



INTERNATIONAL MILKFISH WORKSHOP CONFERENCE

MAY 19-22, 1976 • TIGBAUAN, ILOILO, PHILIPPINES

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CONFERENCE HIGHLIGHTS

The International Milkfish Workshop Conference, sponsored by the Aquaculture Department, SEAFDEC and assisted by the International Development Research Centre (IDRC) of Canada, was held in the Department's main station at Tigbauan, Iloilo, Philippines as a sequel to the National Bangos Symposium held in Manila from July 25-28, 1975.

This conference aimed to consolidate all existing data on milkfish, plan the directions and magnitude of future research related to milkfish and develop a collaborative program on milkfish research among countries, research institutions and other organizations.

Milkfish, which is widely distributed throughout the Indo-Pacific and South China Region, is considered an important food fish, and extensive culture is currently being done in Indonesia, Philippines and Taiwan. One of the major problems of the industry is the inadequacy of fry supply. Milkfish fry are caught in the wild and the wide fluctuations as to amount and seasonal occurrence have hampered rational planning and investment for the development of the industry. This conference, therefore, is significant. The solution to the fry supply problem would contribute immensely to the development of the milkfish industry, and consequently contribute to the protein requirements of a burgeoning population in the region, and also may become an export commodity, thereby increasing the foreign exchange of the countries of the region.

Working papers were requested from the participants which formed the bases of the discussions in the sessions. Six sessions were organized to determine the existing knowledge and information on milkfish, identify the knowledge gaps and determine the priorities and strategies to bridge these gaps.

Session I discussed the behavior and bio-ecology of milkfish in the wild; Session II, egg and larval surveys; Session III, maturation and reproduction in the wild and captive fish; Session IV, egg incubation and larval rearing; Session V, predators, diseases and physiology of stress, and Session VI, research network for countries and institutions engaged in milkfish research.

The conference agreed on the following highest priorities and the countries/institutions which will implement them: (1) physiological maturation/induced breeding (Taiwan, SEAFDEC, Oceanic Institute); (2) racial studies (a) morphology (Japan), (b) biochemical (Oceanic Institute and Hawaii Institute of Marine Biology); (3) larval rearing

(Taiwan, SEAFDEC, BFAR, UP, Hawaii Institute of Marine Biology, Oceanic Institute, India and Indonesia); (4) migratory and spawning studies (SEAFDEC, Oceanic Institute and Thailand); (5) reference repository (SEAFDEC); (6) sex determination (SEAFDEC); (7) cryogenics for males (Taiwan, SEAFDEC); (8) pituitary bank (SEAFDEC); (9) egg and larval survey (SEAFDEC, UP, BFAR, India and Indonesia).

Capsulated research proposals by the representatives of the different countries/institutions, in attendance, were submitted. The proposals included objectives, methodology, institutions involved, approximate cost and personnel requirements.

INTERNATIONAL MILKFISH WORKSHOP-CONFERENCE
Tigbauan, Iloilo, Philippines
May 19-22, 1976

Sponsored by the
Aquaculture Department of the Southeast Asian Fisheries Development Center,
with the Assistance of the International Development Research Centre

INTRODUCTION

The Importance of Milkfish

Milkfish, Chanos chanos (Forsk.) is an important food fish in the Southeast Asian region. Known for its favorable response to culture both in fresh and brackishwater impoundments, it is one of the major products of aquaculture. Extensive culture of this species has been in vogue in Indonesia, Philippines and Taiwan, while experimental and moderate cultures are presently being undertaken in India, Malaysia, Thailand, Khmer, Singapore and Vietnam.

Schuster indicated that the western limit of the geographic range of distribution of milkfish extends from the Red Sea and the eastern coast of Africa through Indonesian and China Seas to the entire breadth of the Pacific Ocean up to the eastern limit consisting of the southwestern coast of the United States of America and Mexico. Within this latitudinal range, it occurs from as far north as the coast of southern Japan to as far south as the coast of eastern Australia and New Zealand.

Total operational area under aquaculture in Southeast Asia is estimated to be about 630,645 ha with a total production of about 470,000 metric tons and a total potential area for development of 12,217,426 ha with an estimated production of about 9 million m.t. Pillay reported that about 80% of the operational area are devoted to the culture of milkfish and other finfish.* According to Liao, the total area devoted for milkfish culture in Taiwan as of 1974 is more than 16,000 ha with a total annual yield of 28,900 tons.**

Total actual pond area for milkfish production is 390,000 ha although Indonesia, the Philippines and Taiwan have the most extensive production with a combined area of 32,000 ha. Total milkfish production is 180,000 m.t. with the three above countries producing 178,900 m.t.

*The Role of Aquaculture in Fishery Development and Management,
FAO, February 1973.

**Working Paper No. 2, citing Taiwan Fisheries Yearbook of 1975.

In 1972, it was reported that the total fish allowance for the SEAsian region is about 8,624,600 m.t., while total fish production from all sources was only 5,302,200 m.t. or a production deficiency of 38.5%. The average per capita intake of fish is 22.6 kg which is below 36.5 kg per capita as required in the Philippines.

Southeast Asia has been considered as having one of the highest rates of population growth. To meet the nutritional needs of a fast growing population at standard levels necessary for healthy living, all efforts towards increasing food, especially protein sources must be encouraged. Southeast Asia is a traditional rice and fish eating region and the development of fisheries as one of the major sources of protein becomes imperative.

Present Status and Problems of Milkfish in Southeast Asia

The three major countries culturing milkfish on an extensive scale are Indonesia, the Philippines, and Taiwan. While Taiwan reports an average production of about 2,000 kg/ha/year, Indonesia, 340 kg/ha and the Philippines has an average yield of 600 kg/ha/year. Indonesia and the Philippines still largely use traditional methods of culture while Taiwan has reportedly improved her technology.

One of the major constraints in the intensification of the development of the industry is the non-availability of adequate fry supply at the time it is needed for stocking. Presently, fry are collected from the wild at certain seasons of the year. The wide fluctuation as to amount and seasonal occurrence has hampered rational planning for investment and development. The Philippines alone, which has 176,000 ha of milkfish ponds and 6,000 ha of fish pens reported a deficiency of half a billion fry in 1974.

Very little information is available on the biology, habitat, migration patterns and spawning habit of the milkfish. It was only in the recent past that some independent studies on milkfish breeding are being done in the Philippines, Taiwan and Hawaii. Egg and larval surveys are also being conducted in Thailand and South India. A UNDP supported program on milkfish and shrimp is being currently undertaken in Jepara, Indonesia.

National Bangos (Milkfish) Symposium

Cognizant of the meager and scattered information about milkfish, a National Bangos Symposium was organized in the Philippines from July 25-28, 1975 under the joint sponsorship of the Philippine Council for Agriculture and Resources Research (PCARR), Bureau of Fisheries and Aquatic Resources (BFAR), University of the Philippines College of Fisheries (UPCF) and the Aquaculture Department, SEAFDEC. About 100 scientists, researchers, academicians, administrators, and practitioners from the government and private sector convened to review existing local literature and benchmark information as to the state of the technology relating to milkfish culture industry.

The symposium attempted to consolidate existing knowledge and experiences on the bangos industry with the hope that the knowledge gap would be identified as part of an integrated research program for the development of the industry. (Proceedings, National Bangos Symposium, July 25-28, 1975)

International Milkfish Conference

As a sequel to this national symposium, the SEAFDEC Aquaculture Department, with the assistance of the International Development Research Centre (IDRC) of Canada, sponsored this International Milkfish Workshop-Conference held in the Department's main station in Tigbauan, Iloilo, Philippines from May 19-22, 1976.

This conference aimed to achieve the following:

- (1) Review existing data on milkfish
- (2) Help plan the directions and magnitude of future research related to milkfish
- (3) Develop a collaborative program on milkfish research among interested research institutions and other organizations.

Working papers were invited from the participants for indicating the extent of studies undertaken in their respective organizations and countries. Six sessions were organized to determine the existing knowledge and information on milkfish, identify the knowledge gaps, and determine the priorities and strategies to bridge these gaps. Milkfish culture practices were not included in the agenda.

CONFERENCE DISCUSSIONS

The workshop-conference was divided into six major areas. In each session, specific working papers were made bases for discussions. The following are the results of the discussions in the different sessions:

SESSION I

Behavior and bio-ecology of milkfish in the wild. The area was divided into subtopics:

A. Taxonomic consideration in relation to racial or specific variability

Most participants agreed that even with the scanty data available, there are strong indications of more than one race in milkfish. Information available in Indonesia and the Philippines show possible racial differences in body depth, and lateral line scale and vertebrae counts.

Electrophoresis of eye lens nucleus protein patterns is being used in Hawaii to examine the possibility of different races in milkfish. However, additional and more detailed studies on morphological characters, population dynamics, biochemical tests such as isozyme and serum or muscle protein studies are still required to be more conclusive.

B. Migration patterns

In the study of migration patterns, no "marking-recapture" studies have been carried out so far. One telemetry study using ultrasonic tags attached to the sabalo has been carried out on four fish in the Philippines and a similar study is being planned in Hawaii.

C. Oceanographic conditions

Data are being collected in the Philippines, Indonesia, Hawaii and Taiwan on sabalo occurrence. In Hawaii, gill nets are mostly used in offshore waters and no traps are used. Preliminary results suggest that sabalo are found more often in the upper three meters of water and capture is better facilitated in murky waters where vision is reduced. Gill nets are also used in Indonesia for catching sabalo. Most of the spawners are caught just near the surface. Catches in the Philippines indicate a greater number of sabalo caught in otoshi-ami offshore than in fish traps closer to shore. In Taiwan, otoshi-ami is used in catching sabalo. In many areas, the number of female sabalo appears greater than males early in the spawning season but this ratio becomes variable later in the season. Problems in identifying any external sexual dimorphic characters have hindered this work.

D. Age and growth studies

Age of the spawning sabalo caught in the Philippines varied from 3-5 years old. It is suggested that more critical studies on age and growth of sabalo are necessary.

E. Food availability and utilization

The milkfish appear to feed on plankton in all areas examined and on benthos in several areas. Whether it is a selective feeder or not is not yet known. Some evidence indicates that adult milkfish browse on attached algae. Some data indicate that the epibranchial organ is involved in digestion.

There is little information on all the above categories for the juvenile up to the sub-adult in the sea before their return to the proximity of the shore during the spawning season.

SESSION II

Egg and larval surveys

A. Past and present activities on milkfish egg and larval surveys

Ichthyo-plankton surveys have been undertaken by most countries bordering the Indian Ocean, the South China Sea and the Pacific Ocean. Cooperative study programs are likewise being done, like the Indian Ocean Programme (I.O.P.) and the Cooperative Study of the Kurishio and Adjacent Regions (CSK Program in the South China Sea).

Probably because of identification problem, no milkfish eggs and larvae have been reported from ichthyo-planktons collected by large research vessels which usually operate in the open seas. It is believed that the milkfish generally spawn close to shore. Some samples collected by these cooperative study programs were just deposited at some plankton sorting centers waiting to be analyzed. Lack of expertise and appropriate instruments often hamper egg and larval surveys, although this is needed by most of the participating countries.

The participants recommend that future egg and larval surveys should cover inshore and coastal waters where the occurrence of milkfish spawners is reported, taking into account influence of races spawning in different seasons.

It is further recommended that the FAO-UNDP International Indian Ocean Fishery Survey and Development Program (IOP), which has carried out large scale and systematic surveys of the fishery resources of the Western Indian Ocean, should include in its ichthyo-plankton analysis studies on the distribution and abundance of milkfish eggs and larvae.

B. Information from fishermen

The discussion leader asked how far information from fishermen can be relied on. While some fishermen may know some things more than the scientist, the latter should not accept everything said as truth.

Some Filipino fishermen in various parts of the Philippines claim that they know when the milkfish spawn and how spawning is related to the lunar periodicity. On the other hand, fishermen in most other countries offer scientists only limited information which most often must be verified.

C. Discussion of working papers in the session

Full description and fine illustration of milkfish eggs and larvae was deemed necessary. It was also believed important to have more data to prove whether the egg described by Delsman (1929) is the real milkfish egg. Accuracy of identification of the so-called milkfish eggs has to be determined through rearing of eggs to the identifiable stage.

Preliminary data on vertical distribution of the egg, daily fluctuations in catch of the egg in confined areas, developmental stages of the egg by time of collection, and ambient water temperature and salinity were presented.

Likewise, the occurrence of larvae much smaller than fry appearing in the beach was reported from otoshi ami at Pandan; however, oceanographical conditions should also be included.

It was suggested that due consideration be given to the migratory pattern of the fry and fingerlings, like the migratory pattern studies in the Vietnamese waters (Kuronuma, 1962). It was further suggested that future survey programs should give due attention to the standardization of the sampling gear and data collection.

SESSION III

Maturation and reproduction in wild and captive fish

A. Reproductive physiology

Very little has been done on the endocrinology, biochemistry and physiology of adult milkfish due to limited availability of sexually mature fish. The result of a chemical study on mullet eggs throughout hydration (tertiary yolk globule stage to spawning) was presented. It was believed that a similar process can presumably take place in milkfish.

Histological slides of developing gonads obtained from 5-6 year old tank-reared and wild milkfish from Tungkang Marine Lab. Taiwan were also shown.

Histological study of gonad development throughout the year has been started in the Oceanic Institute, Hawaii. Gonads for histological examination are also being processed at the SEAFDEC Laboratory at Tigbauan, and preliminary analysis of the tissue section showed both mature and developing oocytes in the ovary at the same time. In males, either spermatozoa or primary spermatocytes were also found.

In order to better understand the milkfish, it was suggested that more studies along this discipline be undertaken.

B. Gonad development

Slides of the ponds in the island of Hawaii from which sexually mature fish has been obtained were also shown. It was mentioned that of the 52 fish examined, the sex ratio of male to female was 1.2:1. All males were fully mature. Four out of 25 females reached full maturity with eggs of tertiary yolk globule stage. Two other females showed the sign of atresia.

It was also reported that the eggs obtained during the spawning season at Pandan had mean egg diameter of 0.8 mm for non-hydrated eggs and 1.2 mm for hydrated eggs. The Hawaii group presented similar egg diameters.

It was also reported that milkfish stay in Lake Naujan for the first two years and later migrate to the sea as sexually immature fish.

Some reports indicate that the fecundity of the fish ranges from about 1-5 million eggs.

It was recommended that studies be undertaken on the environmental conditions and nutritional requirements for rearing and maintaining brood stock.

C. Mating behavior and fertilization of wild fish

No report was presented on the breeding behavior of milkfish or on successful artificial fertilization and development of milkfish eggs.

D. Natural and/or induced spawning of captive fish

No natural spawning of captive milkfish has been reported. Induced spawning has been attempted by the Hawaii, Taiwan and Philippine groups. To date, the groups from Hawaii and the Philippines, using partially purified salmon pituitary gonadotropin (SG-G100) have obtained spawned eggs which were not fertilized. The group from Taiwan, using pituitary hormones and synahorin has obtained maturing eggs in captive fish.

It was explained that SG-G100 is the most potent and reliable hormone to be used not only for egg maturation but also for induced spawning. The standardization of induced spawning of milkfish by SG-G100 was proposed and a regular program of supplying SG-G100 was recommended. Regular collection of pituitary glands from dead sabalo was also suggested.

E. Examination of spawned fish

Examination of spawned fish in the Philippines suggested that milkfish are partially synchronous spawners and multiple spawning may occur in a year. However, it was postulated that this multiple spawning of fish observed in the Southeast Asian region might be due to racial or population differences throughout the milkfish region.

In order to control gonadal development and achieve success in induced spawning in milkfish, understanding of its environmental conditions and nutritional requirements must be undertaken. The importance of tagging experiments and oceanographic surveys to locate spawning grounds and monitor environmental conditions was further emphasized. Techniques of capture, handling and transport of sabalo spawners in Pandan and Tigbauan, Philippines and Hawaii were also discussed.

SESSION IV

Egg incubation and larval rearing

A. Egg incubation

According to Delsman (1929), Chanos eggs are normally about 1.2 mm in diameter and hatch in about 24 hours. The larvae at hatching are about 3 mm in length and grow to about 5 mm in three days when it is ready to feed. When it is 10 days old, it reaches 10 mm in length. At 10-13 mm, they are caught as fry in the commercial fry fishery. The fry grow to 20 mm in about 5 weeks and at this stage the fish lost its larval properties and resemble and behave as juvenile fish.

Information from the collection of milkfish eggs in the waters around Panay Island indicated that although more eggs were collected at surface layers, some were also collected at deeper layers down to 20 meters. The incubation of some of these eggs was found to be about 24 hours which agrees with Delsman's findings. The newly hatched larvae were observed hanging upside down in water and the yolk was used as feed for 2-3 days.

B. Larval rearing

Little, if any, work has been done on larval rearing of *Chanos* from the egg and nutritional requirements of larvae between 5-10 mm are unknown. In the absence of information on food for milkfish between 5-10 mm, participants discussed feeding methods for mullet larvae where information is available. The possibility of feeding larvae on microencapsulated feeds was mentioned and encouraging studies along this line were reported.

Although nutritional requirements for larvae of 10 mm or more in length is unknown, various dried feeds, such as rice bran, dried egg yolk, etc. have been used with practical success. Recent studies in the Philippines showed that Chlorella (a microscopic alga) may be successfully used as food for *Chanos* fry. It was suggested that if early larva (5-10 mm) can also be reared with Chlorella as food, the feeding problem may be solved. The relatively large size of milkfish egg also indicates that rearing early larvae may be easier than with many other marine fish species. However, there is a great need for nutritional studies for all larval stages of this fish.

Discussions were made on the problem of fry mortality from collection to stocking in ponds. Reported average survival rates range from 50% in the Philippines to 95% in Taiwan. Much of this mortality are attributed to methods of collection and handling, predators such as Megalops, Elops, Ophiocephalus, eels, sea snakes that are collected with the fry were mentioned. The difficulty of separating the *Chanos* fry from that of predators was identified as a major problem. The importance of clean facilities, proper care in handling such as avoiding salinity and temperature shock, was stressed. Another problem was poor handling by middlemen who sell fry to the former.

SEAFDEC is currently conducting experiments on "seed banks" for holding *Chanos* fry up to stocking size to increase resistance in handling and to make fry continuously available. One question was raised as to the optimal size for fry for better handling.

The salinity tolerance was reported to be 15% below and 10% above the salinity at which the milkfish fry were living.

SESSION V

Predators, diseases and physiology of stress

A. Superficial injuries following capture

General treatments are usually given following injury to captured fish by using fungicides, bacteriocides, or all-purpose compounds.

B. Shock or stress following capture and handling

Major noticeable symptoms of shock or stress is opaqueness of the covering of the eye with milky film. There is some evidence of gram negative bacterial infection but does not respond to treatment of usual antibiotics. Other symptoms might be gastritis of the stomach and possible heart attack following occlusion of oxygen to brain and fibrillation of the heart. Most effective treatment is to reduce salinity to about 20 ppt for 2-3 weeks. More studies in this field are recommended.

C. General fish diseases

Typical diseases of farmed animals are not yet recorded in literature. Studies are suggested along this field.

D. Parasites

Limited reports of both internal and external parasites are available in existing literature. It is, therefore, necessary to have more systematic studies in this field.

Significant observations have been made during the last 12 months and information on milkfish health and husbandry has increased dramatically. There is, therefore, a continuous need to have standards for the collection and storage of materials including information on origin of materials, such as pond fish, wild fish, environmental data, etc.

SESSION VI

Research network for countries and institutions engaged in milkfish research

A. The network

The delegates by country and by research institutions were consulted on the possibility of establishing a research network and a general consensus was arrived at on the following points:

1. Collaborative research and continuous exchange of information will be maintained among researchers and research institutions.

2. Some kind of arrangement, formal or informal, may be drafted for implementation of the collaborative research and exchange of information.

3. Complete listing of research topics was drafted indicating the countries/institutions' involvement including unilateral or joint action.

4. Listing of high priority research projects was drafted indicating the country/institution having the facilities and expertise.

5. An action research program for the first eight priority areas will be drafted by each country/institution delegate. Under each research area, a consolidated program may be eventually drafted and submitted to the conference chairman.

B. Possible research areas

The conference made a list of research areas, indicating their priority ranking and the countries/institutions which may undertake the study either individually or collectively.

