PROCEEDINGS OF THE GRASS CARP CONFERENCE

Ĩ

Gainesville, Florida

January, 1978

Editor

Jerome V. Shireman

University of Florida

Institute of Food and Agricultural Sciences

PREFACE

Weed infestations of freshwaters in the United States, particularly the southern states, have increased tremendously in the past 10 to 20 years. This is especially true of Florida, where exotic plants such as hydrilla (<u>Hydrilla verticillata</u>) have infested many water bodies. This submerged plant is established in every major watershed in Florida and has spread from Florida through at least eight states into California. Hydrilla is especially troublesome because control methods are costly.

Chemical weed control programs are presently the most effective control measure; however, the cost of herbicides is increasing and restrictions are becoming more stringent. It is evident that even though nuisance aquatic weeds are increasing, we will, by necessity, use less chemicals for weed control in the future.

In order to reduce chemical needs and achieve long-term control, biological control organisms are being investigated. The most promising of these organisms appears to be the grass carp (<u>Ctenopharyngodon idella</u>), for it consumes vast quantities of submerged vegetation; however, the environmental impact of this species on sport fish populations is not well documented.

This symposium was organized to bring researchers from various parts of the world together to discuss the role of grass carp as a weed control organism. The papers contained in these proceedings should provide valuable information and insight into future research needs.

ii

ACKNOWLEDGMENTS

This conference on the grass carp (<u>Ctenopharyngodon idella</u>) came into being because of the foresight, interest and work of a great many people whose participation in the session discussions significantly contributed to the broader knowledge on the topic.

Special credit goes to the authors and session moderators. Many of the authors traveled long distances, bringing to the conference expertise, enthusiasm and information from around the world. Without their contributions there would have been no conference of the caliber achieved.

Funding for the conference was provided by the Institute of Food and Agricultural Sciences (IFAS) of the University of Florida. This funding was made possible largely by the interest and leadership of Dr. K.R. Tefertiller, Vice-President for Agricultural Affairs.

Dr. Robert Q. Marston, President of the University insured the success of the conference by his personal invitation to the distinguished scholars and leaders who participated as speakers and contributed papers.

Dr. Milton Morris, Chairman of the IFAS Editorial Department, receives special credit for his personal effort in planning the conference, providing logistical support, and organizing the proceedings. His assistance and support was continuous and unflagging throughout the entire course of the conference and publication of the proceedings.

Editorial assistance was provided by Janos Z. Shoemyen of the IFAS Editorial Department.

I owe a personal debt to Drs. Haller, Shireman and Sutton who not only contributed papers to the conference but assisted in planning the program, selecting topics, organizing the papers and conducting much of the mundane but necessary business of the conference. In addition, Dr. Shireman served as editor of these proceedings.

> John F. Gerber Program Chairman

i

TABLE OF CONTENTS

Page
Acknowledgments
Preface
Aquatic Weeds in Florida William T. Haller
The Use of Grass Carp in Comparison with Other Aquatic Weed Control Methods J.C.J. von Zon
Grass Carp: The Scientific and Policy Issues Scott Henderson
Research and Management Application of Grass Carp in Iowa Larry Mitzner
Ecological Study of Lake Wales, Florida After Introduction of Grass Carp (<u>Ctenopharyngodon idella</u>) Jerome V. Shireman, Douglas E. Colle, Randy G. Martin 49
Use of the Grass Carp for Control of Hydrilla in Small Ponds David L. Sutton, V.V. Vandiver, R.S. Hestand, W.W. Miley 91
Weed Control and Fish Production Karol Opuszynski
The Role of Herbivorous Fishes at Reconstruction of Ichthyofauna Under The Conditions of Anthropogenic Evolution of Waterbodies B.V. Verigin
Grass Carp Research and Public Policy in England Brian Stott
The Use of Grass Carp for Biocontrol of Aquatic Weeds and Their Implication for Natural Resources and Fisheries in Florida W.W. Miley, J.M. Van Dyke, Dennis M. Riley
The Role and Impact of Introduced Grass Carp (<u>Ctenopharyngodon idella</u> Val.) in the Union of Soviet Socialist Republics and Several Other European Countries W.W. Miley, D.L. Sutton, J.G. Stanley
Natural Reproduction of Grass Carp in the Tone River and Their Pond Spawning Minoro Tsuchiya

	Control of Sex in Fishes, With Special Reference to the Grass Carp Jon G. Stanley
у	Disease Control and Reproduction of Grass Carp in Germany Martin Bohl
	Summary and Conclusions Jerome V. Shireman

!,

•

AQUATIC WEEDS IN FLORIDA

William T. Haller

Aquatic Weeds Research Center

University of Florida

Gainesville, Florida 32611

INTRODUCTION

The topography and semi tropical climate of Florida (latitude 25-31°N) are particularly conducive to the growth and proliferation of aquatic weeds. Warm temperatures result in plant growth nearly all year over much of Florida, and underlying phosphate and carbonate rock provides an abundance of naturally-occurring plant nutrients. Lakes in Florida are characteristically very shallow and contain large littoral areas. South Florida has extensive canal systems designed to control flood waters and allow agricultural use of the productive organic soil around Lake Okeechobee. In general, the majority of the one million hectares of fresh water is composed of fertile, shallow lakes and man-made canals.

The large proportion of littoral areas in Florida lakes provide a large forage base for largemouth bass [Micropterus salmoides floridanus (Lacepede)] and other sport fishes (Barnett and Schneider 1974). Freshwater recreational fishing has been estimated to consist of over 400 million dollars annually, including boat purchases, motors, bait, tackle, and other items. In the 10 year period (1967 to 1977), registration of recreational power boats has nearly tripled from 149,663 boats in 1967 to 422,398 in 1977 (Cato and Mathis 1979). In 1977, 1.33 million Florida residents purchased fishing licenses (or combined hunting and fishing licenses), providing a total of over one million dollars in annual license fees. In addition, many tourists visit Florida for the purpose of recreational fishing. In 1977, 192 thousand nonresident fishing licenses were purchased, bring the state an additional 1.3 million dollars in license fees.

Sport fishermen are not the only beneficiaries of clean Florida waters. Although it is not an extensive industry, a few commercial fishermen derive their livelihood from Florida's fresh waters. By far, the major value of water in Florida is derived from the aesthetics of living near water and the resultant increased land values, and recreational boating, swimming, and related water activities. Irrigation canals permit large-scale agricultural production of sugarcane, vegetables, and citrus, and drainage canals are essential for flood control. In summary, an abundance of clean fresh water is important to Florida's economy. Aquatic plants contribute to productive fish and wildlife populations and concurrently often interfere with navigation, flood control, and other water uses.

AQUATIC WEEDS

Although plants are important to the aquatic environment, an overabundance of plants becomes a hindrance to water activities. Native aquatic plants (<u>Ceratophyllum</u>, <u>Najas</u>, <u>Typha</u>, <u>Pontederia</u>, etc.) occasionally grow excessively and adversely affect water use. However, natural controls such as water level changes, weather extremes, turbidity, or other natural factors frequently maintain native plants at a desirable or tolerable level. If control or management are required, a single herbicide treatment often provides control for two to three years. Currently, the most serious weeds in Florida are the introduced or exotic species such as waterhyacinth (<u>Eichhornia crassipes</u> (Mart.) Solms) and hydrilla (Hydrilla verticillata Royle).

Waterhyacinth was introduced into Florida in the late 1800's and spread rapidly, causing navigation problems in the St. John's River within 10 years after introduction (Zeiger 1962). Hydrilla is a submersed plant which was first found growing in Florida in 1960. For several years it was sold as an aquarium plant and was also spread throughout Florida by boats and trailers (Haller 1976). Hydrilla has a lower light requirement than native submersed plants and consequently grows in deeper water where native plants cannot survive (Van et al. 1976). The vast, diverse littoral zone of Florida lakes, which is so important to sportfish production, is an ideal habitat for waterhyacinth and hydrilla. Hydrilla, because of its lower light requirement, can grow in deeper water than native species and often occupies 80 to 90% of the surface area of many lakes. This profuse growth in effect, increases the littoral zone dramatically, and hinders navigation, flood control, recreational use of water, and can adversely affect sportfish populations (Bennett 1948).

Waterhyacinths are currently under control throughout most of Florida as a result of intensive maintenance programs. These programs rely on the use of herbicides (primarily 2,4-D and diquat) and maintain small fringes of waterhyacinths which are not allowed to grow to troublesome extent. Recent data indicates that on a long-term basis, maintenance control programs result in the use of less herbicide and less detrital or sediment formation (Haller, Univ. of Florida, unpublished data). In order to further reduce the use of chemical control three species of insects (<u>Neochetina brucchi</u> Hustache, <u>Neochetina eichhorniae</u> Warner, and <u>Sameoides albiguttalis</u> Warren) have been released in Florida to retard the growth of waterhyacinths (Zeiger 1979). In addition, a fungal disease is being studied as an additional means of controlling waterhyacinth. Through a combination of effective and relatively inexpensive chemical control and the introduction and development of biocontrol agents, waterhyacinths are currently managed and kept under control.

Hydrilla, on the contrary, is spreading throughout Florida and the United States, and presently there are no methods that are economically feasible to stop its spread. From its initial discovery in Florida in 1960 (Figure 1), it spread to several areas by 1967 (Figure 2), covering an estimated 2,000 ha. In 1977 (Figure 3), hydrilla was present in virtually every major watershed in Florida and was commonly found in about 25% of Florida's fresh waters (250,000 ha). Essentially all waters in the state are threatened by this competitive submersed species. Hydrilla is now present in Alabama, Mississippi, Georgia, South Carolina, Louisiana, Texas, California and Iowa (Figure 4).

There are two major reasons why hydrilla cannot be effectively controlled. Primarily, hydrilla is a submersed species and grows under water where dilution of herbicides requires higher application rates. Hydrilla, therefore, is virtually impossible to control chemically in flowing water. The second reason is the profuse vegetative reproduction and regrowth of hydrilla (Haller 1976). Hydrilla produces underground tubers which are unaffected by control methods and remain viable for many years (Miller et al. 1975). Chemical treatments essentially shear and kill most of the plant biomass in the water column, leaving viable plant crowns, rhizomes, stolons, and plant fragments on the bottom as a source of re-infestation. The rate of regrowth depends upon local conditions of turbidity, water temperature, and etc., but often three chemical treatments a year are required for satisfactory hydrilla control in central Florida.

Ditchbank, emergent, and semi aquatic plants which grow in the transition zone on pond berms and canal banks are also a major problem, particularly in agricultural canals. These plants significantly reduce water flow, provide habitat for mosquitoes, and periodically require control as a part of canal and ditch management.

COST OF AQUATIC WEED CONTROL

It is virtually impossible to determine the precise costs of weeds to society due to the intangible values placed on recreation, aesthetics, health problems, and other values related to weed growth. The economic impact of aquatic weeds is particularly difficult to assess, and likewise, the level of funds to be expended for an economically productive aquatic weed control program cannot be accurately determined. For example, as a lake becomes overcome with a submersed weed, recreational boating, fishing and swimming are greatly reduced. In the immediate lake area, boat sales, gasoline, fishing supplies, food and beverages and a myriad of other items are reduced. The question which arises is, "How much funds should be expended to sufficiently control weeds in the lake to provide an economic return on the weed control effort?" The answer is different for each body of water and is unknown totally.

An estimate of the funds expended for aquatic weed control in Florida has been compiled for 1977 (Table 1). These values combine the cost of all control methods, however, approximately 70% of aquatic weed control is accomplished with herbicides. The remaining 30% is by mechanical means, the majority of which is spent for mowing, cleaning canals and mechanical control of ditchbank vegetation. Submersed weeds are mechanically harvested or mowed in some canals and lakes, but this has not been extensively practiced. More modern, efficient equipment is being developed, however, and mechanical harvesting of hydrilla is becoming more economically feasible in some situations (McGehee 1979). Mechanical control of waterhyacinth is seldom used due to the bulk (up to 150 m.t./ha) and high cost of physical removal. The type of control used in Florida is dictated by economics, or simply the cost of control per hectare per year, and environmental safety. Herbicides are available which are more effective and less expensive than those currently used in aquatic weed control. However, environmental safeguards require that aquatic herbicides have low toxicities and be rapidly biodegradable.

The data presented in Table 1 is an approximation of funds expended for aquatic weed control. Data from public agencies is accurate, however, it is entirely possible that monies spent by the private sector has been underestimated. The \$20,810,000 spent in 1977 is best described as a conservative estimate, due largely to the fact that many large (2,000 ha and larger) citrus, vegetable, and sugarcane farms implement their own aquatic weed control programs in irrigation and drainage canals.

Surprisingly, ditchbank weed control is a very significant problem in relation to other weed problems, and there are no major research programs in progress to study the chemical, mechanical, or biological control of these weeds. Individually, ditchbank weeds require little control, however, collectively, they total a significant 8 million dollars.

Expenditures for ditchbank weeds and waterhyacinth have remained stable over the past 10 years. The rapid expansion of hydrilla has nearly doubled the weed control budget and has brought tremendous attention to aquatic weed control problems.

CHEMICAL CONTROL

There are 113 trade named herbicides registered for use in or near Florida waters. Most of these are formulations of only 8 to 10 active ingredients. For example, 2,4 dichlorophenoxy acetic acid (dimethyamine formulation) is primarily used in waterhyacinth control. In 1977, 304 kl (80,000 gallons) of $2,4-D^{1/}$ was used by public (state, local and federal) agencies in waterhyacinth control programs. The cost of waterhyacinth chemical control programs was estimated to be \$95/ha in 1977, which was divided roughly into three portions; one-third for chemical purchases, one-third for equipment, and one-third for labor expenses.

Hydrilla is the predominant submersed weed requiring chemical control in Florida. Approximately 15,000 ha of hydrilla were treated chemically in 1977 by public agencies. The cost of hydrilla control varies greatly depending upon the herbicide selected, site, density of infestation, etc. Basically, the costs vary between \$190 to \$625/ha with an average expense including labor, equipment and chemicals of approximately \$500 per hectare per treatment. Three herbicides are primarily used; diquat in combination with copper (ratio of 19 1 of diquat: to 38 1 of liquid chelated copper/ha) or 38 to 52 1 of endothall per hectare. In 1977, public agencies used approximately 190 kl (50,000 gallons) of diquat in submersed weed and hyacinth control, 266 kl (70,000 gallons) of copper compounds, and 190 kl (50,000 gallons) of endothall based compounds.

Ditchbank and emergent chemical weed control involves the use of many herbicides but the primary ones used are dalapon (23,000 kg of formulation) on monocots, and various phenoxy compounds on dicots (10- to 15,000 kg formulation). The cost of chemical control of ditchbank weeds is quite variable, but \$190 to \$225/ha is a valid estimate.

^{1/} The active ingredient content of the commonly used formulations are: 2,4-D - 50%, diquat - 35%, liquid copper complexes - 8 to 9%, and endothall 40 - 50% active ingredients.

BIOLOGICAL CONTROL

It is apparent that of the aquatic weeds in Florida, hydrilla is spreading most rapidly and is by far the most expensive to control. The prospects for development of new, inexpensive herbicides or more efficient mechanical harvesting equipment within the immediate future are not great. Consequently, research has to formulate more effective weed control programs with current technology or develop new, alternative control methods.

The discovery and successful widespread use of the alligatorweed flea beetle (<u>Agasicles hygrophila</u> Selman and Vogt) for control of alligatorweed (<u>Alternanthera philoxeroides</u> (Mart.) Griseb.) in the 1960's has provided impetus to study biological control of other aquatic plants (Maddox et al. 1971, Spencer and Coulson 1976). Three insects have already been studied and released on waterhyacinths in the United States, and a host specific fungal pathogen (<u>Cercospera rodmanii</u> Conway) is currently being developed (Freeman 1977, Charudattan 1979).

There are literally hundreds of thousands of insect species in the world, however relatively very few inhabit the submersed aquatic habitat where hydrilla thrives. The search for insect biocontrols for hydrilla has not yet yielded any significant insect candidates (Zeiger, personal communication). Recently, a fungal pathogen (<u>Fusarium culmorum</u>) was isolated from plants from Holland and is most promising for hydrilla control. Extensive field studies are planned beginning in 1979 (Charudattan 1979).

Biological control studies of hydrilla with snails (<u>Marisa cornuaretis</u> L.) has been extensively studied in Florida. It was found that <u>Marisa</u> was not a significant biological control because it was temperature sensitive and very high stocking densities were required (Blackburn, Taylor and Sutton 1971).

Several species of fish have been considered as candidates for the biological control of submersed aquatic weeds (Blackburn, Sutton and Taylor 1971, Legner et al. 1975). The species receiving the most attention for hydrilla control in Florida is the Chinese Grass Carp or White Amur (<u>Ctenopharyngodon</u> idella Val.).

The first field research in Florida was initiated in three small (0.08 ha) earthen ponds in central Florida in 1971. This nonreplicated study (completed in January, 1973) showed that the grass carp at stocking rates of 50 fish/ha was capable of controlling hydrilla without catastrophic effects on the aquatic environment (Haller and Sutton 1976).

Further major research was undertaken by the Florida Department of Natural Resources and the Florida Game and Fresh Water Fish Commission in 1972 (4 pond study). Four natural ponds in widely scattered geographical locations were stocked with grass carp after a one year collection of baseline data. The diverse interpretation of the results of these nonreplicated studies has become widely known by the world's fishery scientists (Beach et al. 1976, Gasaway et al. 1978).

In 1974, six lakes (over 50 ha each) and one reservoir (2,000 ha) were stocked with grass carp to further determine their weed control capabilities and potential environmental impact. This research is still incomplete as three of the lakes have become entirely weed free and three other lakes still contain hydrilla infestations. In one lake, at least, the apparent lack of biocontrol resulted from low residual grass carp populations (Colle et al. 1978). Restocking programs have begun on the remaining vegetated lakes and the reservoir.

Due to the unpredictable results obtained with grass carp, and the unanswered questions concerning its possible impact on sport fish populations, there remains considerable controversy among Florida biologists with regard to the widespread use of the fish in hydrilla control programs. The purpose of this conference is to share our knowledge with the world's experts in the culture, production and biology of the grass carp. The most important questions that arise in the use of the grass carp in Florida are:

- 1. Will the grass carp reproduce under natural conditions in Florida, and if they do, what are the likely environmental consequences?
- 2. Can the grass carp be "managed" so that it controls only hydrilla or a portion of the hydrilla in the littoral zone, or will it remove all vegetation from a lake, including emergent plants?
- 3. Native fish populations in Florida are often found to be nearly 200 to 300 kg per hectare. What is the effect of the addition of twenty- 10 kg grass carp or a doubling of the fish biomass going to have on the carrying capacity of the native fisheries?
- 4. Finally, how will aquatic weed control by grass carp affect phytoplankton, nutrient cycling, and other aspects of water quality?

Currently, the State of Florida allows private possession of the grass carp by individuals with weed problems in lakes of 10 ha or less which are not connected to other water bodies. The rules (16 C-21) allow stocking by permit only in private waters which meet specific criteria (size, weed problems, lack of infall or outfall). Thus, the grass carp is currently in use in golf course ponds, fishery ponds, and waters of similar nature.

The widespread application of the grass carp to solve hydrilla problems in large lakes has been deferred until further studies are conducted. The problem is, that hydrilla continues to spread and cause problems, current control measures are expensive, and the answers to the four questions posed earlier may take several years to answer.

ACKNOWLEDGMENTS

The author gratefully acknowledges the assistance of Bill Baier who compiled most of the information in Table 1 from Legislative Reports and matching fund applications.

LITERATURE CITED

Barnett, Brian S. and R.W. Schneider. 1974. Fish populations in dense submersed aquatic plant communities. Hyacinth Cont. J. 12:12-14.

- Beach, M.L., W.E. Miley, II, T.M. van Dyky, and D.L. Riley. 1976. The effect of the Chinese grass carp on the ecology of four Florida lakes, and its use for aquatic weed control. Final Project Report, Florida Department of Natural Resources, Tallahassee, FL. 246 pp.
- Bennett, G.W. 1948. The bass-bluegill combination in a small artificial lake. Ill. Nat. Hist. Surv. Bull. 24(3):377-412.
- Blackburn, R.D., D.L. Sutton and T. Taylor. 1971. Biological control of weeds. 421-432 pp. In J. Irrigation and Drainage Division, Proc. Amer. Soc. Civil Engineers. Vol. 97, No. IR3.
 - , T.M. Taylor and D.L. Sutton. 1971. Temperature tolerance and necessary stocking rates of <u>Marisa cornuaretis</u> L. for aquatic weed control. Proc. European Weed Res. Conf. 3rd Int. Symp. Aquatic Weed. 79-85.
- Cato, J.C. and K. Mathis. 1979. The Florida Fleet: For Business and Pleasure. Florida Food and Resource Economics Newsletter No. 28. University of Florida, Gainesville, FL. 4 pp.
- Charudattan, R. 1979. Aquatic Weed Control Newsletter. Vol. 4, No. 4. University of Florida, Gainesville, FL. 4 pp.
- Colle, D.E., J.V. Shireman, R.D. Gasaway, R.L. Stetler and W.T. Haller. 1978. Utilization of selective removal of grass carp (<u>Ctenopharyngodon idella</u>) from an 80-hectare Florida lake to obtain a population estimate. Trans. Amer. Fish. Soc. 107(5):724-729.
- Freeman, T.E. 1977. Biological control of aquatic weeds with plant pathogens. Aquat. Bot. 3:175-184.
- Gasaway, R.D. and T.F. Drda. 1978. Effects of grass carp introduction on macrophytic vegetation and chlorophyll content of phytoplankton in four Florida lakes. Fla. Sci. 41:101-109.
- Haller, W.T. 1976. Hydrilla, A Rapidly Spreading Aquatic Weed Problem. Florida Agric. Exp. Stat. Circular S-245. 13 pp.

, and D.L. Sutton. 1976. Biocontrol of aquatic plant growth in earthen ponds by the white amur (<u>Ctenopharyngodon idella</u> Val.). Proc. 4th Int. Symp. Biological Control of Weeds. pp. 261-268.

Legner, E.F., W.J. Hauser, T.W. Fisher and R.A. Medued. 1975. Biological aquatic weed control by fish in the lower Sonoran desert of California. Calif. Agric., Nov., 1975. 8-10.

Maddox, D.M., L.A. Andres, R.D. Hennessey, R.D. Blackburn and N.R. Spencer. 1971. Insects to control alligatorweed. Bioscience. 21:985-991.

McGehee, J.T. 1979. Mechanical hydrilla control in Orange Lake, Florida. J. Aquat. Plant. Manage. 17:In Press.

- Miller, J.L., L.A. Garrard and W.T. Haller. 1976. Some characteristics of hydrilla tubers in Lake Oklawaha. Hyacinth Cont. J. 14:26-29.
- Spencer, N.R. and J.R. Coulson. 1976. The biological control of alligatorweed in the United States of America. Aquat. Bot. 2:177-190.
- Van, T.K., W.T. Haller and G. Bowes. 1976. Comparisons of the phytosynthetic characteristics of three submersed aquatic plants. Plant Physiol. 58:761-768.
- Zeiger, C.F. 1962. Hyacinth obstruction to navigation. Hyacinth Cont. J. 1:16-17.

1979. Operational collection and release of waterhyacinth weevils. J. Aquat. Plant Manage. 17:In Press.

Agency	Hydrilla (x 1000)	Ditchbank ^{1/} (x 1000)	Waterhyacinth (x 1000)
Florida Department of Natural Resources <u>2</u> /	\$1,500	\$1,200	\$ 200
District, city, county $\frac{3}{}$	5,500	5,000	800
Florida Game and Freshwater Fish Commission4/	100	10	1,200
J. S. Army Corps of Engineers $\frac{5}{}$	0	0	800
Private enterprise ^{6/}	2,000	2,000	500
FOTAL	\$9,100	\$8,210	\$3,500

Table1. Expenditures for various types of aquatic weed control in Florida in 1977.

- 1/ Ditchbank weeds include many species of emergent and semi-terrestrial plants such as cattails (Typha sp.), reeds (Phragmites), grasses and sedges (Cyperus, Paspalum, Panicum spp., etc.), pickerelweed (Pontederia sp.), brush (Melaleuca, Cephalanthus, Myrica, Schinus, Salix spp., etc.), and others. Mechanical control (draglines, shovels, etc.) is often used for ditchbank weed control and serves a duel purpose of weed control and cleaning, widening, and deepening canals and ditches.
- 2/ The Florida Department of Natural Resources Bureau of Aquatic Plant Research and Control was established in 1970 and directs the state funding program in cooperation with local city, county, and district agencies.
- 3/ This category represents the local weed control authorities which raise money through local taxes for matching (obtaining) state monies from the Department of Natural Resources.
- 4/ The Aquatic Plant Control Section of the Florida Game and Freshwater Fish Commission is the operational agency for the state. Funds from state, local, and federal agencies provide the Commission with personnel and equipment to control all types of weeds.
- 5/ The U.S. Army Corps of Engineers by public law is charged with maintaining navigation and is the major source of federal funds for aquatic weed control. In 1977 the Corps was only authorized to control waterhyacinths; however, approval for control of additional weed species (<u>Hydrilla and Pistia</u>) was recently received.
- 6/ Private enterprise is the most difficult category to estimate because individuals do not apply for state matching funds. This category includes commercial weed control businesses, corporations, private landowners, etc. Estimates in the Table are undoubtedly conservative, particularly for ditchbank weeds.

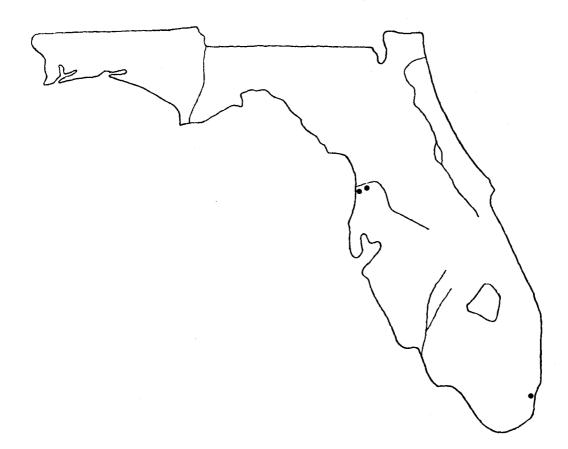
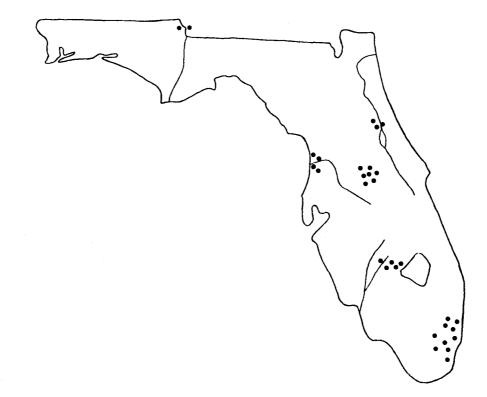
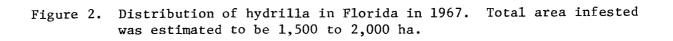


Figure 1. Known distribution of hydrilla in Florida in 1960. The total infestation in both Crystal River and the Miami River was approximately 10 ha.





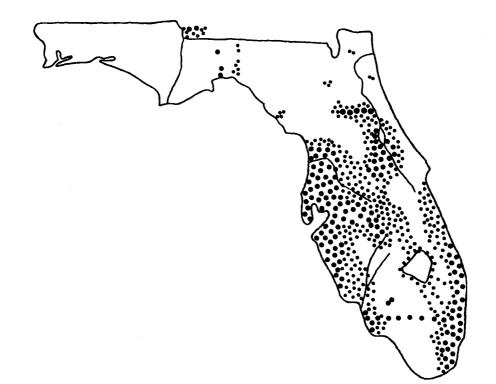
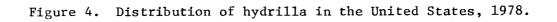


Figure 3. Known distribution of hydrilla in 1978. Larger dots represent dense infestations totaling some 40,000 ha, and smaller dots indicate hydrilla common in the flora of an additional 200,000 ha of Florida's fresh water.





THE USE OF GRASS CARP IN COMPARISON WITH OTHER AQUATIC WEED CONTROL METHODS

J.C.J. von Zon

Centre for Agrobiological Research (CABO)

P.O. Box 14

Wageningen, the Netherlands

INTRODUCTION

Control of nuisance aquatic weeds can be achieved by a number of methods based on the following principles:

- 1. <u>Growth Prevention</u>. This can be done by measures that prevent waterbodies from becoming eutrophicated or removing growth factors for noxious vegetation. Examples are the use of harmless competitive plants (Yeo 1976), stimulation of phytoplankton that fixes light and nutrients (Bungenberd de Jong 1967) or continuous shading by trees or shrubs (Krause 1977, Lohmeyer and Krause 1975), black plastic (Mayhew and Runkel 1962) or cardboard screens (Bernatowicz 1966). The use of preemergence herbicides is also regarded as preventive control, but its duration is usually much shorter than the other methods.
- 2. <u>Growth Retardation</u>. Not many methods have been studied, however, one biological example is the use of shade caused by aquatic plants with big floating leaves (von Zon 1976, 1977). Also growth-retardants have been tried, but have not been very successful.
- 3. <u>Growth Cessation</u>. This principle is most commonly used for aquatic weed control, and can be divided into two catagories: one without harvest, including all chemical and some mechanical methods, and one involving harvest as with some mechanical methods. The latter method is preferred in many situations because at least part of the weed problem is removed, reducing nutrient release in the water.

The use of grass carp (<u>Ctenopharyngodon idella</u>) is intermediate: there is progressive retardation of weed growth, so that growth stops at a certain moment, while some of the plants are harvested.

AQUATIC WEED MANAGEMENT

There are not only different methods for aquatic weed control, but also various types of aquatic weed problems. The control method should be adjusted to the problem, and therefore it is necessary to know the water demands by user groups, and under which conditions the control methods are efficient. The latter can easily be answered by empirical studies such as herbicide concentrations, grass carp stocking densities, solidity of paths for mechanical devices, in relation to water depth and width. Demands are more difficult to quantify because for most uses the criteria are not well known. Of course, all plant material is in principle unwanted in a waterway used exclusively for water transport. This means that the most effective method is best as long as it is not dangerous to crops or the receiving water at the end of the irrigation system. The other extreme can be found when a water is in a naturereserve, where as much species diversity as possible is wanted. Here only the aging process (becoming land) must be retarded.

Most waterways have more than one function. In many developing countries fish production and navigation may be as important as water transport, while in heavily populated parts of the western world often recreation becomes more important than the original agricultural function. In the Netherlands, where 14 million people live on only 4 million ha, the thousands of kilometers of ditches constructed for irrigation and drainage also provide recreation for a million sport fishermen. In consequence, these waters should contain a diverse and sound fish population with good opportunities for breeding and growth. In addition, they play an important landscape role, connect many relatively small nature reserves in the Rhone and Meuse Delta, and have educative value in biology teaching. Also important is their function of providing drinking water for cattle and even humans. It is practically impossible to judge the relative importance of such functions for every ditch, canal, brook, pond or lake and even then, it would be nearly impossible to prescribe maintenance programs for each one. For example, the critical amount of emergent vegetation needed for breeding of pike (Esox lucius), the quantitative relation between weeds and other organisms and the resistance created by various aquatic plant species for water transport are essentially unknown. It is also not possible to predict the amount of transport needed in a certain year, since the weather is not predictable.

In these uncertain situations weed control must be carried out in such a way that everyone is satisfied. This is not possible via the use of rigorous control methods, but only via the way of careful weed management. This means that the following demands must be fulfilled.

- 1. A set of minimum standards for water transport should be formulated, including a realistic small risk. In the Netherlands it seems possible to predict the percentage of the waterway capacity that may be blocked by aquatic weeds in some periods of the year.
- 2. Aquatic weed management should be directed as much as possible to the most vulnerable function of the waterway. In the Dutch situation this is often providing a biotone for many aquatic organisms. Most of these inland waters become a refugium for eutrophic freshwater organisms that originally found their habitat in old river areas which no longer exist. In considering the use of weed control methods, species diversity (especially its constancy and composition in a certain waterbody during the year) plays an important role.

Broad scale weed control methods must be developed. Making allow-3. ance for 1 and 2 above often leads to compromises, however, it will become increasingly possible to adjust weed control systems to local situations especially as more data are collected concerning the positive and negative effects of various control methods, waterway usage and plant biology. This means, that in the course of time aquatic weed management can be adapted to fit common types of natural management which create high local diversity, with high temporal constancy. This type of management is not as futuristic as it may seem. In Dutch road verges, a similar system was introduced with success during the past years: every verge unit has its own prescribed maintenance scheme which takes into account both traffic safety and vegetation type in the surrounding lanscape. As an example, mowing time is adjusted to the time of seedfall of the most important plant species.

With these demands in mind, existing aquatic weed control methods can be compared in a general way, but the emphasis will be placed on the use of grass carp.

EFFECTIVITY OF METHODS

There are various parameters to evaluate the effectiveness of a control method. Generally, only short-term effects are evaluated and rated either "good" (no plants) or "bad" (plants still present). "Moderate" may turn into "bad" very soon. A comparison of the effects of a method with the wanted level of control does not seem practical at the moment, since clear definitions of water levels cannot be given. Still another criteria for judging methods is the duration of the effect. Longer durations are usually considered better, but correlations with the period of the year that control is wanted is usually not made, nor is an adjustment of the method to the growth cycle of the species made. Sometimes species are removed mechanically during the steep part of their growing cycle, so that the effect lasts only a few days, or they are killed or removed only a few weeks before their natural die-off.

When grass carp are applied, criteria for effectiveness must be judged more subtly and more knowledge is necessary of the desired level of control throughout the growing season. Rapid effects cannot be expected, since every plant species that is not consumed in the beginning continues to grow. Rapid effects require mechanical or chemical methods or a very high stocking of grass carp, followed by removal of individuals after a few weeks. With grass carp weed control progresses relatively slow so that stocking should be realized in such a way that good control is achieved in the most critical periods for water transport. In doing so, the duration of the effect in the first year is not important. In temperate regions such a stocking routine is relatively easy, since most aquatic weed problems are present in autumn when rainwater must be drained. Grass carp control vegetation during the summer causing a more acceptable level during the time of critical water transport. A big part of spring and summer weed control with mechanical or chemical methods was considered necessary to prevent weed growth from becoming too dense for efficient control in the autumn.

When method effectiveness is considered over a period of years another criterium other than the rapidity and duration of control is more important, namely the consistance of the weed problem.

In the past, aquatic plants were harvested every year and in succeeding years the same species appeared again, which, as stated before, is a demand for good management. Presently this is still the case where some types of mechanical control are used, at least as long as cut material is harvested in some way. The use of herbicides causes some form of selectivity. The most common example is that control of emergent or floating vegetation with phenoxy-herbicides, nearly always gives rise to a submerged weed problem, which is often worse for some water or waterway functions than the original problem.

The results of years of submerged vegetation control with chemicals can be seen in parts of the Netherlands. In relatively stagnant water, bound nutrients from plants were released after every treatment resulting in hypertrophic conditions. These waters now support only filamentous green or bluegreen algae. Even these algae could be controlled chemically with herbicides, like diuron, but these compounds were not controlling all algal species. The results were very unattractive canals (both from an aesthetic point of view and biological) unusable for nearly every function and with a practically unsolvable algal problem. For various reasons the use of these algicides is now forbidden in the Netherlands (Zonderwijk and von Zon 1974, von Zon and Zonderwijk 1976) and means that users of allowed herbicides must be aware that they can create algal problems that overshadow the original problem.

Grass carp are sometimes reported to be selective in their food choice, although this decreases with age (Cagni, Sutton and Blackburn 1971). Vegetation shifts are, however, in quite another direction and not as detrimental as those seen with herbicides. The reasons are:

- Not all nutrients of consumed plant material flow back into the water; about 50% of the ingested phosphorus and nitrogen is retained in the fish (Nikolskii 1956, Hickling 1966, Mickewicz, Sutton and Blackburn 1972, Fischer and Lyakhnovich 1973, van Rijn, Werumeus Buning and van der Zweerde 1975). As long as the grass carp does not die, these nutrients are withdrawn from the system. The other 50% is only partially available for plant growth, so that severe eutrophication effects do not become visible within the system.
- 2. Shifts towards filamentous algae are not possible, since those plants are consumed with eagerness.
- 3. Plant species that are commonly rejected, even by a mixed population of grass carp, are generally of not much importance, with the exception of water hyacinth. Strongly rejected species are those of the genus <u>Batrachium (Ranunculus</u>), of which many are toxic or raise blisters. Experiments in containers showed that they are consumed to a certain extent after starvation (van Rijn, Werumeus Buning and van der Zweerde 1975); in practice many species of this genus have ended their yearly cycle before they can cause difficulties for water transport (von Zon and Zonderwijk 1975). Another group of unedible species are the Nymphoids (Nymphaea, Nymphoides, Nuphar, Lotus and

others). These plants are consumed when no other species are left, so that selectivity is probably not a matter of unpalatability, but of difficulties in attaining them.

In the Netherlands, an object of study at the moment is whether these Nymphoids, with big floating leaves, can be used in an integrated approach with the grass carp: the big leaves shade submerged species to some extent, probably without interfering with water transport. The fish consumed the remaining submerged material and prevents the floating mat from becoming too dense. In a temperate climate such integration is useful, because summer temperatures can be very different from one year to another. If grass carp are stocked in densities to be effective in a rather cold year, this could lead to starvation in warm summers. The combination of grass carp with floating plants might allow reduction in the initial grass carp stocking density. In cold summers, the mat of floating leaves will increase and in warm summers this mat will serve as a reserve food source for the fish.

The physical and chemical quality of the water is one important factor that can interfere with the success of weed control operations. In general, mechanical control is independent of these factors; however, the effect of herbicides can sometimes be influenced by it. A well-known example is, that herbicides such as paraquat and diquat lose their activity immediately when there is a high content of floating organic matter, which can be increased suddenly by rainfall or improper application techniques. It is clear that biological weed control methods are susceptible to changes in environmental quality. The water must be suitable for fish, which means that the oxygen content should not change very much, and there should be open areas. This may sound very clear, but in practice grass carp are often used by purely technical people for whom this is not the first thought. It is often written that grass carp are not very effective in shallow waters or in often disturbed waters. In experiments where grass carp could not avoid shallow water and disturbances they acclimated to these conditions rather fast (van Starkenburg and van der Zweerde 1976). In practice shallow water areas should be fenced to acheive good effects. In the same experiments no habitation occurred in brackish water (over 500 ppm C1⁻) and food intake was lower than in controls, which means that higher stocking densities were needed to achieve the same effects.

It is difficult to compare the economics of grass carp usage with those of common mechanical and chemical methods. Grass carp are stocked for several years, which means a loss of interest in invested capital, but inflation does not occur. Fences must be constructed and maintained, especially in shallow water where fish kills might occur. Literature data and Dutch experiences indicate that in countries where the fish can be bred the total weed control price will be half of that of chemicals and a quarter of that of mechanical control (Hone 1973, Janichen 1974, von Zon, van der Zweerde and Hoogers 1976).

The different aspects of the effectiveness of various aquatic weed control methods are summarized in Table 1. I concluded that grass carp are an effective, relatively cheap, and lasting weed control agent, provided that the environment is suited for their survival and rapid effects are not wanted.

SIDE EFFECTS

Another reason to use grass carp is that herbicides can be directly or indirectly toxic to various aquatic organisms. Another is the high cost of the mechanical alternative. Grass carp, however, have other side effects that should be studied before their utilization, such as chemical, physical and biological changes of the aquatic environment and the consequences of these changes upon other organisms. During the last few years an enormous amount of data has been collected, but the interpretation of this data is difficult for various reasons.

- 1. In nearly all studies grass carp are compared to an undisturbed control situation. Shifts in biological parameters that are found in such studies have scientific value, but in practice, grass carp will only be introduced where the weed problem cannot be left undisturbed. It is necessary to make comparisons between the side effects of commonly used mechanical and chemical methods and grass carp. One of the rare documented studies, in this respect, is that of Buck, Baur and Rose (1975). They did not find clear biological differences during one year after application of grass carp and diuron in preplanted swimming pools. Some differences in water quality (e.g. higher orthophosphate levels after diuron) indicate that shifts to other problems were less in the pools stocked with grass carp.
- Most studies concerning side effects have been conducted in aquaria 2. and data from such studies cannot be translated directly to field situations (von Zon 1977). Generally the stocking density in laboratory situations is so high that changes are exaggerated (Michewicz, Sutton and Blackburn 1972 and von Zon, van der Zweerde and Hoogers Furthermore, aquaria do not simulate a total ecosystem with 1976). its compensating and buffering capacities. There are indications that changes, which are often based on laboratory studies, are not found in the field. This is true for food selectivity since in an aquarium the supply of plants is not natural and the grass carp are usually very small. It is often observed that macroinvertebrates are consumed in aquaria, which probably results because they are easily caught and are needed for essential amino acids which are lacking in their food supply. The small fish used in these studies might need more animal food (Bailey 1972, Opuszynski 1972). Differences are found between field and aquarium studies as pertains to plankton, which is related to increased nutrients. In laboratory studies with grass carp, the orthophosphate content of the water increases, which leads to blooms of phytoplankton. In the field this is generally not the case (Johnson and Laurence 1973, Rottmann and Anderson 1977) because released nutrients are probably bound in bottom-sediments (Terrell 1975) or in parts of the living aquatic ecosystem, especially fish. Recent reviews on the side-effects of grass carp can be found in von Zon, van der Zweerde and Hoogers 1976, and von Zon 1977.
- 3. The results of grass carp research are interpreted to a great extent by investigators and agencies that are not involved in herbicides and mechanical control and they tend to ask more questions. In fact,

work on the side effects of mechanical control is nearly not carried out, except as pertains to dredging. Chemical control side effects are partly studied by industrial weed specialists (often in the framework of what governments ask for allowance) and partly by biologists. Fishery people have cared little about herbicides and held the view that their side effects were well studied. They generally have not asked for special studies for herbicide allowances. But when grass carp came into reality, fishery people felt capable of judging and asked for more research than has ever been required for other types of aquatic weed control. The usual argument that grass carp are persistent, whereas herbicides are temporary is unrealistic, since curtailment of a herbicide or mechanical treatment program immediately brings back the weed problem.

4. Field methods currently in use do not allow for valid conclusions. Original fish populations are often estimated by rotenone samples, but the impact of these samples on the remaining fish populations is unknown. The influence of grass carp on fish populations is in a few years measured with a new sample. Although drawn from a same area, the situation may be completely different; for example, it may be different to catch fish in a weedy or clean situation (Newton, Merkowsky, Martin, Ellis and Stanley 1976). Moreover, cleaning often stimulates sportfishing, and this may alter the fish population. The same type of difficulties are encountered in sampling macroinvertebrates and plankton.

Summarizing these points, it seems evident that the only way to establish the relative importance of grass carp side effects is a field study in which during a number of years, various commonly used control methods are computed. However, such a study would require so much work that international cooperation is needed. At the moment, policy-people often will not accept data from other countries, since they are not familiar with the names of organisms. However, the structure of aquatic ecosystems with weed problems will be so similar, that the different species occupying niches is not relevant. Therefore, it should be possible to design experiments to answer questions of general importance in order that the results will be applicable to many world situations.

In Table 2 a rough estimation is given of the side effects of various weed control methods in the Netherlands. In this connection, it should be noted that a very clear yearly cycle occurs for most organisms in temperate regions. For many of them, a very important part of the cycle, breeding, is in spring or early summer. The role that aquatic weeds play in the production of these organisms will not be greatly affected by grass carp since the quantitative effects on aquatic vegetation is not important before midsummer if overstocking is prevented (von Zon 1974). The other methods, especially chemical, are often carried out as early in the season as possible, both for maintenance work and for prevention of heavy weed growth. Dense stands of vegetation when treated could cause serious problems (e.g. oxygen depletion by killing all vegetation). In tropical countries, the influence could be more severe because of constant grass carp pressure, although there, a lower stocking density must be compared with a higher herbicide input.

The impact of grass carp on the quantity of other biota in an aquatic ecosystem is mainly temporary (only in the first one or two years) and the diversity is at most only slightly influenced. Therefore, its use for weed control in waters or waterways with different functions is very attractive. The old situation can be restored easily by removing the fish, as has been shown in Dutch experiments. On the other hand, chemical control influences the structure of the living community much more and leads to impoverishment so that a very long restoration time is needed. The effects of persistent herbicides are more severe since they also affect small, nontreated sidewaters that otherwise could act as a biological buffer. The influence of mechanical control is hardly known. If mechanical control is merely an imitation of clearing with handpower it can be expected that the influence on the biota will be small.

SUMMARY

As far as possible from the scarce field experiments comparing the effects of various methods to control aquatic weeds, the following two conclusions can be made.

- 1. The use of grass carp is generally as reliable as mechanical and chemical methods, relatively cheap and lasting, but not fast-working.
- 2. The side effects of this use are at least less severe and anyway less irreversible than those of chemical control.

REFERENCES

- Bailey, W.M. 1972. Arkansas' evaluation of the desirability of introducing the white amur (<u>Ctenopharyngodon idella</u> Val.) for control of aquatic weeds. 102nd Ann. Meet. Am. Fish. Soc. and Int. Ass. Game and Fish Comm., Hot Springs. Memeogr. 59 pp.
- Bernatowicz, S. 1966. The effect of shading on the growth of macrophytes in lakes. Ekologia Polska-Series A. 14:607-615.
- Buck, D.H., R.J. Baur and C.R. Rose. 1975. Comparison of the effects of grass carp and the herbicide Diuron in densely vegetated pools containing golden shiners and bluegills. Prog. Fish-Cult. 37:185-190.
- Bungenberg de Jong, C.M. 1967. Die Anwnedung van Diuron in der Teichwirtschaft. Proc. EWRC 2nd Int. Symp. Aquatic Weeds, Oldenburg. 66-69 pp.
- Cagni, J.E., D.L. Sutton and R.D. Blackburn. 1971. A review of the amur (<u>Ctenopharyngodon</u> idella) as a biological control agent for aquatic weeds. Mimeogr. 17 pp.
- Fischer, Z. and V.P. Lyakhnovich. 1973. Biology and bioenergetics of grass carp (Ctenopharyngodon idella Val.). Pol. Arch. Hydrobiol. 20:521-557.
- $\frac{590.6}{287\rho}$ Hickling, C.F. 1966. On the feeding process in the white amur, <u>Ctenopharyn-</u> <u>z87\rho</u> <u>godon idella</u>. J. Zool. 148:408-419.
 - Höne, U. 1973. Experiment in the use of grass carp for biological weed control in watercourses. Weed Abstr. 24:3020.
 - Jähnichen, H. 1974. Sunkung der kosten bei der Wassenpflanzenbekämpfung durch den Amurkarpfen (Ctenopharyngodon idella). Z. Binnenfisch. DDR 21:85-89.
 - Johnson, M. and J.M. Laurence. 1973. Biological weed control with the white amur. APCP, techn. Rep. 4, Herbivorous Fish for Aquatic Plant Control. E3-E12.
 - Krause, A. 1977. On the effect of marginal tree rows with respect to the management of small lowland streams. Aquatic Botany. 3:185-192.
 - Lohmeyer, W. and A. Krause. 1975. Ueber die Auswirkung der Gehölzbewuchses an Kleinen Wasserläufen. Schriftenz. f. Veget. kunde. 9:105 pp.
 - Mayhew, J.K. and S.T. Runkel. 1962. The control of nuisance aquatic vegetation with black polyethylene plastic. Proc. Iowa Ac. Sc. 69:302-307.
 - Michewicz, J.E., D.L. Sutton and R.D. Blackburn. 1972. Water quality of small enclosures stocked with white amur. Hyac. Contr. J. 10:22-25.
 - Newton, S.H., A.H. Merkowsky, J.M. Martin, J.E. Ellis and J.G. Stanley. 1976. Effects of white amur on populations of warmwater fish. Mimeogr. 30 pp.

Nikolskii, G.V. 1956. In Opuszynski 1972.

- Opuszynski, K. 1972. Use of phytophagous fish to control aquatic plants. Aquaculture. 1:61-74.
- Rijn, C.P.N. van, W.H. Werumeus Buning and W. van der Zweerde. 1975. Onderzoek Naar de Invloed van Graskarper op de Macrofauna van Sloten en Naar de Samenhang Tussen Graskarper (Activiteit). Waterplanten en Waterkwaliteit. Mimeogr. Rep. 104 pp.
- Rottmann, R.W. and R.O. Anderson. 1977. Limnological and ecological effects of grass carp in ponds. Proc. S.E. Assoc. Game and Fish Comm. 30 pp. (In Press).
- Starkenburg, W. van and W. van der Zweerde. 1976. Onderzoek Naar de Invloed van Bezettingsdichtheid, van Waterdiepte en Zoutgehalte en van Verstoring op de Voedselopname en- Conversie van der Graskarper, Alsmede Naar Zijn Gedrag bij Aanwezigheid van Dierlijk Voedsel. Mimeogr. 28 pp.
- Terrell, T.T. 1975. The impact of macrophyte control by the white amur (Ctenopharyngodon idella). Verh. Int. Ver. Limnol. 19:2510-2514.
- Yeo, R.R. 1976. Managing aquatic vegetation by plant competition and antipathic inhibitions. Proc. IV Int. Symp. Biol. Contr. Weeds. Gainesville, 1976. (In Press).
- Zon, J.C.J. von. 1974. The grass carp in Holland. Proc. EWRC 4th Int. Symp. Aquatic Weeds. Wien:128-133.

______. 1976. Status of biotic agents, other than insects or pathogens, as biocontrols. Proc. IV Int. Symp. Biol. Contr. Weeds. Gainesville, 1976. (In Press).

. 1977. Introduction to biological control of aquatic weeds. Aquatic Botany. 3:105-109.

. 1977. Grass carp (<u>Ctenopharyngodon idella</u> Val.) in Europe. Aquatic Botany. 3:143-156.

and P. Zonderwijk. 1975. Chemische Bestrijding van Waterplanten. Waterschapsbelangen. 60:148-149.

. 1976. Diuron en Dichlobenil Voor Teopassing in Water Afgewezen. Waterschapsbelangen. 61:78-80.

, W. van der Zweerde and B.J. Hoogers. 1976. The grass carp, its effects and side-effects. Proc. IV Int. Symp. Biol. Contr. Weeds, Gainesville, 1976. (In Press).

Zonderwijk, P. and J.C.J. von Zon. 1974. A Dutch vision on the use of herbicides in waterways. Proc. EWRC 4th Int. Symp. Aquatic Weeds. Wein:158-163.

GRASS CARP: THE SCIENTIFIC AND POLICY ISSUES

Scott Henderson

Fisheries Research Biologist

Arkansas Game and Fish Commission

INTRODUCTION

I have been asked to discuss the scientific and policy issues involving the use of the grass carp for weed control. I find this somewhat difficult to address because it seems most of the issues raised in this country have been emotional issues. These issues have been based on the lack of understanding of a very complex problem which, as oftentimes presented, is very loosely tied to factual information.

INFORMATION IN THE PRESS

The grass carp has become a "popular" controversy in the United States receiving publicity in widely circulated national magazines and newspapers. We are, according to these articles, discussing a fish that can grow to a maximum size of anywhere from 25-400 lbs., consumes anywhere from 2-12times its body weight per day, and has the ability to jump from 3-12 ft. out of the water. Sensationalism and publicity have blown the controversy way out of proportion.

Some of the popular articles opposing the fish have said, and I quote:

From <u>Shreveport Times</u>, Shreveport, La. - "The grass carp may very well constitute the most alarming situation in the history of fishery research."

From UPI wire service originating in Austin, Texas - "The grass carp could decimate the coastal rice industry if allowed to spread."

From the <u>Galesburg-Register Mail</u>, Galesburg, Ill. - "He feeds by smell rather than by sight, he roils the water and makes it muddy which creates serious diet problems for other fish."

From the <u>Sentinel-Star</u>, Orlando, Fla. - "The fishes effect on largemouth bass is devastating. It does not control noxious weeds."

From the <u>River Hills Traveler</u>, Missouri - "One of the first things discovered about it is that it prefers animal rather than vegetable food. It readily eats the eggs of game fish and rough fish. And finally, when it can't get enough animal matter, it turns to vegetable matter for food and even this is a problem."

By this time, everyone knows that "carp" and "Amur" are synonymous and that both are four letter words.

On the other hand, many of the fish's backers have had no less effect on the controversy. The label "SUPERFISH" certainly added fuel to the fire. Some of the popular articles supporting the fish have said, and I quote:

From <u>Mechanix Illustrated</u> - "As wily a game fish as the trout, as scrappy when hooked as the mighty tarpon, as tasty when cooked as the red snapper. It also has the potential to surpass beef as a source of protein."

From the <u>Star Progress</u>, Berryville, Ark. - "There is no possibility they will reproduce naturally in the U.S. They can easily be caught on hooks baited with grass or moss."

To me the worst of all are those that purport to only present the facts of the controversy so that you as an intelligent individual can decide for yourself. Again, I quote:

From AP wire service originating from Elkhart, Ind. - describes the grass carp as "A large off-white goldfish that grows up to two feet in length."

From the <u>Anniston-Star</u>, Anniston, Ala. - "In Arkansas where the fish is grown on a large scale, those handling it always wear a baseball catchers mask for protection. One death has been attributed to the fish there."

From the Sunday Magazine of the <u>Cleveland Plain Dealer</u> - "At Arkansas hatcheries, employees always wear baseball or fencing masks when working with the fish."

From the <u>River Hills Traveler</u>, Missouri - "The original importation of these fish was by the Soil Conservation Service in Florida."

And there is finally the supreme example of misinformation about these fish. An article in the nationally publicized <u>American Legion Magazine</u> began with a lead that read, "Through the years a monster has been invading our lakes, ponds, and streams - it's the grass carp - a fish introduced into this country from Germany in 1872." The article was complete with a very clear picture of the common or European carp.

SCIENTIFIC ISSUES

The scientific community hasn't been much different than the press in this matter. There appeared to be two factions here from the beginning. Those that support the "Innocent until proven guilty" theory who generally support the use of the fish, and those of the "Guilty until proven innocent" opinion who are generally against its use. Seemingly endless scientific studies of the grass carp can be quoted and the findings interpreted as good or bad, in most cases, depending on your point of view at the outset. If it does nothing else, the grass carp has stimulated the economy in this country by creating more jobs in fishery research in the last 10-15 years than anything else.

The fact is, the overriding questions about the effect of the grass carp remain unanswered. (1) Will the grass carp reproduce in this country? and (2) What will be its impact on the habitat? (usually concerned with speeding up the eutrophication process). These questions do not lend themselves to

the cold objectivity of the classical scientific method. We cannot predict future weather patterns or the subtle effects of high water in high water years or the 100-year flood might have on the reproductive habits of these fish. Certainly not from laboratory or pond experiments. Nor can we predict their impact with complete certainty on any pond, lake or reservoir. There are so many intricate variables within any isolated aquatic system that they are difficult to enumerate much less control and predict. Couple this with the many types of aquatic environments and complicate it further by adding in geographic location and the task becomes enormous. Yet this 100% certainty is what many seem to advocate before using the fish.

COMPREHENSIVE STUDIES

Now, and hopefully without too much bias, I will briefly discuss what I consider to be the most comprehensive study of the grass carp in the United States to date. That is the use of the grass carp in Arkansas. The objective of the Fisheries Division of the Arkansas Game and Fish Commission is to produce the optimum sustained harvest and utilization of the states fisheries resources for the fishing public. Grass carp are important to the utilization of the resource by removing choking aquatic vegetation and allowing fishermen access. We found that the use of the grass carp to allow access is not mutually exclusive of managing the fish population for optimum sustained harvest by the fishing public.

To the present date, more than 100 lakes in Arkansas totaling over 50,000 acres have been stocked with grass carp for aquatic vegetation control. We feel that we have no major vegetation problems in Arkansas and those that were the worst are under control and manageable. We also have some of the best and most diversified fishing in the country.

In the late 1960's, Arkansas' experience with the grass carp began on the Joe Hogan State Fish Hatchery at Lonoke, Arkansas. Successful artificial spawning methods were developed over the first few years and the fish were tried for vegetation control in hatchery ponds with excellent success. This is not to say there were not some minor problems, primarily in handling the fish in the seine during harvest, but these are of little consequence when compared to attempting to harvest a fingerling or production pond choked with vegetation. Stocking rates in hatchery ponds may range from as low as 10 fish per acre to an excess of 1000 fish per acre depending upon the severity of the problem and the length of time available to achieve control. Our stocking rates are also aimed at producing an 8-10" stocker size fish by the time the vegetation in the pond is controlled, then the fish are used for vegetation control in state managed lakes.

After such success in hatchery ponds, some very major problems in the states fishing waters were the next objective. Obviously a lake is not like a hatchery pond and in 1968-69, the first lake in Arkansas was stocked with the grass carp. This was a topographically isolated, shallow, man-made lake. Vegetation control was achieved and other isolated lakes were stocked with the same results. No detrimental effects on the fish populations in the lakes could be determined from routine sampling. Fisherman success in these lakes remained the same or better, likely due to a better access to all areas of the lake. By 1972-73 the grass carp had become a routine management tool for

the control of aquatic vegetation in Arkansas' lakes and has continued to be since that time.

We have never proposed the grass carp as a sport or game fish in Arkansas, however, it is remarkably good eating. After the fish have reached a suitable size and vegetation is controlled, some lakes have been opened to special commercial fishing seasons for removal. This has met with varied amounts of success with the grass carp specifically as the target species. On a statewide basis, in 1976 over 50,000 lbs. of grass carp were taken by commercial fishermen in Arkansas and entered the fresh fish markets in the state. The grass carp remains less than 1% by weight of the total commercial catch of an estimated 6.5 million lbs. in Arkansas during 1976.

During routine population sampling, our district fishery biologists began to notice that, in waters where they were present, grass carp surfaced very soon after rotenone was applied. Further tests in the laboratory and small ponds showed that the grass carp was susceptible to rotenone at approximately the same level as the gizzard shad. This is approximately 1/10 the amount necessary to kill most fish; therefore, the grass carp could be selectively removed from existing fish populations. Even though we have never specifically targeted a kill toward removing grass carp, their sensitivity to rotenone causes many to be seen during sectional fish kills and other renovation projects. In Arkansas the fish management kills are scheduled on Saturdays and the public is invited to pick up and utilize the fish. Needless to say we have excellent public participation and many people have become aware of the table quality of the grass carp. It has often become the trophy fish of the fish kill in those waters where it is found.

In 1976, a survey of 31 lakes totaling more than 16,000 acres (Bailey, In Press) was made utilizing population sample data taken both before and after grass carp were stocked. Some of the parameters looked at were total standing crop, catchable bass and crappie, total standing crop of shad, young of the year of bass and bluegill, and condition factor of catchable bass, bream, and crappie. A look at the overall fish population showed a declining trend in 6 of the lakes, an upward trend in 8 of the lakes, and the remainder fluctuated within limits measured prior to the introduction of grass carp. The only logical conclusion being that the grass carp eliminate the vegetation but other factors such as water level, fertility, and weather have a greater effect on fish populations than does the presence of the grass carp.

The only thing widely agreed upon about the grass carp is that they do eat aquatic vegetation. Our experience has been that weed removal by the grass carp produces no noticeable effect other than what would be experienced from weed removal by any other method. We also believe that a tool that can be used for vegetation control with less environmental impact and expense than chemical and other known methods and be recovered as a part of our fishery resource is a valuable asset to our program.

Granted, there is much we do not know about the fish that we could learn, but where does it end. We can spend years constructing the most complete model possible that should predict with 99.9% accuracy. But in the end it always seems that the model becomes the end instead of the means and the final

step is to stock the fish and see if the model is right. The philosopher ponders the truth, the scientist searches for the truth, but the truth is what happens. The only perfect science known to man is hindsight.

POLICY ISSUES

It seems to me that the issue at hand is to decide if the possible or known values of the fish outweigh the possible or known detrimental effects of the fish and that decision is still further complicated and influenced by the primary use of the water involved. We have been primarily concerned with sportfishing, fish production, and irrigation or drainage; but what about boating and other water related recreational activities, aesthetic qualities, power production, and a multitude of industrial uses. Whatever your perspective toward water use, one overriding problem that spans all uses is nuisance aquatic vegetation and a satisfactory method of controlling it is essential. There are no easy answers.

The one thing I have seen from my experience with Arkansas and our distribution of the fish to various agencies, institutions, and organizations for research purposes is that almost without fail, those who have had experience with the fish in actual field trials have opinions ranging from advocacy to a 'proceed with caution' approach. But none that I am aware of are among those still adamantly opposed to their use.

After nearly 15 years in this country and reams of research reports, every new paper and article still is filled with 'may' and 'might' and every conclusion points out other questions that remain unanswered. There is a saying that goes: The best thing you can do is the right thing, the second best thing you can do is the wrong thing, and the worst thing you can do is nothing. However you see the issues involved, the grass carp controversy goes on and will not be resolved by those who choose to sit back and espouse their opinions and beliefs without some real efforts to substantiate them.

RESEARCH AND MANAGEMENT APPLICATION OF GRASS CARP IN IOWA^{1/}

Larry Mitzner

Chariton Research Station

Route 1, Box 209

Chariton, Iowa 50049

This presentation will cover three aspects of grass carp introductions in Iowa. These are: (1) research, (2) its application to management of aquatic weeds, and (3) the distribution of grass carp in the river systems of Iowa.

The research study objective was to evaluate the effectiveness of grass carp to biologically controlled dense beds of nuisance aquatic vegetation in a 29 ha man-made impoundment in order to improve angler success along the shorelines. This was accomplished by measuring the standing crop of aquatic macrophytes before grass carp were stocked, stocking grass carp and then continuing to measure the biomass and species composition of the macrophyte community. The vital statistics of grass carp including growth, body condition, food consumption, mortality and behavioral activity were described.

The third phase of the investigation was to measure the indirect impact of vegetation control on the water quality, primary production of phytoplankton and the sport fish harvest.

Red Haw Lake, the study area, is a 29 ha man-made impoundment located in south-central Iowa. In July, 1973, 18 grass carp per ha were stocked followed by nine grass carp per ha in July, 1974. Red Haw is a recreational lake surrounded by a state park where public fishing is a major activity. Many areas of the lake are accessible to shore fishermen, yet during the summer months anglers find it nearly impossible to fish from shore because of massive beds of aquatic weeds. This problem exists in many Iowa lakes, resulting in poor use of the fishery resource.

Macrophyte biomass and species composition were measured in 1973 before grass carp were stocked and then monthly during May-September each year thereafter. Divers equipped with SCUBA sampled ten permanently located stations by placing a square frame 0.5 m on each side (0.25 m^2) over the vegetation and extracting all the plants from within the frame. The material was brought to the surface where wet weights were recorded. Samples were preserved for later sorting, identification and measurement of weights for individual plant taxa.

Maximum vegetation biomass occurred prior to grass carp introduction with

I Funds for this study were provided by the Federal Aid in Fish Restoration Act (PL 81:681), Project F-88-R, U.S. Fish and Wildlife Service and the Iowa Conservation Commission cooperating.

an average estimate of 2,438 g/m^2 . Within four years the overall vegetation biomass was reduced to 211 g/m^2 (Figure 1). The greatest reduction in a single season occurred in 1973 when test plot weights were reduced by 73%. Mean sample weight in 1974 was 1,142 g/m^2 decreasing to 445 g/m^2 in 1975 and then to 211 g/m^2 in 1976. Aquatic macrophyte biomass increased greatly in 1977 when the density of grass carp was reduced to an estimated 7 fish per ha.

At first, <u>Potamogeton</u> and <u>Najas</u> dominated the plant community. After three years both groups were nearly eliminated. <u>Potamogeton</u> remained below 100 g/m^2 , while Najas increased greatly in 1976 and 1977 when average plot weight in August was greater than 2,000 g/m² (Figure 2).

Elodea was nearly nonexistent when grass carp were stocked, but became increasingly important in 1974 and 1975 when test plot values in August were about 1,000 g/m² and 500 g/m², respectively. In 1976 and 1977 mean biomass was < 500 g/m². Ceratophyllum never became a problem and by 1976 and 1977 field weights were reduced to values < 100 g/m² (Figure 2).

