fish community. Both species had successfully produced young and the community was estimated to include approximately 1,100 razorbacks and 6,000 bonytail (>15 cm)(www.fort.usgs.gov/products/pubs/11000/11000-A.pdf). The pond's standing crop was 4,350 fish/ha with a biomass of 635 kg/ha. In 2002, bonytail continued to produce young but we did not detect any measurable recruitment during the past two years for razorback sucker.

The following description of work is grouped into four categories. Telemetry studies were conducted on adult bonytail from mid-March through early May. The goal was to examine movements, behavior, and locations frequented by adult bonytail during their suspected spawning period. The second major activity was to use underwater videography to verify suspected spawning sites, monitor spawning behavior, determine timing of peak spawning activity and document possible predators.

The third category included physical measurements, trammel netting, light trapping, water quality monitoring, map surveys and acoustical tests. Mapping survey results will be incorporated into the telemetry results to refine movement data. Data collected for these efforts is currently being analyzed and will be presented in next year's report.

We also conducted a series of predator/prey tank tests to identify potential predators observed in CHLP and to examine suspected nonnative predators found elsewhere in the basin. This work is being conducted at, and with the cooperation of the staff at USFWS Willow Beach National Fish Hatchery and at Achii Hanyo Fish Facility.

BONYTAIL TELEMETRY WORK

Methods

On 18 March 2003, 11 adult bonytail were collected from four sets of trammel nets distributed throughout the pond. Their sizes ranged from 358 to 514 mm. Ten fish (404 to 514 mm [x=453 mm]) were fitted with a sonic micro transmitter (8x35 mm). Due to limited battery life (60 d), transmitters were externally attached to the fish to avoid convalescence that is necessary with surgical implantation (Photo 1).

Transmitters were taped to a wire harness with electrician's tape, which in turn was bracketed by two plastic cable ties. This saddle was secured around the fish's caudal peduncle, a process that took approximately 30 sec. Fish were held overnight and released the following morning. We believe this may be the first time this technique has been used on freshwater fishes.

Photo 1. External attachment technique used to attach sonic transmitters to adult bonytail.

Fish movements were monitored using a directional hydrophone equipped with a magnetic compass. Locations were ascertained from bearings taken from two fixed stations. One monitoring location was a point located on the northern end of the river levee and the second was on the northern end of the high levee; the latter point was accessed by boat.

Following release, study fish immediately sought refuge inside rock interspaces of the high levee. Locations were determined using an omni directional hydrophone attached to a 2.2-m pole that could be slid in large cracks between rocks. The high levee shoreline was divided into 26, 15-m linear zones (A-Z), so fish location within the levee would be consistently identified (Map 1).

We had initially planned to conduct hourly, 24-hour monitoring, however, there were no day-time occurrences of fish in open water and it became obvious that fish were only moving at night. The majority of the monitoring effort therefore focused when fish were active.

Monitoring continued for 8 weeks, starting each Monday and concluding after dawn Thursday. Monitoring began by locating all study fish before sunset. Both monitoring stations were manned at sunset and directional bearings of fish signals were taken from both stations at 15-30 min intervals until after sunrise, when fish retreated back to their daytime hiding places. We also periodically checked fish locations during the day. CHLP was also mapped as part of the telemetry work. Existing bathymetric data existed, however, it needed to be updated and we wanted to see if submergent vegetative cover could also be assessed. The work was conducted during the week of July 21, 2003 by Ken Bovee (Map 1). Depth data was collected by boat using a BioSonics DT-4000 echosounder that had a mobile and base station GPS to track coordinates and develop the data base necessary for the map.

