RELATIONS AMONG TROPHIC STATUS, VEGETATION ABUNDANCE, AND SPORTFISH DREEL FOR 44 FLORIDA LAKES, AND THE USE OF LOW-FREQUENCY SOUND AND FOOD REWARDS AS AN AID IN TRIPLOID GRASS CARP REMOVAL FROM A FLORIDA POND

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**A THESIS PRESENTED TO THE GRADUATE SCHOOL
OF THE UNIVERSITY OF FLORIDA IN PARTIAL FULFILLMENT
OF THE REQUIREMENTS FOR THE DEGREE OF
MASTER OF SCIENCE**

UNIVERSITY OF FLORIDA

May 2002

CHAPTER 3

USE OF LOW-FREQUENCY SOUND AND FOOD REWARDS TO AID IN TRIPLOID GRASS CARP REMOVAL FROM A FLORIDA POND

Introduction

Florida's warm climate and long growing season provide aquatic macrophytes the opportunity for abundant growth. Unfortunately, hydrilla (*Hydrilla verticillata*) and other introduced plants have also flourished as a result of these conditions, interfering with water-use activities throughout the state. In response to these increasing aquatic weed problems, grass carp (*Ctenopharyngodon idella*) were introduced into Florida as a potential biological control agent (Osborne and Sassic 1979; Sutton and Vandiver 1986). In early experiments, grass carp had successfully eliminated submersed vegetation from Lakes Baldwin, Pearl (Shireman et al. 1985; Colle and Shireman 1994) Bell, Clear, and Holden (Leslie et al. 1987), while partially removing vegetation from Lakes Conway (Leslie et al. 1987) and Deer Point (Van Dyke 1994). Based on these experiments, grass carp proved to be an effective biological control agent and a more economical approach to aquatic weed control than herbicides and mechanical methods (Leslie et al. 1987).

Because of concerns of its reproduction and potentially negative impact on sportfish habitat, the grass carp was limited to experimental use in Florida from 1970 to 1984 (Sutton and Vandiver 1986). A solution to the grass carp reproduction problem was obtained when Cassani and Caton (1985) produced a triploid grass carp by exposing fertilized eggs to heat, cold or hydrostatic shock. The result of this process was virtually a sterile, triploid fish that retained the same plant-eating capabilities as the diploid grass carp (Clugston and Shireman 1987; Cassani and Caton 1985).

The controversial issue pertaining to the possible negative impacts that can occur to fisheries and water quality with the elimination of submersed macrophytes, however, still remained. The concerns stemmed from the fact that there was no standard stocking formula for triploid grass carp (Hanlon et al. 2000; Mallison et al. 1994b). Low stocking rates would often lead to insufficient control of vegetation. Higher rates often resulted in the removal of all vegetation. Leslie et al. (1987) reported that stocking rates of more than 10 triploid grass carp per metric ton of vegetation resulted in elimination of vegetation in Florida lakes. Higher stocking densities of triploid grass carp, however, can often lead to elimination of both undesirable and desirable vegetation, while increasing chlorophyll concentrations (Shireman et al. 1984; Leslie et al. 1987; Hanlon et al. 1989). The elimination of vegetation and subsequent increase in phytoplankton populations may be perceived by user groups, such as anglers, as having a negative effect on the sport fishery (Wrenn et al 1994).

In many cases, lake managers and user groups want some submersed vegetation because of its associated benefits. For example, in Chapter 2, largemouth bass anglers had significantly higher effort in lakes with moderate vegetation (40-59%) than lakes with little to none (0-19%). Supplemental stocking of triploid grass carp can be implemented to increase the amount of vegetation removed. However, grass carp have to be removed to reduce grazing pressure once the desired level of vegetation control has been achieved. Studies have shown that active and passive gears normally used by biologists are ineffective in removing grass carp (Schramm and Jirka 1986; Hestand et al.