Vital statistics measured in this investigation were growth, body condition, food consumption, mortality, and behavioral activity. Grass carp were captured with 76-127 mm bar mesh gill nets in shallow embayments in the summer and midwater sets under ice in winter. Lengths, weights and reproductive condition were recorded at the time of capture.

Grass carp grew rapidly from an average length of 320 mm at stocking to 824 mm four years later. Mean weight at introduction was 380 g, while average weight at the end of the 1976 growing season was 6,847 g (Figure 3). Maximum size in the 1976 sample was 890 mm and 10,600 g. Body condition factor, K, ranged from 1.05-2.02, while mean K-factor ranged from 1.13 for the sample in October, 1975, to 1.49 in October, 1973. Mean condition when grass carp were stocked was 1.28, but increased to 1.37 or greater by October. By January and February, mean condition factor again decreased to values in the 1.25-1.30 range.

Grass carp consumed aquatic vegetation almost entirely. Food volume from 28 grass carp in 1973-1975 was measured and items identified along the entire length of the alimentary tract. The plant material in the gut was identified by comparing it to reference material from plants in Red Haw Lake. First, plant leaves and stems were crushed and mounted on slides where the material was cleared and fixed with chloral hydrate and gum arabic solutions. Material from the gut was treated identically and plant fragments were compated with reference material and identified by cell size and morphometry. Vegetation was the main food source, however, < 0.1% of the food content by volume were invertebrates. Traces of filamentous algae, particularly <u>Oedogonium</u> and <u>Spirogyra</u> were found in most of the alimentary tracts.

Aquatic macrophyte fragments in the gut were composed of <u>Potamogeton</u>, <u>Elodea</u>, <u>Ceratophyllum</u> and <u>Najas</u>. In 1973, <u>Potamogeton</u> was most important contributing 65.7% to the diet followed by <u>Elodea</u>, 24%; <u>Najas</u>, 7.1%; and <u>Ceratophyllum</u>, 3.1%. Thereafter, <u>Potamogeton</u> became less important contributing 22.6% to the ration by 1975, while <u>Ceratophyllum</u> increased greatly to 34.5% of the diet. Elodea remained about the same at 20%, while Najas increased to 22.8% in 1974, but decreased to 11.9% of the diet in 1975 (Table 1).

Electivity indexes ranged from total avoidance (-1.00) in some samples to +0.98 for <u>Najas</u> in April and June, 1974. In all samples, except one, the electivity indexes for <u>Najas</u> were positive. <u>Potamogeton</u> was also positive in five of seven periods sampled. There was less selection for <u>Elodea</u> and Ceratophyllum where indexes were less than +0.52 and +0.61, respectively (Table 2).

Biomass of the grass carp population in Red Haw Lake was estimated annually as the product of numerical abundance and mean weight in the population. Mortality was estimated at 33%. Maximum population biomass was attained in 1975 at 1,767 kg. Initial biomass in July, 1973 was about 200 kg which nearly quadrupled by the end of the growing season. The following year 250 additional grass carp were stocked and by October, 1974 the biomass of both introductions was about 1,600 kg. By 1976 growth slowed and mortality became an important factor causing a decrease in biomass to 1,485 kg (Figure 4).

Exact mortality estimates were not determined, but three sources of mortality were identified. Mortality of fish stocked in 1974 was caused mainly due to a weakened condition from parasitism of <u>Gryodactylus</u> and <u>Lernaea</u>. Grass carp were stocked in 28 July and within 15 days, 38 dead fish were found. Three carp were also found by divers on the lake bottom. Aquarium fish from the same stock were also heavily parasitized. Fish from the 1973 introduction were not affected.

The second most numerous cause of mortality was from netting and holding. Grass carp were captured and held in tanks in order to develop surgical techniques for implantation of sonic tags. This activity accounted for the loss of eight fish. Another source of mortality was from electrofishing. Two shocked fish did not recover equilibrium and later six dead fish were found in the same area where electrofishing gear was used.

Behavioral activity was determined by locating and following grass carp with ultrasonic tags surgically implanted in the body cavity. Transmitters were sealed in polypropylene tubes which measured 16 x 60 mm and weighed 8 g in water. Each transmitter emitted a different frequency and impulse rate ranging from 74-76.56 kHz with impulse rates ranging from 46 to 120 per minute.

The recipient fish was anesthetized for 5-8 minutes and placed ventral side up on a V-shaped holding table. A 20-25 mm incision was made 10 mm laterally and parallel to the mid-ventral line. Transmitters were inserted posteriorly between the viscera and body wall followed by closure of the incision with 3-4 sutures of No. 000 braided surgical silk. Battery lift provided a minimum of 120 days of observation.

The nine tagged fish were located by slowly following the lake perimeter approximately 50 m from shore with the hydrophone pointed towards shore. Occasionally the hydrophone was slowly rotated 360° so fish could be located in midwater. The overall distribution of grass carp was summarized by adjusting the number of observations from the fish which were released earlier. Thus, each tagged fish was equally represented in the distribution regardless of the number of times it was contacted. Contact locations were further grouped into four hydrographic components including shallow water (depth < 3 m), midwater (depth \geq 3 m), embayments and the main lake (Figure 5). Fish in the midwater area were confined to the upper lake strata during June-August by a well defined thermocline < 4 m from the surface. Shallow water areas contained 10 ha and were dominated by vegetation, while midwater contained 19 ha. Four embayments comprised 9.3 ha of which 5.7 ha were shallow and 3.6 ha were midwater.

Tagged fish were contacted 412 times which accounted for 62 hours of monitoring. Contact time averaged 9 minutes per fish, but ranged from brief receptions of two minutes to continuous monitoring of 24 hours.

Tagged fish were located in all areas of the lake except near the dam where vegetation was extremely sparse. Shallow water depth (depth ≤ 3 m) was by far the most frequently inhabited area where 251 of 412 contacts were made. In the shallow water segment, 75 contacts were within embayments, while 176 were in the main lake. Locations in deep water comprised the remaining 161 observations of which 63 were in embayments and 98 in the main lake (Table 3).

Chi-square analysis was used to compare observed values with the expected distribution given the assumption there was equiprobability of contacting sonic tagged grass carp in all habitat components. Frequency of contact within each component was adjusted for the size of each component so the observed values were on the basis of contacts per hectare, thereby component size had no influence on the analysis.

Additional analysis showed the significant χ^2 values were due to either a paucity or dominance of observed contacts compared with the expected (Table 4). Large χ^2 values caused by a greater than expected contact frequency showed a significant affinity for a particular area, and conversely, significantly large χ^2 values caused by lower than expected contacts with tagged fish in an area indicated avoidance of that area. Chi-square values which were nonsignificant showed neither avoidance nor affinity for the area.

Overall, the greatest selection was for shallow water of the main lake as seven of the nine fish had an affinity for that particular habitat. Conversely, seven of nine fish avoided deep water within the main lake. Six fish had no particular affinity for midwater within embayments and only two of nine fish had an affinity toward shallow water within embayments.

Usually grass carp were sedentary and moved very little. Movement was in and near weed beds and was slow and erratic, while movement in midwater was rapid and extended over longer distances. Average swimming speed in midwater was 0.12-1.16 m/sec, while maximum speed was 1.46 m/sec.

Two fish showed definite homing tendency toward established activity centers. Both fish were monitored many times leaving and/or returning to an area which they occupied nearly constantly. An example occurred on 10 July when Alice left her activity center twice and was observed returning three times within several hours. Nocturnal activity was not greatly different from diurnal movement.

Phosphates, nitrates, turbidity, alkalinity, pH, and biochemical oxygen demand (BOD) were measured each month commencing in March, 1974 with the samples taken at the surface, 4 m and 8 m at a single location.

Organic phosphates at the surface ranged from 0.21 mg/1 in 1977 to 0.46 mg/1 the previous year. Values in 1974 and 1975 were 0.24 and 0.32 mg/1, respectively. Surface inorganic phosphates showed a similar trend with the highest concentrations in 1976 at 0.41 mg/1. The lowest value occurred in 1974 at 0.28 mg/1 followed by consecutive increases in 1975 and 1976 when average inorganic phosphate concentrations were 0.32 and 0.41 mg/1.

Nitrate nitrogen concentration at the surface was greatest in 1974 at 1.11 mg/l and lowest the following year at 0.29 mg/l. Thereafter there was a gradual increase to 0.34 and 0.50 mg/l in 1976 and 1977. Nitrite nitrogen showed a similar trend with the greatest average concentration in 1974 at 0.021 mg/l followed by two consecutive decreases to 0.009 mg/l and 0.004 mg/l in 1975 and 1976. In 1977 nitrite nitrogen increased to levels near 1974 where the average concentration was 0.017 mg/l.

Turbidity increased during 1974-1976, but then decreased to the lowest level in 1977. Average water clarity in 1974 was 7.24 FTU followed by 10.01 and 10.90 FTU in 1975 and 1976. Average turbidity at the surface in 1977 was 6.30 FTU.

Alkalinity increased each year during the investigation from 109 mg/l in 1974 to 127 mg/l in 1977.

Average pH in 1974 was 7.74 which increased to 7.91 in 1975 and then was followed by a decrease in 1976 to 6.38. Average pH in 1977 was 6.70.

Biochemical oxygen demand decreased from 4.44 mg/l in 1974 to 2.80 in 1976 when it increased to an average of 3.33 in 1977.

Primary production of phytoplankton was measured biweekly by the light and dark bottle technique where a replicate set of bottles were suspended at 1 m intervals from 1-5 m.

Primary production was quite variable during the season (Figure 6) ranging from 0.05 grams of carbon per m² per day (gC/m²/day) to 3.92 gC/m²/day in 1976. Average production in 1974 was 1.91 gC/m²/day nearly identical to the production in 1975 when mean production was 1.90 gC/m²/day. In 1976 average primary production decreased to 1.59 gC/m²/day. A further decline in production occurred in 1977 when the average was 1.29 gC/m²/day.

Primary production was greatest near the surface with an overall average of 0.62 gC/m³/day. Thereafter production values decreased to 0.33, 0.19 and 0.14 gC/m³/day at 2, 3 and 4 m, respectively. Production at 5 m increased slightly to 0.18 gC/m³/day.

The ultimate objective of introducing grass carp in Red Haw Lake was to increase the angler success along the shoreline. The sport fishery was moni-tored during April through 15 September by an expandable census in 1974, 1975 and 1976.

Bluegill, by far, dominated the fishery followed by crappie and largemouth bass. Catch in 1974 was 12,781 followed by 22,372 and 14,638 in 1975 and 1976.

Catch and angler effort were separated by boat and shore anglers. Boat anglers caught far more fish and were more successful than shore anglers. For example, in 1975 shore anglers caught 4,340 fish compared to 18,031 fish for boat anglers and during the same year catch-effort of shore anglers was 0.90 fish/hour compared to 1.97 fish/hour for boat anglers.

Effort for boat anglers remained nearly constant, while effort from shore increased from 2,974 hours in 1974 to 7,181 hours in 1976. While vegetation biomass decreased from about 1,700 g/m² in 1974 to < 100 g/m² in 1975 fishing pressure increased nearly 2.5-fold (Figure 7).

Findings of this investigation showed water quality changed with intensive biological vegetation control. Phosphate concentrations and turbidity increased, but nitrates, nitrites and BOD decreased with higher levels of vegetation control. Alkalinity increased throughout the study and pH showed no particular trend with intensity of vegetation control. Regardless of the changes in water quality average primary production of phytoplankton decreased during the investigation.

The use of grass carp has been expanded to 25 lakes in Iowa where nuisance vegetation has become a problem in the harvest of sport fish. Most of the introductions were made in 1976 and 1977 so results of these stockings are not available. Within a few years the results of introducing 12 to 88 grass carp/ha in these lakes should be known.

The distribution of grass carp of unknown origin in Iowa rivers are shown in Figure 8. One fish was taken in the Mississippi River and verified by Illinois biologists. One carp was taken in a fish renovation project in a backwater of the Des Moines River by Iowa biologists, but most of the reports come from the Missouri River where four fish were captured by commercial fishermen.

	Potamogeton	Elodea	Ceratophyllum	Najas	Unidentified
1973	65.7	24.0	3.1	7.1	0.1
1974	43.3	21.8	13.8	22.8	0
1975	22.6	19.0	34.5	11.9	12.1

Table 1. Percent occurrence of food items in the alimentary tract of 20 grass carp at Red Haw Lake.

	Potamogeton	Elodea	Ceratophyllum	Najas	
1070					
1973 September	+0.27	+0.52	+0.15	-0.79	
1974					
April	-0.08	+0.07	+0.09	+0.98	
June	+0.13	-1.00	-1.00	+0.98	
August	-0.45	-0.55	+0.61	+0.92	
1975					
June	+0.59	-0.72	-0.88	+0.67	
August	+0.84	-0.39	-0.23	+0.22	
September	+0.81	+0.51	-0.30	+0.91	

Table 2.	Electivity indexes of four major food groups consumed by grass	
	carp at Red Haw Lake.	

	Embayme	nts	Main lake			
······································	Shallow	Deep	Shallow	Deep		
Alice	5	2	28	11		
Bertha	21	3	19	3		
Carla	2	5	9	30		
Debra	9	8	16	13		
Ethel	12	6	18	10		
Fay	19	6	15	5		
Gertrude	7	3	25	10		
H e len	0	0	46	0		
Ida	0	30	0	16		
Total	75	63	176	98		

Table 3. Frequency of ultrasonic contacts with nine grass carp in four hydrographic areas at Red Haw Lake.

	Embayme	nts	Main lake			
	Shallow	Deep	Shallow	Deep		
Alice	-	-	+	-		
Bertha	· +	0	+	-		
Carla	-	0	0	0		
Debra	0	0	+	-		
Ethel	0	0	+	-		
Fay	+	0	+	-		
Gertrude	0	0	+	-		
Helen	-	-	+	-		
Ida	-	+	-	0		

Table 4. Affinity (+), avoidance (-) or nonselection (0) of grass carp in four hydrographic components at Red Haw Lake based on χ^2 analysis of contact frequency.

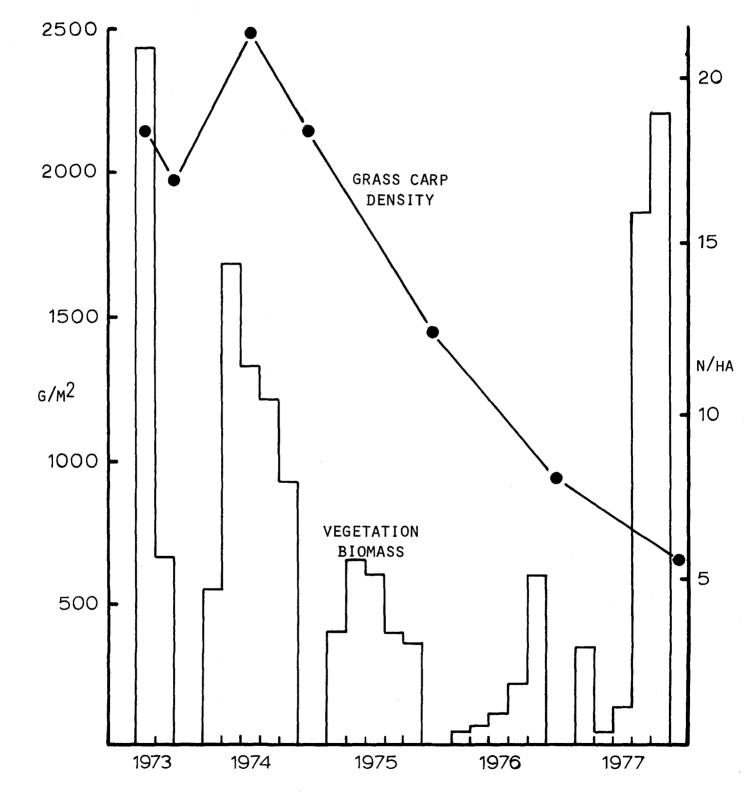
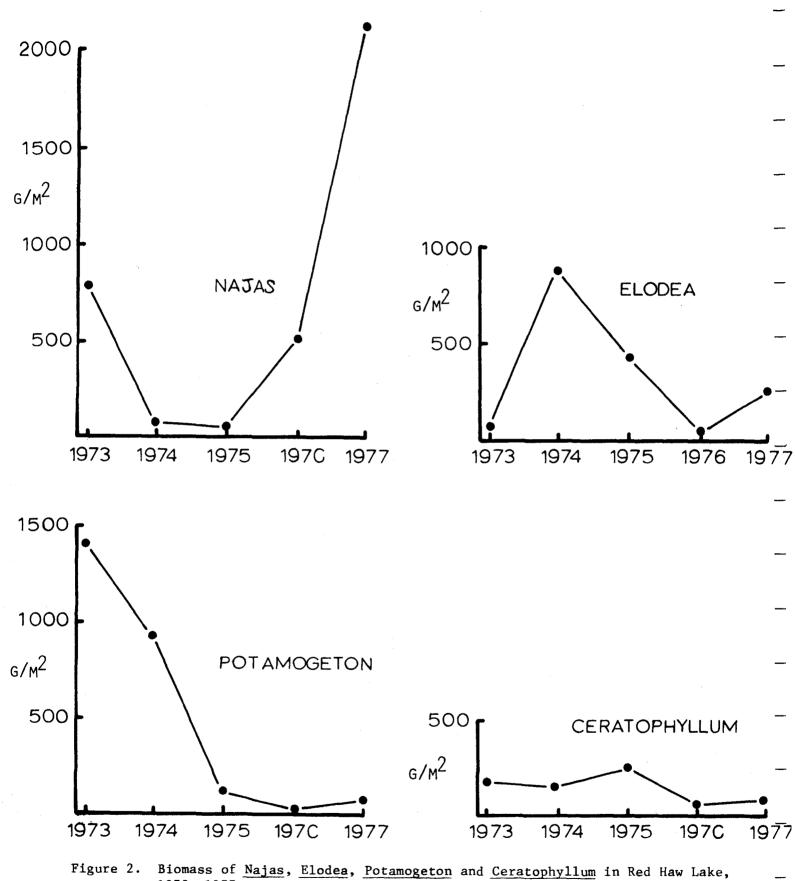
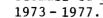
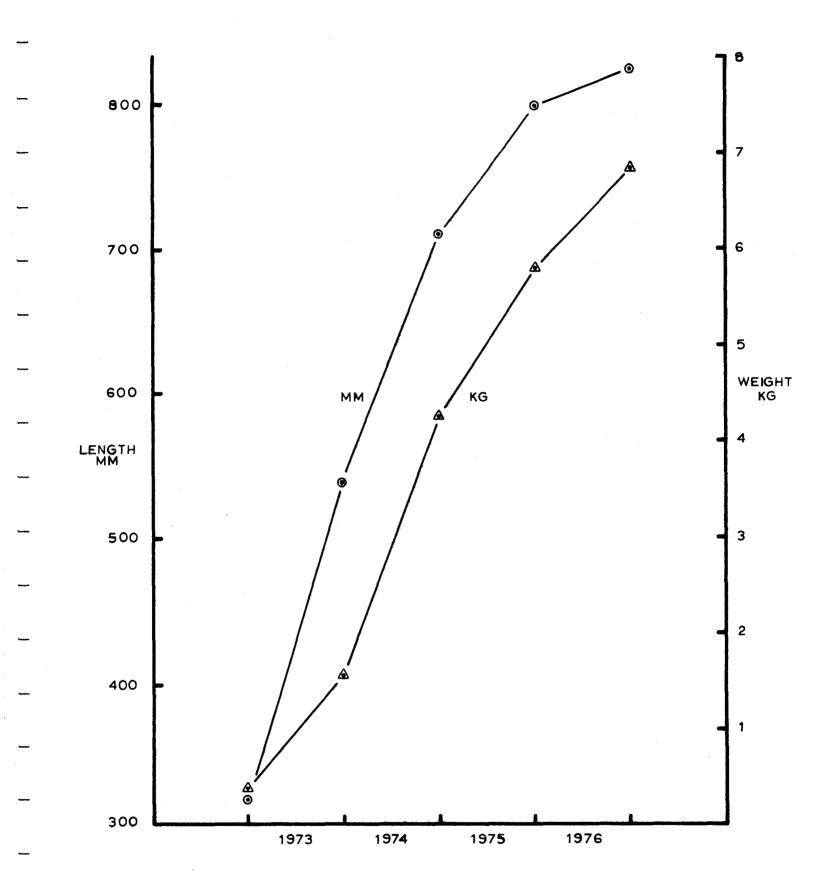
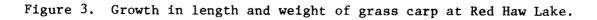


Figure 1. Grass carp density and vegetation biomass 1973-1977 in Red Haw Lake, Iowa.









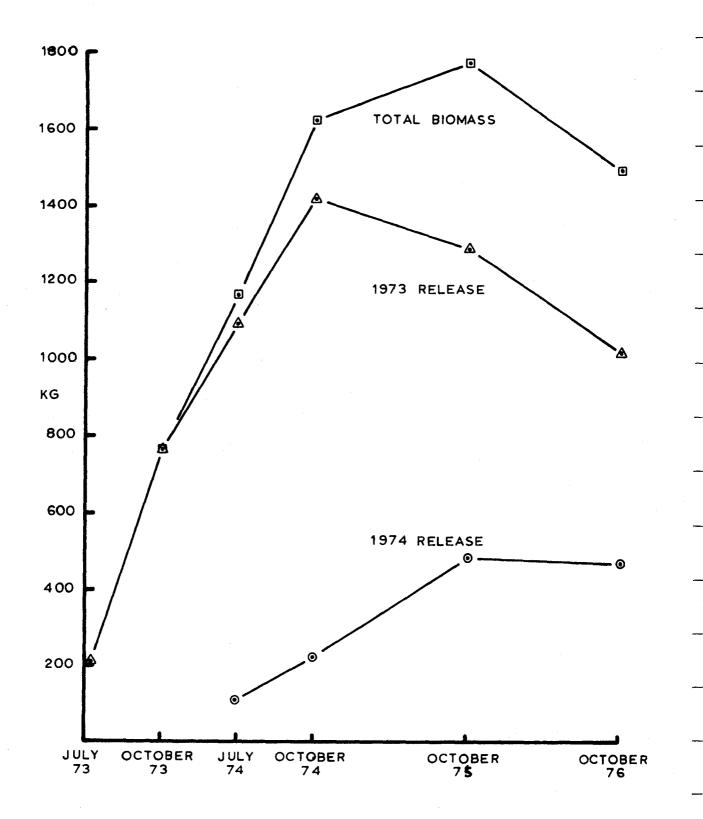


Figure 4. Biomass estimates of grass carp at Red Haw Lake from populations released in 1973 and 1974.

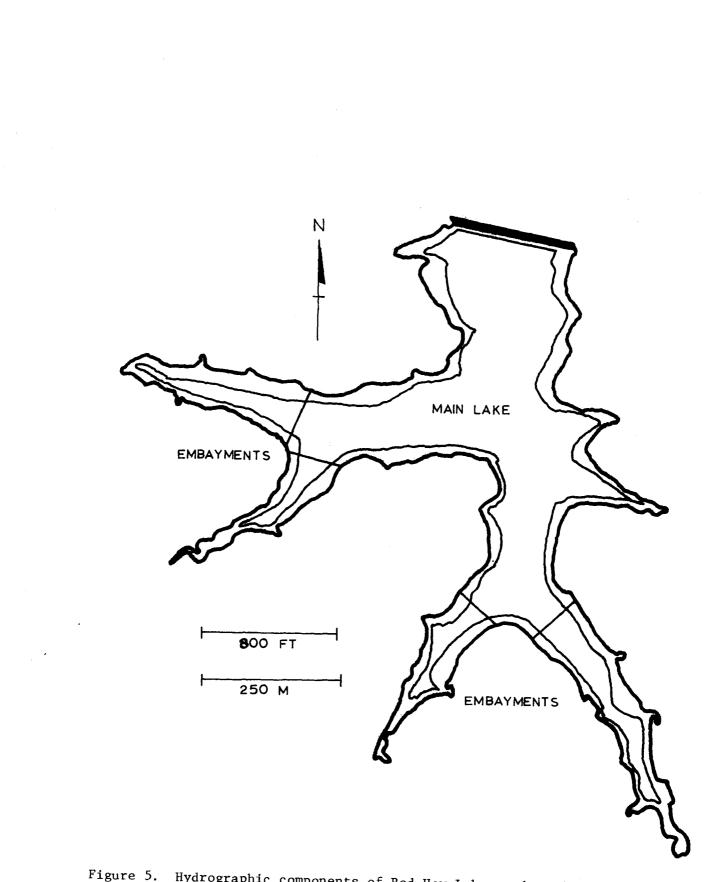
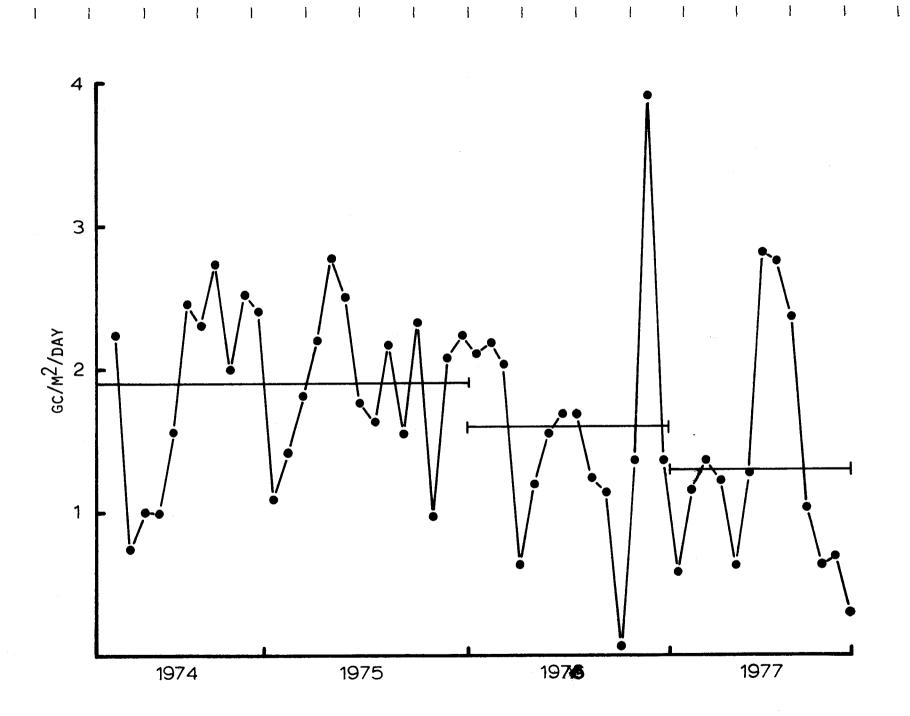


Figure 5. Hydrographic components of Red Haw Lake used to describe the distribution of contact locations for sonically tagged grass carp where deep and shallow water components were separated by the 10 ft contour.



1

1

ł

}

ł

J

ł

ł

١

Figure 6. Primary productivity values for Red Haw Lake, 1974-1977.

46

1

ł

ł

ł

1

1

ļ

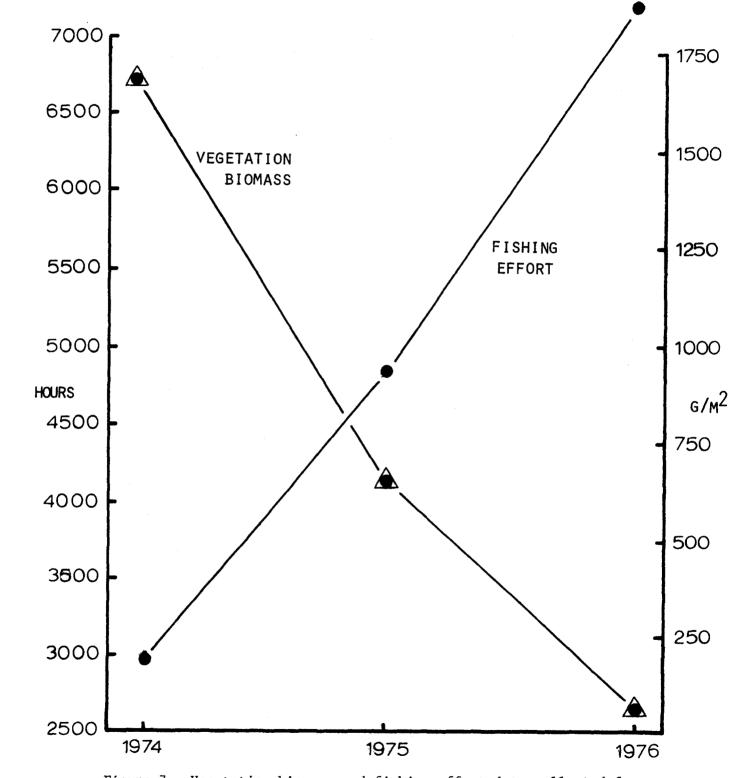
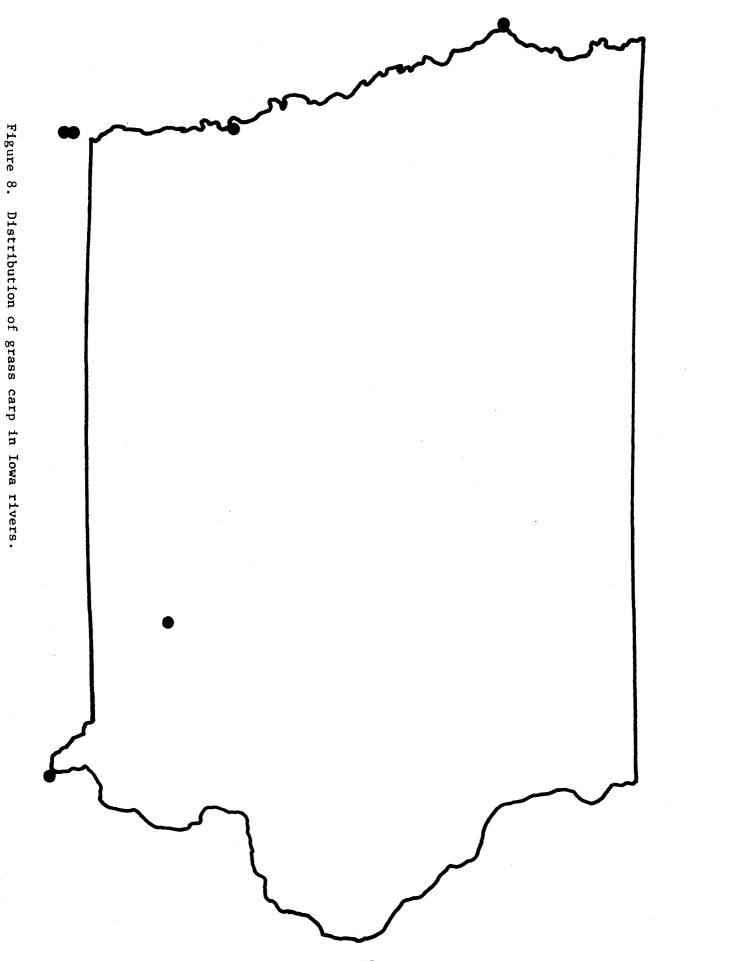


Figure 7. Vegetation biomass and fishing effort data collected from Red Haw Lake, 1974-1977.



ECOLOGICAL STUDY OF LAKE WALES, FLORIDA

AFTER INTRODUCTION OF GRASS CARP (CTENOPHARYNGODON IDELLA)

BY

Jerome V. Shireman, Douglas E. Colle and Randy G. Martin School of Forest Resources and Conservation University of Florida, Gainesville, Florida

INTRODUCTION

Noxious aquatic weeds are a serious problem over much of the United States, especially in Florida where a warm semi tropical climate increases their growth rate. In addition, certain exotic plants, such as water hyacinth (Eichhornia crassipes), Eurasion milfoil (Myriophyllum spicatum) and Florida elodea (Hydrilla verticillata) have become established in Florida waters. Increased nutrification in our rivers and streams, plus a deficiency of natural enemies has resulted in their fast growth rates and rapid spread.

Aquatic weed control in the past has mainly been accomplished through chemical and mechanical means. These methods are expensive, temporary and often degrading to the environment. Biological control would be less expensive, more permanent, and less hazardous. For these reasons, there has been a growing interest in the use of biological control organisms for weed control. This approach has been largely unsuccessful due to the difficulty in finding organisms that either consume or kill large quantities of unwanted plants without harming beneficial plants and other fish. The grass carp (Ctenopharyngodon idella) is being studied for this purpose in Lake Wales, Florida.

STUDY AREA

Lake Wales is a 133.5 ha lake located within the city of Lake Wales, Florida, in Polk County (lat. 27°34'13", long. 81°34'44") within the Lake Wales ridge. This ridge is an area characterized by porous permeable deep sand which does not generally permit surface runoff (Stewart 1966). Most of the circular shaped closed lakes in the Lake Wales ridge are believed to be of solution origin.

Lake Wales has a drainage basin of 6.27 sq km, most of which is located within the urban area. Several storm drains enter the lake causing considerable turbidity near their outlets during and after heavy rains. The lake water level responds to surface runoff, but the lake is connected to the piezometric surface of the Floridan aquifer and shows seasonal fluctuations characteristic of the aquifer. The most outstanding feature of the lake is the dense stand of <u>Hydrilla</u> <u>verticillata</u> which covered approximately 80% of the surface during summer months. Shallows and flats (0.5-1.0 m) are vegetated primarily by <u>Vallisneria americana</u> interspersed with hydrilla. Species of plants along shoreline areas (except for dredged areas along the north shore) are cattail (<u>Typha latifolia</u>), hydrocotyle (<u>Hydrocotyle runculoides</u>), panicum (<u>Panicum</u> sp.), fuirena (<u>Fuirena spp.</u>) and spikerush (<u>Eleocharis sp.</u>).

The lake bottom is similar in characteristic to most solution basins with gradual sloping contours. The maximum depth is approximately 5 meters with a mean depth of approximately 2 meters. The bottom is composed primarily of sand with detrital deposits in deep areas. Several small sink-holes are located along the western shore. One of these is connected to the lake only during high water stages. Study stations were chosen to depict different vegetation types, various water depth, and open water.

Grass carp were introduced into Lake Wales during October, 1974 with an initial stock of 11,053 fish (150 to 300 mm TL) by the Florida Game and Fresh Water Fish Commission. Because hydrilla was not controlled by the first stocking, an additional 50,000 grass carp fingerlings (100 to 150 mm TL) were introduced in January 1977 by the Florida Game and Fresh Water Fish Commission.

Sampling Stations

Eleven stations were selected by the Florida Game and Fresh Water Fish Commission prior to the initiation of the study. Two additional stations were added one month (Station 9) and three months (Station 8) into the study (Figure 1). For statistical data analysis, stations were pooled into six "collection sites" based on similarity of vegetation type and water depth.

Site 1

Stations 2 and 11 make up site 1 which represents the <u>Vallisneria</u> community. This site is characterized by relatively shallow water (1 to 2 m), with <u>Vallisneria</u> being the dominant vegetation, intermixed with small amounts of hydrilla. Percent light transmittance was always > 1.1% and oxygen levels > 4.6 mg/l at the bottom. Bottom type is composed of 100% sand.

Site 2

Stations 1 and 4 represent site 2 which typifies the shallow water hydrilla community. Station depth ranged from 2 to 3 m with <u>Hydrilla</u> being the only submerged vegetation. Bottom type is 60% sand, 40% muck. Bottom percent light transmittance was always > 1.1% and bottom oxygen levels ranged from 0.8 to 2.3 mg/l seasonally.

Site 3

Stations 3 and 5 are hydrilla stations with a depth of 3 to 4 m. <u>Hydrilla</u> surface matted during the warmer months for the first two years of the study, but has exhibited a decrease during the third year. Bottom light transmittance was < 1% during seasons of peak hydrilla growth, with bottom oxygen

levels ranging seasonally from 1.3 to 8.0 mg/l. The bottom is composed of 75% sand, 25% muck.

Site 4

Stations 6 and 7 ranged from 3.5 to 4.75 m deep, and are characterized by near surface mats of dense hydrilla during the warmer seasons. Hydrilla has decreased at these stations during the third year. Site 4 is characterized by bottom light readings being < 1% for all seasons except the winter of 1975. The depth to compensation point (1% light transmittance) has increased with the corresponding reduction in hydrilla height. Bottom oxygen levels fluctuate seasonally from 0.0 to 6.9 mg/l. The bottom is composed of 90% muck, 10% sand.

Site 5.

Station 9 is located in the cove at the southeast end of the lake. Hydrilla has remained at or near the surface during the warmer seasons throughout the study. The station is located near the opening of a storm drain, which causes considerable turbidity during heavy rains. Station depth ranged from 2.5 to 3.5 m, with bottom type being 50% sand and 50% muck.

Site 6

Stations 8, 10, 12 and 13 represent the deep water hydrilla site located in the center of the lake. Station depths ranged from 3.7 to 5.1 meters, depending upon the water level stage. This site was characterized by dense hydrilla surface mats during the first two years of the study, followed by complete elimination of all hydrilla during the third year. Depth to the compensation point has doubled with the elimination of hydrilla from site 6, with bottom light levels also doubling due to less shading from hydrilla. Bottom oxygen levels have shown an increase during the warmer seasons with elimination of hydrilla. Bottom type is 75% muck and 25% sand.

METHODS

Oxygen and temperature measurements were determined monthly with a Yellow Springs temperature and oxygen probe. Hydrogen ion concentrations were taken with a portable pH meter, while light penetration was determined with a Whitney-Montedero undewater photometer. Percent light transmission was determined by comparing incident light striking a deck cell with that of an underwater cell, from these readings the compensation point was determined.

Water chemistry values were determined from samples collected from the entire water column. All water samples were analyzed at the Game and Fresh Water Fish Commission, Eustis Water Chemistry Laboratory. Water samples were collected during one day, placed on ice, transported to the Eustis laboratory and determined the following day. These determinations included ortho-phosphate, total phosphate, sulfate, ammonia nitrogen, nitrate nigrogen, organic nitrogen, chloride ion, iron potassium, sodium, magnesium, calcium, turbidity, and chlorophyll a. Duplicate zooplankton samples were collected monthly at each major station at one meter increments with a Van Doran bottle. The two liter samples obtained in this manner were concentrated and brought to the laboratory for identification and enumeration. Three to ten counts were made of each sample in a Sedgewick rafter counting cell the first year and a Ward 5 ml. plankton wheel thereafter. Organisms were identified to species when possible. Data were analyzed with analysis of variance according to season, depth, site and time. Duncan's multiple range tests were used to determine significant differences between catagories with significance at ≤ 0.05 . Step-wise multiple regression methods were used to determine relationships among all parameters.

Duplicate benchic samples were collected at all stations with a 232.3 $\rm cm^2$ Ponar dredge and preserved in ethanol and rose bengal stain. Stained organisms were easily seen during separation. The entire sample was washed through a series of sieves. All organisms retained in a U.S. standard #30 seive were counted and identified.

Fish were sampled quarterly for weight-length and food habit analysis^{1/}. These were diel samples, where fish were collected for one hour every two or three hours over a 24 hour period. Ten fish of each major species were collected at each interval, weighed to the nearest 0.1 gram and measured to the nearest mm. Stomachs were removed for food habit analysis. This procedure was utilized from February, 1975 through June, 1976. Thereafter, diel samples were designed to sample larger segments of the three major sport fish populations. Weight-length relationships were determined for these fish.

RESULTS

Physical Parameters

Water level, depth to compensation point, and mean vegetation height^{2/} for site 4 and 6 are shown in Figures 2 and 3, respectively. The two sites represented the areas of heaviest hydrilla infestation at the initiation of the study. Depth to the compensation point was found to be directly related to hydrilla height at both sites, and was greatly reduced during periods of peak hydrilla growth. Hydrilla levels fluctuated almost identically during the first two study years, with growth peaks in the warmer seasons, and a characteristic winter dieback. During the third study year (spring 1977) hydrilla was eliminated at site 6 and has not reappeared. A 90% reduction in hydrilla height occurred at site 4 during the third year, and surface mats have not occurred elsewhere in the lake, whereas numerous mats occurred during the initial two years. The reduction in hydrilla coincides with the second stocking of grass carp into Lake Wales.

Oxygen and Temperature

Oxygen and temperature profiles indicate slight seasonal temperature stratification with more pronounced seasonal oxygen stratification (Figure 4).

2 / Obtained from Florida Game and Fresh Water Fish Commission, Lake Wales.

<u>1</u> / Food analysis being conducted by Florida Game and Fresh Water fish Commission personnel.

Oxygen stratification was strongly dependent upon the amount and type of vegetation present. At the shallower <u>Vallisneria</u> site (Site 2) oxygen levels remained above 5 mg/l throughout the year, with only slight stratification occurring during the warmer months. Oxygen stratification was more pronounced at the hydrilla sites, with the bottom reading approaching 0 mg/l during periods of maximum hydrilla growth (summer 1975 and 1976). The elimination of hydrilla during 1977 at site 6 corresponded with an increase in bottom oxygen levels. Surface oxygen values for 1977 decreased when compared to the two previous years. The trend in oxygen to increasing bottom levels and decreasing surface readings may indicate more uniform lake mixing due to the decrease in vegetation height. Rottmann and Anderson (In Press) observed similar increases in oxygen levels when surface mats of vegetation were removed by grass carp.

Water Quality

The location within the lake from which samples were collected did not significantly effect any of the 16 water quality parameters monitored. Therefore, samples from each station were pooled within seasons for further statistical analysis. Sulfate, pH, and potassium were the only parameters that differed significantly and appeared to be correlated with hydrilla reduction. Sulfate levels were significantly reduced the last two study years; however, there were no significant differences between the two years. Analysis of variance testing revealed no significant seasonal or chronological trends for magnesium, iron, conductivity and turbidity, ortho phosphate, total phosphate and nitrate nitrogen.

In 1977, pH was significantly lower than the two previous years. Values ranged from 8.2 to 9.3 in 1975 and 1976, while values in 1977 were from 7.4 to 7.7. The elevated pH levels in 1975 and 1976 are the result of high utilization of free CO, and possibly bicarbonate by hydrilla. Although values were below lethal limits for bluegills, 10.4, (Trama 1954) during 1975 and 1976, they were higher than values considered optimum for aquatic fauna. Of United States waters that support good fish fauna, only 5 percent have pH values less than 6.7, 50 percent have pH less than 7.6, and in 95 percent the pH is less than 8.3 (Hart et al. 1945). Rottmann (1976), Buck et al. (1975), and Beach et al. (1976) reported significant decreases in pH values after grass carp controlled the vegetation in ponds and pools.

Potassium levels increased 300 to 400% in 1977 as opposed to prior years (Figure 5). Values in 1977 were significantly greater than all seasons except the winter of 1976. Avault et al. (1968) reported increases of greater than 300% in potassium levels after grass carp introduction. Lembi et al. (1978) attributed the significant increases in potassium levels after the introduction of grass carp to low incorporation of potassium into fish tissue, and slow removal from the water column. They concluded that potassium levels could be used as indicators of grass carp feeding. Lake Wales results substantiate the potassium theory, as the significant potassium increases coincided with hydrilla reduction in 1977.

Organic and ammonia nitrogen levels varied seasonally, but were not significant through years. Significant seasonal peaks occurred in the spring of 1976 and 1977 and the winter of 1975. The seasonal extremes were not significantly different from each other, with a range of 1.05 to 1.15 mg/1. Ammonia nitrogen also exhibited significant seasonal peaks in each study year; however, among years, they were not significantly different. These peaks occurred in the fall 1974, fall 1976, and the spring and summer of 1977, with values of 0.087, 0.096, 0.097, and 0.105 mg/1 respectively.

Sodium, calcium, and chloride levels differed significantly through seasons (Figure 6); however, these differences were found to be seasonally cyclic for each study year. Sodium values usually peaked during the winter and spring months in all years, with lower levels occurring in the fall and summer. Although Reid (1961) states the limnological importance and behavior of sodium and potassium are quite similar; in Lake Wales, sodium did not serve as an indicator of grass carp feeding as did potassium. Calicum and chloride seasonal cycles were characterized by significant winter peaks coinciding with the winter senescense of hydrilla, and significantly lower levels in summer when hydrilla was at its maximum. Seasonal cycles within 1977 were not characterized by the variation evident in the previous years. The trend toward stabilization in 1977 was probably related to the hydrilla decrease.

Chlorophyll a concentrations vary within species and with environmental and nutritional factors that do not necessarily reflect phytoplankton standing crop (Weber and Strumm 1963); they do, however, provide insight into primary production. Significant peaks occurred in chlorophyll a concentrations during the fall, 1975, summer, 1976, and spring, 1977. There were no significant differences among these peaks, thus indicating a seasonal cycle that was similar in all study years. Rottmann (1976), Beach et al. (1976) and Terrell (1975), reported insignificant changes in chlorophyll a values and/ or no apparent algal blooms after grass carp had controlled submerged vegetation. The introduction of grass carp has, however, resulted in algal blooms (Alikunhi and Sukumaran 1964, Prouse 1969).

Zooplankton

Statistical analysis of zooplankton populations was confined to the following predominant groups of net plankton: calanoid and cyclopoid copepods, nauplii; eight cladocera species, <u>Diaphanosoma brachyurum</u>, <u>Daphnia</u> <u>ambigua</u>, <u>Alona costata</u>, <u>Camptocerus certirostris</u>, <u>Chydorus sp.</u>, <u>Macrothrix</u> <u>rosea</u>, <u>Ceriodaphnia pulchella</u> and <u>Bosmina coregoni</u>; and <u>rotifers</u> (predominantly <u>Keratella</u> and <u>Corochilus</u>). These organisms were present in all seasons with the exception of <u>Daphnia ambigua</u>, <u>Macrothrix rosea</u>, <u>Camptocercus</u> <u>rectirostris</u>, and <u>Chydorus</u> sp. which did not occur in either one or two seasons. Two additional cladocerans (<u>Simocephalus</u> and <u>Eurycercus lamellatus</u>) were recorded in plankton samples, but their occurrences were sporadic and could not be analyzed statistically. Analysis of variance revealed no consistant significant site differences within any of the zooplankton categories; therefore, sites were pooled within seasons for analysis.

Calanoid copepods and the cladoceran <u>Diaphanosoma</u> <u>brachyurum</u> were the only zooplanktors that revealed significant chronological change, with both groups consistently decreasing through time. Other zooplankton groups

exhibited only significant seasonal differences, which were repetitive for each study year. Calanoid copepods peaked during the summer, 1975, then began a significant decline during the summer of 1976, with a reduction in mean numbers of organsism per liter from 39.4 to 16.2 after the summer of 1976 (Figure 7). <u>Diaphanosoma</u> density (Figure 8) has shown a significant temporal decrease throughout the study. Seasonal peaks in <u>Diaphanosoma</u> numbers reflect this chronological decline; mean density during the summer of 1975 was 75.2 organisms per liter, fall of 1976, 17.8, and spring 1977, 9.0 organisms per liter.

The great majority of limnetic communities are characterized at one time by one numerically dominant copepod and cladoceran species (Pennak 1957). Ceriodaphnia pulchella was the dominant cladoceran collected during the winter of 1975 with Diaphanosoma brachyurum being second in abundance. For the remainder of 1975 Diaphanosoma was dominant, however, when it began to decline in 1976, Bosmina coregoni became most abundant during the winter seasons, with Ceriodaphnia pulchella dominant during the spring of 1976 and 1977 and the summer of 1977. Macrothrix rosea was the most abundant cladoceran during the summer of 1976. The shift in dominance from Diaphanosoma brachyurum (length 2-3 mm, Shireman 1976) to the smaller Bosmina coregoni (length 0.3-0.5 mm, Pennak 1957) and Ceriodaphnia pulchella (length 0.5 to 0.7 mm, Pennak 1957) may be a direct effect of habitat change or an indirect effect due to increased predation on the larger zooplanktors after vegetation decreased. Lake Wales has shifted from an entirely littoral habitat characterized by 100% cover of submerged vegetation to a more classical littoral-limnetic community. Edmondson (1959) noted Diaphanosoma brachyurum is a littoral cladoceran associated with marshes and weedy margins of lakes, whereas Bosmina coregoni is characterized as a limnetic species. Brooks (1968) found introductions of obligate planktivores resulted in immediate shifts in cladoceran dominance from larger Daphnia species to the much smaller cladoceran Bosmina longirostis; Brooks also noted reductions in the larger calanoid copepods. He felt introductions of facultative planktivores would also cause shifts to smaller cladocerans, but the change would be to intermediate cladocerans such as Ceriodaphnia rather than the smaller plantors such as Bosmina. Nordlie (1976) indicated considerable variability in species composition occurred among the three central Florida lakes he studied. He suggested that differences may be due to the presence of predators, for the lakes with the greatest number of predators were characterized by small cladocera. The decrease in submerged vegetation in Lake Wales may have resulted in an increased rate of predation upon the larger net planktors (calanoid copepods and the cladoceran Diaphanosoma brachyurum) which resulted in decreased density and a shift to dominance by the smaller cladoceran Bosmina coregoni.

Significant summer peaks in density occurred for the cladocerans <u>Alona</u> <u>costata</u> and <u>Macrothrix rosea</u>, and cyclopoid copepods. When mean water temperatures were 29.5 <u>C</u>. <u>Alona</u> and <u>Macrothrix</u> populations were characterized by residual densities (< 5 organisms per liter) during all seasons except for summer blooms. Although significant summer increases of <u>Alona</u> occurred in the summer of 1975 and 1976, they did not increase in 1977. The highest density of cyclopoid copepods occurred during the winter of 1976, however peak numbers during 1975 and 1977 were documented in the summer samples. When cyclopoid numbers within seasons are compared, against years, a slight decrease in numbers is evident during 1977; however, only yearly peaks were

significant. The summer peak of 1977 (40.3 organisms per liter) was significantly lower than the winter peak of 1976 (116.1) and the summer peak of 1975 (63.5). Copepod nauplii (Figure 7) closely parallel the seasonal variations of the adult cyclopoids. Greatest density occurred during the summer of 1975, winter of 1976, and winter and summer of 1977. Unlike adult cyclopoid copepods, numbers did not decrease significantly in 1977. The winter peak of 1976 was significantly greater than the other seasonal peaks.

"Winter blooms" were found for <u>Bosmina coregoni</u> and <u>Daphnia</u> <u>ambigua</u> when mean water temperatures ranged from 20.0 C. in 1975 to 15.0 C. in 1977. Both species were characterized by tremendous increases in numbers during the winter, with reminant populations occurring during the other seasons. Seasonal differences were cyclic for all years, with chronological differences occurring only for <u>Bosmina coregoni</u> which increased the last two study years. <u>Bosmina</u> winter peaks have significantly increased from 23.7 organisms per liter in 1975 to 184.2 and 176.6 in 1976 and 1977 respectively.

<u>Ceriodaphnia pulchella</u> and <u>Chydorus</u> sp. both exhibited significant spring peaks in all study years which was preceded by an initial winter density increase, followed by significant decreases to residual populations during the summer and fall. Ceriodaphnia annual maximum densities have consistently increased throughout the study. The winter peak of 1975 (36.9 organisms per liter) was significantly lower than the spring maxima of 59.4 in 1976 and 75.5 organisms per liter in 1977. <u>Chydorus</u> numbers have not shown any significant change throughout the study.

<u>Camptocercus rectirostris</u> increased significantly in abundance during the fall, 1974 and 1976, but significant chronological trends in abundance did not exist. <u>Camptocercus</u> was a minor cladoceran with respect to abundance, with maximum numbers being only 9.4 organisms per liter. Its density was usually less than 3.6 organisms per liter excluding the periods of "fall bloom".

Rotifers also exhibited a significant fall abundance peak, but only in 1975, which was due primarily to massive blooms of a colonial rotifer, <u>Conochilus unicornis</u>. Rotifers exhibited the widest variability of any net plankters, ranging in density from 13.5 organisms per liter in the summer of 1976 to 709.4 organisms per liter in the spring of 1977. The predominant genera, excluding the fall sample of 1975, was <u>Keratella</u>, which was entirely responsible for the rotifer bloom in 1977. Mean numbers of rotifers per liter were significantly greater in 1977 than any other season, except for the fall bloom in 1975 of <u>Conochilus unicornis</u>. Their density remained greater than 315 organisms per liter throughout the entire year.

Stepwise regression analysis (Table 1) was performed to identify significant environmental factors that influenced net plankton abundance, and to predict the effect changes in these parameters might have upon zooplankton numbers. Variables found to be significant with only one or two planktors included magnesium, iron, organic nitrogen, potassium and, surprisingly, temperature. This might indicate that iron, magnesium, organic nigrogen and potassium levels were never low enough or conversely high enough to limit net plankton abundance. The lack of significance for temperature, except for <u>Camptocercus rectirostris</u>, seems to indicate the seasonal cycles observed in net plankton abundance are not, in effect, temperature related. However, the relationship between temperature and zooplankton may be a nonlinear function, rather than a linear one, which was used in the stepwise analysis.

Calcium and orthophosphate levels apparently exhibited depressing effects upon the zooplankters they were correlated with, while sulfate levels were positively correlated with four of the eleven organisms tested (Table 1). Chlorophyll a and pH were significant with 54% of the dependent variables. Clorophyll a levels were used to estimate phytoplankton density. Therefore a positive relationship between chlorophyll a and zooplankters might indicate that these organisms are dependent upon phytoplankton as a food source.

Copepod nauplii, <u>Alona costata</u>, <u>Bosmina coregoni</u>, <u>Daphnia ambigua</u> and <u>Chydorus sp. had significant positive relationships to phytoplankton density</u> whereas <u>Ceriodaphnia pulchella</u> was negatively correlated (Table 1). Cyclopoid and calanoid copepod adults, <u>Diaphanosoma brachyurum and Macrothrix</u> rosea correlations were not significantly related to phytoplankton abundance.

Hydrogen ion significantly declined during the study, as did calanoid copepods and <u>Diaphanosoma</u>. Both <u>Chydorus</u> and <u>Alona</u> were positively correlated with pH. Winter blooms of both <u>Bosmina</u> and <u>Daphnia</u> occurred during the winter when pH was low.

Correlation with water depth indicate vertical distribution whenever significance occurred (Table 1). Nauplii were negatively correlated indicating they were distributed at lower depth; whereas <u>Camptocercus rectirostris</u>, <u>Bosmina coregoni</u>, <u>Daphnia ambigua</u>, and <u>Ceriodaphnia pulchella</u> occurred near the surface. The lack of significance with depth for the remaining net plankters indicate they were randomly distributed within the water column.

Examination of r^2 values indicate that less than 50% of the variation for any organism was accounted for by the variables tested. The remaining variation was probably due to other environmental factors as well as to sample variation (Table 1).

Benthos

A species list of benthic organisms collected from Lake Wales with their frequency of occurrence is presented in Table 2. Statistical analysis was performed only upon taxons having frequent occurrences. These included: Chaoborus larvae; Chironomidae larvae; the amphipod Hyalella azteca; the bristle-worm Naidae and other Oligochaetia; trichopteran larvae (Orthotrichia, Oxeythira, Polycentropus, and Leptocella); and gastropods (Gyraulus parva, Helisoma duryi, and Physa). Chaoborus larvae distributions were highly significant, with maximum densities occurring at sites 4 and 6. Gastropod populations (Helisoma duryi and Physa) and trichopterans (Polycentropus and Leptocella) were most abundant at Vallisneria sites; while the gastropod Gyraulus parva exhibited significant preference for both Vallisneria and shallow water Hydrilla collection sites. Chironomid larvae and oligochaetes were more abundant in the samples collected in less than 4 meter depths (Sites 1, 2, 3); with both having significant density reductions in all seasons in the deep water locations (Sites 4 and 6). The trichopteran larvae, Oxyethira and Orthotrichia, did not exhibit site preferences. Significant chronological increases occurred in Chaoborus larvae and Oligochaetes beginning with the summer sample of 1976. The trichopterans, Polycentropus and Leptocella, both increased significantly in abundance during 1976 and 1977. Hyalella azteca and <u>Gyraulus</u> parva both underwent significant population declines. Amphipods

were not collected at either site 4 or 6 during 1977 and <u>Gyraulus</u> numbers began to deceline significantly during the spring of 1976. This decline continued for the remainder of the study. <u>Physa</u>, chironomid larvae, <u>Orthotrichia</u> and <u>Oxyethira</u> populations did not exhibit significant chronological changes in abundance and revealed only seasonal peaks that were repetitive in all study years.

Chaoborids

Differences in Chaoborus larvae numbers were highly significant for sample locations. Chronological changes occurred at the deeper sample sites, where the larvae were most abundant (Figure 9). Maximal densities were obtained during the summer and fall samples with corresponding decreases during winter collections. Vallisneria flats (Site 1) were characterized by having extremely low Chaoborus numbers throughout the study and significant chronological changes in abundance did not occur at this site. Although numbers were not statistically significant, a trend was evident in the spring and summer of 1977 towards increased Chaoborid densities. Chaoborus abundance at the shallow hydrilla stations (Site 2) closely parallel Vallisneria-Chaoborus densities except for significant seasonal peaks in the summer and fall of 1975 and 1976. Although the number of Chaoborus collected during the summer of 1977 was not significantly different, it appears the cyclic summer and fall abundance of phantom midge larvae in 1975 and 1976 will be repeated in 1977. Chaoborus larvae were usually not the dominant benthic invertebrate collected at Vallisneria stations, but were the most abundant benthic organism within the shallow hydrilla site during the summer density peak of 1975. The deeper hydrilla stations (Sites 3,4,5, and 6), during all seasons, had significantly greater phantom midge populations than the inshore sites. Two of these stations (4 and 6) generally had the greatest phantom midge density and dominance throughout the study, with both revealing a significant chronological increase. Chaoborus numbers have also increased at mid-depth hydrilla stations (Site 3), but in only one season were the numbers significantly greater (summer 1977). The organisms were predominant at site 3 only during the winter of 1975, whereas they became the most numerous organism during all seasons in 1976, and during the spring and summer samples of 1977. Phantom midge populations at site 4, began increasing during the summer of 1976, with densities remaining above 740 $organisms/m^2$ through the summer of 1977. Chaoborid abundance was greatest during the summer of 1977 at site 4 (2280 organisms/ m^2) and were the dominant benethic organism during all seasons at this site, except for the winter sample of 1976. Benthic cove samples (Site 5) are not presented in Figure 10; however, significant chronological increases occurred in chaoborid densities at this site due to a 50% increase in the seasonal peaks of 1976 and 1977. Significant peaks in abundance occurred during the winter and summer collections of 1976 (588.5 organisms/m²) and the spring and summer samples of 1977 (624.4 organism/ m^2), as opposed to the lower maximum in the summer of 1975 (236.8 organisms/ m^2). Examination of cove sample benthic populations also indicated a chronological change, with seasonal dominance shifting from chironomids and Chaoborus prior to the 1976 summer sample to total Chaoborus dominance for the remainder of the study. Chaoborus densities have increased consistently in the deeper offshore sample locations (Site 6), with progressive significant increases in yearly peaks (Figure 9). Phantom midge densities were greater than any other benthic organisms collected during all seasons at the deep water stations, except for the fall of 1974 and the spring of 1975.

Chironomids

Chironomid larvae abundance exhibited little significant chronological change at any of the collection sites (Figure 10). Significant site differences occurred seasonally, with an overall trend towards higher chironomid densities at inshore locations (1 and 2). Chironomid densities varied greatly at Vallisneria stations, with numbers usually being as great or significantly greater than the other sites. A significant increase in abundance occurred from the summer of 1975 to the winter of 1977, with both the remaining seasons of 1975 and 1977 being not statistically different from each other. Chironomids were the dominant benthic invertebrate in the Vallisneria collections prior to the summer of 1976. Shallow water hydrilla samples were characterized by relatively large chironomid larvae densities, but significant yearly differences were not indicated. Midge larvae were the dominant benthic organisms at Site 2 during the collections of 1975 and 1976, and the winter of 1977. Chironomids were the most abundant benthic invertebrates at site 3 during 1975, however a shift to Chaoborid dominance occurred during both 1976 and 1977. Midge abundance at the two offshore hydrilla sites (4 and 6) fluctuated identically. Both sites had significantly fewer chironomid larvae than the shallower stations. Chironomids were the most abundant benthis organism in only three seasons at the offshore site; winter 1976 (Site 4), fall 1974 and spring 1975 at site 6. Significant yearly cycles of abundance occurred at both sites with density peaks in the winter of 1976 and 1977 at site 4, and during the fall samples of every year at site 6. No significant difference occurred in the magnitude of the peaks among years at either site. Midge abundance did not change significantly at site 5 throughout the study, but significant differences occurred between the fall peaks of 1974 and the summer peaks of 1976. Chironomids were the dominant organism at this site prior to the summer of 1976.

Oligochaetes

Significant yearly increases in oligochaete abundance occurred at all sample sites (Figure 11). Site differences were also significant with large populations occurring at the shallow sample locations (Sites 1, 2, and 3). Prior to the summer samples of 1976, oligochaetes were confined primarily to Vallisneria sites; however, beginning with the summer 1976, a marked increase in oligochaete abundance was recorded at all locations. After spring 1976, oligochaetes replaced chironomids as the dominant benthic invertebrate at site 1 and remained the most abundant organism at site 1 for the duration of the study. Oligochaete densities revealed high significant temporal differences in the shallow hydrilla samples with densities increasing from less than 30 organisms per square meter prior to the summer of 1976, to over 340 organisms per square meter for the duration of the study. Oligochaetes were not found in appreciable amounts at the deep hydrilla sites (3-5) until summer 1976, when their abundance increased significantly at all sites. Oligochaetes were dominant only in the benthic samples of site 3 during the winter of 1977; their numbers, however, never approached Chaoborus densities at any of the other deep water hydrilla sites. Pennak (1953) states aquatic oligochaetes occupy a niche which is equivalent to that of the terrestrial species, and feed on bottom mud mixing it as effectively as earthworms mix surface layers of garden and meadow soils. The increase in oligochaetes cannot be explained, but may be related to increased detrital deposits associated with hydrilla control in Lake Wales. The bristly-worm, Naidae, a freshwater oligochaete, also increased significantly

after the summer of 1976. Prior to this collection it had been collected only during two seasons. <u>Naidae</u> were present during all seasons after the summer of 1976, but abundance was not related to site differences.

Amphipods

Hyalella azteca revealed strong significance with respect to site, being predominant in the shallower stations. Amphipods were present during all but one season within the <u>Vallisneria</u> and shallow water hydrilla sites, whereas a marked yearly decrease occurred in abundance at the deeper sites. <u>Hyalella azteca</u> was not collected at the midwater sites, 4 and 6, during 1977. The elimination of amphipods at these sites was due to reduced hydrilla. Amphipods are strongly thigmotactic and react negatively to light, with daylight hours spent in vegetation or hidden under and between debris and stones (Pennak 1953). The elimination or reduction of hydrilla at the deeper sites rendered this area unsuitable for amphipod habitation.

Gastropods

Benthic gastropod populations were composed primarily of Gyraulus parva, with fewer numbers of <u>Helisoma</u> duryi and <u>Physa</u> sp. <u>Gyraulus</u> densities were significantly greater within Vallisneria and shallow water hydrilla sites during all seasons except the fall of 1975 and winter of 1976. Significant peaks in abundance occurred at the 3 to 4 meter hydrilla stations. The peak in Gyraulus abundance during the fall of 1976 and winter of 1976 closely parallels the increase in Gyraulus numbers in hydrilla samples collected during this same sample period. A significant chronological decrease occurred at all sample sites after the spring collection of 1976 and they were not collected during the summer and fall of 1976 and the spring and summer of 1977. Gyraulus was prevalent in the winter samples of 1977 at all sites, with densities equivalent to the previous years. Helisoma duryi was associated with Vallisneria, as 70% of them were collected in Vallisneria. Helisoma abundance did not change significantly through years; but significant seasonal density peaks were evident in the summer of 1975, the spring of 1976, and the winter of 1977. Physa occurred infrequently and only at Vallisneria and shallow water hydrilla stations. They were present in just five seasons: summer of 1975; spring, summer and fall of 1976; and spring of 1977.