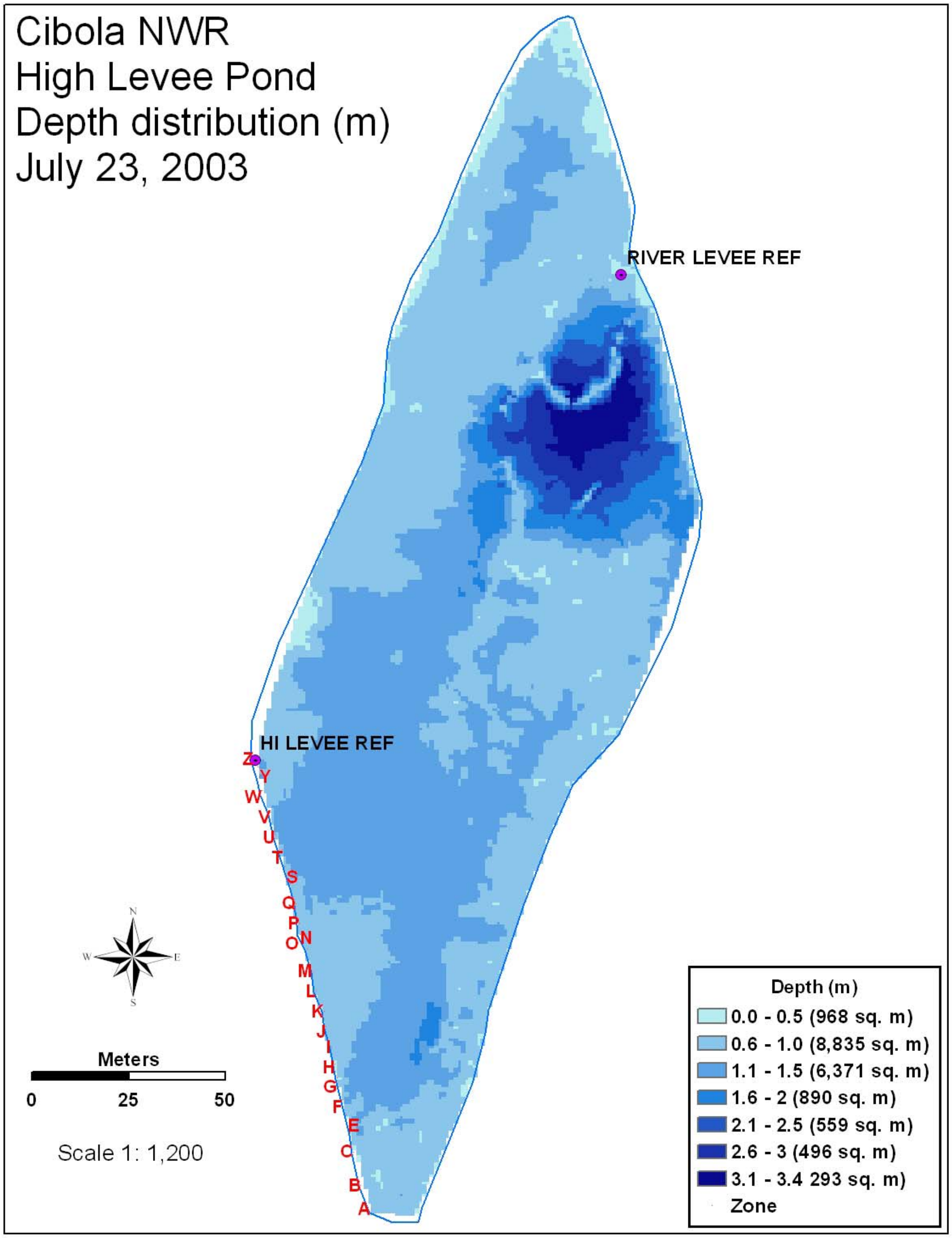
Results and Discussion

Telemetry data are still being analyzed, however, some trends were obvious. All study fish moved during the course of the study; there was no evidence to suggest any mortalities occurred during the study. Some fish frequented open water almost every night while others did not. Study fish always spent daylight hours inside the high levee (Photos 2 & 3). Weak and changing signal strength suggested fish were penetrating the levee several meters and were possibly moving around inside the rock passages. Individuals also exhibited a high degree of fidelity to specific zones or cavities.

Photos 2 & 3. Probing the crevasses of large riprap along the high levee to locate cavities where adult bonytail hide during daylight hours in Cibola High Levee Pond.

None of the fish were detected outside of the levee during daylight hours. Typically, fish did not leave their refuge before 30 min after sunset and returned no later than 30 min before sunrise. While fish moved throughout the pond, some individuals appeared to prefer specific areas.

We discovered that the disturbance of an outboard motor or even rowing caused these fish to retreat back into the riprap. Bonytail were quite sensitive to any type of disturbance.



Map 1. Bathymetric map of Cibola High Levee Pond, Arizona-California showing telemetry reference point (vegetative cover has not yet been added).

VIDEOGRAPHY

Methods

Spawning activities and fish behavior were recorded using a black and white underwater video camera and VHS recorder. The video system consisted of both underwater and surface components. A camera head the size of a soda can was attached to a small bipod fixed to the end of a 3-m piece of steel conduit. We discovered that bonytail avoided visible artificial light, so an infrared spot light (300 W infrared-880 µm) was attached next to the camera for illumination at night. This equipment was wired to a monitor, a VHS recorder, and a 12-V DC battery that powered the system (Photo 4).

Photo 4. Video camera monitor, recorder, and power source set-up on the CHLP river levee.

The camera was aimed by simply turning the system on, lowering it into the water and positioning it using the conduit from shore. Monitoring sessions contained varying proportions of substrate and water column. The viewing area varied due to camera angle, turbidity, and lighting. During daylight hours we could see fish clearly for 1-2 m but at night this distance was often reduced to <30 cm. The relative size of the monitoring area was estimated based on the average size of razorback sucker (50-cm) and tadpoles (75-mm) seen in the frames. Monitoring areas fell into four size categories: 30 X 30 cm, 45 X 45 cm, 60 X 60 cm, and 90 X 90 cm.

The camera was set-up in one of three general locations: 1) the deep portion of the pond off the river levee where razorback were spawning, 2) the area where bonytail spawned the past two years, and 3) at two control sites where spawning had not been observed nor suspected, at the toe of the high levee and at a portion of undisturbed river levee. Underwater activity was recorded on 2-hour VHS videocassette tapes.

Recordings were reviewed using a VHS editor, which time-referenced each 2-hour tape recording. Recordings were paused at precise 5-min intervals. Fish and tadpoles were counted for that single frame and then the tape was played for 60-sec as additional organisms were counted. Based on these counts and area assumptions, the occurrence of razorback sucker, bonytail, and bullfrog tadpoles were converted to organism/m^2 for single frame counts. Number of organisms/m^2 provides an approximation of density while organisms/1-min provides an indicator of activity since a single organism could be viewed more than once. Unique behavior was also noted in the monitoring log.

Results and Discussion

More than 124 hours of underwater monitoring was recorded. All but four hours (control) were directed at bonytail and razorback sucker spawning activity.

Razorback Sucker. Razorback suckers were already spawning when field activities started 18 February, 2003. Groups of adults were seen schooling along the river levee. Actual spawning events caused plumes of sediment to 'boil' to the surface and then gradually disperse. Snorkeling revealed the substrate was comprised of cobble and large gravel. Spawning occurred on the levee's toe, or the deepest area of the pond. Disturbed substrate extended 3 m from the levee's toe and ran parallel to the levee for approximately 30 m (90 m^2). Depth ranged from 1 to 2 m and gradually increased to 2-3 m as water elevation increased in response to rising river stage. Surrounding substrates were covered with approximately 10 cm of fine sediment.

Spawning continued well into March as eggs were discovered on the 19th and larvae were collected on 24 March. Water temperature during spawning ranged from 15.3 to 20.1˚C (bottom temperature, Fig. 1). Spawners had dispersed and bonytails were finally observed in the area by 1 April, 2003.