A tabulated list of areas is hereby included as follows:

<u>Priority</u>	<u>Work Areas</u>	<u>Who is working at present</u>
1	Maturation/induced breeding	ABGIK
1	Racial studies (morphology)	ABFG
1	Migratory and spawning activities	BG
1	Larval rearing	ABCEGIKF
1	Reference repository (literature)	B
1	Sex determination	BJ
1	Cryogenics for males	AB
1	Pituitary bank	B
1	Egg and larval surveys (identification)	BCFHI
2	Nutrition (broodstock)	ABCEGI
2	Capture methods and transport of fry	ABCEIK
2	Capture of adults	BGE
3 & 2	Parasites and diseases	GI
3	Egg biochemistry	G
3	Stress (shock)	GFA

Legend:	A - TML (Taiwan)	G - OI (Hawaii)
	B - SEAFDEC	H - CMFRI (India)
	C - UPCF	- CIFRI (India)
	D - PCARR	I - SCRC (Indonesia)
	E - BFAR	J - JAPAN
	F - HIMB (Hawaii)	K - TDOF (Thailand)

CONFERENCE RECOMMENDATIONS

The final session consolidated the different recommendations on priorities and strategies to be undertaken. From the nine priorities assigned to the different countries and institutions, the following studies, in capsulated proposals, were prepared and submitted by the delegates of the country/institution concerned which included objectives, methodology, institutions involved, approximate costs, personnel and publications.

The titles of the different studies are:

1. Physiological maturation/induced breeding
2. Racial studies
 - a) Morphology
 - b) Biochemical
3. Larval rearing
4. Migratory and spawning studies
5. Reference repository
6. Sex determination
7. Cryogenics for males
8. Pituitary bank
9. Egg and larval survey

The individual capsulated proposals follow in the succeeding pages.

STUDY TITLE NO. 1 Reproductive Physiology and Induced Breeding

DURATION 5 Years

RESEARCH OBJECTIVES

1. To define the optimum spawning procedure (for salmon gonadotropin, specifying correct time for treatment, dosage, dose rate, response, etc.)
2. To determine the cost effectiveness of readily available hormones.

METHODOLOGY

1. By determination of natural reproductive physiology (for both sexes) from immaturity.
2. By determination of responses to hormone treatment.
3. By testing reactions to salmon pituitary gonadotropin.
4. By testing reactions to other cheaper hormones.

INSTITUTIONS INVOLVED

Tungkang Marine Laboratory
SEAFDEC Pandan
Oceanic Institute
SCRC (Indonesia)

APPROXIMATE COSTS

Capital - approximately \$200,000
Recurrent - approximately \$400,000/year

TRAINING

The training program will be arranged whenever necessary, both short term (2-3 months) and long term (MSC PhD).

PERSONNEL

Present personnel in ongoing projects in above organization.

PUBLICATIONS

As available.

STUDY TITLE NO. 2-A Racial Study (Morphology)

DURATION 3-5 Years

RESEARCH OBJECTIVES

The milkfish ranging widely in the Indo-Pacific waters from the Red Sea to the Mexican Coast is believed represented by different races geographically or population micro-geographically. The races or populations have to be identified precisely in order to establish the basic ground on which all the biological problems are constructed.

METHODOLOGY

A format with detailed instruction is established as the start line of the study. Secondly, the document above is circulated to the institutions involved requesting the data filled in. Thirdly, the data are analyzed by the hand of the institution selected.

INSTITUTIONS INVOLVED

A number of them will be involved. At present, they will not be named except those mentioned during the conference.

APPROXIMATE COSTS

Capital -
Recurrent - \$2,000/year

TRAINING

PERSONNEL

2 bio-mathematicians

PUBLICATIONS

Scientific Journals available

STUDY TITLE NO. 2-B Biochemical Taxonomy of Chanos chanos

DURATION 5 Years

RESEARCH OBJECTIVES

To ascertain racial differentiation of Chanos chanos throughout its distribution range and thereby assist in selective breeding of the fish.

METHODOLOGY

Electrophoretic and other appropriate biochemical techniques

INSTITUTIONS INVOLVED

Oceanic Institute, Hawaii
Hawaii Institute of Marine Biology

APPROXIMATE COSTS

Capital - none
Recurrent - \$30,000/year for 5 years (\$150,000)
(training, travel, 1/2 time technicians,
collection and preparation of material)

TRAINING

One trainee/year to Hawaii

PERSONNEL

Dr. J. Kuo
Dr. A. Smith
Dr. J. Shacklee

PUBLICATIONS

To be expected through Scientific Literature

STUDY TITLE NO. 3 Larval Rearing

DURATION

Objective I - 3 years (after spawning techniques developed) Simul-
Objective II - 3 years (" " " ") taneous

RESEARCH OBJECTIVES

1. To develop rearing techniques for larvae between hatching and 10 mm (Induced spawning may be prerequisite) - 3 yrs.
2. To improve larval rearing techniques from 10 mm to metamorphosis. (Wild-caught fry can be used) - 3 yrs.

METHODOLOGY

1. Conduct studies of empirical feed development, nutritional requirements, maintaining optimal water quality, determining optimal physical conditions, rearing tank design.
2. Conduct studies of empirical feed development, nutritional requirements, optimal holding facilities, density, diseases of wild-caught fry, optimal physical conditions.

INSTITUTIONS INVOLVED

1. Tungkang Marine Laboratory, Taiwan, Oceanic Institute, SEAFDEC, HIMB (advisory), Indonesia
2. Tungkang Marine Laboratory, Taiwan, Oceanic Institute, SEAFDEC, HIMB (advisory), UP, BFAR, India (CIFRI, CMFRI), Indonesia (SCRC)

APPROXIMATE COSTS (\$ US) PER YEAR*

	TML	OI	SEAFDEC	HIMB	SCRC	UP	BFAR	INDIA
Capital	20,000	0	0	0	10,000	2-3,000	₱50,000	21,000
Recurrent	5,000	0	0	5,000	25,000			4,000

TRAINING

After research had led to improved larval rearing techniques, training programs at the institutions actively involved are strongly indicated.

* '0' indicates that ongoing projects in these areas are already funded.

Study Title No. 3 (Cont'd.)

PERSONNEL

TML	OI	SEAFDEC	HIMB	SCRC	UP	BFAR	INDIA
5	3	2	1	1 scientist 2 junior re- searchers	2 scien- tists 1 tech- nician	4-sci- entists 2 labor- ers 2 guards	2 scien- tists 4 junior scien- tists Field Staff

PUBLICATIONS

Reports and journal articles.

STUDY TITLE NC. 4 Migration and Spawning

DURATION 3 Years

RESEARCH OBJECTIVES

1. To establish the environmental conditions for spawning.
2. To understand the life history and migratory patterns of the fish.
3. To simulate the conditions in the hatchery for establishing broodstock in captivity.

METHODOLOGY

1. By determination of natural locations of spawning sites through tagging and tracking.
2. By determination of seasonal migrations of fish through tagging.
3. By experimental manipulation in the hatchery of environments.
4. By study of plankton in probable spawning areas.

INSTITUTIONS INVOLVED

SEAFDEC Pandan
Oceanic Institute

APPROXIMATE COSTS

Capital - \$50,000 Tag development and testing
per field operation
Recurrent - \$15,000 (including tag purchases).

TRAINING

Simple instructions for field operators following nomination of project leader(s).

PERSONNEL

US/Canada based development team.
One or two national project leaders.
Field operators (and local fishermen).

PUBLICATIONS

STUDY TITLE NO. 5 Reference Repository

DURATION 5 Years

OBJECTIVES

1. To collect all published literature on milkfish.
2. To build up a Regional reference library which would be the center from where information on publications on milkfish research/reprints could be made available to research workers/ Institutions in Southeast Asian countries and other countries engaged in milkfish research.
3. The Reference repository will also collect literature, scientific reports and publications on other related species.

METHODOLOGY

1. An updated bibliography on milkfish will be prepared from available published bibliographies, biological abstracts, Aquaculture Abstracts and Proceedings of National and International Symposia on milkfish.
2. Index card will be prepared on Authors as well as on different subjects and items of research on milkfish.
3. Authors of published papers/publishers will be requested to send reprints which may be obtained free/purchased or photostat copies obtained of reprints which are not readily available.
4. Lists of literature/publications available at the Reference repository will be sent monthly/quarterly to all Institutions engaged in milkfish research.
5. Xerox copies of relevant papers will be sent to researcher at their request.
6. Occasionally annotated bibliography of available literature will be circulated to milkfish researchers.
7. Invite workers from various institutions to supply any additional information on milkfish work and papers published for inclusion in the Reference library.

Study Title No. 5 (Cont'd.)

INSTITUTIONS INVOLVED

The reference repository will be established at SEAFDEC at its Tigbauan Research Station, Iloilo, Philippines.

All other Institutions engaged in milkfish research will be involved.

APPROXIMATE COSTS

Capital - US \$60,000

Recurrent - US \$130,000 (Total for 5 years)

TRAINING

- a. Librarian may be sent for training abroad for a period of 3 months.
- b. Library assistant may receive local training for a period of 3 months.

PERSONNEL

1. One Editor for editing publications, newsletter, bulletin, etc.
2. One Librarian
3. One Library Assistant
4. One Typist and one duplicator

PUBLICATION

The Reference repository may bring out newsletters, annotated bibliography and bulletins to disseminate up-to-date information on the on-going research and progress made on milkfish research in various Institutions/countries.

STUDY TITLE NO. 6 Sex Determination

DURATION 2 Years

RESEARCH OBJECTIVES

To determine the sex of milkfish from external characteristics.

METHODOLOGY

Examination of external morphology of all milkfish killed during capture or transport and of those which die or are killed in holding tanks. Determination of the sex of these fish on autopsy. Correlation of sex with morphological data.

INSTITUTIONS INVOLVED

All Institutions which obtain milkfish.

APPROXIMATE COSTS

Capital No extra costs. Part of ongoing projects.

Recurrent

TRAINING

Short-term training (2-3 months)

PERSONNEL

Present personnel.

PUBLICATIONS

As available.

STUDY TITLE NO. 7 Cryogenic Preservation of Milkfish Sperm

DURATION More than five (5) years

RESEARCH OBJECTIVES

The main purpose of this study is to ensure the availability of milkfish semen anywhere and anytime it is needed.

METHODOLOGY

Same method which is used for preservation of sperm from other fish.

INSTITUTIONS INVOLVED

SEAFDEC, TML (Taiwan)

APPROXIMATE COSTS

Capital - US \$4,000

Recurrent - US \$2,000/year

TRAINING

Short-term training (2-3 months)

PERSONNEL

Present personnel.

PUBLICATIONS

As available.

STUDY TITLE NO. 8 Pituitary Bank

DURATION 2 Years

RESEARCH OBJECTIVES

To develop adequate quantities standardized pituitary gonadotropin preparations suitable for least cost breeding of milkfish and other food fishes.

METHODOLOGY

1. Collection of salmon pituitaries.
2. Collection and preservation of pituitary glands from all dead mature milkfish
3. Collection of pituitaries from common carp and other fishes.
4. Preparation of various levels of fractionated pituitaries.
5. Laboratory and field testing of above.

INSTITUTIONS INVOLVED

1. IDRC (B.C. Research)
2. SEAFDEC
3. Oceanic Institute/TML

APPROXIMATE COSTS

Capital

Recurrent - US \$200,000/year

Total - US \$400,000

TRAINING

Training of staff in field testing.

PERSONNEL

1 Endocrinologist 2 mm

PUBLICATIONS

One field and laboratory results.

STUDY TITLE NO. 9 Egg and Larval Survey

DURATION 3 Years

RESEARCH OBJECTIVES

1. To determine the natural locations of spawning grounds.
2. To establish the environmental condition, both topographical and oceanographical, for spawning.
3. To establish fluctuation in spawning activity in relation to lunar/tidal phase.
4. To understand early life history and precise knowledge on proper identification of egg and larvae.
5. Laboratory rearings of larvae obtained from the eggs to the identifiable stage.

METHODOLOGY

1. By larval net sampling, both horizontal tows at different layers and vertical tows.
2. Collected eggs and larvae to be described and reared to the identifiable stage.

INSTITUTIONS INVOLVED

SEAFDEC, Other institutions interested are UPCF, BFAR, CMFRI and CIFRI (India), SCRC (Indonesia)

APPROXIMATE COSTS

Capital

Recurrent - US \$20,000/year

TRAINING

Field operator regarding towing the net - 1 year
Laboratory assistant regarding sorting - 1 year
Scientist - identification of major group of fish eggs - 3 months

PERSONNEL

Two scientists, field operators and crew (or local fishermen)
Two laboratory assistants

PUBLICATIONS

WELCOME ADDRESS

by

Domiciano K. Villaluz*

On behalf of the Aquaculture Department of the Southeast Asian Fisheries Development Center and the International Development Research Centre of Canada, who are jointly sponsoring this First International Milkfish Workshop Conference, I bid you a warm welcome. It is also my pleasure to convey to you the cordial greetings of our staff who feel highly honored to be host to such a distinguished group of scientists -- world renown in the science of aquaculture.

We are especially thankful to those of you who have detoured from your regular route to attend the FAO KYOTO Technical Conference on Aquaculture on May 26 to June 2, in order to participate in this workshop and share with us your knowledge in the breeding and cultivation of fishes under controlled conditions, with emphasis on milkfish, Chanos chanos Forskal.

Milkfish or bangos is the major food fish in this country and is the mainstay of our fishpond and fishpen industry. This is also extensively cultivated in our neighboring countries, particularly in Indonesia and in Taiwan. In the Philippines, over 170,000 hectares of fishponds and 6,000 hectares of fishpens are devoted to milkfish cultivation. The rate of harvest in fishponds is reported to be from 800 to 2,000 kilos per hectare, while in Laguna de Bay, harvest from fishpens ranges from 5,000 to 10,000 kilograms per hectare. The most critical problem that faces the fishpond and fishpen industry of the Philippines today is the inadequacy of milkfish fry supply, for nature alone cannot cope with the needs of the industry's fast expansion and development. Only through spawning and propagation under controlled conditions may we be able to remedy the situation.

Studies on the spawning of Sabalo (milkfish spawner) have been undertaken as early as 1937 in the Philippines. The milkfish seed production project of the SEAFDEC Aquaculture Department, with financial assistance from the International Development Research Centre, was started in May, 1975. Pandan was chosen as the project site

*Chief, Aquaculture Department of the Southeast Asian Fisheries Development Center (SEAFDEC).

because of the presence of an otoshi-ami, which was reported to have caught a big number of Sabalo every year, the peak season of which is from March to May. The main objectives of the project include the following: (1) To determine the spawning areas and the pattern of migration of larvae and fry of milkfish; (2) To monitor the ecological conditions of the area under investigation; (3) To develop techniques for catching, handling, transporting and domestication of Sabalo; (4) To determine the sexes externally and the different stages of gonadal development; (5) To breed sexually mature Sabalo under confinement and to induce breeding with the use of gonadotropin and other hormonal extracts; and (6) To hatch the eggs and rear the fry produced from breeding experiments.

Those of you who have seen the Pandan project must now be aware of our problems and our need for further knowledge and expertise. With your presence, we hope that from our combined wisdom and experiences, there may arise some concrete and effective plan of cooperation for this kind of research. We expect that our cooperative efforts shall lead to a better understanding of the work being undertaken by our own different laboratories. We have to recognize the fact that scientific isolation has no more place in the world today. All of us are aware that science and technology have increased the opportunities for international understanding and cooperation by reducing the distance between nations, as witnessed by some of you who have travelled from your own countries to the Philippines.

We hope that during this conference, we shall develop confidence to enable us to strengthen our faith in the efficacy of international collaboration in our search for solutions to common problems; that we shall dedicate the application, that is the practical application of our science, with goodwill for mankind; and that the impact of our deliberations will prove, beyond doubt, the decisive role of scientific research in the achievement of progress, so that aquaculture science would gain more, not only moral, but especially, material support from funding institutions, including governments.

COMMENTS ON THE INTERNATIONAL MILKFISH
WORKSHOP-CONFERENCE

by

Andres Mane*

On behalf of the private sector and on my own, I wish to congratulate the conferees in bringing the workshop to a successful end. The meetings have been very orderly and cooperation, sincerity, and goodwill have pervaded in all the sessions. The participants have contributed their share unselfishly in the resolution of the issues under consideration which more than anything else was responsible to the success of the conference.

I would like also to express our appreciation and gratitude to SEAFDEC and IDRC for sponsoring this International Milkfish Workshop-Conference and in bringing in a galaxy of talents that participated. This conference has brought the cause of the bangos industry on an international level. It should be remembered that a national bangos symposium had been held from July 25-28, 1975 co-sponsored by SEAFDEC with almost the same objectives.

It will be desirable and the industry will welcome it very much if conferences like this could be held regularly on a continuing basis.

It is, indeed, very gratifying that this International Milkfish Workshop-Conference was conceived and carried out. The bangos industry, more than ever, is faced with the serious lack of seed-fish for growing to commercial sizes. This holds back and even retards its development and progress.

In the Philippines, milkfish occupies a premiere position as far as production of fish in general is concerned. It also plays an important role in the economy and well-being of our people. In 1974 for example, the production of this fish was about 100 million tons, second only to that of the round scad (galunggong) produced by the commercial fisheries, which was 166 million tons. It is estimated that about 200 thousand people are employed directly in the production of bangos and about

* Former Commissioner, Philippine Fisheries Commission and presently member, Board of Directors, Fishpen Owners Association of the Philippines.

five times that number in the secondary and tertiary industries. A start has been made to export the fish in order to participate in earning foreign exchange badly needed in the economic development of the country.

In view of the importance of the bangos, its production has been highly specialized such that certain segments have become distinct industries in themselves. Among these are: the bangos fish fry industry, the fingerling or bangos nursery industry, and the fishpond and fishpen rearing industries. These industries are beset with problems inherent to each but most serious of all and which forms the common denominator is the lack of seed-fish. These problems, I am sure, are not only true in the Philippines but in other bangos producing countries as well.

The bangos fry produced in the country in 1974 that went into production stream was estimated at 900 million. In that year, the requirement at 600 kg production per hectare was 1.4 billion, a deficiency of 500 million or 36 per cent; at 800 kg production per hectare, the requirement would have been 1.72 billion, a deficiency of 800 million or 48 per cent; and at 1,000 kg production per hectare the requirement would be 2.14 billion, a deficiency of 1.24 billion or 53 per cent.

Indications are that the production of bangos fry in this country has about reached its maximum level as the fishery, which is its only source, has already attained full maturity. In the face of improved technology and expanded areas of production, the problem of lack of seeds becomes more and more acute, if not critical.

It is in the solution of this problem that the private sector is greatly interested in. It, therefore, looks at this symposium with the greatest hopes and expectations, that its problem may eventually find solutions. The efforts at controlling the propagation of bangos fry through induced breeding that had been deliberated upon in the meetings has been very heartening to us, especially, with the initial success along this line in the experiments in Pandan.

It is a source of gratification that proposals and recommendations had been advanced for the international collaboration and coordination of the researchers in the propagation and induced spawning of bangos.

We in the private sector, therefore, are entertaining the highest hopes that the resolution and recommendations that have been arrived at in this conference would be implemented soonest by action programs. We are greatly consoled that, in the not too distant future, our problem of lack of seed fish shall have been solved.

CLOSING ADDRESS

by

Felix R. Gonzales*

It is a great honor on my part to have been invited to this gathering of distinguished scientists and experts and share with you my own thoughts about the significance of this Conference to the development of milkfish industry in Southeast Asia particularly the Philippines.

We in the Philippines have shown keen interest in the outcome of this Conference especially in terms of new directions in research which this Conference aims to provide in view of the importance of milkfish as a food fish among the masses in the Southeast Asian region.

As you are aware, our country is endowed with the natural conditions most suitable for milkfish culture and our people depend mainly on milkfish as a source of protein in their diet. With an estimated annual production of 100,000 metric tons, our milkfish industry alone generates a total of P425 million per year.

Cognizant of the vital role of the milkfish industry in the Philippine economy, President Ferdinand E. Marcos has expanded his support to the fisheries sector as evidenced, among others, by the huge yearly contribution of the Philippine Government to the SEAFDEC Aquaculture Department. President Marcos has also increased the budget of the Bureau of Fisheries and Aquatic Resources from P19 million in 1973 to P54 million in 1976, or an increase of 65% over a period of three years. Moreover, President Marcos issued in May 1974 a decree designed to accelerate the development of the fishing industry. The necessary incentives for the full development of the milkfish industry have been provided by the Philippine Government and it is for us in the fisheries sector to do our part to pursue this national objective with fervor and zeal.