Caddisflies

Caddisflies were represented primarily by the genus <u>Polycentropus</u>, with fewer numbers of case dwelling <u>Leptocella</u>, <u>Orthotrichia</u>, and <u>Oxyethira</u>. <u>Polycentropus</u> and <u>Leptocella</u> populations were highly significant as to site specificity, with densities being greater in <u>Vallisneria</u> for all but two seasons. These organisms exhibited significant winter peaks during each study year, with maximum densities (422.5 organisms/m²) occurring during the winter samples of 1976. A significant yearly increase in <u>Polycentropus</u> and <u>Leptocella</u> occurred during 1976 and 1977 as opposed to 1975, with 1976 collections significantly larger than the other years. <u>Oxyethira</u> and <u>Orthotrichia</u> did not occur in benthic samples prior to the fall sample of 1975, and exhibited no consistent significant site preference. Significant peaks in population abundance, for both genera, occurred in the summer and fall of 1976 and the winter of 1977; followed by a reduction in numbers during spring and summer of 1977. They occurred only at Vallisneria sites.

Benthic Correlations

Stepwise regression analysis (Table 3) was performed with benethic organisms to determine interrelationships with various environmental parameters. Conductivity, sulfates, magnesium, iron, nitrate nitrogen, ammonia nitrogen, and orthophosphate levels did not significantly effect the abundance of any of the benthic organisms. Significant positive relationships occurred between station depth and Chaoborus larvae densities. Bottom temperature and water level were also positively correlated with their abundance. They attained maximas during the warmest seasons. Oxyethira and Orthotrichia larvae were the only other benthic invertebrates positively related to depth. The remaining invertebrates were negatively influenced by depth, indicating preferences for shallow water. Chlorophyll a and chloride levels were positively correlated with benthic invertebrate numbers; other water quality parameters were negatively correlated with benthic invertebrates (Table 3). Oxygen, temperature and percent light at soil-water interfaces were positively correlated with certain organisms (Table 3). The gastropod, Gyraulus, was positively correlated with bottom oxygen levels. Pennak (1953) states that gastropods, under experimental conditions, cannot tolerate anaerobic conditions for more than 48 hours. Percent light transmittance was correlated with Polycentropus and Leptocella numbers. These organisms were found almost exclusively in Vallisneria flats where values for light transmittance were always much greater than any of the other sample sites. This correlation, although significant, may not be valid, but distributions may be due to habitat type. Bottom temperature was positively related to numbers of Chaoborus larvae, Oligochaetes, Polycentropus larvae and Leptocella larvae; both Chaoborus and Oligochaetes reached maximal densities during the warmer spring and summer months in all study years. However, the positive relationship with Polycentropus and Leptocella is surprising, for both were characterized by winter peaks during all study years.

Fish

Analysis of quarterly electrofishing data were confined to the three principal sport fishes collected: largemouth bass (Micropterus salmoides) bluegill (Lepomis macrochirus), and redear (Lepomis microlophus). Analysis of covariance utilizing logarithmic transformations was conducted to assess chronological changes in weight-length relationships. Fulton's coefficient of condition K=Weight in grams/ (Total length in millimeters)³ was calculated within seasons and size classes. In order to confine analysis to individual growth stanzas, fish were divided into biological size classes based upon literature values for shifts in food habits, sexual maturity, and age. Size classes were: 50-100 mm, 101-150 mm, and > 150 mm for bluegills and redears; and 75-150 mm, 151-250 mm, 251 - 350 mm, and > 350 mm for largemouth bass. Statistical analysis was limited to size groups in which ten or more individuals were present.

Sample size was adequate for all species during six seasons to test for sex differences in weight-length relationships. Analysis of covariance tests indicated no significant differences in any season or size class for bluegills (Talbe 4); significant differences in elevations occurred in three seasons. Regression line elevations for female bluegills were significantly greater during the summer of 1975 for fish 101 - 150 mm; indicating that females were heavier than males for a given length within the size class. Elevations for male bluegills > 151 mm were significantly greater during the winter of 1975 and summer of 1976. Sample size was adequate to assess sex differences in the redear 101 - 150 mm size group. Within this size group significant elevation differences occurred in the summer of 1976; indicating males were significantly plumper than females (Table 5). No significant slope differences between male and female redear were found in any of the other seasons.

The influence of sex, on weight-length relationships, was tested for < 251 mm. Larger bass were not sacrificed for food habits and therefore were not sexed except during the spring 1975 sample. Bass > 350 mm in this sample (1975) exhibited no significant sexual differences in line slopes or elevations. Significant slope differences were found only during the summer of 1976 for bass 151 - 250 mm (Table 6). These differences were probably due to unequal fish lengths within the size class, as male bass were generally larger. Van Der Avyle and Carlander (1977) state that in age-specific regression based on fish collected over a relatively short time period, the regression slope is a function of the difference between slow growing and fast growing individuals rather than growth of individuals. Elevation differences were significant for bass 151-250 mm in the winter collection of 1975, indicating males were plumper on the average for any given length.

For chronological weight-length analysis, sexes were pooled within seasons and analysis of covariance was conducted comparing equivalent seasons for the three study years. Examination of bluegill mean coefficients of condition (Table 7) indicates a chronological increase in condition, and an accompanying significant difference in elevation of weight-length regression for the two larger size classes. Sample size was sufficient during the spring of 1976 and 1977 for assessment of weight-length relationships in bluegills 50-100 mm. No significant slope or elevation differences occurred among these seasons for bluegills 50-100 mm. Significant slope differences for the two larger size classes were found only in the winter collections of 1975 and 1977. Length distributions within the size class reveal significance was due to the perponderence of longer fish in the 1977 collection. A significant increase occurred with each progressive year, with both largest condition values, and significantly higher elevations of weight-length regressions occurred only during 1977, with peak K(TL) occurring during the spring.

Redear coefficients of condition and weight-length relationships paralleled bluegills; with significant elevation differences also occurring only in the two larger size classes (Table 8). Only one significant slope difference was found for smaller redear (50-100 mm), which occurred between the spring of 1976 and 1977. However, neither of the slopes for the spring samples of 1976 and 1977 were significantly different from 1975. A trend toward maximal condition values in 1977 was evident during all seasons for the two larger redear size classes. Weight-length relationships also revealed significantly higher elevations for 1977; with the exception of the summer sample of redears > 151 mm in which no significant yearly differences occurred. Seasonal differences in K(TL) were noted in both 1975 and 1977 for redears > 151 mm; maximal values occurred in the summer of 1975 and spring of 1977.

Significant chronological increases in elevation of the weight-length regressions occurred in all size classes of largemouth bass; however, the majority of the significance was confined to bass <350 mm (Table 9). Coefficients of condition also revealed an increasing trend to maximal values in 1977. Analysis of covariance among years within seasons did not show significant differences in slopes for any of the regression lines. Significant elevation differences, in all cases, were characterized by higher elevations during all seasons in 1977, when compared to the two previous years. No significant elevation differences were found among years during the summer and winter for largemouth bass > 350 mm.

Comparisons of condition factors with other studies indicate values for Lake Wales bass, bluegill and redear were below the norm during the two years of heavy Hydrilla infestation. Carlander (1977) gives a central 50% range of K(TL) of 1.78-2.05 based upon the mean of studies conducted throughout North America. K(TL) for Lake Wales bluegills only approached or exceeded Carlander's mean K(TL) values during 1977, the year of reduced Hydrilla coverage. Coefficient of conditions for Lake Wales redear were also lower than values reported by Carlander, with K(TL) values approaching the norm only in 1977. Condition factors for Lake Wales largemouth bass did not increase as greatly as did values for larger bluegills and redears. Significant differences in weight-length relationships were confined primarily to bass < 350 mm. K(TL) values for Lake Wales bass were lower, in all study years, than those reported by Cooper et al. (1962) for Florida largemouth bass, and were equivalent to the lower range values given by Clugston (1964) for Florida waters. Both authors demonstrated correlation between condition and growth, with slow growth accompanied by a continuously low condition factor. Increasing condition factors probably indicate growth improvement for Lake Wales largemouth bass < 350 mm; however, the increase in condition was not as great in bass > 350 mm. This might indicate that condition and growth rates for Florida largemouth bass > 350 mm are not as severely influenced by conditions created by dense hydrilla mats.

Coefficient of condition and weight-length regression analysis indicated the dense stands of Hydrilla present in 1975 and 1976 possibly had a depressing effect upon bluegill and redear > 100 mm and on largemouth bass < 350 mm. Krummrich (1976) concluded, in a literature review of the relationship of submerged vegetation to fish population dynamics, that it is totally unwise to attempt management of fish populations around overabundant submerged vegetation. Dense beds of Hydrilla in several Florida lakes rarely contained numerous piscivorous fish; however, groups of largemouth bass were frequently observed at the periphery of the dense vegetation (Barnett and Schneider 1974). They concluded that dense stands of aquatic vegetation were the principle habitat for large populations of forage and juvenile game fish. Bennett (1948), studying a small Illinois lake, found that annual production of fish decreased with an increase in percent cover of Potamageton foliosus. Growth rates of bluegill also decreased in Bennett;s study; however, dieoff of the pond weed was not followed by increased bluegill growth. The average coefficients of condition and the growth rates of largemouth bass did reveal a marked increase after plant dieoff, which he attributed to prey being more vulnerable to bass. Ricker (1942) reported no association evident between growth rate of bluegills and abundance of aquatic vegetation in a series of Illinois ponds. Cope et al. (1969) found that redear sunfish gained weight faster after vegetation was controlled with dichlobenil. In another study, Cope et al. (1970) reported that bluegills grew faster in ponds where vegetation was controlled with 2, 4-D;

more food was made available because of vegetation removal and thinning of the fish population in high treatment ponds.

Both Childers (1967) and Trautman (1957) stated that redear require both relatively clean water and aquatic vegetation for their introductions to be successful. Data from Lake Wales indicated that excessive quantities of dense <u>Hydrilla</u> can result in both reduced condition and therefore reduced growth of redears.

SUMMARY AND CONCLUSIONS

During the third study year (spring 1977) hydrilla was eliminated at site 6 and a 90% reduction has occurred at site 4. Surface mats have not occurred during the third year, whereas, they were numerous during the initial two years. The reduction in hydrilla coincides with the second stocking of grass carp.

The reduction in hydrilla has affected both oxygen profiles and the depth to the light compensation point. Oxygen values at the surface decreased during 1977, but increased near the bottom, indicating better mixing during the third year. The light compensation point was found to be directly related to the amount of hydrilla. As hydrilla decreased the compensation point was found nearer the bottom. This indicates that although lake water levels have increased, enough light is available near the bottom, at all depths, for hydrilla growth. Recent recorded tracings (April 1978) indicate that hydrilla is growing at the deep water sites.

Three water quality parameters, sulfate, pH, and potassium, changed significantly through time. Sulfate and pH levels both decreases potassium levels increased 300-400%. Both Avault et al. (1968) and Lembi et al. (1978), found similar increases in potassium levels. Lembi et al. stated that increased potassium levels were an indication of grass carp feeding. Our results substantiate their conclusions. Lowered pH values during 1977, also indicate vegetation reduction. Rottmann (1976), Buck et al. (1975) and Beach et al. (1976) reported similar findings in their studies after vegetation control by grass carp. Other water quality parameters showed cyclic changes within each year, but yearly differences were not significant.

Major zooplankton groups were analyzed statistically and the abundance of calanoid copepods and the cladoceran, Diaphanosoma brachyurum, changed significantly with both groups decreasing through time. Other zooplankton exhibited significant seasonal differences during each study year. A shift in species dominance was noted during the study. During the winter of 1975, Ceriodaphnia pulchella was dominant with Diaphanosoma brachyurum subdominant. For the remainder of 1975 D. brachyurum was dominant. During 1976 and 1977 dominance shifted from larger cladocerans to the smaller Bosmina coregoni and C. pulchella. This shift may be due to habitat change since D. brachyurum is associated with marshes and weedy margins of lakes, whereas, B. coregoni is a limnetic species (Edmondson 1959). The decline of the larger species might also have been due to increased predation. Brooks (1968) noted that shifts from large planktors to smaller ones occurred with increased predation. Brooks (1968) noted that shifts from large planktors to smaller ones occurred with increased predation. Less hydrilla might have allowed small fish to become more efficient predators.

The abundance of benthic organisms was related to water depth and vegetation type. As an example, <u>Chaoborus</u> larvae were more abundant at the deeper sites. Gastropods and the trichopterans <u>Polycentropus</u> and <u>Leptocella</u>, were more abundant at Vallisneria sites. Two other trichopterans, <u>Oxyethira</u> and <u>Orthotrichia</u>, did not reveal site preferences. Generally chironomid larvae densities remained unchanged during the study, whereas <u>Chaoborus</u> larvae and oligochaetes increased.

Analysis of quarterly electrofishing data was confined to largemouth bass, bluegill and redear sunfish. The data, analyzed according to size class, indicated that sex influenced condition factor only during a few seasons. For chronological weight-length analysis, sexes were pooled within seasons and size classes. Bluegill and redear coefficients of conditions were significantly greater for the two larger size groups during 1977. Significant chronological increases in elevation of the weight-length regressions occurred in all size classes of largemouth bass; however, the majority of the significance was confined to bass < 350 mm. Although differences in slopes for the regression lines were not significant among years, significant elevation differences occurred during all seasons in 1977. Coefficient of condition and weight-length regression analysis indicated the dense stands of hydrilla present in 1975 and 1976 had a depressing effect upon bluegill and redear > 100 mm and on large mouth bass < 350 mm.

ACKNOWLEDGEMENTS

The authors thank Mr. Robert Gasaway, Mr. Randy Montegout, Mr. Robert B. Juul, Mr. Douglas DuRant and Mr. Robert Stetler for their assistance with field and laboratory work. This project was funded by the Florida Department of Natural Resources.

LITERATURE CITED

- Alikunhi, K. H. and K. K. Sudumaran. 1964. Preliminary observations on Chinese carps in India. Proc. Indian Aca. Sci. Section B. 60: 71-189.
- Avault, J. W., Jr., R. W. Smitherman and E. W. Shell. 1968. Evaluation of eight species of fish for aquatic weed control. FAO (F.A.O.U.N.) Rep. 44VIIE-3:109-122.
- Barnett, B. S. and R. W. Schneider. 1974. Fish populations in dense submerged plant communities. Hyacinth Control J. 12:12-14.
- Beach, M. L., W. W. Miley, II, J. M. Van Dyke and D. M. Riley. 1976. The effects of the Chinese Grass Carp {<u>Ctenopharyngodon idella</u> (Val.) } on the ecology of four Florida lakes, and its use for aquatic weed control. Final Rpt. Fla. Dept. of Nat. Res.
- Brooks, J. J. 1968. The effects of prey size selection by lake planktivores. Systematic Zoology. 17:273-291.
- Buck, H. D., R. J. Baur and C. R. Rose. 1975. Comparison of the effects of grass carp and the herbicide Diuron in densely vegetated pools containing golden shiners and bluegills. Prog. Fish-Cult. 37:185-190.
- Carlander, K. D. 1977. Handbook of Freshwater Fishery Biology, Volume II. The Iowa State Univ. Press. Ames, Iowa.
- Childers, W. F. 1967. Hybridization of four species of sunfishes (Centrarchidae). Ill. Nat. Hist. Surv. Bull. 29:159-214.
- Clugston, J. P. 1964. Growth of the Florida largemouth bass, <u>Micropterus</u> <u>salmoides floridanus</u> (Le Sueur), and the northern largemouth bass, <u>M. S. salmoides</u> (Lacepede), in subtropical Florida. Trans. Amer. Fish. Soc. 93:146-154.
- Cooper, E. L, E. Hidu and J. K. Anderson. 1963. Growth and production of largemouth bass in a small pond. Trans. Amer. Fish. Soc. 92: 391-400.

Cope, O. B., J. P. McCraren and L. L. Eller. 1969. Effects of dichlobenil in two fishpond environments. Weed Science. 17:158-165.

, E. M. Wood and G. H. Wallen. 1970. Some chronic effects of 2, 4-D on the bluegill (Lepomis macrochirus). Trans. Amer. Fish Soc. 99:1-12.

Edmondson, W. T. ed. 1969. H. B. Ward and G. C. Whipple. Fresh-water biology, 2nd. ed. John Wiley and Sons, New York.

- Hart, W. B., P. Doudoroff and J. Greenback. 1945. Evaluation of toxicity of industrial wastes, chemicals and other substances to fresh-water fishes. Water Contr. Lab. Atlantic REfininc Co., Philadelphia, Penn.
- Krummrich, J. T. 1976. Partial annotated bibliography on relationship of submerged vegetation to fish population dynamics. Fla. Game and Fresh Water Fish Comm. Mimo. Rept. 13 pp.
- Lembi, C. A., B. G. Ritenour, E. M. Iverson and E. E. Forss. 1978. The effects of vegetation removal by grass carp on water chemistry and phytoplankton in Indiana ponds. Trans. Amer. Fish. Soc. 107:161-171.
- Nordlie, F. G. 1976. Plankton communities of three central Florida lakes. Hydrobiologia. 48:65-78.
- Pennak, R. W. 1953. Fresh-water invertebrates of the United States. The Ronald Press Co., N. Y.
- _____ 1957. Species composition of limnetic zooplankton communities. Limnol. Oceanog. 2:222-232.
- Prouse, G. A. 1969. The role of cultured pond fish in the control of eutrophication in lakes and dams. Verh. Internat. Verein. Limnol. 17:714-718.
- Reid, G. K. 1961. Ecology of inland waters and estuaries. Reinhold Pub. Corp., N. Y. 375 pp.
- Ricker, W. E. 1942. The rate of growth of bluegill sunfish in lakes of northern Indiana. Invest. Ind. Lakes, Streams. 2:261-214.
- Rottmann, R. W. 1976. Limnological and ecological effects of grass carp in ponds. M. S. Thesis., Univ. of Missouri, Columbia, Mo.
- and R. O. Anderson. In Press. Limnological and ecological effects of grass carp in ponds. Proc. S. E. Assoc. Game and Fish Comm. 30.
- Shireman, J. V. 1976. Ecological study of Lake Wales, Florida after introduction of grass carp (<u>Ctenopharyngodon idella</u>). Annual Rpt. Fla. Dept. Nat. Res.
- Stewart, H. G., Jr. 1966. Ground-water resources of Polk County. Fla. Geol. Sur. Rept. No. 44:170 pp.
- Terrell, T. T. 1975. The impact of macrophyto control by the white amur (<u>Ctenopharyngodon idella</u>). Verh. Int. Ver. Limnol. 19:2510-2514.
- Trama, F. B. 1954. The pH tolerance of the common bluegill (Lepomis macrochirus, Raf.). Phila. Aca. Nat. Sci., Not. Nat. 256 pp.

Trautman, M. B. 1957. The fishes of Ohio. Ohio State Univ. Press. 683 pp.

Van Der Avyle, M. J. and K. D. carlander. 1977. Seasonal variation in weight-length relationships of largemouth bass in a 3-hectare reservoir. Proc. Iowa Acad. Sci. 84:35-38.

_

Weber, W. J. and W. Stumm. 1963. Mechanism of hydrogen ion buffering in natural waters. J. Am. Wat. Wks. Asso. 55:1553-1578.

Table 1. Relationships between zooplankton and environmental factors significant at $P \le 0.05$ in stepwise multiple regression (+= positive relationship; -= negative relationship).

	Cyclopoid copepods	Calanoi d copepods	Nauplii	Diaphanosoma	Alona	Camptocercus	Bosmina	Daphnia	Ceriodaphnia	Macrothrix	Chydorus
рН		÷		+	+		-	-	· · · · · · · · ·		+
Chlorophyll a		x	+		+		+	+	-		+
Sulfate	÷		+	+ ·		+					
Turbidity	+		-			÷	-				
Calcium		-			-		-	-			-
Magnesium		+									
Sodium						-			+		. +
Potassium			+								+
Chloride							+	+			
Iron			+								
Nitrate Nitrogen				-			÷	+ .			
Ammonia Nitrogen	-		+				.	-			+
Organic Nitrogen		-									
0 t thophosphate			-				-	-	-		
Total Phosphate		+	+			-	+		+		
Temperature						-					
Depth			-			+	+	+	+		
Water Level							+	+	-		
r ²	.05	.21	.23	.18	.16	.16	.47	.32	.18	.10	.15

Taxon	Percent Frequency of Od	CUTTORCA
	Tercent frequency of o	
Coelenterata		
Hydra	2	
Turbellaria		
Planaria	7	
Nematoda	4	
Annelida		
Naididae	13	
Other Oligochaeta	40	
Hirudinea	7	
Amphinodo		
Amphipoda Huglolla arteen	21	
Hyalella azteca	21	
Decapoda		
Palaemonetes	2	
Hydracarina	8	
Ephemeroptera larvae		
<u>Callibaetis</u>	2	
Caénis	2	
. Centroptilum	<u>≤</u> 1	
Odonata larvae	2	
Anisoptera	2	
Perithemis		•
<u>Aphylla</u>		
Panatala		
Zygoptera	2	
Enallagma	۷	
Irichoptera larvae		
Oxyethira		
Orthotrichia	{9	
Polycentropus	· · · · · · · · · · · · · · · · · · ·	
Leptocella	{11	
Lepidoptera larvae		
Eoparagyractis	3	
^o leoptora	^	
Coleoptera <u>Bidessus</u> larvae	2	

Table 2. Percent frequency of occurrence of benthic invertebrates collected from Lake Wales.

Table 2. Cont.

axon	Percent Frequency of Occurrence
iptera	
Culicidae	
Chaoborus larvae	70
Chaoborus pupae	8
Tendipedidae larvae	78
Pseudochironomus	
Dicrotendipes lobus	
Chironomus	
Tendipedidae pupae	9
Ceratopogonidae	≤1
astropoda	
Gyraulus parva	16
Helisoma duryi	12
Physa	2
elecypoda	
Sphaeridae	<u><</u> 1
Sphaeridae	<u><</u> 1

Table 3. Relationships between the dominant benchic organisms and environmental factors significant at the $P \le 0.05$ in stepwise multiple regression (+ = positive relationship; - = negative relationship).

	lae			lia	Supus		
	Chironomidaè larvae	Chaoborus 1arvae	Hyalella azteca	Oxyethirus Urthotrich larvae	Polycentropus Leptocella	Oligochaeta	Gyraulus
рН				_			
Chlorophyll a							+
Turbidity				-			
Calcium			-			+	
Sodium		-				-	
Potassium				-			
Chloride				+			
Organic Nitrogen				-	· ••••		
Total Phosphates					* .	-	
Bottom Oxygen							+
Bottom Temperature		+			+	+	
Bottom Light					+		
Station Depth	-	+	-	+	· _	_	
Water Level	-	+					
r ²	. 32	.26	.05	.20	. 31	.36	.08

Table 4. Comparison of mean coefficients of condition, K(TL), between sexes within season and size classes for bluegill in Lake Wales. (Coefficients followed by same letter did not have significant elevation (intercept) differences at $P \le 0.05$).

	Size Class						
	50 - 1	00	100 - 1	L50	> 1.	50	
Season	Male	Female	Male	Female	Male	Female	
Spring 1975			1.44 ^a	1.46 ^a	1.49	1.44*	
Spring 1976	1.39*		1.61*	1.49*	1.63 ^a	1.62 ^a	
Summer 1975	1.43*		1.44 ^a	1.49 ^b	1.48	1.47*	
Summer 1976			1.57*	1.61*	1.65 ^a	1.47* 1.52 ^b	
Fall 1975	1.63*		1.53*	1.46	1.48	1.42*	
Vinter 1975			1.50*	1.45*	1.50 ^a	1.41 ^b	

Table 5. Comparison of mean coefficients of condition, K(TL), by season, size class, and sex for redear in Lake Wales. (Coefficients followed by same letter did not have significant elevation (intercept) differences at $P \le 0.05$.)

50 – 1	100	<u>Size C1</u> 100 -			_
	100	100	1.50		
		100 -	150	> 1.	50
Male	Female	Male	Female	Male	Female
1 594	1 504	1 / ca	1 //a	1 40*	1.41
1. 52*	1.52"				1.55*
		1.03-	1.02-	1.00	1.00*
	1.54*	1.44 ^a	1.46 ^a	1.66*	1.51*
		1.52^{a}	1.43^{b}	1.59	1.52*
	1.53*	1.43 ^a	1.47 ^a	1.42*	1.55*
		1.41 ^a	1.39 ^a	1.42*	1.38*
	1.52*	1.52* 1.52* 1.54* 1.70* 1.53*	1.65^{a} $1.54* 1.44^{a}$ $1.70* 1.52^{a}$ $1.53* 1.43^{a}$	$\begin{array}{cccc} 1.65^{a} & 1.62^{a} \\ 1.54^{*} & 1.44^{a} & 1.46^{a} \\ 1.70^{*} & 1.52^{a} & 1.43^{b} \end{array}$	1.65^{a} 1.62^{a} 1.55 $1.54*$ 1.44^{a} 1.46^{a} $1.66*$ $1.70*$ 1.52^{a} 1.43^{b} 1.59 $1.53*$ 1.43^{a} 1.47^{a} $1.42*$

Table \oint . Comparison of mean coefficients of condition, K(TL), by season, size class, and sex for largemouth bass in Lake Wales. Coefficients followed by same letter did not have significant elevation (intercept) differences at P \leq 0.05.

	Size Class							
	75 -	150	151 -	- 250	250 -	- 350	> 3	50
Season	Male	Female	Male	Female	Male	Female	Male	Female
Spring 1975	0.92 ^a	0.92 ^a	0.87 ^a	0.94 ^a	1.03*	1.05*	1.26 ^a	1.34 ^a
Spring 1976	0.99 ^a	0.96 ^a	1.01 ^a	0.99 ^a	1.08*	1.04*	1.30*	1.35
Summer 1975	0.89 ^a	0.92 ^a	0.92 ^a	0.92 ^a	1.04*	1.11*	1.03*	1.33*
Summer 1976	1.15*		1.061	1.031	1.25*	1.14*	1.72*	
Fall 1975	0.94 ^a	0.99 ^a	0.95 ^a	0.96 ^a			1.44*	1.33*
Winter 1975	0.90 ^a	0.93 ^a	0.94 ^a	0.91 ^b	1.12*		1.35*	1.32*

¹Significant slope differences between sexes due to larger length of males.

Table 7. Mean coefficients of condition, K(TL), by season and size class for bluegill in Lake Wales. (Coefficients followed by same letter did not have significant elevation (intercept) differences at $P \leq 0.05$ within equivalent seasons and size classes.)

	Size Class					
Season	50 - 100	100 - 150	> 1.50			
Spring 1975 Spring 1976 Spring 1977	1.58 ^a 1.49 ^a	1.44 ^a 1.61 ^b 1.67 ^c	1.48 ^a 1.62 ^b 1.81 ^c			
Summer 1975 Summer 1976 Summer 1977	1.49* 1.51	1.47 ^a 1.59* 1.60 ^b	1.47 ^a 1.58 ^b 1.73 ^c			
Fall 1975		1.47	1.47			
Winter 1975 Winter 1977	1.38	1.47 ^a 1.58 ^b	1.47 ¹ 1.70 ¹			

¹Significant slope differences between seasons.

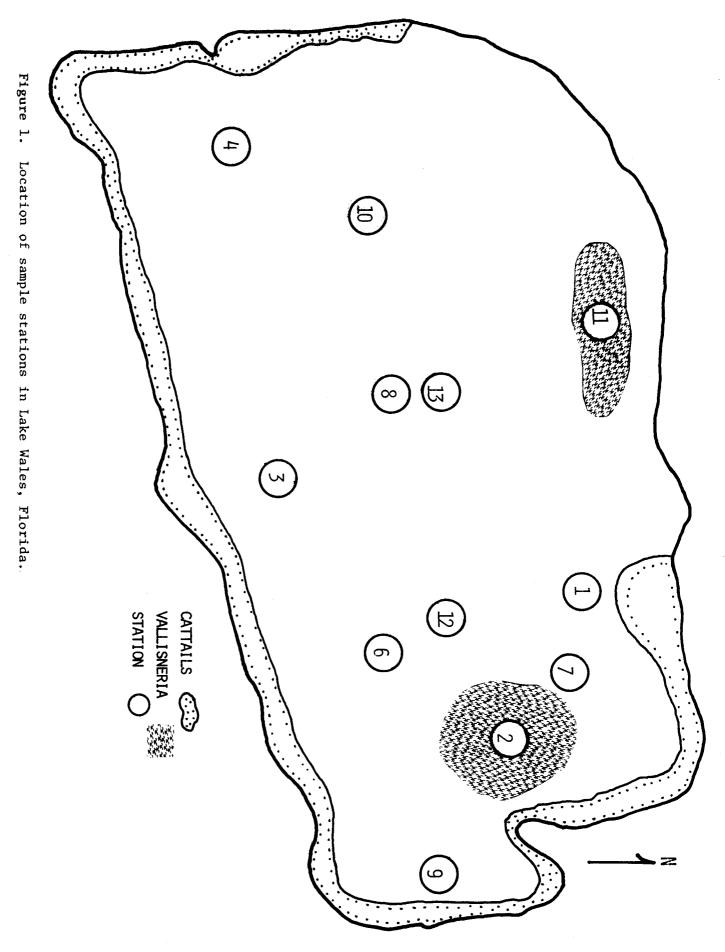
Table 8. Mean coefficients of condition, K(TL), by season and size class for redear in Lake Wales. (Coefficients followed by same letter did not have significant elevation (intercept) differences at $P \leq 0.05$ within equivalent seasons and size classes.)

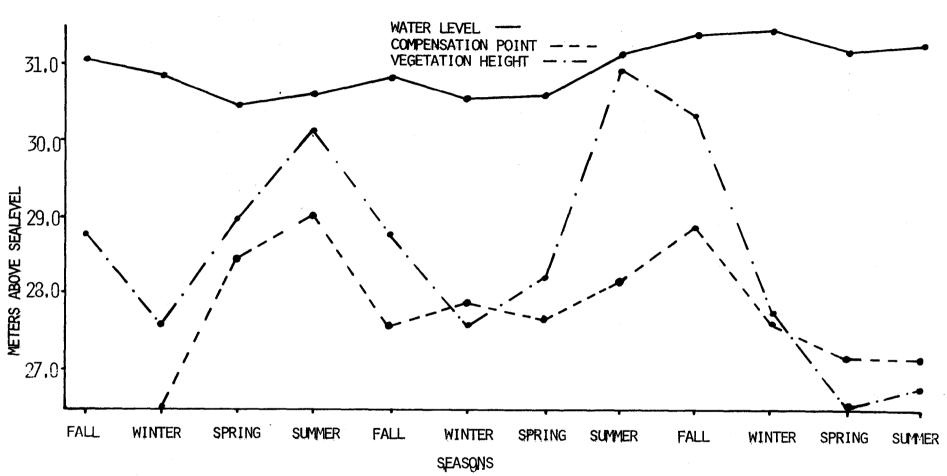
	Size Class				
Season	50 - 100	100 - 150	> 150		
Spring 1975	1.52 ^a	1.44 ^a	1.41 ^a		
Spring 1976	1.64 ^{a1}	1.62 ^b	1.54 ^b		
Spring 1977	1.63 ^a 1	1.63 ^b	1.41 ^a 1.54 ^b 1.81 ^c		
Summer 1975		1.44 ^a	1.60 ^a		
Summer 1976	1.62 ^a	1.470	1.60 ^a		
Summer 1977	1.62 ^a	1.60 ^c	1.60 ^a 1.72 ^a		
Fall 1975	1.52*	1.46	1.49		
Winter 1975	1.46*	1.40	1.43 ^a		
Winter 1977	1.47	1.50*	1.55 ^b		

¹Significant slope differences within seasons.

Table 9. Mean coefficients of condition, K(TL), by season and size class for largemouth bass in Lake Wales. (Coefficients followed by same letter did not have significant elevation (intercept) differences at $P \le 0.05$ within equivalent seasons and size classes.)

	Size Class					
Season	75 - 150	150 - 250	250 - 350	> 350		
Spring 1975	0.92 ^a	0.91 ^a	1.03 ^a	1.31 ^a		
Spring 1976	1.01 ^b	1.00 ^b	1.08 ^a	1.35,al		
Spring 1977		1.12 ^c	1.19 ^b	1.41 ^b		
Summer 1975	0.91 ^a	0.92 ^a	1.06*	1.20 ^a		
Summer 1976		1.03*	1.16*	1.31*		
Summer 1977	1.14 ^b	1.06 ^b	1.14	1.32 ^a		
Fall 1975	0.99	0.95		1.42*		
Winter 1975	0.91	0.92 ^a	1.12*	1.32 ^a		
Ninter 1975	1.14*	1.03 ^b	1.38	1.38 ^a		





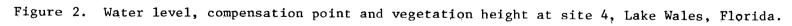
I

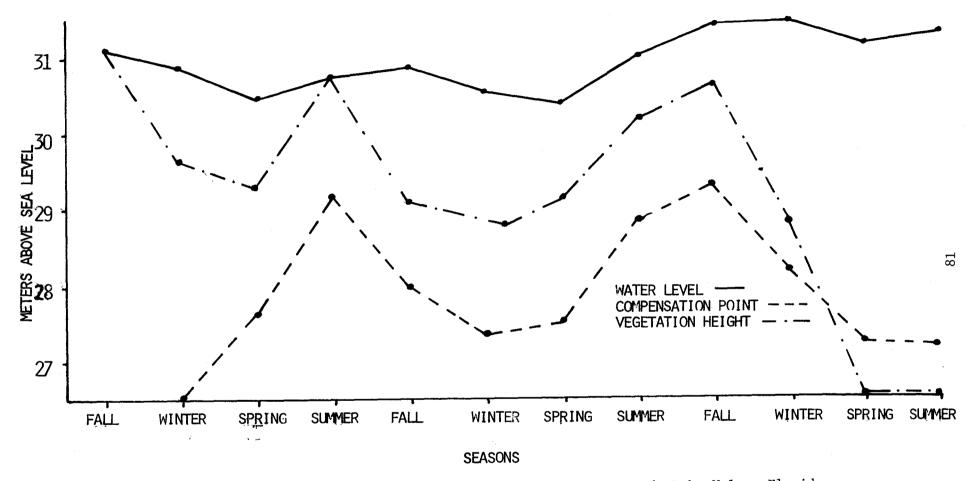
ŀ

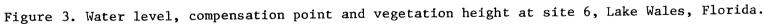
)

ł

I







L

I

1

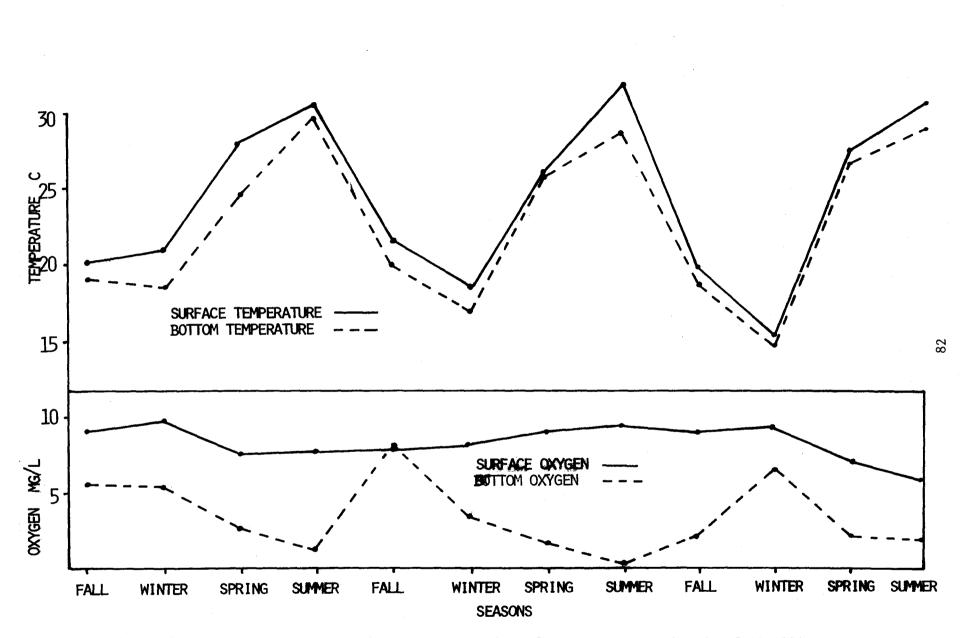
1

I

1

1

ł



Ì

١

1

1

1

1

I

ł

I

Ι

ł

1

ļ

1

Figure 4. Surface and bottom oxygen and temperature values for Lake Wales, Florida, fall 1974 through 1977.

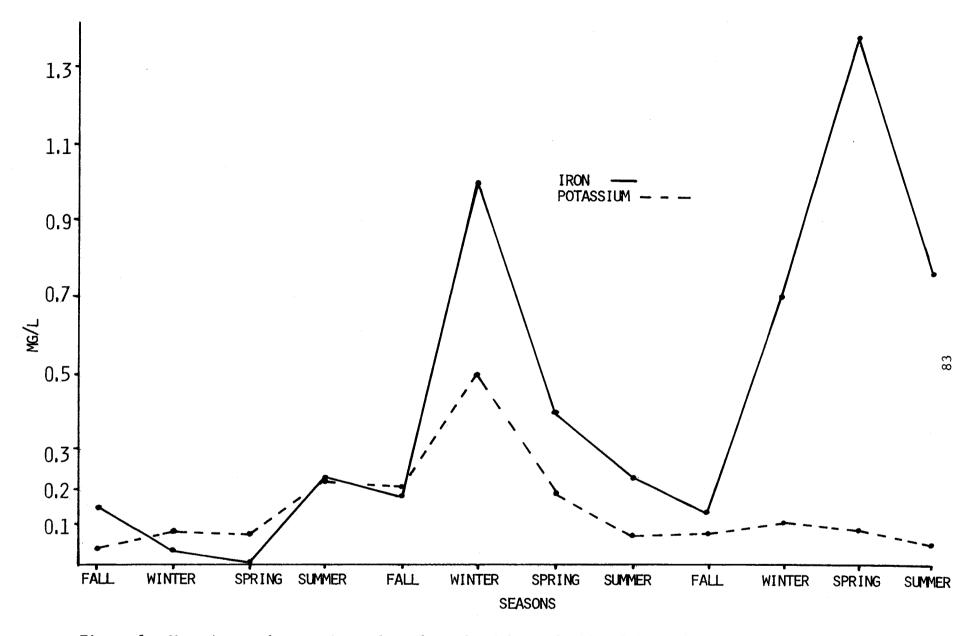


Figure 5. Mean iron and potassium values for Lake Wales, Florida, fall 1974 through summer 1977.

ł

1

ļ

J

1

ł

1

1

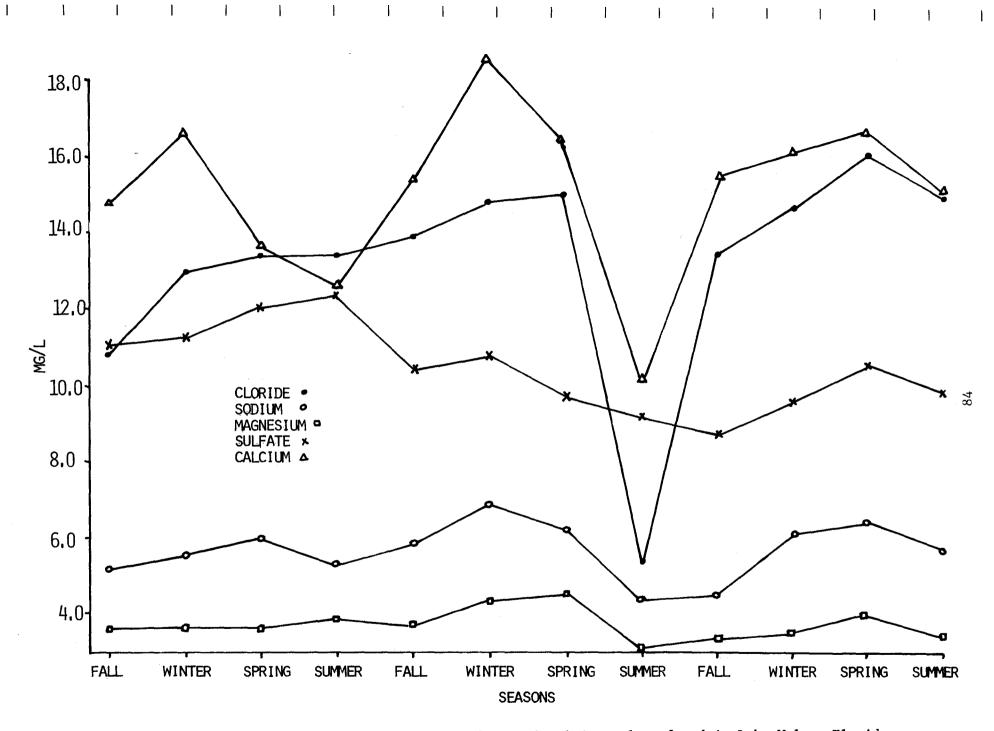
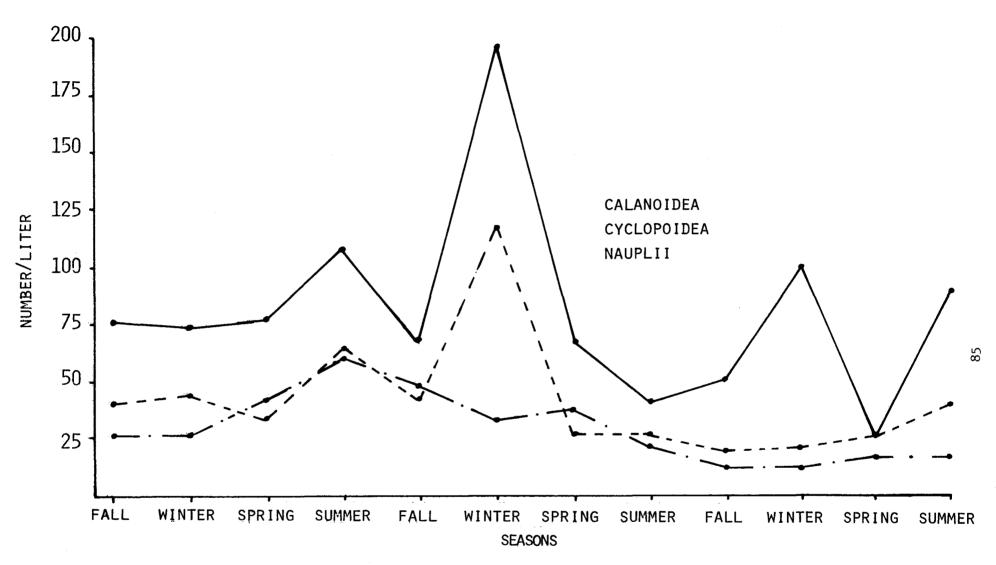


Figure 6. Mean chlorida, sodium, magnesium, sulfate and calcium values found in Lake Wales, Florida, 1974 - 1977.



ļ

I

I

ł

1

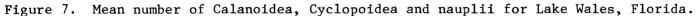
1

ŧ

ł

1

ł



1

ł

1

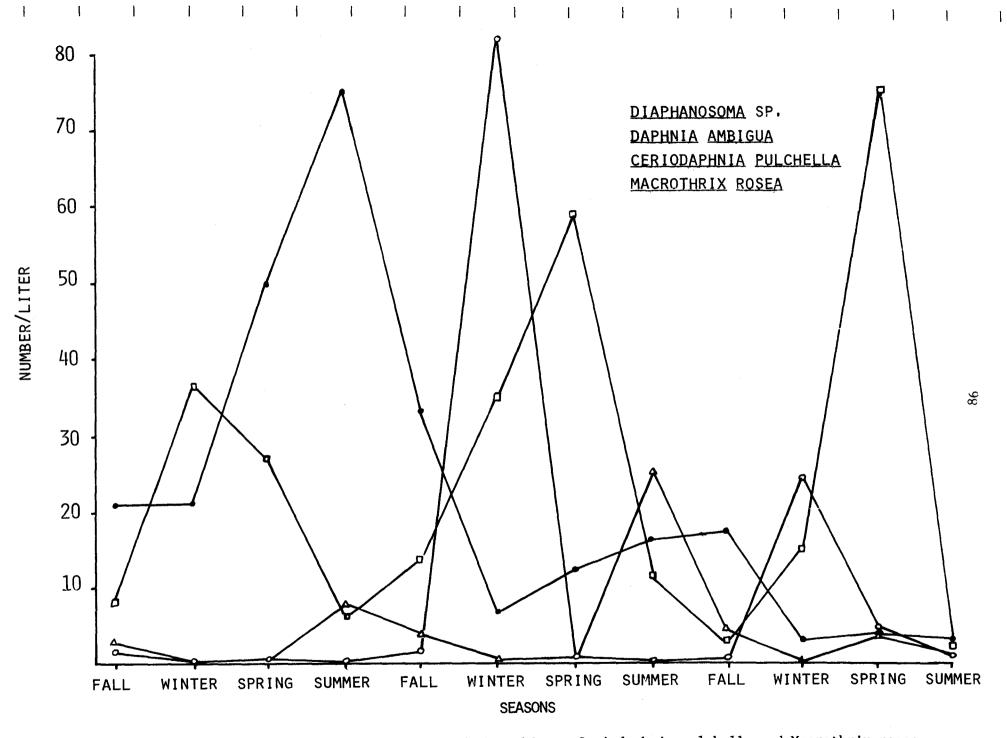
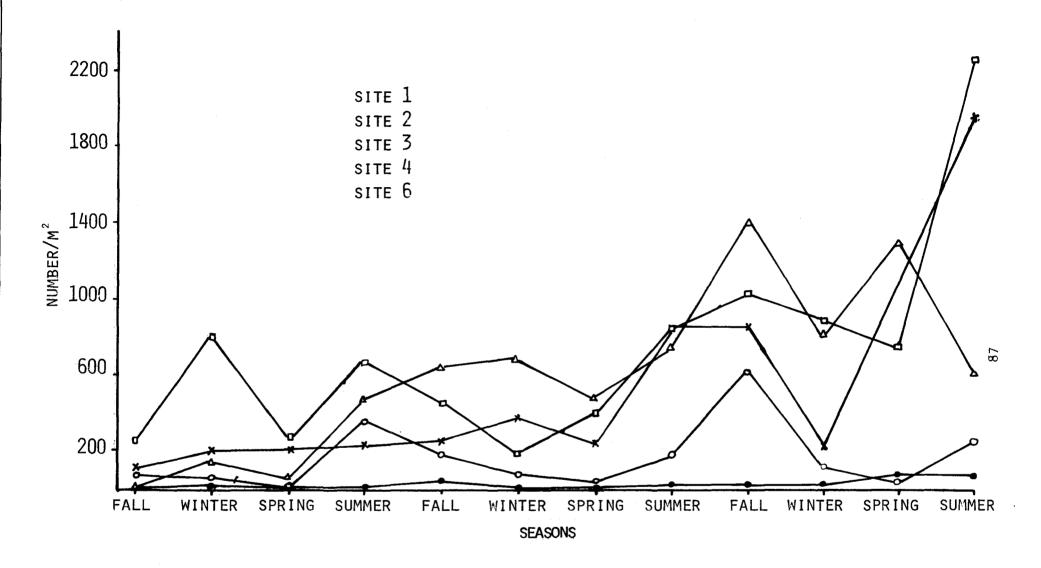
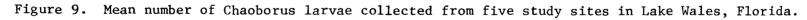


Figure 8. Mean number of <u>Diaphanosoma</u> sp., <u>Daphnia</u> <u>ambigua</u>, <u>Ceriodaphnia</u> <u>pulchella</u> and <u>Macrothrix</u> <u>rosea</u> for Lake Wales, Florida.





}

}

}

}

)

}

}

}

}

1 } }

)

}

}

1

I

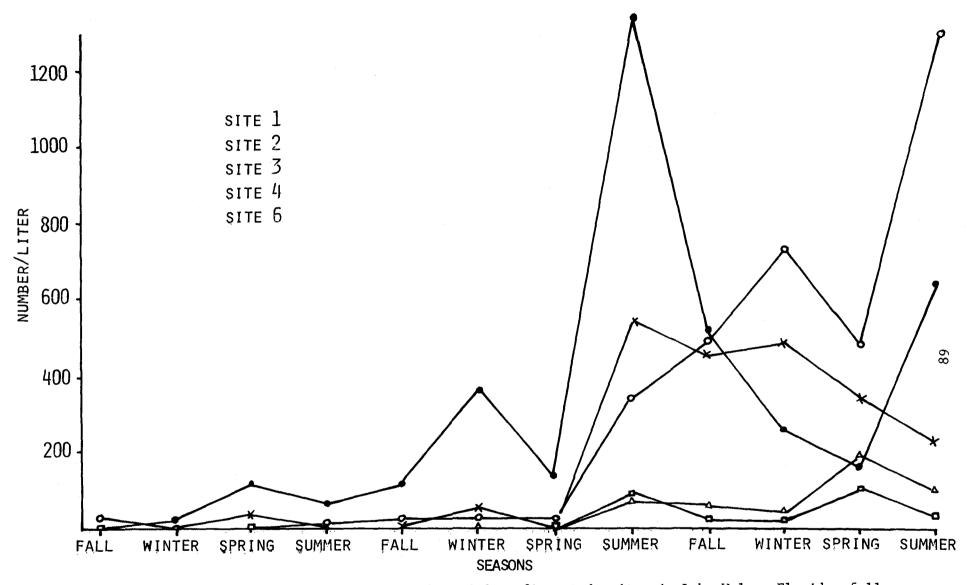


Figure 11. Mean number of oligochaetes collected from five study sites in Lake Wales, Florida, fall 1974 through summer 1977.

l

۱

ł

ł

l

ł

1

ł

ł

ł

l

l

ł

1

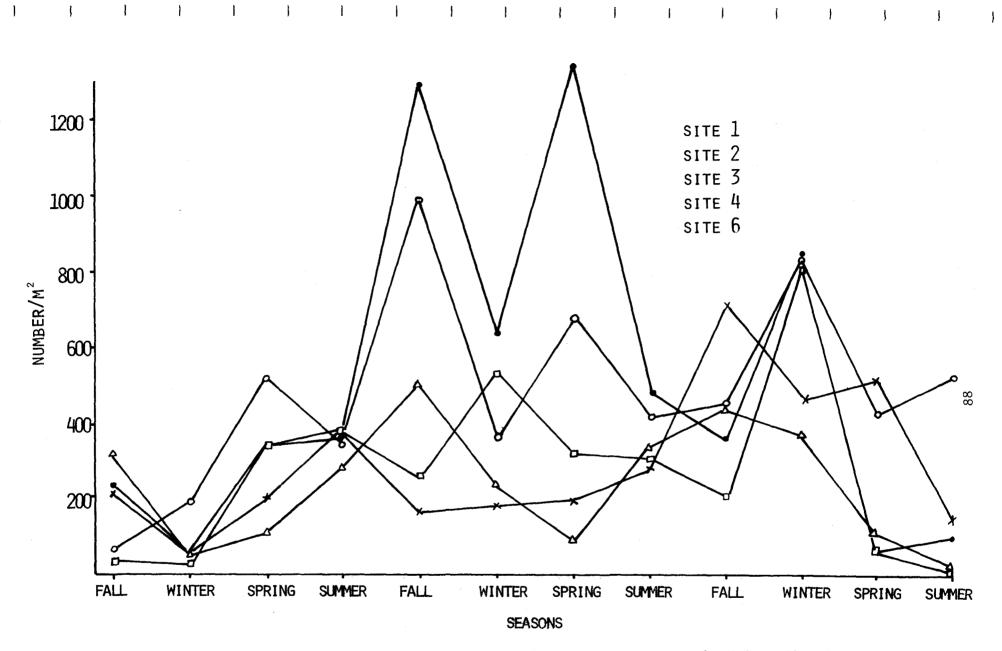


Figure 10. Mean number of chironomids collected from five study sites in Lake Wales, Florida.

USE OF THE GRASS CARP FOR CONTROL OF HYDRILLA IN SMALL PONDS $\frac{1}{2}$

David L. Sutton, V.V. Vandiver, Jr.,

R.S. Hestand, and W.W. Miley, II

Associate and Assistant Professors, University of Florida Aquatic Botanist, Florida Game and Fresh Water Fish Commission Biologist III of the Florida Department of Natural Resources

INTRODUCTION

Studies on biological control of aquatic weeds have increased in recent years. While other control methods provide only temporary relief, biological control offers the potential of exerting a constant pressure on the target species. Many different organisms have been and are being studied for their potential to control aquatic weeds. The grass carp (<u>Ctenopharyngodon idella</u> Val.) appears to be a promising means for control of submersed weeds (Cross 1969; Hickling 1965; Sutton 1977; and Swingle 1957).

Hydrilla (Hydrilla verticillata Royle) was introduced into Florida in the late 1950's and has spread rapidly and has become the major submersed weed problem in Florida. The grass carp is one of the most promising organisms being investigated for its potential to control hydrilla.

CONTROL OF HYDRILLA IN ORANGE COUNTY PONDS

Following the stocking of grass carp in small ponds in central Florida for hydrilla control, it was found that high stocking rates of grass carp would be necessary to control dense infestations of hydrilla (Sutton 1974). In this study, one-quarter of each of four ponds 0.08 ha in size were planted with hydrilla, vallisneria (Vallisneria americana Michx.), chara (Chara sp.), and southern naiad (Najas guadalupensis Spreng. Magnus).

Within two years, hydrilla became the dominant plant in the control pond without fish, while the ponds stocked with 137 to 360 grass carp per ha contained no hydrilla at the end of the test period. Vallisneria remained in the ponds in the presence of up to 900 kg of grass carp per ha. Even though grass carp will eat vallisneria, this plant is not a preferred food. This study suggested that through careful management of the grass carp it

L/ Cooperative study of the University of Florida Agricultural Research Center at Fort Lauderdale; the Florida Game and Fresh Water Fish Commission, Fisheries Research Laboratory at Eustis; the Florida Department of Natural Resources Bureau of Aquatic Weed Control at Tallahassee; and the United States Department of Agriculture, Science and Education Administration, Federal Research, Aquatic Plant Management Laboratory at Fort Lauderdale. Published as Journal Series Number 1096 of the Florida Agricultural Experiment Station.

might be possible to encourage growth of certain aquatic plants while using the grass carp to stress growth of the hydrilla.

SMALL POND STUDIES

1. FPL Pond. A 0.4-ha pond which had a history of excessive hydrilla growth was stocked on 30 September 1975 with 40 grass carp averaging 0.5 kg in weight (100 fish per ha) (Table 1). At the time of grass carp stocking, hydrilla was estimated to cover approximately 80% of the surface area.

On 25 March 1976, 39 grass carp averaging 1.67 kg were removed from the FPL pond using rotenone. Hydrilla growth was no longer visible at the surface of the pond. Grass carp had grown at a rate of 0.19 kg per month or 6.6 g per day.

The FPL pond was restocked on 18 May 1976 with five fish of an average weight of 1.89 kg which is equivalent to 12 fish per ha or 24 kg per ha. By the end of the 1976 growing season, hydrilla had not regrown to the surface. This stocking rate kept the hydrilla growth in check.

A survey of the FPL pond was begun on 15 December 1976 to monitor the presence and relative abundance of vegetation. The survey consisted of stretching two lines across the pond and sampling with a long-handled probe every 1.5 m along each line. The average of the two lines recorded as the percent frequency of occurrence of vegetation along the transect lines are presented in Table 2.

Hydrilla accounted for 13% of the vegetation occurring along the transect line in December 1976, but only 2% for the same month a year later. The most abundant plants in the pond were chara, southern naiad, the common bladderwort (<u>Ultricularia macrorhiza</u> B.). These plants remained relatively stable although bladderwort did increase 10% from December 1976 to December 1977.

A vegetation survey was not conducted prior to grass carp stocking in this pond; therefore, the frequency of occurrence of vegetation cannot be compared precisely to that present in the pond before the grass carp were stocked. Based on the previous history of the pond where hydrilla was seen at the surface of the pond for several years, the stocking of grass carp had reduced the growth of hydrilla to a low level. Hydrilla did not grow to the surface during the 1976 or 1977 growing season. The stocking of 12 grass carp (24 kg of fish) per ha was sufficient to prevent reestablishment of hydrilla for two growing seasons.

2. BCC Pond. A 0.6-ha pond on the Broward Community College campus had a history of hydrilla problems. This pond was treated with herbicides in the spring of 1975, but by June of that year the pond was heavily infested with hydrilla around the edges. On 27 June 1975 the pond was stocked with 10 grass carp averaging 0.5 kg, on 10 September 1975 with 10 fish averaging 0.5 kg, and on 30 October 1975 with 10 fish averaging 0.8 kg each for a total of 30 grass carp. One-third of the pond was again treated with herbicides on 17 December 1975 to help control the hydrilla.

Hydrilla was not present at the surface of the pond in the spring of 1976. The pond was rotenoned on 27 May and 27 grass carp averaging 4.1 kg were removed. The pond was restocked in June 1976 with five grass carp averaging 2.0 kg each. Hydrilla did not grow to the surface for the remainder of the year, and a vegetation survey revealed that the hydrilla growth had been checked by the restocking of grass carp (Table 3). Hydrilla was a minor portion of the vegetation in the pond during 1976 and 1977.

3. Vo-Ag Pond. In order to follow closely the removal of hydrilla from a small pond by the grass carp and their subsequent effect on the vegetative propagules of this submersed plant, 19 grass carp averaging 1.08 kg were placed in this 0.25 ha pond on 15 October 1976. The pond contained slightly over 80% hydrilla as based on the transect lines (Figure 1) with hydrilla growing to the surface. The abundance of hydrilla was relatively stable through the January sampling date. By March 1977, the hydrilla was starting to disappear with a rather dramatic decrease from May to July.

All 19 grass carp were removed on 7 December 1977 using rotenone. At that time the fish weighed an average of 4.27 kg. The weight of the fish on a per ha equivalent was 82 kg when stocked and 329 kg when removed. During this 418-day period the fish grew at a rate of 0.23 kg per month or 7.6 g per day.

The number of vegetative propagules in this pond was estimated by collecting 25 core samples every 2 months (Figure 2). The core samples were obtained with a device which removed a portion of the hydrosoil composed of a surface area of 86 cm² by about 20 cm in depth. Cores were washed on a small mesh screen which allowed the propagules to be separated from the hydrosoil.

The number of turions and tubers was estimated at 29.8 and 12.6 million per ha, respectively, at the time the grass carp were stocked (Figure 2). After 1 year the number of turions and tubers was estimated at slightly under 400 thousand and 3.1 million per ha, respectively.

A comparison of the sizes of tubers for the November 1976 and 1977 sampling dates shows a reduction in number for all sizes for this time period (Figure 3). Core samples from the November 1976 sampling time contained tubers as small as 4 mm, not shown in the graph, while none smaller than 7 mm were found a year later. Also, no tubers 14 mm in size were found in the November 1977 core samples.

These data suggest that one of the initial effects of the grass carp is a reduction in the number of turions. Additional studies will be required to evaluate the long term effect on the tubers as well as on the turions.

4. Hillcrest Pond. The approved herbicides, diquat and copper sulfate, were applied to this 0.31-ha pond during January 1976 and again in February 1977 to control hydrilla infestation. These applications of herbicides controlled the hydrilla. On 7 April 1977 five grass carp averaging 3.0 kg were stocked in the pond. Hydrilla did not reappear after this stocking.

Core samples (15) collected 3 February 1977 were used to estimate the number of hydrilla vegetative propagules prior to stocking of grass carp for

regrowth control. The number of tubers and turions was estimated at 1.9 million and 150 thousand per ha, respectively. On 21 December 1977, another set of core samples (25) was collected which indicated the number of tubers was 2.1 million and the number of turions was 92 thousand per ha. These results are similar to that for the Vo-Ag Pond in that one of the short-term effects of the grass carp for regrowth control is more pronounced on reducing the number of turions than the number of tubers.

5. Hamlet Pond. In order to evaluate growth of grass carp on the basis of sex, 10 female and 10 male 3-year-old fish were placed in this 0.49 ha pond. These fish had been selected from a group of 3-year-old fish so that the initial weight of both sexes were the same at the time of stocking. The 20 fish averaged 1.6 kg when they were placed in Hamlet Pond on 29 April 1976. Hydrilla was abundant during most of the growth period, but the pond did not contain any of these plants growing up to the surface of the water when the fish were removed on 18 February 1977.

The female grass carp had increased to an average weight of 4.2 kg during this 295-day period, while the males averaged 2.3 kg. The average daily growth rate was 14.9 g and 7.8 g, respectively for female and male fish.

DISCUSSION

The previous history of the ponds served as the bench mark for evaluating the impact of grass carp feeding. Since dense infestations of hydrilla had been present and repeated applications of herbicides were necessary to keep the hydrilla under control, it is assumed that excessive growth of hydrilla would have occurred if the ponds had not been stocked with grass carp. One the hydrilla infestation was controlled with herbicides a stocking rate of 8 to 12 grass carp per ha was effective in keeping hydrilla from rapidly establishing itself, at least for two growing seasons.

Small grass carp are very likely to be preyed upon (Gasaway 1977); therefore, for long-term stocking, large fish 0.5 kg or larger in weight may be required. The growth rate of female grass carp was almost twice that of males. This is in agreement with other studies (Hickling 1967). However, we did not determine whether females consumed twice the amount of vegetation as males. Information of this type will be useful in determining optimal numbers for stocking uniform populations of fish.

Most of the food preference studies (Avault 1965; Gidumal 1958; and Pentlow and Stott 1965), have been conducted with small fish or in small containers. In the small ponds we used, the large grass carp were either seeking out the regrowth of hydrilla or at least consuming it sufficiently so that other plants were competitive with hydrilla.

Stocking with low numbers of grass carp allowed desirable native vegetation to grow. In the FPL and BCC Ponds, the plants that grew were ones that became established naturally. For example, some vallisneria, a desirable aquatic plant, was present in the BCC Pond prior to application of the herbicide and stocking with grass carp. Once the dense growth of hydrilla was under control, vallisneria could grow. Additional studies are needed to evaluate the possibility of using the grass carp to keep regrowth of hydrilla

at a low level while encouraging growth of desirable plants which either develop naturally or are transplanted to the body of water. In this way it may be possible to replace problem weeds with desirable aquatic plants.

Short-term effects of stocking low numbers of grass carp are: (1) preventing regrowth of hydrilla, and (2) reducing the number of turions. Since the tubers grow below the surface of the hydrosoil, as opposed to the turions which fall off the plant and lay on top of the hydrosoil, the tubers may remain viable for a long period and serve as a source reservoir of propagules to reinfest the body of water. The long-term effect of low stocking rates of grass carp for regrowth control will require additional study.

ACKNOWLEDGMENTS

Supported in part by the Center for Environmental Programs, University of Florida, Gainesville. A number of individuals assisted with the collection of data and other work related to this study. The Authors wish to thank the following individuals for their assistance: Diane Johnston, Jay Gaus, Terry Lott, Chris Carter, Lowell Trent, Andrew Leslie, Jo Peacock, Dennis Riley and Jess Van Dyke.

REFERENCES

- Avault, J.W. 1965. Preliminary studies with grass carp for aquatic weed control. Prog. Fish-Cult. 27:207-209.
- Cross, D.G. 1969. Aquatic weed control using grass carp. J. Fish. Biol. 1:27-30.
- Gasaway, R.D. 1977. Predation on introduced grass carp (<u>Ctenopharyngodon</u> idella) in a Florida lake. Fla. Sci. 40:167-173.
- Gidumal, J.L. 1958. A survey of the biology of the grass carp (<u>Ctenopharyn-godon idella</u> (C & V). Hong Kong Univ. Fish. J. 2:1-6.
- Hickling, C.F. 1965. Biological control of aquatic vegetation. Pest Articles and News Summary. 11:237-244.
 - 1967. IV. On the biology of a herbivorous fish, the white amur or grass carp, <u>Ctenopharyngodon</u> idella Val. Proc. R. Soc. Edinb. 70:62-81.
- Pentlow, F.T.K. and B. Stott. 1965. Grass carp for weed control. Prog. Fish-Cult. 27:210.
- Sutton, D.L. 1974. Final report to the Florida Department of Natural Resources on the project: Control of aquatic plant growth in earthen ponds by the white amur. Fort Lauderdale ARC Res. Rep.-FL74-2. 75 pp. mimeographed.