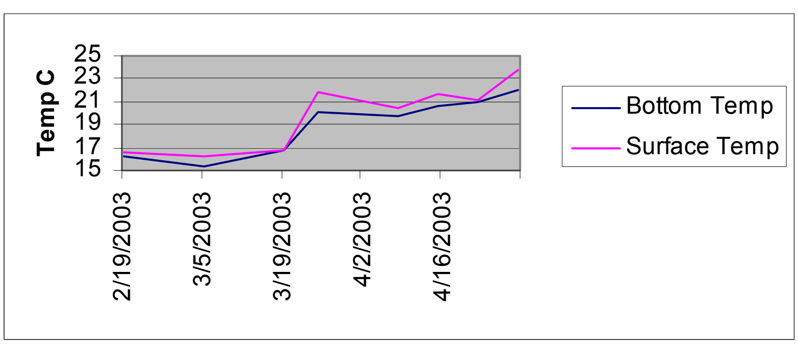
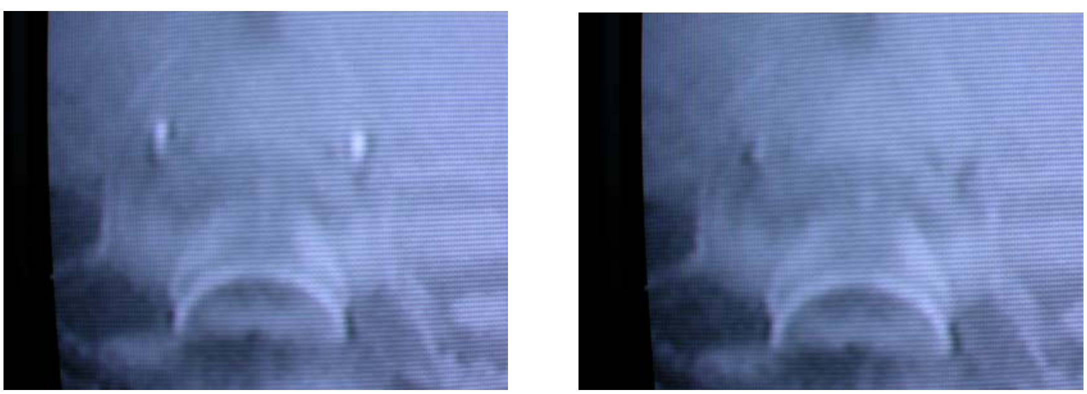


Figure 1. Water temperature taken at the surface and bottom of Cibola High Levee Pond, Arizona-California.

Videography revealed spawning characteristics similar to those described by Minckley (1973, 1983). Fish, mostly males (80-90%), 'cruised' over the spawning area and often lay on the bottom presumably awaiting the arrival of ripe females. Typically, females attracted several males who would position their bodies against the females' posterior. Spawning occurs when the fish settled to the bottom and violently vibrated in unison for 2 to 4 sec. During this process, gametes were released, mixed, and driven into the interspaces of the gravel. The event caused considerable turbidity, which immediately attracted other razorback suckers that apparently fed on exposed eggs. Fish continued to feed in the area for 5 to 10 min, the period during which we assumed all exposed eggs were consumed.

The density of spawning razorback suckers observed on the spawning site averaged 4.76 fish/m^2 (range of 1.19 - 9.0; n=286). Based on the size of the area (90 m^2) and the 2002 population estimate of 1,100 fish, this would suggest that nearly one third of the sucker population was typically in the spawning area.

Razorback suckers would often lie quietly on the bottom in front of the camera. The species has the ability to roll and expose the reflective lining (sclera) of its eyes which causes a distinctive white reflection (Photos 5 & 6). This unique peculiarity proved to be relatively common (n=>50). Its purpose, if any, is not understood, however, it is difficult to imagine such a distinctive display not having some purpose for such a highly camouflaged fish.



Photos 5 & 6. A comparison of a razorback sucker "winking" and not winking its eyes.

Bonytail. We did not witness any daytime schooling over spawning sites as we did last year. Bonytail were not seen from the bank or by snorkeling. Spawners were recorded by underwater videography during the second week of April. Fungused eggs were found between the 8th of April and the 6th of May, which suggests spawning occurred during most of April when water temperatures ranged from 20.4 to 21.6 C° (Figure 1).

Spawning occurred along the river levee shoreline at the same locations where eggs were found last year. Eggs were not visible; however, they were found in the interspaces of small riprap where beaver had been active. The majority of spawning occurred on the central portion of the levee, at the terminus of a large beaver trail and in relatively shallow water (<50 cm). The three suspected spawning sites were relatively small in area (<3 m^2) and limited to disturbed areas. Selection of previously cleaned substrate is not uncommon for other desert cyprinids (Mueller 1984).

Bonytail appear to be very interactive and social. Smaller fish (10-15 cm) were constantly over the spawning area, especially during the night. These fish would form loose schools as they would dart from place to place. Social interactions generally included darting about, chasing and nudging one another with their snout, and foraging for food.

Bonytail densities (video estimates) over known spawning sites averaged 17.44 fish/m^2 (3.6 to 55.4, n=222); considering the relatively small spawning area (<9 m^2), this suggests that only a small portion (5%) of the community spawned at any given time (2002 estimate: 6,000 bonytail >15 cm). Unlike razorback suckers, individual spawning may be shorter in duration (hours versus days) and number of events. It is also quite possible that we have not discovered all the areas used by spawners.

It was difficult to determine what was actual spawning because of all the activity. By all indications, spawning was infrequent and rapid. The video recorded two definite spawning acts. The act would often only take 1-2 sec and involved several fish (3-7) and would be followed by the telltale feeding frenzy by several dozen egg scavengers. One event involved a larger, presumed female that was tightly flanked by two smaller males. The three fish dipped in unison to the substrate, with their backs arched. Immediately, two dozen other bonytails aggressively drove into the gravels to feed. The second observation was of a larger group of fish. Again, 4 to 5 smaller males flanked a larger female. They rested their heads in a depression and pressed their bodies together and tails slightly elevated. Unlike the first sighting, these fish finned vigorously for nearly 2 sec, which again triggered a feeding frenzy involving about 4 dozen fish.