1987; Bonar et al. 1993; Mallison et al. 1994b). Other studies have found that the use of fish management baits or herding of grass carp to be a little more effective than traditional methods (Bonar et al. 1993; Mallison et al. 1994a), but herding may be less efficient, whereas, the use of fish management baits may face opposition due to the by-kill of non-targeted fish.

Maniwa (1975) used sound to attract schooling marine fishes off the ocean floor for harvesting purposes. Other sound studies have found that many fish are attracted to their own feeding sounds and in some cases to other sounds (Hashimoto and Maniwa 1966). Willis (2000) found that triploid grass carp could be conditioned to respond to their own feeding sound and low frequency sounds in tanks. Therefore, my objective was to determine the effectiveness of attracting and then removing grass carp from a Florida pond with the aid of sound and food.

Methods

Study Site

A single, 0.1-ha pond located at the University of Florida's Department of Fisheries and Aquatic Sciences facility in Gainesville, Florida was used in this study. The pond had a dock extending out approximately three meters from the center of the north bank and an aerator in the center of the pond. The bottom was composed of a thick, flocculant sediment overlying clay. There was no submersed vegetation. Emergent vegetation was comprised of pennywort (*Hydrocotyle spp*.), torpedo grass (*Panicum repens*), alligator weed (*Alternanthera philoxeroides*), cattail (*Typha spp*.), and primrose (*Ludwigia spp*.). This pond was selected because it lacked submersed vegetation, which would optimize the probability of sound and food being successful in this experiment.

Protocols

I initially drained and seined the pond to remove and transfer any live fish that were in the pond. When catch rates were drastically reduced, rotenone was applied at 1 mg/L to remove any remaining fish. The pond was refilled and sufficient time was allowed for the degradation of the rotenone before any grass carp were stocked.

Floats from an old blocknet were placed into the corners of the pond to delineate feeding areas where the triploid grass carp were to respond. The floats extended along the surface from the midpoint of each side of the pond to the midpoint of the adjoining sides, which made each of the feeding areas 1/8 of the pond area.

Once the setup was complete, 103 fingerling triploid grass carp were stocked into the pond in February 2000. Total length (TL) of the stocked grass carp ranged from 13.1 to 27.5 cm (mean TL = 19.9 cm) and weights ranged from 28 to 220 g (mean weight = 96 g). Prior to the initiation of the training, the grass carp were given several weeks to get acclimated to their new environment. This also provided them with sufficient time to become conditioned to eating floating catfish food (the reward) which was tossed on to the surface of the pond.

Fish training, conducted in the pond, was the operant conditioning protocol used by Willis (2000). Operant conditioning is a training technique in which desired responses are rewarded. Sound, with food rewards, was used to get the grass carp to respond to a desired area in the pond.

A Crass noise simulator (model S44) was used to produce a series of pure-tone low-frequency (600 hz) bursts that were three seconds long, separated by one-second pauses. These bursts (approximately 25) were recorded from the Crass noise simulator onto a cassette tape using a tape recorder/player (Realistic model CTR-100). The taped sound was played by the same tape recorder/player, amplified by an 80-watt amplifier (Realistic model 12-1870A) and transmitted through the water of the pond by a J-9 transducer (an underwater speaker; Naval Undersea Warfare Center, Newport, Rhode Island). Within the pond, sound transmission was checked at various distances from the transducer using a hydrophone (LC-10 Model, Celesco Ind., Inc.) connected to a receiver with headphones to ensure detection by the grass carp. The sound became weaker as the distance from the transducer increased, but it was detectable at the furthest, opposing corner.