A prediction has been made that by the year 2000 the total fish consumption in the Philippines per year will increase from the present 1.3 million metric tons to 2.4 million metric tons, or an increase of 46% over a period of 24 years. This means that we in the Philippines must come up with an average incremental fish production of about

* Director, Bureau of Fisheries and Aquatic Resources (Philippines) and concurrently SEAFDEC Council Director for the Philippines.

100,000 metric tons per year to meet the fish requirements in the year 2000. Efforts must, therefore, be made to develop the necessary technology for the mass cultivation and propagation of milkfish fry in this region; unless this problem is solved, the milkfish industry will be unable to meet this targetted incremental production per unit hectare.

On behalf of the Philippine Government and the SEAFDEC Aquaculture Department, I wish to thank the International Development Research Centre (IDRC) of Canada for its generous assistance in funding milkfish studies currently being undertaken at the SEAFDEC Aquaculture Department. Hopefully, with the success of these studies the Department will be able to perform its mandate of sharing the necessary technology to other SEAFDEC member countries as well as other countries in need of such technology.

Lastly, on behalf of the organizers of this Conference, may I extend the congratulations to each one of you for the insights you have contributed to the problems of milkfish research and development. We appreciate your personal concern in providing new information to fill in the gap in our knowledge of the biology of the milkfish because your efforts in this regard will help solve certain production problems that presently hamper the milkfish industry. With your fruitful and enriching participation, this Conference will help pave the way for the removal of obstacles to insure adequate fish supply in the future.

ONGOING RESEARCH STUDIES ON MATURATION AND SPAWNING
OF MILKFISH, CHANOS CHANOS AT THE BRACKISHWATER
SHRIMP AND MILKFISH CULTURE APPLIED RESEARCH
AND TRAINING PROJECT, JEPARA, INDONESIA

by

K. H. Alikunhi*

Abstract

The paper gives an account of the research work carried out at Jepara, Indonesia, on induction of maturity of milkfish in ponds and enclosures, and procurement of the spawners from the wild for seed production by hypophysation. Seven to eight years old pond grown milkfish were found sexually immature. Experiments are being conducted for growing and inducing maturity in 1-2 years old milkfish in fertilized ponds with regular tidal flow of water and also under regular hypophysation program. Milkfish spawners collected from sea had a few males in oozing condition and females mostly spent.

Introduction

The UNDP/FAO project on Brackish Water Shrimp and Milkfish Culture Applied Research and Training, located at the Shrimp Culture Research Centre, Jepara (Central Java) Indonesia is actively involved in studies on maturation and spawning of milkfish as a dependable method for augmenting seed production of this widely cultivated fish. Cognizant of the limited natural supply of milkfish seed, the project, with the active support of two FAO fishery biologists, is actively engaged in:

- i) prospecting and locating new fry collection centres;
- ii) improving collection gear for increasing fry collections;
- iii) attempting induction of maturity of milkfish in ponds and enclosures;
- iv) collecting milkfish spawners from the sea for hypophysation and fry production.

Activities under items iii and iv above are pursued under the leadership of M. K. K. Sukumaran, Fishery Biologist (Induced Breeding) and the following information is extracted from the report he is now preparing.

*Project Manager, UNDP/FAO, Brackish Water Shrimp and Milkfish Culture Applied Research and Training, Jalan Taman Kartini, P. O. Box 1, Jepara, Central Java, Indonesia.

Induction of Maturity in Ponds

Large milkfish, traditionally grown in deep tambaks, in east Java, were examined during the season when similar size specimens in the sea were ordinarily sexually mature. One lot of 20 specimens examined, ranged from 560 mm/1.4 kg. to 1030 mm/10.5kg. None of these specimens was sexually mature. Some of the specimens appeared markedly stout and weighed heavier than normal specimens of corresponding size; e.g., 950 mm/12 kg; 800 mm/9.5 kg; 600 mm/3.25 kg. Dissection and examination of such specimens showed a remarkable increase in fat deposition, particularly on the abdominal wall. According to pond owners, the large specimens weighing 10-12 kg are 7-8 years old.

Though information so far available does not show that Chanos normally attains sexual maturity in enclosed brackishwater ponds, there are stray records from such waters, of specimens showing early stages of gonadal development. Experiments were, therefore, initiated holding 1 1/2 year old milkfish, weighing 600 g and over in selected tambaks with regular exchange of tidal water, under a specified regime of fertilizer inputs and monthly injection of carp pituitary gland extract, for increasing growth and possible induction of gonadal maturation. Under identical conditions of habitat and population the injected lot shows better growth.

Larger specimens weighing up to 1.5 kg and 2 1/2 years old are also being held in ponds with regular tidal flow of water to see if gonads will respond to regular fertilizer inputs and program of compounded artificial feeds. Two ponds, each one hectare in area, with 1.0 to 1.2 meters depth of water, under a program of regular manuring with organic and inorganic fertilizers have also been stocked with year-old milkfish to grow them to larger size for further studies.

Floating Enclosures

A floating enclosure, 6 m x 6 m x 6 m, with an extra 1.5 m height of netting projecting above water level, to prevent fish from jumping out, was put up in the sea near the Research Centre and 1.5 years old milkfish were introduced into the enclosure at the rate of 1/sq m. The enclosure was made of (2.54 cm) mesh polyethylene netting. Large plastic cans and bamboo frames were used to keep the enclosure floating. Fish were daily fed with a compounded, artificial feed cake, kept in a feeding tray. They readily accepted the feed and markedly improved in size during a period of three months, when heavy monsoon rains and wind badly buffeted the enclosure in the open,

unsheltered location and enabled the fish to escape. A new, more sheltered location for the enclosures has now been selected and four enclosures are being commissioned for further trials.

Spawners Sanctuary

To produce large number of spawners for commercial operations and to make them available when required, large impoundments in suitable, sheltered locations along the coastal sea and stocking them with yearling or even bigger specimens of milkfish appear promising. A program of developing such a sanctuary of Karimun Jawa, about 90 km north-west of Jepara, in the Jawa sea is now under active consideration.

Collection of Spawners from the Sea for Hypophysation

Schools of milkfish spawners have been reported from several parts of the coastal sea around the Indonesian archipelago, though there are no records of milkfish among sea fish landings. Arrangements made with provincial fishery administrations and fishermen resulted in the procurement of milkfish spawners. The details are given in Table 1.

Table 1. Details of milkfish spawners procured from three centers.

Place of Procurement	Date	Total Length (mm)	Weight (kg)	Sex	Remarks
South Sulawesi					
	12-12-72	103	9.0	Male	
	12-12-72	105	10.2	Female	
	12-12-72	102	8.7	Female	
	11-17-73	107.5	11.2	Males	Four male specimens studied
		to	to		
	11-17-73	115	12.8		
	11-17-73	103	10.6	Female	Weight of ovaries: 1472 g
	11-17-73	112.5	12.3	Female	Spent?
Tayu, C. Jawa					
	6- 9-74	107.5	10.0	--	
Manco, C. Jawa					
	9-11-75	96	6.35	Male	Oozing; testes: 155 g
	9-19-75	93.5	6.5	Male	Oozing; testes: 390 g
	10-20-75	101	9.0	Female	Ovaries: 450 g
	11-21-75	91	7.2	Female	Spent

Milkfish spawners are reported frequently caught by Karimun Jawa and dried/smoked ovaries examined indicated that the specimens were in fairly advanced stages of maturity.

Fishing for spawners was also organized at Karimun Jawa from May, 1975, using 4-7 inch (10.2-17.8 cm) bar gill nets, made of 210 D/12 - 15 nylon twine. So far 17 specimens were caught, of which 7 ranging from 1.4 to 2.75 kg in weight were immature while the rest were adult fish, 87.5 mm/ 4.75 kg to 102 mm/7.9 kg. in size. Females were all spent; while males were mature but not oozing.

When 4-4.5 bar gil nets were used the smaller specimens were gilled. The larger specimens were only entangled in the net and had no marks of gilling.

By periodic examination of the nets after setting, freshly gilled specimens could be taken out alive and held alive for several hours in specially designed cradles inside floating kapa nets in the sea.

Remarks

It appears quite practicable to catch milkfish spawners from the sea from areas where they are known to frequent. However, for hypophysation, mature male and female spawners are to be available simultaneously and to ensure this organized fishing - gill netting and possibly purse-seining, employing several vessels and a team of technical workers prepared to handle the spawners in the sea itself are required. Getting the spawners in the proper stage of maturity for hypophysation now appears the most difficult problem. Availability of suitable injection material could also become problematic, since it is extremely difficult to get mature or maturing specimens of species closely related to Chanos for pituitary glands collection. A limited stock of pituitary glands of Hilsa toli from Bombay has already been collected for trials. Both Hilsa toli and Hilsa macrura are available in Indonesian waters and might possibly be the best donor species for hypophysation of milkfish. With the present level of technology in spawning, hatching of eggs and larval rearing, of mullet and penaeid shrimps, actual spawning, hatching and rearing of milkfish should not present insurmountable problems, particularly since Chanos hatchings are of relatively large size.

EFFECTS OF SALINITY ON GROWTH OF YOUNG MILKFISH,
CHANOS CHANOS*

by

Fei Hu and I-Chiu Liao**

Abstract

Growth of young milkfish was studied at different levels of salinity over a period 68 days. Results suggested that young milkfish reared in freshwater or less saline sea water grew faster than in sea water. The increase in body weight was neither due to the increase in water content nor increase in feeding rate. The difference in growth rate might be attributed to the deviation from the original acclimating salinity. Mechanisms of the effect of salinity in retarding or accelerating milkfish growth should be investigated in the future.

Introduction

Milkfish culture in Taiwan dates back to the end of Ming dynasty, more than three hundred years ago. Milkfish is one of the most important food fish in Taiwan. According to the Fisheries Yearbook of 1975 published by the Taiwan Fisheries Bureau, the culture area of milkfish is more than 16,000 ha which is about 31.4% of the total area under aquaculture in Taiwan. The estimated annual yield of milkfish in 1974 was 28,900 tons. While extensive work has been done on the various cultural aspects of this economically important species (Chen, 1971), very little information is available on its physiology and ecology (Lin, 1969; Tsai et al., 1970). The present study is one of a series of physiological and ecological experiments carried out on milkfish during 16 September to 23 November 1975, in order to find out the optimum salinity range for the optimum growth of young milkfish.

* Contribution A No. 32 from the Tungkang Marine Laboratory.

**Mr. Hu is a Senior Specialist, while Mr. Liao is a Senior Specialist and Director at the Tungkang Marine Laboratory Taiwan Fisheries Research Institute, Tungkang, Pingtung, Taiwan.

Materials and Methods

Milkfish fry, presumably from the same spawning stock, were collected near the shoreline of the Tungkang coastal area with a triangular scoop net in June, 1975. They were brought alive to the laboratory and then acclimatized to a salinity of about 15‰ at water temperature ranging from 29 to 30°C in the outdoor cement tank for about three months. During this period and later experimental period, fish were fed with compound feed. The composition of the compound feed is given in Table 1. The amount of feed given each day was enough for satiation. 300 young milkfish were transferred to indoor plastic tanks (0.5 ton capacity) for the experiments. They were divided into 15 groups of 20 fish each for testing of different salinities ranging from 2.32 to 37.06‰. After being weighed, each group was acclimated to the present levels of salinity through intermediate salinities, by adding sea salt to freshwater. Then each group was divided into two subgroups with approximately equal mean body weight. Experiments were carried out in 30 covered gray PVC aquaria (50 cm x 25 cm x 21 cm in deep), each containing 10 fish in 26 liters of water with aeration and filter system. Water temperature, salinity and ration were recorded daily for each group (Fig. 1 and Table 2). Water in the experimental aquaria was renewed partially with prepared water of same salinity whenever needed. Experiments were conducted under natural illumination. Growth data were obtained by weighing all fish from each aquarium periodically. Wet body weight was recorded on a single pan Mettler P 1210 balance. After the final measurement, four to six individuals of different sizes from each group were sacrificed for water content analysis.

Results and Discussion

Mean wet body weight of fish in every group at each weighing is listed in Table 2 and illustrated in Fig. 2. Data obtained are divided into four groups of closer salinity range, i.e., 2.32-9.25, 10.77-18.74, 19.75-29.12 and 30.05-37.06 ‰ and those of the same group are plotted in the same panel. Fig. 2 shows that milkfish reared in low salinity of 2.69 ± 0.37 ‰ grew faster than those reared in other salinities, the gross general pattern of increase in weight was similar. It was, however, observed that increase in weight somehow slowed down in all groups after the fourth weighing probably owing to the decrease in water temperature (Fig. 1). To check if the different increase in weight at various levels of salinity

Table 1. The composition of compound feed for young milkfish.

Ingredient	%
White fish meal	18
Shrimp meal	10
Soybean	25
Yeast	20
Flour	15
Wheat germ	10
Soybean oil	4
Vitamin mixture	1
Total	<u>100</u>

Table II: Water temperature, salinity, ration and summarized growth results.

Tank number	1,1'	2,2'	3,3'	4,4'	5,5'	6,6'	7,7'	8,8'	9,9'	10,10'	11,11'	12,12'	13,13'	14,14'	15,15'
Temperature (°C) (mean ± S.E.)															
Designated salinity (‰)															
Actual salinity (‰) (mean ± S.E.)															
Initial number of fish															
Initial mean body weight (g)															
Daily ration (g) per fish during 1st period															
Mean body weight (g) at end weighing															
Growth rate (%)* and specific growth rate (x10 ⁻²)** at end weighing															
Daily ration (g) per fish during 2nd period															
Mean body weight (g) at 3rd weighing															
Growth rate (%)* and specific growth rate (x10 ⁻²)** at 3rd weighing															
Daily ration (g) per fish during 3rd period															
Mean body weight (g) at 4th weighing															
Growth rate (%)* and specific growth rate (x10 ⁻²)** at 4th weighing															
Daily ration (g) per fish during 4th period															
Final mean body weight (g)															
Growth rate (%)* and specific growth rate (x10 ⁻²)** at final weighing															
Averaged growth rate (%)															
Averaged feeding rate (%)***															
Survival number															

* Growth rate = $\frac{W_t - W_0}{t} \times 100\%$

** Specific growth rate = $\frac{W_t - W_0}{W_0 \cdot t}$

*** Averaged feeding rate = $\frac{R}{W_0 + W_t} \times 100\%$

W_t : Final mean body weight.

W₀ : Initial mean body weight.

t : Period in days.

R : Averaged daily ration.

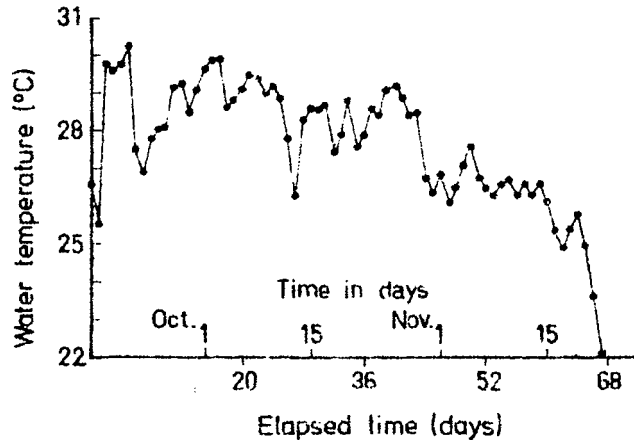


Fig. 1 Fluctuation of water temperature during the experimental period.

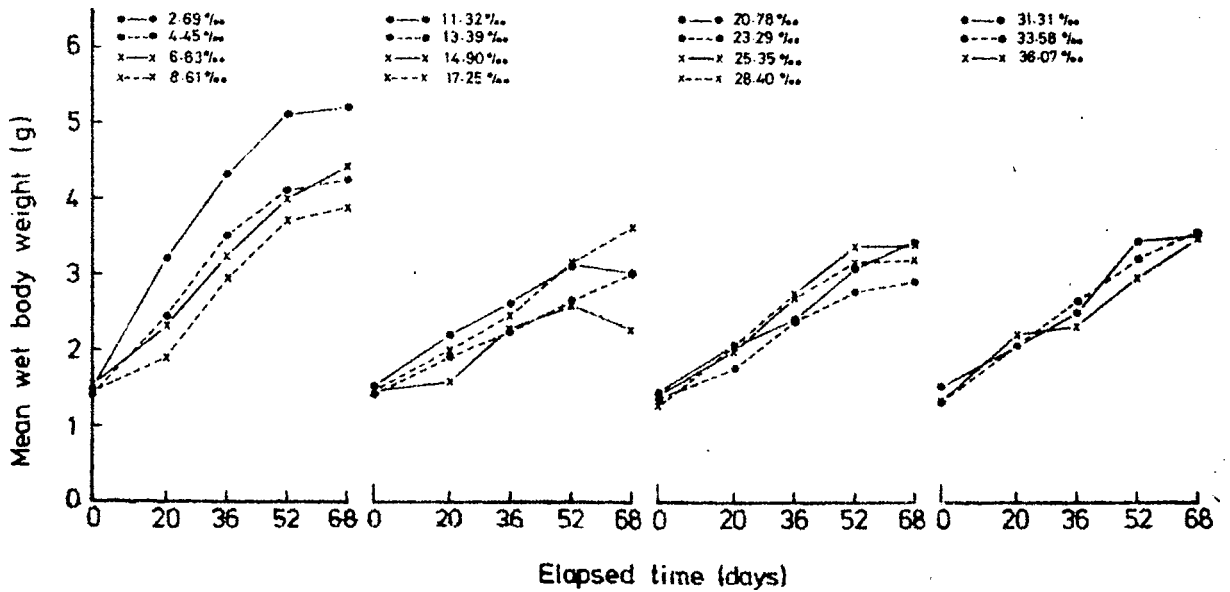


Fig. 2 Mean wet body weight of milkfish reared in different salinities.

was the result of losing or gaining water, water content was analyzed and data are presented in Fig. 3. Although the water content of young milkfish reared in different salinities for 63 days were rather different, no relationship was indicated between the water content and salinity of rearing water. It might be concluded that the increase in weight of young milkfish in lower salinity was not due to the increase in water content but to other components. It implies that growth of milkfish in lower salinity is of practical significance.

The specific growth of young milkfish in different salinities are shown in Fig. 4A. It is clear that the specific growth rate during each period varied a lot and the disparity in specific growth rate attributed to ration effect was limited, as Fig. 4B was taken into consideration. Fig. 5 shows the average growth rate, average feeding rate and the mean ration per fish over the whole experimental period. It is found that fish in salinity of $14.90 \pm 0.73^{\circ}/\text{oo}$ grew least, whereas fish in $2.69 \pm 0.37^{\circ}/\text{oo}$ had the lowest feeding rate and the highest growth rate, almost 3.5 times of the former one. It indicates again that different rations offered in this experiment had a restricted effect on the growth of young milkfish.

The milkfish used in the experiments were reared under similar conditions of water temperature, light, space, and food. Only the ration and salinity levels were different. As mentioned earlier, ration had restricted effect on the growth of fish in the experiments. Fig. 6 shows the total growth increment in young milkfish reared in various salinities as compared to that of salinity $14.90 \pm 0.73^{\circ}/\text{oo}$. From this, it is clear that change in salinity was good for growth, and that change into lower salinity was better than that of higher salinity. A number of studies have shown that the growth rate of some euryhaline organisms decrease suddenly at salinities other than 5 to $8^{\circ}/\text{oo}$, despite the ability of these forms to survive over a long period of time in a wider salinity range (Khlebovich, 1969). Chidambaram and Unny (1946) noticed a remarkable variation in the growth rate of milkfish belonging to the same stock when reared in water of different salinities. The rate of growth was maximum, (61 cm in about one year) in freshwater; whereas it was less in brackishwater and much less in confined seawater. In the present study, an appreciable decrease in the growth rate was observed in salinities above $8^{\circ}/\text{oo}$.