Other "Factors & Organisms". The videos revealed that razorback sucker were spawning over substrate composed of a mixture of small fines to cobble, while bonytail were spawning over more coarse material, composed of small to large angular rip-rap. This difference in material size may influence survival. Horns and Magnuson (1981) reported that substrate composition was an important factor determining lake trout egg survival. They found that crayfish consumed large numbers of trout eggs. Savino and Miller (1991) showed a correlation between substrate size and survivability of lake trout fry.

We also observed high densities of bullfrog tadpoles and to a lesser extent crayfish feeding among spawning fish. This prompted us to examine their possible role in this pond's ecology.

<u>Bullfrog Tadpoles.</u> Tadpoles are often associated with poor production in hatchery ponds, however, until recently, there has been little evidence to suspect them of direct predation. Tadpoles have been traditionally considered microphagous feeders. Petranka and Kennedy (1999) recently reported that many anuran larvae are macrophagous predators, feeding on a wide variety of macroinvertebrates. They also presented data showing that tadpole larvae can be cannibalistic and feed on other anuran species.

There are few reports of tadpoles feeding directly on fish. A veterinarian reported on the Internet that high densities of tadpoles had actually killed adult goldfish (www.fishdoc.co.uk). Boyd (1975) reported that bullfrogs interfered with fish production, but showed no evidence of direct predation. Another paper reported toad tadpoles had consumed on average 17 white catfish larvae per day (Nguenga et al. 2000). The catfish were found to be most vulnerable during the first 6 days after hatching. Underwater monitoring, confirmed by snorkeling, indicated that tadpole densities were inversely proportionate to depth. Densities ranged from 0.9 tadpoles/m^2 (0 to 9.26, *n*=286) in the deeper (2 to 3 m) razorback sucker spawning site, to 2.8 tadpoles/m^2 (0.45 to 5.22, n=48) at the control sites (non-spawning, 1 m depth), to a high of 3.7 tadpoles/m^2 (0 to 17.49, n=222) along shore (bonytail spawning site, 50 cm depth) (Table 1). Based on control densities (2.8 tadpoles/m^2), it is estimated that CHLP could support nearly 57,500 animals. We feel this is conservative, since densities in cattail stands is much higher and not include in these estimates. Based on predation rates (17 white catfish larvae/day) reported by Nguenga et al. (2000) it is conceivable that tadpoles could consume nearly 1 million fish larvae per day in CHLP.

Table 1. Average densities of bullfrog tadpoles (TP), razorback suckers (RZB), and bonytail (BT) determined using video footage taken at razorback sucker and bonytail spawning sites and control sites at Cibola High Levee Pond.

Location	Date	TP/m^2	RZB/m^2	BT/m^2	RZB/min	BT/min	*N*
RZB site	2/19 to 4/1/2003	0.89	4.26	0.00	10.64	0.19	286
BT site	4/2 to 4/16/2003	3.66	0.23	17.44	0.38	22.33	222
Control	4/7 to 4/8/2003	2.84	0.00	0.00	0.00	0.73	48

Based on this information, we initiated laboratory tests to determine if bullfrog tadpoles and crayfish would eat sucker eggs and larvae. These tests will be further refined next year.

PHYSICAL MEASUREMENTS, LIGHT TRAPPING AND NETTING

Methods

Physical water quality parameters were measured using a Hydrolab each trip. Readings were taken by boat at 1-m depth intervals at the ponds deepest location.

Light traps were set to determine the presence of fish larvae, however, their use was restricted to periods following telemetry work because it was believed their presence could affect fish behavior. Light traps were set near suspected spawning areas for intervals of two hours and overnight. Samples were examined in the field. Fish larvae were separated from other organisms and preserved for later analysis.

CHLP was surveyed on July 23, 2003 and a resulting bathometry map was produced (Map 1). This map will be further refined to show submergent vegetation, information that will be presented next year.

Trammel nets were set March 17 to capture adult bonytail for the telemetry study. Only large-meshed (7.5-cm stretched bar) nets were used to avoid capturing small fish. We also set nets following the study to recapture study fish in order to examine and remove transmitters. Immediately following the study (May 7) we set trammel nets parallel to the High Levee at 04:00 hours to intersect fish returning to their cavities.

Realizing our opportunity to recapture large bonytail, we collected gut samples from fish <40 cm using a nonlethal method of stomach and intestinal irrigation that was developed for adult humpback and roundtail chub (Wasowicz and Valdez 1994, Valdez and Hoffnagle 1999). We duplicated this technique using vinyl tubing and a hand water pump (Wasowicz and Valdez 1994). Water was forced through the intestinal track, which flushed consumed material out of the fish's anal vent. The samples were collected, preserved, and are being examined in the laboratory.

Results and Discussion

Hydrolab Measurements. Limnological parameters all appeared normal. CHLP remained generally well mixed and parameters remained within acceptable limits. Temperatures ranged between 16 and 30 ˚C, dissolved oxygen ranged from 7.7 to 10.8 mg/L, conductance ranged from 1207 to 1234 µm/cm, and pH ranged from 7.1 to 7.6.

Fish Larval Collections. Larval light traps were periodically set to determine the presence of fish larvae. Last year we were unable to collect any razorback sucker larvae and with the absence of juvenile razorback suckers in the collection, we speculated that either spawning or larval survival had not been successful. Light traps were again used, except during the bonytail telemetry study. We discovered that adult bonytail avoided light so in order not to influence chub movements, light traps were not used. Subsequent sampling collected both bonytail and razorback sucker larvae.

Razorback Sucker Larvae. Ten larvae were collected by light trap during the week of March 24, 2003. The prolarvae were collected adjacent to the razorback spawning area, which was along the river levee. Six were preserved for later identity.

The larvae were confirmed as razorback sucker (Snyder 1981) and ranged in size from 8.5 to 9.6 mm (avg. 9.0 mm). One larvae had been actively feeding and had a full gut of small *Daphnia*. Their distinctive eye spot made it possible to count individuals; the larvae had recently consumed 25 daphnia (Photo 7).