1977. Grass carp (<u>Ctenopharyngodon</u> idella Val.) in North America. Aquatic Bot. 3:157-164.

Swingle, H.S. 1957. Control of pondweeds by use of herbivorous fishes. Proc. South. Weed Conf. 10:11-17.

	Date	Fish per ha	Weight (kg) per ha
Α.	Hydrilla control		
	30 September 1975	100	51
	25 March 1976	98	163
в.	Regrowth control		
	18 May 1976	12	24

Table 1. Schedule for grass carp stocked and removed from the FPL Pond for hydrilla control and for controlling the regrowth of hydrilla and other species. Table 2. Frequency of occurrence (%) of aquatic vegetation in the FPL Pond.

Aquatic vegetation	15-Dec-76	22-Apr-77	15-Jun-77	21-Sep-77	15-Dec-77
<u>Hydrilla verticillata</u> Royle	13.0	2.2	5.4	1.2	2.0
<u>Chara</u> sp.	31.5	42.2	32.4	35.2	36.1
Najas quadalupensis (Spreng.) Magnus	30.1	26.7	35.7	35.2	27.8
<u>Utricularia macrorhiza</u> B.	16.4	13.3	22.2	26.1	26.4
Eleocharis sp.	2.7	2.2	1.1	1.1	2.1
Bacopa sp.		2.2		_	2.1
<u>Nitella</u> sp.	- <u></u>		1.1		
Panicum repens L.		2.2	1.1		1.4
Polygonum sp.			0.5	0.6	
<u>Stenotaphrum</u> <u>secundatum</u> (Walt.) Kuntze ^{<u>a</u>/}			0.5	-	1.4
Ludwigia peruviana L.					0.7
Scintello sp.		—		0.6	
Bare hydrosoil	6.3	9.0			

 $\frac{a}{2}$ Not an aquatic plant but grows at the edge of a pond and tolerates flooding.

l

1

(

(

Aquatic vegetation	15-Dec-76	22-Apr-77	15-Jun-77	21-Sep-77	15-Dec-77 ^a /
<u>Hydrilla</u> verticillata Royle	12.1	8.3	0.8	4.8	4.4
Chara sp.	_				0.9
Eleocharis sp.	0.9				
Bacopa sp.		_			3.5
Cyprus sp.	0.9	<u> </u>			_
Lippia sp.		· · · · · · · · · · · · · · · · · · ·			2.6
<u>Stenotaphrum</u> <u>secundatum</u> (Walt.) Kuntze ^b /			2.3	—	14.8
Ludwigia peruviana L.	0.9		4.5		
Vallisneria americana Michx.	14.7	19.3	17.6	34.6	13.9
Panicum repens L.	11.2	11.0	9.2	7.7	12.2
Hydrocotyle umbettata	7.8	3.7	11.5	3.9	9.6
Bare hydrosoil	51.5	57.5	49.5	49.0	38.1

Table 3. Frequency of occurrence (%) of aquatic vegetation in the BCC Pond.

 \underline{a}' Rise in water level flooding plants around the edge of the pond.

 $\frac{b}{b}$ Not an aquatic plant but grows at the edge of the pond and tolerates flooding.

66

[

T

{

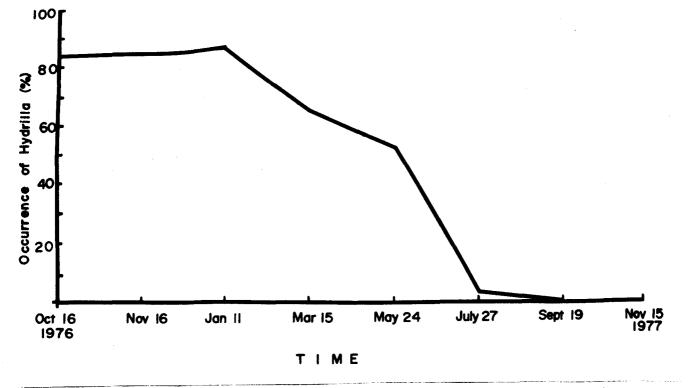
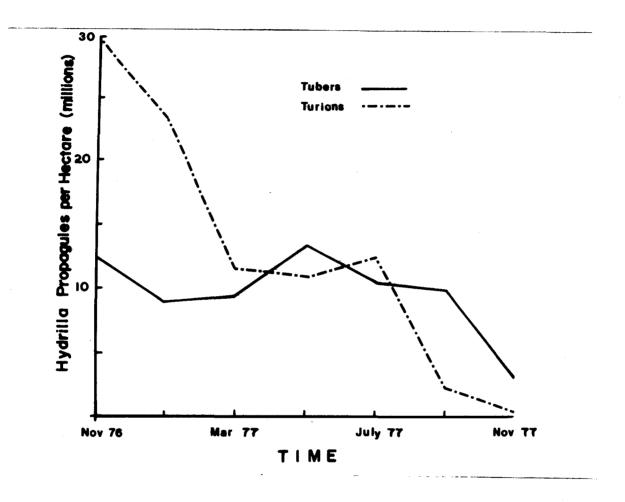
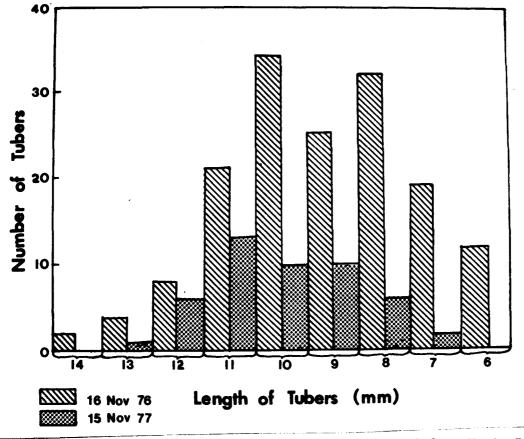
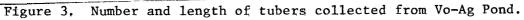


Figure 1. Percent occurance of hydrilla in Vo-Ag Pond as determined by line transect.









WEED CONTROL AND FISH PRODUCTION

Karol Opuszynski

Inland Fisheries Institute, Department of Fish Culture

Zabieniec near Warsaw, 05-500 Piaseczno, Poland

INTRODUCTION

The control of undesirable aquatic vegetation by means of herbivorous fish species appears to be a promising possibility. A desirable effect from the point of view of human interests could possibly be achieved without the danger of chemical methods, and furthermore avoiding labor outlay and the costs of mechanical vegetation control. An additional advantage of a biological method is that useless plant matter is transformed into fish meat utilized directly as a food product, for sport fishing or as feed for other animals of economic importance.

In introducing new fish species it is necessary to analyze their food habits and efficiency of food utilization, as also the food relationship with indigenous species on order to foresee, as far as possible, the effect of these new species on the aquatic vegetation and on fishery production.

Another problem is that of dewatering the influence of the introduced fish species on the water ecosystem as a whole. The immediate advantages gained in bringing in such species to remove undesirable plant groups might well result in more serious, natural and economic consequences.

PHYTOPHAGOUS FISH SPECIES DISCUSSED IN THE PAPER

Since both in Poland as well as in Europe there is a lack of indigenous phytophagous fish species of economic importance, the following Asiatic species were imported from the Soviet Union in 1964: grass carp, <u>Ctenopharyngodon idella</u> (Val.), silver carp, <u>Hypopthalmichthys molitrix</u> (Val.), and bighead carp, <u>Aristichthys nobilis</u> (Rich.). More intensive studies were hitherto carried out on the first two species mentioned, with more intensive studies on the third species planned for the future. The present paper presents the results of studies conducted in Poland on the background of available world literature.

FOOD HABITS AND FOOD UTILIZATION

Grass carp fry initially feed exclusively on animal feed consisting chiefly of plankton crustaceans and Chironomidae larvae. Transition from animal to plant food takes place at a weight of 1.1-1.8 g and body length (longitudo corporis) of 36-43 mm (Opuszyński 1969) or total length (longitudo totalis) of 49 mm (Ciborowska 1972). Some data indicate that this transition depends, apart from the size of fish, also upon water temperature. Scheer et al. (1967) consider that the grass carp feeds on zooplankton up to a water temperature of 15° C, and on plants above this temperature. The proportion of macrophytes ingested increase with growth, the larger individuals living mainly on macroflora. In two-year old fish reared in ponds, the animal food represented less than 1% of all the diet (Figure 1). It was also reported by Boruckij (1952), who studied the food of grass carp 33-58 cm long in the Amur River, that their alimentary tracts were filled with macroflora. The author states that since grass carp feeds on macroflora together with leaves and plant stems, all living organisms such as algae, <u>Rotatoria</u>, Chironomidae larvae, Oligochaetes and other aquatic organisms pass to the alimentary tract. Their number and biomass was, however, negligible in comparison with those of the macroflora.

The type of plant eaten changes with the development of grass carp. Initially its food consists of phytoplankton, filamentous algae, <u>Fontinalis</u> sp., <u>Charales</u> and small species of flower-bearing plants such as <u>Lemna</u> sp., <u>Potamogeton pectinatus</u> L., <u>Elodea canadensis</u> Rich. Larger fish weighing more than 250 g also eat emergent plants.

Food selection of grass carp with respect to aquatic plants was studied under experimental aquarium conditions as well as in the natural environment (Stroganov 1963, Verigin et al. 1963, Penzes and Tolg 1966, Drupauer 1967, Fischer 1968). The results of these experiments are not, however, unequivocal which is probably the result of widely varying experimental conditions. Different sizes of fish and various water temperatures during the experiments make these results difficult to compare. Further, different types of plants were used for food selection.

In general, the foods most desired by grass carp are soft submerged plants. The aquatic plants which are readily consumed include <u>Elodea canadensis</u> Rich., <u>Ceratophyllum demersum</u> L., <u>Potamogeton pectinatus</u> L. and <u>Myriophyllum specatum</u> L. The suitability of other plant species for grass carp is still under discussion. This concerns emergent plants and filamentous algae, which frequently occur in great abundance and create considerable difficulty in the management of fisheries and proper use of water bodies for other purposes.

According to Stroganov (1963) Typha sp. and Phragmites communis Trin., at a height above one meter are not consumed by the grass carp. On the other hand, Aliev (1963) and Verigin et al. (1963) are of the opinion that these two plants are readily eaten. Grass carp first breaks a tall <u>Phragmites</u> <u>communis</u> by catching its leaves and then consumes them together with the soft upper part of the stem. <u>Typha</u> is broken at the base of the shoot and then eaten in toto. Similarities in reed breaking and <u>Typha</u> consumption of <u>Schoeneplectus lacustris</u> L. by grass carp over one kg in weight were observed in our ponds. This does not mean, however, that these plant species are always readily eaten by the fish.

Filamentous algae are most frequently mentioned as the most attractive food for grass carp (Avault 1965, Prihodke and Nosal 1966, Penzes and Tolg 1966). It has been reported by other authors that filamentous algae and <u>Hydrodictyon reticulatum</u> (L.) are not eaten willingly (Stroganov 1963, Tsharyiev and Aliev 1966) or are completely neglected (Ilin and Solovieva 1965). Reluctant consumption of filamentous algae by grass carp has been confirmed in our studies (Opuszyński 1969). In 1965, in ponds, a massive

development of filamentous algae appeared. Quantities of algae were so great that they represented a considerable obstacle while fishing with nets. Despite this fact the amount of algae in grass carp food was small. During the whole season the algae accounted for, on the average, 9% of the total food intake, whereas the consumption of macroflora amounted to 72% of total food intake. When given a choice, grass carp clearly prefer macroflora to algae.

From the point of view of utilizing the grass carp for controlling aquatic vegetation of ponds, an important problem is that of its consumption of feed designed for other fish species. Stevenson (1965) suggests that grass carp may prefer pellets over aquatic plants. This is not supported in our studies, in which the fish were fed on sorghum, the fodder generally used in fisheries in Poland. Analysis of alimentary tracts of grass carp revealed that the share of plants ingested was greater than the fodder. Similar studies were done for fry. The proportion of these plants ingested since fodder for fish was first added, amounted to 86 and 81% of the total food intake of fry in 1965 and 1966, respectively. In one-year old fish macroflora and filamentous algae represent 81% of the food intake during the whole season. Twoyear old fish consume an even greater proportion of macroflora amounting to 91% of their diet. In experiments with two-year old fish the distribution of fodder and plants was similar. There were no aquatic plants in the ponds and these two types of food were introduced at two sites in the pond. During the whole period of investigation fodder was found to dominate in the food of fish only on July 29, 1965 (Figure 1). These results may indicate that under conditions of intensive culture and feeding of fish, grass carp can be used also for controlling aquatic plants.

QUANTITY OF FOOD CONSUMED

Food intake is often given as a percentage of body calorific content, or as a ratio of wet weight of food eaten to wet weight of fish, or simply as wet weight of food consumed. Lukanin (1959) reports that grass carp weighing about 2000 g consume 500-2300 g of fresh vegetation. Verigin (1963) ascertained that daily intake varied from 115 to 1350 g fresh weight per kilogram of fish, depending on the species of plant eaten. Cure (1970) maintains that the grass carp eats its own weight of plants a day at a temperature of 20-28 C. Zolotova (1971) states that the average daily intake of grass carp on a plant diet is 30% of fish body weight.

Fischer (1968) investigated the daily ration of one-year old grass carp kept under laboratory conditions and fed various plants normally eaten by it. The values obtained varied from 660 to 9786 calories per individual, depending on the species composition of the diet. The daily intake of grass carp weighing 20 - 50 g was 3000 - 56,000 calories, when fed on a pure plant diet of one species, while that of 86 - 107 g fish on a pure animal diet was 3000 -10,000 calories. However, a mixed animal-plant diet gave astonishing results (Fischer 1973), the intake being higher than for animal food alone (Figure 2). Food intake increased uniformly with fish body weight when a mixed animalplant diet was fed to excess. However, when plants only were supplied food intake increased considerably with change of fish weight from about 240 to 300 g. Within this relatively narrow weight range daily intake increased threefold from 16 to 45 Kcal. In a 10 day experiment the fish fed on animal food gained about 4% in weight and the fish fed on plant food lost about 2%. According to Fischer's and Lyakuovich's (1973) considerations it is possible that after reaching a weight of about 300 g there is an increased demand for some "minimum factor" (a vitamin or an aminoacid) which results in increased food intake and retarded growth.

There are interesting results of experiments in which grass carp were fed different quantities of animal and plant food (Fischer 1973). The fish faced with an excess of animal food (Figure 3) did not consume it in a constant rate. In the first two series when the food supply seemed to be sufficient, since the fish does not consume it wholly, the amounts of <u>Tubificidae</u> eaten were similar. When the availability of plants was diminished the fish consumed fewer animals. In the reverse situation, however, when animal food was limited and plant food was in excess, the consumption of food did not decrease. The plant food (given in excess) was consumed in greater quantities when the supply of animal food was diminishing. However, when converted to calories (Figure 4) the amount of plant food eaten remained practically unaffected (differences range from 4 to 4.5 Kcal.). From these data (Fischer and Lyakuovich 1973) concluded that the grass carp needs a certain minimum of plant food, which facilitates the ingestion and probably digestion of animal food.

Water temperature exerts an important influence on the amount of food consumed. In feeding fry with <u>Chara</u> sp. and <u>Fontinalis</u> <u>antipyretica</u> L. Opuszyński (1967a) found that the diurnal amount of plant matter consumed at temperatures from 16 to 28 C increased more than twofold and was from 0.5 to 1.03 g/gram of fish body weight. At temperatures from 22 to 28 C the diurnal increase in food consumed was similar for fry and two-year old fish, and reached 29 and 30%, respectively. The significant correlation (r = 0.79) between the amount of hydrilla (<u>Hydrilla verticillata</u> Royle) consumed by the grass carp and water temperature was estimated by Sutton (1974).

Food Conversion and Efficiency

The food conversion ratio, that is the ratio between food intake and growth of fish, was determined for three year old grass carp (Opuszyński 1969). Since vegetation did not develop in the ponds, practically the whole amount of plant food consumed was supplied from outside. The initial stock of grass carp weighing 116 kg/ha consumed from 6652 to 7406 kg/ha of wet plant weight. Consumption of green plant matter per 1 kg of initial stock ranged from 57 to 64 kg at food conversion coefficients of from 25 to 35.

Higher ratios were obtained in feeding grass carp fry weighing 8-10 g and two year old fish weighing 91-109 g with water plants under aquarium conditions (Opuszyński 1967a). Food conversion coefficients in this case depended upon the water temperature and ranged from 78 to 92 (Figure 5). These high food conversion coefficients appear to indicate that the fry had not yet changed from feeding on zooplankton to plant food, and probably still required considerable additions of animal protein in their diet for rapid growth.

Food coefficients for two year olds were more than twofold lower, and were from 36 to 35 at a temperature range of 22-28 C. They were likewise less variable within this range than fry. A considerable increase in the food conversion coefficient was observed at a temperature of 35 C. This temperature was very close to the lethal temperature of 36.6 C (Opuszyński 1967b). Poor growth was not the result of lower food rations. No studies were carried out to determine if the increased food conversion coefficient was caused by poor digestion and food assimilation or by increased maintenance requirements (standard metabolism) of the fish at this high temperature.

Food conversion coefficients averaging 30 were obtained by Verigin et al. (1963). The fish used in his experiment weighed 170-260 g, and the water temperature ranged from 30 to 34 C. Stroganov (1963) in his experiments with fish ponds having a water temperature of not more than 24 C, obtained coefficients ranging from 14 to 21, with an average of 18. These results, similarly as those cited previously for experiments in ponds (Opuszyński 1969), are probably lower because invertebrate fauna consumed by the grass carp in ponds was not taken into consideration.

Fish food conversion coefficients might likewise result from the high water content in the tissues of aquatic plants. Sutton (1974) called attention to this fact when feeding hydrilla to grass carp. The food conversion coefficient calculated for the wet mass of hydrilla was 62. Samples taken for dry weight determinations indicated that the hydrilla contained 91.8% moisture. In other words, 23.2 g of dry hydrilla produced 4.6 g of fish weight. The conversion relationship showing the amount of hydrilla consumed divided by fish growth, indicated that 5.04 g of dry hydrilla were required for each 1.0 g increase in growth of these fish.

Fischer (1972) calculated a food assimilation and energy budget for grass carp fed on animal and plant food under aquarium conditions. As animal food tubificids (<u>Tubifex</u> <u>tubifex</u>) were used and as plant food lettuce (<u>Lactuca</u> <u>sativa</u>). Fish fed exclusively with plants had a gross production efficiency index $K_1 = 2.2\%$ (this is the proportion of food intake used for fish growth) whereas those 2.2% fed with animals $K_1 = 12.5\%$. The assimilability of plants was also very low and amounted to about 20%.

Fischer and Lyakuovich (1973) assumed that under natural conditions, where fish consume both plant and animals, growth was caused mainly by animal proteins and by lipids of both animal and plant origin. It is more likely that carbohydrates (mostly in plants) and proteins of plant origin are used chiefly for energy metabolism. Glycogenesis probably occurs to a marked extent.

Fischer's values produced under laboratory conditions with excess food and a strict monospecific diet are probably lower than those for natural conditions. Nevertheless, it can be concluded from these data of the importance played by addition of animal feed to the diet of grass carp, and of the probable drop in food utilization for growth as the share of the animal food component is decreased.

SILVER CARP

Food Quality

The silver carp is also a phytophagous fish which filters its food

from the water by means of gill rakers embedded in the gill arches. The space between gill rakers in adult fish is 20 to 25 microns (Voropajev 1968). This species also feeds on zooplankton during its initial period of life (Opuszyński 1969, Ciborowska 1972). It was found that phytoplankton became the essential food component when fry reach a body length (longitudo corporis) of over 32 mm and a weight of 0.7-0.8 g (Figure 6). In contrast to the grass carp, vegetation does not always constitute the main food component of older silver carp. As can be seen from Figure 1, zooplankton was the predominant food of two year old silver carp in ponds.

The food conposition of the silver carp is very variable. Detritus makes up 90-99% of the food in spring in the Amur River, whereas in summer during the peak period of blooms it drops to 5-15% with zooplankton constituting 1% and phytoplankton the remaining part (Boruckij 1950, Verigin 1950). Depending upon the type of natural waters and fishery ponds, some authors found chiefly phytoplankton in the digestive tracts of silver carp, while others, detritus (Boruckij 1973). Opuszyński (In Press) found that the percent share of phytoplankton and zooplankton drops while detritus increases in the food as density of the fish in ponds grows from 1.5 to 12 thousand individuals per ha. The percent share of phytoplankton and zooplankton in consecutive densities of silver carp was 10.1, 4.3, 3.9, 1.6, 1.4.

Food selectivity of the silver carp is a controversial question. Some authors (Salar 1967, Lupaceva 1969, Tarasova 1970, 1971) are of the opinion that phytoplankton dominant in the environment constitutes the basic food of fish. These authors compare the activity of silver carp to a "natural plankton net". On the other hand other authors found distince selectivity of the fish with respect to specific groups and species of algae (Savina 1965a and b, Kajak 1977, Opuszyński In Press). These authors are of the opinion that the silver carp cannot by any means be compared to a plankton net filtering mechanical particles of a given size. There is no universal opinion as concerns consumption of blue-green algae. These algae are considered as being both willingly consumed (Muchamedova 1974), and as avoided by the fish (Omarov and Lazareva 1974, Kajak 1977).

Quantity of Food Consumed

A number of authors when investigating the nutrition of silver carp determined the amount of food found in the digestive tracts. This amount is in general expressed in 0/000 as the index of filling, or the amount of feed multiplied 10 thousand times per unit of body weight of the fish under study. The index of filling for the silver carp from the Amur River was found to be $57 - 582 \ 0/000$ (Boruckij 1973), and from ponds as follows: 502 - 547 (Savina 1965b), 142 - 143 (Omarov 1970), $80 - 570 \ 0/000$ (Tarasova 1970). Similar values were obtained in our investigations (Table 1).

In order to determine the actual amount of food consumed by silver carp it is necessary to know the rapidity with which food passes through the digestive tract. Omarov (1970) found that at a water temperature of 23 C filling and emptying of the digestive tract takes place 6 times diurnally. By multiplying the amount of food found in the digestive tract of silver carp by 6, the diurnal food ration of this species ranges from 17 to 20% of the body weight.

Food Conversion and Efficiency

Opuszyński (In Press) found that the index of food conversion efficiency for the silver carp was 2-6% (Table 2). Without doubt this index is low. Attention is likewise called to the fact that the value of this index drops as density of fish increases. There are not data in the literature with which the values obtained could be compared.

BIGHEAD CARP

I have little to say as concerns the bighead carp. As shown by Opuszyński (In Preparation) zooplankton is of greater significance as food than for silver carp (Figure 7). This might possibly be due to the sparser spacing of filtering rakers than in silver carp. The spacing of filtering rakers in the bighead carp ranges from 20 to 60 microns (Vovopajev 1968).

FOOD RELATIONSHIPS WITH INDIGENOUS FISH

In view of the fact that in reality there is a lack of herbivorous fishes in the European ichthyofauna, food competition between indigenous fish species and introduced Asiatic ones can be limited by comparing the amount and type of animal food consumed by these species. Comparisons were made on the animal food composition of carp, grass carp and silver carp raised jointly in ponds (Opuszyński 1968, 1969). The carp seemed to be the most typical representative of indigenous ichthyofauna, as this is a species basically consuming all species of invertebrates on which other autochtonic fish feed.

Animal food was characterized by means of the index of filling/quotient of animal food weight and fish weight in the 0/000/ and also by the index of food convergence after Shorigin. The latter is the sum of lower percent shares of food consitutents appearing in the food of two fish species under comparison. The index varies within the limits from 0 - when the food composition in the species compared is entirely at variance - to 100 when it is exactly the same.

The index of filling for animal food in grass carp fry in the respective years under consideration was only 2.6 to 4.7 times less than that for carp fry (Table 3). At the same time the convergence of food composition in the two species was high and ranged from 33.2 to 51.8. The index of filling in older grass carp individuals showed a downward trend. In the stocks of twoyear old fish, it was 11.4 times less than that in carp. A high convergence of animal food composition in carp and grass carp was due chiefly to the fact that both fish species fed on the same groups of Chironomidae larvae. Larger species of Cladocera and Copepods were also important (mainly <u>Daphnia longis</u>pina Mull) although in grass carp food it tended to decrease with fish growth.

With regard to silver carp, the index of filling varied considerably as compared with carp. It was 1.7 (fry during the cold season of 1965) to 12.1 times less than that of carp and grass carp in individual experiments. The convergence of food composition in carp and silver carp was less marked than that in carp and grass carp. This convergence in individual years under observation ranges from 3.7 to 25.4 (Table 3). Crustacean plankton invariably represented the basic animal food constituent in silver carp. The amount of crustacean plankton tended to increase as fish aged. It accounted for 98% of the animal food of two-year old silver carp, and 93% of all the crustacean plankton weight was attributed to Bosmina longirostris Mull.

The development of <u>B</u>. <u>longirostris</u> on a mass scale should be looked upon as a harmful side effect of intensified carp production. These crustaceans which are consumed by carp only to a very small extent, feed on phytoplankton which serves for a staple food of other animal organisms significant in the carp diet. Therefore, the fact that the silver carp feeds on <u>B</u>. <u>longirostris</u> seems to be of double advantage. In addition to a better utilization of food resources for production, it may have a restrictive effect on the numerical increase of this crustacean.

INFLUENCE ON THE ECOSYSTEM

Control of Aquatic Plants

The effectiveness of grass carp in controlling plant overgrowth in bodies of water depends on a number of factors, the most important probably being water temperature, composition of plant species and individual weight of fish and stock density. Elevated water temperature and stock density augment the effectiveness of grass carp in controlling aquatic plants. Man's contribution to the solution of this problem is limited to controlling stock density.

According to investigations carried out under the moderate climatic conditions of Poland there is a general opinion that the grass carp can hamper the development of macrophytes to a great extent, but it cannot completely solve the problem of overgrowth of inland water (Opuszyński 1969, 1972, Bernatowicz and Wolny 1969). Two factors limiting grass carp utilization for aquatic weed control are food selectivity and the relatively high temperature needed for active feeding. Effective control could be obtained using a very high stocking density with a high initial weight of individual fish. This, however, due to slow growth of fish under such conditions, would not be economically justified in countries where grass carp is a marketable fish.

It should be stated that partially positive results were obtained even in a relatively cold climate in England (Stott and Robson 1970). The mean daily water temperature was 15.8 C with a range of 8.5 - 21.5 C. A stock density giving a mid-season biomass of approximately 300 kg of two-year old fish reduced plant growth to about 50% of its potential. Fish growth was not satisfactory (the mean weight increment was 37.8 g), and the mean survival was only 61%. In the moderate climate of the Moscow area, ponds did not show excessive overgrowth during the entire season with a stock of 200 - 600 grass carp/ha (500 - 700 kg/ha). The initial mean weight of individual fish was over 1 kg. After exhaustion of the preferred plants, grass carp ate the "compulsory plants" (plant species which are not consumed in the presence of other food). The growth rate of fish was slow.

The optimistic evaluation of the effectivity of grass carp in the control of aquatic plants in the German Democratic Republic is given by Janichen (1976). According to this author the following premises must exist for the successful application of grass carp: (1) sufficient water quality; (2) water temperatures higher than 15-16 C; (3) water depths of at least 0.3 m with areas of 1 m and deeper; (4) exclusion of waters with single-species-colonization, for instance by <u>Stratiotes aloides</u>, <u>Ranunculus</u> sp. or with an abundant development of <u>Nymphaceae</u>; (5) objects suitable to prevent any migration of fishes; (6) fishes with a minimum age of two years; and (7) mean stocking rate of 200 kg/ha, which is to be reduced or raised due to the extent of plant density.

The effectiveness of grass carp for plant control increases with an increase in temperature. A good example is represented by the results obtained in the moderate climatic zone in water heated by effluents from a thermal power station. Attempts to use grass carp for this purpose were carried out in a heated lake in Poland. About 5000 grass carp of individual weight 250 -1000 g were introduced to this lake of 154 ha. The fish considerably reduced the plants in the lake. The only plant that remained was Nuphar luteum Sm., which is reluctantly eaten by grass carp. In order to learn if these plants disappeared as a result of the introduction of phytophagous fish, a portion of the littoral zone was separated by means of a wire net 25 x 10 m in size. Plants grew in abundance only in this zone, where no fish could enter. In the middle of July the water level in the lake was increased and the edge of the net was about 30 cm above the water surface. As a result, several grass carp of about 6 kg jumped over the net. Three days later no plants except Najas sp., which is reluctantly eaten by these fish, were seen in the isolated area (Horoszewicz, personal communication).

Stocking grass carp into water bodies heated by thermal power stations in a moderate climate as well as water bodies in a hot and tropical climate may solve the problem of overgrowth in these waters without the use of the traditional methods. The only condition is that the vital requirements of the fish must be met.

Phytoplankton

Massive algae blooms are the most visible and troublesome symptoms of rapidly progressing eutrophication of surface waters resulting from human activities. The problem of controlling communities of plankton algae is one of highest priority.

Silver carp, because of food habits, may be a way to control phytoplankton by direct consumption by a suitably large population of fish (Prowse 1969, Vovk 1974). Other authors (Opuszynski 1972, Grygierek 1973, Barthelmes 1975a), although optimistic as to the role of the silver carp in this respect, call attention to the possibility of more complicated effects due to this species on the biocenosis beyond the direct relationship of silver carp and phytoplankton. Experimental studies allowing for an evaluation of the role of the silver carp in counteracting eutrophication processes are, however, few in number, and their results discrepant (Januszko 1972, 1974; Kajak et al. 1975).

Recent studies by Januszko (In Press) showed increases in phytoplankton biomass in ponds stocked with carp and silver carp as compared to ponds treated as controls and stocked with carp only. The stocking density with silver carp was 4, 8 and 12 thousand fish per ha. The control group of ponds showed an average seasonal phytoplankton biomass of 16 g/m³ whereas ponds with successive silver carp densities showed 21, 18 and 22 g/m^3 . The dynamics of seasonal changes in phytoplankton biomass are shown in Figure 8.

Changes occurred within the plankton algae community as silver carp stocking densities were increased. The percental share of diatoms increased and Chlorophyceae decreased. In consecutive groups of ponds the biomass of diatoms increased 32, 42 and 66% as compared to the control ponds. The decline in the biomass of Chlorophyceae of 18, 35 and 37% in consecutive groups of ponds as compared to the control ponds was due chiefly to the three to fivefold drop in the biomass of the dominant species, <u>Chlorella minutissima</u> Fott, which is a small algae (3 microns). It passes easily through the gill filtration apparatus. The average weight of plankton algae was higher in ponds with silver carp than in the controls.

Other experiments differ in their results from those under discussion herein. The influence of silver carp on plankton and benthos was investigated in enclosures of plastic foil $(2.5 \times 2.5 \text{ and } 1.5 \text{ m} \text{ in depth})$ in a eutrophic lake. A fourfold drop in the phytoplankton biomass and twofold growth of the benthos biomass was found in the enclosures with silver carp, as compared to non-stocked control enclosures (Kajak et al. 1975).

Januszko (1974) found considerable effect of bighead carp on the biomass and composition of algal communities. In the ponds stocked with 1500 bighead carp/ha and 2000 common carp/ha there was more than a twofold increase in algal biomass in comparison with control ponds stocked with carp alone. The mean seasonal biomass value in the control group was 32 g/m^3 and in the group with bighead carp 67 g/m³. In control ponds blue-green algae biomass (Cyanophyta), mainly Anabaena flos aquae, increased innumerably (Figure 9).

Influence on Fish Production

Studies on the effect of phytophagous fish on pond production have been carried out in various European countries. In general, all investigators emphasized the possibility of obtaining additional yield without a diminition in production of carp. Little is known, however, as to the validity of the data describing the magnitude of this additional production, since only in a few studies have control ponds been used.

Experiments conducted to determine the effect of additional stocks of fry and one- and two-year old phytophagous fish on carp production were conducted in Poland (Opuszyński 1969). The results of these experiments are presented in Figure 10. Each column represents the mean from three experimental ponds similar in area and depth. The ratio of carp to phytophagous fish was 1:1. The stocks of phytophagous fish were composed of 50 percent grass carp and 50 percent silver carp. It is seen from Figure 10 that in both years the additional stock of phytophagous fish caused a decrease in production of carp fry, although in 1966 the total polyculture production was 338 kg/ha higher than in monoculture. Considerable differences between the results of both years can be explained in terms of different thermal conditions as 1965 was an exceptionally cool year.

Carp production was not decreased when older phytophagous fish were stocked. These fish gave an additional yield of 330-370 kg/ha, representing an increase of 20-25 percent. It should be emphasized, that in the fish farm, in which the experiments were conducted with one- and two-year old fish, the growth of the silver carp was much slower than in other farms in Poland. This was probably due to exceptionally high percentages of blue-green algae in the phytoplankton.

The main reason for reduced carp fry production in polyculture situations seems to have been food competition between carp and grass carp. This was evident when we analyzed the index of filling for animal food and the convergence of food composition.

Stocking silver carp within the range of 4 to 12 thousand two-year old fish per hectare resulted in a decline in carp production. This decline occurred at the lower silver carp stocking rates, whereas density increases of from 8 to 12 thousand individuals per hectare did not cause a further drop in carp production (Figure 11). Lowered carp production was caused by poor fish growth rather than increased mortality. Increased silver carp densities resulted in reduced individual weight increments as silver carp production increased. The highest silver carp stocking density increased total production of ponds by almost 800 kg/ha in high density carp ponds.

Causes for the decrease in carp production cannot be attributed only to direct food interrelations between the two fish species. Although the silver carp caused a decrease in the numbers of zooplankton (Grygierek, In Print), the numbers and biomass of Chironomidae larvae (Wasilewska, In Print) increased. In all probability, complex ecological mechanisms might have caused changes in behavior as a result of fish density. Similar results have been described previously (Ivlev 1955, Nelsson 1967).

> Influence on Some Elements of the Environment and the Biocenosis

Grass carp

In addition to reduction of aquatic weeds, grass carp may induce considerable changes in biocenosis. This process has not been subjected to detailed study until recently.

Taking into consideration the previously cited data on food consumption and assimilation, I assume that an initial stock of grass carp weighing 100 kg/ha discharges approximately 5000 kg/ha of excrements into the environment during one growing season under temperate climatic conditions (May to September). Grass carp possess strong pharyngeal teeth with sharp masticating surfaces. As a result the food becomes highly disintegrated. According to Hickling (1966) the plant particles after passing through the alimentary tract are smaller than 3 sq. mm. Thus, the feeding activity of grass carp releases into the environment large quantities of highly disintegrated and only partly digested plant parts. This process may cause intensive fertilization of water bodies, as grass carp excrements are easily decomposed by micro-organisms. As a result the released nutrients may cause development of phytoplankton. Cure (1971) reported increased eutrophication of a pond following introduction of grass carp at a stocking rate of about 70 two-year old fish/ha. From year to year the pond became more and more eutrophic. Primary plankton production

increased gradually, as macrophytes diminished. Before stocking, in 1967, the pond was unproductive with a primary plankton production in the range of -6.7 and +1.0 g $0_2/m^2/day$, in 1968, after stocking and until 1970, the pond was considered as mesotrophic-eutrophic (+2.6 to 7.1 g $0_2/m^2/day$ and even highly eutrophic +11.1 g $0_2/m^2/day$). Cure (1971) maintains that in ponds strongly overgrown with macrophytes, non-drainable and with a thick mud bottom, grass carp at a moderate rate are able to substitute for organic and mineral fertilization.

Removal of vascular plants can result in disruption of the ecological equilibrium in a water body since these plants can effectively assimilate biogenic compounds of autochthonic and allochthonic origin. In consequence, reduction of macroflora by grass carp may indirectly contribute to the development of other groups of aquatic plants, thus leading to undesirable phenomenons such as water blooms.

Stocking of water bodies with grass carp may directly cause alterations in species composition and fish abundance. This was observed in a heated lake in Poland as the spawning grounds of fish laying their eggs on the aquatic plants was destroyed (Backiel, personal communication).

Silver Carp

The importance of biological methods in counteracting eutrophication processes led to the decision to undertake investigations pertaining to these methods by a group of workers of the Inland Fisheries Institute in Poland. It was decided to increases stocking densities with silver carp to very high levels in a number of ponds (Figure 11), and to investigate the influence of such densities on selected elements of the environment and the biocenosis. These investigations were carried out on physico-chemical conditions (Piotrowska, In Press), bacteria (Kruger, In Press), phytoplankton (Januszko, In Press), zooplankton (Grygierek, In Press), benthos (Wasilewska, In Press), fish health (Peitrzak, In Press), and food relationships and fishery production (Opuszyński, In Press).

The presence of silver carp in the ponds resulted in a number of environmental and biocenosis changes. Changes in particular factors varied with respect to their course, magnitude and frequency but were not directly correlated with silver carp stocking density, since consecutive densities of stocking did not always result in a gradual intensification of change (Figure 12).

When compared to the control group the greatest effect of the silver carp was noted with respect to the chlorophyll content of the water, primary production and the number of bacteria. As concerns the groups of bacteria under study, proteolytic and ammonification bacteria in the water, and total numbers of bacteria and denitrification bacteria in sediments showed the strongest reaction to environmental changes. Smaller changes of a positive character were found with respect to oxygen content, biochemical oxygen demand (BOD₅), pH values (an average 8.1 in control ponds and 8.3 in ponds with silver carp), organic phosphorus content, total number of bacteria, phytoplankton biomass and benthos number and biomass.

As to negative changes due to the influence of silver carp the following

should be mentioned: lower levels of nitrogen, phosphorus, and dissolved carbonates; lower levels of accumulation of organic carbon, nitrogen and total phosphorus in the sediments. The only, but very strong negative change in the biocenosis, consisted of a drop in the numbers and biomass of zooplankton.

According to our investigations the role played by the silver carp in the circulation of matter appears to consist of ingesting dead and living organisms, decomposing them and rapidly transporting them to the bottom in the form of excrements. Fish excrements are, then, acknowledged to be a good medium for bacteria, undergoing rapid mineralization in the water. It can be concluded from these deliberations that as silver carp stocking rates are increased, the inflow of partially decomposed plant matter should likewise increase. Simultaneously, an increasingly smaller part of the algal biomass (per biomass unit of fish) would be incorporated into fish flesh and removed from the ecosystem (Table 2). Hence, a basic effect of introducing very high stocking densities of silver carp would consist of a considerable acceleration of circulation of matter in the water. Mosevic and Rodhe (Wrobel 1965) are of the opinion that the rapidity with which circulation of matter takes place is of greater importance than the direct supply of mineral nutrients.

Results of our investigations carried out in carp ponds do not indicate any possibility of controlling algal blooms by means of large stocks of silver carp. The reasons for this are as follows: (a) low effectiveness of feeding on algae due to detrital feeding; (b) elimination of zooplankton, which in turn feeds on phytoplankton; (c) more rapid circulation of biogens; and (d) reduced weight increments of fish as stocking densities are increased.

As mentioned above, Kajak et al. (1975) found in a eutrophic lake a significant drop in the phytoplankton biomass due to silver carp; however, in their experiment the factors causing rapid circulation of matter and biogens to the water depths were lacking when compared to our experiments. Circulation of water and the presence of a large stock of benthic fish constitute the factors in question. The diurnal stratification cycle in ponds corresponds to an annual cycle in lakes, and the carp is a fish known for its intensive mixing of sediments. In all probability, the lack of detritus in the water was the reason that silver carp in plastic foil enclosures did not cause a plankton bloom. Finally, our ponds were intensively fertilized with mineral fertilizers which constituted a basic difference between the two experiments.

Kajak (1976) suggests that the very heavy pressure of silver carp on phytoplankton can, in certain circumstances, efficiently improve the water purity in lakes, at least for some period of time. The feces of silver carp sink rather rapidly (1-3 m/sec) (Barthelmes 1975b), thus they leave the epilimnion before significant decomposition and nutrient release takes place. In comparatively stagnant environments, the nutrients were thus removed from circulation for months. In the water bodies with a definite thermocline, the nutrients would not recycle during the stagnation period.

CONCLUSIONS

Efforts were made in this paper to present an overall picture of the effects of phytophagous fish on the environment and biocenosis of water bodies, and not limiting it only to the influence of these species to selected plants. The grass carp under north, central and east European climatic conditions can contribute to the control of plants in inland waters. The grass carp can in many cases effectively substitute present mechanical and chemical methods of control in warm and tropical climatic conditions, and also in water bodies heated by effluents from thermal power plants.

Considerable eutrophication can be one of the consequences of introducing the grass carp. Removal of vascular plants, high food rations connected with low food assimilation, and rapid decomposition of excrements might possibly accelerate circulation of biogenic substances in the water body. As a result of these precesses considerable bloomings of algae can be expected.

In order to counteract the above mentioned phenomena it appears sensible to introduce fish species which feed on phytoplankton. We have endeavored to show, however, that the effect of the silver carp and the bighead carp on the ecosystem is more complex than was at first assumed. At least in some cases these fish can cause further growth in the phytoplankton biomass and evoke unfavorable changes in its species composition.

Counteracting eutrophication processes by means of phytophagous fish is of great scientific and practical importance, and for this reason requires further intensive investigations. As concerns the influence of herbivorous fish on fishery production, favorable results can be expected at least in such simple and controlled human ecosystems as warm-water fish ponds.

REFERENCES

- Aliev, D.S. 1963. Trial of using grass carp for aquatic weed control. In: Problemy Rybokhoziaistvennego ispolzovanija rastitelnojadnych ryb v vodoemach SSSR. Aschabad: 203 - 208 (In Russia).
- Avault, I.W. 1965. Preliminary studies with grass carp for aquatic weed control. Prog. Fish-Cult. 4:207-208.
- Barthelmes, D. 1975a. Elemente der Sauerstoffbilanz in Karpfenteichen, ihre Wirkingsweise sowie die Optimierungsmoglichkeiten durch Silberkarpfen. Z. Binnenfischerei DDR. 12:355-363.
 - 1975b. Zur Phosphatregeneration durch Silverkarpfen. In: Suspendiertes Material in Binnen und Kustengewassern, Reinhardsbrum. 11-12.
- Bernatowicz, St., Wolny, P. 1969. Fisheries Botany, PWRiL, Warszawa, 420 (In Polish).
- Boruckij, E.W. 1950. Materials on feeding of silver carp (<u>Hypophthalmichthys</u> <u>molitrix</u> Val.). Proc. of Amur Ichthyol. Expedition, 1945-1949. Vol. 1:319-331. Moskva (In Russian).
 - 1952. Materials on feeding of <u>Ctenopharyngodon idella</u> and <u>Plagiognathops microlepis</u> in the basin of Amur River. Proc. of Amur Ichthyol. Expedition 1945-1949. Vol 3:505-511. Moskva (In Russian).
 - 1973. Feeding of silver carp (Hypopthalmichthys molitrix Val.) and bighead (Aristichthys nobolis Val.) in natural waters of ponds of SSSR. In: Trofologia Vodnych Zinotnych, "Nauka," Moskva:299-322.
- Ciborowska, J. 1972. The food of phytophagous fish (<u>Ctenopharyngodon idella</u> Val., <u>Hypophthalmichthys molitrix</u> Val. and <u>Aristichthys nobilis</u> Rich.) reared with common carp in fry ponds. Rocz. Nauk Roln. H-94-2:41-58 (In Polish).
- Cure, V. 1970. The development of grass carp (<u>Ctenopharyngodon idella</u> Val.) in Frasinet Pond. Bull. Cerc. Pisc. 4:31-49. (In Rumanian).
- 1971. Primary planktonic production in the Frasinet Pond before and after stocking with grass carp. Bul. Cerc. Pisc. 1:3-8. (In Rumanian).
- Fischer, Z. 1968. Food selection in grass carp (<u>Ctenopharyngodon idella</u> Val.) under experimental conditions. Pol. Arch. Hydrobiol. 15:1-8.
 - 1972. The elements of energy balance in grass carp (<u>Ctenopharyn-godon idella</u> Val.). Part 3. Assimilability of porteins, carbohydrates, and lipids by fish fed with plant and animal food. Pol. Arch. Hydrobiol. 19:83-95.

Fischer, Z. 1973. The elements of energy balance in grass carp (<u>Ctenopharyn-godon idella</u> Val.). Part 4. Consumption rate of grass carp fed on different type of food. Pol. Arch. Hydrobiol. 20:311-320.

Fischer Z. and V.P. Lyakhnovich. 1973. Biology and bioenergetics of grass carp (<u>Ctenopharyngodon idella</u> Val.). Pol. Arch. Hydrobiol. 4:521-557.

Grygierek, E. 1973. The influence of phytophagous fish on pond zooplankton. Aquaculture. 2:197-208.

In Press. The influence of silver carp, <u>Hypopthalmichthys</u> <u>molitrix</u> (Val.), on eutrophication of the environment of carp ponds. Part 4. Zooplankton. Rocz. Nauk Roln. 99-H-2.

Hickling, C.P. 1966. The feeding process in the white amur. J. Zool. (London). 148:408-419.

Ilin, W.M. and L.M. Solovieva. 1965. Production and wintering of yearlings of phytophagous fish. Vopr. Prad. Ryb. 13:11-21.

Ivlev, V.S. 1955. Experimental ecology and fish feeding. Piscepromuzdat, Moskva. 251 pp (In Russian).

Januszko, M. 1972. Algae in ponds with the fry of common carp and phytophagous fish. Rocz. Nauk Roln. 94-H-2:81-97. (In Polish).

1974. The effect of three species of phytophagous fish on algae development. Pol. Arch. Hydrobiol. 21:431-454.

In Press. The influence of silver carp, <u>Hypopthalmichthys</u> <u>molitrix</u> (Val.), on eutrophication of the environment of carp ponds. Part 3. Phytoplankton. Rocz. Nauk Roln. 99-H-2.

Janichen, H. 1976. Combat against macrophytes by <u>Ctenopharyngodon idella</u>. Proc. Internation. Symposium on Eutrophication. Karl-Marx-Stadt. GDR:37-45.

Kajak, Z. 1976. Considerations on the influence of fish on the functioning of lake ecosystem and resulting water quality. Proc. International Symposium on Eutrophication. Karl-Marx-Stadt. GDR:51-59.

1977. Studies on selectivity of silver carp, <u>Hypopthalmichthys</u> molitrix (Val.). Ekol. Pol. 2:227-239.

Kajak, Z., J.I. Rybak, I. Spodniewska and W.A. Godlewska-Lipowa. 1975. Influence of the planktonivorous fish <u>Hypopthalmichthys molitrix</u> (Val.) on the plankton and benthos of the eutrophic lake. Pol. Arch. Hydrobiol. 2:301-310.

Krupauer, V. 1967. Food selection of two-year old grass carp. Bul. VUR Vodnany. 1:1-11 (In Czech.).

- Kruger, D. In Press. The influence of silver carp, <u>Hypopthalmichthys</u> <u>moli-trix</u> (Val.), on eutrophication of the environment of carp ponds. Part 2. Bacteria. Rocz. Nauk Roln. 99-H-2.
- Lukanin, A. 1959. Experiments on growth of white amur in Angelinski ponds. Ryb. Khoz. 35:82-83. (In Russian.)
- Lupaceva, L.I. 1969. Feeding of silver carp in ponds of Cjurupinskoj Fish Breeding Farm. Ryb. Choz. Keiv. 9:35-37. (In Russian.)
- Muchamedova, A.F. 1974. Feeding and growth of young silver carp in ponds Cimlianskoj Fish Breeding Farm. Izv. Gosud. Naucno-issled. Inst. Ozer. Recn. Ryb. Choz. 88:231-245.
- Nilsson, N.A. 1967. Interactive segregation between fish species. In: The biological basis of freshwater fish production. Blackwell Scientific Publications, Oxford and Edinburg. 259-315.
- Omarov, M.O. 1970. Daily food ration of silver carp, <u>Hypopthalmichthys moli</u>trix (Val.). Vopr. Ichtol. 3:580-582. (In Russian.)
- Omarov, M.O. and L.P. Lazareva. 1974. Feeding of silver carp in waters of Dagestan. Gidrobiol. Z. 10:100-103. (In Russian.)
- Opuszyński, K. 1967a. Feeding of <u>Ctenopharyngodon</u> idella Val. on aquatic plants under aquarium conditions. Rocz. Nauk. Roln. 90-H-3:453-462. (In Polish.)

1967b. Comparison of temperature and oxygen tolerance in grass carp <u>Ctenopharyngodon idella</u> Val., silver carp <u>Hypopthalmichthys</u> molitrix Val., and mirror carp Cyprinus carpio L. Ekol. Pol. 17:385-400.

1968. Carp polyculture with plant-feeding fish: Grass carp (<u>Ctenopharyngodon</u> idella Val.), and silver carp (<u>Hypopthalmichthys</u> molitrix Val.). Bul. Acad. Pol. Science. 11:677-681.

1969. Production of plant-feeding fish (<u>Ctenopharyngodon</u> <u>idella</u> Val. and <u>Hypopthalmichthys molitrix</u> Val.) in carp ponds. Roczn. Nauk. Roln. 91-H-2. (In Polish.)

1972. Use of phytophagous fish to control aquatic plants. Aquaculture. 1:61-73.

In Press. Silver carp, <u>Hypopthalmichthys molitrix</u> (Val.), in carp ponds. I. Fishery production and food relationship. Ekol. Pol. 27.

In Press. Silver carp, <u>Hypopthalmichthys</u> molitrix (Val.), in carp ponds. III. Influence on ecosystem. Ekol. Pol. 27.

Penzes, B. and I. Tolg. 1966. Aquaristische Untersuchung des Pflanzenverbrauches des Graskarpfen (<u>Ctenopharyngodon idella</u> Cuv. et Val.) Z. Fish. Deren Hilswiss. 1/2:131-137.

- Pietrzak, B. In Press. The influence of silver carp, <u>Hypopthalmichthys moli-</u> <u>trix</u> (Val.), on eutrophication of the environment of carp ponds. Part VI. The state of health of fish. Roczn. Nauk Roln. 99-H-2.
- Piotrowska, W. In Press. The influence of silver carp, <u>Hypopthalmichthys</u> <u>molitrix</u> (Val.), on eutrophication of the environment of carp ponds. Part 1. Physico-chemical conditions. Roczn. Nauk Roln. 99-H-2.
- Prihodko, V.A. and A.D. Nosal. 1966. Production and breeding of grass carp and silver carp in Ukraine. In: Phytophagous fish. Moscov: 40-52. (In Russian.)
- Prowse, G.A. 1969. The role of cultured pond fish in the control of eutrophication in lakes and dams. Verh. Int. Ver. Limnol. 17:714-718.
- Salar, V.M. 1957. The phytoplankton in ponds of Moldavia and its significance in the feeding of phytophagous fish. In: VII Zjazd Hydrobiologow Polskich:119-120. (In Polish.)
- Savina, R.A. 1965a. Filter-feeding of silver carp. Vopr. Ichtiol. 1:135-140. (In Russian.)

1965b. Feeding and growth of silver carp in ponds. Vopr. Prud. Rybov. XIII:47-54. (In Russian.)

- Scheer, D., H. Jahnichen and K. Grahl. 1967. Beogachtungen bei der Haltung von ein = und zwei = sommigen Graskarpien (<u>Ctenopharyngodon</u> <u>idella</u>) in kleinen Teichen in Gebiet von Karl-Marx-Stadt. Dt. Fish. Ztg., Radebeul. 14:141-146.
- Stevenson, I.H. 1965. Observations of grass carp in Arkansas. Prog. Fish-Cult. 4:202-206.
- Stott, B. and T.O. Robson. 1970. Efficiency of grass carp (<u>Ctenopharyngodon</u> <u>idella</u> Val.) in controlling submerged water weeds. Nature (London). <u>5248:870.</u>
- Stroganov, N.S. 1963. Food selection in grass carp. In: Problemy Rybochoziaistviennogo Ispolzovania Rastitielnojaknych ryb v vodoemach SSSR. Aschabad:181-191. (In Russian.)

Sutton, D.L. 1974. Utilization of hydrilla by white amur. Hyacinth Control Journal. 12:66-70.

Tarasova, O.M. 1970. Feeding and food relationship of two-year old silver carp and common carp in ponds. Ryb. Choz. Kiev. 11:30-33. (In Russian,)

1971. Feeding of the silver carp in ponds of experimental fish farm "Nivka", Ryb. Choz. Kiev. 13:51-53. (In Russian.)

Tsharyiev, R. and D.S. Aliev. 1966. The use of grass carp for water weed control at the ponds stocked with carp. Rybochziaistviennoe Osvoienie Rastitielnojadnych Ryb. Moscov:77-82. (In Russian.) Verigin, B.V. 1950. The development of young silver carp with connection in biology. Proc. of Amur Ichthyol. Expedition, 1945-1949. 1:303-318.

1963. The present state and perspectives of utilization of silver carp and grass carp in water bodies of USSR. In: Problemy Rybokhoziaistviennogo Ispolzovania Rastitielnojadnych Ryb v Vodoemach SSSR. Aschabad, Izd. Akad. Nauk Turkmenskoj SSSR:20-38. (In Russian.)

- Verigin, B.V., V. Nguen and D. Nguen. 1963. Food selection and daily ration of grass carp. In: Problemy Rybokhoziaistviennogo ispolzovanija rastitielnojadnych ryb v vodoemach SSSR. Aschabad, Izd. Akad. Nauk Turkmenskoj SSSR:192-194. (In Russian.)
- Voropajev, N.V. 1968. Morphological features, feeding and some production data of the silver carp, the bighead carp and their hybrids. In: Novyje Issledovanija po ekologii i razvedeniju rastitelnojadnych ryb. Izd. "Nauka", Moskva:206-217.
- Vovk, P.S. 1974. Possibility of utilization of the silver carp, <u>Hypopthal-michthys molitrix</u> (Val.) for increasing fishery production and decreasing the eutrophication level in Dneprovsk Impoundments. Vopr. Ichtiol. 3: 406-414. (In Russian.)
- Wasilewska, B. In Press. The influence of silver carp, <u>Hypopthalmichthys</u> <u>molitrix</u> (Val.), on eutrophication of the environment of carp ponds. Part V. Benthos. Rocz. Nauk Roln. 99-H-2.

Wrobel, S. 1965. Causes and consequences of pond eutrophication. Acta. Hydrobiol. 7:25-52.

Zolotova, E.K. 1971. On possibility of utilization of grass carp against overgrowing the reservoirs. In: Razvitie Prudovogo Rybovodstva i Rastitelnoje Osvoenie Vodoemov i Vodochranilisc. Moskva, Izd. WNIPRCh: 44-45. (In Russian.)

		Stock Density of Silver Carp (thous. indiv./ha)							
Food			968	1974					
		1.5	3	4	8	12			
Wet weight					<u> </u>				
Index	%000	180	202	171	229	283			
Average per fish	mg	1291	1707	2746	3653	3806			
Biomass of zoo- plankton and phytoplankton									
Index	%000	17	10	7	3	4			
Average per fish	mg	131	73	106	57	53			
Biomass of Phytoplankton									
Index	%000	4	6	5	3	3			
Average per fish	mg	26	49	85	53	48			

Table 1. Average amount of food found in the digestive tract of Silver Carp.

		Stock Density of Silver Carp (Thous. indiv./ha)		
		4	8	12
Average Fish Increment	in g/day (A)	1	0,7	0,5
Food Ration	in g/day (B)	16	22	23
K _{fc} (A) (B)	in %	6	3	2

Table 2. Index of food conversion efficience (K_{fc}) for Silver Carp in different stock densities.

1

!

ł

Index	Fish Species	Fry			2-year old fish	3-year old fish
	rish species	1964	1965	1966	1965	1966
	С	103,2	43,2	20,6	20,5	
Filling for animal food	Gc	38,8	9,2	5,5	1,8	1,0
	Sc	13,5	24,8	1,7	2,8	2,4
ow many times less than	Gc	2,6	4,7	3,7	11,4	
n Carp	Sc	7,6	1,7	12,1	7,3	
nimal food convergence	C and Gc	33,2	39,5	51,8	36,8	
	C and Sc	3,7	23,2	25,4	5,2	

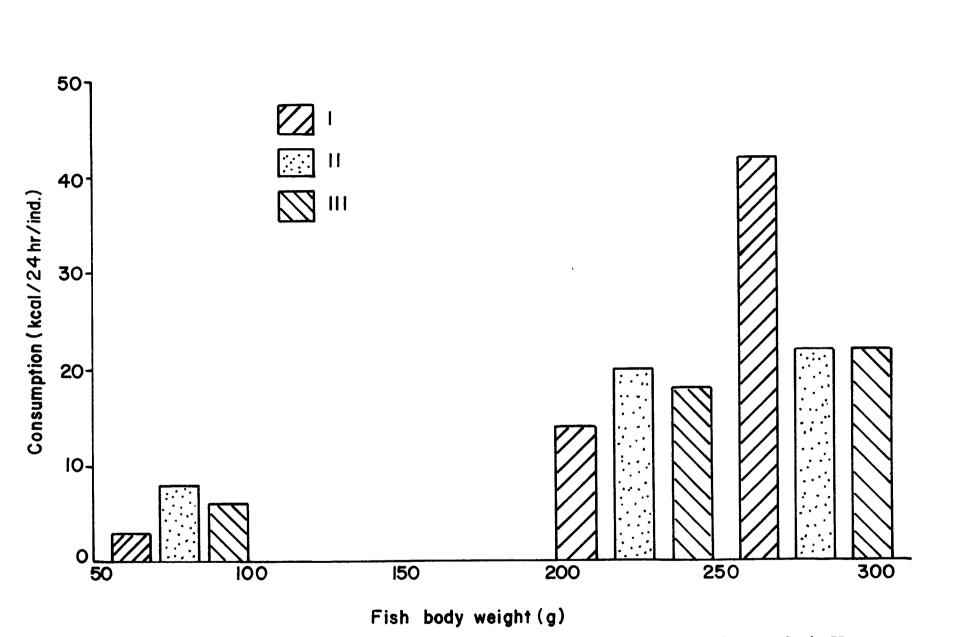
1 1

Table 3. Comparison of indices of filling for animal food and animal food convergence in Carp, Grass Carp and Silver Carp. (C = carp, Gc = Grass carp, Sc = Silver carp.)

Date	Phyto- plankton	Filamentous Algea	Rooted Plants	Sorghum	Zoo- plankton	Benthos
July 8, 1965						
July	42.4252 2010-1451 - 1455 - 145				Artunanyara England	
July 29, 1965			773 273 77111112			
July	र स्ट्रा इन्हरू इन्हरू					
30, 1965						
Aug.	19 20					
Sept. 3, 1965						
Sept. d	<u></u>					
Average för All Seasön	1050-2 ³	3	<u>27773</u>	51 51 		шо

carp silver carp silver carp

The percent food composition of two-year old carp, grass carp and silver carp. Figure 1.



ł

}

}

}

]

1 1

1

ł

1

}

ł

I

1

1

Figure 2. Dependence of consumption rate on the type of food supplied. I - excess of plant food, II - excess of plant + animal food, III - excess of animal food (Fischer and Lyakhnovich 1973).

1

}

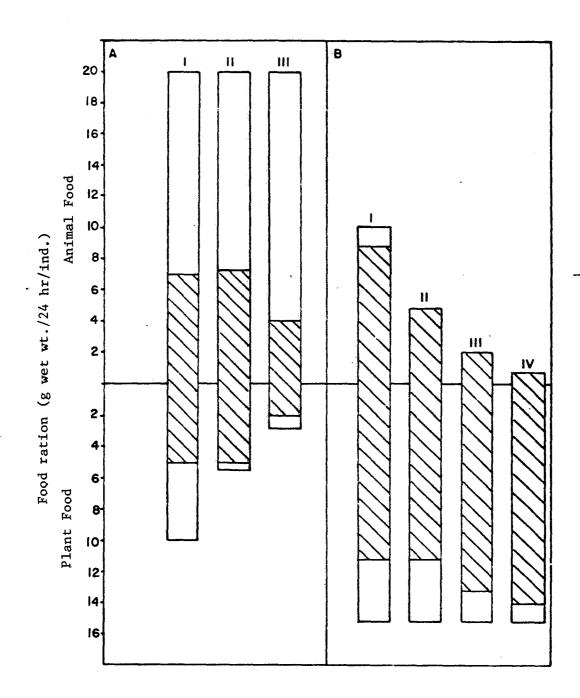


Figure 3. Dependence of consumption rate on quantity and quality of food supplied. 1-food consumed, 2-food remained, Aexcess of animal food, B-excess of plant food (Fischer 1973).

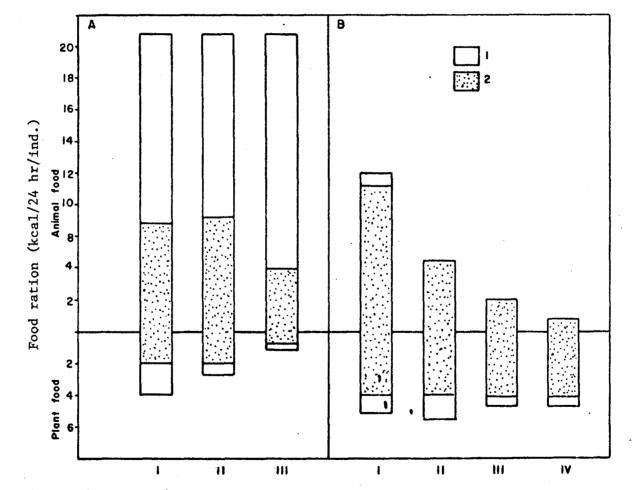
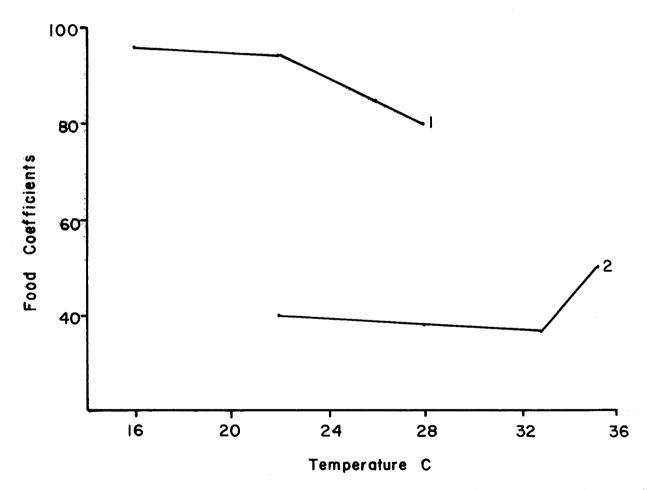


Figure 4. Dependence of consumption rate on quantity and quality of food supplied. 1-food remained, 2-food consumed, A-excess of animal food + diminishing ration of plant food, B-excess of plant food + diminishing ration of animal food (Fischer and Lyakhnovich 1973).



ł

1

1

ł

ł

1

}

Figure 5. Food coefficients for one- and two-year old fishes, <u>Ctenopharyngodon idella</u> Val., in various water temperatures: 1-one-year old fish, 2-two-year old fish, a-food coefficients.

1

)

ł

1

I

1

Ł

1

Data Date	Fitoplankton Phytoplankton	Glony nitkowate Algea	Makrofity Macrophyts	Pasza Food	Zooplankton	Bentos
2.VII.1966						2223
28.VII. 1966						
26.VIII.I966			11111		<u></u>	
				3	<u> </u>	8

)

arp [1] grass carp [1] silver carp

Figure 6. Percent composition of fry food in 1966; a - carp, b - grass carp, c - silver carp.