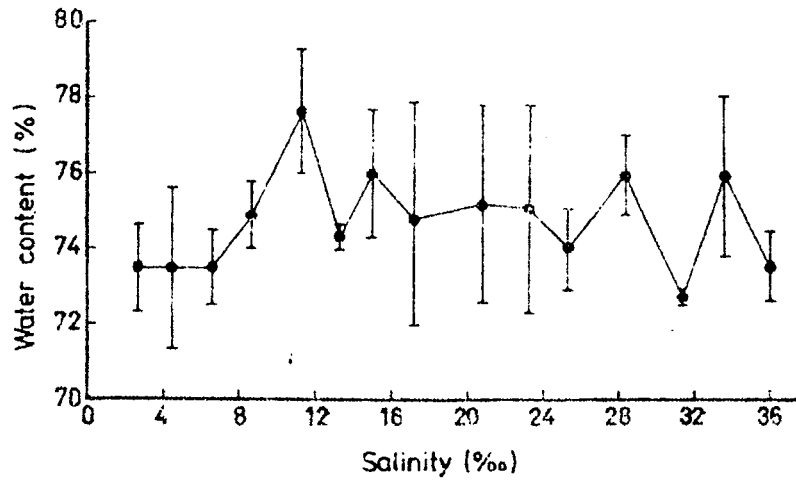


Fig. 3 Water content of young milkfish reared in different salinities.

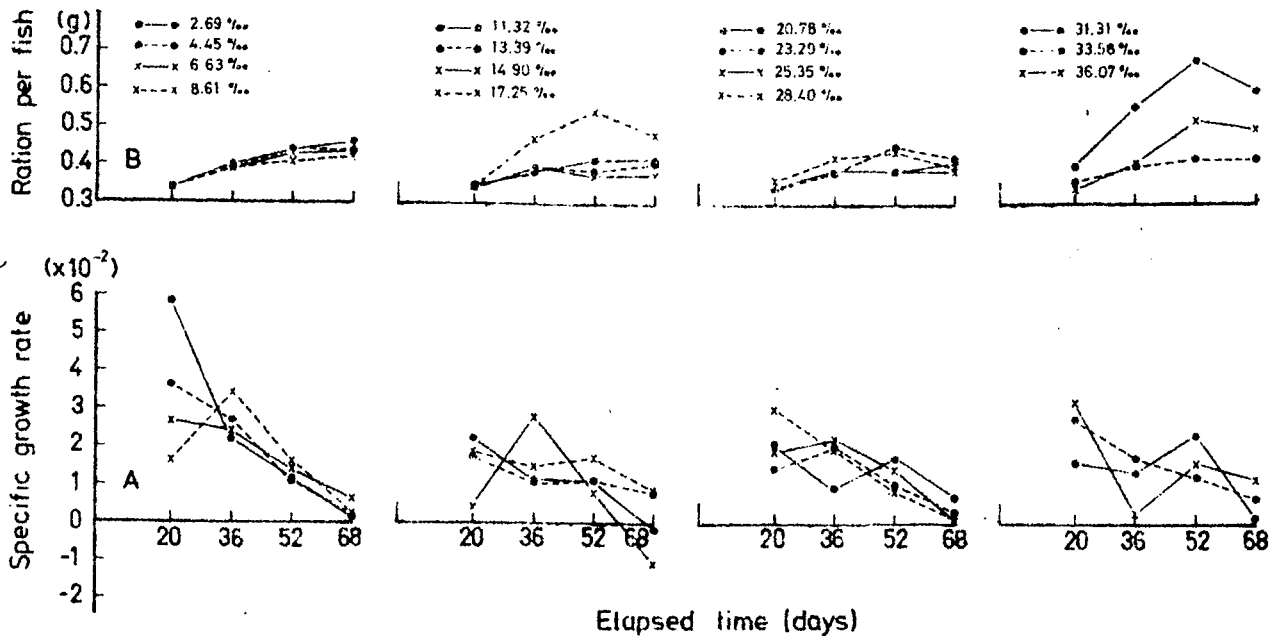


Fig. 4A Specific growth rate of young milkfish reared in different salinity.
 B Ration per fish in different salinities.

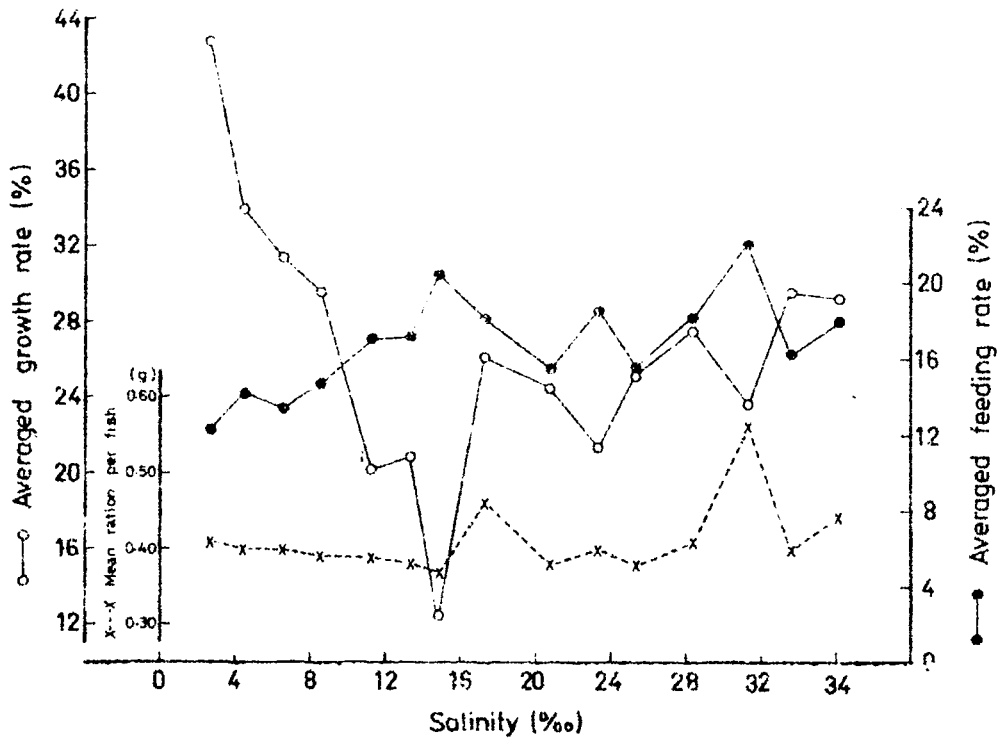


Fig. 5 Averaged growth rate and averaged feeding rate of young milkfish reared in different salinities. Mean ration per fish was presented as a reference.

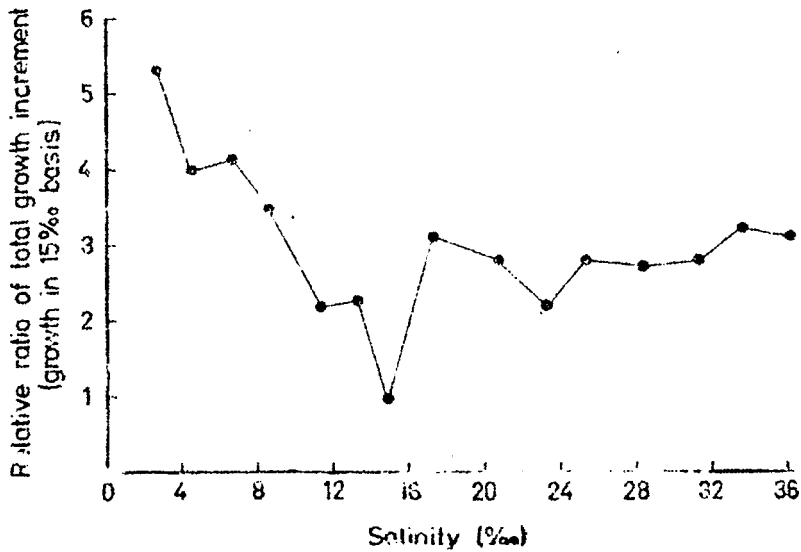


Fig. 6 Total growth increment obtained from young milkfish reared in various salinities as compared with that obtained in 14.90 ± 0.73 o/oo.

In Taiwan, the traditional milkfish farming faces a serious problem since it is no longer possible to increase the production. Modification or improvement in culture techniques is urgently needed. Determining the optimum salinity range for each growth stage could conceivably be of more value and large scale trials are necessary for confirming the present finding. Thus knowledge gained from these physiological and ecological studies, no doubt, would help increasing the production in milkfish farming.

Acknowledgments

Our thanks are extended to Mr. S. R. Liang for the experimental set-up, to Mr. D. L. Lee for the formula of the compound feed to Miss S. W. Lou for her analysis of water content and to Miss C. C. Tseng for her assistance in drawing. The authors are also indebted to Miss L. S. Pan and Mr. C. S. Lee for their help throughout the experimental period.

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PRELIMINARY NOTES ON THE SALINITY PREFERENCE
OF MILKFISH, CHANOS CHANOS, FRY

by

J.V. Juario and W.E. Vanstone*

Abstract

Vertical salinity gradient columns were used to investigate the salinity preference of milkfish fry. Newly captured fry showed a preference for 32 ‰ salinity. Fry which had been in captivity for one to five days, at 12 or 22 ‰ salinity, had no salinity preference between waters of 12, 22 or 32 ‰ salinity.

Introduction

Milkfish, Chanos chanos, fry are traditionally captured at a length of 11-14 mm along the shoreline and transported to brackishwater fish farms where they are grown to marketable size. The fry that are not captured presumably return to sea and are seldom seen again in any manner until they return as spawning adults. Several variations of this pattern occur. Documented reports show some fry enter river-lake systems and spend their first year or two in the lake and return to the sea as sexually immature fish. There are other reports which indicate that some fry stay for a time in mangrove swamps.

As an aid to the milkfish culture, several investigations have been made on the salinity tolerance of milkfish fry and fingerlings (Juliano and Rabanal, 1963; Anon. 1972, 1973a and 1973b) but no salinity preference studies have been made on milkfish fry. In order to understand better the physiology of this species, a study has been initiated to determine the salinity preference of newly captured fry and possible changes in their preference with age. This report contains the initial results of such a study.

Materials and Methods

For this study, milkfish fry were collected daily from the shore waters adjacent to the laboratory at Mag-aba, Pandan, Antique Province, Central Philippines, between May 4 to 10, 1976. Some 100 fry ranging

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in length from 11 to 14.5 mm were selected at random from about 500 fry collected on May 4 and placed in an aerated glass aquarium (30 cm x 15.5 cm x 20 cm) containing diluted sea water at a salinity of 12 ‰. A second group of 100 was maintained in an identical aquarium but at a salinity of 22 ‰. These fry were fed with rotifer, Brachionus sp., daily at 0830 hours and 3/4 of the water in each aquarium was changed daily at 0800 hours.

The salinity preference experimental tanks were plastic aquaria (33 cm x 18 cm x 23 cm) placed on a table in a curtained corner of the laboratory. A 1.22 m single fluorescent tube was located 2 m above the tanks.

A 7 cm layer of sea water of salinity 32 ‰ was placed at the bottom of one experimental tank. A 7 cm layer of 22 ‰ diluted sea water was then floated on top of the 32 ‰ water and a 7 cm layer of 12 ‰ was floated on top of the former layer. A layer of 21 cm of water at one salinity was placed in the second tank which served as the control.

Three tests were conducted each day: test I, with fry newly collected from the shore waters (salinity, 32 ‰); test II, fry "acclimatized" at 22 ‰; and test III, fry "acclimatized" at 12 ‰. The control tanks for each test contained water at salinities of 32, 22 and 12 ‰, respectively. For each test, four fry were placed in each of the gradient and control tanks. Observations on the position of the fry were started one hour later and every 20 minutes thereafter for 80 minutes. Salinities of each water layer were determined after each test, surface temperatures recorded and the fry removed, measured and preserved in 5% sea water formalin.

Results and Discussion

Upon introduction of fry into the experimental or control tanks, it was observed that they immediately swam to the bottom of the tank. On reaching the bottom, they appeared to be very confused. However, they quickly calmed down and it was assumed that the one-hour adjustment period was more than sufficient for recovery from the stress of handling and introduction into new surroundings. In both the control and experimental tanks, occasional schooling was observed in all groups of fry but most of the time the fry swam individually.

Newly captured fry swam freely up and down through the whole column of water in the control tank (32 ‰) but remained in the 32 ‰ layer in the experimental tank. Fry maintained in water with salinities of 12 and 22 ‰ for 1 to 5 days did not exhibit a preference for any particular salinity but swam at random throughout the control and experimental tanks. However, it was observed that these "acclimatized" fry tended to stay longer at the bottom of each tank and appeared to be nibbling or searching for detritus.

The results obtained are still tentative but similar studies on other species (Baggerman, 1960, 1963; Otto and McInerney, 1970; Hain, 1975; Schulz, 1975) have shown that salinity preference is functionally related to the physiological state of individual fish.

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PHYSIOLOGICAL FUNCTIONS OF THE EPIBRANCHIAL ORGAN
OF MILKFISH FROM THE POINT OF ITS ONTOGENY

by

Takeichiro Kafuko and Yukimasa Kuwatani*

Abstract

The epibranchial organ of milkfish was believed for long to be a respiratory organ similar to that in Anabantoids. Later on, the structure was considered to be an accessory to the digestive system. The observations made by the authors on the physiological functions of the organ supports the latter views. This finding, probably would provide a new field of research on milkfish.

Introduction

Hyrtil (1863) was the first author to describe and draw a diagram of the pharyngeal organ of Lutodeira chanos (Chanos chanos) as a respiratory organ, called "die accessorishchen kiemen organe".

Since then many scientists have studied the structure of the organ mostly in lower teleosteam herbivorous fishes such as the elopiform and clupeiform fishes. They believed that the organ had a respiratory function similar to that in anabantoids. Heim (1935), however, corrected this concept by studying the structure of the organ concluding the organ to be an accessory to the digestive system.

Recently, Bertnar, et. al. (1969) summarized the studies along this line, and moreover, expanded our knowledge of the Clupeiformes (Clupeidae and Engraulidae), Gonorhynchiformes (Chanidae, Gonorhynchidae, Kneriidae, and Phractolaemidae), Osteoglossiformes, Salmoniformes (Salmonidae), and Cypriniformes (Characidae, Distichodontidae, Citharinidae, Curimatidae, Hemiodontidae, Prochilodontidae). They classified the structures into seven types.

Besides the contribution of Hyrtil, (1863) there are only a few papers dealing with the epibranchial organ of milkfish such as (Monod, 1949, 1961, 1963; Kapoor 1954; and Takashi, 1957). Among them, Kapoor attempted detailed studies on the anatomy

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and histology, and came to the same conclusion that the epibranchial organ of milkfish functioning principally as accessories to feeding and digestion. This was based on the finding that the inner surface of the pocket is lined with striated, squamous epithelium carrying a number of mucous cells.

Observations

We have observed a speck of food wrapped in mucous in both the organs. We also studied the structures which allow the passage of the food before and after coming to the organ. From these studies we gather the following:

The food accumulated between two rows of gill rakers on each gill arch are sucked into the organ under an automatic movement. The water sucked along with the food into the organ is ejected through the marginal canal which runs from the inner wall of the blind sac to the outer part of the gill rakers in buccal cavity. Then, the accumulated food is conveyed to the oesophagus. Enzyme which might exist in the organ are not yet elucidated. Thus, it appears that the organ is a part of the digestive system.

Surprisingly, the rudimentary epibranchial organ appears before any other parts of the digestive system and is observed in as early stage as 14 mm fry. The relationship between the development of the epibranchial organ and the number of gill rakers was found as follows:

Total length	Weight	Number of gill rakers	Epibranchial organ	Length of intestine
14mm	--	14	rudimentary	about 7mm(straight)
19mm	--	177	complete	19mm
62mm	--	250	-do-	(not examined)
	300g	430	-do-	(not examined)

Remarks

From the findings as mentioned above, we may attribute, reasons for, higher production in different milkfish culture systems especially because of the existence of this specialized organ and its function. The other words, we consider that the functional study of the epibranchial organ from the point of biochemical and habitual aspects provide a new field on milkfish problems for searching rational culture techniques from fry to adults.

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THE OCCURRENCE OF MILKFISH CHANOS CHANOS FRY IN
PANDAN BAY, ANTIQUE, FROM 21 MAY TO 25 JUNE, 1975

by

S. Kumagai, A.C. Villaluz,
L.B. Tiro, Jr. and W.E. Vanstone*

Abstract

Milkfish fry were collected at the Pandan shoreline and 500 meters offshore. While shore-caught fry were uniform in size those captured offshore varied in size and stage of development.

Introduction

The culture of milkfish, Chanos chanos, is a major industry in many Southeast Asian countries particularly Indonesia, Philippines and Taiwan. For centuries, milkfish fry have been captured along the shoreline and reared to marketable size in fish ponds and more recently in net enclosures.

A great deal is known about the yearly fluctuations in fry abundance and the climatic and oceanographic conditions prevailing during the peak seasons of their occurrence in various geographic regions of Southeast Asia and the Pacific Islands. Similarly, the culture of milkfish has been well documented and there are several ongoing research programs designed to assist the industry.

In contrast to available knowledge on milkfish in captivity, very little is known of its life cycle in nature. To assist in overcoming this lack of knowledge, the Aquaculture Department of the Southeast Asian Fisheries Development Center (SEAFDEC), has undertaken a concentrated study of Chanos chanos in the wild. From 13 May to 26 June, 1975, SEAFDEC stationed a study team at Mag-aba, Antique, Philippines (Fig. 1) to survey the occurrence of milkfish in Pandan Bay and to lay the groundwork for future studies. The selection of this site was prompted by the fact that 75% of the milkfish fry captured in Panay Island come from Antique Province (Villaluz, 1975) and with the existence of one otoshi-ami, a large Japanese-designed bag net (Manacop, 1975), with a 30-m-deep bag located in 30m deep and 500m offshore from Mag-aba, Pandan Bay. In

*Mr. Kumagai and Mr. Villaluz are researchers of the SEAFDEC Aquaculture Department; Mr. Tiro is a research aide of the same Department; while Dr. W.E. Vanstone is a scientist with the SEAFDEC-IDRC Milkfish Project.

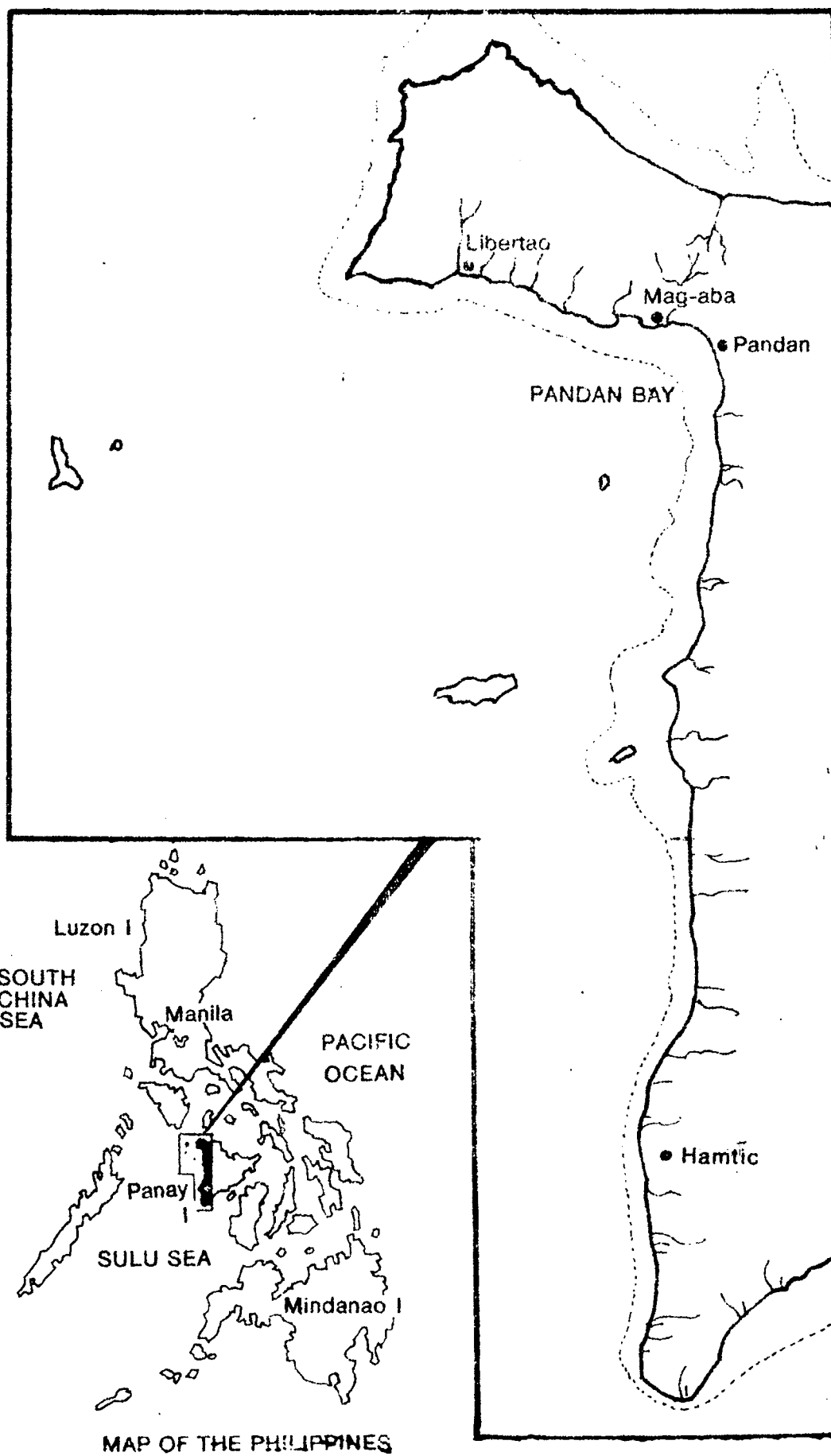


Fig.1 Area of study

addition, there are two otoshi-ami offshore from Libertad, 20km west of Mag-aba and several fish corals in the area. Both the otoshi-ami and the fish corals capture adult milkfish or sabalo during the fishing season from January to June.