Bonytail. Light traps were set only two nights during the week of May 6, 2003. Four larvae were taken from traps set along the river levee during the bonytail spawn. Lengths ranged from 7.8 to 15.8 mm (avg. 11.1 mm) and could contain both species. Identification of these larvae is pending.

Trammel Netting. Trammel nets were set on May 7 to recapture bonytail used in the telemetry study. The following night, nets were set prior to sunset to capture fish leaving their cavities. A total of 28 bonytail were captured during the two efforts, and four had been used in the telemetry study (Table 2). This effort was repeated July 1 and 2 in an attempt to recapture the remaining 6 study fish. Catch rates (CPUE) for bonytail were nearly twice at high in May as for the other collection efforts. Only 3 bonytail were captured in July, the lowest catch rate. It appears during the heat of the summer, fish activity may drop dramatically, as fish become inactive.

Table 2. Trammel net catch rates (CPUE) for bonytail and razorback suckers captured in Cibola High Levee Pond, AZ-CA.

	Bonytail		Razorback suckers	
Date	Number	CPUE *	Number	CPUE *
3/17/2003	11	1.37	12	1.50
5/7/2003	28	2.27	3	0.24
7/1-2/2003	3	0.17	6	0.40

* fish/hour/100 m^2 of net

Overall, the study fish were in good shape, and we did not observe any external parasites. We found that the tag attachment of recaptured study fish had chafed the dermis around the caudal peduncle. It appears the cable tie teeth ‘sawed’ through the skin due to swimming action. This might be remedied by switching to a wider banding material that does not have teeth or use something less ridged, like electrician’s tape. Otherwise this approach appears to have excellent potential for short-term studies.

Dietary Samples. The gut contents of 30 fish were collected (Photo 8). The 20 fish collected just prior to dawn contained the most material. Samples have not been fully examined but it was obvious they contained crayfish, remains of small fish, vegetation, and small particle debris. The ten fish examined just after dusk contained very little gut contents. We recovered tapeworm contents from 4 (13%) of the 30 fish.

We occasionally found dead fish. One razorback male was found during the spawning season and there was no apparent reason for its death. I observed a tuberculate and highly fungused male during the telemetry study. The fish spent about 10 min on the surface next to the boat. It appeared the fish was curious about the hydrophone. Five dead adults (1 BT, 4 RZB) were found during the week of July 22. Again the fish showed no external evidence of disease, parasites, or wounds. These fish had not bloated and appeared to have died recently. The single BT was a telemetry study fish that still retained its transmitter. The cable ties had left a wound, similar to those fish collected earlier. The wound was clean, not inflamed or infected and did not appear to have caused the fishes death. The size of the fish, the fact that they died near or at the same time suggests some event (i.e., lightening, poisoning) may have caused their death.

Photo 7. Larval razorback sucker intestinal track filled with 25 daphnia (note eye spots).

Photo 8. Gut contents of an adult bonytail being flushed into a pan.

PREDATOR/PREY TANK TESTS

Few biologists question the predatory role of nonnative fishes. Substantial funding and effort is currently being expended toward the removal of large predators, such as largemouth bass, northern pike, and channel catfish (McAda 1997, Jackson and Badame 2002, Modde and Fuller 2002). Unfortunately, systematic research has not been conducted to determine the extent of the problem or whether removal is even feasible.

Removal programs have become quite popular with management agencies and the present trend is generally toward removing large recreational predators. Hundreds of thousands of game fish have been removed. Unfortunately, there is little or no data to suggest native communities have benefited.

One of the purposes of targeting large predators is to reduce the reproductive capabilities. Unfortunately, there is growing evidence that while the abundance of large individuals has decreased, there has been a responding increase in smaller and intermediate-sized predators (Smith and Brooks 2000, Demers et al. 2001, Davis 2003). It appears standing crop has remained unchanged, but removal efforts have simply shifted size distribution of these populations.

This response may actually create predation risks for small fish that are actually worse than no action at all. The majority of native fishes are disappearing before they reach a few days old---at most a few weeks old---which suggests they are being lost by smaller predators rather than larger ones. Typically, large predators feed on large prey while small predators feed on small prey. The optimal foraging theory predicts that predators will choose prey sizes giving the highest energy return per time spent foraging. In simple terms, adult eagles don't waste time chasing flies. All evidence suggests that extremely young native fish are not being lost to the large predators.

The increase of small predator densities may actually increase predation pressure, especially in nursery habitats. Small predators have better access to shallow and densely vegetated habitats and they also compete for food and space. It is possible small nonnative fish pose more of a predatory threat to early life stages than large predators (Nesler 2002).

There is very little information available regarding the role of small nonnatives fishes. Ruppert et al. (1993) raised concerns about the role of adult red shiner on native larvae in the Yampa and Green Rivers. Unfortunately, the problem is not just limited to predatory fish. The detrimental aspect of bullfrog and nonnative crayfish introductions is currently being examined in amphibian restoration programs. These same problems appear evident for native fishes in CHLP.

The objects of our test are two fold: 1) to determine the role of nonnative species (tadpoles and crayfish) in Cibola High Levee Pond, and 2) examine the role of small nonnative species in general, in the lower Colorado River mainstem.

Methods

Razorback sucker larvae and fry were supplied by USFWS while nonnatives were provided by agency hatcheries, captured nearby, or purchased from aquaculturists. Tests were conducted in 10 and 30-gal tanks located at Willow Beach and Achii Hanyo Fish Hatcheries.

Bullfrog Tadpole-Fish Egg Test. During routine field sampling, several adult razorbacks freely expressed gametes in the holding tank. Four hundred of these eggs were salvaged for predation experiments with bullfrog tadpoles obtained from CHLP.

Four 10-gal aquarium tanks were filled with 7 gal of water and aerated. One hundred razorback eggs were placed in each tank. Twenty-five tadpoles were placed in each of three tanks, leaving one as a control (no tadpoles). Lengths of 10 tadpoles were measured in each group (Table 3). Water temperatures ranged from 20 to 22˚C. The experiment started Thursday afternoon and ended Monday morning, approximately 90 hours later. We were unable to make daily observations.