ł

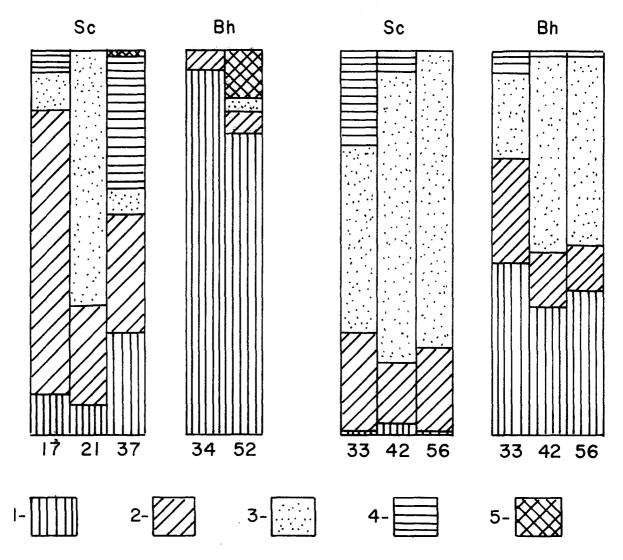


Figure 7. Percent food composition of the silver carp (Sc) and the bighead carp (Bh). Ponds number 17, 21, 37 - Sc in monoculture, 34, 53 - Bh in monoculture; 33, 42, 56 - Sc + Bh in polyculture; 1 - zooplankton, 2 - Chlorophyta, 3 - bacillariaphycene, 4 - Cyanophyta, 5 - Euglenophyta.

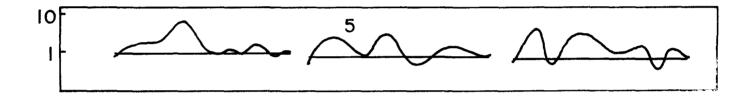


Figure 8. Influence of the silver carp on algae biomass in carp ponds during the season /IV - IX 1974/ The horizontal line - average biomass of algae in control groups of ponds stocked with carp alone; from left to right - the consecutive stock density of the silver carp; 4, 8 and 12 thousand ind./ha.

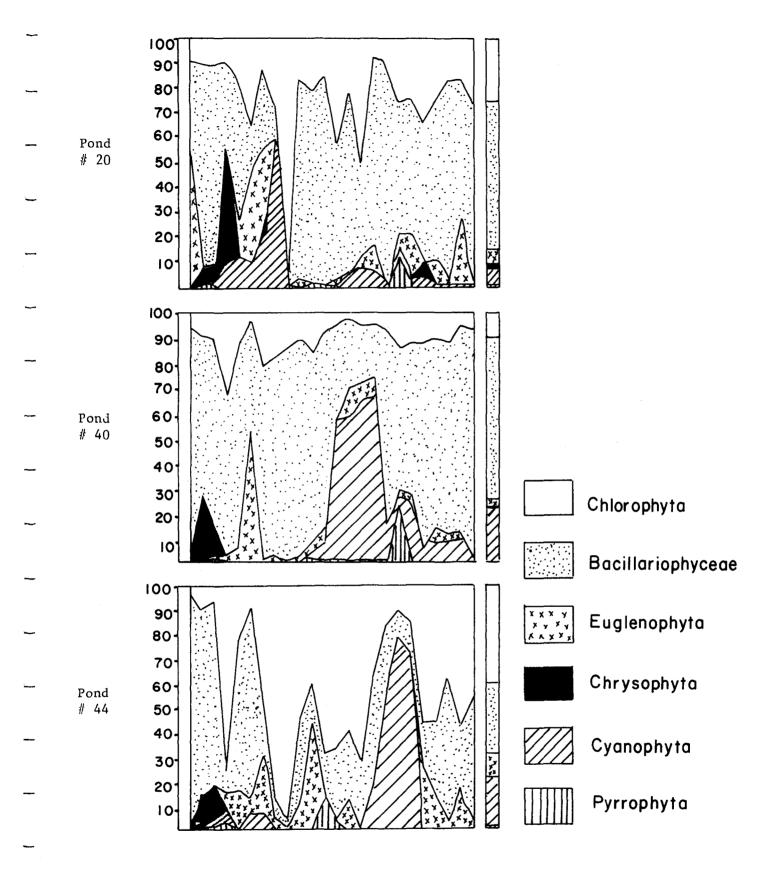


Figure 9. Influence of the bighead carp on the development of Cyanophyta. Ponds number: 20, 40, 44 - carp alone; 26, 32, 48 - carp + bighead carp.

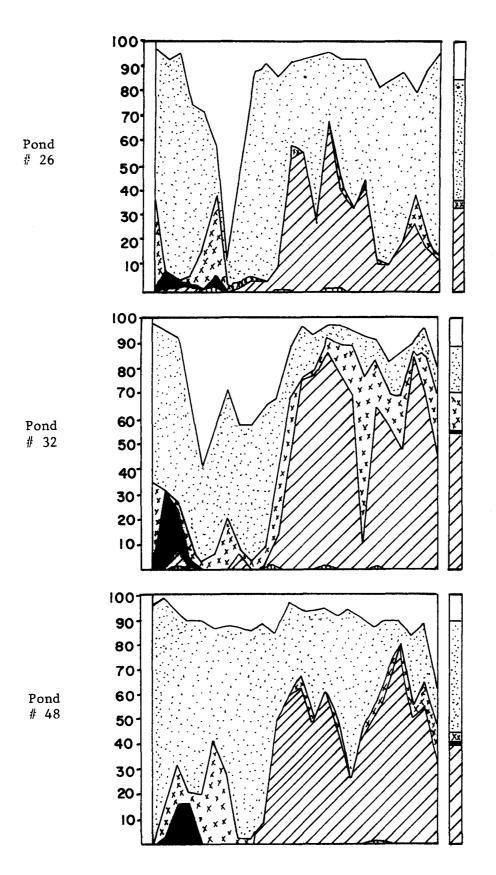


Figure 9. Continued

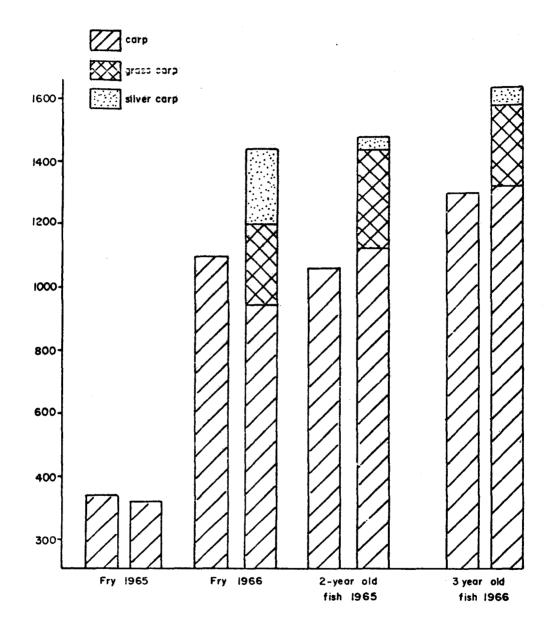


Figure 10. Yield per ha in mono- and polyculture. Each column represents the mean from three experimental ponds.

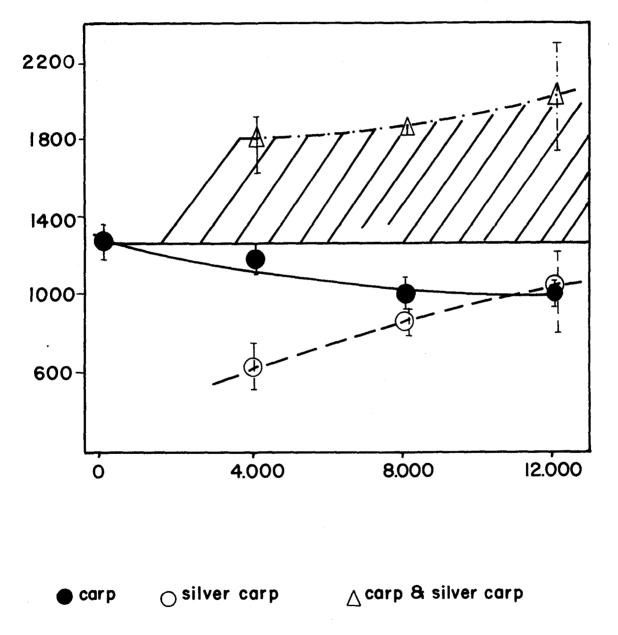
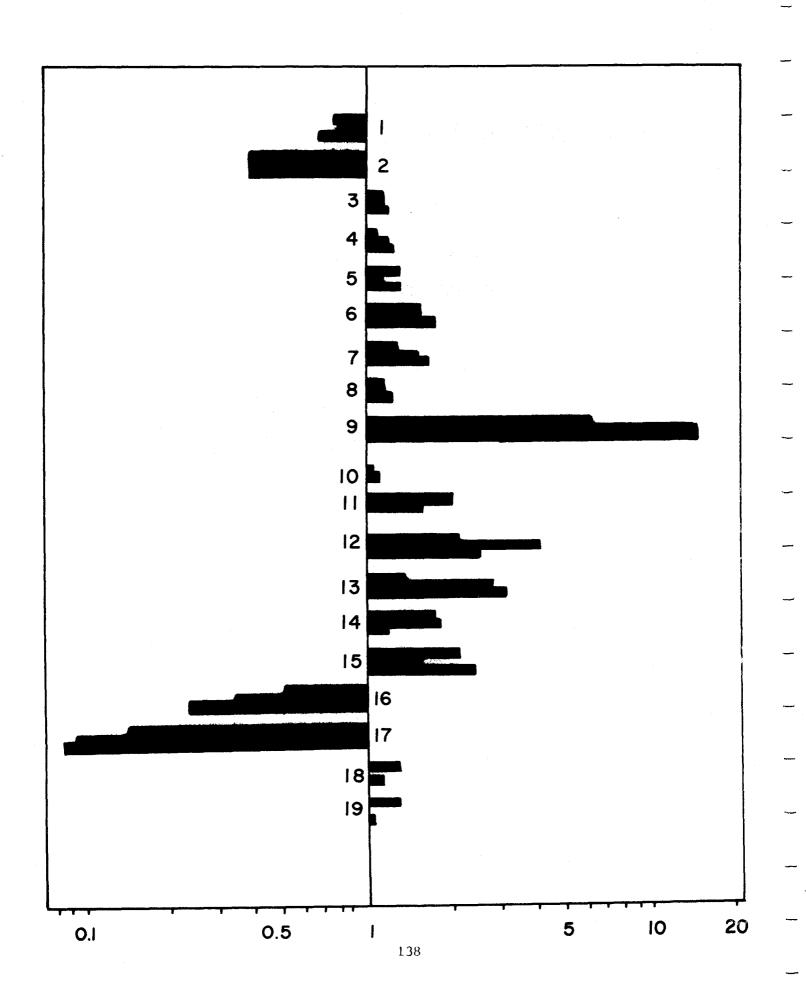


Figure 11. The carp production and total production under increasing stock density of the silver carp in 1974. Stock density of the silver carp/ind./ha/ stock density of the carp always the same.

Figure 12. The influence of various stocking densities with silver carp on average seasonal values of some environmental and biocenotic factors.

Logarithmic scale, 1 corresponds to the value of a given factor in the control group of ponds with carp only; the three bars correspond to the three consecutive increasing densities of silver carp; the highest bar shows the particular value at lowest density of silver carp; 1- ammonium nitrogen in the water, 2- phosphates in water, 3-BOD5, 4- soluble oxygen, 5- phytoplankton biomass, 6- chlorophyll, 7- gross primary production, 8- destruction, 9- net primary production, 10- total number of bacteria in water, 11- number of proteolytic bacteria in water, 12- number of ammonificating bacteria in water, 13- total number of bacteria in sediments, 14- number of proteolytic bacteria in sediments, 15- number of denitrifying bacteria in sediments, 16- zooplankton numbers, 17- zooplankton biomass, 18- benthos numbers, 19- benthos biomass.



THE ROLE OF HERBIVOROUS FISHES AT RECONSTRUCTION OF ICHTHYOFAUNA UNDER THE CONDITIONS OF ANTHROPOGENIC EVOLUTION OF WATERBODIES

B.V. Verigin

Laboratory of Ichthyology

Faculty of Biology, Moscow State University

INTRODUCTION

Man's economic activities cause great changes in the appearance of our planet. Inland waterbodies (freshwater) comprise a small portion of the land mass; however, large volumes of this water are used by many industries. Therefore, management of these waterbodies by man has influenced them greatly. Inland freshwater fisheries must be developed which will maintain waterbodies in their present condition and also take into account future anthropogenic effects.

Hydrotechnical construction causes hydrological changes which result in two opposite processes. The natural hydrological regime of rivers is changed as they are turned into reservoir systems, which cause lakes with slow flow. These reservoirs are created for energy, transportation and irrigation purposes and are rather different in their area, depth and other characteristics. Peculiar to our country is the presence of considerable numbers of large, rather shallow, reservoirs. They were created on the lowland rivers of the south slope (the Dnieper, the Don, the Volga) and occupy large areas in the most populated regions.

On the other hand, the number of artificial channels is increasing. They are created for irrigation, water supply, soil drying, transportation and other purposes. Among these channels there are some as large as the Karakum Channel, North-Crimea Channel, Channel Dnieper-Donbass, Dnieper-Krivoi Rog, Irtysh-Karaganda and others. There are also small channels such as these used for irrigation and collection of irrigation water from fields and drainage ditches are important and compose many hundreds of kilometers.

Water chemistry is changed in some degree by changing the hydrological regime. But in most cases, the change in water quality is caused by man's activity on the land that surrounds the waterbody. These changes in water chemistry are rather well known as water pollution results because of different poisons that have been washed from the fields by effluents from chemical and other plants. However, there are more complicated water chemistry changes that we cannot call "pollution". As a result of economic activity, a large amount of biogenes are also added to waterbodies. These are mainly compounds of nitrogen and phosphorus. These are sources of nutrients for aquatic plants which assimilate the suns energy and become the base for all other links to the aquatic ecosystem.

Enrichment by nutrients, as a result of man's activity, is called anthropogenic eutrophication. This process may be a positive one, within definite limits, as it increases biological and economic productivity of waterbodies, but when hypereutrophication results, this process becomes negative. The decomposition of huge masses of higher plants and algae causes the oxygen levels to sharply decline. Water becomes unusable for fish life and for drinking. This is termed secondary pollution.

The types of anthropogenic effects on waterbodies are variable in historical aspect. Originally the most salient factor was hydrotechnical construction. This developed from primitive irrigation systems and waterflows, in ancient times, to the present day constructions such as the Kara-Kum Channel and cascades on large rivers.

DISCUSSION

Thermal Effluents

During the last decade, simultaneously with population growth, industry growth and intensification of agriculture, excessive eutrophication has increased sharply. Together with hydrotechnical and chemical factors there is added a new strong anthropogenic factor - a thermal one. It results from the development of thermal and atomic energy. Only 30 - 35% of the energy contained in fuel converts into electroenergy. About 15% is lost in the air and 50 - 55% in the water. This results from vapor condensation as the water in condensors is warmed by 8 - 10 C. The warm water of atomic stations is 1.7 - 2.0 times greater than from thermal ones. Thus, the amount of warm water has increased greatly. It is known that in the near future, in the United States, 30% of the annual river flow will pass through condensors or 100% during the low-water period. The amount of water that presently flows to waterbodies will be enough to boil the cascade of waterbodies on one of our large rivers. The amount of heated water in both countries which waterbodies receive will increase several times.

Aquatic Vegetation

These anthropogenic factors cause ecosystems and the biological equilibrium to change. The principle effect of ecosystem disbalance is an excessive development of aquatic vegetation, submerged and partly submerged higher plants (macrophytes), and algae (microscopical unicellular and colonial algae). There is another after effect, but it is less important. Algae creates biological obstruction to water use in different branches of the economy. These obstructions often reach such proportions that water cannot be used normally and cause factories to shut down.

Overgrowth of shallow water by vegetation reduces water quality and also causes additional evaporation of valuable moisture, especially in southern irrigation channels. Overgrowth of channels at thermal stations reduces their ability to cool water, which provokes unnecessary use of fuel. The most serious damage results, however, from the overgrowth of artificial irrigation channels and collector-drainage system channels.

Excessive development of algae ("flowering") presents a serious problem for water supply and other water use. This is especially true as concerns waterbodies that are subjected to anthropogenic eutrophication. For the struggle against aquatic plants different mechanical and chemical methods are utilized, but these methods do not solve the problem because mechanical removal of vegetation is very unprofitable, and chemical methods lead to water pollution.

Herbivorous Fish

Recently, because of the development of methods for the artificial reproduction of the Far-East herbivorous fishes [grass carp, <u>Ctenopharyngodon</u> <u>idella</u> (Val.), silver carp, <u>Hypopthalmichthys molitrix</u> (Val.), and bighead, <u>Aristichthys nobilis</u> (Rich.)], we use biological methods for aquatic plant control. Great success has been obtained against aquatic macrophytes. Stocking of commercial, irrigation and technical waterbodies with grass carp allows us, in a short time, to get rid of soft submerged vegetation and rigid partly submerged ones. It is not my purpose to speak in detail of the use of the fish, but some ecological effects of these introductions will be mentioned. The primary effect of the removal of macrophytes in lakes and reservoirs is the improvement of conditions for zooplankton and fishes; zooplanktophage survival is increased.

Apprehensions have been expressed in the literature about the possibility of zoogenous succession and phytocenosis of high plants in waterbodies. However, we do not have exact observations of this phenomenon. One publication indicated that buttercup increased after introduction of grass carp. This is a littoral plant found in shallow areas inaccessible to grass carp. Charophyta also develop in ponds where grass carp are introduced. Young grass carp do not control Charophyta, but large fishes do not allow it to develop. When it became evident that grass carp did not eat water ladies thumb (<u>Polygonum amphibium</u>), we feared that waterbodies would be overgrown with it. But we soon realized that water ladies thumb did not appear in ponds inhabited by grass carp because the latter destroyed its sprouts.

The destruction of vegetation also effect animal populations inhabiting a waterbody. The quantity of blood-sucking mosquitoes is reduced as the larvae are deprived of shelter. The number of small trash fishes inhabiting vegetation is also decreased. There is apprehension that destruction of plants damaged the reproduction of fishes spawning there, but for the present these apprehensions are not confirmed. Introduction of grass carp into the Khauzkhan waterbody, in Turkmenia, did not reduce catches of native fishes. The reason being that some balance was maintained. Although spawning places were reduced, coarse fishes did not eat eggs and larvae in as great amounts. There may be other reasons, but the above mentioned one does not mean that the reduction of vegetation does not effect the ichthyocenosis. The use of grass carp must take place under those conditions where we can regulate its abundance and in such waterbodies where the full reconstruction and cultivation of ichthyofauna can be provided.

Anthropogenic Eutrophication

The struggle against microscopical algae that causes the "flowering" of water is more complicated. This problem is closely connected with anthropogenic eutrophication. I have mentioned this phenomenon before but I will examine it in more detail. In the recent literature I have not succeeded in finding a strict definition of this concept. However, as to the cause of this phenomenon, its effect, and the role of different factors, the definitions of numerous scientists are practically the same. Summing them, the modern definition of "anthropogenic eutrophication" must be formulated as the increased trophic conditions in the waterbody which are under the effect of enrichment by chemical biogens resulting from man's economic activity on the surrounding area which changes water quality for the process of economic water use.

Examining this phenomenon, it becomes clear that additional application of biogens intendifies biological processes in waterbodies, which leads to the growth of masses of living matter in the ecosystem. Increasing production which increases the ecosystem mass is characterized by the rate of anthropogenic eutrophication.

We know that the rate of biological productivity is determined not only by the mass of the ecosystem but also by the turnover rate. The same as a percentage of the capital, the productivity is determined by the size of "capital" and the rate of turnover. The rate of biological cycles in aquatic ecosystems depends initially on the temperature regime. Until recently this rate has been determined by the peculiarities of climate and is a natural factor, therefore, it has not been necessary to take temperature into account when considering the phenomenon of anthropogenic eutrophication (Figure 1).

As mentioned before, in the near future and especially in perspective, with the development of thermal and atomic energy, the amount of additional heat will grow and hence the rate of biological cycles in ecosystems will increase. Therefore, the anthropogenic effect on the productivity of waterbodies will consist of two processes: 1) increase in productivity at the expense of an increase in ecosystem mass, and 2) an increase in productivity at the expense of the acceleration of biological turnover of this mass. Under these new conditions, it is logical to modify the concept of "anthropogenic eutrophication". It can be considered as two processes, namely, the entrance of additional heat to the system which I term thermal eutrophication, which increases biological turnover in ecosystems. The derivative of these processes will be the phenomenon of anthropogenic eutrophication.

Anthropogenic eutrophication can more strictly be defined as the biological productivity of aquatic ecosystems are increased by the effect of man's economic activities on the surrounding watershed and results in degradation of water quality and makes the water uneconomical for use.

The reverse of this is the phenomenon of anthropogenic pollution which is defined as <u>deterioration of water quality at the place of habitation and</u> <u>the object of economical use</u>. It can be primarily chemical and primarily thermal as well as secondary or eutrophicated. Secondary pollution appears as a result of anthropogenic eutrophication, (chemical or thermal) which leads to unnecessary production of organic matter by autotrophic plants. Unbalanced ecosystems arise in which the primary link is hypertrophicated and the productivity of the next link falls behind.

Ichthyofauna Reconstruction

The task of creating a balanced ecosystem can be solved by reconstruction of its ichthyofauna. The native ichthyofauna of our eutrophicated waterbodies consists primarily of relatively cold resistant non-productive fish species that are the producers of the third and fourth order, i.e., the zoophages or predators. These species must be replenished or replaced by highly productive warm-water fish species that are the producers of the second order, i.e., phytophages. The most important second order species are those which use phytoplankton and detritus as food.

Eutrophication resulting from chemical and thermal introductions into aquatic ecosystems are currently considered to be a negative anthropogenic effect which decreases the productivity and water quality of these systems. Ichthyofauna reconstruction can turn these negative effects into positive ones that increase not only economic productivity, but also water quality.

The negative effect of chemical eutrophication is evident in our large waterbodies or the south slope rivers: the Dnieper, the Don, the Volga. Therefore, a mass introduction of silver carp was planned and a rather small introduction has been made. There herbivorous fishes can take the lead role in the fisheries of these waterbodies. They grow rapidly, adding a kilogram or more a year, which is much more than any of the native fishes. Only in relatively small waterbodies, where on a unit of area a rather large sample was introduced, has there been large fishery effect. The fish productivity of these waterbodies has increased from some kg/hectare to 2-3 centners/ But it is not quite clear how silver and bighead carp can restrain the ha. production of algae and improve water quality. The problem arises as a result of eutrophication. Blue-green algae develop which comprises 30% of the phytoplankton production. They are considered as toxic and inedible plants. Thus, some question arises as to whether these fishes can effect the development of blue-greens. Food habit studies indicate that both species avoid blue-green algae.

Only precisely conducted observations can determine the capability of the silver and bighead carp in controlling different taxons of algae. Their food preference is affected by the size of the algae, as they prefer large cells and colonies. These data are interesting and indicate that they do not simply remove a definite biomass but they repress algae reproduction.

It was shown experimentally that silver and bighead carp assimilate bluegreen algae as they do all other groups, however, the theoretical and experimental principles of the quantitative estimation of the ecological role of phytoplanktophagous fishes has not been studied sufficiently. These investigations will be continued.

I would like to reiterate that the use of herbivorous fishes does not supress eutrophication, but the direction of the productive processes and the form of biological production are changed. Instead of useless algae, we obtain important fish production, and rationally utilize anthropogenic eutrocation. The same argument can be presented for thermal eutrophication. In the future, warm water from electropower stations will be used for fisheries purposes. Fish for stocking purposes will be reared in heated ponds using artificial food. Research indicates that introduction of Far-East herbivorous fishes to these waterbodies gave good results.

Heated water leads to a sharp increase in growth rate, and rapid maturity. Mass introduction of these fishes to cooling pools increases their productivity to 2-4 centner/hectare. There is also a positive result after introduction of African herbivorous tilapia to cooling pools. Undoubtedly the Indian gigant carps and some other phytophagous fishes of the south-eastern part of Asia might also grow well in these conditions.

The introduction of herbivorous fishes is only the first step toward ichthyofauna reconstruction. A third trophic link which includes warm water zoophages (buffalo, catfishes and some other fishes of the United States) could also be added.

Polyculture is intensively developed in pond fisheries. In the future we can consider reconstructing entire cooling pool ecosystems by utilizing more thermophilous organisms. By experimentation and study of these relatively small cooling pools we can obtain information that will allow for ichthyofauna reconstruction in larger waterbodies where electropower plants will be constructed.

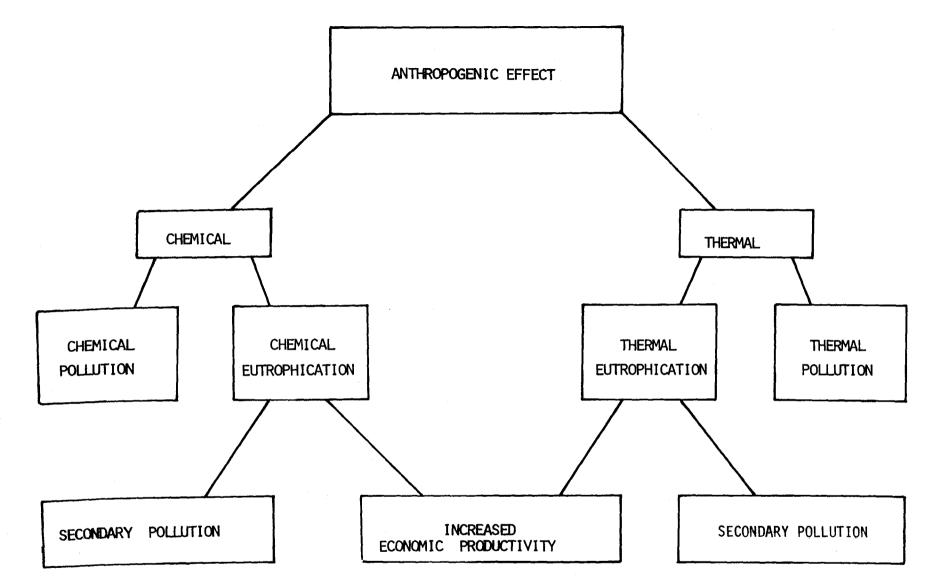


Figure 1. Anthropogenic effects and how they relate to water quality.

}

GRASS CARP RESEARCH AND PUBLIC POLICY IN ENGLAND

Brian Stott

Salmon and Freshwater Fisheries Laboratory Ministry of Agriculture, Fisheries and Food Whitehall Place, London, SW1a 2HH, England

INTRODUCTION

Clearly, this paper will deal with United Kingdon experience only, for while some of the discussion and controversy which has surrounded the grass carp in other countries is apparent, firsthand knowledge of their situations is obviously unavailable. However, from what has been published the situation in the United States and indeed other countries, has been rather similar to that in the UK; grass carp have had their protagonists, more or less cautions, and their antagonists. Some of the latter have been very outspoken, which is not necessarily a bad thing, but occasionally their case has been served badly by wild overstatements and sometimes sheer mis statements. A classic example was a popular article attacking grass carp as an undesirable exotic but the characteristics of the fish described were obviously those of the common carp (Cyprinus carpio L.).

EXOTIC SPECIES

Being islands, the UK has a comparatively limited fauna, including freshwater fish. Generally, also, there is public recognition that exotics can be undesirable or even dangerous; the risk of rabies reaching England from continental Europe via smuggled dogs and cats has received wide publicity. Perhaps stemming from this awareness there has been little public pressure in favor of introducing exotic freshwater fish during the last two decades or so. Occasionally the angling press will carry an article or print a letter in favor of black bass (<u>Micropterus salmoides</u> Lacepede) but these raise little support. Also, there have been well publicized problems with exotic animals: the coypu (<u>Myocastor coypus Kerr</u>), the mink (<u>Mustella vison</u> L.) and the grey squirrel (<u>Sciurus carolinensis Gmelin</u>).

The only exotic freshwater fish which has been successfully introduced into the UK in recent times has been the pike-perch, or zander, (Stizostedion <u>lucioperca</u> L.). This was imported from Germany by the Duke of Bedfore in 1878 (Buckland 1883), but it remained localized until 1963 when it was introduced into the Great Ouse system in East Anglia. From here its spread has been rapid, but the effect on fisheries is difficult to determine; as always the decision between "good" and "bad" in freshwater fisheries involves a value judgment in which objective standards are impossible to apply. It appears that the effect zander have had in England "depends on whether or not you like them" (Dr. R.S.J. Linfield, Regional Biologist, Anglian Water Authority, Personal Communication).

RESEARCH PROGRAMS AND POLICY

It was against this generally conservative background that grass carp (<u>Ctenopharyngodon idella</u> Val.) were imported for experimental work in the UK. In about 1963 the then Chief Officer of Salmon and Freshwater Fisheries, the late F.T.K. Pentelow, was persuaded by the late C.F. Hickling that grass carp might be useful in helping with aquatic weed control. At that time, and it is the same today, the only legislation controlling the importation of freshwater fish was the Diseases of Fish Act, 1937, under which the Ministry of Agriculture, Fisheries and Food (MAFF) could grant licenses. Initially, a license was granted to the Central Electricity Generating Board to import some 15,000 fingerling grass carp from the Far East, which were stocked into the cooling lagoon of a thermal power station on the north-west coast of England where weeds were impeding the circulation of water. Unfortunately, the experiment was not successful for the fish did not survive. The reason was thought possible due to predation by the resident brown trout population of the effect of brackish water, or perhaps both.

Some fish from the consignment, however, were kept in the laboratory where they grew and were eventually given to MAFF. These were the fish wer used in our first experiment in a pond in southeast England (Pentelow and Stott 1965). This trial had a very simple objective; to determine if the fish would in fact eat weeds under our climatic conditions and it was quite successful. Great care was taken in the selection of the site, the pond was spring-fed and the stream flowing from it discharged directly into an estuary where the water quality was so poor as to make the survival of any escapees very doubtful. Naturally, the outflow was screened. The pond was on private ground, well concealed and overlooked by a private house whose occupants, when told of the experiment, readily agreed to keep an eye open for unauthorized intruders.

Following this experiment, a limited number of fingerlings were imported by MAFF in 1965 for more trials. These came in by air from the Far East, but great difficulties were experienced in keeping the fish alive. Several consignments were received but they were invariably in poor condition on arrival and often infected with Ichthyophthirius multifiliis and Gyrodactylus sp. Consequently it was only when fish became more readily available in Europe (Krupauer 1971) that a sufficient stock was obtained enabling more work to be carried out. Two thousand one- and two-summer fish were brought in by road from Hungary in 1976 but these were more than could be accommodated in the Ministry's London laboratory and it was necessary to place the fish in water which were not directly under MAFF supervision. Luckily England is well provided with "stately homes", large country houses situated in private parkland and almost invariably with lakes well supervised by resident staff. These have been very useful for grass carp work. Part of the first Hungarian consignment was stocked in a pond at Woburn Abbey with the interested consent of His Grace the Duke of Bedford, the great grandson of the Duke who introduced zander years earlier.

These fish were subsequently used in the first real experiments which were to examine the relationship between stocking rates and degree of weed control obtained (Stott and Robson 1970). From this point it became apparent that the fish might have a real role to play on controlling aquatic weeds in the UK and this related directly to MAFF's responsibility for and interest in land drainage of agricultural areas throughout the country – an extremely important function in relation to our production. Although the grass carp work remained under the control of MAFF, and was carried out by it, Salmon and Freshwater Fisheries Laboratory, the then recently formed Aquatic Weed Section of the Agricultural Research Council's Weed Research Organization became involved in collaborative research and, of course, MAFF's Land Drainage Division was kept informed of progress.

Although, as explained above, the importation of freshwater fish could be controlled by MAFF only under the Diseases of Fish Act, 1937, the river authorities (local government bodies controlling fisheries, and responsible for pollution control), local land drainage and other water functions (in river basin areas) had received powers under the Salmon and Freshwater Fisheries Act, 1972, enabling them to prohibit the introduction of fish into their areas for whatever reason. By this time, through publications and papers presented at scientific meetings whether published (Stott 1967) or not, the work on "the grass-eating carp" was becoming widely known. The popular media, newspapers, radio and television, all seemed to be quite fascinated by the idea behind the work and considerable interest was aroused. This was undoubtedly welcome, but it led to scores of letters sometimes wanting more information, which was gratifying, but more often making requests for supplies of fish which could not be met. Neither MAFF nor the local river authorities wanted the fish to become widespread. We had to explain to the enquirers that MAFF was discouraging the spread of grass carp until our investigations were completed. This was invariably accepted as logical and proper; indeed the appreciation by the public of the need for a thorough examination of the potential of grass carp and the ecological effects of its grazing has been a great help to the Laboratory.

From the scientific viewpoint, however, it was by no means certain how thorough an ecological study could be made with the limited resources available, but it seemed sensible to concentrate, at least initially, on the effect grass carp grazing might have on indigenous fish species. The really big issue, the possibility that the fish might breed in our water, and spread to become a nuisance which would be impossible to eradicate and expensive to control was, of course, not open to experimentation but by the early '70's from the increasing amount of published work on the fish, particularly in the USSR and Eastern Europe, it was becoming clear that breeding did not readily occur outside the native range and this was very interesting from our point of view. Remarks such as artificial reproduction being "the main and decisive link in their economic development and exploitation" (Vinogradov et al. 1965), were encouraging.

The progress of our work carried out on the effect grass carp grazing might have on other fish species was rather disappointingly slow. Suitable sites, having an adequate population and variety of indigenous species, were difficult to find, for at that time security both from point of grass carp escaping and human interference was given high priority. The earlier work on stocking rates had been done in ponds surrounded by a specially erected barbed wire fence as very little was known about the fish. Some work was, however, successfully completed and bream (<u>Abramis brama</u> L.) were shown to grow better in the presence of grass carp than by themselves and survive no worse. There were reports in the literature of similar results with several species (Opuszynski 1968, 1971). Work at sites using fish other than bream was less successful; a series of divided pond experiments taught us little except that grass carp could and would easily leap over the nets we had used to keep populations apart.

FOOD PREFERENCE

Emerging from earlier work on weed control an interest was developed on the food preferences of the fish and this was continued at the Weed Research Organization. There was a certain amount of information available in the literature (Stroganov 1963; Verigin et al. 1963; Zolatova 1966; Krupauer 1967; Fischer 1968; Edwards 1974) and it seemed clear that the fish, particularly when small and at lower ambient temperatures, preferred soft vegetation to hard fibrous plants and that some plants seemed to be avoided. Our own work (Stott and Robson, unpublished data) confirmed this. Thus it was possible that the usefulness of grass carp might have practical limitations and that through selective grazing the fish might actually increase weed biomass in a mixed stand of weeds by removing the competition of the preferred species and thus allowing other plants to increase. This liklihood would be greater at the lower stocking rates designed to reduce the plant biomass to, say, 25-50% of its potential. Indeed it has recently been suggested that aiming to stock to achieve a 50% reduction of plant growth potential may be very difficult or even impossible (Fowler and Robson, in preparation).

REPRODUCTION PROBLEMS

In addition to fears that weed control by grass carp might be very difficult to manage, the problem of the fish reproducing naturally still caused concern. Experiments were reported on monosexual culture (Stanley et al. 1975; Stanley 1976a) and this could be a possible approach. However, with more publications giving details on natural breeding outside the normal range of the fish (Inaba et al. 1957; Aliev 1965; Motendov 1969; Vinogradov and Zolotova 1974; Mitrofanov 1975) it began to seem less and less probable that the grass carp would spawn naturally in the UK. Certainly water temperature in some rivers (e.g. R. Trent) receiving a number of heated effluents from power stations might occasionally become high enough, but water velocities are too low to keep the pelagic eggs in suspension for a distance of the order of 200 km which is apparently desirable (Stanley 1976b) and sudden rises in water level, which are apparently necessary to initiate spawning, could only be caused by rain which inevitably decreases water temperature. The rather outside chance that power station cooling water discharges might themselves be suitable spawning sites in other, more swiftly flowing rivers was discounted when it was shown that at temperatures below 20 C the eggs either failed to hatch or, if the did hatch, the embryos were deformed and non viable (Stott and Cross 1973).

WEED CONTROL

Another aspect of the work in the UK needs mentioning. It was never assumed that the grass carp would be the complete answer to our weed control problems and conventional methods would still be needed in some circumstances even if biological control became a practicality. Weed cutting would barely affect the fish but clearly the tolerance of the fish to herbicides was

important. The fish is quite tolerant and Tooby et al. (in preparation) have shown that it is in no danger from the herbicides cleared for use in the UK when used at the recommended dosages (Table 1) although its appetite may be temporarily reduced (Table 2) at some concentrations.

CONTROL COSTS

Despite the potential value of grass carp for weed control its usefulness would be limited if it proved to be very expensive and this aspect has been given some attention. For UK conditions it has been assumed that a 50% reduction in weed growth potential is acceptable and can be achieved and maintained by a fish density of between 125-200 kg/ha. From data on fish growth and an assumed constant annual mortality of 50%, the changes in the biomass of a cohort of 12,500 one-summer grass carp, mean weight 10 g, is such that (assuming no limitation due to food shortage, etc.) an initial stocking of 125 kg will control 17 hectares/year (Table 3). Currently 125 kg of one-summer fish imported from Yugoslavia (the only country able to satisfy the recently imposed UK health certificate requirements) cost 1,750 (\$3150), making the cost of control 103 (\$185) per hectare per year excluding labor. This sum compares very favorably with the aquatic herbicides cleared for use in water in the UK but it is more than the cost of cutting (Table 4). The grass carp costs in this model are believed to be overestimated because the growth data were taken from fish in suboptimal conditions and also large grass carp may well exhibit lower mortality rates than the one assumed. In addition, almost half the cost of the fish was due to transportation and associated costs which could be reduced significantly if the fish were to be artificially bred in the UK. The model is a very simple one, however, and labor charges need to be incorporated in the estimates, nevertheless the indications are that biological control could be economically attractive.

RESEARCH POLICY

It is clear that there is a limit to the amount of useful information which can be gained from the small-scale investigations which have so far been carried out in the UK and, in view of the possible benefits which biological control might bring to weed control, it has been recommended that the grass carp trials should be increased and extended (Stott 1977). A steering committee has recently been established by MAFF through which water authorities (the successors to the river authorities mentioned above) are cooperating with MAFF and the Weed Research Organization in carrying out larger field trials to provide more information and experience with the fish. A shortage of grass carp is holding up the rapid implementation of these plans and a source of new supplies which can meet our health requirements is being sought. It is realized that larger trials hold some risks, for well-meaning but ill-advised anglers may try to take fish and put them in open waters but the chances of damage are not great if breeding does not take place. The next few years are going to be very interesting.

SUMMARY

The current position with regard to grass carp in the UK can be summarized:

- 1. The fish seems to be potentially useful for water weed control and could be used in conjunction with the aquatic herbicides currently used.
- 2. The ecological impact of the fish is poorly understood, but since the chances of natural breeding taking place is believed to be low, the risk of permanent damage is virtually nil.
- 3. The economics look promising but better data on natural growth and mortality rates are needed.
- 4. More information is needed on the food preferneces of the fish under natural conditions.

REFERENCES

- Aliev, D.S. 1965. The breeding of the white amur (<u>Ctenopharyngodon idella</u> Val.), silver carp (<u>Hypopthalmichthys molitrix</u> Val.), and bighead (<u>Aristhichthys noblis</u> Rich.) in the Amu-Darya Basin. Vop. Ikhtiol. 5:593-599. (In Russian.)
- Buckland, F. 1983. Natural history of British fishes. Society for Promoting Christian Knowledge. London. 420 pp.
- Edwards, D.J. 1974. Weed preference and growth of young grass carp in New Zealand. N.Z.J. Mar. Freshwater Res. 8:341-350.
- Fischer, Z. 1968. Food selection in grass carp (<u>Ctenopharyngodon idella</u> Val.) under experimental conditions. Pol. Arch. Hydrobiol. 15:1-8.
- Fowler, M.C. and T.O. Robson. In Preparation. The effects of the food preferences and stocking rates of grass carp (<u>Ctenopharyngodon idella</u> Val.) on mixed committees.
 - Inaba, D., M. Nomura and M. Nakamura. 1957. Preliminary report on the spawning of grass carp and silver carp in the Tone River, Japan and the deposition of their eggs. J. Tokyo Univ. Fish. 43:81-96.
- X Krupauer, V. 1967. Food selection of two-year old grass carp. Bull. V.U.R. Bodnany. 1:1-11. (In Czech.)
 - 1971. The use of herbivorous fishes for ameliorative purposes in Central and Eastern Europe. Proc. Eur. Weed Res. Coun. 3rd Int. Symp. Aquatic Weeds. 95-103.
 - Lucey, J., T.E. Tooby, M. Lee and C. Allchin. In Preparation. The effect of sub-lethal concentration of aquatic herbicides on the consumption of Elodea by grass carp, (Ctenopharyngodon idella Val.)
 - Mitrofanov, V.P. 1975. On the natural reproduction of grass carp (<u>Ctenophar</u>yngodon idella Val.) in the Ili River. Vopr. Ikhtiol. 15:1039-1045.
 - Motenkov, Y.M. 1969. The results of settling phytophagous fish in natural waters in Krasnodar Krai. Sb. nauch-Tekh. Inf. Krasnodar Fil Vses Naucho-Issled Inst. Prud. rub. Khoz. 1:59-61. (In Russian.)
 - Opuszynski, K. 1968. Carp polyculture with plant feeding fish: grass carp <u>Ctenopharyngodon idella</u> (Val.) and silver carp <u>Hypopthalmichthys moli-</u> <u>trix</u> (Val.). Bull. Acad. pol. XVI, 11:677-681.
 - 1971. Present state and prospective in culture of phytophagous fish in Europe. Proc. Symp.: New ways of freshwater fisheries intensification. Fish. Res. Inst. Vodnany.

Pentelow, F.T.K. and B. Stott. 1965. Grass carp for weed control. Prog. Fish-Cult. 27:210. Stanley, J.G. 1976a. Female homogamety in grass carp (<u>Ctenopharyngodon</u> <u>idella</u> Val.) determined by gynogenesis. J. Fish. Res. Bd. Canada. <u>33:1372-1374</u>.

1976b. Reproduction of the grass carp (<u>Ctenopharyngodon</u> idella Val.) outside its native range. Fisheries. 1:7-10.

- Stanley, J.G., J.M. Martin and J.B. Jones. 1975. Gynogenesis as a possible method for producing monosex grass carp (<u>Ctenopharyngodon idella</u> Val.). Prog. Fish-Cult. 37:25-26.
- Stott, B. 1967. Aquatic weed control by grass carp (<u>Ctenopharyngodon idel</u>la Val.). Proc. 3rd Br. Coarse Fish Conf., Liverpool Univ. 62-65 pp.
- 1977. On the question of the introduction of the grass carp (<u>Ctenopharyngodon</u> idella Val.) into the United Kingdom. Fish. Mgmt. 8:63-71.
- Stott, B. and D.G. Cross. 1973. A note on the effect of lowered temperatures on the survival of eggs and fry of the grass carp <u>Ctenopharyngodon idella</u> (Valenciennes). J. Fish. Biol. 5:649-658.
- Stott, B. and T.O. Robson. 1970. Efficiency of grass carp (<u>Ctenopharyngodon</u> idella Val.) in controlling submerged water weeds. Nature (Lond.) 226: 870.
- Stroganov, N.S. 1963. Food selection in grass carp. In: Problems of the Fisheries Exploitation of Plant-eating Fishes in the U.S.S.R. Ashkhabad. 181-191. (In Russian.)
- Tooby, T.E., J. Lucey and B. Stott. In Preparation. The tolerance of grass carp (<u>Ctenopharyngodon idella</u> Val.) to aquatic herbicides with a view to this fish being used in integrated aquatic weed control.
- Verigin, B.V., N. Nguen and D. Nugen. 1963. Data on the food selectivity and the daily ration of the white amur. In: Problems of the Fisheries Exploitation of Plant-eating Fishes in the U.S.S.R. Ashkhabad. 192-194. (In Russian.)
- Vinogradov, V.K., L.V. Erokhina, A.G. Konradt and G.I. Savin. 1965. Biotechnical aspects of the culture of phytophagous fishes. Proc. All-union Scient. Res. Inst. Pond Fish Cult. 13:3-9. (In Russian.)
- Vinogradov, V.K. and Z.K. Zolatova. 1974. The influence of grass carp on aquatic ecosystems. Gidrobiol. Zh. 10:72-84.
- Zolatova, Z.K. 1966. On the question of selectivity in the feeding of the white amur carp (<u>Ctenopharyngodon idella</u> Val.). Proc. All-union Sci. Res. Inst. Pond Fish Husbandry XIV:39-49. (In Russian.)

Active Ingredient	Herbicide Trade Name	% Composition	96 hr. L.C.50 (mg/1)	Max. Concn. of active in- gredient (or formulation) in water 1 m deep if recom- mended rate is applied to surface (mg/1).
Dalapon		85	30,000	3.0 (3.5)
2,4-D Amine Salt		50	1,350	0.4 (0.8)
Diquat	Aquacide	13.5	1,400	2.0 (14.8)
Terbutryne	Clarosan	100 tech.	6.2	0.1
Glyphosate	Roundup	36	14	0.8 (0.5)
Dichlobenil	Casoron	100 tech.	9.5	1.0

I

Table 1. Toxicity, 96 hr. L.C.₅₀ values, for various aquatic herbicides to grass $carp^{1/}$.

 $\frac{1}{2}$ Data from: Tooby, T.E. et al. (in preparation).

1

ſ

Herbicide	Recommended dosage (active ingredient) (mg/1)	Reduced feeding observed at; (mg/1)	Food intake depression
Dichlobenil	1.0	1.6	80
Diquat	2.0	18.2	70
Cyanatryn	0.2	1.75	70

Table 2. Reduction in amount of <u>Elodea</u> canadensis eaten by grass carp in water containing aquatic herbicides.

1/ Data from: Lucey, J. et al. (in preparation).

Time	Age of Fish	No.	Mean wt. (g)	Biomass (kg)	Biomass + 4
lst Spring	0+	12,500	10	125	
2nd Spring	1+	6,250	78	488	122
3rd Spring	2+	3,125	230	719	180
4th Spring	3+	1,563	470	735	184
5th Spring	4+	781	820	640	160
6th Spring	5+	391	1,300	508	127
7th Spring	6+	195	1,800	351	88
8th Spring	7+	98	2,300	225	56

Table 3. Change in biomass of a cohort of 12,500 one-summer fish in a one hectare water body, assuming no limitation on growth.

Herbicide (mg/1)	Cost \$	
"Clarosan" (1% terbu tryne)		
Recommended dose 0.05	229 - 257	
Max. permitted dose 0.01	445 - 502	
"Casoron GSR" (22% dichlobenil)		
Recommended dose 1.0	504	
"Aquacide" (2,4-D Amine salt)		
Recommended dose 1.0	310 - 324	
Max. permitted dose 2.0	581 - 610	
Manufacturer's max recommended dose 8.0	2,212 - 2,324	
Mechanical cutting including weed removal	131	

Table 4. Costs, including labor, of an annual herbicided treatment, and biannual mechanical cutting, of aquatic weeds in a standard channel 1 km long, 10 m wide and 1 m deep.

THE USE OF GRASS CARP FOR BIOCONTROL OF AQUATIC WEEDS

AND THEIR IMPLICATION FOR $\frac{1}{2}$ NATURAL RESOURCES AND

FISHERIES IN FLORIDA

Woodward W. Miley II, Jess M. Van Dyke and Dennis M. Riley

Biologist III Florida Department of Natural Resources

Biologist II Florida Department of Natural Resources

Biologist III Florida Department of Natural Resources

ABSTRACT

The results of two studies to evaluate the ecological impact of grass carp (<u>Ctenopharyngodon idella</u> Val.) in Florida are discussed. Although one of the studies is still in progress, the other has been completed. After one year's background data had been collected, four ponds ranging from 2.03 to 12.15 ha were stocked with grass carp at a rate of 67 kg/ha and monitored for an additional two years. Changes in fish populations are discussed. Adverse effects were noted in two ponds, but it is concluded that the grass carp was not directly responsible. It is recommended, however, that grass carp not be stocked in ponds with sparse vegetation. One of the ponds with a heavy initial infestation of the plant <u>Hydrilla verticillata</u> Royle supported an extremely healthy native fish population at the end of the study in spite of the presence of a huge biomass of grass carp (511 kg/ha).

The study in progress is being conducted on four lakes ranging from 36 to 2,024 ha. In this study, the grass carp has demonstrated the ability to completely control hydrilla with a stocking rate of 49 fish per ha. It is postulated that the effects of the grass carp on water quality are much less than would be expected by other forms of aquatic weed control. In a 2,024 ha reservoir, the grass carp has not controlled Eurasian watermilfoil (Myrio-phyllum spicatum).

INTRODUCTION

Any discussion of grass carp (<u>Ctenopharyngodon idella</u> Val.) in Florida should be put into perspective by preceding it with a short discussion of hydrilla (<u>Hydrilla verticillata</u> Royle). Florida received the South African aquatic plant through the aquarium industry in about 1960. By 1965 hydrilla was established in approximately 4,049 ha of Florida waters (Figure 1). In 1970 this aquatic macrophyte was reported in about 20,243 ha (Figure 2) and

Cooperative study by the Department of Natural Resources Bureau of Aquatic Plant Research and Control in Tallahassee, the University of Florida in Fort Lauderdale and Gainesville, the Florida Game and Fresh Water Fish Commission in Tallahassee, and Orange County Pollution Control at Orlando. in 1976 was found in an estimated 283,401 ha of Florida's 1.01 million ha of fresh water (Figure 3). Approximately 80,972 ha of the infestation was considered dense.

RESULTS AND DISCUSSION

Florida has been researching the grass carp since 1968. One of the earliest and certainly the most controversial, studies was the Four Pond Study. One of the objectives of this research was to determine the impact of the grass carp on native sport fishes. Two of these ponds (Pasco and Suwannee) were designated as fisheries impact ponds and the fish populations were sampled. However, due to shortcomings in experimental design and technique, fisheries impact conclusions are confusing. Based on blocknet data, Pasco Pond experienced a 91% reduction by weight in the largemouth bass population. Suwannee Pond showed a 74% reduction in harvestable size bass. Population structures and indices were altered. An analysis of the data for population structures and dynamics indices yielded a confusing fact. Of 21 parameters comparable by blocknet, nine went in exactly opposite directions. If a cause-and-effect relationship existed between the presence of grass carp and the adverse effects experienced in the fish populations of these ponds, the direction of change should be consistent between ponds. The degree of change would be an individual expression of the particular pond, however, the direction should be consistent.

After reevaluation of the raw data, an alternative explanation to the drastic changes in the fish populations of Suwannee and Pasco Ponds became apparent. In Suwannee Pond, the rotenone was not contained within the blocknet samples and an estimated 272.2 kg of fish were killed outside the net. A sample of approximately one-third of the kill showed 57% by number was largemouth bass. Weight extrapolations showed 68% of the total largemouth bass biomass of the lake were killed in this sampling. Trammel nets were set in Pasco Pond and combined with blocknets removed 46% by weight of all largemouth bass in or above the 25.4 cm class. This represented a 29% removal by weight of the total largemouth bass population. Quarterly rotenone shoreline samples were taken in these two ponds for two years, and semiannual electrofishing was conducted for the same period. It is probable that over-zealous sampling was responsible for the adverse effects on the fish populations of these two ponds.

As a systems check, the total fish population at renovation from a third pond (Broward) was analyzed and compared with the final populations of the two fisheries ponds. Baseline data were not available for Broward Pond, but a comparison of the final populations is subjectively useful. Broward Pond has unique value in the fisheries study for three reasons. First, it was the only pond that had a large initial standing crop of submerged macrophytes (predominantly hydrilla) making it the only type of lake that would be intentionally stocked with grass carp for weed control. Secondly, the fish populations of Broward Pond were not subjected to the same intense sampling pressure as were Pasco and Suwannee Ponds. Thirdly, Broward Pond contained a large population of grass carp at the termination of the study.

Broward Pond was heavily infested with weeds, thus offering protection from predators and food for rapid growth of the grass carp. These factors resulted in excellent survival of grass carp. At renovation, 86 grass carp per ha (410 kg/ha) were recovered from an initial stocking of 116 per ha. During the last 10 months of the study, approximately 50 grass carp were removed from this pond for stomach content analysis. If included in the population, these fish would bring the residual grass carp population to approximately 111 individuals (511 kg) per ha. By comparison, 6.9 grass carp/ha (3.03 kg/ha) and 1.7 grass carp/ha (3.6 kg/ha) were removed from Pasco Pond and Suwannee Pond, respectively (Table 1).

The bass population in Broward at renovation consisted of 442 individuals per hectare (111 kg/ha), with evidence of good reproduction and recruitment (Table 2). Admittedly, there was no baseline data, but had we initially sampled this pond as intensively as the other two, perhaps the fish population would not have been so healthy. Broward was a relatively new pond and its fish population was probably in a state of expansion. However, other lakes in Florida, Lake Talquin for example, have bass populations recorded that exceed 112 kg per hectare according to blocknet samples (Ware and Smith 1972).

The panfish population was also excellent in this pond. There were more than 611 harvestable adults per hectare (Table 2). Good reproduction and recruitment were evident in these species. The grass carp has been accused of devastating the shallow water fishes. Total renovation values showed 5819/ ha, 8191/ha, and 37,072/ha of shallow water fishes for Pasco, Suwannee and Broward Ponds, respectively (Table 2). The pond with the largest grass carp biomass also had the largest standing crop of shallow water fishes.

Although there are no baseline diversity data for Broward Pond, the fish population appeared to be in good condition at renovation. The pond has a high Shannon-Wiener value (1.93) and a low dominance index (0.372). Pasco Pond showed values of 1.23 and 0.67 for Shannon-Wiener and Simpson indices, respectively, while Suwannee showed 0.81 and 0.78 for these parameters (Table 3). If 7.0 and 1.7 grass carp per hectare were adversely affecting the sport fish populations in Pasco and Suwannee Ponds, respectively, it would seed that Broward Pond with 111 grass carp/ha (511 kg/ha) would not have the good structure, dynamics, and bioindices exhibited by the fish populations of that pond.

The conclusions that can be drawn from this study are clouded by several factors. First, it is apparent that initial sampling in Pasco and Suwannee Ponds did considerable damage to the fish populations prior to grass carp stocking. Secondly, background fisheries data are based solely on blocknet sampling. Although blocknet sampling may have been the best technique for estimating the initial fish population, the wide disparity between final blocknet estimates and the correct assessment of the fish populations obtained by total renovation at study termination, casts serious doubts on the accuracy of the blocknet technique. Finally, the apparent changes in the fish populations of Pasco Pond was adversely affected; the changes in the fish population of Suwannee were mixed; and the final game fish population in Broward Pond was excellent.

An explanation for the differences in the final condition of the fish populations in Broward, Pasco and Suwannee Ponds may be found in the dissimilarities in their initial macrophyte communities. The relatively sparse amount of submerged aquatic vegetation in Suwannee and Pasco Ponds must have played an integral part in their ecosystems as refuge for small fish and substrate for invertebrate fauna. The elimination of this essential amount of vegetation by the grass carp would be expected to be harmful to the fish populations. Broward Pond, however, had an abnormally abundant standing crop of submerged macrophytes before the grass carp were stocked. Even after this vegetation was removed, the robust stand of emergent vegetation remaining must have been sufficient to fill the necessary roles of macrophytes in the ecology of the pond. It is, therefore, not surprising that the native fish populations of Broward Pond were healthy even with an enormous residual population of grass carp.

The grass carp is being studied in Florida for the specific purpose of controlling plants in lakes, like Broward Pond, that have an over-abundant biomass of submerged vegetation. The grass carp does not belong in ponds with normal or sparse macrophyte communities. Fortunately, small grass carp do not appear capable of surviving in such lakes. Although 96% of the grass carp survived in Broward Pond, only 3% survived in Pasco Pond and only 1% in Suwannee Pond (Table 1).

According to research conducted in the Game and Fresh Water Fish Commission, the Department of Natural Resources, and the University of Florida, it appears that the low survival rate of grass carp in Pasco and Suwannee Ponds was due to predation by largemouth bass. The dense vegetation in Broward Pond offered a refuge and food for rapid growth, thus enhancing survival of the grass carp.

Although aquatic macrophytes are a very necessary part of the aquatic ecosystem, excessive vegetation is known to have an adverse effect on sport fish populations. Bennett (1962) reported that a 50 percent cover of Potemogeton caused a 58% reduction in fish yield even though fishing pressure was up 157% for the same period. In his paper, Bennett concluded that vegetation can sometimes be directly responsible for overpopulation and stunting of fish populations. Heman, Campbell and Redmond (1969) and Michaelson (1970) experienced similar results. Buck, Baur and Rose (1975) showed better fish production in pools where vegetation had been reduced by grass carp relative to heavily vegetated control pools and pools where a herbicide had been used to reduce vegetation. Other authors, Thomaston (1962), Pfeiffer (1967), Buck and Thoit (1970), Ebert (1972), Hichman and Congdon (1972), Rasmussen and Michaelson (1972), and Barnett and Schneider (1974) have documented the adverse effects of excess vegetation on fish populations. In a 1966 report available from the Florida Game and Fresh Water Fish Commission, it is recommended that troublesome aquatic weeds such as Elodea spp. be allowed to cover no more than two percent of the water surface. Fishery biologists in the Commission presently believe 60% or more cover of hydrilla in lakes is beneficial to sport fish populations (Forrest Ware, personal communication). Regardless of the proposed usage of a particular water body, you can get too much aquatic vegetation. It appears obvious then that "leave hydrilla alone" is not among our choices.

Traditionally, Florida contends with the aquatic weed problem in three ways: water level fluctuation, mechanical control and chemical treatment. Drawdowns are very effective in weed control and have many beneficial side effects for fish and wildlife. Unfortunately, relatively few water bodies have the proper physical configuration to facilitate dewatering. Cost for mechanical removal ranges from \$988 to \$3705 per hectare per treatment and in some cases requires two to four treatements per year. With a plant such as hydrilla, that reproduces by fragmentation, mechanical methods can result in a more widespread problem. In Florida cost for chemical control of aquatic plants varies from \$247 to \$711 per hectare per treatment and again, multiple treatments are sometimes needed for year round control. Side effects of chemical control are often undesirable. The liquid formulation of hydrothol 191, effective on hydrilla at 3 ppm, has been shown to have a 24 hour TLm at 0.2 ppm for bluegill (Blackburne et al. 1971). Recent Dutch studies have shown that Diuron at a concentration of 0.1 ppm resulted in lack of fish ovarian development and heart damage (Hans von Zon, personal communication). Diuron has also been shown to block development of such zooplankters as Daphnia (Kersting 1976). Adverse side effects of herbicides on fish were noted by van Dord et al. (1974). Also with chemical control, 100% of the nutrients available in the plant are returned to the system for further plant growth.

In theory, biological control has many desirable attributes. First, one treatment costing as low as \$50/ha can be effective for many years. Second, the large standing crop of plants is removed slowly. Finally, plant nutrients are tied up in the bodies of the control agents. However, the purposeful introduction of any exotic weed control agent, especially in an ecosystem such as Florida's, should be carefully analyzed, and the decisions must be based on facts generated by solid research.

After the Four Pond Study, the Florida Department of Natural Resources (DNR) initiated a cooperative large lake study with the Florida Game and Fresh Water Fish Commission (GFC) and the University of Florida. Of the Eight Lakes Project, the DNR is responsible for research on three lakes (Bell, Clear and Holden) and shares joint responsibility of GFC on Deer Point Lake. On these lakes it is the responsibility of DNR to monitor weed control and water quality. Lake Bell in Pasco County is a 36 ha lake and was stocked with 1,780 grass carp. Clear Lake, also in Pasco County, is a 64 ha lake and received 3,180 grass carp. Lake Holden in Orange County, a 103 ha lake, received 5,080 grass carp. This stocking represents a rate of 49 fish per hectare. The lakes were stocked in October 1974. Deer Point Lake in Bay County is a 2,025 ha open-ended reservoir which was stocked with approximately 100,000 grass carp.

Control was obtained more rapidly and more completely in Lakes Bell, Clear and Holden than had been planned (Figure 4). Lake Holden in particular, received over-control. This is a culturally eutrophic lake with the sediments and water column overloaded with nutrients. Lake Holden receives a calculated 6,186 kg of total nitrogen and 707 kg of total phosphorus annually (Dawkins 1977). The Department of Natural Resources is currently removing grass carp from this system in an effort to reach a balance and allow beneficial vegetation to return to portions of the littoral zone. It has been demonstrated by Sutton et al. (1978) that hydrilla can be removed from a system by grass carp and/or chemicals, and replaced by eel grass.

The last of the grass carp to be stocked in Deer Point Lake were stocked in September of 1977. However, the bulk of the 100,000 fish were stocked in February, 1975. Illinois pondweed (Potamogeton illinoensis), which was present in problematic proportions, is greatly reduced. However, the primary target plant, Eurasian watermilfoil, <u>Myriophyllum</u> <u>spicatum</u>, is still present at pregrass carp levels (Figure 4). Pondweed is preferred by grass carp over Eurasian watermilfoil, and now that the Illinois pondweed has been controlled, a reduction in milfoil is expected. After the seasonal "die off" this winter, it is expected that the grass carp will control milfoil regrowth, if sufficient numbers of fish remain in the lake.

The main research query of this portion of the Eight Lake Study involves the effects of biological weed control, with grass carp, on water quality. Although significant changes occurred from 1975 to 1976, there appears to be little indication of a cause-and-effect relationship (Tables 4 and 5).

Trends are more noticeable when 1976 and 1977 values are compared. Lakes Bell and Holden show an overall increase in nutrient related parameters, and Lake Clear experienced a decline in the same parameters. Several possible explanations exist for the trends noted in these lakes and are probably due to a combination of several factors. The most feasible of the explanations involves the relative amounts of nutrient run-off received by each lake. As discussed earlier, Lake Holden is a culturally eutrophic lake receiving both domestic and industrial nutrients. Lake Bell is almost completely surrounded by homes and also receives industrial run-off. Clear Lake has very few homes surrounding it and much more marginal vegetation to act as a buffer zone. Therefore, the improvement in water quality in Clear Lake is probably due to the relatively low rate of nutrient input from its watershed coupled with the reduction of nutrient cycling from the bottom mud due to the removal of macrophytes. Another possible explanation lies in the relative depths of these lakes. Lakes Bell and Holden have an average depth of 3.05 and 3.87 m, respectively, with deep (11.58 m) holes biasing the average. Clear Lake has an average depth of 4.5 m and a more uniform bottom morphology. It is possible that the deeper areas of Clear Lake act as a "nutrient sump", thus reducing nutrient availability. It is also possible that the dredge holes in Lakes Bell and Holden serve this same function and without them, parameter values would have been exaggerated. An important point is that with biological control, some of the nutrients are converted to fish flesh whereas all nutrients are returned to the water column or hydrosoil when chemicals are used.

ACKNOWLEDGMENTS

The authors wish to express their sincere gratitude to Andrew J. Leslie, Jo McIntosh, and Gerry Kobylinski for assisting in the collection and analysis of the data and for editing this research paper.

6.9/3.0	-97/-96
1.7/3.6	-99/-95
111.2/510.9	-4/+760

Table 1. Number and weight of grass carp stocked and recovered from the three study ponds.

		Pasco Pond			Suwannee Pond			Broward Pond		
	number	number harvestable	weight (kg)	number	number harvestable	weight (kg)	number	number harvestable	weight (kg)	
Largemouth bass Micropterus salmoides	70.2	16.1	8.7	828.4	33.4	27.7	433.1	181.3	110.9	
Pan fish Lepomis macrochirus L. microlophus L. gulosus Pomixis nigromaculatus	7131.9	226.8	37.3	13099.7	589.0	57.1	7249.0	1346.6	178.6	
Brown bullhead Ictalurus nebulosus	99.6	49.4	18.0	239.1	227.7	55.8	393.7	101.3	13.3	
Total	7269.0	292.3	64.0	14167.2	820.1	140.6	8085.8	1629.2	302.8	
Shallow water species 🍙	5819.3			8190.5			37072.2			
Grass carp Ctenopharyngodon idella	6.9		3.0	1.7		3.6	111.2		510.9	

1

1

- 1

Table 2. Composition of principal sportfish species per hectare at study termination.

ſ

Table 3. Shannon-Wiener index of diversity and Simpson's index of dominance for Pasco, Suwannee and Broward Ponds at the time of study termination.