This note is based on a comparison of data on milkfish fry captured along the Mag-aba shoreline with those captured in the otoshi-ami near Mag-aba from 21 May to 25 June, 1975.

Materials and Methods

From 21 May to 25 June, 1975, mixed species of fry were collected daily by means of a 3 meter long by 1 meter deep seine net 3m x 1m along 50m of the Mag-aba shoreline between 0800 and 0900 hours dragged by two men. At 1200 hours and again in 1600 hours, mixed species of fry collected with a scoop net fitted with NGG-54 nylon meshed scoop net from the otoshi-ami at Mag-aba as the commercial fishermen hauled it to the surface. Salinity and temperature of water sampled at 5, 15 and 30m depth below the otoshi-ami during the 1200 lifting of the net were noted and reported elsewhere (Tiro et.al., 1976). All fry samples were preserved in 5% seawater formalin for one to three days prior to sorting and measuring the milkfish fry. Also, some of the "shore-caught" milkfish fry obtained on 11 June were placed in an aquarium containing sea water (32 o/oo) and fed with a mixture of polished rice washings and mixed plankton obtained from plankton tows along the shoreline. Fifty per cent of the volume of water in the aquarium was changed daily and sub-samples of these fry were examined every second day until 25 June.

Results and Discussions

During the period from 21 May to 25 June, 1975, 522 milkfish fry ranging in total length from 11 to 14.3mm were captured along the shoreline and 1500 fry ranging in length from 5.8 to 14.6mm were obtained from the otoshi-ami (Figs. 2 and 3).

While this report covers a period of only 36 days the trend in the data presented in Fig. 2 suggests that the daily number of fry caught by the otoshi-ami was greater one to two days prior to the new and full moons whereas the greater daily catch along the shoreline occurred two to three days after the new and full moons. Miyagami (1971), Carbine (1948), and Kuronuma and Yamashita (1962), reported that the peak in the number of fry captured along the shoreline do in fact follow the lunar cycle with the greatest abundance occurring from one to two days before and up to three days after the new and full moons. This pattern is modified with

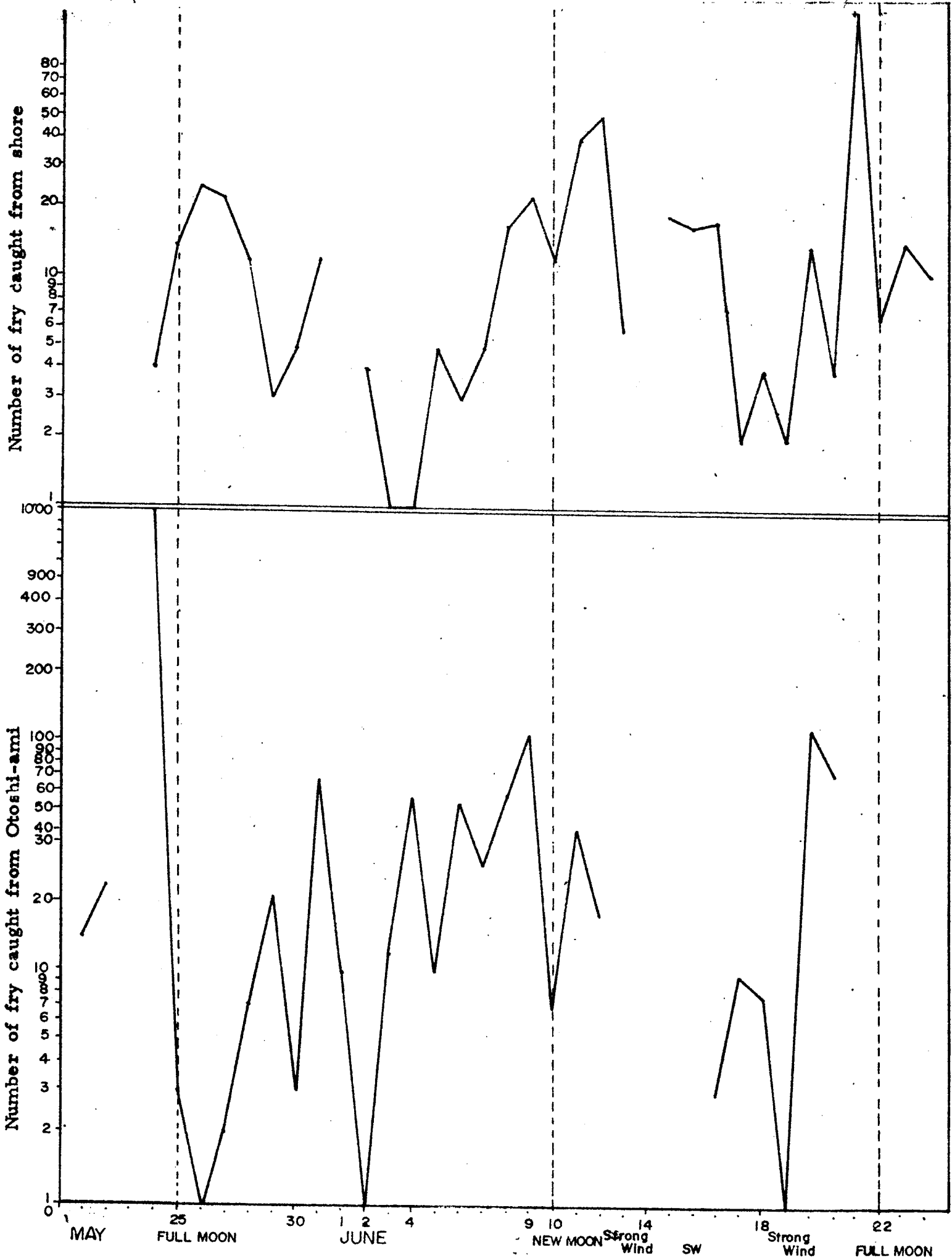


Fig. 2. Occurrence and abundance of Chanos fry at two stations

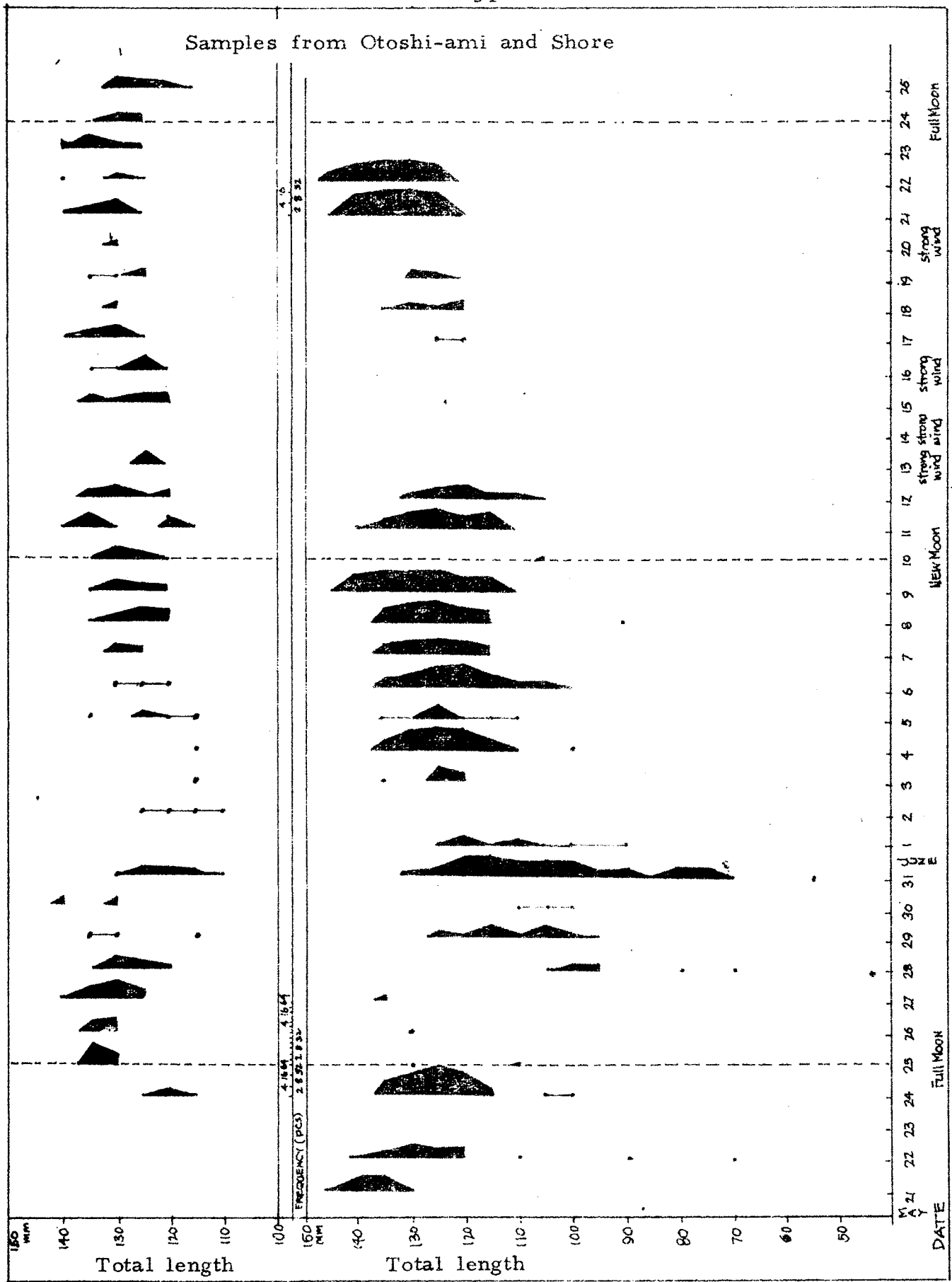


Fig. 3. Length and frequency of Chanos fry

the abundance of fry being lower during these periods when there is a large movement of surface water toward the shore if the high tides are accompanied by high offshore winds. In contrast to the above reports, local fry collectors and fisheries officers in Southeastern Luzon Island and Antique Province on the West coast of Panay Island state that the peaks in fry gathering usually occurs two to three days after the new and full moons. While these observations may be local situations, they are supported by the present findings.

The total length of the fry (Fig. 3) were 1.5 to 2mm shorter than those reported by Delsman (1929) at most stages of development. While our fry of 5.8mm total length appeared identical to Delsman's 6.0mm fry, our fry of 8, 9 and 10mm from the otoshi-ami and our 11 to 14.6mm fry also from the otoshi and from the shoreline were similar to his pelagic larvae of 10, 11 and 12mm and his shore-caught fry of 13mm.

Although Delsman obtained larvae of 6 to 12mm total length in surface plankton hauls, we were unable to obtain any milkfish larvae or fry by this method in the vicinity of the otoshi-ami or further offshore from it. Because of shallow water over a coral reef we were unable to tow our plankton nets closer than 500m from the shore. However, we did obtain larvae and fry at different stages of development from the otoshi-ami after the bag was lifted through 30m of water. From our limited observations, it appears that milkfish fry and larvae collected from Mag-aba 500m or more offshore, are located somewhere in the mid-water layers and that they drift in this layer from the breeding grounds towards the shore while growing to a total length of 9 to 10mm. At this size they have well developed dorsal and anal fins and probably air bladder and float to the surface near the edge of the coral reef. The fry are then carried to the shore by tidal currents and winds and collected along the shoreline as 11 to 14mm fry.

As reported by Delsman (1929), and confirmed in this study, ventral fins are absent from newly captured shoreline fry. These fins, however, were present in some specimens after 10 days and in all specimens after 14 days of aquarium rearing (Table 1).

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Table I. Length measurement mean of three samples and development of ventral fins of aquarium reared milkfish fry.

Date	Days in Aquarium	Total Length (mm)	Pre-dorsal Length (mm)	Preanal Length (mm)	PDL TL	PAL TL	Remarks
1975 June 11	0	13.1	-	-	-	-	
	13	13.3	7.8	9.2	0.582	0.688	
	15	12.6	7.5	8.7	0.597	0.689	
	17	13.0	7.5	8.9	0.572	0.683	
	19	13.5	7.8	9.2	0.581	0.680	
	21	13.3	7.9	8.9	0.594	0.673	Ventral fins present in some fish
	23	13.4	8.1	9.3	0.606	0.689	
	25	14.0	8.1	9.6	0.582	0.684	All specimens have ventral fins

REVIEWS AND RECOMMENDATIONS ON CURRENT AND
FUTURE RESEARCH ACTIVITIES FOR THE
BIOLOGICAL INVESTIGATIONS ON MILKFISH

by

Katsuzo Kuronuma*

Foreword

The notes written herewith are presented in compliance with the "Proposed Agenda Items for the Milkfish Workshop-Conference at Iloilo, Philippines, May 19-22, 1976", and given in the order as denoted in the agenda above.

It is mentioned that the reviews or impressions on the current research activities be limited mainly to the contents in Part III, "SEAFDEC-IDRC MILKFISH RESEARCH PROJECT. Milkfish (SEAFDEC) IDRC File 3-P-74-0146, 1975: 62 pp., Fig.", and also, that recommendations for future investigations be within the framework of the reviews or impressions for each agenda.

The present writer proposes to establish an additional agenda item, VIII. Documentation (Presentation and recording of works; Report writing; Bibliography).

I. Behaviour and bio-ecology of milkfish in the wild

A. Taxonomic considerations in relation to racial or sub-specific variabilities

It is believed that Chanos chanos occurring widely in the Indo-Pacific waters extending from the Red-Sea to the Mexican coast is represented by different races geographically or populations micro-geographically, although the problem had not attracted the attention of systematic ichthyologists. The present writer observed in Indonesia and the Philippines that there are two types of milkfish as shown by photographs taken, one with greater depth of body and higher lateral line scale count and the other with slenderer body and lower scale count. Since no examinations were made on the specimens the fact may be considered as accidental. The two seasons of appearance of fry of these different types along the coast if definitely determined, might suggest the different races or populations existing.

*President, Ichthyological Society of Japan, Tokyo, Japan.

Recommendation: In view of its importance to fisheries work, racial or population analysis will be needed for obvious reasons, and the analysis to be conducted may vary in scope depending on the configuration and extension of coastal line. From the standpoint of pure biology, a semi-global program in racial analysis of milkfish may be thoroughly evaluated.

The racial or population analysis of fish will be made possible under three methodological categories, morphological (morphometrical measurement and meristic counting), ecological and biochemical studies on various stages of the fish. The program of studies will be made by referring to the literature on salmon, sardine, herring, mackerel, etc. The methodology of studies is explained in an article by S.J. Holt (Report of the International Training Center on the methodology and techniques of research on mackerel (Rastrelleiger), Bangkok, 20 Oct. - 28 Nov. IPFC/C58/W28, 1958).

B. Migration pattern

The movement of the milkfish fry from the spawning ground toward the shore water is apparent. However, the present information about the migration is scanty and not supported by real evidence. Furthermore, internal and external factors responsible for the movement are not understood. The migration of adult milkfish in the sea is not known either, and the observations made so far are limited in the context of space and time.

The hypothetical comment for the migration in spawning of milkfish was presented by the present writer (K. Kuronuma and M. Yamashita, 1962), but the comment was derived indirectly through the appearance of the fry in shore water. There are many observations on the movement of young or sub-adult fish in ponds or in associated water systems, but it has not been confirmed whether they demonstrate similar behaviour in natural sea water.

The collection of fry made at Pandan (1975 Project, Pt. III) shows that those fish in offshore (otoshi-ami site) and those in shore waters are evidently identical as population. The apparent difference in size between the two lots of fry may suggest the growth which occur during migration. But the work does not give information on the migration of fry in sea water.

Recommendation: Similar to the population analysis noted in the previous section (IA), the subject on migration of fish from juvenile to sabalo may be treated from the local standpoint or on local population. Such limitation may be justified in view of fisheries research.

The migratory behaviour of fish from larvae to fingerlings can be traced by extensive and intensive collection of samples. Larva-net is commonly used. Depending on the facilities and gear available, the collection will be made by vertical hauling of net or towing of the gear in specific layers of water. The collection as such, attempted in different waters, will supply the data on which migratory behaviour may be understood. The size of stock among different waters may also be estimated.

The studies on moving or migration of fish from sub-adult to sabalo stages can be made by "marking-recapture" method. It may be needless to mention that the present method can be adopted under the condition that fishery regulation has been worked out and fishermen's collaboration has been promised. Perhaps the method will be an answer to the problem.

C. Oceanographic conditions

The natural environmental conditions, physico-chemical and biological, influences, if not usually determine all the biological phenomena taking place in the water. Milkfish is not an exception in this respect. Thorough understanding of the life of fish can be hardly achieved without knowledge of environmental factors surrounding the fish.

In the field work at Pandan (1975 Project), temperature and salinity of the water were measured during the operation of otoshi-ami (over a few months). The water currents, tidal and sea, if measured during the work there might have supplied more valuable information for the understanding of fry and sabalo. Observations on wind, cloudiness, water-transparency and precipitation should also have been made.

Recommendation: It is considered as a general rule that oceanographical survey or observation if made without association with milkfish investigation would contribute little to the knowledge on bio-ecology of the fish.

The working program for oceanographic observation will be made very carefully depending on locally accumulated experiences and objectives, not speaking of technical problems involved. The survey vessels of the Department (SEAFDEC I and II) will play leading part in such project.

D. Age and growth studies (on sabalo)

Work has been started at Pandan (1975 Project, Sec. 3). It may be noted on age and growth at first that the term "morphological measurements" given in the title needs clarification. The term denotes measurements on various body parts of fish as understood among ichthyologists and fishery biologists, working especially on identification of species, races, population, etc. (See I.A., also paper by Liao [1971, Fig 1]). In the same report there are length-weight measurements, but not the morphological measurements.

As to the age determination of Chanos the report discussed the problem based on the reading of scales. Admitting that the techniques involved in fish scale reading are extremely complex, the determination attempted appears not fully convincing. The adoption of a paper on silvercarp was not explained. The spawning mark, if there is any, is not discussed. The age as determined in Taiwanese sabala (Liao, 1971) appears conflicting or not coinciding at least with that of Pandan fish.

Scale reading, if attempted on the milkfish culture in fishponds or lakes, might have supplied valuable background knowledge to the work.

The length-weight relationship in sabalo trapped by otoshi-ami (1975 Project, Sec. 3, Tables II and III) shows very wide ranges both in males and in females the fact being well noted if all the data are plotted on a graph. It may be worth trying to calculate the body weight of fish without gonad (gonad weight is subtracted from the total body weight), the length-weight relationship is sought. A linear relationship might be shown by doing the above.

The growth of sabalo appears hardly discernible because age determination is still ambiguous. The size-frequency method may hardly be applied to sabalo because of its catches.

The development of gonads examined on the sabalo at Pandan was treated in the report only preliminarily, and further analyses of the data may be recommended.

Recommendation: The methods adopted at Pandan Field Station are believed adequate. Some suggestions may be presented as follows: 1) effort will be expanded further for the collection of materials; 2) for the analyses of the data the workers will read more literature relevant to the subject concerned in order to make more accurate interpretation of the problem and full utilization of the data; 3) similar and collaborative studies may be implemented at different field stations including Hamtik, Oton, Pandan and possibly Naujan Lake. It must be remembered that the otoshi-ami is not the sole fishing gear to catch adult milkfish.