Each day a session was conducted in which the food pellets were tossed, sound was produced and the response time of the triploid grass carp was documented. After a session was conducted, the transducer was moved to another corner for the next day's session. At the beginning of the experiment, food pellets were tossed first and 20 seconds later the sound was transmitted (stage 1). When the response times of the grass carp to the food pellets and sound quickened, the time between food release and sound transmission was reduced to 10 seconds (stage 2), until both were given at the same time (stage 3). Sound in the first two stages was used to get the grass carp to associate sound with food. Once the association between sound and food strengthened, the purpose of sound was to quicken response times by signaling when and where the food would be thrown. The length of each stage varied from a week to a month, depending on the grass carp response times. Because of the reduced number offish observed feeding at later stages, an additional 149-fingerling triploid grass carp were stocked in June 2000. The sound and food sessions and stages were repeated until response times became quicker.

Once the triploid grass carp showed quicker responses to the sound and food pellets, a trap area was set up in the northeast corner of the pond. The trap area consisted of a 6-mm bar mesh blocknet attached to wooden posts that extended across corner of the pond in a similar fashion as the floats used during the training sessions. Between two of the posts, which were off-centered to the right when facing the southwest corner of the pond, a 0.91-m x 2.44-m trap door was constructed using 1.91-cm PVC pipes, a 1.2-m piece of blocknet, steel rings, nylon rope, and a pulley. The trap door was held on the bottom of the pond by the weight of the steel rings and pulled up during the experiment.

The J-9 transducer was placed into the northeast corner of the pond with the front of the transducer facing the southwest corner of the pond. It was placed approximately 1 m from the north bank and 3 m from the east bank of the pond so that it was completely submersed. This allowed the transducer to maintain the same temperature as the surrounding water, which prevented the occurrence of air bubbles and reduction of the transmission distance.

Sound and food rewards, given at the same time, were used to lure the triploid grass carp into the netted area. After the carp were observed feeding inside the trap area, the trap door was pulled up, confining the feeding fish. While confined in the trap area, a 6-mm bar mesh seine, that was approximately 8 m long, was pulled through the enclosed area several times to remove the grass carp. Captured fish were transported to 950-L fiberglass tanks for holding until all removal trials were completed. Fish in the holding tanks were given a different food supplement, Silvercup (sinking food pellets with 40% crude protein), due to the floating catfish feed draining out of the standpipes. Also, no sound was produced prior to feeding the grass carp in the holding tanks.

When grass carp ceased to respond to the sound and food rewards, the pond was drained and seined to determine the number of grass carp remaining in the pond. Seined fish were counted and placed into aerated hauling boxes until no more grass carp were seined from the pond. Once all grass carp were removed, they were immediately put back into the pond while it was being refilled. Fish in the holding tanks were given a partial right-pectoral-fin clip and then placed into the same pond on the day following the pond draining and seining. The removal process was repeated a day or two later to allow the grass carp to recuperate from any stress related to handling.

Temperature and dissolved oxygen levels were monitored daily using a YSI (model 57) oxygen meter due to their effects on grass carp feeding behavior.

Analyses of Grass Carp Removal

The effectiveness of this removal method was assessed based on the percent of grass carp removed. The percentage of carp removed was determined by the following equation:

% Removed = No. Caught in Trap Area / (No. Caught in Trap Area + No. Remaining in Pond).

Results and Discussion

Triploid grass carp removal percentages ranged from 61% during the first trial to 32% during the third trial (Table 3-1). Although these results range widely, they are within the range (4 to 79%) of removal rates obtained during the fish management bait study conducted by Mallison et al. (1994a). The use offish management baits or sound and food rewards seem to be effective for large-scale removal of triploid grass carp, but the use offish management bait may face scrutiny related to by-kill of non-targeted sportfish species.

During the first trial, I believe more fish should have been caught. Seventy-eight of the fish taken in the first trial were captured in the first seine pull. The remaining four removal attempts yielded only one grass carp, though many fish were observed feeding in the enclosed area. Inspection of the blocknet barrier indicated that many grass carp probably escaped through a 0.3-m x 1.0-m gap between the bottom of the net and pond bottom located near the trap door. This problem was corrected during the first pond draining and prior to trial two.