		₩~₩~₩₩₩₩₩₩₩₩₩₩₩₩₩₩₩₩₩₩₩₩₩₩₩₩₩₩₩₩₩₩₩₩₩		
	Pasco	Suwannee	Broward	
Shannon-Wiener	1.23	0.81	1.93	
Simpson	0.67	0.78	0.37	

	Lakes					
Parameters	Bell	Clear	Holden	Deer Point		
Dissolved Oxygen	+0.164	+0.304	+0.075	+0.0751/		
Temperature	-0.089	-0.007*	-0.019*	-0.158		
Conductivity	+0.007*	-0.950	-0.144	-0.942		
Turbidity	+0.001*	+0.011*	+0.717	+0.722		
Extinction Coefficient	-0.064	+0.833	-0.876	+0.824		
рН	-0.325	+0.846	+0.039*	-0.051		
Alkalinity	+0.018*	+0.904	+0.847	+0.724		
Chlorophyll	-0.126	+0.895	-0.266	-0.035* ² /		
Ortho Phosphate	-0.066	-0.088	-0.068	-0.514		
Total Phosphorus	+0.085	+0.160	+0.257	-0.251		
Kjeldahl Nitrogen	+0.252	+0.146	-0.302	-0.760		
Nitrate	+0.955	+0.320	-0.178	-0.041*		
Total Zooplankton	+0.038*	+0.048*	-0.188	+0.385		

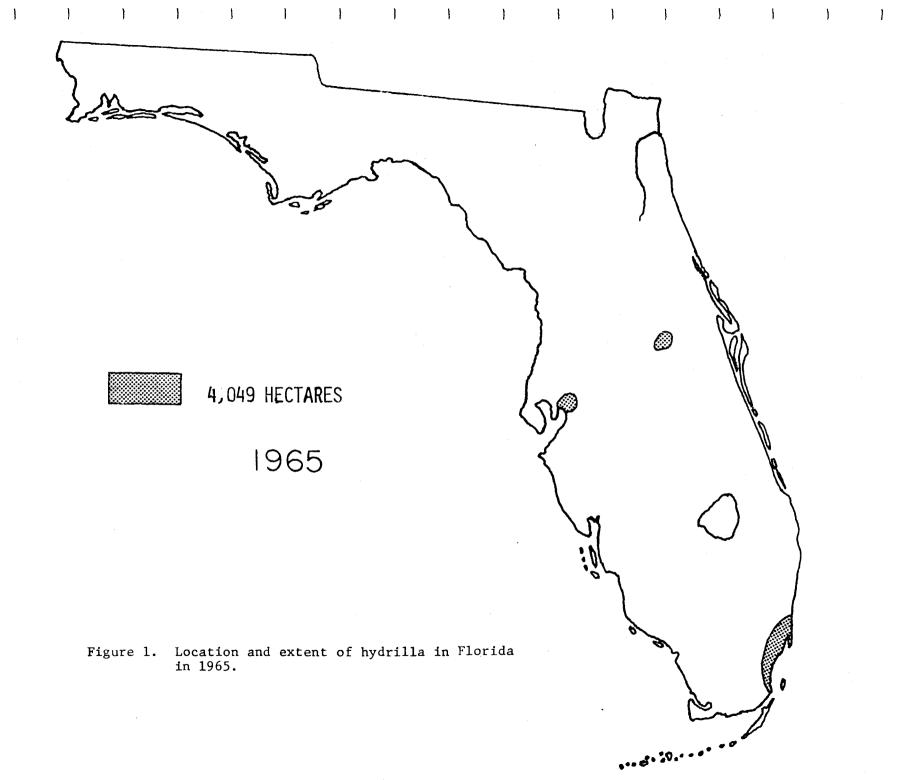
Values for Student's t statistic (P) for paired data from Lakes Table 4. Bell, Clear, Holden and Deer Point Reservoir (March 1975 - December 1975 X March 1976 - December 1976).

 $\frac{1}{2}$ Sign (+ or -) indicates direction of change. $\frac{2}{2}$ * indicates significance at P = 0.05.

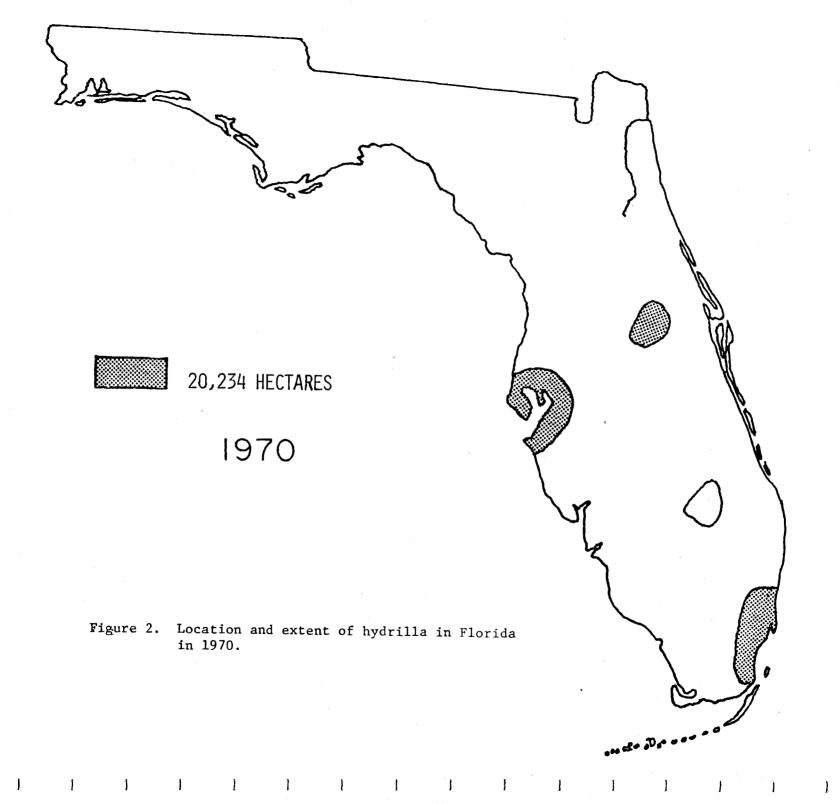
	Lakes				
Parameter	Bell	Clear	Holden	Deer Point	
Dissolved Oxygen	+0.038*	-0.602	-0.220	-0.197 ¹ /	
Temperature	-0.671	+0.625	+0.316	+0.010*2/	
Conductivity	+0.001*	_0.103	+0.114	+0.040*	
Turbidity	+0.001*	-0.980	+0.015*	+0.388	
Extinction Coefficient	+0.897	0.000	10.040	0.005	
Coefficient	+0.097	-0.088	+0.340	-0.085	
рН	+0.002*	-0.343	+0.575	+0.014*	
Alkalinity	+0.001*	-0.193	+0.343	+0.189	
Chlorophy11	+0.283	-0.049*	+0.136	-0.417	
Ortho Phosphate	+0.272	-0.399	+0.258	-0.290	
Total Phosphorus	+0.039*	-0.049*	-0.180	+0.382	
Kjeldahl Nitrogen	+0.577	-0.266	+0.023*	+0.001*	
Nitrate	+0.811	-0.422	+0.840	-0.779	
Total Zooplankton	-0.167	_0.381	+0.460	-0.354	

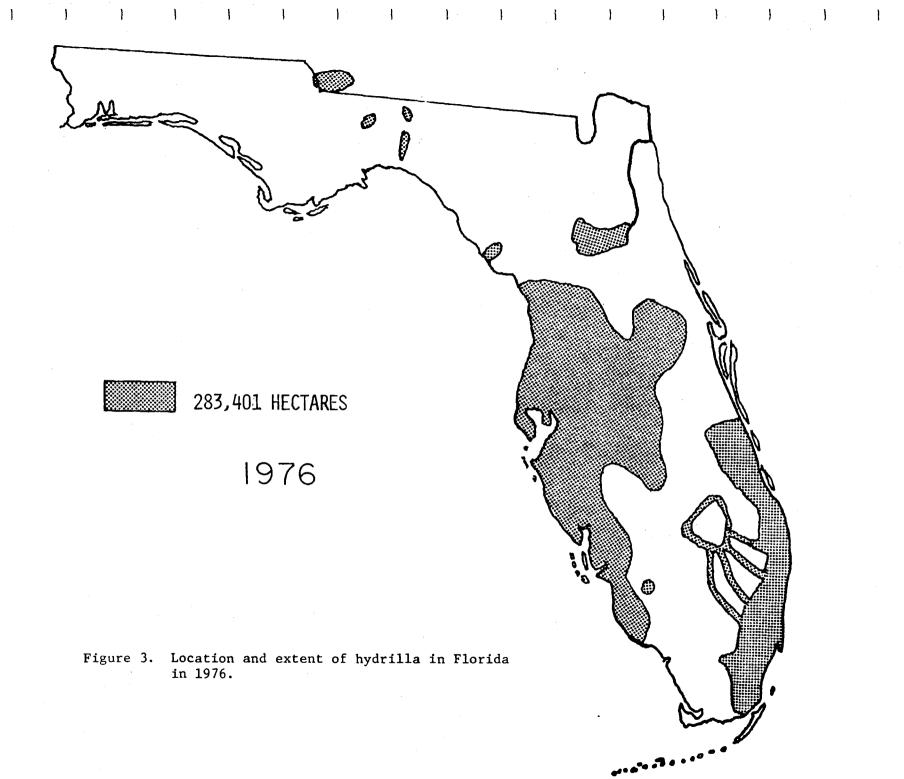
Values for Student's t statistic (P) for Paired data from Lakes Table 5. Bell, Clear and Holden and Deer Point Reservoir (March 1976 -December 1976 X March 1977 - December 1977).

 $\frac{1}{2}$ Sign (+ or -) indicates direction of change. $\frac{2}{2}$ * indicates significance at P = 0.05.



)

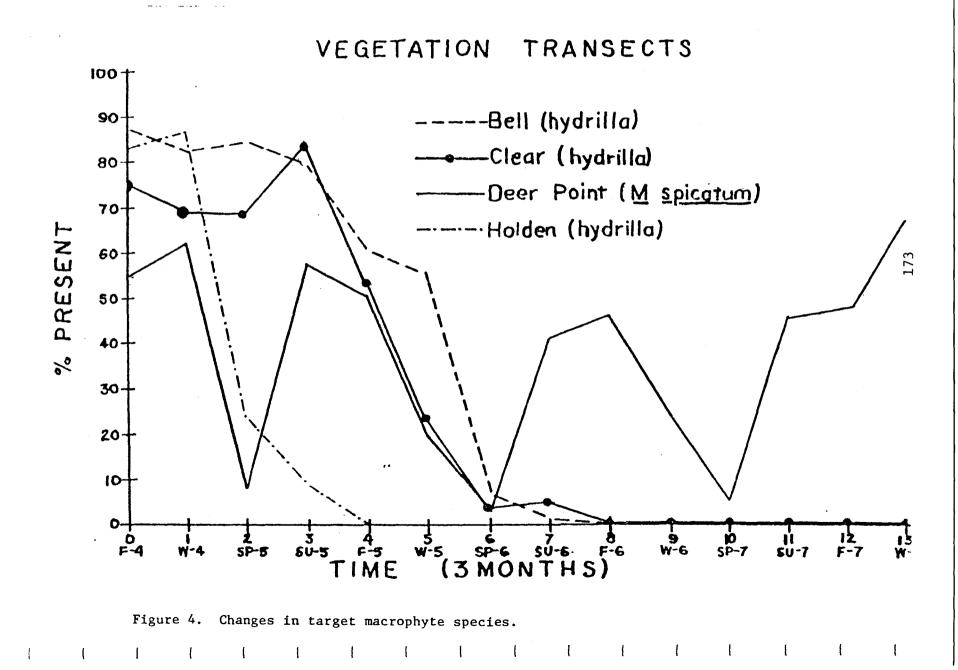




)

1.72

)



LITERATURE CITED

- Barnett, B.S. and R.W. Schneider. 1974. Fish populations in dense submersed plant communities. Hyacinth Cont. J. Vol. 12
- Bennett, G.W. 1962. Management of artificial lakes and ponds. Reinhold Pub. Co., N.Y. 283 pp.
- Blackburn, R.D., K.A. Boyer and C.E. Timmer. 1971. Phytotoxicity of four formulations of the alkylamine salt of endothall on <u>Hydrilla</u> <u>verticillata</u> on fish. Hyacinth Cont. J. 9(1):55-58.
- Buck, D.H., R.J. Baur and C.R. Rose. 1975. Comparison of the effects of grass carp and the herbicide Diuron in densely vegetated pools containing golden shiners and bluegills. Prog. Fish-Cult. 37:185-190.
- , and C.F. Thoit. 1970. Dynamics of one-species populations of fishes in ponds subjected to cropping and additional stocking. Nat. Hist. Surv. Bull. 30:165 pp.
- Dawkins and Associates. 1977. Lake Holden storm water management and restoration survey. Engineer's Proj. 056-S02. Dawkins and Assoc. Orlando, Fla. 55 pp.
- Dord, D.C. van, B.J. Hoogers, and J.C.J. von Zon. 1974. Studies on the sideeffects of herbicides used in the aquatic environment. Proc. EWRC. 4th Int. Symp. Aq. Weeds. Wien:173-179.
- Ebert, V.W. 1972. Determination of the effects of fertilizer-induced plankton turbidity supplemented by herbicides on submerged aquatic plants. Job. Compl. Rep. Fed. Aid Proj. F-15-R-7.
- Heman, M.L., R.S. Campbell and L.C. Redmond. 1969. Manipulation of fish populations through reservoir drawdown. Trans. Amer. Fish. Soc. 98:293-304.
- Hichman, G.D. and J.C. Congdon. 1972. Effects of length limits on the fish populations of five North Missouri lakes. Symp. Overharvest and Mgt. of Largemouth Bass in Small Impoundments. N. Cent. Div., Am. Fish. Soc. Special Pub. No. 3. 84-94 pp.
- Kersting, K. 1976. The use of microsystems for evaluation of the effect of toxicants. Hydrobiol. Bull. 102-197.
- Michaelson, S.M. 1970. Dynamics of balanced and unbalanced bass-bluegill populations in ponds in Boone County, Missouri. M.A. Thesis.
- Pfeiffer, P.W. 1967. Some physical, chemical and biological characteristics of Greenbo Lake. Kentucky Fish. Bull. No. 32.
- Rasmussen, J.L. and S.M. Michaelson. 1972. Attempt to prevent largemouth bass overharvest in three Northwest Missouri lakes. 1974 Symp. Overharvest and Mgt. Largemouth Bass in Small Impoundments. N. Cent. Div. Am. Fish. Soc. Special Pub. No. 3. 69-83 pp.

- Sutton, D.L., V.V. Vandiver, Jr., R.S. Hestand and W.W. Miley, II. 1978. Use of grass carp for control of hydrilla in small ponds. Proc. Int. Symp. Grass Carp, Univ. of Fla., Jan. 1978.
- Thomaston, W.W. 1962. The results of population alteration and factors affecting balance in farm ponds in Georgia. Proc. 16th Conf. S.E. Assoc. Game and Fish Co.. 361-369 pp.
- Ware, F.J. and S.L. Smith. 1972. Study V. Lake Talquin investigations. <u>In:</u> Annual Progress Report for Investigations Project. Lake Management and Research. D.J. F-12-13. 1-25 pp.

THE ROLE AND IMPACT OF INTRODUCED GRASS CARP

(CTENOPHARYNGODON IDELLA VAL.) IN THE

UNION OF SOVIET SOCIALIST REPUBLICS AND

SEVERAL OTHER EUROPEAN COUNTRIES¹

W.W. Miley II, D.L. Sutton, and J.G. Stanley

Biologist III, the Florida Department of Natural Resources

Associate Professor, University of Florida

Unit Leader, Maine Cooperative Fishery Research Unit

INTRODUCTION

Florida supports an abundance of freshwater aquatic plants that are an essential part of the aquatic ecosystem, but often these plants grow to such an extent they interfere with other important water uses. A rapidly growing exotic, hydrilla (<u>Hydrilla verticillata</u> Royle), has spread widely and caused great damage. There is a need to manage this plant. Herbicides are effective in retarding growth and is the most commonly used method for treating hydrilla infestations. Herbicides, however, provide only temporary relief because plants immediately grow back from the reproductive structures that are not killed. In some cases undesirable side effects are seen; oxygen depletions, algal blooms, and secondary fish kills.

Needed was an effective and economical control method with minimal environmental impact, one that would approximately restore the system to the condition found before the invasion of hydrilla. Several biocontrol methods were considered but one, the grass carp (<u>Ctenopharyngodon idella</u> Val.), appeared to have considerable merit because it was reasonably selective and economical. However, the grass carp is controversial because, among other reasons, it is in the same family of fishes as the carp (<u>Cyprinus carpio L.</u>), a species that is generally considered a pest in the United States. There is concern of an adverse impact, but most of the uncertainty centers on the reproductive potential and whether the grass carp will achieve large destructive populations.

To learn more about the reproductive biology, environmental impact, and harvest techniques, two of the authors (W.W. Miley II and D.L. Sutton) met with scientists and inspected areas in the Netherlands, Poland, Czechoslavakia, Federal Republic of Germany (West Germany), Austria, the United Kingdom, and the Soviet Union. Jon Stanley took part in the U.S.S.R. part of the trip

¹ Cooperative study of the Florida Department of Natural Resources, Bureau of Aquatic Plant Research and Control at Tallahassee; the University of Florida Agricultural Research Center at Fort Lauderdale; and the Maine Cooperative Fishery Research Unit of Maine, Orono.

and visited Czechoslavakia.

Some of the people we visited were present at this symposium and have given first hand accounts of grass carp in their respective countries. However, D.S. Aliev from the U.S.S.R., among others, was unable to attend the conference and we would like to present some of the information we were told and of the observations made in the various countries. Some of these experiences can perhaps be applied to understanding the place for grass carp in Florida.

RESULTS AND DISCUSSION

In the Netherlands most of the 150,000 hectares of surface water is in the form of canals. Canals are multi purpose with the main function to move water from lands below sea level to pumps. Aquatic plants impede water flow and therefore control measures are needed. Generally chemical and/or mechanical methods are practiced but alternatives are being sought because mechanical removal causes turbidity due to bottom composition, and herbicides leave chemical residues; both methods may cause oxygen depletion. Many herbicides have been taken off the market due to adverse effects on fish and other aquatic life but those still allowed are regulated closely.

Research on the grass carp in the Netherlands is under the guidance of Hans von Zon, who attended this conference and presented details of his work. Preliminary work with the grass carp suggested minimal environmental damage. Macrofauna species diversity was undiminished during a three-year study and diversity of plankton was maintained without the disappearance of any species after the grass carp was introduced to canals. Dramatic changes in water quality was not observed except in small aquaria. Impact studies of native fishes is currently in progress but interference with spawning or production is not anticipated.

In Poland, aquatic plants are a problem mainly in cooling ponds of power plants. Herbicides and mechanical control methods were used in the past but have been entirely replaced by biocontrol with the grass carp. The degree of control is regulated by grass carp stocking rate. The grass carp ameliorated some of the adverse effects caused by heated water and escaped individuals apparently have had no noticeable effects in non target areas.

Most of Poland's 60,000 hectares of ponds and lakes are stocked with grass carp. The country also has two large rivers, the Vistula and the Odra. At times these are turbid, fast flowing waters with annual temperature ranges from just above freezing to 28 C. The grass carp has been in these rivers, though not intentionally stocked, for approximately 12 to 13 years in small numbers and natural reproduction has not occurred. Dr. Opuszynski and Professor Dr. Backel, of the Department of Inland Fisheries, do not believe the fish is capable of successful reproduction in the rivers of Poland. Karol Opuszynski attended the conference and presented information on the culture and biological consequences of grass carp.

In Czechoslavakia, grass carp research in aquaculture was done at the Fisheries Research Institute at Vodnany and in natural systems at the Laboratory of Fisheries Research at Bratislava. Grass carp were introduced in

Czechoslovakea in the mid-sixties, primarily for aquaculture and stocks are now maintained by artificial spawning. In addition some fish entered the country by immigration up the Danube River from escapes from fish farms. The main use of the grass carp is in controlling aquatic vegetation in polyculture fish production ponds. Grass carp are also harvested and are a preferred food item. Last year only 20 metric tons were produced on the fish farms in Czechoslovakia.

Fishery scientists have not recorded adverse effects of grass carp in ponds or rivers. Although the grass carp has not caused any negative impacts, a tapeworm introduced along with the grass carp has caused considerable damage to the valuable carp fisheries in some ponds.

In West Germany the grass carp is being studied at the Bundeforschungsanstalt fur Fischerei in Ahrensburg under the direction of Volker Hilge and in southern Germany at the Bayerische Landesanstalt fur Wassenforschung at Wielenback under the leadership of Dr. Martin Bohl, who attended the conference and presented a paper on parasites of grass carp.

The grass carp has been studied for some time in West Germany but the technology is in about the same stage of development as in Florida. Effective control of aquatic weeds in small bodies of water has been achieved without adverse effects and the grass carp is now routinely used for local aquatic plant problems. Investigations are currently being conducted on the feasibility of using grass carp in large water bodies and reservoirs.

The grass carp is used extensively for control of aquatic weeds in Austria, and has been stocked in most lakes and ponds in the lowlands. This fish has been in Austrian rivers, which are very cold in summer due to melting glaciers, for about 12 years with no evidence of natural reproduction. In hatcheries this fish has been artificially reproduced for the past seven years. Grass carp can be bought and stocked by anyone in Austria without a permit. While in Austria, we visited Dr. Alexander von Mensel at his private research hatchery facility near Wettmannstatten. Dr. von Mensel ships grass carp to several countries including the United States.

Weed control recommendations in Austria with grass carp are 400 to 600, six to ten centimeter fish per hectare if no predators are present. At this stocking rate weed control can be expected in one year. If pike or other predator fish are present, the recommended rate is 200 to 300, 700 gram fish per hectare.

The people of Great Britain are very conservation oriented and freshwater resources are highly valued. Sport angling is a popular pastime with the British. Similar to the Netherlands, British aquatic weed problems occur in the rivers and canals of the Fens, reclaimed lands that are below sea level. Alternatives to the temporary and expensive mechanical and chemical techniques are being sought.

Great Britain is still in an experimental phase with the grass carp. Early work has shown promising potential for biocontrol using the grass carp. Two hundred-twenty kilograms of grass carp per hectare could be expected to reduce aquatic vegetation by fifty percent of its potential. In an impact

study on the native bream, <u>Abramis</u> brama, grass carp was found to have no effect on survival of this species. The bream showed increased growth rates in direct proportion to the density of grass carp.

In the Soviet Union research work on grass carp was conducted at numerous locations and this fish has been stocked widely for aquaculture, weed control in canals and cooling reservoirs, and commercial fisheries. The attempts to establish the grass carp in rivers is enlightening relative to whether reproduction is possible in the United States. Several river systems have been stocked annually with millions of fingerlings without resulting in self-sustaining populations. The most intense effort at naturalization was in the Volga River and the result was almost total failure, the population reached only one fish per ten hectares after 20 years of stocking. Spawning was observed but recruitment was negligible. In the Kuban River spawning stocks were established, but subsequent construction of the large Kuban Reservoir has made further reproduction doubtful. In other rivers, the Syr Darya and Ili, reproduction was sufficient to perhaps sustain small populations but additional hatchery stockings were still made. However, further stocking in the Ili River is no longer recommended by fishery biologists. The report of reproduction in the Terek River was of herbivorous fish and may or may not have included grass carp, the Soviet workers we visited do not know.

Of the five locations where spawning occurred, only one, the Kara Kum Canal, had massive successful reproduction. Here the population built to a peak, and then declined to about half the maximum, due perhaps to the change in habitat of eggs laid in the flowing water of the canal and the large number of young produced in Kelif Reservoir, the nursery area, the fish did not pervade the entire canal system. Large numbers are still stocked, mostly in small side canals or temporary reservoirs.

Based on the information from Soviet biologists and from the scientific literature we wrote a review of spawning requirements as pertains to whether spawning could occur in the United States (Stanley, Miley and Sutton 1978). The reproductive requirements for spawning, egg incubation, and larval survival are summarized:

- 1. Brood stock must be in good nutritional and physiological condition.
- 2. During the breeding season the adults migrate to the spawning habitat, flowing water of rivers or canals.
- 3. A focal point for congregation of ripe adults is needed, usually with turbulance such as occurs downstream from an island, river confluence, or below a dam.
- 4. After egg laying, turbulent upwelling is needed to keep the eggs suspended until they are water hardened and become more buoyant.
- 5. Flowing water with a velocity of 1.0 to about 1.5 meters per second must be present to carry the semi-pelagic eggs, although eggs have been collected in water moving as slow as 0.6 meters per second.

- 6. Optimum temperatures for sexual maturation, spawning, and egg incubation are 22 C to 26 C, but reproduction has been observed at temperatures as low as 18 C and as high as 31 C, with increased deformities and mortality toward the extremes.
- 7. The incubation time depends on the temperature; hatching occurs in about 26 hours at optimum temperatures, about 40 hours at 20 C and about 20 hours at 30 C.
- 8. The flowing water must occur over sufficient distance so that eggs remain suspended for the entire incubation. Theoretically 50 kilometers is the minimum distance and 90 kilometers the optimum (at optimum temperatures and flow rates), but the observed average is about 85 kilometers (eggs may be caught in back eddies and hence drift less distance than predicted by the current at midchannel).
- 9. Vegetated lakes adjacent to the river or reservoirs located downstream serve as nursery grounds for young.
- 10. The first food of the newly hatched fry (at about two days age) is rotifers, followed by arthropod zooplankton, and after several weeks, aquatic vegetation.
- 11. The grass carp apparently has poorly developed behavioral mechanisms for escaping predators.

The information on environmental impact of grass carp in natural systems was not as complete as on spawning requirements because the problem had not been addressed with rigorous research in the Soviet Union. Many tests in fish ponds established that the grass carp had beneficial or no effects on companion species, except at high stocking rates where decreased growth of carp was observed. The few observations made in natural systems indicates a benign effect; adverse effects have not been identified. Grass carp in natural systems generally did not destroy all vegetation and there was no noticeable effect on commercial species. In the semi-natural system of the Kara Kum Canal grass carp reduced plant masses that formerly died, decomposed and caused oxygen depletion. Oxygen improved after grass carp introduction and apparently was accompanied by an increased fish production; at least the harvest of fish increased. Pike (Esox lucius) might have declined due to vegetation control. In only one pond, heavily stocked with grass carp, was there a decrease in native fishes; pike and perch (Perca fluvialtilis) were eliminated. One Soviet scientist pointed out that if vegetation were eliminated from natural systems some important commercial species that lay eggs on plants would be effected. However, no documentation of such adverse effects have been made in open systems because the grass carp failed to destroy aquatic vegetation.

Algal blooms reported after the grass carp removed vegetation in experimental ponds are the exception. Generally, there were no observed increases in algal populations after stocking of grass carp. Of course, silver carp that feed on phytoplankton are always stocked with grass carp in the Soviet Union and this perhaps compensated for any tendency toward algal blooms. Capturing the grass carp from large bodies of waters and production ponds was difficult and not completely solved. One technique was to wait until cool water of 15 C or less made the fish sluggish, then they could be easily netted.

CONCLUSIONS

Based on the findings of our trip, a similar trip to Asia by Dr. William Haller and Mr. Bill Bailey, and review of the literature, we predict that the grass carp may shed eggs in the United States, probably in the Mississippi River system, but except for possible local concentrations, the probability of establishing a large population of grass carp is remote. We further predict that, in the areas of Florida where the grass carp will be stocked, they will have considerably less adverse effects on the aquatic ecosystem relative to other methods of weed control. In dense stands of hydrilla, the grass carp should have an ameliorative effect on sport fish.

REFERENCES

Stanley, J.G., W.W. Miley II, and D.L. Sutton. 1978. Reproductive requirements and likelihood for naturalization of escaped grass carp in the United States. Trans. Am. Fish. Soc. 107:119-128.

NATURAL REPRODUCTION OF GRASS CARP IN THE TONE RIVER AND THEIR POND SPAWNING

Minoru Tsuchiya

Chief of Fishculture

Saitama Prefectural Experiment Station

Kazo-shi, Saitama-Ken, Japan

INTRODUCTION

Natural reproduction of grass carp (<u>Ctenopharyngodon idella</u>) in the Tone River was considered a rare case, because it was thought that this fish only reproduces within its own original habitat, Mainland China. The original spawning occurred 30 years ago. Since then natural reproduction has continued. During this period, however, the catch of grass carp has been remarkably decreased by environmental changes in the Tone River, especially the rapid change in the quantity and quality of the water, due to its increased use, and the development of swampy areas around the river. In this paper the recent condition of natural reproduction of the fish and the method of pond spawning being operated to supplement natural resources of the fish are reported.

NATURAL REPRODUCTION IN THE TONE RIVER

Transplantation and Natural Reproduction in the Tone River

Record of transplantation

Twice between the years of 1943 and 1945, a total of 23,000 fry, mainly grass carp, were introduced into Lake Kasumi, Lake Kita and the Tone River. The fry, from the Yang-tze River in Mainland China, were between 6-10 cm in length and 3-12 g in weight (Anon. 1958).

Catch in fingerlings

In September, 1948, 18 fingerling grass carp (8.8-25.2 cm) and 9 silver carp were caught in Lake Kasumi and Lake Kita (Tange 1949). Also, 14 fingerling grass carp and 3 silver carp were captured in the Watarase River which connects with the Tone River (Nakamura 1949). From the results of age-examination of these fishes, they were presumed to have naturally reproduced in the Tone River system. Moreover, during investigations between 1950 and 1952, additional grass carp fingerlings were seen along with bighead carp and black carp. It was confirmed that all these species were reproducing in the Tone River (Akiba 1955).

Collection of eggs and fingerlings and confirmation of the spawning ground

For the first time, in 1954, floating eggs and hatched fries were collected at Rokkaku, Inashiki county, in Ibaragi prefecture (Akiba 1955). In 1955, floating eggs were captured upstream at Otonemachi, Kitasaitama county, Saitama prefecture. Furthermore, in 1956, it was observed that adult grass carp and silver carp caught during spawning were spawning between Manume-machi, Osato county and Sukamura, Kitasaitama county, in Saitama prefecture (about 10 km) (Tsuchiya and Takahashi 1956) (Figure 1). Therefore, grass carp, silver carp, bighead carp and black carp transplanted from Mainland Chima, naturally spawned five years after their transplantation and have continued spawning constantly.

Spawning Habits and Conditions of Breeding

Spawning ground location

In the investigations during 1956-1965, the upper limit of the spawning ground was at Manume-machi, Osato county (165 km up-stream from the river mouth) and the downstream limit at Sakai-machi, Saruchima county in Ibaragi prefecture. The distance in approximately 25 km. Unitl 1960 the spawning ground was near the upstream part of the river between Manume-machi and Haneoi-city. The spawning site began shifting downstream due to the increase of gravel in that area. Moreover, the construction of the Otone Zeki Weir (154 km upstream from the river mouth), started in 1964, making it impossible for adult carps to migrate upstream from the Weir. The spawning site was confined to the area between Otone-machi and Sakai-machi after the completion of the Otone Zeki Weir in 1968, even though there are fishpaths in the Weir (Suzuki and Tsuchiya 1957 - 1966).

Environment at the spawning site

The river at the spawning area has a silt bottom, a width of 800 - 1000 m and a 400 - 600 m water flow width during normal flow. The depth is 1.5 - 3.0 m deep in the channel and 0.5 - 1.0 m deep for three-fourths of the water flow width. At the time of spawning, however, the water level rises 0.5 - 2.0 m from the usual level and water flow width increases. At Kirikashi, located in the spawning site, the discharge rate becomes 400 - 2300 m³/sec during the spawning period, whereas the annual average is 200 - 250 m³/sec. The flow velocity is 80 - 100 cm/sec in the channel during normal flow, but during increasing discharge, it becomes faster.

Migration for spawning

Adult carps start migrating from Lake Kasumi and Lake Kita, their original habitat, and the downstream area of the Tone and Edo River toward the spawning ground prior to the spawning season. Migration usually starts at the beginning of April. By the middle of May the first adult carp arrives at the spawning ground. Since 1963, a sluice gate, which is for the prevention of backflow and salt pollution was established on the Tokiwa River (the entrance of Lake Kasumi and Lake Kita). In 1970, the Kakozeki Weir was built on the main stream of the Tone River (18.5 km upstream of the river mouth), and it has been difficult for the adult carps to migrate. Thus, most of the adult fishes arrive at the spawning site immediately before the spawning starts. Immediately after spawning, the fishes start moving downstream and return to the downstream area after several days.

Period of spawning

According to investigations, from 1956 to 1965, the spawning period lasts three months from June to August, with the most active spawning taking place between the end of June and the middle of July. The spawning periods for the last five years (1973-1977) have been during nearly the same dates as shown in Figure 2. The annual average has been 3.3 times and large numbers of eggs were spawned each time. This does not include data for the times of little spawning activity.

Primary factors for spawning

The main factor initiating spawning is a rapid increase in water level caused by heavy fain in the upstream river. Spawning will begin under circumstances where the water level rises 0.5 - 2.0 m with muddiness and a water temperature reaching from 19 - 23 C. Even though the rising water with turbidity occurs due to rain and melted snow, the spawning activity does not take place without an increase in water temperature. The relationship between fluctuation of water level and spawning days from 1973 to 1977 is shown in Figure 2. Transparency of the water ranges between 0.2 - 0.4 m and pH between 6.9 - 7.1 during the spawning period.

Spawning activity

Four different kinds of carps, (grass carp, silver carp, bighead carp, and black carp) spawn simultaneously at the same spawning ground. Silver carp are observed more easily than the others since they spawn near the surface. It is difficult to observe the other three carps spawning, because their numbers are few and they tend to spawn in a somewhat deeper strata. All four species of adult carps are captured at the spawning ground and since all produce floating eggs, it is obvious that the four carps spawn simultaneously at the same spawning site. The duration of spawning lasts almost all day. After spawning the adult female fish drift downstream in a cyncopic state from exhaustion.

Drift of the eggs and hatched fries

The spawned and fertilized eggs soon start absorbing water and expand. After 1-1.5 hours they become semi-floating eggs 5.0-5.5 mm in diameter and drift downstream. As distribution of the eggs in the flow is almost uniform, they can be captured at all depths throughout the river. According to the 1964 investigation, the hatching location for the floating eggs was in the main stream of the Tone River near Sahara City, Chiba prefecture. The average drifting velocity of the eggs was 2.7 km/hr (Kasebayashi, Kafuku and Nakano 1966). The fry which hatched near this area slowly drifted downstream and were influenced by tide. They entered a swamp, rice fields and a channel where they grew to young fishes.

Large numbers of eggs also drifted into the Edo River which is connected with the Tone River at Sakai-machi, the downstream limit of the spawning ground. Near Noda City, Chiba prefecture, 22 km downstream from the junction of the Tone and Edo Rivers, large numbers of eggs are collected at present time and they are used for artificial reproduction. Because most of the floating eggs in the Edo River are useless for natural reproduction, because the Edo River is shorter in length than the Tone River, the collection of the eggs is permitted in this river.

Also the number of drifting fry per one spawning is estimated to be 17×10^8 , but the number of spawning eggs deduced from density of the float-ing eggs is assumed to be 60×10^6 .

A Tendency and Prediction of the Carp Resources in the Tone River System

Decreasing grass carp numbers

Grass carp and silver carp catches by years in Lake Kasumi and Lake Kita are shown in Table 1. Since 1960, the catches of grass carp have decreased and they are rarely captured now.

Ratio change of grass carp fry to total numbers of all carp fry

The ratio of numbers of grass carp fry to total numbers of all carp fry obtained from the collection of the floating eggs and the number of the fry hatched from the floating eggs (1958-1964) is shown in Table 2. Since 1964, the ratio has been assumed to be 3% although an accruate investigation has not been made.

Factors assumed to diminish catch

The fry hatched in the main stream of the Tone River around Sahara City enter swamps, rice paddy fields and channels in the downstream area where they grow to adulthood. The environment of these rearing grounds have been changed rapidly by river improvement and land development since 1960. In other words, due to improvement of water banks, channels, sluice gates and reclamation of swampy areas, soft water weeds needed for growth of the fingerlings have decreased.

Decrease of water weeds due to use of chemical weed killers

Water weeds, needed for growth of grass carp, in the downstream areas and lakes decreased because of increased use of the weed killer, PCP, between 1960 and 1970. Water weeds in the main stream of the Tone River have increased since the use of PCP has been restricted.

Influence of the construction of dams and weirs

Since 1976, the dams constructed for the purpose of increasing water supply and flood control have been completed at seven different locations in the upstream area of the Tone River system and five more dams are under construction or planning. Moreover, the Otone Weir and Kako Weir were constructed in 1968 and 1970, respectively. Owing to the construction of these dams and weirs, the following influences have been observed:

- The habitat for fingerlings and adult fishes in the river was decreased by less water discharge.
- (2) Since the Otone Weir was built, at the center of the original spawning ground, the spawning site moved downstream and was narrowed.
- (3) The Kakozeki Weir made it difficult for the adult carps to migrate upstream to the spawning ground.

Influence of increased eutrophication in Lake Kasumi and Lake Kita

Around Lakes Kasumi and Kita, located near Tokyo, many houses and factories have been built recently. Their drainage discharge into the lakes increased eutrophication. An abnormal increase in phytoplankton caused by increased eutrophication has been constantly observed for the last several years. It followed that, in 1973 and 1974, many cultivated carps perished. The vast increase of phytoplankton in lakes lowers water transparency and decreases the growth of submerged plants. This is advantageous to silver carp which feed on phytoplankton, but it is thought to be one of the main reasons why herbivorous grass carp have remarkably decreased (Figure 3).

Future prediction

As mentioned above, absolute numbers of grass carp have been decreasing since 1960, and in the present situation it is doubtful their numbers will increase. The causes, as pointed out previously, are due to environmental changes. Therefore, if the irrigation plan for the Tone River, causing more and more water demand, is carried out, natural reproduction will not take place.

PRESENT SITUATION OF ARTIFICIAL REPRODUCTION SUCCESS BY POND SPAWNING

Background for the Necessity of Collecting Eggs by Artificial Spawning

For several years, since the spawning site in Saitama prefecture was discovered in 1956, the Saitama Prefecture Fisheries Experimental Station has carried out the production of eggs based on artificial mating of the adult carps caught at the spawning ground during spawning. By this means, however, it was difficult to capture adult fishes with mature eggs. Thus, since 1958, egg production has depended on the collection of floating eggs. Yet, as mentioned before, the proportion of grass carp floating eggs was low. It was therefore, recognized that collection of eggs from adult fishes directly is the only way to obtain sufficient grass carp eggs.

Progress of the Research on Artificial Spawning

Between the years 1960 and 1967, adult fishes migrating for spawning were captured near the spawning ground and were induced to spawn by injecting the pituitary of the grass carp, silver carp and so on. For the first several years adult fishes were held for spawning in the river after injections, but this did not produce good results due to injury during catch and cultivation. Since 1964, adult fishes were anesthetized with MS-222 immediately after capture and were carried to an experimental pond where they were given a hormone injection. This method worked well and for the first time fry were obtained from artificial spawning. The method of collecting eggs from wild adult fishes under anesthesia continued until 1967.

Artificial spawning of mature fishes reared in a pond

The method of anesthetizing wild adult fishes, until 1967, was applied to cultivated mature fishes being reared since 1960, and succeeded in artificial spawning. Moreover, in the same year, albino grass carp were found among the fry which were artificially hatched from the eggs of the wild mature fish. In 1976, we were successful in developing albinos from these fry.

Conversion from the method of artificial fertilization to pond spawning

From 1964 to 1969, artificial fertilization techniques were applied. In this method the adult fishes kept in a pond after injection were ovulated after a certain time interval. Eggs were collected and fertilized utilizing the dry-leading method. As a result, however, this method injured the adult fishes very badly and caused stress. It was also difficult to calculate the proper spawning time. Because of these reasons, the method of pond spawning has been used since 1970.

Pond Spawning

Adult fish

Cultured adult fish, over five years old, 5-20 kg in weight are used. Adult fishes are given fresh weeds and feed with low protein, and their genital organs are allowed to develop. In looking from the rear, externally, fish with expanded bellies and swollen excretory openings are chosen. The ideal ratio of sexes is 1.0 to 1.5-2.0 in the spawning pond. After spawning the adult fish are given a mixture of feed added to fresh weeds, and they are allowed to recover for the next spawning.

Hormone injection

The pituitary of wild silver carp is most effective. Pituitary taken from mature fish obtained in spawning condition is dried with acetone and ground. Pituitaries taken from silver carp during the winter (January through March) are also effective. On usage, the ground pituitary is dissolved with 0.6% salt water and used as an injection liquid.

Quantity of injection and number of injections

Females were injected with 5-10 mg/kg of pituitary extract; males were given one half this amount. Injections are given inside the abdomen through the muscles at the base of the pectoral fin. The mature fish are kept in shallow water, therefore, they should be injected carefully and quickly without injury. Although injections can be given twice, once every six hours, a single injection is usually sufficient.

Spawning pond

A pond $100 - 250 \text{ m}^2$ in area and 1 - 1.5 m deep is sufficient though it varies with the size of the adult fish and number of fishes. After the injection, water is discharged into the pond to stimulate the mature fish. The discharge should be $50 - 100 \text{ m}^3/\text{h}$ for a pond 250 m^2 in area. Ten to twenty females and 15 to 30 males can be treated in a pond of this size.

After the first injection, spawning begins after 20 hr in 20 C water, 15 hr for 22 C water and 12 hr for 24 C water. Spawning usually lasts from 6-12 hr. Spawning activity can be observed many times from dawn until sunrise. Active spawning tends to take place during the night.

Egg collection

Floating eggs are collected in an egg collecting net (a fine meshed net) installed at the outlet manhole. Since the eggs immediately after spawning are small in size, they will be discharged after a certain time period. Because of this, during spawning, only surface water is discharged; during egg collection, bottom water is discharged.

Hatching

The hatching tank shown in Figure 4 is used to hatch the eggs. The egg membrane has a tendency to be easily broken as compared to the naturally spawned eggs. The results of grass carp spawning in ponds by hormone injection for the last several years is shown in Table 3.

Raising of the Fingerlings

For the period between egg yolk absorbtion and the beginning of normal swimming, fry should be allowed to drift in a rearing tank within the hatching tank. Because the fry tend to lie over one another in stationary water during this period, attention should be given on avoiding death from suffocation.

The fingerlings are transferred to either cagenets or rearing ponds where feeding begins. A rearing pond made of concrete with $30 - 100 \text{ m}^2$ area and 30 - 50 cm deep is used. Cagenets are effective in larger ponds. This net is 4 m^2 (2 x 2 m) and 1 m deep and is hung on bamboo poles. The number of net meshes at the beginning is more than 30 meshes. The mesh should be changed into a larger size as the fry grow. A solution of boiled egg-yolk and compound feed (powder) dissolved in water is sprayed over them. The fingerlings are raised with this feed for 2-3 weeks until they become 2.0-2.5 cm in length.

Raising of breeding fishes

Breeding fishes are raised in a large rearing pond of 10-20 acres and are fed on a compound powder feed and crumbling feed. During the rearing period, between July and October, they are sent as breeding fish at 5-20 cm in length on request.

Complications

The following problems are encountered during artificial spawning and fry culture:

- (1) Determination of the most effective age of mature fish for spawning.
- (2) The rearing environment and management of feeds to develop genital organs of mature fish.
- (3) The appropriate time for injection of brood fish.
- (4) Development of effective methods for collecting eggs in pond spawning.
- (5) Defining the price of pituitary glands and securing them.
- (6) Improvement of hatching methods.

REFERENCES

- Akiba, D. 1955. The reproduction of grass carp at Tonegawa. Abstract. National Symposium on (fish) Breeding and Reproduction in Lakes and Rivers.
- Anonymous. 1958. Association for the Protection of Japanese Water-Products Resources. Water-Products Resources. Special report on grass carp.
- Kasebayashi, N, T. Kafuku and I. Nakano. 1966. The ecology of young grass carp and silver carp at Tonegawa river/lake areas. Kasumigaura and Kitaura Water-Products Office, Ibaraki County. Investigation Research Report.
- Nakamura, M. 1949. The fish yield of small grass carp and silver carp at Watarase River area. Research Institute of Resources Science. Report No. 14.
- Suzuki, K and M. Tsuchiya. 1957 1966. Study of the artificial seed-collection of grass carp. Saitama Water-Products Experiment Station. Business Reports Nos. 9, 11, 13, 15, 17, 19, 21, 23 and 25.
- Tange, F. 1949. The reproduction of herbivorous fish and grass carp near Kasumigaura and Kitaura. Investigation, Information, Water-Products Division.
- Tsuchiya, M and K. Takahashi. 1956. Study on the artificial egg-collection of herbivorous fish. Saitama Water-Products Experiment Station. Business Report No. 6.

	Silve	r Carp	Grass Carp		
Year	Catch	Ratio	Catch	Ratio	
1956	4	21.1	15	78.9	
1957	6	42.9	9	57.1	
1958	2	22.2	7	77.8	
1959	8	14.5	47	85.5	
1960	6	50.0	6	50.0	
1961	48	94.1	3	5.9	
1962	64	100.0	0	0	
1963	87	96.7	3	3.3	•
1964	128	93.4	9	6.6	
1965	278	97.5	7	2.5	
1966	491	99.2	5	0.8	
1967	1,528	99.5	8	0.5	
1968	549	99.8	1	0.2	
1969	295	99.0	3	1,0	
1970	304	100.0	0	0	
1971	241	100.0	0	0	
1972	66	98.5	1	1.5	
1973	94	95,9	4	4.1	
1974	119	95.2	6	4.8	
1975	415	98.3	7	1.7	

Table 1. Grass carp and silver carp catches in Lake Kasumi and Lake Kita.

Table 2. Percent of grass carp fries collected from hatching.

	Year						
	1958	1959	1960	1961	1962	1963	1964
Percent	39.5	26.2	9.4	12.9	25.0*	4.9	34.3
						15.0*	19.0*

* indicates fries collected from natural river.

195

Date	Female Parents						Time	Water	······································	Survival
	Heads	Avg. Body Weight	Pitultar Kind	y Hormone Dose	Injection time	Starting time of spawning	required for spawning	temp. in pond	Number of eggs (volume)	erate after 24 hrs.
6-24-49	18	12 kg	acetone dried	5 mg/kg 5 glands/	4 - 5:00 pm	7:00 am	15 h	19.8- 21.5°	4.98 mil- 1ion	64.5%
	4	12 kg	fresh	head				С	(311 1)	
6-23-50	15	13 kg	acetone dried	5 mg/kg	2:45- 4:00 pm	5:00 am	14 h	20.0- 21.5° C	2.85 mil- lion (155 1)	60.9%
6-21-51	10	15 kg	acetone dried	5 mg/kg	4:30 - 5:00 pm	10:00 am	17 h	18.2- 19.8°	3.70 mil- lion	48.0%
	6	15 kg	fresh	36 - 226 mg/head	JUGU Par			C	(200 1)	
7-4-52	21	12 kg	acetone dried	6 mg/kg	3:30 - 4:00 pm	3:30 am	12 h	24.5- 24.8° C	3.79 mil- lion (209 l)	63,1%

1

{

١

ł

. 1

1

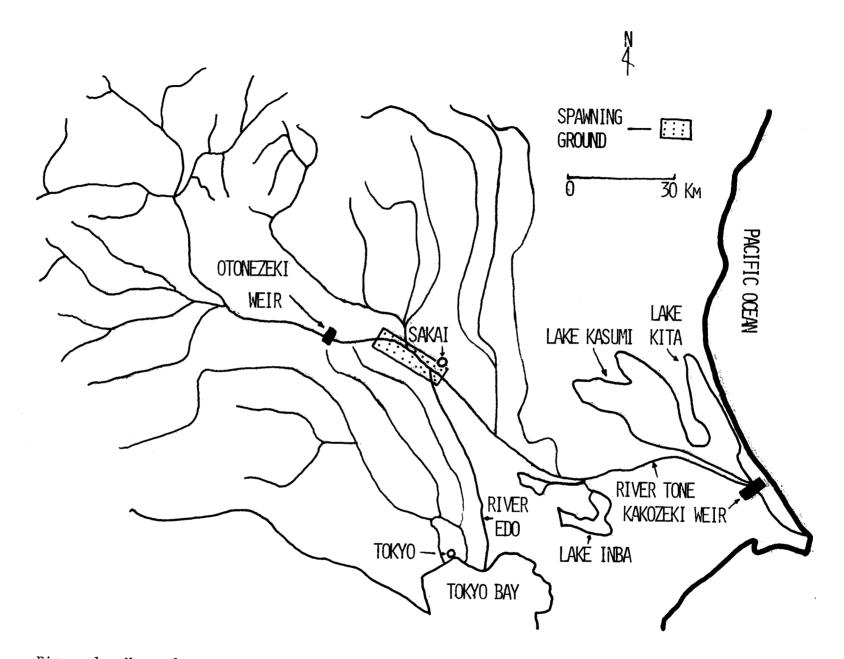
Table 3. Results of grass carp spawning in ponds by hormone injection.

· (

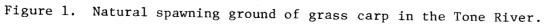
1

l

ł



ļ



1.

1

197

}

1

1

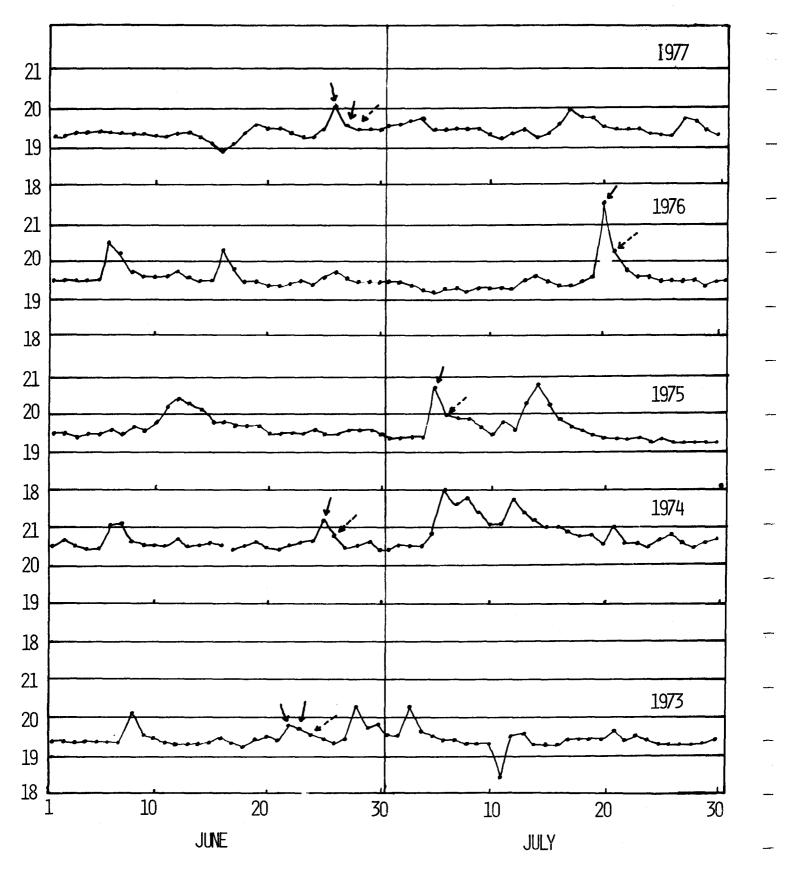
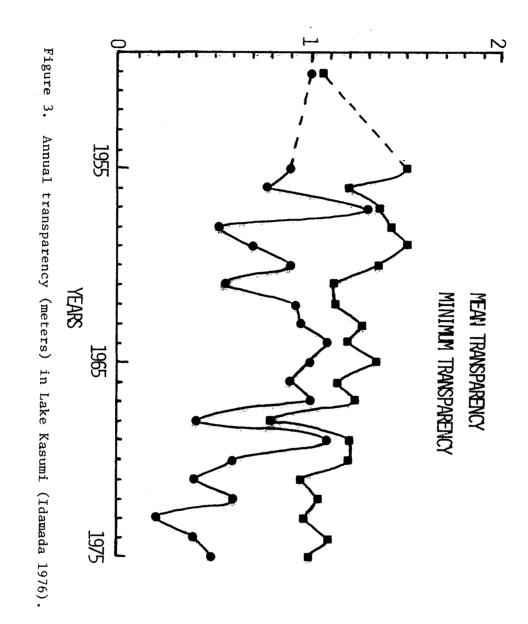
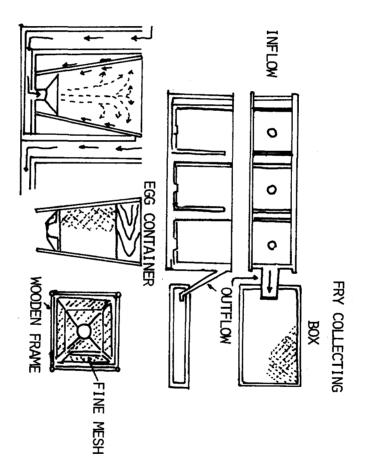
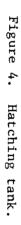


Figure 2. Water level fluctuation (meters) of the Tone River as observed at Suka, near Otonezekii Weir. Solid arrows represent spawning; dashed arrows represent egg collection.







CONTROL OF SEX IN FISHES, WITH SPECIAL REFERENCE TO THE GRASS CARP

Jon G. Stanley

Main Cooperative Fishery Research Unit, $\frac{1}{1}$ Department of Zoology

and Migratory Fish Research Institute, University of Maine, Orono, ME 04473

ABSTRACT

Some aquatic weed problems in the United States could be corrected by the grass carp (<u>Ctenopharyngodon idella</u>) but there is fear that the fish would reproduce and become a pest. Fish incapable of spawning or producing offspring are needed. A review of the literature revealed that techniques may be available for accomplishing such sex control. Fish can be sterilized if eggs are treated with cytochalasin to make the fish polyploid. Monosex grass carp have been produced by gynogenesis, but only in small numbers sufficient to stock small bodies of water. An attempt to develop monosex grass carp by sex reversal was unsuccessful. Sterilization with radiation or chemosterilants is temporary in most fish species. Hormonal treatment has been reported to cause sterility. The development and application of a failsafe technique for rendering grass carp incapable of reproducing would ensure against naturalization.

INTRODUCTION

Weed problems in many waters in the United States could be corrected by the use of a biological control agent such as the grass carp (<u>Ctenopharyngodon</u> <u>idella</u>). Biological weed control acts more gradually than chemical or mechanical methods, and consequently avoids problems of chemical residues and oxygen depletion due to decomposing vegetation. However, many environmentalists, including myself, believe that despite the advantages of biological control of aquatic plants, the use of the grass carp introduces a risk element not present in other methods; the fish might reproduce and become a permanent pest. In my opinion, widespread stocking of grass carp should be curtailed until the risks can be more critically assessed. Potential spawaning habitat must be identified and long-term environmental effects must be identified.

One way to avoid uncertainty about permanent effects would be to use grass carp incapable of spawning of producing viable offspring, or individuals that are all of one sex. Efforts to produce monosex grass carp began in 1972 at the Fish Farming Experimental Station at Stuttgart, Arkansas, and attempts to sterilize grass carp were made at the University of California at Davis. I will attempt to summarize and evaluate potential methods used to control sex of various fishes and recommend procedures for developing nonreproducing grass carp.

 $\frac{1}{1}$ The Unit is jointly supported by the University of Maine, the Maine Department of Inland Fisheries and Wildlife, and the U. S. Fish and Wildlife Service.

If sterilized fish or fish incapable of reproducing had been available, the controversy generated by the introduction of the grass carp into the United States could have been largely avoided. Assurance against reproduction would remove many objections to this fish into new waters, because if the grass carp became a pest, they would soon die off and the system would revert to pretreatment conditions. The state of the art has not developed to the point where nonreproducing fish can be placed in the hands of managers but this review will inform those in authority that reproduction in grass carp can potentially be controlled.

RADIATION STERILIZATION

Rationale

Radiation from radioisotopes or X rays causes chromosome damage, especially in actively dividing cells. Because the rapidly proliferating germ cells in gonads are more sensitive than most cells, radiation can cause sterility. To radiosterilize grass carp, a dose of radiation must be chosen that completely incapacitates gonadal germ cells, with minimal damage to other systems needed for survival and good health. The literature on radiosterilization is reviewed and evaluated relative to the possible effectivness for sterilizing grass carp (Table 1). To my knowledge, radiosterilization of grass carp has not been attempted.

Internal Radioactivity

Injection of radioactive substances that emit beta or gamma rays has been used to experimentally produce sterility in fish. Vivien (1953a, 1953b) sterilized the guppy (<u>Poecilia reticulata</u>) and swordtail (<u>Xiphophorus helleri</u>) by the injection of radioactive phosphorus, a beta ray emitter. Srivastava and Rathi (1967) castrated the Indian catfish (<u>Heteropneustes fossilis</u>) by injection of cobalt , which produces gamma rays.

Radioactive materials added to the water and absorbed by fish may affect reproduction. Krumholz (1956) observed that white crappie (Pomoxis annularis) disappeared and golden redhorse (Moxostoma erythrurum) were greatly diminished in White Oak Lake after contamination with nuclear wastes, largely Sr⁹⁰. Addition of this beta emitting isotope to the environment inhibited gonad formation in young medaka, Oryzias latipes (Hyodo-Taguchi and Egami 1970), and destroyed most oocytes in the marine goby, Chasmichthys glosus (Hyodo-Taguchi et al. 1971). In the Atlantic salmon (Salmo salar) the primary sex cells were depressed after the fish was placed in water containing Sr⁹⁰ (Migalovskii 1971). Retarded sexual development also resulted from holding guppies in tritiated water (Erickson 1971). In the above laboratory studies the investigators did not indicate whether sterility was permanent.

Placing fish in isotope solutions or injecting them with radioactive material is not an acceptable sterilization method because of hazards to human health and the environment. Even though sterilized grass carp might be introduced only for weed control, there is no assurance against some being caught and consumed, or of other food chain contamination; therefore radiation must be applied from an external source.

Gamma Ray Sources

Two radioactive elements, Co^{60} and Cs^{137} , emit gamma rays that can be directed into fish to cause radiation damage without contamination. The use of these sources might be a convenient way to sterilize grass carp.

Of the two gamma sources, Co^{60} is the most commonly used to study effects of ionizing radiation. The best and most pertinent study relative to grass carp was done by Cherfas (1962) on carp (<u>Cyprinus carpio</u>). She observed that gonads were abnormal in carp that received 1 kR 2 to 3 years earlier, when they were larvae. Half of the 2-year-olds and 10 percent of the 3-yearolds were completely sterile. Lower doses of 50 to 300 R of gamma rays caused a less lasting effect. These findings on carp are perhaps more applicable to grass carp because of the similar life span, size, and the close taxonomic relationship of the two species.

Gamma rays from radiocobalt produced temporary sterility in male bluegills (Lepomis macrochirus) and stopped development of new oocytes in females at a dose of 1 kR (Ulrikson 1969). Similar treatment of rainbow trout (Salmo gairdneri) depressed sexual maturation (Tashiro 1972). Gamma radiation from Cs^{137} at 0.5 to 1 R produced temporary sterility in Oryzias latipes (Michibata 1976). High levels of exposure given to the guppy by Purdom and Woodhead (1973) caused sterility in almost all fish receiving 5 to 8 kR of gamma radiation from Co^{60} . Fecundity was reduced about 50 percent in fish retaining their fertility--mainly those that received 4 kR. In chinook salmon (Oncorhynchus tshawytscha) 0.4 kR from Co^{60} had no significant effect on the gonads, whereas 0.8 kR depressed gonadal development in the fry (Bonham and Donaldson 1972).

X Rays

Radiations emanating from a cathode tube (X rays) are similar to gamma rays. Powerful X rays have been available for a long time and effects on the reproductive system have been studied. Most people are aware that X rays cause sterility but the effects on the reproductive system of fish are not as well known (reviewed by Golovinskaya and Romashov 1958, Romashov and Golovinskaya 1960).

One of the earliest radiation studies was performed on the guppy. Application of 0.7 to 3.5 kR caused testicular degeneration (Natali 1940), a finding which was later confirmed in a more detailed study by Kobayashi and Yamamoto (1971). The most extensively studied fish was the medaka. Fish treated with 2 kR of X rays produced no fertile eggs (Egami and Hyodo 1965, Konno and Egami 1966), and most individuals were completely sterile (Egami and Hodo-Taguchi 1969). Irradiation destroyed the youngest oocytes in the ovary but oocytes already formed were less sensitive (Srivastava 1966), because radiation affects interphase in meiosis more than it affects cells in later stages of development (Hamaguchi and Egami 1975, Hamaguchi 1976). The maturation of the testes in the male was similarly inhibited by x-radiation (Hyodo-Taguchi and Egami 1974); also, there is differential sensitivity of the various stages of oogenesis (Hyodo-Taguchi and Egami 1976). Of relevance to grass carp are studies on the radiation effects in other cyprinids. One of the first radiation studies showed that X rays adversely affected the gonads of the goldfish, <u>Carassius auratus</u> (Samokhvalova 1938, confirmed by Etoh 1964). But again, the most useful study was one by Cherfas (1962) on the carp. Forty percent of the fish exposed to 0.6 kR as fingerlings were sterile at 3 and 4 years of age, and all had abnormal gonads. At 5 years (3 years after irradiation), however, only 10 percent were still sterile. This study suggests that sterilization may not be practical for grass carp because sterility is apparently not permanent, and growth is retarded (discussed later).

The pink salmon (<u>Oncorphynchus gorbuscha</u>) irradiated with 0.35 kR of X rays showed an initial response, but after 7 months gametogenesis again became active (Persov 1975). Males were more severely affected than females, and some were sterile. Similar findings were reported earlier by Kobayashi and Mogami (1958) for fingerling rainbow trout given 0.5 kR of X rays; ovarian size was reduced for several months. Adult rainbow trout, irradiated 3 months before spawning, developed ripe eggs but significant mortalities occurred in the developing embryos and fry (Foster et al. 1949).

In most studies, irradiation doses of 1 kR of X rays or gamma rays were used. Higher doses gave about the same results. Irradiation with 1 to 16 kR of X rays caused ovarian damage in the loach, <u>Misgurnus anguillicaudatus</u> (Egami and Aoki 1966). Tilapia were sterilized with 32 kR of gamma rays (A1 Daham 1970) but not by 3.3 kR (Nelson et al. 1976). Irradiation with 0.5 to 4 kR destroyed oocytes in the marine goby (Hyodo-Taguchi et al. 1970) and a similar range of doses destroyed spermatocytes and spermatozoa in the male (Hyodo-Taguchi and Egami 1971). The irradiation of both parents in <u>Fundulus</u> heteroclitus resulted in death of all progeny (Rugh and Clugston 1965).

Recovery of Fertility

Because sterilization with ionizing radiation is often temporary, its practicality is limited. The guppy recovered spermatogeneis 4 months after irradiation with 0.7 to 3.5 kR (Natali 1940). The ovaries of rainbow trout irradiated with 0.1 R recovered in 166 days, and irradiation with 0.5 R was effective for only 273 to 315 days (Kobayashi and Mogami 1958). Radiosterilization of tilapia with 32 kR was successful, but at lower doses the gonads were suppressed for only 128 to 153 days (A1-Daham 1970). Gonads of medaka irradiated with 2000 R did not differ from controls 20 days after treatment (Solberg 1938), presumably because proliferation of cells from the spermatogonia repopulate the succeeding cells types (Michibata 1976). In the female medaka, irradiation at first prevented germ cell proliferation but repopulation occurred 12 to 20 days after hatching (Hamaguchi 1976). In carp, irradiation in doses up to 1 kR sterilized some individuals but almost all developed functional gonads 1 to 2 years after the control fish had become mature (Cherfas 1962). Production of mature sperm resumed 120 days after sterility was induced in bluegills receiving 1 kR (Ulrikson 1969). In the marine goby, the ovary was restored to normal function in only 24 days after doses up to 4 kR Taguchi et al. 1970) and spermatogonia began proliferation at 19 to 24 days (Hyodo-Taguchi and Egami 1971). Repair systems are more effective during early embryogenesis than during oogenesis (Pechkurenkov 1976).