E. Food availability and utilization

Very detailed study was attempted for the subject by stomach content analysis (1975 Project, Sec. 4), presenting a valuable contribution to the biology of chanos.

A statement (p. 37) ". . . results of which may be used as background data for the milkfish seed production project" may not sound convincing, because the stomach content was examined only on sabalo, but not on fry or fingerlings in natural sea water.

Another personal comment raised by the present writer is as follows. 'Milkfish in the sea water is plankton feeder' is fully understood. The statement (p. 42)". . . they have no specific preference for plankton on any water level" deserves attention. The occurrence of various forms of plankton in sea water or their habitats were not studied nor explained based on existing literature.

Recommendation: The food habit of fish in natural environment is an important subject for study, and one of the techniques often adopted in the study is stomach-content analysis. There are a number of useful papers on the subject, of which 2 publications below may be useful as a reference. Lagler, K.F. (1952) "Freshwater Fishery Biology" and Pilley, T.V.R. (1952) "A critique of the methods of study of food of fishes", Journ. Zool. Soc. India, 4(2): 185-200.

Efforts should be made to study the food habits of chanos in fry and fingerling stage, and also to become familiar with literature along the same line.

II. Maturation and reproduction in wild and captive fish

A. Reproductive physiology

This problem had not yet been taken up in the SEAFDEC project.

Recommendation: The knowledge on milkfish, especially the biological problem may be said to be meager or almost nil, since the investigation on this problem is still new. The program of the study naturally will start from the survey of literature. Here again, there are many publications on this subject of fish biology, and the paper below may be found useful in the initial stage of literature review: Holiday, F.G.T. (1963) "The behaviour and physiology of herring and other clupeoids. In: Advance in marine biology, 1: 261-404, Academic Press. Also, contact and subsequent request for technical papers should be made from the Laboratory of Fish Physiology, Department of Hydrobiology, Institute National de la Recherche Agronomique. 78350, Jouy-en-Jones, France.

B. Gonadal development

The problem on milkfish has been touched by examining ovaries and testes of fishes collected at Pandan (1975 Project, Sec. 3) and UPCF Inland Fisheries Project. The studies made so far may be said to be still in its initial stage of work as to its scope and technique.

Recommendation: The techniques required and the interpretation of the data will be studied at first by reading the publications given below. Holt, S.J. (1956) "Report of the International Training Center on the methodology and techniques of research on mackerel (*Rastrelliger*). IPFC/C58/WP 28". Breder, C.M. and R.E. Rosen (1966) "Modes of reproduction in fishes. Am. Mus. Nat. Hist." K. Yamamoto (1966-1968) "Collection of papers on the reproduction of fish, Tokyo, Japan.

C. Mating behaviour, etc.

The problem has not yet been touched up to date beyond prospects or imagination concept.

Recommendation: Similar suggestion can be given to refer to the following: Kuo, C.M. and others (1974) "A procedural guide to induce spawning in grey mullet (Mugil cephalus Linnaeus). Aquiculture, 3" Liao, I.C. (1970) "Experiments on induced breeding of the grey mullet, Mugil cephalus L. A paper in Coastal Aquaculture Symposium, IPFC 14th Session, Bangkok, Nov. 18-27, 1970".

D. Natural and/or induced spawning, etc.

Same as in the previous section as the problem has not been touched.

Recommendation: Same as given above (I.C.).

E. Examination of spawned fish

The problem dealt with in this section may be better focused on the subject of how to keep sabalo under captivity. The present writer believes that actual work on the subject is carried out currently at Pandan, of which he is unfamiliar. However, he has been informed that several researchers of the Department made observation tours to Honolulu (Oceanic Institute) and Taiwan (Tungkang Marine Biology Station), and in each institution, experiments are being done to keep sabalo in tanks. It is hoped that the observations made by them will produce valuable information on the works now going on at Pandan and elsewhere in the future.

Recommendation: The present writer has no particular suggestion to make at the moment aside from reporting the following: At the Oceanic Institute (Honolulu) where he made a visit in September 1975, several sabalos nearly one meter long were kept in wooden tanks fed by running sea water. The sabalos were swimming in circle very calmly probably because they were content with the water and food (vegetative) provided. Some of the sabalos kept in the tank were said to have been taken from fishponds in the island, and this should interest the workers in the Philippines.

III. Egg and larval surveys

A. Identification of milkfish eggs and larvae

The eggs and larvae of the species were described by Delsman more than 40 years ago. Since then, no detailed studies on the morphology have been made. This fact alone will require to include full description of eggs and larvae with fine illustrations in the report

(1975 Project). The fact that local fishermen and fry dealers of milkfish are well acquainted with their commodities they will not be confused with the significance of scientific documentation.

Recommendation: Full description and illustration (photograph and drawing) will be made on eggs and the larvae in successive stages based on the materials collected in sea water or reared in tanks. One paper noted here may supply a valuable suggestion to the work above. Matsubara, K. (1942) "On the metamorphosis of a clupeoid fish, Pterothrissus gisu Hilgendorf. Journ. Imp. Fish. Inst., Tokyo, 35(1); 16 pp., 6 figs."

B. Occurrence of larvae

The milkfish fry were collected in 1975 at Pandan, Hamtik and Oton by the Department, and the report (1975 Project, Sec. 2) covers the work done at Pandan only. The present writer received a copy of the draft report by P. Gabasa entitled *Milkfish fry investigation on the Antique coast, Panay Island, Philippines, May - June, 1975*, 14 pp. and a number of figures.

A few comments on the report from Pandan may be given herewith. On Fig. 2 (p. 21) the ranges in the size (T.L.) of larvae are different in the samples scooped in otoshi-ami from those seined in beach water, the former represented by smaller fishes at least in May and June. Whether the difference in size is reflected by gear selectivity or is it indicating the growth of fry is not explained. Oceanographical conditions are not presented thus making it difficult to analyze the occurrence of the fry in relation to environmental factors. Apparently, tidal fluctuation does effect the abundance of shoals both in offshore and shore waters, but the picture at Hamtik is not so simple. In the report from Pandan, milkfish fry were worked on but no other fishes which should have also been collected were mentioned, nor plankton community in the local sea water explained.

Recommendation: The investigation of Chanos fry will have to be programmed on a national scope because of the importance of the industry. The program could be formulated through careful and detailed planning of field work required: In the planning, the following items may be considered -- site selection, period of operation, unit-collecting-gear, unit-effort, land facilities, not speaking of manpower. In this connection, the experience of the present writer (Kuronuma, K. and M. Yamashita, 1962) may supply some useful suggestions.

The final report of milkfish fry investigation in 1975 will have to incorporate all the works done by the Department presenting the data with care.

IV. Egg incubation and larval rearing of chanos

Within the limited knowledge held by the present writer, the problems involved in the subject have not been touched, nor are published information available. Therefore no commentary notes can be afforded at the moment.

Recommendation: Since the studies on incubation techniques and rearing of larvae are now projects so-to-speak in the Department, the study will start from the planning of the study program, but not from 'happy thoughts'. As emphasized repeatedly by the present writer the consultation with literature will be the starting line in the planning, and he will suggest to become familiar with some publications on mullet and herring as introduced elsewhere in the present writing. It may be predicted that the Station at Tigbauan will be the center of actual experiments, where shrimp will be successfully hatched and its larvae will be reared with high survival rate. It is to be remembered that this type of experiments is highly influenced by the facilities provided in addition to techniques.

V. Pathology and physiology

The above subjects may be said to be practically unknown in so far as Chanos is concerned.

Recommendation: Since the subjects are broad and quite heterogeneous besides being rather different from other subjects discussed before, an entirely new project will be established or created. The present writer believes that the project will not be undertaken together with other subjects. The Department will also need professional biologists if the project should be taken up at this time.

VI. Status of present research

VII. Future plan of action

The two above-mentioned problems are better treated within the same category of considerations and ideas, that he also expressed his evaluation, comment and recommendation of each subject mostly from technical points of view in previous pages. Further, it seems apparent that the two problems will have to be treated or considered

from two angles, technical and administrative as well as political in a sense. However, the concepts held by the writer will be given here in the order of original agenda items, but with more bearing on administration.

(1) Researches conducted within the Department

(a) The system in the actual conduct of researches will have to be more solid so that each worker engaged in milkfish investigations can identify himself on the nature and level of the work he is involved in the over-all study conducted by the Department. A pyramidal system of works will have to be formulated more clearly.

(b) Researchers will have to utilize library facilities more extensively. The library on the other hand will make effort to collect publications needed by researchers.

(c) Research vessels will be placed under the maintenance section of the Department, but the captain will be detailed with the research section.

(2) Researches conducted at the national level

(a) Intimate collaboration is needed among research institutions, or organizations existing in the nation -- SEAFDEC, UPCF, BFAR, MSU and probably SCSFDCP.

(b) At present it is observed that the same subject of study is carried out by different institutions with apparently no mutual understanding. The joint studies and the studies shared will have to be defined clearly.

(3) Prospects of international collaboration

(a) It may be easily understood that international collaboration on milkfish investigation is a golden rule.

(b) Conducting a campaign among other nations will have to be made based on an attractive prospectus reinforced by technical aspects of highest quality. The timing of the campaign must be cleverly calculated.

(c) The campaign will be extended to broad areas of the Indo-Pacific and not confined only to the Southeast Asia.

(d) The campaign will be made more efficiently through the channels of international organizations like IPFC, IOFC, SPFC, SCSFDCP, PSA, FAO, etc.

(e) For the time being the present writer is inclined to believe that it is appropriate to hold a meeting in an international level tentatively entitled "International Training Center on the Technique of Milkfish Biological Research", which will follow the present Workshop Conference.

(f) It is to be remembered that IPFC have had a sub-committee within the organization in the past.

VIII. Documentation

A. Presentation and recording of works

The Aquaculture Department of SEAFDEC has started actual work since 1973 or so at the MSU Biological Station in Mindanao. Now, it is apparent that records on many works had been drafted. The records are not limited to those describing the works conducted, but also include other types of recordings on the activities of the Department personnel, study and training tours abroad and locally, lectures delivered or attended, etc.

Under such circumstances it will be highly recommended to exert effort to assemble all kinds of records regardless of the nature of its contents. The records, will be properly classified and deposited in the library and be accessible to reading or examination.

Referring to the format of various records issued by the Department the present writer observes that those which have been circulated so far are too bulky, and they are not classified as to the nature of substance. In some cases, administrative content is combined together with research substance, resulting in some criticisms that the Department is a research and training institution.

B. Report writing

The type of reporting on the researches conducted may be classified roughly into two -- Progress Report and the (final) Report. In this sense the reports on the works done and issued so far by the Department may all fall on the so-called Progress Report.

It is hoped that some of the works conducted (e.g. Chanos fry collections in 1975, and shrimp rearing in the pen) will be published as a final report as well as a real scientific paper. It is also hoped that such a report will be printed in a Journal published by the SEAFDEC AQUACULTURE DEPARTMENT.

C. Local common names of Chanos chanos

This writer made some compilation of common names of the milkfish used in several countries and different places in the Philippines. The list is attached as Annex 1.

D. Bibliography

It is needless to emphasize here the meaning of bibliography in any research activity, and the present writer observes that a scientific paper or book which is not reinforced by a review of literature is very often unacceptable.

In this sense the library must be well provided with publications, and the researchers must always open their eyes widely in literature hunting.

Attached as Annex 2 is a bibliography compiled by this writer.

Local Common Names of Chanos chanos

compiled by K. Kuronuma

Awa-Awa (Hawaii)
Baulo, Bolu (Celebes)
Belanak-sembawa (N. Borneo)
Binni Al-bhr (Iraq)
Milkfish (English)
Banglis (Philippines)
Bangles (Philippines)
Banglot (Philippines)
Banglus (Philippines)
Bangus (Philippines)
Bangos (Philippines)
Ca Mang (S. Vietnam)
Ga-tin (Burma)
Bandang (Malaysia)
Jangos (Malaysia)
Pisong-pisong (Malaysia)
Pla-Nuah-Chan (Thailand)
Plai-meen (Ceylon)
Pal-meen (Tamil)
Ikan Bandeng (Java)
Vaikka (Sinhaleese)
Sabalo (Spanish in Mexico and Philippines)
Sabahee (Taiwan and Japan)

Note: Correction and addition highly appreciated.

BIBLIOGRAPHICAL SOURCES ON THE BIOLOGY AND CULTURE
OF MILKFISH, CHANOS CHANOS

Compiled by Katsuzo Kuronuma, April 1976*

An attempt has been made to compile the present bibliography as comprehensively as possible within the limits of source materials available at hand. It is admitted that many indispensable works may have been escaped in the listing. Also, attention may be drawn to the fact that a large number of references included here have been quoted from different sources. Hence, the presentation of these may be lacking in consistency of style of citations and other notations.

It is hoped, however, that the present list of bibliography on milkfish covering more than 250 items will mark a starting line toward the completion of the full listing of the publications, referring to this valuable and interesting species of fish occurring widely in the Indo-Pacific sea areas.

It may be added that the present list is submitted to be used during the "Milkfish Workshop-Conference" at Iloilo, Philippines, May 19-22. An effort will be made in due time to prepare a more comprehensive and critically checked revised edition.

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NOTES ON THE CAPTURE, HANDLING AND TRANSPORT
OF THE SABALO OR ADULT MILKFISH,
CHANOS CHANOS (FORSKAL)

by

R. Mateo, O. Paraan and L. Rodriguez

Abstract

The paper describes a simple and inexpensive method of handling and transporting wild sabalo captured from the sea.

Introduction

The sabalo is a fast swimmer and a very wild fish. It is usually trapped in fish corrals locally known as "baklad" during the peak months of April, May and June. It is also caught in the "otoshi-ami" or the Japanese trap net which is a very effective gear recently introduced in the Philippines. Live sabalo can be caught with gill nets also provided the fish is lifted at the right time.

Handling the sabalo in the otoshi-ami or fish corral often presents problems as the fish is very excitable and injures itself by jumping. The first step, therefore, before taking up any program on artificial propagation of milkfish is to develop a technique of transporting and handling sabalo caught from the wild. The paper describes the method followed to solve this important problem.

Materials and Methods

For the study, all the three types of fishing gears as mentioned earlier have been used. The experimental sabalo were caught from 1) otoshi-ami in Pandan Bay, Antique in May, 1975; 2) two fish corrals or "baklad", measuring 200-300 m (approx.) from the shore of Tigbauan, Iloilo in May, 1976; and 3) gill nets from Lusaran, Nueva Valencia, Guimaras in April and May, 1976.

In the first two fishing gears an extra net was used to cover the fish trap to keep the sabalo from escaping. With the slightest

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provocation, the sabalo would jump as high as 8-10 ft (2.66-3.05 m) making its escape easy if no protective net is used. In the third fishing gear, however, immediate careful removal of the fish caught in the net is essential so as to prevent further damage to the fish.

As soon as the sabalo are caught, transporting them presents another serious consideration. In our preliminary studies which were conducted early in May 1975, 2-phenoxy-ethanol tranquilizer was used but the milkfish spawners died shortly after the treatment.

In the middle of May 1975, a simple but more effective method of transporting sabalo alive from the otoshi-ami was developed. The fish was simply put inside a wooden tank with sea water, turning its ventral side up and its dorsal side down, in an inverted position.

Upon reaching the shore, the fish was laid on a trough of double "M" metal frame, lined with urethane foam (Plate 1). The nape and tail of the fish fitted snugly on the trough, thereby holding it securely in the inverted position (Plate 2). The fish was tied further after wrapping a thin plastic sheet and a urethane foam around its belly in order to prevent any, injury and to secure it during the transport.

Then, the entire set-up was kept inside a one-ton PVC tank (Plate 3). The PVC tank, filled with sea water contained two to four sabalo during every transport. It is also continuously aerated with pure oxygen and cooled down to about 20°C to reduce the basal metabolic rate of the fish.

In May 1976, sabalo were handled using basically the same technique as previously developed in Pandan. In order to minimize body injuries a fine-meshed plankton net was utilized in transferring the sabalo from the fish corral to a wooden box (Plate 4) in a motorized boat. This box was designed with "V" slots lined with foam to support and stabilize the inverted fish.

To secure adequate supply of oxygen and remove traces of previously applied topical medication (Betnovate C and Whitfield ointments), sea water was changed manually every 3-5 minutes while in transit. The abdomen of every fish was examined by pressing gently for possible eggs or milk. Morphological measurements were also taken.

From time to time, several batches of sabalo were brought to the Igang Sea Farming Station (Plate 5) and to the

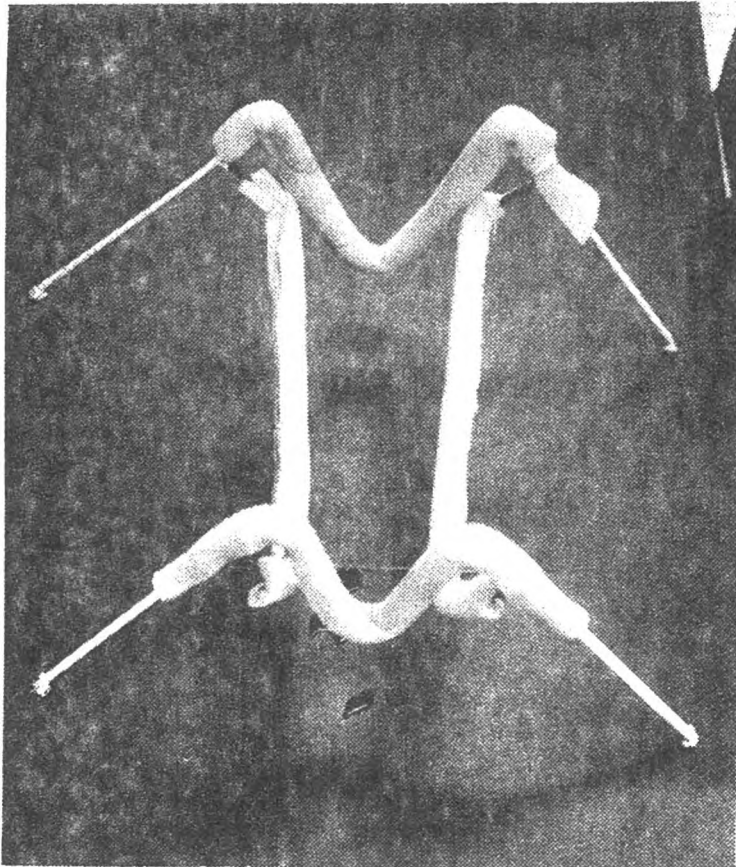


Fig. 1. Double "M" metal frame for holding sabalo.

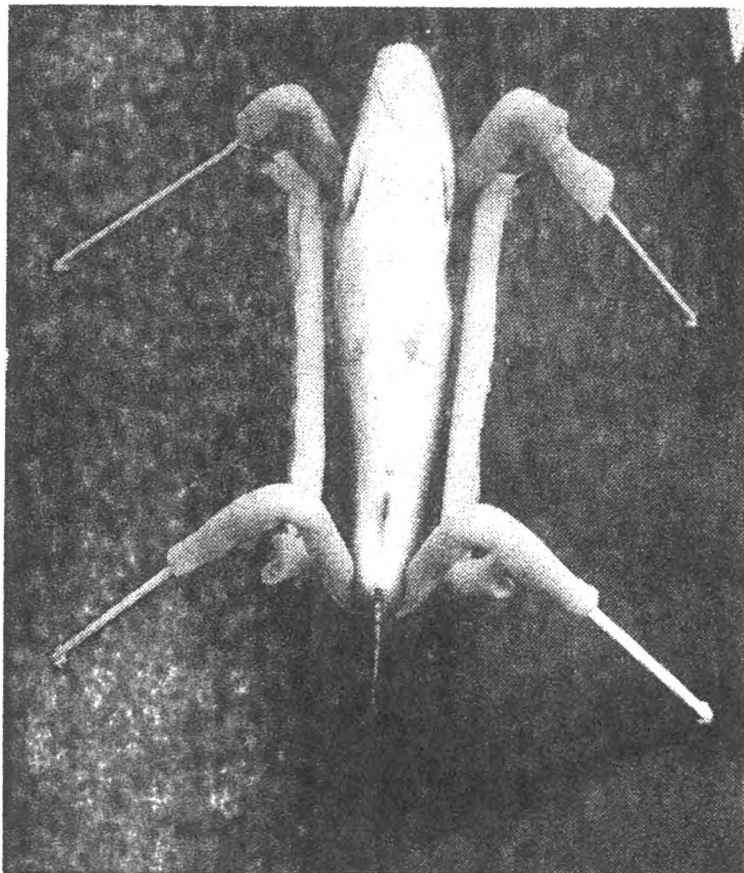


Fig. 2. Sabalo in inverted position on double "M" metal frame.