Crayfish-Razorback Fry. Tests were conducted examining crayfish predation on razorback sucker fry. Three sets of tests were conducted using four 10-gal tanks. The experiments were designed to determine if razorback sucker fry utilized overhead cover and to examine associated predation rates.

Table 3. Bullfrog tadpoles used in egg predation experiments.

Bullfrog tadpoles	Tank 1	Tank 2	Tank 3	Tank 4
Wet weight (g)	102	108	104	Control
Length range (mm)	54-83	48-89	46-82	
Average length (mm)	75	75	73	

Each tank was elevated (10 cm) on one end using a block of wood and filled with 7 gal of water. This provided both a shallow (10 cm) and deep (20 cm) end. Plastic, imitation hydrilla was used in two experiments to determine if fry were attracted to the cover regardless of water depth. On one test the cover was placed near the surface of the shallow end and in the second test it was placed in the deep end.

Razorback sucker fry were counted and measured and 20 were placed into each aquarium. Two crayfish were then placed in each of three tanks. Fry were fed brine shrimp (*Artemia*) once a day and their numbers were counted after 24, 48, and 72 hours.

Potential Predators of Razorback Larvae. To examine potential predation by multiple predator species, eight 30-gal tanks were set up at Achii Hanyo and Willow Beach hatcheries. Predators were obtained from Willow Beach, captured within the lower Colorado River basin, or acquired from aquaculturists (Table 4). The fish farms gave us fish that had been in ponds and had not yet switched to an artificial diet. However the rainbow trout and bonytail from Willow Beach hatchery were fed artificial flakes or pellets throughout their life. When predators were brought into the experiment, we switched their diet to frozen bloodworms ad libidum. To make the experiment more conservative, predators were not starved before each trial; instead they were fed approximately 0.2-0.5 g of bloodworms/tank within one hour of beginning of a trial and fed every few hours throughout the trial as well. Razorback larvae were fed live brine shrimp (*Artemia*) or artificial larval food supplied by the hatchery.

When razorbacks were <30 mm, all experimental tanks contained 20 razorback larvae. When razorbacks reached 30-50 mm, experimental tanks contained a single prey treatment of 20 razorbacks or a mixed-prey treatment of 10 razorbacks and 10 fathead minnows (Table 4). We carefully measured prey for the mixed treatments so that they were all a similar size. The purpose of adding fathead minnows to the experiment was to determine if predators indicate a preference between the two prey species, or if the non-native prey affects predation on razorbacks.

At least two control tanks (prey species with no predator) were used in each predator trial. All treatment tanks contained four predators of the same species. Trials ran for at least 12 hours and ended when we removed predators and counted surviving larvae.

For the experiments using the more fragile fish under 30 mm, once we determined that they fit within a 1-cm size class, a subsample of larvae were measured to mm to reduce the stress on all fish before subjecting them to a predation experiment. For the experiments using 30-50 mm fish, measurement methods varied. We continued to measure a subsample on the bluegill trial. However, we realized that obtaining accurate measurements of all prey fish before and after the experiment would allow us to compare the size of starting fish compared with the size that survived. This information could be used to determine if predators were size-selective. We also measured head width and depth of prey to compare with gape measurements of predators.

Results and Discussion

<u>Bullfrog Tadpole-Fish Egg Test.</u> Videography at CHLP indicated that tadpoles were present and actively feeding among bonytail and razorback sucker spawners. This triggered our curiosity regarding their role in the pond and possibly the wild, which led to the tank tests. Trammel et al. (2002) suspected they might be contributing to fish losses in the Upper Colorado Basin, but their examination of intestinal contents provided no evidence to support their concerns.

Table 4. Potential predators used in chronological order in razorback larvae trials, 2003.

Predator species	Predator size range (mm)	Predator biomass g/m3	Razorback larvae size range (mm)	Dates and duration of trials	Origin of predators
Red shiner	46– 65	TBA	9 - 12	21-22 Mar; 26 hours	Virgin River
Bonytail	57-75	TBA	9 - 12	23-24 Mar; 24 hour	Willow Beach Hatchery
Lepomis sp.	37-57	TBA	12 - 16	26-27 Mar; 24 hours	Colorado River below Davis Dam
Rainbow trout YC-1	40-51	TBA	12 - 15	27-28 Mar; 14 hours	Willow Beach Hatchery
Bullfrog tadpole	72-87	TBA	10 - 15	11-12 Apr; 24 hours	CHLP
Yellow bullhead	91-138	TBA	10 - 15	12-13 Apr; 18 hours	Irrigation ditches surrounding Achii Hanyo Hatchery
Rainbow trout Adult	123-182	TBA	18 - 28	28-29 May; 14 hours	Willow Beach Hatchery
Bluegill	75-115	TBA	31 - 39	24-27 Jun; 63 hours	Park Moabi, Colorado River
Channel catfish	88-126	TBA	30 - 50	10-14 Jul; 88 hours	Hopper-Stephens hatchery, AK
Largemouth bass	62-79	TBA	30 - 45	1-2 & 14-16 Jul; 24 and 39 hours	Anderson Fish Farms, AK

Tank experiments suggest that tadpoles (n=75) had consumed all the razorback sucker eggs (n=300) within 90 hour. Predation was 100%.

There has been a great deal of suspicion but little evidence that tadpoles are competitors or possibly predators on fish eggs and larvae (Boyd 1975, Kane et al. 1992). A glimpse at the literature provides ample evidence that tadpoles feed on frog and salamander eggs (Morin 1983, Kupferbert 1997, Petranka et al. 1998). It has also been reported that tadpoles excrete a growth inhibitor that interferes with fish spawning (Boyd 1975) and that toxicity from eating frog eggs could cause reproductive failure in other anuran species (Petranka and Kennedy 1999). We could find only two references of tadpoles feeding on fish larvae (Savino and Miller 1991, Nguenga et al. 2000).