The percentage of triploid grass carp removal declined during trials two and three but increased during trial four. One possible explanation is water temperature. Trials one and two were conducted between the middle of August 2000 and late October 2000 when pond temperatures ranged from 20 to 31.5 C. Based on the findings of Clugston and Shireman (1987), these trials were conducted within the optimum feeding range of triploid grass carp (20-26 C). Trial three was conducted during December 2000 when pond temperatures ranged between 12.5 and 20.5 C, well below optimum foraging temperatures. Because the observed aggressive feeding behavior of the grass carp declined, trial four did not begin until March 2001 when the water temperature approached 20 C. However, the reason for the decline of carp removal in trial two is unknown.

The decline in percent removal of triploid grass carp may also be related to the subsequent decline in carp density. At the higher densities (>1200 grass carp/ha) larger percentages of grass carp were removed than at lower densities (<1100 grass carp/ha). A similar relationship was found by Mallison et al. (1994a). At higher densities, there was less food available per fish, which potentially resulted in more aggressive feeding.

Constant handling of the grass carp might have deterred the grass carp from responding in subsequent retrials. For example, Mallison et al. (1994a) found that previously tested triploid grass carp that survived ingestion of rotenone pellets avoided the fish management bait in later trials. Also, the grass carp in my holding tanks were given sinking food pellets with no sound. This could have possibly weakened the association between sound and food rewards and therefore, decreased the chances of recapture in subsequent retrials. But, based on the partial right pectoral fin clips of the fish captured in trial one, 49% of the fin-clipped fish were recaptured during trial two, and 72% (38 of 53) of all fish caught in trial two were recaptures. In later trials, it became increasingly difficult to recognize fin-clipped fish, but 11 of 33 fish (33%) in trial three were believed to be recaptures based on structural differences in the right pectoral fins. These results suggest that handling of the grass carp and the lack of sound reinforcement in the holding tanks had negligible impact on recapturing the fish.

Mortality of the grass carp in this study was high considering the lack of predatory fishes in either the pond or holding tanks. Low dissolved oxygen during August and September 2000, resulting from aeration failure and handling stress related to draining and seining events accounted for 20 carp deaths. Eleven grass carp were killed or removed from the holding tank by birds or mammals (e.g., bobcats *Lynx rufus*). Bald eagles (*Haliaeetus leucocephalus*) and ospreys (*Pandion haliaetus*) took at least eight fish. Other wildlife predators present included the great blue heron (*Ardea herodias*), little blue heron (*Egretta caemled*), great egret (*Casmerodius albus*), double-crested cormorant (*Phalacrocorax auritas*), Florida softshell turtle (*Trionyx ferox*), and American alligator (*Alligator mississippiensis*). Ross and Johnson (1999) found that double-crested cormorant predation on fingerling lake trout (*Salvelinus namaycush*) was as high as 13.7% following a stocking event and cited other authors where depredation rates by cormorants were as high as 50% shortly after release offish. Thus, cormorants could have consumed many of the fingerling grass carp shortly after both stocking events.

This study has shown that a proportion of triploid grass carp may be removed from a 0.1-ha pond using low frequency sound and food rewards. However, in this study, the same fish and a single pond were used throughout the experiment, thus I lacked the true replication of my treatments necessary for statistical validation. Replication of this sort has been termed 'pseudoreplication' by Hurlbert (1984). However, the removal percentages were within the range reported by Mallison et al. (1994a), yet more effective than large-scale removal attempts reported using other methods (Schramm and Jirka 1986; Hestand et al. 1987; Bonar et al. 1993; Mallison et al. 1994b). So, if statistical validation is desired, additional studies are warranted, but the evidence available suggests the use of sound and food rewards should be field tested on a small lake.