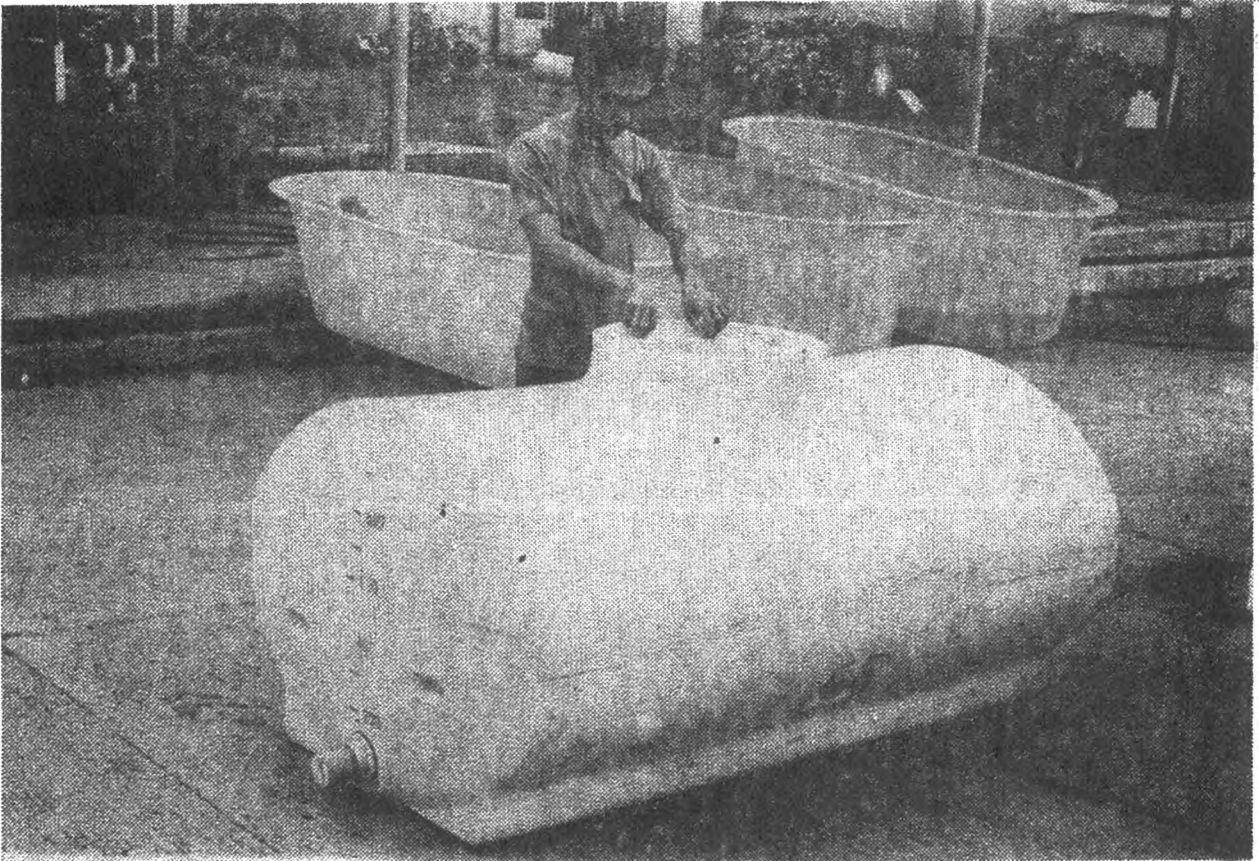


Fig. 3. PVC tank used in the transport of sabalo.

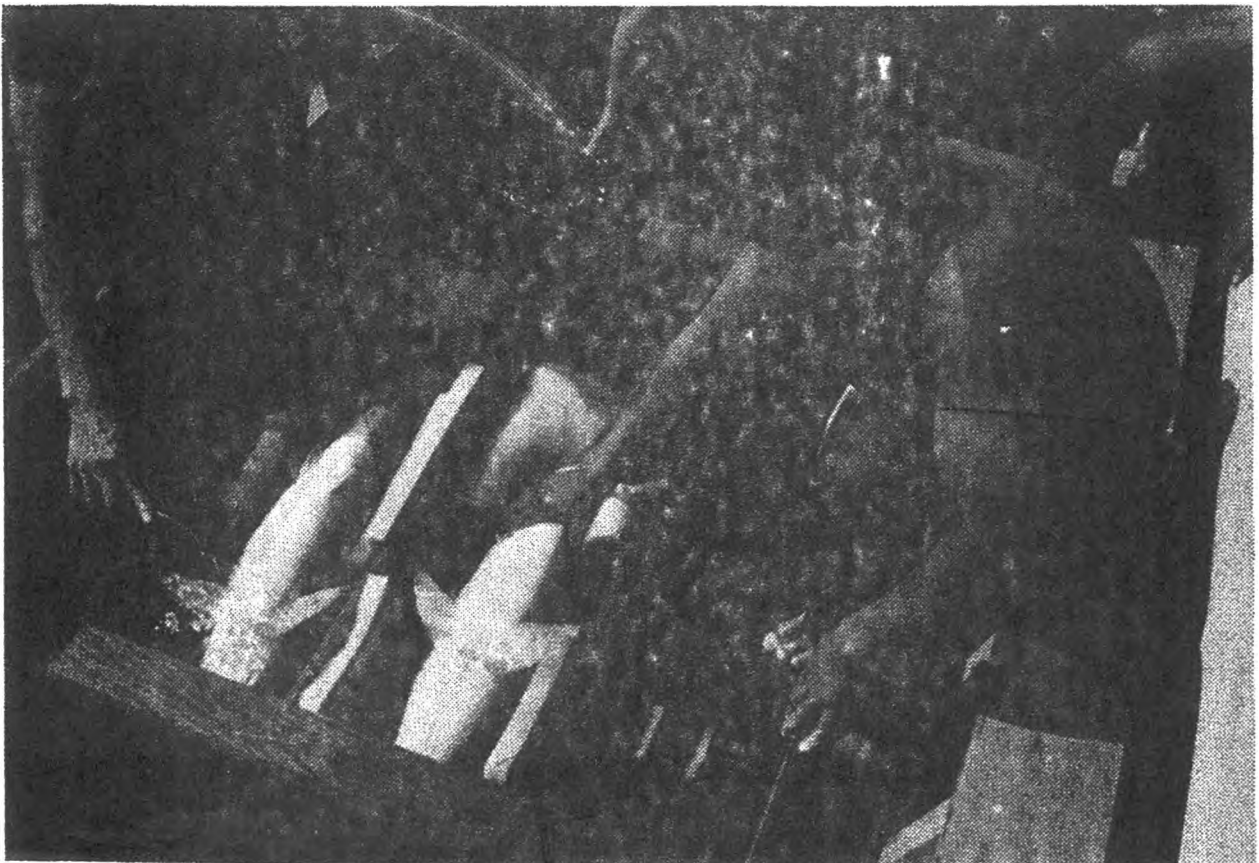


Fig. 4. Wooden tank with "V" slots for holding sabalo.

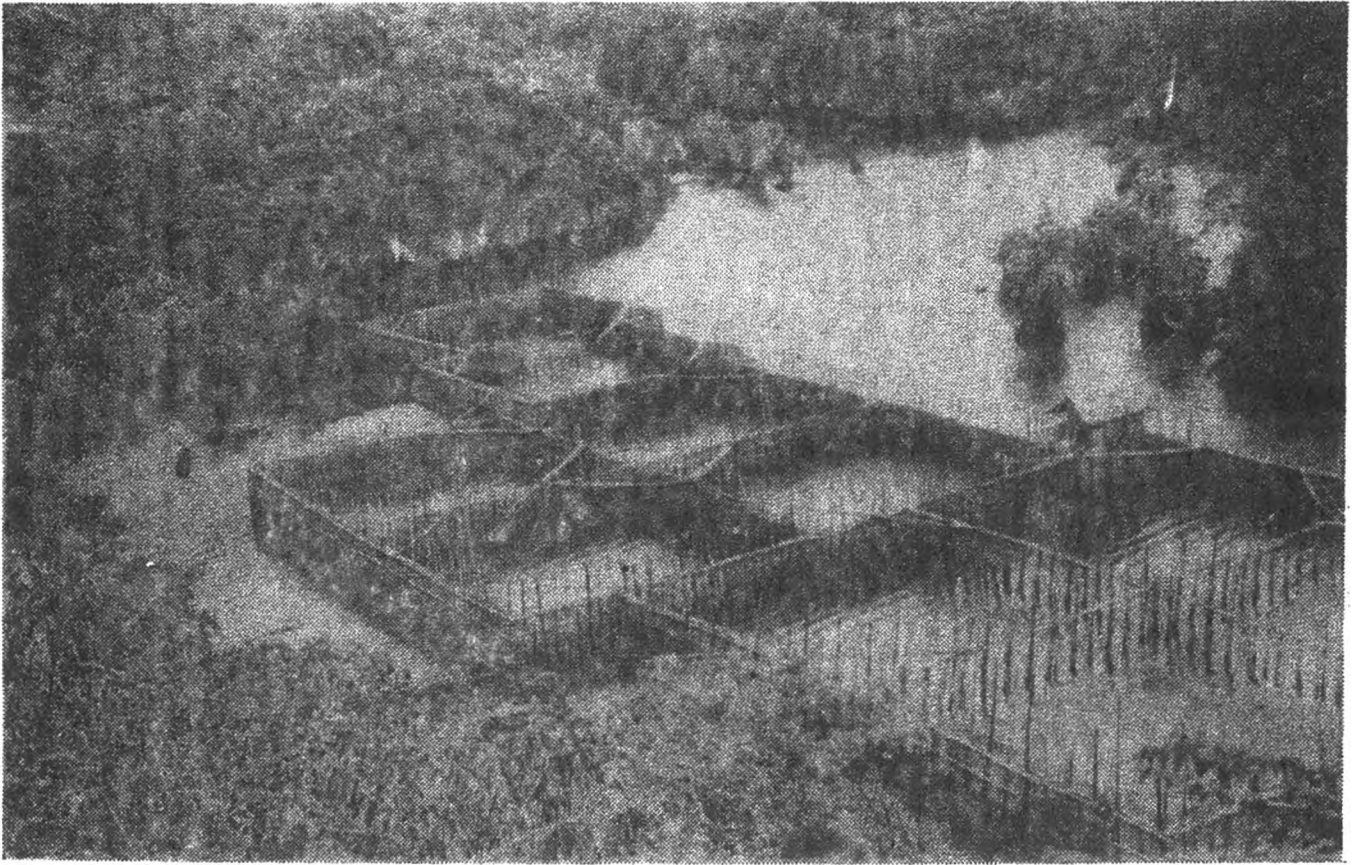


Fig. 5. Fishpen in Igang

Tigbauan Hatchery which are about 13 nautical miles and 200 m respectively from the catching site. Boat transport to Igang took about 1 1/2 hours, compared to Tigbauan which took only 3-5 minutes.

Results and Discussion

It has been observed from the above stated experiments that like highly domesticated Cyprinus, the sabalo also became passive and quiet when placed in an inverted position. This handling technique, therefore, facilitated transport even without the use of tranquilizers.

Several trials were made in 1975 at the SEAFDEC Pandan Station (Table 1). Out of the 13 sabalo which were transported, 10 survived the transport stress during the 200 km trip over rough road for four to five hours, and lived for at most 2 days in the Tigbauan hatchery. This result is significant because the sabalo that had been kept in the otoshi-ami for over a week duration were already badly injured even before handling and transport. The fish kept on bumping their heads against the net in their futile attempt to escape. As a result, the heads and bodies were badly bruised and in some cases the dematocranium was already exposed.

In 1976, sabalo transport operation continued from the fish corrals in Tigbauan and the gill nets in Lusaran to the Igang Station. In addition, sabalo were also transported from the fish corrals to the Tigbauan hatchery.

All the fish that were transported by the inverted method reached their destination alive (Tables 2, 3 & 4). However, some of them died of injuries due to their aborted attempts to escape out of the bamboo fish pens in Igang (Plate 5). Three sabalo died after hurting themselves when they jumped consecutively over one or two adjacent compartments.

As of May 20, 1976, two sabalo are still alive in Igang and six in Tigbauan. One sabalo in Igang has been kept alive in captivity for 22 days while another sabalo in Tigbauan for 16 days.

Between the first day of transport to about five to six days later, the adipose tissue covering the the sabalo's eye and side of the head, together with the dorsal surface of the head, would turn opaque

from its original translucent state. At this stage, the fish also became practically blind, and one could catch it with his bare hands.

Thereafter, the eyecover of the fish started recuperating gradually to its original translucent condition in about five to seven days.

The oldest live sabalo in Tigbauan which was caught on 4 Ma, 1976 was transferred four days later from the open hatchery tank containing sea water to the roofed hatchery tank No. 5 with salinity of 14‰ since the fish developed opaqueness of the eyecover. On 11 May 1976, the eyecover started clearing and by 16 May 1976, recovered completely.

It was, therefore, thought that reducing the salinity did the trick in effecting recuperation. So we decided to transfer to the brackishwater RH tank No. 5 the other 3 sabalos caught recently on 14 and 15 May and having opaqueness of the eyecovers; but inspite of the transfer to lower salinity the opaqueness continued.

Table 1. Result of sabalo transport from the otoshi-ami in May, 1975 in Pandan

Trial No.	Total No. of Sabalo transported	No. died on the way	No. survived the trip to Tigbauan	Length of survival at Tigbauan hatchery (in hours)
1	3	1	2	40 1/2
2	2	0	2	48 42
3	4	2	2	24 37
4	2	0	2	25 36
5	2	0	2	25 28
	13	1	12	40-longest

Table 2. Result of sabalo transport from the fish corrals to Igang in April and May, 1976.

<u>Trial No.</u>	<u>Date collected</u>	<u>Total no. of sabalo transported</u>	<u>No. died on the way</u>	<u>No. survived the trip</u>	<u>Length of survival (in days)</u>
1	30 Apr 1976	1	0	1	Still alive*
2	2 May 1976	1	0	1	3
3	3 May 1976	1	0	1	2
4	5 May 1976	1	0	1	1

*For 20 days as of 20 May 1976.

Table 3. Result of sabalo transport from gill net in Lusaran to Igang in April and May, 1976.

<u>Trial No.</u>	<u>Date collected</u>	<u>Total no. of sabalo transported</u>	<u>No. died on the way</u>	<u>No. survived the trip</u>	<u>Length of survival (in days)</u>
1	28 Apr 1976	1	0	1	Still alive*
2	6 May 1976	1	0	1	3
3	10 May 1976	1	0	1	1

*For 22 days as of 20 May 1976.

Table 4. Result of sabalo transport to the Tigbauan hatchery.

<u>Trial No.</u>	<u>Date collected</u>	<u>No. of sabalo</u>	<u>No. died on the way</u>	<u>No. survived the trip</u>	<u>Length of survival (in days)</u>
1	4 May 1976	2	0	2	4 Still alive*
2	8 May 1976	4	0	4 Sti	1 1 Still alive (2)*
3	9 May 1976	3	0	3	1 1 Still alive (1)*
4	14 May 1976	1	0	1	Still alive*
5	15 May 1976	2	0	2	Still alive*

*As of 20 May 1976.

A PRELIMINARY STUDY ON THE PURIFIED TEST DIET FOR
YOUNG MILKFISH, CHANOS CHANOS*

by

Dong-Liang Lee and I-Chiu Liao**

Abstract

In studying the nutritional requirements of young milkfish, experiments were conducted to develop a purified test diet. Mixtures of the purified constituents tested were: vitamin-free casein, vitamin-free gelatin, supplemented with L-tryptophan and L-cystine as the protein sources; shark liver oil and soybean oil as the fat sources; and dextrin as the carbohydrate source. Mineral mixture and vitamin mixture were also added.

The results showed that a test diet containing vitamin-free casein supplemented with L-tryptophan as the protein source, was best for the growth of young milkfish. Soybean oil was found to be a better source of fat. Vitamin mixture (4%) and mineral mixture (10%) were observed to promote growth in young milkfish. A purified test diet consisting of vitamin-free casein 60%, L-tryptophan 0.5%, soybean oil 10%, vitamin mixture 4%, mineral mixture 10%, carbohydrate and others 16% was thus suggested for young milkfish.

Introduction

Milkfish farming was well established in Taiwan for more than three hundred years and had played an important role in the island's food economy ever since. However, there is hardly any information available on the nutritional requirements of milkfish. It is fundamental to develop a purified test diet which maintains the normal growth.

The present study is largely based on the purified test diet tried on eel (Arai et al., 1971) with some modification. In this preliminary investigation, an attempt has been made to find out an ideal source of protein and fat, and the optimum quantity of vitamin mixture and

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mineral mixture needed for the normal growth of young milkfish. The effect of supplementary amino-acid on the growth of milkfish was also studied.

Materials and Methods

Experimental fish and feeding method

The experimental fish were young milkfish originally imported from Indonesia in October, 1974. They were overwintered in fishpond and reared in cement tank with purified diet for six weeks prior to the initiation of the present study. Fish of about 1.7 g in body weight were chosen and transferred to grey PVC aquaria (50 cm x 25 cm x 15 cm deep), each containing 25 fishes. Aeration and running water at a velocity of 1.5 liter/min were provided throughout the experiment. During the experiment period, water temperature and salinity ranged from 25 to 28°C and from 24 to 30‰ respectively. At 9:00 in the morning, fish were fed with the compound feed formed in the shape of noodles by pressing through 50 ml nylon syringes. The feeding was repeated every 2.5 hrs, totaling four times each day. The aquaria were cleaned every two days and once in every two weeks the body weight of experimental fish was taken after anesthetizing in 1.0 ppm MS-222 solution.

Test diets

The main constituents of test diets are vitamin-free casein of Difco Laboratory (U.S.A.) and vitamin-free gelatin of the Matheson Company Inc. (U.S.A). Mineral mixture and vitamin mixture were added as recommended for chinook salmon Halver, (1975) except that the amount of vitamin added was doubled. In making the test diets, vitamin-free casein, dextrin, mineral mixture, vitamin mixture, amino acids and carboxymethyl cellulose were mixed in mortar first and then oil and solution of vitamin-free gelatin in water were added. The compound feed was preserved at -20°C in a freezer.

Results and Discussion

Experiment on protein

The effect of various protein on the growth of young milkfish was investigated. The composition of five test diets including various contents of vitamin-free casein and vitamin-free gelatin as protein sources for this experiment is given in Table 1. Each diet had a total of 60% protein content.

Growth of young milkfish that fed on these five test diets for 36 days is shown in Table 2 and illustrated in Fig. 1. There was best growth of fish in Lot 1 that fed on diet with only vitamin-free casein as protein source. The higher the content of vitamin-free gelatin in the test diet was, the poorer the growth obtained. The probable reason might be that gelatin was hard for young milkfish to digest or that the amino acid imbalance influenced the growth rate of milkfish in the presence of gelatin.

Experiment on fat

The effect of different sources of fat in the test diet on the growth of young milkfish was investigated. The test diet consisting of vitamin-free casein, vitamin-free gelatin, vitamin mixture, mineral mixture and dextrin was supplemented with 10% fat. Six test diets with various amount of shark liver oil and soybean oil were made (Table 3.) for this experiment.

Data on the growth of young milkfish fed on these six test diets for 36 days is tabulated in Table 4 and illustrated in Fig. 2. The best growth was observed in Lot 6 of the diet with only soybean oil as fat source. Poorer growth resulted when fed with lower content of soybean oil and higher content of shark liver oil. The worst growth of fish was observed in Lot 10 fed on the test diet containing only shark liver oil as the fat source. The longer the term for feeding, the slower the growth rate was. In Lots 9 and 10 with higher content of shark liver oil, 30% of experimental fish showed injury of fins after being reared for 36 days. This may probably be due to the deficiency of essential fatty acids or poisonous effect of peroxide of shark oil liver.