<u>Crayfish-Razorback Sucker Fry.</u> We witnessed similar losses with crayfish. Within 72 hour, 2 crayfish had killed 96.7% of the razorback sucker young (16.2 mm) in the 'No Cover' experiment (Table 5). Unfortunately, the tests were inconclusive concerning the role of cover. Predation rates in the 'Deep Cover' tests were lower than the control (70%). The shallow tank tests were delayed and we were unable to use razorback sucker fry of the same size. The young razorbacks were substantially larger (20 mm versus 16

mm). We believe the larger fish were better at avoiding predators than their younger cohorts.

There is ample evidence that crayfish can be effective competitors and predators of native fishes (Minckley and Craddock 1961, Guan and Wiles 1997, Carpenter 2000). Crayfish can be aggressive predators of fish eggs and juveniles (Horns and Magnuson 1981, Savino and Miller 1991, Carpenter 2000). The tank tests supported previous literature.

These tests will be repeated next year, using fish of the same size groups with complete series of various sizes; hopefully we will also expand tests to include bonytail.

Table 5. Results of tanks tests examining predation rate of razorback sucker fry by crayfish in the presence and absence of protective cover.

	Tank 1	Tank 2	Tank 3	Tank 4	Predation Loss
No Cover (RZB length 16.2 mm, crayfish 34 mm)					
Start	20	20	20	20	0%
24 hours	20	4	6	15	58.3%
48 hours	20	0	0	5	91.7%
72 hours	20	0	0	3	96.7%
Deep Cover (RZB length 16.1 mm, crayfish 48.9 mm)					
Start	19	20	20	20	0%
24 hours	19	20	16	16	13.3%
48 hours	19	13	10	6	35.0%
72 hours	19	9	4	5	70.0%
Shallow Cover (RZB length 20.5 mm, crayfish 54.7)					
Start	20	20	20	20	0%
24 hours	20	19	20	20	1.7%
48 hours	20	18	18	20	3.3%
72 hours	20	28	17	20	8.3%

Potential Predators of Razorback Larvae. All tested predators consumed razorback suckers. Larvae suffered 100% mortality in both rainbow trout tests; mortality was >85% in tanks with bonytail, red shiner, and yellow bullhead (Table 6). Bullfrog tadpoles and *Lepomis* sp. (bluegill and/or redear <6 cm) had low predation rates; this is likely due to the smaller gape of these animals compared to other predators.

Table 6. Predation rates ($\bar{x}$ ± SE) in single-prey (RZB*) and mixed-prey (RZB + FHM) treatments.

Predator	Number of treatment tanks	Percent predation (Instantaneous predation rates **)	
		Razorback larvae	Fathead minnows
Razorback larvae 10-16 mm			
Red shiner	4 RZB	87.5 ± 9.5 (0.168 ± 0.018)	---
Bonytail	4 RZB	88.8 ± 2.4 (0.185 ± 0.005)	---
Lepomis sp.	5 RZB	64.0 ± 17.1 (0.133 ± 0.036)	---
Rainbow trout YC-1	4 RZB	100 ± 0.0 (0.357 ± 0.0)	---
Bullfrog tadpole	5 RZB	7.0 ± 3.4 (0.015 ± 0.007)	---
Yellow bullhead	4 RZB	98.75 ± 1.3 (0.274 ± 0.003)	---
Razorback larvae 18-28 mm			
Rainbow trout Adult	6 RZB	100 ± 0.0 (0.455 ± 0.0)	---
Razorback larvae 30-50 mm			
Bluegill	3 RZB	18.3 ± 4.4 (0.015 ± 0.003)	---
	2 RZB + FHM	55.0 ± 35.0 (0.022 ± 0.014)	5.0 ± 5.0 (0.002 ± 0.003)
Channel catfish	3 RZB	50.0 ± 7.6 (0.028 ± 0.004)	---
	3 RZB + FHM	63.3 ± 14.5 (0.018 ± 0.004)	13.3 ± 3.3 (0.004 ± 0.001)
Largemouth bass	5 RZB	58.0 ± 4.4 (0.099 ± 0.006)	---
	6 RZB + FHM	66.7 ± 13.6 (0.050 ± 0.006)	63.3 ± 7.2 (0.051 ± 0.003)

* RZB = razorback sucker, FHM = fathead minnow
** number of fish consumed/predator/hour, based on duration given in Table 4

Comparing predation rates between predatory species is complicated by the different rates of satiation in experiments with larger prey and the variation in test duration (see Table 4). Also, in the bluegill test, the predators appeared to be unusually uninterested in the prey, possibly due to shock from handling before the experiment. This test will be repeated in 2004. Instantaneous predation rates (number of razorback larvae consumed/predator/hour) account for variations in test duration. Predation rates varied from 0.015 for bullfrog tadpoles to 0.455 for adult rainbow trout (Table 6).

Mortality in control tanks was insignificant: it occurred in 6 of 29 control tanks (a total of 580 larvae), with a total of 7 deaths. Thus mean mortality in controls was 1.03% ± 0.46 SE). All but one dead larvae were <15 mm.

In mixed-prey experiments (channel catfish, largemouth bass, bluegill), the fathead minnows used at the beginning of the experiment were the same size as the razorback suckers (separate variance t-tests: $p \geq 0.1$; Table 7). Surviving prey were also not significantly different in size, suggesting that the predators were not size-selective for the prey sizes they were presented with.

Table 7. Size of prey ($\bar{x} \pm$ SE) in mixed-prey tanks with predators at beginning and end of experiments. Separate variance *t*-tests compare differences in size by prey species. NM = Not measured.