Another concern is whether the grass carp will prefer the food reward hi the presence of desirable vegetation. Though my pond lacked submersed vegetation preferred by grass carp, an adjacent pond, located at the same facility, gave me insight to this question. The pond (0.1 ha) is one of two ponds used for sturgeon research, but was densely infested with hydrilla. At least 16 triploid grass carp had been stocked (160 fish/ha). Observations indicated the grass carp were eating Silvercup, a high-protein, sinking food pellet, intended for the sturgeon while the hydrilla continued to spread.

Therefore, I believe this removal method of using sound and food rewards to lure grass carp into an entrapment area can be effective in systems with submersed vegetation.

The amount of time that the triploid grass carp retain the association between sound and food rewards is another concern. Retention times would have an impact on stocking densities. The shorter the retention time, the higher the stocking density required due to the need for quicker vegetation consumption and carp removal. Thus, it would increase the cost for vegetation control. Conversely, longer retention times allow fewer carp to be stocked because vegetation consumption time and grass carp removal can be prolonged. Lower stocking densities would decrease the cost of this removal technique. Such a study again would be warranted, but the grass carp retained the association between sound and food long enough (about 9 months) not to preclude a larger field test.

Based on the results of this study, future experiments should determine the feasibility of this removal technique using sound and food rewards in small, vegetated lakes. Fingerling triploid grass carp could be trained to associate sound with the food rewards at the hatchery or in an isolated pond until the desired response times and/or grass carp size are attained. The trained fish could then be stocked into the lake. Once the desired level of vegetation control is achieved, sound and food rewards should be used to lure the carp into an entrapment area or device for removal from the lake.

Management Considerations

This study demonstrated the use of low frequency sound and food rewards were effective aids in removing up to 61% of the stocked triploid grass carp. Using the protocol by Willis (2000), triploid grass carp were lured into a trap area throughout the experiment. Because many fish were recaptured in subsequent trials, handling stress would probably have negligible effects on using the same fish in other systems.

Temperature and stocking density effect the feeding behavior of grass carp. Grass carp are more likely to be more aggressive feeders during warmer temperatures and at higher densities. The size of the grass carp stocked will effect stocking densities because small grass carp (< 250 mm TL) are more susceptible to predation. Smaller fish should be stocked at higher densities to compensate for predation and achieve the desired level of macrophyte control. Therefore, it may be more cost-effective to stock grass carp ranging in size between 300 and 350 mm TL because it would reduce fish loosed to predation (Shireman et al 1978).

The future use of grass carp as long-term biological control agents depends on an efficient and economically feasible method of removal. The use of positive reinforcement may be the best option. The next step should be to train grass carp in small ponds and then move them to a small, vegetated lake to determine the feasibility of this technique in an operational situation. The size of the water body and the distribution of dense vegetation areas may dictate the number and locations of sound and trap setup areas. In lakes, grass carp have been known to establish home ranges, which are usually centered around dense vegetation mats (Clapp et al. 1994; Mallison et al. 1994a). Such cases may require the use of more power and/or an increase in pulse length to increase transmission distance, moving the sound source and trap area around, or the use of multiple sound and trap devices.

In this study, seining was the removal method of choice. I felt this minimized stress on the fish and increased the possibility of using them again. Coves and pond corners would make good entrapment areas since the shoreline would provide most of the containment boundary. However, lake morphology and bottom debris in many circumstances may not permit the use of an entrapment area or seining as the removal method. Cages may be the best option. Lake managers should take these factors into consideration when planning to stock and remove grass carp from a lake.

Table 3-1. Percent of triple id grass carp removed from the study pond (0.1 ha). "Total fish" represents the number removed from the trap area plus the number remaining in the pond at the end of each trial.

Trial Number	Removed from Trap Area	Total Fish	Percentage
1	79	129*	61.2
2	53	128	41.4
3	33	102	32.4
4	22	56 **	39.3

* My total fish estimate was 111 fish, however there had to be at least 129 triploid grass carp present based on trial two and the death of one carp in the holding tanks.

** I removed a total of 56 carp but several were observed in the pond after refilling it.