If sterilization is to be permanent, high doses of radiation must be used. For grass carp the minimum dose and best time for irradiation are not known.

Side Effects of Irradiation

Doses that effectively sterilize fish cause deleterious effects (Egami and Hyodo 1965). The most serious effects are gene mutations and chromosomal aberrations, which in turn cause mortality and reduced vigor. If sterilization of grass carp were successful, it could reduce survival or the effectiveness of the fish in controlling vegetation.

Very high doses of irradiation cause acute radiation disease and prompt death, whereas the levels needed for sterilization act more subtly. Fingerling chinook salmon had significant delayed mortality after receiving 2.5 kR (Bonham et al. 1948). In medaka, delayed mortality and reduced life span were observed at doses between 0.1 and 1 kR (Egami and Etoh 1973).

Irradiation may cause changes that reduce survival potential. A reduction in white blood cells after irradiation was noted in Atlantic salmon (Kosheleva 1971) and rainbow trout (Watson et al. 1959, Nakatani and Foster 1963, Strand et al. 1977). After irradiation the susceptibility of goldfish to disease caused by <u>Aeromonas salmonicida</u> increased (Shechmeister et al. 1962, Watson et al. 1963). Irradiated fish might be less resistant to any disease because of the depression of the immune system.

Irradiation may cause subtle changes in behavior that might not be noticed by the investigator but which could reduce survival. During x-irradiation with 7 to 18 kR, golden shiners (<u>Notemigonus crysoleucas</u>) became more active (Scarborough and Addison 1962) and x-irradiation of the larvae of the cichlid <u>Aequidens portalegrensis</u> produced a noticeable increase in activity (O'Brien and Fujihara 1963). After irradiation with tritium, the larvae of two marine fishes lay almost motionless on the bottom (Ichikawa and Suyama 1974). A cobalt-60 tag implanted on rainbow trout reduced activity levels in winter but increased it in spring and fall (Lichtenheld 1967). Irradiation reduced aggressive behavior in <u>Cichlasoma nigrofasciatum</u> (Schroder 1973, Holtsberg 1973). Obviously, more data are needed before definite conclusions can be drawn about long-term effects of irradiation on behavior.

The study by Cherfas (1962) is probably most germane to grass carp sterilization. Irradiation did not appear to reduce the survival of carp but growth was depressed about 20 percent by gamma irradiation of fingerlings. Older fish were not affected. Viability and growth might have been more severely affected by doses that would produce permanent sterility. The growth of bluegills (Ulrikson 1969) and chinook salmon (Welander et al. 1948, Bonham et al. 1948) was also reduced after irradiation. Growth was also reduced in rainbow trout fed radioactive substances (Watson et al. 1959, Nakatani and Foster 1963) or given x-irradiation (Welander 1954). In Atlantic salmon embryos, low doses of x-irradiation increased metabolic activity of Atlantic salmon embryos (Neustroev and Podymakhin 1966), whereas higher doses depressed respiration of loach embryos (Nikolskaya and Grudnitskii 1970). Although these studies were limited to acut effects on embryos, alterations in metabolism or inhibition of cell reproduction may account for depressed growth.

Irradiation-Induced Sex Reversal

One result caused by irradiation is the nearly complete destruction of the gonads. The gonads regenerate however, and again become functional. The process of regeneration may involve re-differentiation, in which the normal sex determining mechanism is greatly weakened. Often the regenerated gonad is of the opposite sex. Radiation sex reversal was first mentioned by Natali (1942a, 1942b) for poeciliids in which the ovary took on the appearance of a testis. Females of Xiphophorus were induced to form a gonadopodium by x-rays although there was little histological change in the ovary (Vivien 1950). After the treatment of young guppies with tritiated water, the proportion of males increased (Erickson 1971). In the bleak (Alburnus alburnus) females were transformed into males after exposure to low levels of uranium for 400 days (Stroganov and Telitchenko 1958). In irradiated chinook salmon, ovaries resembled poorly developed testes (Bonham and Donaldson 1972). Functional sex reversal occurred in Platypoecilus maculatus irradiated with X rays, but in the opposite direction: genetic males became females (Anders et al. 1969). A breeding test showed that this sex reversal was physiological rather than caused by genetic mutation. Hermaphrodites also may be produced (Egami and Hyodo-Taguchi 1969).

Safety and Regulation for Radiation Use

Radioactive materials and x-ray machines can cause serious health hazards, such as acute symptoms of leucopenia, hemorrhage, loss of hair and vomiting. Chronix exposure to low levels of radiation has been implicated in cancer. The routine use of X rays of CO^{60} needed for a program of sterilization should be directly supervised by someone trained in health physics.

If the radiation source were a radioactive isotope, such as cobalt 60, licensing by the U. S. Atomic Energy Commission is needed. For such licensing the operator must have specific training and experience in the use of radioactive material (Anon. 1967). Furthermore, adequate testing and monitoring equipment is required.

Evaluation of Radiosterilization

Sterilization of grass carp with X rays or CO^{60} is feasible. A radiation dose must be found that maximizes sterility and minimizes side effects. Tests are needed to establish the proper dose, age and conditions under which it can be most effectively applied. However, radiosterilization is not a practical procedure for absolute protection against reproduction, because of the possibility of gonad regeneration.

206

CHEMOSTERILIZATION

Rationale

Functioning of the reproductive system depends on cellular synthesis of proteins and nucleic acids, mobilization of raw materials for gamete formation, and stimulation and coordination by several hormones of the endocrine system. Pharmaceuticals that interfere with any of these biochemical or hormonal processes effectively block successful reproduction. Chemicals that block reproduction in fish are classified as follows:

- 1. Mutagens that modify DNA in the chromosomes and have effects similar to radiation.
- 2. Cytotoxins that kill the gametogenic cells or the gametes.
- 3. Hormone antagonists which block the normal action of hormones, disrupting coordination of the endocrine system.
- 4. Hormones when given in excess quantities or at critical times during the life cycle may upset normal sex differentiation and leave the fish sterile.
- 5. Mammaliam antisera formed in response to exposure to gamete proteins may cause sterility when injected into fish.

Mutagens

Little work has been done with chemical mutagens in fish. Three mutagens (1, 4 bis-diazoacetyl butane, dimethyl sulfate, and N-nitrosoethyl urea) were effective in carp for producing chromosomal aberrations, gene mutations, and morphological changes (Tsoi et al. 1974). Although survival was adequate, sterility was not produced. Sperm from rainbow trout and <u>Coregonus peled</u> treated with dimethyl sulfate resulted in embryos and fingerlings with many genetic defects (Tsoi 1969, 1972). Although these compounds have potential, sterilization of fish has not been accomplished. Chemical mutagens would be no more effective than irradiation.

Cytotoxins

Several chemicals have been extensively studied as sterilants for insects. The different agents that have chemosterilant activity probably do not have a common mode of action (Borkovec 1974). One possible mechanism of action is the induction of lethal mutations (cytotoxins that cause lethal mutations could be considered mutagens).

Chemosterilants have significant effects on fish. The size of the gonads of the killifish <u>Fundulus majalis</u> was somewhat reduced after the fish were treated with 10 to 1000 mg/liter of apholate (Eisler 1966).

Apholate also caused weight loss. Another chemosterilant, tris (1-aziridiny1) phosphine oxide, abbreviated TEPA, inhibited reproduction in the guppy (Stock and Cope 1969). The testes were reduced and the sperm produced was probably inactive. A temporary reduction in brood production resulted, although TEPA caused no apparent histological changes in the ovary. In tilapia, of two chemosterilants tested by Al-Daham (1970), metapa caused severe testicular atrophy and tretamine resulted in inviable sperm. Another sterilant with similar structure to the four above, P, P-Bis (1-aziridiny1)-N-methy1-phosphinothioic amide (bisazir), sterilized both sexes of the sea lamprey (Petromyzon marinus) at doses of 100 mg/kg (Hanson and Manio 1978). Eggs and sperm were produced and embryos developed, but all died before hatching.

Pesticides

A well known effect of chlorinated hydrocarbon pesticides is the inhibition of reproduction. The failure of reproduction in fish after exposure to sublethal doses of DDT is well documented (Burdick et al. 1964; Cuerrier et al. 1967, Macek 1968). Use of a persistent pesticide for the treatment of grass carp is not feasible, however, chiefly because it could not be registered for such use.

Parathion, and organophosphorous insecticide, temporarily sterilize the guppy (Billard and de Kinkelin 1970). Brief exposure had no effect, but spermatogenesis was suppressed in fish held for 40 days in water containing 0.1 to 10 mg/liter of the insecticide. Spermatogenesis was restored 34 days after cessation of treatment. Because the effects were temporary, parathion would not be a practical compound for use in managing grass carp.

Hormone Antagonists

Several chemicals are analogs or agonists of natural hormones. Methallibure (I.C.I. 33,838) was first used in fish to block production of sperm and eggs in the goldfish and the stickleback <u>Gasterosteus aculeatus</u> (Hoar et al. 1967). It had the same effects in the guppy (Martin and Bromage 1970, Billard et al. 1970, Pandey 1970, Pandey and Leatherland 1970) and molly <u>Poecilia</u> <u>latipinna</u> (Van Den Hurk and Van de Kant 1975, Van Den Hurk and Testerink 1975). Male and female gamete production was blocked in two marine species, the sea perch, <u>Cymatogaster aggregata</u> (Wiebe 1969) and the firetail gudgeon, <u>Hypseleostris galii</u> (MacKay 1973). Although methallibure has not been used to curb reproduction under other than laboratory conditions, it is effective in two species that commonly overreproduce--tilapia (Hyder 1972) and pumpkinseed, Lepomis gibbosus (Kramer 1972).

The hormone agonist, cyproterone acetate, decreased the uptake of testosterone by the testes in rainbow trout (Schreck 1973). This chemical fed to medaka at 0.5 mg/g of food resulted in a sex ratio of 15 females to 5 males (Irons and Schreck 1974). Several other inhibitors are available but have not been tested in fish. One interferes with the releasing factor produced by the hypothalamus that stimulates the release of gonadotropin hormones by the pituitary (de la Cruz et al. 1967). Hormone agonists are not likely to be effective in grass carp, however, becasue the effects are temporary.

Hormone Induced Gonadal Degeneration

Treatment of fish with natural or synthetic hormones (usually androgens) either early in the life cycle or at high doses may cause gonads to degenerate or fail to develop. Such treatments produce consistent effects in some fish. In juvenile coho salmon (Oncorphynchus kisutch) the addition of methyltestosterone to the diet caused an initial hypertrophy followed by degenerative changes in the testes, but had no effect on the ovary (Fagerlund and McBride 1975). Methyltestosterone caused degeneration of the testes and suppression of the ovaries in chum salmon (Oncorphynchus keta) and pink salmon (Yamazaki In rainbow trout, 4-chlorotestosterone caused the testes to degenerate 1972). and the ovaries to become smaller (Hirose and Hibiya 1968b), whereas methyltestosterone induced complete sterility that lasted for 2 years (Yamazaki 1976). Some of the rainbow trout similarly treated by Jalabert et al. (1975) and Johnstone et al. (1978) were sterile. Johnstone et al. (1978) sterilized some Atlantic salmon by treating embryos and fry with methylestosterone added to the water and feed. These studies suggest that treatment of salmonids with androgens may be a feasible method for inhibiting sexual development.

Consistent results were not obtained by androgen treatment in a number of other fishes. They synthetic androgen, 4-chlorotestosterone acetate caused degeneration of the testis and suppression of the ovary in the goldfish (Hirose and Hibiya 1968a). Another androgen, ethinyltestosterone caused ovarian inhibition in the guppy (Eversole 1941). Methyltestosterone, the androgen most frequently used, often caused sterility in the guppy (Takahashi 1975e). When given to the guppy at the proper time this androgen arrested ovarian development and elicited its differentiation into a testis (Dzwillo 1965, Clemens et al. 1966) and sterility was observed in some individuals. Injections of this androgen sterilized some, but not all wrasse, Halichoeres poecilopterus (Okada 1964). The response to exogenous androgen depends on the dose. Methyltestosterone fed to the medaka at doses less than 25 µg of feed resulted in small sterile ovaries; at doses of about 250 µg both sexes were castrated; and at intermediate doses, females became functional males (Yamamoto 1958). Oogonia were present in the gonad of the medaka before they were fed methyltestosterone; the ovaries degenerated and then grew into perfect testes (Kawamoto 1973).

Several studies have been conducted on cichlids. In <u>Tilapia aurea</u> the addition of methyltestosterone to the water caused involution of the gonads in some individuals (Eckstein and Spira 1965) and its addition to feed caused gonad degeneration (Guerrero 1975a). Jalabert et al. (1974) reported that methyltestosterone sterilized <u>Tilapia macrochir</u> but masculinized <u>T. nilotica</u>. In the paradise fish (<u>Macropodus opercularis</u>), methyltestosterone blocked gonad formation of all fish at some doses (Vanyakina 1972). Relatively small doses of adrenosterone caused the degeneration of all ovaries and most testes in <u>Tilapia nilotica</u> (Katz et al. 1976). In addition to these studies numerous attempts have been made to reverse the sex of fish by using exogenous androgens. There is, however, no clear pattern of androgen action in fishes.

Androgens have been fed to grass carp in attempts to reverse sex, but not to cause sterility (discussed later). The feeding of methyltesterone had no effect (Stanley and Thomas 1978)., whereas implanation of the androgen produced some degenerative effects (Jensen et al. 1978).

The effects of estrogen on fish have been extensively studied, and a depressing effect on the gonad has occasionally been reported. The artificial estrogen, stilbestrol, inhibited the testes and stimulated formation of ovotestes in the guppy (Berkowitz 1941). Estrodiol had a lesser effect. The same two estrogens had similar effects in the medaka, besides the production of many sterile individuals (Yamamoto and Matsuda 1963). Estrodiol inhibited oocyte development in <u>Gambusia</u> (Ishii 1961). Stilbestrol caused complete involution of the gonads of <u>Tilapia aurea</u> (Eckstein and Spira 1965). Estrogen treatment suppressed spermatogenesis and caused destruction of testicular lobules in <u>Monopterus</u> albus (Tang et al. 1974a).

Immunological Techniques

Fish can be sterilized by antisera produced by mammals that have been injected with fish reproductive products. Development of such technology is progressing in the control of human fertility and could prove to be useful in fish sterilization. However, little work has been done on fish. In perch, an extract from the testes distinctly interacted with extracts from the ovaries (Kothbauer and Schenkel-Brunner 1974). A specific substance was present in the gonads that could be used to prepare antibodies. Sea lampreys injected with antisera from rabbits that had received injections of gamete or gonad preparations yielded eggs and sperm which in some tests reduced embryonic survival (John H. Howell, Hammond Bay Biological Station, personal communication).

Safety and Registration

The safety of chemicals used to effect changes in sex or growth depends on the identity of the chemical, the amounts used, and the time elapsed after treatment before the fish are consumed. Administration of low levels of hormonal substances to young fish long before they might be caught and eaten is not likely to pose health hazards. Continuous feeding until the time of release for weed control, however, could result in residue levels that exceeded established tolerance levels.

Use of chemicals in fisheries in the United States requires registration with either the U. S. Food and Drug Administration or the U. S. Environmental Protection Agency (Cumming 1975). The efficacy of the candidate chemical must be proved and its breakdown products must be identified. The chemical and its metabolities must be tested for toxicity and carcinogenicity. Residue levels in the tissues must be identifiable. The collection of the background information needed for registration generally costs about \$2 million.

The costs of registering a chemical for fishery use can be reduced in two ways. If the chemical is widely used for other purposes most of the required data may already exist; if the chemical is used in minute amounts, long before the fish becomes large enough for human consumption, some of the requirements might be waived. A stronger case could be made if the agent were

210

a naturally occurring compound. Extensive data are available on diethylstibestrol, but it is a suspected carcinogen and is no longer approved for use as feed additive or for treatment in water. As an example of the difficulty in successfully registering a compound, not a single chemosterilant has been approved for use in controlling insects (A. B. Borkovec, personal communication) despite extensive testing.

Evaluation of Chemosterilants

Sterilization of grass carp with chemosterilants is not likely to be effective because the action of chemicals on the gonads of fish is usually temporary. After withdrawal, the gonads regenerate (sometimes as the opposite sex). In a fish with a long life cycle, such as grass carp, fertility will probably return and the effort will have been fruitless. Techniques in which chemical agents are used are not likely to be practical because of the high cost of collecting data for regisration. For this reason alone, chemosterilants or synthetic hormones are not recommended for use in the production of sterilized grass carp.

BREEDING FOR NON-REPRODUCTION

Rationale

Sterility in fish has been observed in triploids and in hybrids between related species. Breeding to produce polyploid animals that would be incapable of forming normal gametes is the most likely development for managing grass carp. Hybridization requires crossing grass carp with a species with dissimilar chromosomes or different numbers of chromosomes. The hybrid is sterile because synapse between homologous chromosomes is not possible; the resulting gametes are abnormal. The difficulty is that sterility results only if the parental species are not closely related, and most offspring of such crosses die.

Triploid Sterility

Fish with three sets of chromosomes are expected to be sterile because homologous chromosomes would be unable to synapse in gametogenesis. Triploidy has been induced in the stickleback (Swarup 1956, 1959a, 1959b), the sturgeon (Vasetskii 1967) and <u>Tilapia aurea</u> (Valenti 1975) by cooling the eggs immediately after fertilization. Such thermal shock also produced a triploid <u>Coregonus lavaretus</u> among 30 eggs (Svardson 1945). In the same study a single triploid was observed among 161 cold shocked eggs of an Atlantic salmon X brown trout hybrid. Cold shock has also produced triploidy in hybrids of plaice X flounder (Purdom 1972). Polyploids were induced in Atlantic salmon (<u>Salmo salar</u>) with the chemical, cytochalasin B (Refstie et al. 1977), but not by cold shocks (Lincoln et al. 1974).

The possibility of sterility was reported in a few studies of triploids. The ovaries of the triploid hybrid plaice X flounder contained few oocytes, compared with those of the diploid (Purdom 1972) and at sexual maturity (age IV) the gonadal volume was only 10% of that of controls (Purdom 1976). Triploid hybrids between Poecilia formosa and P. sphenops were sterile (Schultz and Kallman 1968). A single triploid of the California roach (<u>Hesperoleucas</u> <u>symmetricus</u>) was an immature fish, and was difficult to determine whether the small gonads were due to sterility or immaturity (Gold and Avise 1976). Sterility in the brook trout (<u>Salvelinus fontinalis</u>) was the result of polyploidy (Allen and Stanley 1978).

There is another possible explanation of how triploid fish could reproduce. In theory a triploid individual cannot form gametes, but there are many examples of triploid fish that do successfully reproduce. These triploid fish are naturally occurring species that consist only of female individuals and that reproduce by gynogenesis, a process in which the spermatozoan does not contribute genetic material. These triploid fish include several forms of <u>Carassius auratus</u> in Japan, and on the Aurasian continent (Astanin and Podgornii 1968, Kobayashi 1976, among many other references). In North America several forms of <u>Poeciliopsis</u> are triploids (Schultz 1967, Vrijenhoek and Schultz 1974). The block to gamete formation has been overcome in two ways. In <u>Carassius auratus</u> only one of the two divisions in meiosis occurs (Cherfas 1966) and in <u>Poeciliopsis</u> sp. an extra mitosis, but without cell cleavage, preceeds meiosis to give hexaploidy (Cimino 1972).

If artificially triploid fish could be produced, it is not likely that they would spontaneously reproduce. No doubt triploidy has arisen many times in nature and in only a few cases did a mechanism evolve that allowed for propagation.

Most methods for inducing triploidy have not resulted in all offspring being triploid. Even if half the progeny became triploid, as in some of the tests by Vasetskii (1967), the procedure would not be practical for grass carp. The remaining normal fish would be full fertile and difficult to separate when at the size the species is usually stocked. Fortunately there are two methods that could generate all triploids.

One procedure for obtaining triploidy in 100% of the offspring is crossing a diploid with a tetraploid; the diploid parent produces the usual haploid gamete the tetraploid parent a diploid gamete. The two gametes combined make a triploid. The difficult part of this procedure is the production of the tetraploid parent. Purdom (1972) was not successful in inducing tetraploids in hybrids of plaice and flounder. A few tetraploid progeny were found in chilled eggs of <u>Tilapia aurea</u> (Valenti 1975). In Atlantic salmon Refstie et al. (1977) found many tetraploid cells in larvae hatched from eggs treated with cytochalasin B. If germ cells consisted of tetraploids the gametes would indeed be diploid. I was unable to locate other references on induced tetraploidy, although tetraploid individuals and cells are not uncommon in natural populations of fish (F. L. Roberts, personal communication).

The second method used to develop polyploids is to treat eggs with cytochalasin B, as was done in the study by Refstie et al. (1977). Allen and Stanley (unpublished) found that most fish hatches from Atlantic salmon

eggs treated with cytochalasin B were polyploid mosaics. The cellular composition was similar to that found in sterile brook trout (Allen and Stanley 1978). Obviously, if this method were tried on grass carp, research would be needed on dosages and time of treatment. In my opinion, the induction of triploid sterility is the most feasible way to produce sterile grass carp.

Hybrid Sterility

Hybridization of animals often results in sterility because the dissimilar chromosomes of the hybrid are unable to synapse in the meiosis necessary for reproduction. There are examples of sterility in fish. Most intergeneric hybrids in salmonids e.g. between <u>Salvelinus</u>, <u>Salmo</u>, and <u>Oncorhynchus</u>, are sterile and survival is low (Suzuki 1974). Even within a genus, hybrids between Atlantic salmon and brown trout may have reduced fertility. In Sweden, this hybrid was almost completely sterile, and the progeny produced died shortly after hatching (Svardson 1945). In Ireland, crosses of sea-run Atlantic salmon with sea-run brown trout yielded hybrids that were fully fertile (Piggins 1965a, 1965b). The cross between landlocked Atlantic salmon and brown trout gave abnormal embryos if brown trout eggs were used and no embryos if salmon eggs were used (Buss and Wright 1956). A table given by MacCrimmon et al. (1974) tabulates fertility and infertility of barious crosses within the family Salmonidae.

Many sterile hybrids also may be polyploid, and it is not clear which condition causes sterility. Triploid sterile hybrids were reported in the cross between <u>Poecilia formosa</u> and <u>P. sphenops</u> (Schultz and Kallman 1968) and between plaice, <u>Pleuronectes platessa</u>, and flounder, <u>Platichthys flesus</u> (Purdom 1972, Purdom et al. 1976). Also sterile hybrids between rainbow and trout and brook trout (Buss and Wright 1957) were triploid (Capanna et al. 1974).

The reason polyploid hybrid sterility is especially revelant to grass carp is that hybrids of this species are usually polyploid (Vasilev et al. 1975, Stanley 1976a). It is unclear whether these hybrids are sterile (Stanley 1976b). The question is immaterial, however, because of low hybrid progeny viability. If eggs were derived from the grass carp, the hybrid progeny invariably perished (Makeeva and Sukhanova 1966). In the reciprocal hybrid (carp female), fry were more viable and usually began feeding but all died later (Kobayashi and Mizumoto 1950, Makeeva and Sukhanova 1966). Kuronuma (1955) and Makeeva (1968, 1969) reported that hybrids lived about a month. A few individuals have been reared to several months of age (Makeeva and Verigin 1974, Stanley 1976a). My hypothesis is that all ordinary diploid hybrids die and that the rare individuals, that by accident receive an extra set of chromosomes survive. However, Makeeva (1976) found some diploid hybrids along with triploids. Hybrid polyploids have no obvious use for managing aquatic vegetation. Hybrids between grass carp and silver carp (<u>Hypophthalmichthys molitrix</u>), bighead carp (<u>Aristichthys nobilis</u>), white amur bream (<u>Parabramis pekinensis</u>), and black amur bream (<u>Megalobrama terminalis</u>), have been produced (Makeeva and Sukhanova 1966). To my knowledge no attempt has been made to rear these hybrids to stockable sizes not to test them as weed control agents. Hybrid sterility has been reported in the Cyprinidae in crosses between carp and <u>Labeo rohita</u> (Alikunhi and Chaudhuri 1959), and between <u>Rhodeus ocellatus</u> Acheiolgnathus limbata (Suzuki 1965a).

Evaluation of Breeding Sterility

Sterile hybrids are not likely to be practical because of poor viability. Making sterile triploids is the best possibility for controlling sex in grass carp to ensure against any possibility of reproduction and naturalization. I recommend that research be initiated to breed for triploid sterility so that if further escape of grass carp to open systems occurs the individual will not reproduce.

Breeding for triploids requires two major research efforts. First a study must be conducted to show that triploids are viable and sterile. Second, tetraploid parents must be produced. Of all the possible ways to develop sterile fish, this is most feasible because it is cheapest, safest, and most likely to yield permanently sterile fish.

GYNOGENETIC MONOSEXES

Rationale

Artificial gynogenesis and also parthenogenesis are easily induced in fish, generally resulting in haploid embryos that have one set of chromosomes derived from the mother. However, some progeny may be diploids due to duplication of the maternal chromosomes. In many species of fish, including grass carp, sex is determined by chromosome inheritance, two x chromosomes yielding a female and an x and a y a male. Because gynogenetic offspring receive inheritance only from xx genotypic female, offspring are expected to be all females.

It might be possible to produce sufficient numbers of diploid gynogenetic grass carp for testing and perhaps for operational use. I must clearly state at the outset that such all-female fish are fully fertile and would be expected to reproduce if they spawned with escaped males. In many waters in the United States, the use of monosexes can no longer be recommended because males are known to be present (Pflieger 1978). In Florida, monosex fish are now being used in Lake Conway near Orlando (Theriot 1977).

Natural Gynogenesis

Several populations of fish are reported that reproduce by natural gynogenesis. These populations are all characterized by having only females, and with plastic mating habits that allow mating with a contiguous closely

related form. Some naturally gynogenetic fish are triploid, others diploid. Some are widespread and some restricted to a local environment. Gynogenesis as a mode of reproduction apparently has arisen many times in fish, amphibians, and reptiles. The adaptative advantages of gynogenesis is that the average fecundity per individual of the population is double that of populations with males.

Hubbs and Hubbs (1932), first discovered gynogenesis in fishes in the all-female <u>Poecilia</u> formosa. This species has been shown to be a hybrid of <u>P. latipinna</u> and <u>P. mexicana</u>, on the basis of morphological (Hubbs et al. 1959), and electrophoretic evidence (Abramoff et al. 1968). Hybrids of <u>P. latipinna</u> and <u>P. mexicana</u> are morphologically similar to <u>P. formosa</u> (Hubbs and Hubbs 1946). Another gynogenetic species was synthesized in the laboratory by Schultz (1973). Breeding is accomplished by mating with males of the progenitor sympatric species. The possibility of genetic recombination is greatly limited in fish that reproduce in this fashion, and hence gynogenetic populations consist of different clones (Kallman 1962). Artificial gynogenetic fish should also be more homozygous and could have reduced adaptability.

The naturally hynogenetic silver crusian carp (<u>Carassius auratus gibelio</u>) is widely distributed in Eurasia and mates with the males of several related species, such as carp and crucian carp (<u>Carassius carassius</u>). Goloyinskaya and Cherfas (1975) are of the opinion that the gynogenetic populations are of hybrid origin. <u>Carassius auratus</u> is probably one of the parental species; the other is unknown. Other species of naturally gynogenetic fish have been reported but the only well documented case is for Poeciliopsis (Schultz 1973).

Hybrid Gynogenesis

Gynogenesis may occur in crosses between dissimilar species that do not normally reproduce by this mode. This phenomenon was first reported by Roosevelt (1881) in an attempted cross of the striped bass (<u>Morone saxatilis</u>) and the American shad (<u>Alosa sapidissima</u>). There are several cases of gynogenesis among remote hybrids. Embryos from the cross of the pike perch (<u>Stixostedion lucioperca</u>) and the bream (<u>Abramis brama</u>) were identical with those of pike perch (Kryzhanovskii et al. 1953). Brook trout eggs fertilized with brown trout sperm apparently developed gynogenetically (Buss and Wright 1956). Gynogenesis was observed in eggs of black buffalo (<u>Ictiobus niger</u>) and bigmouth buffalo (<u>Ictiobus cyprinellus</u>) fertilized with sperm from goldfish (Stanley et al. 1975). Similar results were found in plaice eggs fertilized with flounder sperm but not in the reciprocal cross (Purdom and Lincoln 1974).

Hybrid gynogenesis also occurs in the family Cyprinidae, including grass carp. Makeeva (1968) and Alieva (1967) reported gynogenetic grass carp from eggs fertilized with carp milt. Silver carp eggs fertilized with carp sperm develop and produce larvae that appear to be silver carp and have silver carp isoensymes (Burlakov et al. 1973). Goldfish eggs developed gynogenetically after fertilization with sperm from white bass, <u>Morone</u> chrysops (Stanley et al. 1975). There are rare gynogentic bighead carp among the progeny of a cross between bighead carp and carp (Makeeva 1976). Apparently hybrid gynogenesis can be induced in many species by crossing forms that are not closely related, but the percentage yield is too meager for such gynogenesis to be practical for the production of monosex carp.

Radiation Gynogenesis

A more dependable way to obtain artificial gynogenesis is by irradiation of the sperm to completely denature the paternal chromosomes, but without destroying the flagella or microtubules that are the components responsible for activating development (Iwamatsu and Ohta 1974). Usually the resulting zygote is haploid and has limited ability for development and survival, as first reported by Oppermann (1913) for the brown trout and later by Neifakh (1959) for the weatherfish (Misgurnus fossilis).

Diploid gynogenesis was observed in eggs of sturgeon (<u>Acipenser</u> <u>guldenstadti</u>), sterlet (<u>Acipenser</u> <u>ruthenus</u>), and beluga (<u>Huso</u> <u>huso</u>) treated with x-irradiated sperm (Romashov et al. 1963). These studies wer extended to the weatherfish, in which Romashov and Belyaeva (1964), (1965) attempted to increase the percentage of diploids by thermal shocks of the eggs. Diploidy in gynogenetic carp induced by x-irradiated sperm was reported by Golovinskaya (1969) and Chervas (1975). Gamma irradiation also inactiviates DNA in sperm, and such treated sperm stimulated gynogenetic development in rainbow trout (Vassileva-Dryanovoska and Belcheva 1959) and in brown trout, plaice and flounder (Purdom 1969).

In 1972 a project was initiated at the Fish Farming Experimental Station, Stuttgart, Arkansas, to produce monosex grass carp by gynogenesis. In the initial efforts Stanley and Sneed (1974) used x-irradiation to denature spermatic DNA. These efforts were successful but x-radiation proved inconvenient because an x-ray unit was not available at the station. In all later efforts, ultraviolet (UV) light from an ordinary germicidal lamp was used. The first unit used delivered 1.0 m/W/cm^2 at the surface of the milt and required 15 to 60 minutes for irradiation (Stanley et al. 1975, Stanley 1976a, 1976b). In later studies a UV unit with a power of 6.5 m/W/cm^2 was used, which required less than 10 minutes to denature the spermatic DNA (Stanley 1976c). Although the percentage yield of grass carp was low, several thousand monosex grass carp were produced from the millions of eggs that were treated. During the 5 years of the study 58,177,000 grass carp eggs treated with irradiated milt from carp yielded 45,233 larvae (Thomas 1977).

If gynogenetic grass carp are to be useful for aquatic vegetation control there must be assurance that growth or performance is not depressed by a high degree of homozygosity that may accompany the diploid condition. Because chromosomes are derived only from the mother, homogeneity should be greater than in fish produced by normal fertilization. The degree of homozygosity is even greater because it is believed that diploidy is restored by circumvention of the second meiotic division (Romashov and Belyaeva 1965, Purdom and Lincoln 1973), thus producing a zygote with sets of sister chromosomes. All gene loci should be homozygous, except for chromosomal crossovers. Gynogenesis has been suggested as a method of producing highly homozygous fish (Purdom 1970, Stanley and Sneed 1974, Golovinskaya et al. 1977). Highly homozygous grass carp might have reduced survival and growth and thus have reduced effacacy in controlling vegetation relative to bisexual fish. Purdom and Lincoln (1973) found that gynogenetic plaice had reduced survival and Cherfas (1975) found the same in gynogenetic carp but that growth was about the same as normal fish. Romashov et al. (1963) reported reduced viability in gynogenetic sturgeons. Thomas and Carter (1977) stated that gynogenetic grass carp were expected to grow at about the same rate as normal fish. The high degree of homozygosity apparently is not realized because of a high rate of chromosonal crossovers of several loci (Purdom et al. 1976, Cherfas 1977).

Because gynogenetic grass carp are produced by stimulating egg development with carp sperm there is a possibility that some carp genes are inherited. If grass carp were not pure they could be less effective than normal fish in controlling weeds. Burlakov et al. (1973) noticed patroclinous inheritance in some embryos of presumed gynogenetic silver carp and Makeeva (1975) also reported inheritance of isozyme bands from the father in presumed bynogenetic bighead carp. Morphological (Stanley and Jones 1976) and biochemical parameters (Stanley et al. 1976) were examined and unquestionably gynogenetic grass carp are pure forms with no indication of inheritance from the male carp.

At the beginning I was not certain that gynogenetic grass carp would be all female. Some fish have sex determined by a different inheritance pattern. <u>Xiphophorus maculatus</u> from Mexico has female heterogamety, female wy, male yy (Gordon 1947), as do several species of tilapia: <u>Tilapia hornorum</u>, <u>T. aureau</u>, <u>T. macrochir</u>, <u>T. variabilis</u> and <u>T. leucostica</u> (Mires 1977). The same inheritance pattern probably determines sex in some sunfish, <u>Lepomis</u> sp. (Childers 1967). Gynogenesis in species with female heterogemety should give two kinds of progeny, males with a yy genotype and superfemales with a ww genotype. These females crossed with yy males would produce all female progeny.

Of 350 gynogenetic grass carp examined, all were females (Stanley and Thomas 1978). Gynogenetic carp are also all females (Cherfas 1975), except that one male was found among eight questionable individuals (Golovinskaya et al. 1977). Gynogenetic carp are completely capable of reproducing (Tsvetkova 1971, Golovinskaya et al. 1974), as are gynogentic grass carp (Stanley 1976d).

A limitation in the production of gynogenetic grass carp is the relatively low yields. Less than 1 percent of the eggs treated become diploid. A method is needed to increase the rate of diploidization. It is well known that thermal shock of eggs interferes with normal cell divisions of meiosis. Romashov and Belyaeva (1965) induced increased number of diploids in the weatherfish with cold and warm shocks. Cold shocks also promoted diploidization in flatfish (Purdom 1969, Purdom and Lincoln 1973). I used both warm and cold treatments in an attempt to improve production of diploid gynogenetic grass carp, but the treatments were essentially unsuccessful. Exposure of eggs to 5 or 10 C for 15, 30, or 45 min reduced the survival of gynogenetic fish (Stanley et al. 1975). In only one lot of eggs from one female was there a relatively large increase in diploids (16%) compared with the percentage in controls (2.8%) held constantly at 23 C. Milder cold treatment of 12, 14, 16, and 18 C has little effect on the production of diploids (Stanley 1976b).

Moderately high temperature was more consistent in improving diploidization in gynogenesis. Grass carp eggs were killed in 5 min by temperatures of 35 C but tolerated temperatures of 31 C for several minutes and 28 C for at least 45 min. In a series of four tests, grass carp eggs were exposed to 31 C for 1 min, beginning at 1, 2, 3, 4, 5, 6, 7, 8, 9, and 10 min after fertilization. In each of the 10 treatments, 1000 eggs were used. In three of these tests the number of diploids increased in larvae hatched from eggs exposed 2-3 or 3-4 min after fertilization. However, the yield was only 1.5 to 2.7 times greater than that in the controls. In a series of two tests the same time intervals were used but the temperature was 28 C. At this temperature the peak yield was in eggs shocked for 1 min beginning 5 min after fertilization. In one test the percentage was 0.7 in the controls, 1.8 at 4-5 min, and 2.7 at 5-6 min. The effective treatment time corresponded exactly to the anaphase-to-telephase period, during the expulsion of the seond polar body (Bobrova 1969). In a third test series in which thermal shocks were at 28 C, but for 3 min beginning a 0, 2 and 4 min, the yield of diploids was 0.9% in the controls, only 0.1%, in eggs shocked at minute 0-3, 0.4% at minute 2-5, and 4.7% (a 5-fold increase over the controls) at minute 4-8. Thus exposure to high temperatures offers the possibility for moderate increases in the yield of diploid gynogenetic grass carp.

The percentage of gynogenetic diploids varied greatly in lots of eggs from different females. In grass carp the yield ranged from 0 to about 4% of the eggs treated. Variation was similar in the weatherfish (Romashov and Belyaeva 1965) and carp (Cherfas 1975). Romashov and Belyaeva (1965) speculated that the difference in response was inherited in the weatherfish. Such inheritance almost surely holds true for carp because the percentage diploids was much higher in the second generation of artificial gynogenesis (Cherfas 1975). However, in grass carp Stanley (1976d) observed no difference between the two generations. If the tendency toward diplodization is heritable in grass carp it must be only weakly so.

Evaluation of Gynogenesis

Gynogenesis is an inefficient method for large scale production of monosex fish, but sufficient numbers could be produced to stock at experimental sites, such as Lake Conway in Florida. Monosex grass carp do not provide absolute security against naturalization because populations of male and female grass carp have already been widely distributed. The addition of monosex fish to the existing population in open water could cause populations to reach the critical levels believed necessary to elicit sexual maturation and spawning (Stanley et al. 1978). The disappointing yield of diploids in gynogenesis might be increased by treating eggs with cytochalasin B, and I recommend that such tests be conducted.

MONOSEX HYBRIDS

Rationale

In crosses between different species hybrids are sometimes largely of a single sex. This is because the parental species have different systems for determining sex. Crossing a male having a yy genotype with the female of a xx genotype will yield xy hybrids. One y sex chromosome determines maleness when paired with a x chromosome from the related species, but a female if paired with the w sex chromosome of its own species. Because the two alternate systems are not available in grass carp it is of academic interest only in this paper, and discussion will be brief.

All Male Hybrids

Crosses between the redear sunfish (Lepomis microlophus) and bluegill produced only male progeny (Childers 1967). In tilapia, crosses between any five males species and any of four female species resulted in all males (see review by Mires 1977). In Cyprinidae the cross between <u>Gnathopogon</u> japonicus and <u>Biwia zezera</u> gave only male progeny (Suzuki 1965b). In <u>Xiphophorus maculatus</u> both types of sex determining mechanisms exist within the same species and a cross between xx females and yy males resulted in all male progeny (Kallman 1965).

Hybrid monosexes are not known in crosses involving grass carp.

SEX REVERSAL

Rationale

The administration of sex hormones to juvenile fish can override the genetic determination of sex. The generally accepted hypothesis concerning the mechanism of action is that the exogenous sex hormones act as an inducer on the presumptive germinal tissue. It is supposed that treatment must coincide with the period of sex differentiation. However, in several experiments (some cited in earlier sections) the gonad was clearly differentiated but degenerated after treatment, and later differentiated into that of the opposite sex. This second pattern may be followed in natural sex reversal. In fish some cells may remain undifferentiated and these could differentiate during treatment.

To induce sex reversal in grass carp or other fish, appropriate sex hormones are administered during the time of sex differentiation to ensure complete functional induction of the desired sex. Less effective, and apparently not possible in some species, is sex transformation in individuals already having ovaries or testes. If a treatment was 100% effective, these fish could be used directly in management without danger of reproduction; however, often not all individuals are transformed and some members of the opposite sex remain available for mating.

To avoid the problem of incomplete sex reversal, a second generation must be produced in which a sex-reversed fish is mated to a normal individual to produce 100% monosex fish. For a species with female homogamety, such as the grass carp, androgens are administered to juveniles and the sex-reversed genotypic females (Phenotypic males) are then mated to ordinary females to produce all-female progeny. In species with male homogamety the juveniles are given estrogen and then sex-reversed males are mated to normal males to give all-male progeny. In both treatments test crosses are needed to distinguish the sex-reversed individuals in the test lots from the fish that would normally have been of that sex.

For grass carp 2 or 3 years are required to rear the fish from the test cross to a size where sexes can be distinguished. I propose using gynogenetic grass carp for androgen treatment, thus cirvumventing the need for the test cross. Any male found in a treatment would be a sex-reversed female (because without treatment all gynogenetic grass carp are females). The sex-reversed genotypic female mated with a normal female would sire all female progeny: $(xx \ X \ xx \ - xx)$.

Natural Sex Transformation

Sex transformation has frequently evolved and the occurrence and analyses of sex transformations were reviewed thoroughly by Atz (1964). Yamamoto (1969), Chan (1970), Reinboth (1970), and Warner (1975). The original literature was not consulted. Sex transformation occurs in 5 orders and 13 families of fish and is best studied in the Serranidae, Sparidae, and Labridae. The general pattern of development is that both male and female elements are present in every gonad and that first one tissue, and then the other, is active. Because this pattern of hermaphrodism evolved independently several times, it must have been derived from a basic pattern occurring in fish with the normal mode of sex expression. Thus natural transformations are relevant to artificial manipulations of sex expression.

Knowledge of the mechanisms of natural sex inversion suggest ways for artificially controlling sex. The difficulty is that steroidal sex hormones are the most effective agents for eliciting sex changes artificially whereas they were impotent in promoting natural reversal in the ricefield eel, <u>Monopterus albus</u> (Tang et al. 1974a) or in <u>Sparus auratus</u> (Colombo et al. 1972). Lutenizing hormone induced precocious sex reversal in the ricefield eel (Tang et al. 1974b), which is ineffective in artificial sex reversal. However, natural and artificial sex reversal share one characterisitic: the bipotential gonad.

Artificial Sex Reversal

Sex has been reversed in both directions in fish of several taxonomic groups, when an exogenous hormone was administered at the appropriate period in the life cycle. It is generally believed that hormones must be given during the period of sexual differentiation (Yamamoto 1969, Vanyakina 1969, Schreck, 1974).

Early efforts to reverse sex were inconsistent because treatment often began after sex differentiation (Yamamoto 1969). By treating early in life the sex of several fish were successful reversed. Males of medaka were induced to develop into females by adding estrogen to the diet: estrodial -17 beta, stilbestrol (Yamamoto and Matsuda 1963), estrone (Yamamoto 1959) and estriol (Yamamoto 1965). Females were induced to become males by adding androgens in the diet: methylestosterone (Yamamoto 1958), androstenedione (Yamamoto 1968), and testosterone propionate (Yamamoto et al. 1968). By combining various genetic crosses with hormone treatments every possible genotype and phenotype was produced, all of which were fully viable and grew about equally (Fineman et al. 1974).

In the guppy attempts to induce sex reversal failed until hormones were given before birth (Dzwillo 1965, Takahashi 1975a, 1975b). However, certain doses (25-50 ug/1) of 11-ketotestosterone caused complete reversal if fish were treated for 35 days, beginning at birth (Takahashi 1975c). Also, genetic males were feminized by adding ethinylestradiol to the diet fed for 30 days after birth (Takashi 1975d).

The sex-reversal technique has been used as a method for producing monosex fish for fish farming. Cichlids are an important group of pondfishes in tropical regions, but overpopulation causes stunting, and therefore reduced yields of harvestable-sized fish. The sex of <u>Tilapia mossambica</u> was reversed by giving methyltestosterone in the feed (Clemens and Inslee 1968, Nakamura 1975, Guerroro 1975a, 1976). This same androgen also effectively caused females of <u>T</u>. <u>nilotica</u> to differentiate as males but was ineffective in <u>T</u>. <u>macrochir</u> (Jalabert et al. 1974). Methyltestosterone and ethynyltestosterone, but not dehydrotestosterone, were effective in causing <u>T</u>. <u>aurea</u> with female genotype to differentiate as males (Guerrero 1975b). The sex of <u>T</u>. <u>zillii</u> was not reversed after oral administration of either methylestosterone (Guerrero 1975a) or ethynyltestosterone (Guerrero 1976). Thus the sex of some species can be readily reversed, whereas the sex of other related forms given the same dose under the same conditions was not affected.

Sex change in the opposite direction has also been elicited in cichlids. <u>Tilapia mossambica</u> was completely feminized by oral administration of ethinylestradiol (Nakamura and Takahshi 1973). Estradiol caused female production in <u>Hemihaplochromis</u> <u>multicolor</u> (Hackmann and Reinboth 1974). There is a paradoxal effect of androgen in causing feminization. In <u>T</u>. <u>mossambica</u> methyltestosterone and testosterone propanate caused feminization, although ovarian development was inhibited, whereas in <u>T</u>. <u>heudeloti</u> male characters were induced, even though a female gonad was retained (Hackmann 1974). In <u>Hemihaplochromis multicolor</u> these same two androgens caused some males to exhibit female characteristics (Hackmann and Reinboth 1974). This same peculiar effect was seen in <u>T</u>. <u>mossambica</u> given high doses of methyl-testosterone; males become females but females were not affected (Nakamura 1975).

Until recently, sex reversal was largely unsuccessful in salmonids. Treatment of brown trout with progesterone, estradiol, and testosterone failed to cause sex reversal (Ashyby 1957). In horai masu, a variety of rainbow trout, the feeding of methyltestosterone caused a preponderance of males, but in ordinary rainbow trout it caused complete sterility (Yamazaki 1976). Most hermaphrodites and sterile rainbow trout were found in rainbow trout given methyltestosterone at 1 to 6 months of age (Jalabert et al. 1975). The clue to success is to begin treatment very early, even before feeding begins, by immersing the eyed eggs in solutions (Johnstone et al. 1978). In rainbow trout 100% females were induced with a combination of immersion and feeding of estradiol. Hermaphrodites developed when treatment began at the start of feeding. In Atlantic salmon, Johnstone et al. (1978) found 100% females in all fish regardless of when estradiol was given. For the opposite reversal in rainbow trout, methyltestosterone induced mostly males but the sex of some females was not reversed, and some hermaphrodites were produced (Johnstone et al. 1978). Atlantic salmon were 100% males, regardless of whether treatment began before or after hatching.

Sex reversal in cyprinids was first mentioned by Natali and Natali (1947) in carp, accoridng to Vanyakina (1969). The sex of goldfish was reversed in both directions (Yamamoto and Kajishima 1968). By crossing the sex-reversed females with normal females high yields of all female broods were produced. Later studies produced all possible genotypes, proving that the yy genotype was fully viable (Yamamoto 1975). By crossing this yy supermale with a normal female, all-male progeny were produced.

Sex Reversal in Grass Carp

The system of mating sex-reversed with normal fish is proposed for mass production of monosex grass carp but in which sex-reversed gynogenetic grass carp are mated with normal females. The use of gynogenetic progeny ensures that all were of the female genotype.

Grass carp were treated with methyltestosterone by adding it to the diet at either 30 or 60 mg/kg of feed. The feed was ground for feeding to fry and fingerlings were fed unground crumbles. The length of treatment and the age at which treatment began and ended was varied in an attempt to deliver the androgen at the time of sex differentiation, which was not known. Eleven different treatment regimes were used, all totally without success; not a single male was observed (Stanley and Thomas 1978). All 350 grass carp were females. At least sex is stable in the grass carp; spontaneous sex reversal is unlikely.

None of the treatments began before 7 days of age nor extended beyond 20 weeks. Possibly the attempted sex reversal in grass carp did not include the critical period in which the gonad is sensitive to androgen. On the basis of the findings described above for salmonids I believe that treatment should have been started earlier by adding androgen to the aquarium water.

Jensen et al. (1978) failed to produce sex reversal in grass carp, using implantation of pellets containing methyltestosterone. However some fish had depressed gonadal tissue. At least these fish showed a response to the methyltestosterone treatment.

Evaluation of Sex Reversal

The sex reversal technique has great potential for producing monosex fish, although direct use of hormone-treated fish is not possible because the chemical inducing agents are not registered for fishery use. This problem is avoided by stocking the progeny of sex reversed fish.

Sex reversal of grass carp was not successful with two different techniques. Even if sex had been reversed the value of the method would have been small, since many grass carp have already escaped to open water and males are presumably available for mating.

SUMMARY

- 1. Radiosterilization is temporary, and therefore a poor choice for a a long-lived fish.
- 2. Chemosterilants have potential for preventing reproduction of fish but could never be registered for fishery use.
- 3. The breeding of triploid fish by crossing tetraploids with diploids offers the best way to prevent reproduction, but more research is needed.
- 4. Monosex grass carp produced by gynogenesis are available, but only in small numbers. Increased production is not likely.
- 5. Sex reversal to produce monosex grass carp was unsuccessful, but has good potential in other species.
- 6. Stocking monosex grass carp in waters where escape and mingling with bisexual populations is possible could add to the naturalization potential. Sterilized grass carp would be safer.

REFERENCES

- Abramoff, P., Darnell, R. M., and Balsano, J. S. 1968. Electroporetic Demonstration of the Hybrid Origin of the Gynogenetic Teleost Poecilia formosa, Am. Natur. 102(928): 555-558.
- Al-Daham, N. K. 1970. The Use of Chemosterilants, Sex Hormones, Radiation, and Hybridization for Controlling Reproduction in Tilapia species. Ph.D. Thesis, Auburn University, Auburn, Ala. 176 p.
- Aliev, D. C. 1967. Morphology of Young-of-the-year Hybrids of Phytophagous Fish. Vopr. Ikhtiol. 7(1): 191-194 (in Russian).
- Alikunhi, K. H., Chaudhuri, H. 1959. Preliminary Observations on Hybridization of the Common Carp (<u>Cyprinus carpio</u>) with Indian Carps. Proc. Indian Sci. Congr. 46.
- Allen, S. K., Stanley, J. G. 1978. Reproductive Sterility in Polyploid Brook Trout, <u>Salvelinus fontinalis</u>. Trans. Am. Fish Soc. 107(3): 474-478.
- Anders. A., Anders. F., Rase, S. 1969. SY Females Caused by X-Irradiation. Experientia 25(8): 871.
- Anon. 1967. How to Get a License to Use Radioisotopes. Atomic Energy Commission, U. S. Government Printing Office, Washington, D. C.
- Ashby, K. R. 1957. The Effect of Steroid Hormones on the Brown Trout (<u>Salmo trutta</u> L.) during the Priod of Gonadal Differentation. J. Embryol. Exp. Morphol. 5(3): 225-249.
- Astanin, L. P., Podgornii, M. I. 1968. Features of the Fertility of <u>Carassius carassius</u> (L). and <u>C. auratus gibelio</u> (Bloch). J. Ichthyol. 8(2): 209-214).
- Atz, J. W. 1964. Intersexuality in Fishes. In: C.N. Armstrong and A. J. Marshall (eds.), Intersexuality in Vertebrates Including Man, Academic Press, New York: 145-232.
- Berkowitz, P. 1941. The effects of Estrogenic Substances in the Fish (Lebistes reticulatus). J. Exp. Zool. 87(2): 233-243.
- Billard, R., de Kinkelin, P. 1970. Stérilisation des testicules de Guppies par des Doses non Léthales de Parathion. Ann. Hydrobiol, 1 (1): 91-99.
- Billard, R., Breton, B., Jalabert, B. 1970. Inhibition de la Spermatogeneses du Guppy (Poisson Cyprinodontidae) par le Méthallibure. Ann. Biol. Anim. Biochim. Biophys. 10: 511-515.
- Bobrova, Yu. P. 1969. On Development of the Gonads and the Process of Fertilization in the White Amur. In: Cherfas, B.I. (ed.), Genetics, Selection and Hybridization of Fish. Akad. Nauk. SSSR. Transl. N.T.I.S., Springfield, VA: 123-128.

- Bonham, K., Donaldson, L. R. 1972. Sex Ratios and Retardation of Gonadal Development in Chronically Gamma-Irradiated Chinook Salmon Smolts. Trans. Am. Fish. Soc. 101(3): 428-434.
- Bonham, K., Donaldson, L. R. Foster, R. F., Welander, A. D., Seymour, A. H. 1948. The Effect of X-rays on Mortality, Weight, Length, and Counts of Erythrocytes and Hematopoietic Cells in Fingerling Chinook Salmon, <u>Oncorhynchus tshawytscha</u> Walbaum. Growth 12: 107-121.
- Borkovec, A. B. 1974. Mechanism of Action of Alkylating and Nonalkylating Insect Chemosterilants. Am. Chem. Soc. Symp. Series 2, Mechanism of Pesticide Action: 130-135.
- Burdick, G. E. Harris, E. J., Dean, H. J., Walker, T. M., Skea, J., Colby, D. 1964. The Accumulation of DDT in Lake Trout and the Effect of Reproduction. Trans. Am. Fish. Soc. 93(2): 127-136.
- Burlakov, A. B., Makeeva, A. P., Rybarov, I. N. 1973. Isozyme Composition of Hybrid and Gynogenetic Forms of Some Cyprinids at Early Stages of Ontogenesis. In: Kirpichnikov, V. S., Troshin, A. S., Fridlyanskaya, I. I. (eds.), Biochemical Genetics of Fish. Institute of Cytology Academy of Sciences USSR, Leningrad: 85-90 (in Russian).
- Buss, K. W. Wright, J.E., Jr., 1956. The Results of Species Hybridization within the Family Salmonidae. Prog. Fish-Cult. 18(4): 149-158.
- Buss, K. W., Wright, J. E., Jr. 1957. Appearance and Fertility of Trout Hybrids. Trans. Am. Fish. Soc. 87: 172-181.
- Capanna, E., Cataudella, S., Volpe, R. 1974. An Intergeneric Hybrid between the Rainbow Trout and the Fresh-water Char (Salmo gairdneri X <u>Salvelinus fontinalis</u>). Boll. Pesca Piscic. Idriobiol. 29(2): 101-106.
- Chan, S. T. H. 1970. Natural Sex Reversal in Vertebrates. Phil. Trans. Roy. Soc. London B. 259: 59-71.
- Cherfas, N. B. 1962. Radiation Disease of the Gonads of Carp. Vopr. Ikhtiol. 2(1): 104-115 (in Russian).
- Cherfas, N. B. 1966. Meiotic Analysis of Unisexual and Bisexual Forms of Crucian Carp. Tr. Vses. Nauchno-Issled. Inst. Prud. Rybn. Khoz. 14: 63-82 (in Russian).
- Cherfas, N. B. 1975. Investigations of Radiation-induced Diploid Gynogenesis in the Carp: I. Experiments on Obtaining the Diploid Gynogenetic Progeny in Mass Quantities. Genetika 11(7): 78-86 (in Russian).

- Cherfas, N. B. 1977. Investigation of Radiation induced Diplois gynogensis in the Carp (Cyprinus carpio L.): II. Segregation with Respect to Certain Morphological Characters in Gynogenetic Progeny. Genetika 13(5): 811-820.
- Childers, W. F. 1967. Hybridization of Four Species of Sunfishes (Centrarchidae). Bull. Ill. Nat. Hist. Surv. 29(3): 159-214.
- Cimino, M. C. 1972. Meiosis in Triploid All-female Fish (Poeciliopsis, Poeciliidae). Science 175: 1484-1486.
- Clemens, H. P., Inslee, T. 1968. The Production of Unisexual Broods by <u>Tilapia mossambica</u> Sex-reversed with Methy Testosterone. Trans. Am. Fish. Soc. 97(1): 18-21.
- Clemens, H. P., McDermitt, C., Inslee, T. 1966. The Effects of Feeding Methyl Testosterone to Guppies for Sixty Days after Birth. Copeia 1966(2): 280-284.
- Colombo, L., Del Conte, F., Clemenze, P. 1972. Steriod Biosynthesis in vitro by the Gonads of <u>Sparus auratus</u> L. (Teleostei) at Different Stages during Natural Sex Reversal. Gen. Comp. Endocrinol. 19(10: 26-36.
- Cuerrier, J. P., Keith, J. S., Stone, E. 1967. Problems with DDT in Fish Culture Operations. Natural. Can. 94: 315-320.
- Cumming, K. B. 1975. History of Fish Toxicants in the United States. In: Eschmeyer P. H. (ed.), Rehabilitation of Fish Populations with Toxicants: A Symposium, North Central Division, Am. Fish. Soc.: 5-12
- de la Cruz, A., Coy, D. H., Vilchez-Martinez, J. A., Arimura, A., Schally, A. V. 1967. Blockage of Ovulation in Rats by Inhibitory Analogs of Luteinizing Hormone-releasing Hormone. Science 191 (4223): 195-197.
- Dzwillo, M. 1965. Uber den Einflub von Methyltestosteron auf primare und Sekundare Geschlechtsmerkmale wahrend vershiedener Phasen der Embryonalentwicklung von Legistes reticulatus. Verh. Dtsch. Zool. Ges. 41: 471-476.
- Eckstein, B., Spira, M. 1965. Effect of Sex Hormones on Gonadal Differentiation in a Cichlid, <u>Tilapia aurea</u>. Biol. Bull. 129: 482-489.
- Egami, N., Aoki, K. 1966. Effects of X-irradiation of a Part of the Body on the ovary in the Loach, <u>Misgurnus anguillicaudatus</u>. Annot. Zool. Jap. 39(1): 7-15.
- Egami, N., Etoch, H. 1973. Effect of X-irradiation During Embryonic Stage on Life Span in the Fish, <u>Oryzias latipes</u>. Exp. Gerontol. 8(4): 219-222.

- Egami, N., Hyodo, Y. 1965. Inhibitory Effect of X-irradiation on the Development of the Ovaries of the Fish, Oryzias latipes, in Sexually Inactive Seasons. Annot. Zool. Jap. 38: 8-11
- Egami, N., Hyodo-Taguchi, Y. 1969. Hemaphroditic Gonads Produced in <u>Oryzias latipes</u> by X-radiation during Embryonic Stages. Copeia 1969: 195-196.
- Eisler, R. 1966. Effects of Apholate, an Insect Sterilant, on an Estuarine Fish, Shrimp, and Gastropod. Prog. Fish-Cult. 28(3): 154-158.
- Erickson, R. C. 1971. Effects of Tritiated Water and Other Sources of Ionizing Radiation on Mortality, Growth, Male Characters, Courting Behavior and Thermal Death of <u>Poecilia reticulata</u>, the Guppy. Ph.D. Thesis, Univ. of Washington, Seattle. 174 p.
- Etoh, H. 1964. The Effects of Partial Irradiation of the Goldfish, <u>Carassius auratus</u> with a Massive Dose of ⁶⁰ Co²y-rays. Zool. Mag. Tokoyo. 73: 239-241.
- Eversole, W. J. 1941. The Effects of Pregeninolone and Related Steriods on Sexual Development in Fish (Lebistes reticulatus). Endocrinology 28: 603-610.
- Fagerlund, U. H. M., McBride, J. R. 1975. Growth Increments and Some Flesh and Gonad Characteristics of Juvenile Coho Salmon Receiving Diets Supplemented with 17 α-methytestosterone. J. Fish Biol. 7(3): 305-314.
- Fineman, R., Hamilton, J., Chase, G., Bolling, D. 1974. Length, Weight and Secondary Sex Character Development in Male and Female Phenotypes in three Sex Chromosomal Genotypes (XX, XY, YY) in the Killifish, <u>Oryzias latipes</u>, J. Exp. Zool. 189(2): 227-234.
- Foster, R. F., Donaldson, L. R. Welander, A. D., Bonham, K., Seymour,A. H. 1949. The Effect on Embryos and Young Rainbow Trout from Exposing the Parent Fish to X-rays. Growth 13: 119-142.
- Gold, J. R., Avise, J. C. 1976. Spontaneous Triploidy in the California Roach <u>Hesperoleucus</u> symmetricus (Pisces: Cyrpinidae). Cytogenet. Cell Genet. 17(3): 144-149.
- Golovinskaya, K. A. 1969. Artificial Gynogenesis in Carp. In: Cherfas, B. I., (ed.), Genetics, Selection, and Hybridization of Fish. Akademiya Nauk SSSR. Trans 1. N.T.I.S., Springfield, VA: 74-78.
- Golovinskaya, K. A. Cherfas, N. B. 1975. Fish Natural and Artificial Gynogenesis. Ichthyologia 7(1): 11-15.
- Golovinskaya, K. A., Romashov, D. D. 1958. Effect of Ionizing Radiation on the Development and Reproduction of Fishes. Vopr. Iktiol. 2(11): 16-38 (in Russian).

- Golovinskaya, K. A., Belyaeva, V. N., Cherfas, N. B. 1977. Artificial Gynogenesis in Fishes and Its Possible Use in Selection. In: Prob. Eksper. Biol., Nauka Press, Moscow: 100-114.
- Golovinskaya, K. A., Cherfas, N. B., Tsvetkova, L. I. 1974. The Results of Estimation of Reproductive Function of Female Carp of Gynogenetic Origin. Trud. Vses. Nauchno-Issled. Inst. Prud. Rybn. Khoz. 23: 20-26 (in Russian).
- Gordon, M. 1947. Genetics of <u>Platypoecilus maculatus</u> IV. The Sex Determining Mechanism in Two Wild Populations of Mexican Platyfish Genetics 32: 8-17.
- Guerrero, R. D. III. 1975a. Sex-reversal of Tilapia. FAO Aquaculture Bull. 7(3-4): 7.
- Guerrero, R. D. III. 1975b. Use of Androgens for the Production of Allmale <u>Tilapia</u> <u>aurea</u> (Steindachner). Trans. Am.Fish. Soc. 104(2): 342-348.
- Guerrero, R. D. III. 1976. <u>Tilapia mossambica</u> and <u>T</u>. <u>zilli</u> Treated with Ethynyltestosterone for Sex Reversal. Kalikasan (5(2): 187-192.
- Hackmann, E. 1974. Einfluss von Androgenen auf die Geschlechtsdifferenzierung Verschiedener Cichliden (Teleostei). Gen. Comp. Endocrinol. 24(1): 44-52.
- Hackmann, E., Reinboth, R. 1974. Delimitation of the Critical Stage of Hormone-influenced Sex Differentiation in <u>Hemihaplochromis</u> <u>multicolor</u> (Hilgendorf) (Cichlidae). Gen Comp. Endocrinol. <u>22: 42-53.</u>
- Hamaguchi, S. 1976. Changes in the Radiation Response of Oogonia in the Embryos and Fry of the Fish <u>Oryzias latipes</u>. Intern. J. Radiat. Biol. 29(6): 565-570.
- Hamaguchi, S., Egami, N. 1975. Post-irradiation Changes in Oocyte Populations in the Fry of the Fish <u>Oryzias latipes</u>. Intern. J. Radiat. Biol. 28(3): 279-284.
- Hanson, L. H., Manion, P. J. 1978. Chemosterilization of the Sea Lamprey (<u>Petromyzon marinus</u>). Tech. Rep. Great Lakes Fish. Comm. (in press).
- Hirose, K., Hibiya, T. 1968a. Physiological Studies on Growth-promoting Effect of Protein-anabolic Steriods on Fish. I. Effects on Goldfish. Bull. Jap. Soc. Sci. Fish. 34: 466-472.
- Hirose, K., Hibiya, T. 1968b. Physiological Studies on Growth-promoting Effects of Protein-anabolic Steroids on Fish. II. Effects of 4-chlorotestosterone Acetate on Rainbow Trout. Bull. Jap. Soc. Sci. Fish. 34: 473-478.