	All Prey Beginning of experiment (mm)		Surviving Prey End of experiment (mm)	
Predator	Razorback sucker	Fathead minnow	Razorback sucker	Fathead minnow
Bluegill	35.0 ± 0.9 n=8	36.8 ± 0.5 n=8	NM	NM
	t=1.79; df=10.6; *p*=0.10			
Channel catfish	41.7 ± 0.7 n=30	41.6 ± 0.6 n=30	44.7 ± 1.2 n=11	42.3 ± 0.6 n=25
	t=-0.11; df=55.2; *p*=0.92		*t*=-1.8, df=15.5; *p*=0.09	
Largemouth bass	39.4 ± 0.6 n=30	40.2 ± 0.6 n=29	40.6 ± 0.6 n=19	41.9 ± 0.6 n=15
	t=0.87, df=56.4, *p*=0.39		*t*=1.56, df=31, *p*=0.13	

When examining mixed-prey treatments only, there was no difference in percent predation for razorback suckers in single-prey versus mixed prey experiments (separate variance t-tests; $p > 0.15$; Table 6). Considering the difference in percent predation by bluegill, the lack of statistical difference is likely due to the small sample size (n=2); repeating the bluegill test in 2004 appears worthwhile.

The mixed-prey tests suggest that bluegill, channel catfish, and largemouth bass do not prefer razorback suckers over fathead minnows, nor that razorback suckers are more vulnerable to predation when in the presence of fathead minnows.

CONCLUSIONS

CHLP continues to be an amazing place to study. Sadly, it remains the only oxbow community where the early life stages of these two species can be studied. We have uncovered some interesting information but so much more needs to be discovered.

There are specific locations or habitat types that are being used by specific live stages. Adult fish are utilizing the entire pond. Spawners of both species are using rock and gravel found along or on the river levee that has been previously disturbed. During the past two years, there has been no evidence to suggest spawning occurs elsewhere. However, in past years when high water flooded the access area, bonytail there were observed exhibiting spawning behavior (M. Thorson, FWS, personal communication). The use of other areas of the pond by spawners, especially at various river stages, cannot be ruled out. These species are remarkably adaptive.

The telemetry study revealed that large bonytail utilized the large cavities found in the riprap of the high levee during daylight hours. The use of this area by all ten study fish suggests dark cover is an important component of the adults preferred habitat. Historically, large rock talus was rare in this area of the basin, however, woody debris, large snags, root wads, and drift piles were prevalent but have been lost with the deforestation of the river's riparian community (Minckley and Rinne 1985). This type of cover appears to be absent from the river today.

The use of cavities may have been an effective survival strategy to reduce avian predation, but today, with the presence of channel and flathead catfishes, this behavior may put bonytail at greater risk. These factors merit further examination.

Fish larvae and juveniles are routinely found utilizing areas that have dense overhead cover. This preference may have predatory implications. Young fish are most frequently seen in or near beaver den entrances where water depths are normally a meter or more. Small schools of larvae and juveniles were observed sharing shallower habitats with crayfish, which had their claws extended and were actively trying to capture them. Added depth may provide young fish a margin of space or safety.

Tank tests revealed an alarming predation rate for small nonnative predators that included crayfish, amphibians, and fish. Small predators are far more numerous, have easier access to shallower and more densely vegetated habitats than their larger cohorts. They are also relatively immune to most mechanical removal techniques. It is becoming painfully obvious that nonnative predation is an overwhelming obstacle to recovery that we may never fully resolve.

Bonytail school, and the size of these schools decrease with age. Relatively large (>100 fish) schools of larvae and fry were found under the protection of overhead cover. We also observed tight schools of small (<10 cm) bonytail that numbered up to 50 fish. Schools of older juveniles (10-15 cm) were fewer in number and more dispersed in nature. The underwater camera occasionally captured bonytail >20 cm, but these events were rare (5 fish/hour). One event involved a small school (5) of large adults.

Bonytail of all sizes were cover-oriented and nocturnal in nature. While we observed bonytail using the deeper portions of the pond during the day, their numbers dramatically increased at night. They often fed on the surface and by using spotlights we could detect them throughout the water column, where we presume they were feeding on larger plankton.

Larger fish (>30 cm) were far more rare and were only observed at night. One large bonytail was observed attacking and driving a relatively large crayfish from a spawning site.

Next Years Work

Telemetry equipment will be used to further describe the daily behavior and use of cover by bonytail. We are examining the possible use of extremely small transmitters on juveniles (15-20 cm). The goal would be to determine behavior and diel movements, especially in terms of habitat and cover use. This work is planned for the next two years.

Sampling will increase in an effort to gather more information pertaining to growth rates and habitat use. We plan to use large winged hoop nets to determine the movement of various life stages of bonytail and razorback sucker. We plan to examine the gut contents of more bonytail to get a better handle of the extent of the tapeworm infestation and to determine what smaller bonytail are eating.

Sampling will also incorporate fish acoustics in an attempt to gather more data pertaining to daily movements, densities, sizes, and spatial distribution. We are planning a minimum of 4 seasonal surveys.

Tank experiments will continue to examine potential small predators of razorback sucker. We plan to conduct additional tank tests examining the rate of egg and larvae predation by tadpoles as well as possible behavioral differences between razorback sucker and bonytail juveniles. We also plan to conduct experiments examining the interactions of large and medium sized predators in relationship to larval native fish losses.

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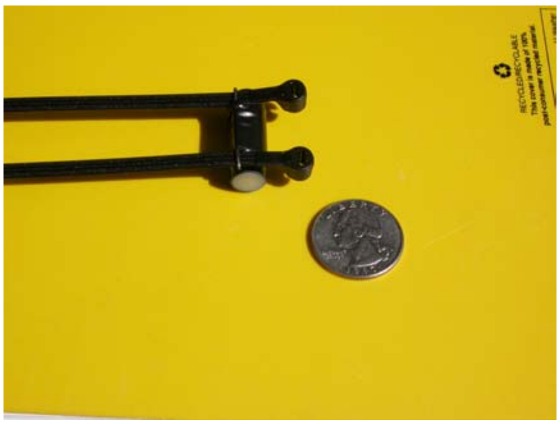
Photographs



Plastic cable tie harness holding a micro sonic transmitter.

Wound resulting from wearing a transmitter harness for two months.

Comparison of female (L) and male (R) bonytail genitalia.