- Hoar, W. S., Wiebe, J., Wai, E. H. 1967. Inhibition of the Pituitary Gonadotropic Activity of Fishes by a Dithiocarbamoylhydrazine derivative (I.C.I. 33, 828). Gen Comp. Endocrinol. 8(1): 101-109.
- Holtzbert, S. 1973. Change in Agressive Readiness in Post-irradiation generations of the Convict Cichlid Fish, <u>Cichlasoma nigrofasciatum</u>. In: Schroder, J. H., (ed.), Genetics and Mutagenesis of Fish. Springer-Verlag, New York: 173-176.
- Hubbs, C. L., Hubbs, L. C. 1932. Apparent Partenogenesis in Nature, in a Form of Fish of Hybrid Origin. Science. 76: 628-630.
- Hubbs, C. L., Hubbs, L. C. 1946. Experimental Breeding of the Amazon Molly. Aquarium J. 17: 4-6.
- Hubbs, C. L. Drewry, G. W. Warburton, B. 1959. Occurrence and Morphology of a phenotypic Male of a Gynogenetic Fish. Science 129: 1227-1229.
- Hyder, M., 1972. Endocrine Regulation of Reproduction in Tilapia. Gen. Comp. Endocrinol. Suppl. 3: 729-740.
- Hyodo-Taguchi, Y., Egami, N. 1970. Inhibitory Effects of β-rays from ⁹⁰Sr -⁹⁰Y on Gonad Formation in Embryos of <u>Oryzias latipes</u>. Zool. Mag. 79: 185-187.
- Hyodo-Taguchi, Y., and Egami, N. 1971. Notes on X-ray Effects on the Testis of the Goby, <u>Chasmichthys glosus</u>. Annot. Zool. Jap. 44(1): 19-22.
- Hyodo-Taguchi, Y., Egami, N. 1974. Inhibitory Effect of x-irradiation on Maturation of Testes of Fish, <u>Oryzias latipes</u>, in Sexually Inactive Seasons. Radiat. Res. 59: 305.
- Hyodo-Taguchi, Y., Egami, N. 1976. Effect of X-Irradiation on Spermatogonia of the Fish, Oryzias latipes. Radiat. Res. 67(2): 324-331.
- Hyodo-Taguchi, Y., Egami, N. Mori, N. 1971. Histological Effects of β -rays from ⁹⁰ sr ⁹⁰Y on the Ovary of the Marine Goby, Chasmichthys glosus. J. Fac. Sci. Tokyo Univ 12(2): 337-344.
- Hyodo-Taguchi, Y., Egami, N., Yoshimura, N. 1970. Histological Effects of X-ray Irradiation on the Ovary of the Marine Goby Chasmichthys glosus. J. Fac. Sci. Tokyo Univ. 12(1): 47-54.
- Ichikawa, R., Suyama, I. 1974. Effects of Tritiated Water on the Embryonic Development of Two Marine Teleosts. Bull. Jap. Soc. Sci. Fish. 40(8): 819-824.
- Irons, H. T., Schreck, C. B. 1974. Sex Determination of Japanese Medaka (Oryzias latipes) by an Antiandrogen. VA. J. Sci. 25(2): 65.

- Ishii, S. 1961. Effects of Some Hormones on the Gestation of the Top Minnows, J. Fac. Sci. Tokyo Univ. 9(2): 278:290.
- Iwamatsu, T., Ohta, T. 1974. Cleavage-initiating Activities of Sperm Fractions Injected into the Egg of the Medaka, <u>Oryzias</u> <u>latipes</u>, J. Exp. Zool. 187(1):3-11.
- Jalabert, B., Billard, R., Chevassus, B. 1975. Preliminary Experiments on Sex Control in Trout: Production of Sterile and Simulataneous Self-fertilizable hermaprodites. Ann. Biol. Animal Biochim. Biophys. 15: 19-28.
- Jalabert, B., Moreau, J., Planquette, P., Billard, R. 1974. Sex Determination in <u>Tilapia macrochir</u> and <u>Tilapia nilotica</u>: Effect of Methyltestosterone Administered in Fry Food on Sex Differentiation; Sex Ratio of the Offspring Produced by Sexreversed Males. Ann. Biol. Animal Biochim. Biophys. 14(4-B): 729-739.
- Jensen, G. L., Shelton, W. L., Wilken, L. O. 1978. Use of vethyltestosterone Implants to Control Sex in Grass Carp and Their Effect on Growth. Proc. Fish. Cult. Sect., Am. Fish. Soc. (in press).
- Johnstone, R., Simpson, T. H., Youngson, A. F. 1978. Sex Reversal in Salmonid Culture. Aquaculture 13(2): 115-134.
- Kallman, K. D. 1962. Population Genetics of the Tynogenetic Teleost, Mollinesia formosa (Girard). Evolution 16: 497-504.
- Kallman, K. D. 1965. Genetics and Geography of Sex Determination of the Poeciliid fish, <u>Xiphophorus</u> <u>maculatus</u>. Zoologica N.Y., 50: 151-190.
- Katz, Y., Abraham, M., Eckstein, B. 1976. Effects of Androsterone on Gonadal and Body Growth in <u>Tilapia nilotica</u> (Teleostei, Cichilidae). Gen. Comp. Endocrinol. 29(3): 414-418.
- Kawamoto, N. 1973. Some morphological observations of the gonads in the Young Medaka (<u>Oryzias latipes</u>) treated with Sex Steriods. Zool. Mag. (Tokyo) 82(1): 29-35.
- Kobayashi, H. 1976. A Cytological Study on the Maturation Division in the Oogenic Process of the Triploid Ginbuna (<u>Carassius</u> <u>auratus langsdorfi</u>). Jap. J. Ichthyol. 22(4): 234-240.
- Kobayashi, J., Yamamoto, T. 1971. Effects of Irradiation on Spermatogenesis in the Guppy, <u>Lebistes reticulatus</u>. Tokushima J. Exp. Med. 18: 21-27.
- Kobayashi, S., Mizumoto, S. 1950. Studies on the Hybrid <u>Ctenopharyn-godon idellus</u> (Cuvier and Valenciennes) and <u>Cyprinus carpio L.</u> Sci. Rep. Shiga Fish. Exp. Sta. 1: 10-13.

- Kobayshi, S., Mogami, M. 1958. Effects of X-irradiation upon Rainbow Trout, <u>Salmo irideus</u>. III. Ovary Growth in the Stages of Fry and Fingerlings. Bull. Faculty Fish. Hokkaido Univ. 9(2): 89-94.
- Konno, K., Egami, N. 1966. Notes on the Effects of X-irradiation on the Fertility of the Male of <u>Oryzias</u> <u>latipes</u> (Teleostei, Cyprinodontidae). Annot. Zool. Jap. 39: 63-70.
- Kosheleva, V. V. 1971. Change in Peripheral Blood in the Embryos and Larvae of Atlantic Salmon under the Influence of Radioactive Contamination of the water and X-rays. AEC translation 7418 from Tr. Polyar Nauchno-Issled.: 75-88.
- Kothbauer, H., Schenkel-Brunner. H. 1974. Haemagglutinine aus Weiblichen Fischgonaden: Zur Frage ihrer Biologischen Funktion-Hemmversuche mit Maennlichen Gonaden Z. Immunitaets-Forsch. Exp. Klin. Immunol. 147(1): 87-90.
- Kramer, B. 1972. Behavioral effects of an Antigonadotropin, of Sexual Hormones, and of Psychopharmaka in the Pumpkinseed Sunfish, <u>Lepomis</u> gibbosus (Centrarchidae). Experientia 28(10): 1195-1196.
- Krumholtz, L. A. 1956. Observations on the Fish population of a Lake Contaminated by Radioactive Wastes. Bull. Mus. Nat. Hist. 110(4): 281-368.
- Kryzhanovskii, S. G., Disler, N. N., Smirnova, E. A. 1953. Ecologicomorphological Regularities in the Development of Percoid Fish. Tr. Inst. Morf. Zhiv. 10(3): 138.
- Kuronuma, K. 1955. Do Chinese Carps Spawn in Japanese Waters? Proc. Indo-Pac. Fish. Counc. 5: 126-130.
- Lichtenheld, R. W. 1967. Effect of Light, Temperature and Gamma Radiation on the Locomotor Activity of Juvenile Steelhead Trout (Salmo gairdneri). Diss. Abstr. 27B: 4606-4607.
- Lincoln, R. F., Aulstad, D., Grammeltvedt, A. 1974. Attempted Triploid Induction in Atlantic Salmon (<u>Salmo salar</u>) Using Cold Shocks. Aquaculture 4: 287-297.
- MacCrimmon, H. R., Stewart, J. E., Brett, J. R. 1974. Aquaculture in Canada, the Practice and the Promise. Bull. Fish. Res. Board. Can. 188: 1-84.
- Macek, K. J. 1968. Reproduction in Brook trout Salvelinus fontinalis Fed Sublethal Concentrations of DDT. J. Fish. Res. Board. Can. 25 (9): 1787-1796.
- MacKay, N. J. 1973. The Effects of Methallibure (I.C.I. 33,828) and Thiourea on Gametogenesis in the Firetail Gudgeon, <u>Hypseleotris</u> galii. Gen. Comp. Endocrinol. 20(2): 221-235.

- Makeeva, A. P. 1968. Hybridization of the Common Carp with Phytophagous Fishes. Probl. Ichthyol. 8(2): 294-297.
- Makeeva, A. P. 1969. Characteristics of Embryonal and Fry Development in Hybrids of Some Pond Cyprinidae. In: Cherfas, B. I. (ed.), Genetics, Selection, and Hybridization of Fish. Academy of Sciences USSR, Moscow. Transl. NTIS. Springfield, VA: 148-174.
- Makeeva, A. P. 1975. The Phenomenon of Hybrid Gynogenesis in Fishes. J. Ichthyol. 15(1): 72-81.
- Makeeva, A. P. 1976. On the Nature of Matroclinous Individuals among the Offspring of Remote Hybridization of Fishes. Genetike 12(11): 72-82.
- Makeeva, A. P., Sukanova, A. I. 1966. Development of Hybrids of Phytophagous Fishes. Vopr. Ikhtiol. 6(3): 477-497.
- Makeeva, A. P., Verigin, B. V. 1974. Hybridization of Carp <u>Cyprinus</u> <u>carpio L. with White Amur <u>Ctenopharyngodon idella</u> (Val.). Vopr. <u>Ikhtiol. 14(2): 290-296</u>.</u>
- Martin, P., Bromage, N. R. 1970. The effect of methallibure on spermatogenesis in Poecilia reticulata. J. Fish Biol. 2: 47-51.
- Michibata, H. 1976. The role of spermatogonia in the recovery process from temporary sterility induced by gamma-ray irradiation in the teleost <u>Oryzias latipes</u>. J. Radiat. Res. 17(3): 142-153.
- Migalovskii, I. P. 1971. Development of fish eggs and the early period of gametogenesis in the embryos and larvae of the Atlantic salmon under conditions of radioactive contamination of water. AEC translation from Tr. Polyar Nauchno-issled: 36-52.
- Mires, D. 1977. Theoretical and practical aspects of the production of all male tilapia hybrids. Bamidgeh 29(3): 94-101.
- Nakamura, M. 1975. Dosage-dependent changes in the effect of oral administration of methyltestosterone on gonadal sex differentiation in <u>Tilapia</u> mossambica. Bull. Fac. Fish. Hokkaido Univ. 26(2): 99-108.
- Nakamura, M., Takahashi, H. 1973. Gonadal sex differentiation in <u>Tilapia mossambica</u>, with special regard to the time of estrogen treatment effective in inducing complete feminization of genetic males. Bull. Fac. Fish. Hokkaido Univ. 24(1): 1-13.
- Nakatani, R. E., Foster, R. F. 1963. Effect of chronic feeding of Sr-90-Y-90 on rainbow trout. U.S. Atom. Energy Comm. HW-SAss55. 14 pp.

- Natali, V. F. 1940. The influence of Roentgen rays on the reproductive glands of male Lebistes reticulatus. Ychenye Zapiski Mosk. Gos. Ped. Instr. 22(1): 30-45.
- Natali, V. F. 1942a. Changes in the Ovaries of Fishes of the Poeciliidae Family under the Influence of X-rays in Connection with the Problem of Sex Inversion. Biull. Eksperim. Biol. Med. 14(4): 57-61.
- Natali, V. F. 1942b. On the Influence of Roentgen Rays on the Reproductive Glands of Female Fish (Lebistes reticulatus Pet.). Biull. Eksperim. Biol. Med. 13(5-6): 56-59.
- Natali, V. F., Natali, A. I. 1947. Development and Differentiation of the Gonads in Carps in Connection with the Problems of Transformation of the Sexes. Ycenye Zapiski Mosk. Gos. Ped. Inst. 40(3):
- Neifakh, A. A. 1959. Effects of Ionizing Radiation on Early Development of Fish. Trud. Inst. Morf. Zhiv. 24: 134-159.
- Nelson, S. G., Andersen, A. C., Momeni, M. H., Yeo, R. R. 1976. Attempted Sterilization of Sexually Undifferentiated Fry of <u>Tilapia</u> <u>zilii</u> by ⁶⁰ Cobalt Gamma-ray Irradiation. Prog. Fish-Cult. 38(3): 131-134.
- Neustroev. G. V., Podymakhin, V. N. 1966. On the Respiration of Salmon Roe (Salmo salar L.) Under Conditions of Radioactive Contamination of the Water. Radiobiology 6(1): 189-192.
- Nikolskaya, I. S., Grudnitskii, V. A. 1970. Influence of X-irradiation and Inhibitors of Nucleic Acid Aynthesis on the Respiration and ATP Content on Loach Embryos. Dokl. Biol. Sci. USSR. 194: 506-508.
- O'Brien, R. T., Fujihara, M. P. 1963. Effects of Deuterium Oxide and Radiation on Yeast and Fish. In: Hanford Biology Research Annual Report., USAEC HW-7600: 187-195.
- Okada, Y. A. 1964. Effects of Androgen and Estrogen on Sex Reversal in the Wrasse, <u>Halichoreres poecilopterus</u>. Proc. Jap. Acad. 38(8): 541-544.
- Oppermann, K. 1913. Die Entwicklung von Forelleneiern nach Befruchtung mit Radiumbestrahlten Samenfaden. Arch. Mikroskop. Anat. 83: 141-189.
- Pandey, S. 1979. Effects of Methallibure on the Testes and Secondary Sex Characters of the Adult and Juvenile Guppy <u>Poecilia</u> reticulata Peters. Biol. Reprod. 2: 239-244.

- Pandey, S., Leatherland, J. F. 1970. Comparison of the Effects of Methallibure and Thiourea on the Testes, Thyroid, and Adebohypohysis of the Adult and Juvenile Guppy, <u>Poecilia reticulata</u>. Can. J. Zool. 48: 445-450.
- Pechkurenkov., V. L. 1976. Chronic Irradiation of Loach Embryos (<u>Misgurnus fossilis</u> L.) Incubated in EDTA and Caffeine. Radiobiologiya. 16(4): 587-592 (in Russian).
- Persov. G. M. 1975. Differentiation of Sex in Fish. Leningrad University Press, Leningrad. 148(in Russian).
- Pflieger, W. L. 1978. Distribution and Status of the Grass Carp (<u>Ctenopharyngodon idella</u>) in Missouri Streams. Trans. Am. Fish. Sox. 107(1): 113-118.
- Piggins, D. J. 1965a. Salmon and Sea Trout Hybrids. Salmon Res. Trust. Ireland, Rep. for the Year Ended 31st Dec. 1964: 27-37.
- Piggins, D. J. 1965b. Salmon and Sea Trout Hybrids. Atl. Salmon J. (Fall issue): 3-5.
- Purdom, C. E. 1969. Radiation-induced Bynogenesis and Androgenesis in Fish. Heredity 24(3): 431-444.
- Purdom, C. E. 1970. Gynogenesis A Rapdi Method for Producing Inbred Lines of Fish. Fish. News Internat. 9(9): 29-30.
- Purdom, C. E. 1972. Induced Polyploidy in Plaice (<u>Pleuronectes platessa</u>) and Its Hybrid with the Flounder (<u>Platichthys flesus</u>). Heredity 29(1): 11-24.
- Purdom, C. E. 1976. Genetic Techniques in Flatfish Culture, J. Fish. Res. Board Can. 33(4): 1088-1093.
- Purdom, C. E. Lincoln, R. F. 1973. Chromosome Manipulation in Fish. In: Schroder, H. (ed.) Genetics and Mutagenesis of Fish. Springer-Verlag, New York: 83-89.
- Purdom, C. E., Lincoln, R. F. 1974. Gynogenesis in Hybrids within the Pleuronectidae. In: Blaxter, J.H.S. (ed.), The Early Life History of Fish. Springer-Verlag, Berlin: 537-544.
- Purdom, C. E., Woodhead., D. S. 1973. Radiation Damage in Fish. In: Schroder, (ed), Genetics and Mutagenesis of Fish. Springer-Verlag, New York: 67-73.
- Purdom, C. E., Thompson, D., Dando, P. R. 1976. Genetic Anaysis of Enzyme Polymorphisms in Plaice (Pleuronectes platessa). Heredity 37(2): 193-206.

- Refstie, T., Vassvik, V., Gjedrem, T. 1977. Induction of Polyploidy in Salmonids by Cytochalasin B. Aquaculture 10: 65-74.
- Reinboth, R. 1970. Intersexuality of fish. Mem. Soc., Endocrinol. (Hormones and Environment). 18: 515-544.
- Romashov, D., D., Belyaeva, V. N. 1964. Cytology of Radiation Gynogenesis and Androgenesis in the Loach (<u>Misgurnus fossilis L.</u>). Dokl. Biol. Sci. 157: 403-506.
- Romashov, D., D., Belyaeva, V. N. 1965. Increased Yield of Diploid Gynogenetic Loach Larvae (<u>Misgurnus fossilis L.</u>) Induced by Temperature Shock. Byull. Mosk. Obshch. Ispyt. Prir. (Biol.) 70 (5): 93-109 (in Russian).
- Romashov, D. D., Golovinskaya, K. A. 1960. Radiation Biology and Genetics of Fish. Sb. Itogi. Nauk., Biol., Nauk (3): 324-343 (in Russian).
- Romashov, D. D., Nikolyukin, N.I., Belyaeva, V. N., Timofeeva, N. A. 1963. Possibilities of Producing Diploid Radiation-Induced Gynogensis in sturgeons. Radiobiology 3(1); 145-154.

Roosevelt, R. B. 1881. Hybrids. Trans. Am. Fish. Soc. 9: 8-13.

- Rugh, R., Clugston, H. 1965. Effects of Various Levels of Xirradiation on the Gametes and Early Embryos of <u>Fundulus heteroclitus</u>. Biol. Bull. 108: 318-325.
- Samokhvalova, G. V. 1938. The effect of Roentgen Rays on Fish (Guppy, Swordtail and Goldfish). Biol. Zhur. 7: 1023-1034.
- Scarborough, B. B., Addison, R. G. 1962. Conditioning in Fish, Effects of X-irradiation. Science 136(35170: 712-713.
- Schreck, C. B. 1973. Uptake of ³H-testosterone and Influence of an Antiandrogen in Tissues of Rainbow Trout (Salmo gairdneri). Gen. Comp. Endocrinol. 21: 60-68.
- Schreck, C. B. 1974. Hormonal Treatment and Sex Manipulations in Fishes. In: Schreck, C. B., (ed.), Control of Sex in Fishes. Extension Division, Virginia Polytechnic Institute and State University, Blacksburg, VA: 84-106.
- Schroder, J. H. 1973. Teleosts as a Tool in Mutation Research. In: Schroder, J. H., (ed.), Genetics and Mutagenesis of Fish. Springer-Verlag, New York: 91-99.
- Schultz, R. J. 1967. Gynogenesis and Triploidy in the Viviparous Fish Poeciliopsis. Science 157: 1564-1567.

- Schultz, R. J. 1973. Unisexual Fish: Laboratory Synthesis of a "Species". Science 179: 180-181.
- Schultz, R. J., Kallman, K. D. 1968. Triploid Hybrids Between the All-female Teleost <u>Poecilia</u> formosa and <u>Poecilia</u> sphenops. Nature 219: 280-282.
- Shechmeister, I. L., Watson, L. J., Cole, V. W., Jackson, L. L. 1962. The Effect of X-irradiation on Goldfish. I. The Effect of X-irradiation on Survival and Susceptibility of the Goldfish, <u>Carassius auratus</u>, to Infection by <u>Aeromonas salmonicida</u> and <u>Gyrodactylus spp. Radiat. Res. 16(1): 89-97.</u>
- Solbert, A. N. 1938. The Susceptibility of the Germ Cells of Oryzias latipes to X-radiation and Recovery after Treatment. A. Exp. Zool. 78: 417-439.
- Srivastava, P. N. 1966. Effect of Ionizing Radiation on the Ovaries of Japanese Medaka, <u>Oryzias latipes</u> (T and S). Acta Anat. 63(3): 434-444.
- Srivastava, P. N., Rathi, S. K. 1967. Radiation-induced Castration in the Female Teleostean Fish, <u>Heteropneustes</u> fossilis Bloch. Experientia 23(3): 229-230.
- Stanley, J. G. 1976a. Production of Hybrid, Androgenetic, and Gynogenetic Grass Carp. Trans. Am. Fish. Soc. 105(1): 10-16.
- Stanley, J. G. 1976b. Production of Monosex White Amur for Aquatic Plant Control. Contract Rep. A-76-1, U.S. Army Corps of Engineers, Washington, DC 41 p.
- Stanley, J. G. 1976c. A Review of Methods for Obtaining Monosex Fish and Progress Report on Production of Monosex White Amur. J. Aquatic Plant Manage. Soc. 14: 68-70.
- Stanley, J. G. 1976d. Female Homogamety in Grass Carp (<u>Ctenopharyngodon idella</u>) Determined by Gynogenesis. J. Fish. Res. Board Can. 33(6): 1372-1374.
- Stanley, J. G., Jones, J. B. 1976. Morphology of Androgenetic and Gynogenetic Grass Carp, <u>Ctenopharyngodon idella</u> (Valenciennes). J. Fish. Biol. 9(6): 523-528.
- Stanley, J. G., Thomas, A. E. 1978. Absence of Sex Reversal in Unisex Grass Carp Fed Methyltestosterone. Pages 194-199 In R. O. Smitherman, W. L. Shelton, J. H. Grover, eds. Culture of Exotic Fishes Symposium, Am. Fish. Soc., Auburn, Alabama.

- Stanley, J. G., Sneed, K. E. 1974. Artificial Gynogeneis and its Application in Genetics and Selective Breeding of Fishes, In: Blaxter, J. H. S. (ed.), The Early Life History of Fish. Springer-Verlag, Berlin: 527-536.
- Stanley, J. G., Biggers, C. J., Schultz, D. E. 1976. Isozymes in Androgenetic and Gynogenetic White Amur, Gynogenetic Carp, and Carp-Amur Hybrids. J. Hered. 67(3): 129-134.
- Stanley, J. G., Martin, J. M., Jones, J. B. 1975. Gynogenesis as a Possible Method for Producing Monosex Grass Carp (Ctenopharyngodon idella). Prog. Fish-Cult. 37(1): 25-26.
- Stanley, J. B., Miley, W. W., Sutton, D. L. 1978. Reproductive Requirements and Likelihood for Naturalization of Escaped Grass carp in the United States. Trans. Am. Fish. Soc. 107(1): 119-128.
- Stock, J. N., Cope, O. B. 1969. Some Effects of TEPA, an Insect Chemosterilant, on the Guppy, Poecilia reticulata. Trans. Am. Fish. Soc. 98(2): 280-287.
- Strand, J. A., Fujihara, M. P., Burdett. R. D., Poston, T. 1977. Suppression of the Primary Immune Response in Rainbow Trout (Salmo gairdneri), Sublethally Exposed to Tritiated Water during Embryogenesis. J. Fish. Res. Board Can. 34(9): 1293-1304.
- Stroganov, N. S., Telitchenko, M. M. 1958. The Chronic Effect of small Doses of U-238 on Bleak Gonads. Byul. Mosk, Obshch. Ispytat. Priorody. Otd. Biol. 63(4): 154 (in Russian).
- Suzuki, R. 1965a. Notes on the Bitterling Hybrids Produced in an Aquarium. Bull. Freshwater Res. Lab. 15(1): 49-58.
- Suzuki, R. 1965B. Hybridization Experiments in Cyprinid Fishes. IX. <u>Gnathopogon japonicus x Biwia zezera</u> and Reciprocal Crosses between <u>Pseudorasbora parva</u> and <u>Biwia zezera</u>. Bull. Freshwater Fish. Res. Lab. 14(2): 91-95.
- Suzuki, R. 1974. Inter-crossings and Back-crossings of F₁ Hybrids Among Salmonid Fishes. Bull. Freshwater Fish. Res. Lab. 24(1): 11-32.
- Svardson, G. 1945. Chromosome Studies on Salmonidae. Medd. Undersokn Anst. Satvattensfisk. Stockh. 23: 1-151.
- Swarup, H. 1956. Production of Heteroploidy in the the Three-spines Stickleback <u>Gasterosteus</u> aculeatus (L.). Nature, London, 178: 1124-1125.

Swarup, H. 1959a. Production of Triploidy in <u>Gasterosteus</u> aculeatus (L.). J. Genet. 56: 129-142.

- Swarup, H. 1959b. Effect of Triploidy on the Body Size, General Organization and Cellular Structure in <u>Casterosteus</u> aculeatus (L.). J. Genet. 56: 143-55.
- Takahashi, H. 1975a. Process of Functional Sex Reversal of the Gonad in the Female Guppy, <u>Poecilia</u> <u>reticulata</u>, Treated with Androgen Before Birth. Devl. Growth Diff. 17(2): 167-176.
- Takahashi, H. 1975b. Functional Masculinization of Female Guppies, <u>Poecilia reticulata</u>, Influenced by Methyltestosterone Before Birth. Bull. Jap. Soc. Sci. Fish. 41: 499-506.
- Takahashi, H. 1975c. Masculinization of the Gonad of Juvenile Guppy, <u>Poecilia reticulata</u>, Induced by 11-ketotesterone. Bull. Fac. Fish. Hokkaido Univ. 26(1): 11-22.
- Takahashi, H. 1975d. Functional Feminization of Genetic Males of the Guppy, <u>Poecilia reticulata</u>, Treated with Estrogen after Birth, Bull. Fac. Fish. Hokkaido University 26(3): 223-234.
- Takahashi, H.1975e. Modification of the Development of Female Reproductive Organs in the Guppy, Poecilia reticulata, following an Androgen Treatment in their Juvenile Period. Bull. Fac. Fish. Hokkaido Univ. 25(3): 174-199.
- Tang, F., Chan, S. T. H., Lofts, B. 1974a. Effect of Steriod Hormones on the Process of Natural Sex Reversal in the Rice-field Eel, <u>Monopterus albus</u> (Zuiew). Gen. Comp. Endocrinol. 24(3): 227-241.
- Tang, F., Chan, S. T. H., Lofts, B. 1974b. Effect of Mammalian Luteinizing Hormone on the Natural Sex Reversal of the Ricefield eel, <u>Monopterus albus</u> (Zuiew). Gen. Comp. Endocrinol. 24(3): 242-248.
- Tashiro, F. 1972. Effects of Irradiation ⁶⁰ Co Gamma-ray on the Maturation of Rainbow Trout. Bull. Jap. Soc. Fish. 38(8): 793-797.
- Theriot, R. F. 1977. The large-scale Operations Management Test (LSOMT) with the White Amur at Lake Conway, Orlando, Florida. Proc. Res. Planning Conf. Aquatic Plant Control Prog. A-77-3: 87-96.
- Thomas, A. E. 1977. Culture Techniques for the Production of Monosex White Amur. Proc. Res. Planning Conf. Aquatic Plant Control Prog. A-77-3: 177-186.
- Thomas, A. E., Carter, R. R. 1977. Survival of Monosex Grass Carp in Small Ponds. Prog. Fish-Cult. 39(4): 184.

- Tsoi, R. M. 1969. The Effect of Nitrosomethyl urea and Dimethylsulphate on Sperm of the Rainbow Trout (Salmo irideus Gibb.) and the Peled (Coregonus peled Gmel.). Dokl. Biol. Sci. USSR. 189: 849-852.
- Tsoi, R. M. 1972. Chemical Gynogenesis of <u>Salmo</u> irideus and <u>Coregonus</u> <u>peled</u>. Genetike 8(2): 185-188 (in Russian).
- Tsoi, R. M., Menshova, A. I., Golodov. Yu. F. 1974. The Specificity of Chemical Mutagens. Effects on Spermatic Elements of Cyprinus carpio L. Genetika. 10(2): 68-72 (in Russian).
- Tsvetkova, L. I. 1971. Investigation of the Progeny from Gynogenetic carp female. Trud. Vses. Nauchno-Issled. Inst. Prud. Rybn. Khoz. 18: 234-239 (in Russian).
- Ulrikson, G. U. 1969. Use and Effects of Cobalt 60 for Sterilization of Bluegills (Lepomis macrochirus). Diss. Abstr. 30B: 2473.
- Valenti, R. J. 1975. Induced Polyploidy in Tilapia aurea (Steindachner) by Means of Temperature Shock Treatment. J. Fish Biol. 7: 519-528.
- Van Den Hurk, R., Van de Kant, H. 1975. The Effect of Methyltestosterone, 11-ketotestosterone and Methallibure on Gonadotropic Cells, Leydig Cells, Spermatogenesis and the Epithelium of the Intratesticular Efferrent Duct System of the Juvenile Male Black Molly (<u>Mollienisia latipinna</u>). Proc. K. Ned. Akad. Wet. Ser. C. Biol. Med. Sci. 78(3): 275-285.
- Van Den Hurk, R., Testerink, G. J. 1975. The Effect of Methallibure and Methyltestosterone on Gonadotropic Cells, Leydig Cells and the Intratesticular Efferent Duct System of the Adult Male Black Molly <u>Mollienisia latipinna</u>. Proc. K. Ned. Akad. Wet. Ser. C. Biol. Med. Sci. 78(3): 265-274.
- Vanyakina, E. D. 1969. Genetics of Sex Determination and Some Problems of Hormonal Regulation of Sex in Telests, In: Cherfas, B. I. (ed.), Genetics, Selection and Hybridization of Fish. Akad. Nauk, USSR. Trans. N.T.I.S., Springfield, VA: 25-41.
- Vanyakina, E. D. 1972. Action of Methyltestosterone on Sexual Differentiation in the Paradisefish (<u>Macropodus opercularis L.</u>) and Guppy (<u>Lebistes reticulatus</u> R.). Dokl. Biol. Sci. USSR. 202: 42-44.
- Vasetskii, S. G. 1967. Changes in the Ploidy of Sturgeon Larvae Induced by Heat Treatment of Eggs at Different Stages of Development. Dokl. Biol. Sci. USSR. 172: 23-26.

- Vasilev, V. P., Makeeva, A. P., Ryabov, I.N. 1975. On Triploidy of Remote Hybrids of Carp (Cyprinus carpio L.) with other representatives of Cyprinidae. Genetika, 11(8): 49-56 (in Russian).
- Vassileva-Dryanovska, O., Belcheva, R. 1965. Radiation Gynogenesis in <u>Salmo</u> irideus Givv. C. R. Acad. Bulg. Sci. 18(4): 359-362.
- Vivien, J. 1950. Masculinisation des Femelles de Xiphorphores par Action des Rayon X sur la Gonade, Compt. Rend. Acad. Sci. Paris, 231: 1166-1168.
- Vivien, J. H. 1953a. Action du Phosphore Radioactif sur les gonades des Cyprinodontes vivipares: Lebistes et Xiphophores. Compt. Rend. Soc. Biol. 147: 1459-1462.
- Vivien, J. H. 1953b. Sterilisation Totale des Gonades apres Traitement par le Phosphore Radioactif, chez les Cyprinodontes: <u>Lebistes</u> et Xiphophores. Compt. Rend. Aca. Sox., Paris. 236: 2172-2174.
- Vrijenhoek, R. C., Schultz, R. J. 1974. Evolution of a Trihybrid Unisexual Fish (<u>Poeciliposis</u>, Poeciliidae). Evolution. 28(2): 306-319.
- Warner, R. R. 1975. The Adaptive Significance of Sequential Hermaphroditism in Animals. Am. Natur. 109(965): 61-82.
- Watson, D. G., George, L. A., Hackett, P. L. 1959. Effects of Chronic Feeding of Phosphorus-32 on Rainbow Trout. In: Annual Report for 1958. USAEC HW--59500: 73-77.
- Watson, L. J., Shechmeister, I. L., Jackson, L. L. 1963. The Effect of X-irradiation on the Hematology of <u>Carassius auratus</u>. Physiol. Zool. 36(4): 370-382.
- Welander, A. D. 1954. Some Effects of X-irradiation of Different Embryonic Stages of the Trout (Salmo gairdnerii). Growth. 18: 227-255.
- Welander, A. D., Donaldson, L. R., Foster, R. F., Bonham, K., Seymour, A. H. 1948. The Effects of Roentgen Rays on the Embryos and Larvae of the Chinook Salmon. Growth. 12: 203-242.
- Wiebe, J. P. 1969. Endocrine Controls of Spermatogenesis and Oogenesis in the Viviparous Seaperch Cymatogaster aggregata Gibbons. Gen Comp. Endocrinol. 12(2): 267-275.
- Yamamoto, T. 1958. Artificial Induction of Functional Sex-reversal in Genotypic Females of the Medaka (Oryzias latipes). J. Exp. Zool. 137(2): 227-263.

- Yamamoto, T. 1959. The Effects of Estrogen Dosage Level upon the Percentage of Sex-reversals in Genetic Male (XY) of the Medaka (Oryzias latipes). J. Exp. Zool. 141: 133-153.
- Yamamoto, T. 1965. Estriol-induced XY Females of the Medaka (Oryzias latipes) and Their Progenies. Gen. Comp. Endocrinol. 5: 527-533.
- Yamamoto, T. 1968. Effects of 17a-hydroxyprogesterone and Androstenedione upon Sex Differentiation in the Medaka, <u>Oryzias latipes</u>. Gen. Comp. Endocrinol. 10(1): 8-13.
- Yamamoto, T. 1969. Sex Differentiation. In: Hoar, W. S., Randall. D. J., (eds.), Fish Physiology, Academic Press, New York: 117-175.
- Yamamoto, T. 1975. A YY Male Goldfish from Mating Estrone-induced XY Females and Normal Males. J. Hered. 66(1): 2-4.
- Yamamoto, T., Kajishima, T. 1968. Sex Hormone Induction of Sex Reversal in the Goldfish and Evidence for Male Heterogamety. J. Exp. Zool. 168(2): 215-221.
- Yamamoto, T., Matsuda, N. 1963. Effects of Estrodiol. Stilbestrol and Some Alkyl-carbonyl Androstanes upon Sex Differentiation in the Medaka, <u>Oryzias latipes</u>. Gen. Comp. Endocrinol. 3(2): 101-110.
- Yamamoto, T., Takeuchi, K., Takai, M. 1968. Male-inducing Action of Androsterone and Testosterone Propionate upon XX Zygotes in the Medaka, <u>Oryzias latipes</u>. Embryologia (Nagoya). 10: 142-151.

Yamazaki, F. 1972. Effects of Methyltestosterone on the Skin and Gonads of Salmonids. Gen. Comp. Endocrinol. Suppl. 3: 741-750.

Yamazaki, F. 1976. Application of Hormones in Fish Culture. J. Fish. Res. Board. Can. 33(4): 948-958.

Radiation	Dose (kR)	Species	Effects on Reproduction	References
Beta rays p ³²	internal	guppy, swordtail	sterility	Vivien 1953a, 1953b
Beta rays H ³	in water	guppy	retarded sex development	Erickson 1971
Beta rays Sr ⁹⁰	in water	medaka	inhibited gonads	Hyodo-Taguchi & Egami 1970
Beta rays Sr ⁹⁰	in water	golby	oocytes destroyed	Hyodo-Taguchi et al. 1971
Beta rays Sr ⁹⁰	in water	Atlantic salmon	sex cells depressed	Migalovskii 1971
Gamma rays Co ⁶⁰	internal	Indian catfish	sterility	Srivastava & Rathi 1967
Gamma rays Co ⁶⁰	1	carp	sterilized 1-2 years	Cherfas 1962
Gamma rays Co ⁶⁰	1	bluegill	temporary sterility	Ulrikson 1969
Gamma rays Co ⁶⁰	0.4	rainbow trout	depressed maturation	Tashiro 1972
Gamma rays Co ⁶⁰	0.8	chinook salmon	inhibited gonads	Bonhnam & Donaldson 1972
Gamma rays Co ⁶⁰	5-8	guppy	most were sterilized	Purdom & Woodhead 1973
Gamma rays Co ¹³⁷	0.5-1	medaka	temporary sterility	Michibata 1976
x rays	2	medaka	infertile eggs	Egami & Hyodo 1965, Konno & Egami 19
x rays	2	medaka	most were sterile	Egami & Hodo-Taguchi 1969
x rays	0.7-3.5	guppy	testicular degeneration	Kobayshi & Yamamoto 1971
x rays	0.6	Carp	sterility for 2-3 years	Cherfas 1962
x rays	0.36	pink salmon	some sterile males	Persov 1975
x rays	0.5	rainbow trout	reduced ovaries	Kobayashi & Mogami 1958
x rays	1-16	Loach	ovarian damage	Egami and Aoki 1966
x rays	32	tilapia	sterilized	Al-Dahan 1970
x rays	0.5-4	goby	temporary sterility	Hyodo-Taguchi et al. 1970 Hyodo-Taguchi & Egami 1971

Table 1. Summary of studies that mention radiosterilization of fishes given various doses of radiation. Exact doses cannot be determined for fish injected with or held in isotype solutions.

242

(

í

1

Į

1 . 1

DISEASE CONTROL AND REPRODUCTION OF

GRASS CARP IN GERMANY

Martin Bohl

Bavarian Water Research Agency

Experimental Station of Fishery Biology, D-8 2, Wielenbach i.OB.

INTRODUCTION

Grass carp were first imported into the Federal Republic of Germany from Hungary in 1964 - it was only a "good handful" of fry. The Wielenbach research station received the second load of 3-year old herbivorous fishes in 1966 (Bohl 1967). In the following years more fish were imported; not only from Hungary but from other countries. The many risks that are involved in introducing foreign species are now well known. There are not only ecological problems but also great dangers from disease. In contrast to the known disease problems, which are connected to the trade of indigenous fishes, the importation of foreign species often causes more trouble. These latter problems will be discussed.

NEW DISEASES IMPORTED WITH GRASS CARP

Not only new infectious diseases but also new parasites can be introduced by the import of fish, not native to our country. Most fish epidemics are not confined to one species. Therefore, the spread of fish epidemics caused by the import of non native fish is as great as those caused during the importation or trade of indigenous fish. Much more dangerous is the introduction of new parasites by the imported foreign fishes. However, it is only those parasites able to survive in a new biotop which will find conditions suitable for developing or where intermediate hosts occur. Fortunately, this rarely happens. Only during the first years after the introduction of grass carp into Russia were some species of parasites such as Crytobia branchialis, Capillaria amurasis found (Bauer et al. 1969). To these parasites the metacercaria of Metagonimus Yokogawai can be added. This metacercaria develops and becomes mature in the intestine of humans and of carnivorous mammals. Obviously the necessary intermediate hosts, certain molluscs, were missing in their new surroundings in Europe. Normally, only parasites become indigenous which do not need intermediate hosts or which are able to develop in several species or in a wide spread species. The introduction of new parasites to another country would not be so disturbing if these parasites remained with the imported species, but very often they are able to develop or live on other species of fish. In this respect, the recently invaded fish may be more severely damaged because they did not have occasion to develop immunity. The following helminth parasites have been introduced into the German Federal Republic by imported grass carp:

 Bothriocephalus gowkongensis Yeh, 1955; the name of this species has been corrected by Korting (1975b) to <u>B</u>. <u>acheilognathi</u> Yamaguti, 1934. in two week intervals under German carp pond conditions.

Another application-method used in Iran (Mokhayer 1976a) is 1 mg Yomesan per 1 g body weight of fish is cannulated into the first portion of the digestive tract. In a similar manner dibutyl-tindilaurate has been tried in New Zealand (Edwards and Hine 1974). If fish will not ingest food, this method is very helpful in cases of heavy tapeworm infestation.

Prophylaxe

Prophylactic measures involve controlling the <u>Bothriocephalosis</u> by disconnecting the life cycle. In this case eggs must be killed either by draining or disinfection of ponds. Kulow (1973) reported that the eggs of <u>Bothriocephalus</u> die very quickly in drained ponds at a temperature under 18 C. Disinfecting ponds thoroughly with quicklime (2.5-4 tons/ha) or chloride of lime (600 kg/ha) in combination with prophylactic measures are successful in ridding a pond of Bothriocephalus.

KHAWIOSIS

As already mentioned, the second of the two imported parasites was also a cestode. It is a caryophyllaeid. In routine examination it is very difficult to differentiate this new parasite, <u>Khawia sinensis</u> (Hsu 1935) from our European species <u>Caryophyllaeus fimbriceps</u> or <u>laticeps</u>. For identification the parasite should be fixed in 70% alcohol and stained with alauncarmine (50 mg ammonia potassium and 1 mg carmine heated in 1 L of water for 15 minutes and filtered). Before staining the parasite must be rinsed thoroughly in aqua dest. The preparation is stained for 5-30 minutes and rinsed again.

Microscopically the two parasites can be differentiated: The vesicles of the testes and vitellaria begin about 2-4 mm behind the scolex of <u>Khawia sin-</u><u>ensis</u>, whereas the pre- and postovarian vitellaria occur in the cortical parenchyma in <u>Caryophyllaeus</u>, some postovarian vitelline follicles occur in the medullary parenchyma in <u>Khawia sinensis</u>. This can easily be seen in cross section. Obviously the difficulty in identification is one of the reasons that in the GFR <u>Khawia</u> has been found in only a few fish populations. Up to 60 parasites have been found in one fish. When fish are heavily attacked a catarrhalic or catarrhalic-hemorrhagic enteritis results. If secondary infections follow there are usually high losses. The life cycle requires oligochaeta (Tubificidea) as intermediate hosts and invasion is highest in early summer. The procercoid remains in the tubificid and is able to invade fish for about one year. During late summer (August) the cestodes leave the intestine and die.

The primary final hosts for this parasite are one year old carp and grass carp. Khawiosis is controlled like Bothriocephalosis¹.

After the Grass carp conference the following report was published: "Successful treatment of carp caviosis by means of Taenifugin in Czechoslowakia by Tesarcik, J. and A. Prouza, 1977. Bul VURH Vodnany 8(4):26-31. Taenifugin granulat: piperazin-niclosamide, sodium chloride and wheat-food-flour. 2. Khawia senensis Hsu 1935 (Körting 1974).

BOTHRIOCEPHALOSIS

The most dangerous species is the pseudophyllidian tapeworm, <u>Bothrioce-phalus acheilognathi</u>. While it damages the grass carp very little, it can develop in young European mirror carp. In some cases almost 100% of the one-year old carp are attacked.

In case of a heavy infestation, growth is reduced considerably. In the summertime mortality can reach about 30% and during the winter about 90% (Kulow 1973). Up to 60 tapeworms have been found in one fish. In Russia up to 3,000 in one fish. In this case the entire digestive tract is stuffed with tapeworms. This often results in a catarrhalic or catarrhalic-hemorrhagic enteritis. These young carp will appear small and very cachetic as compared to normal ones. In Germany, we have not found a heavy infection in grass carp, in contrast to the experiences in Iran (Mokhayer 1976b). The scolex of this tapeworm possesses two bothrias. The adult worm sheds eggs into the fish gut and into the water. The coracidium hatches at 25 C water temperature during the third day. Further development to the procercoid stage takes place in copepods (intermediate hosts) and is completed after 8-10 days (Korting 1975a). The life cycle of this tapeworm is completed when infected copepods are eaten by carp and other susceptible species. In the intestine it completes its development from procercoid to a plerocercoidlike stage after 20-21 days to the adult tapeworm of Bothriocephalus acheilognathi.

In Germany, this tapeworm has not been found in fishes other than carp and grass carp. In Russia, this parasite also lives in crucian carp (<u>Carassius carassius</u>), gibel (<u>C. auratus gibelio</u>), white bream (<u>Abramis brama</u>), barbel (<u>Barbus barbus</u>), chrup (<u>Aspius aspius</u>), wels (<u>Silurus glanis</u>), and species of gambusia (Bauer et al. 1969). Reasons for the rapid spread are: 1) a relatively simple life cycle; 2) several cyclops species as intermediate hosts; which are widely spread over the world; and 3) the wide range of final hosts.

To prevent damage in fish and further spread, it is necessary to control Bothriocephalosis.

CONTROL OF BOTHRIOCEPHALOSIS

Direct Therapy

There are several medications for direct therapy:

Phenothiazin (1 g/kg body wt per oz; 3 time ia intervals of 2 days).

Di-n-butyl-tinoxyd (0.5 g/kg body wt 1 to 3 times a day for 3 days (Hnath 1970).

The best medication is $Mansonil^R$, a niclosamide-piperazin-salt (a synonym is Yomesan^R, used in human medicine). This substance is mixed into the food; 500-600 g/100 kg dried pellets, 1.5% of body weight is fed 2 to 3 times

Cryptobiasis. Against the first two protozoans malachite green is successful (0.15 ppm; 1-3 times in intervals of two days). Sometimes in therapy malachite green is combined with 24 ppm formaldehyde. According to Russian experiments (Zobel 1975) treatments with brilliant green and violet K (arylmethan-colour substances) are also very effective in concentrations of 0.15-0.2 ppm (water temperatures 7-12 C) against Ichthyophthirius, Childonella and other parasites. Against the gillworm Masoten^R (Metrifonal, 0.5 ppm = 5 kg/ hectare average water depth of 1 m) is very effective. Short term baths can also be given with sodium chloride (2.5%, 10 minutes) or better results are obtained with formaldehyde (20-25 ml of 40% formaldehyde in 100 litres of water for 30 minutes) (Bohl 1975).

We have also had losses of grass carp fingerlings due to cryptobia, a flagellate protozoa. During heavy infestations fish showed typical "Scale detrusion". On the sides of their bellies there were many small hemorrhagics and the skin was slimy. The epithelium of the mouth was swollen with many cryptobia present. In some cases they were found only on the gills.

PROPHYLAXE FOR INTRODUCED PARASITES

Imports of cyprinids, which are already on food, should be denied entry. Fry are excluded from this edict if they are hatched in closed warmwater systems and are fed on artificially produced food. The reason for this rule is that all species of fish could introduce several parasite species into the country. Older fish carry more parasites. In spite of intensive diagnostic controls of imported fish, we found that generally only 25% of the possible parasites are found.

During a period of quarantine of 2 to 4 years about 75% of the parasites are found. About 100% are found after 5 to 10 years. The most dangerous parasites, Bothriocephalus, Khawia and Philometra belong in these categories.

In spite of increasingly stronger control regulations and consequent quarantine, 27 different species of parasites were introduced into Russia (Bauer et al. 1969). Rinsing the intestine may help considerably when diagnosing intestinal parasites of living fish (Faina and Par 1977). A small plastic hose is introduced into the cranial part of the digestive tract and is connected to a syringe in order to rinse the intestine with water. Carp, with a weight of more than 20 grams can be examined by this method. It is more difficult with grass carp. Food as well as cestodes will be washed from the intestine. This may not be the best method for diagnosis, but in some cases it is helpful.

INFECTIOUS DISEASES

As previously mentioned, infectious fish diseases can be introduced and spread by foreign fish. It is known that grass carp can be affected by infectious dropsy, which may sometimes result in heavy losses.

The dropsy-complex is divided into two types: "spring viremia", the acute form - the infectious agent being <u>Rhabdovirus carpio</u> - and erythrodermatitis, the chronic form - the infectious agent being a subspecies of <u>Aeromonas sal</u>-<u>monicida</u> var. <u>achromogenes</u>. A specific enteritis is also observed in the

OTHER PARASITOSIS

Because laws do not exist for the control of fish imports, fish epidemics are uncontrolled. If major losses do not occur, imported fish will be without health-control. Therefore, we may assume that other foreign parasites have been introduced into our country by the imports of grass carp or carp. In this manner, 27 species of parasites have been introduced into Russia (Bauer et al. 1969). Most of them are protozoans and are not usually as important²/.

Another example of a dangerous parasite is a nematode: <u>Philometra lusi-ana</u> (Vismanis 1966). The red colored female penetrates through the skin where the posterior end can often be seen. The males usually enter the caudal part of the airbladder and destroy it. Fish infected in this manner lose their equilibrium.

DISEASES WHICH AFFECT GRASS CARP IN EUROPE

Not only are indigenous fishes endangered by imported disease agents or parasites, but imported fishes are endangered because they may not be immune against our indigenous parasites and other disease agents.

We know of some cases where grass carp have been heavily infested with digenetic trematodes. From Rumania (Fadulescu and Georgescu 1963) and the German Democratic Republic (Mattheis 1967) heavy invasions of metacercariae of <u>Diplostomum</u> <u>spathaceum</u> have occurred, which causes parasitic-cataract ("Wurmstar"). Grass carp are invaded 5 to 25 times as much as carp which are the same age and reared in the same ponds. In Hungary the situation is similar (Szakolczai and Molnar 1966) with about 200-300 parasites per host. Grass carp are more susceptible to these metacercaria than carp.

In the German Democratic Republic grass carp were strongly invaded by a tetracotylous metacercaria (<u>Apharyngostrigea</u> curnu) in the abdomen (Mattheis and Odening 1969). Along with other species of fish the grass carp is the second intermediate host for this parasite. Obviously the grass carp is preferred by the miracidia, for they develop very well. The first intermediate host is the common pondsnail (<u>Planorbis</u> sp.). The final hosts are heronbirds. A heavy infestation by a similar species of metacercaria (<u>Tetracotyle percae fluviatilis</u>) caused heavy losses in Hungary (Szakolczai and Molnar 1966).

These tetracotylosis are controlled by interrupting the life cycle. In this case the best method is to control the snails with molluscicides (Bayluscid^R, 0.5 ppm) or possibly with snail eating fish like <u>Mylopharyngodon piceus</u>, and by keeping away the final hosts (waterbirds). Other widespread parasitosis which endanger the population of the grass carp are: <u>Ichthyophthiriasis</u> (white spot), Chilodonellosis, Dactylogeirosis, and

^{2/} Latest experiences have shown that <u>Balantidium ctenopharyngodonis</u> belonging to the suborder Heterotrichaea - was found in the intestines of grass carp and common carp at our research station (Klein, 1978; pers. communication).

Mattheis, T. 1967. Vorlaufige Untersuchungen an einsommerigan Graskarpfen (Ctenopharyngodon idella). Dt. Fischerei-Ztg. 14(5):151-157.

Mattheis, T. und K. Odening. 1969. Die Metacerkarie von Apharyngostrigea cornu bei Graskarpfen (<u>Ctenopharyngodon idella</u>) in der DDR. Z. Fischerei NF. 17(587):481-496.

Mokhayer, B. 1976a. The treatment of Bothriocephalosis in grass carp. Fiv. It. Piscic. Ittiop. 11(4):119-121.

1976b. Fish diseases in Iran. Riv. It. Piscic. Ittiop. 11(4):123-128.

- Radulescu, B. und R. Georgescu. 1963. Zitiert nach Nattheis und Odening (1969).
- Shireman, J.V., D.E. Colle and R.W. Rottmann. 1976. Incidence and treatment of columnaris disease in grass carp brood stock. Prog. Fish-Cult. 38(2):116-117.

Szalay, M. and L. Horvat. 1977. Mikrobiologische Probleme bei der Fortpflanzung der pflanzenfressenden Fische. Osterr. Fischerei. 30(11/12): 177-180.

Szakolczai, J. und K. Molnar. 1966. Veterinarmedizinische Untersuchungen an den in Ungarn eingeburgerten pflanzenfressenden Fischarten. Z. Fischerei NF. 14(1/2):139-152.

Vladimirov, V.L. 1969. Method of antibiotics-depot in fish body. Iswestia Gosnior ch. Vol. 69. p. 76-79. Leningrad (In Russian with Engl. summary).

Zobel, H. 1975. Die Entwicklung der Karpfenteichwirtschaft der UdSSR. Z. Binnenfischerei DDR. 22(7):199-207. grass carp.

To prevent secondary infections the following treatments with antibiotics are advisable: by feed, injections or baths (Amlacher 1970, Bohl 1970).

a) Medicated feed - Oxytertracyclin 3500-7500 mg/kg food 10-14 days (50-75 mg/kg body wt, 7-10 days). The lowest concentration which helps against Aeromonas punctata in the blood is 0.1 γ .

b) Injection - Chloramphenicol or Oxytetracyclin 20-25 mg/kg fish in 0.5-1 ml aqua dest. intraperitoneal. The depot-application of antibiotics 400-500 mg Chloramphenicol/kg body weight, mixed with 1 ml mixture of parafin-oil (80%) and Ianolin (20%) intraperitoneal is the best protection against an outbreak of infectious diseases (Vladimirov 1969).

c) Bath - 100 mg/l for severe cases and normally 60-80 mg/l Chloramphenicol up to 36 hours with aeration. Sulfadiazin 100-250 mg/l water, long duration bath. Resistance tests should be taken to determine the most effective therapeutics.

In some cases branchiomycosis broke out. The fungus <u>Branchiomyces</u> sanguinis grew very rapidly; high mortalitys often occurred.

Therapy with quicklime (150-250 kg/ha) or copper sulfate (about 1 mg/l; toxicity depends on water chemistry) can be used.

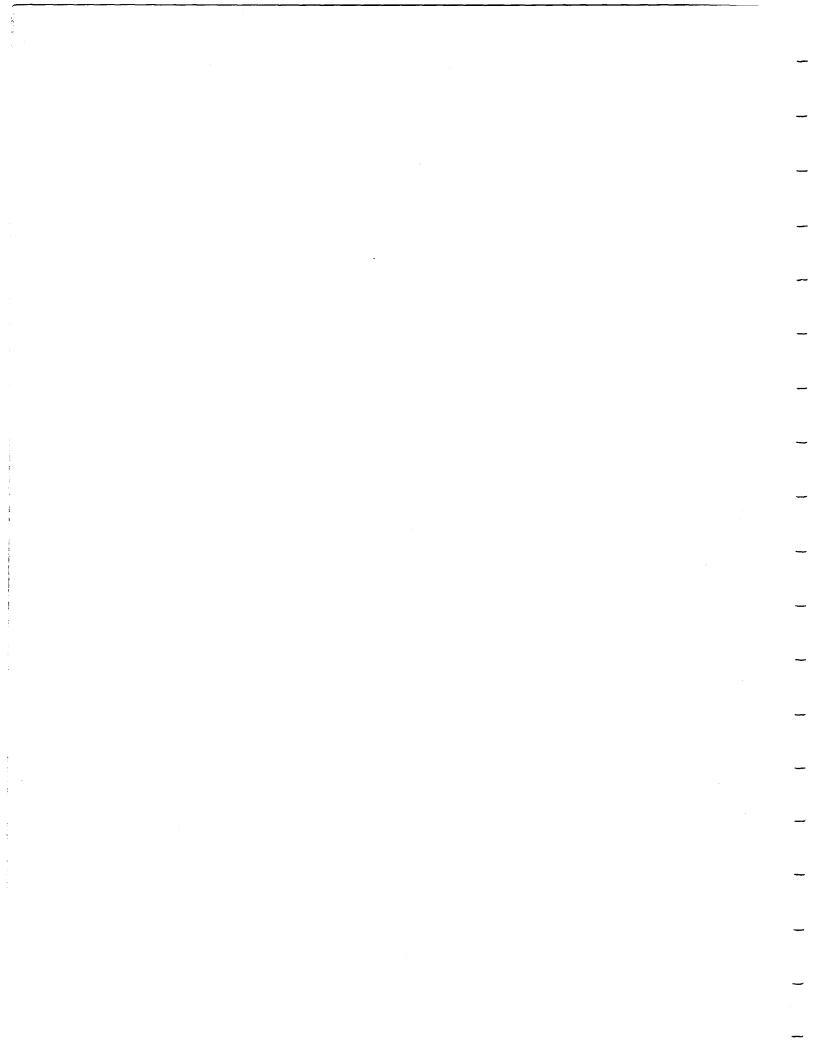
Furthermore, a colleague in Munich isolated a rhabdovirus from grass carp collected in the River Danube. This rhabdovirus was previously unknown (Ahne 1975). This virus is highly infectious (up to 100% mortality) in grass carp fry, but not in older ones and not in carp. It differs in serological characteristics from RVC and shows a resemblance to pike fry virus (Ahne 1977, personal communication). It should also be mentioned that in the United States they have had trouble with columnaris disease (Shireman et al. 1976).

Russian experiences have shown that the RVC is most likely not transferred by eggs and sperm. For this reason, artificial breeding and rearing of fry can take place when hygenic methods are followed. Together with colleague Ahne, from Munich, we shall work on the different aspects of these problems this season.

REPRODUCTION OF GRASS CARP

At our research station we perform experiments on the biotechnical reproduction of grass carp in a warm water recirculation plant. This will enable us to obtain disease-free fry and become independent of imports. Since 1974, grass carp have been bred at Wielenbach. Until recently, our warmwater plant was small and our work was limited. Nevertheless, this year we will start a large warmwater plant. One cycle with activated sludge, two others with trickling filters.

Our present research plant uses a gravel filter system-it is a very simple system, but for our purposes it was sufficient.



REFERENCES

- Ahne, W. 1975. A rhabdovirus isolaged from grass carps (<u>Ctenopharyngodon</u> <u>idella</u>). Arch. Virology 48. p. 181-185.
- Amlacher, E. 1970. Textbook of Fish Diseases. T.F.H. Publications, London.
- Antalfi, A. und I. Tölg. 1971. Graskarpfen Pflanzenfressende Fische Donau-Verlag, Gunzburg.
- Bauer, O.N., V.A. Musselius and J.A. Strelkow. 1969. Die Parasiten und Drankheiten von <u>Ctenopharyngodon idella</u>, <u>Hypopthalmichthys molitrix</u> und <u>Aristichthys nobilis</u> bei der Aufzucht in Teichwirtschaften der UdSSR. Z. Fischerei, NF 17(1-4):205-213.
- Bieniarz, K. and P. Epler. 1976. Preliminary results of the in vivo studies on ovarian reabsorption in carp (<u>Cyprinus carpio L.</u>). J. Fish. Biol. 8(6):449-451.
- Bohl, M. 1967. Erste Erfahrungen mit dem Grasfisch (<u>Ctenopharyngodon idella</u>) in Wielenbach. AFZ 92(21):657-660.

1970. Prophylaxe und Therapie der Infektiosen Bauchwassersucht in "Die Infektiose Bauchwassersucht (IBW) der Karpfen". Munchner Beitrage zur Abwasser-, Fischerei- und Flußbiologie. R. Oldenbourg, Munchen-Wien. Bd. 20:112-129.

1975. Die Kiemenwurmkrankheit (Dactylogyrose, die gefurchteste Parasitose bei der Karpfenbrut). Fisch und Umwelt. 1:67-80.

- Edwards, D.J. and P.M. Hine. 1974. Introduction, preliminary handling, and diseases of grass carp in New Zealand. N.Z.J. Mar. Freshwater Research. 8(3):441-454.
- Faina, R. and O. Par. 1977. The use of the rinsing of the digestive tract of live carps for the diagnosis of bothriocephalosis (In Czechose). Bul. VURH, Vodnany.
- Hnath, J.G. 1970. Di-n-butyl tin oxide as a vermifuge of Eubothrium crassum (Eloch, 1779) in rainbow trout. Prog. Fish-Cult. 32(1):47-50.
- Körting, W. 1975a. Larval development of <u>Bothriocephalus</u> sp. (Cestoda: Pseudophyllidea) from carp (<u>Cyprinus carpio</u> L.) in Germany. J. Fish Biol. 7:727-733.
- 1975b. Aspekte zum Bandwurmbefall. Fisch und Umwelt. 1: 81-87.

Kulow, H. 1973. Sowjetische Erfahrungen uber die Bothriocephalosis, Khawiosis und Philometrosis. Z. Binnenfischerei DDR. 10(9):263-268.

SUMMARY AND CONCLUSIONS

Jerome V. Shireman

Aquatic Weed Research Center

University of Florida

Gainesville, Florida 32611

The opening paper given by Dr. Haller discussed Florida's aquatic weed problems, and interest in grass carp. Grass carp are being evaluated in Florida primarily as a control agent for hydrilla (<u>Hydrilla verticillata</u>) which was introduced during the early 1960's. By 1967 hydrilla was established in approximately 2,000 ha, and by 1977 was found in freshwaters totaling near 250,000 ha. Dr. Haller emphasized the importance of establishing control methods that are not degrading to the environment and methods that will not harm Florida's sport fishery which is a sizable industry.

Dr. von Zon (Holland) stated that aquatic plant control could be achieved by methods based upon the following principles: growth prevention, retardation and cessation. In this meeting we have been concerned with and discussed grass carp as a bio-control agent. The grass carp, according to von Zon, encompasses all three methods as there is a progressive retardation of weed growth, which causes growth to stop and finally weeds are harvested. He stated that the plant control method utilized should be adjusted to each situation and user group and that plant erradication was not advocated. In Holland, grass carp control vegetation more cheaply than either chemical or mechanical control methods, however, the control is not nearly as fast. He continued by stating that grass carp may be unfairly evaluated, as the chronic effects of other control methods (chemical and mechanical) have not been studied. He concluded that the side effects were less severe using grass carp and less irreversible than chemical methods.

Mr. Ware of the Florida Game and Fresh Water Fish Commission expressed great concern over the use of grass carp for vegetation control in Florida waters. This concern is based on the fragility of aquatic ecosystems and he used Lake Apopka in Central Florida as an example. This large lake, because of largescale removal of the vegetation and other environmental changes is characterized by continued phytoplankton blooms. His concern is for the maintenance of good sport fish populations in Florida lakes and feels that indiscriminate removal of aquatic weeds from these systems will degrade the sport fishery.

Other speakers expressed optimism concerning the grass carp as a biocontrol organism. According to Mr. Henderson, Arkansas utilizes the fish routinely for vegetation control and has not seen adverse effects from its usage.

Mr. Stott stated that Great Britain has guarded optimism concerning grass carp, but it is potentially useful for weed control and could be used in conjunction with approved herbicides. Although the environmental impact of the fish is not fully understood, the risk of permanent damage to waters in Great Britain is slight because it is doubtful that natural reproduction will occur. He suggested that more large-scale trials should be attempted by local authorities in his country in order to more fully evaluate the fish.

Consideration should be given to the area into which grass carp are stocked. The paper presented by Miley et al. addressed this problem. Grass carp were stocked into four Florida lakes ranging in size from 5 to 30 acres. Adverse effects to native fish populations were noted in two sparsely vegetated ponds, whereas in a third pond that contained considerable amounts of submerged vegetation (hydrilla) and a large residual grass carp population, a viable sport fish population still existed at the termination of the study. It is unfortunate that base line fishery data was not collected for this pond. The authors concluded that grass carp should not be stocked in ponds containing "normal" or sparse vegetation.

Natural reproduction of grass carp was discussed by several speakers. Stanley, Sutton and Miley summarized a recent European trip. They reported that grass carp reproduced naturally in Russian rivers, but made up a small percentage of the fishes occurring in these rivers. Russian researchers noted, however, that northern pike (Esox lucius) and yellow perch (Perca fluvialtilis) were eliminated in one pond heavily stocked with grass carp. Dr. Haller visited Asia and reported the findings of his trip. Grass carp are reproducing in the Tone River, Japan and apparently reproduced one year in the Ah Tien Reservoir on Twain, natural reproduction has not occurred subsequently in this reservoir. Although Dr. Haller did not visit the Phillipines, another member of the group (Mr. William Bailey) visited the Panpanga River. It was not definite as to whether grass carp reproduced in this river. Mr. Leslie discussed the probability of natural grass carp spawning in Mexico. A trip was made to Mexico by Florida Department of Natural Resources personnel and the conclusion was that grass carp had spawned in the Rhio Balsas River. Dr. Tsuchiya discussed natural grass carp reproduction in the Tone River, Japan. He indicated that the construction of wiers (dams) has changed the spawning site of grass carp and that silver carp now make up a greater percentage of the reproducing Chinese carp populations. The change in composition was due to more favorable conditions for silver carp survival.

From the above discussions it was concluded that the following conditions are necessary for natural reproduction: (1) Adult fish must be in good physiological condition; (2) Water temperature must be above 18 - 19 C; (3) Water velocity must be above 0.8 m/sec; (4) Turbulence near the spawning site is necessary; (5) The river must be at least 50 km long; (6) Fry and fingerlings must have a food source near where hatching occurs; (7) Turbidity increases might be necessary and stimulate spawning through light intensity; and (8) A rise in water level usually precludes spawning which might influence water quality, making conditions more suitable for spawning.

Dr. Tsuchiya also discussed grass carp spawning techniques used at their stations; adult fish over five years of age weighing 5-20 kg are used.