The fat requirement usually differs in different species of fish. For example, Halver (1957) found that corn oil was best for chinook salmon, while compound of corn oil and fish liver oil was found best for rainbow trout and eel by Halver and Coates (1957) and Arai et al.

Table 1. Composition of the test diets in experiments with protein

Lot No.	1	2	3	4	5
Vitamin-free casein	56	51	46	41	36
Vitamin-free gelatin	0	5	10	15	20
Soybean oil	8	8	8	8	8
Shark liver oil	2	2	2	2	2
Vitamin mixture*	4	4	4	4	4
Mineral mixture	4	4	4	4	4
Dextrin	14.5	14.5	14.5	14.5	14.5
L Tryptophan	0.5	0.5	0.5	0.5	0.5
L-Cystine	1	1	1	1	1
Carboxymethyl cellulose	10	10	10	10	10
Water	200	200	200	200	200

*Vitamin-free casein was used as carrier for vitamin mixture and the amount of vitamin mixture added to each diet was the same as that reported by Halver (1957).

Table 2. Growth of young milkfish in experiment with protein.

Lot no.	Number of fish	Average body weight (g)		Growth rate (%)
		Initial	Final	
1	25	1.65 \pm 0.64	3.09 \pm 0.90	86.93
2	25	1.77 \pm 0.64	2.70 \pm 0.96	52.19
3	25	1.60 \pm 0.45	2.53 \pm 0.85	58.22
4	25	1.59 \pm 0.49	2.76 \pm 0.99	73.80
5	25	1.56 \pm 0.43	2.36 \pm 0.92	51.67

Table 3. Composition of the test diets in experiment with fat.

Lot No. Ingredients	6	3	7	8	9	10
Vitamin-free casein	46	46	46	46	46	46
Vitamin-free gelatin	10	10	10	10	10	10
Soybean oil	10	8	6	5	2	0
Shark liver oil	0	2	4	5	8	10
Vitamin mixture*	4	4	4	4	4	4
Mineral mixture	4	4	4	4	4	4
Destrin	14.5	14.5	14.5	14.5	14.5	14.5
L-Tryptophan	0.5	0.5	0.5	0.5	0.5	0.5
L-Cystine	1	1	1	1	1	1
Carboxymethyl cellulose	10	10	10	10	10	10
Water	200	200	200	200	200	200

*Vitamin-free casein was used as carrier for vitamin mixture and the amount of vitamin mixture added to each diet was the same as that reported by Halver (1957).

Table 4. Growth of young milkfish in experiment with fat.

Lot No.	Number of fish	Average body weight (g)		Growth rate (%)
		Initial	Final	
6	25	1.74 ± 0.48	2.79 ± 0.98	59.88
3	25	1.60 ± 0.45	2.53 ± 0.85	58.22
7	25	1.59 ± 0.46	2.41 ± 0.92	51.19
8	25	1.63 ± 0.49	2.25 ± 0.86	38.37
9	25	1.38 ± 0.31	2.03 ± 0.45	46.78
10	25	1.77 ± 0.53	2.22 ± 0.68	25.42

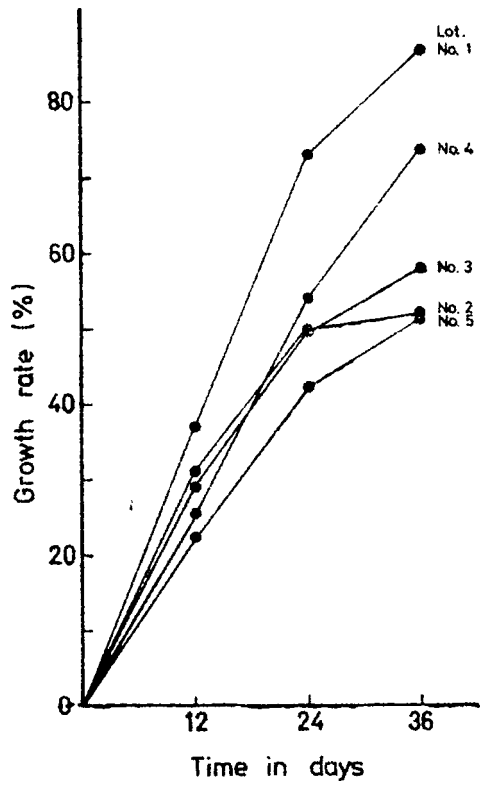


Fig. 1 Body weight gain in various lots of experiment on protein.

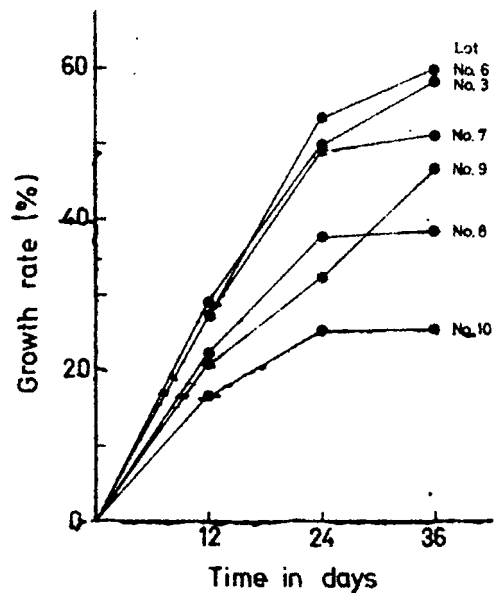


Fig. 2 Body weight gain in various lots of experiment on fat.

(1971) respectively. The growth of both rainbow trout (McLaren et al., 1947) and yellowtail (Tsukahara et al., 1967) was not good when fed with corn oil alone as the fat source but could be improved with supplement of fish liver oil. Yone et al. (1971) found that sea bream grew well with fish liver oil as its only fat source. However, the present experiment demonstrated that soybean oil was quite good for the growth of young milkfish.

Experiment on vitamin mixture and mineral mixture

In order to study the effect of vitamin mixture and mineral mixture on the growth of young milkfish, the experiment was done by following the prescription of Halver (1957) on chinook salmon. As shown in Table 5, four test diets containing various quantity of vitamin mixture ranging from 0% to 6% and five test diets containing various quantity of mineral mixture ranging from 2% to 10% were tested.

The results obtained after rearing for 36 days showed that young milkfish grew better as the vitamin mixture content was increased up to 4% but the growth rate was lower when increased to 6%. The retardation of appetite and body weight started on the 12th day in Lot 11 which were fed on the vitamin-free test diet. It was followed by high mortality which increased to 50% on the 36th day. The result is presented in Table 6 and illustrated in Fig. 3.

As has been noted in the present experiment, young milkfish showed low appetite, decreased body weight and high mortality when vitamin in diet was insufficient. Although vitamin is essential for fish nutrition, it is generally believed that overdose of vitamin is not of any use.

The result of experiments conducted on the requirement of mineral mixture is presented in Table 6 and illustrated in Fig. 4. There was no remarkable difference in growth among young milkfish fed on the diet containing various quantity of mineral mixture. Only from Lot 17 was obtained and so it seemed that 10% mineral mixture would be beneficial for young milkfish.

It is well known (Phillips, 1959), that fish get the necessary minerals from environmental waters through gills and skin. Such capacity, however, varies from species to species. Wolf (1951) found no bad effect when rainbow trout was fed with mineral-deficient diet

Table 5. Composition of the test diets in experiment with vitamin mixture and mineral mixture.

Ingredients	Experiment of vitamin mixture				Experiment of mineral mixture					
	Lot No.	11	12	3	13	14	3	15	16	17
Vitamin-free casein		50	48	46	44	46	46	46	46	46
Vitamin-free gelatin		10	10	10	10	10	10	10	10	10
Soybean oil		8	8	8	8	8	8	8	8	8
Shark Liver oil		2	2	2	2	2	2	2	2	2
Vitamin mixture*		0	2	4	6	4	4	4	4	4
Mineral mixture		4	4	4	4	2	4	6	8	10
Dextrin		14.5	14.5	14.5	14.5	16.5	14.5	12.5	19.5	8.5
L-Tryptophan		0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5
L-Cystine		1	1	1	1	1	1	1	1	1
Carboxymethyl cellulose		10	10	10	10	10	10	10	10	10
Water		200	200	200	200	200	200	200	200	200

* Vitamin-free casein was used as carrier for vitamin mixture and the amount of vitamin mixture added to each diet was the same as that reported by Halver (1957).

Table 6. Growth of young milkfish in experiment with vitamin mixture and mineral mixture.

Lot No.	Number of fish	Average body weight (g)		Growth rate (%)
		Initial	Final	
Experiment 11	25	1.85 ± 0.62	---	---
of vitamin 12	25	1.62 ± 0.31	2.32 ± 0.57	43.56
mixture 3	25	1.60 ± 0.45	2.53 ± 0.85	58.22
13	25	1.60 ± 0.45	2.21 ± 0.69	37.86
Experiment 14	25	1.34 ± 0.18	2.15 ± 0.48	60.32
of mineral 3	25	1.60 ± 0.45	2.53 ± 0.85	58.22
mixture 15	25	1.53 ± 0.39	2.31 ± 0.70	50.58
16	25	1.59 ± 0.60	2.42 ± 0.94	52.01
17	25	1.73 ± 0.56	2.92 ± 0.99	68.00

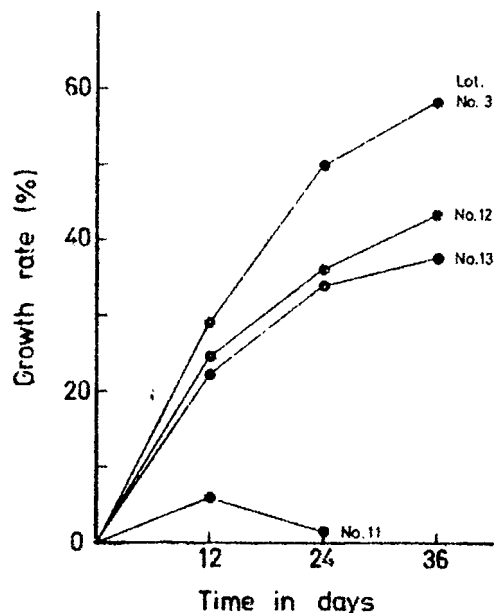


Fig. 3 Body weight gain in various lots of experiment on vitamin mixture.

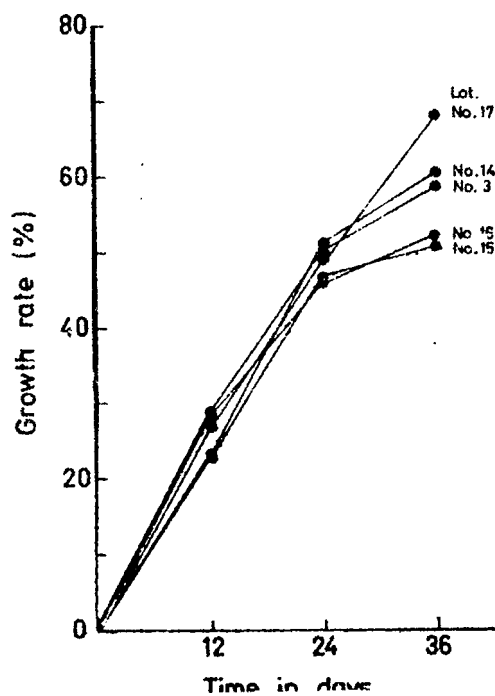


Fig. 4 Body weight gain in various lots of experiment on mineral mixture.

for a long period of 23 weeks. On the contrary, Arai et al. (1971) reported the syndrome of low appetite hindered growth and high mortality in eel fed on mineral-deficient diet in the second week. They obtained best growth in the group fed with 8% mineral mixture. Young milkfish required approximately 10% mineral mixture in diet, close to that found in eel.

Experiment on supplement of amino acids

It is noted in the experiment on protein that milkfish grew well when fed on the diet with vitamin-free casein as the only protein source. For demonstration the effect of vitamin-free casein supplemented with crystalline L-amino acid, seven test diets with various amount of tryptophan, cystine, methionine and threonine as shown in Table 7 were tested.

The experiment was conducted for 28 days and it was found that all groups with supplementary amino acid had better growth than those without it (Table 8 and Fig. 5). Especially, fish in Lot 21 with 0.5% tryptophan showed the most satisfactory growth. From these results, it is believed that the amino acid balance would be more suitable for young milkfish after the supplement of tryptophan made the tryptophan content in casein more complete. Lots 19, 20 and 23 showed a decreased effect of supplementing with 0.5% tryptophan and additional cystine, methionine and threonine that might have changed the amino acid balance. Similar result was reported by Arai et al. (1971) in their study on test diet for eel. There was significant increase of eel growth when fed on the diet containing casein and gelatin as protein sources supplemented with 0.5% tryptophan and 1% cystine. However, an additional 0.5% threonine and 1% methionine showed the bad effect on its growth.

From the results obtained in above experiments, a purified test diet consisting of vitamin-free casein 60%, L-tryptophan 0.5%, soybean oil 10%, vitamin mixture 4%, mineral mixture 10%, carbohydrate and others 16% was suggested for young milkfish.

Acknowledgements

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Table 7. Composition of the test diets in experiment with supplement of amino acids.

Ingredient	Lot No.	18	19	20	21	22	23	24
Vitamin-free casein		51.5	50	50	51	50.5	49.5	49.5
Soybean oil		10	10	10	10	10	10	10
Vitamin mixture*		4	4	4	4	4	4	4
Mineral mixture		10	10	10	10	10	10	10
Dextrin		14.5	14.5	14.5	14.5	14.5	14.5	14.5
L-Tryptophan			0.5	0.5	0.5		0.5	
L-Cystine			1				1	1
L-Methionine				1		1		1
L-Threonine							0.5	
Carboxymethyl cellulose		10	10	10	10	10	10	10
Water		200	200	200	200	200	200	200

*Vitamin-free casein was used as carrier for vitamin mixture and the amount of vitamin mixture added to each diet was the same as that reported by Halver (1957).

Table 8. Growth of young milkfish in experiment with supplement of amino acids.

Lot No.	Number of fish	Average body weight (g)		Growth rate (%)
		Initial	Final	
18	20	4.06 ± 1.45	5.02 ± 1.69	23.63
19	20	4.14 ± 1.34	5.16 ± 1.61	24.70
20	20	3.95 ± 1.29	5.18 ± 1.88	31.06
21	20	3.77 ± 1.33	5.06 ± 2.15	33.99
22	20	3.89 ± 1.10	4.94 ± 1.80	26.79
23	20	3.82 ± 1.25	4.95 ± 1.68	29.58
24	20	4.12 ± 1.27	5.28 ± 1.82	28.05

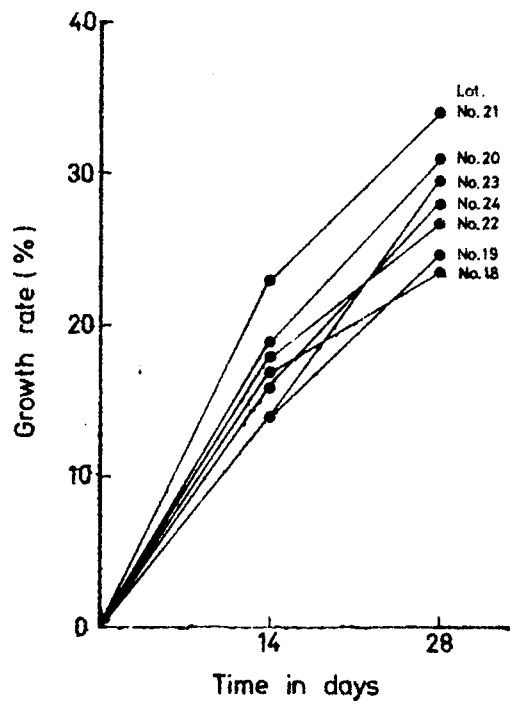


Fig. 5 Body weight gain in various lots of experiment on supplement of amino acids.

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A PRELIMINARY REPORT ON THE GONADAL DEVELOPMENT
OF ADULT MILKFISH, CHANOS CHANOS, REARED IN TANK*

by

I-Chiu Liao and Yea-Sha Chang**

Abstract

Milkfish is one of the most important food fishes in Taiwan. There are more than 16,000 ha of culture area and over 160 millions of fry are needed for milkfish farming industry every year. The fry are collected from the sea and also imported from other countries. However, due to several environmental factors, there is unpredictable fluctuations in the occurrence of these wild fry. In recent years, the demand for milkfish fry has gone up considerably owing mainly to the fast-growing populations, the natural resources being so limited that there is insufficient supply of stocking materials of this important foodfish.

To solve the problem of shortage of milkfish fry, Tungkang Marine Laboratory started the preliminary work on artificial propagation of milkfish in 1970. In addition to capturing wild spawners, the Laboratory has also been raising the adult milkfish in tanks for this objective. After being reared for six years, one male and one female were dissected on 11 April 1976. The male had ripe sperms; the testes weighing 4.63 g with the GSI of 0.12. The gonad of the female weighed 21.20 g with the GSI of 0.66 and part of the ovarian oocytes was found to be at the oil droplet (yolk vesicle) stage. Judging from the condition of maturity of the above female, the feasibility of raising tank-reared spawners was ensured. It is believed that this is the first attempt on the world and is the prelude to successful artificial propagation by using tank-reared milkfish as spawner.

Introduction

The milkfish, *Chanos chanos* is one of the best cultivable fishes in Taiwan, being euryhaline, disease resistant, popular and most economical to grow. There are more than 16,000 ha of area under milkfish culture in Taiwan. At a stocking density of 10,000 per hectare, the annual requirement for milkfish fry exceeds 160 million. In addition, the

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demand for milkfish fry is increasing because young milkfish is found to be an ideal longlining bait in recent years and also because a great many milkfish fry and fingerlings often die of frigid weather during overwintering. Besides, there is unpredictable fluctuations in the occurrence of wild fry due to many environmental factors which further limit the quantity of stocking materials of milkfish.

Usually, only 85% of the total requirements of fry in Taiwan is captured from the wild. The deficiency is made up by importing fry from the Philippines and Indonesia. Recently, supply of seed in both countries are limited. In 1974, only 13% of the demand was collected in the Philippines and most milkfish ponds remained idle (Schmittou, 1975). Such a serious downward tendency will undoubtedly become a big problem in milkfish farming industry.

With a view to supplying an adequate quantity of fry, Tungkang Marine Laboratory initiated the preliminary work on artificial propagation of milkfish in 1970. In addition to capturing wild spawners, the Laboratory has been raising milkfish in cement tanks since then. After being reared for five years, one male and one female adult milkfish were dissected on 13 May 1975. Again on the sixth year of rearing, one male and one female were sacrificed on 11 April 1976. The results were highly encouraging. The male aged five years had mature sperm. The gonadal development of the female of six years of age was more advanced than that of five years. Its gonad weighed 21.20 g with the GSI of 0.66. Part of its ovarian oocytes was found to be at oil droplet (yolk vesicle) stage. Judging from the condition of said gonadal development, the feasibility of raising tank-reared milkfish as spawners and the prospect for their successful artificial propagation are fairly ensured.

Materials and Methods

The materials from the present study were wild milkfish fry collected in the shoreline of Tungkang coast in 1970, and had been reared in cement tanks at Tungkang Marine Laboratory for five and six years.

Two hundred fishes were reared in a rectangular cement tank (10 m x 20 m x 1.5 m deep) for the first three years. Thirty fishes were randomly selected from these and transferred to a round cement tank (8.25 m in diameter, and 1.5 m in depth) (Plate I-1). In the fifth year, one pair of fishes were dissected for investigation on gonadal development. Again, twenty fishes were chosen and continued to be reared in round cement tank of the same size.

The fishes were fed daily in the morning with a compound feed, the composition of which is given in Table 1. The amount given was just sufficient for their satiation.

Aeration was provided throughout the rearing period and water was placed when necessary. The replacement was made more frequently in summer because of the fast growth of phytoplankton. The fluctuation of average water temperature and salinity of rearing water in last one and a half year is shown in Fig. 1. The water temperature ranged from 19.2 to 29.8°C and salinity from 15.78 to 31.18 ‰.

Table 1. Composition of compound feed.

<u>Feedstuff</u>	<u>Fish meal</u>	<u>Rice bran</u>	<u>Yeast</u>	<u>Soy bean</u>	<u>Wheat flour</u>	<u>Soy bean oil</u>	<u>Multiple vitamins</u>
%	25	20	20	15	19	0.5 ~ 1.0	0.1 ~ 0.2

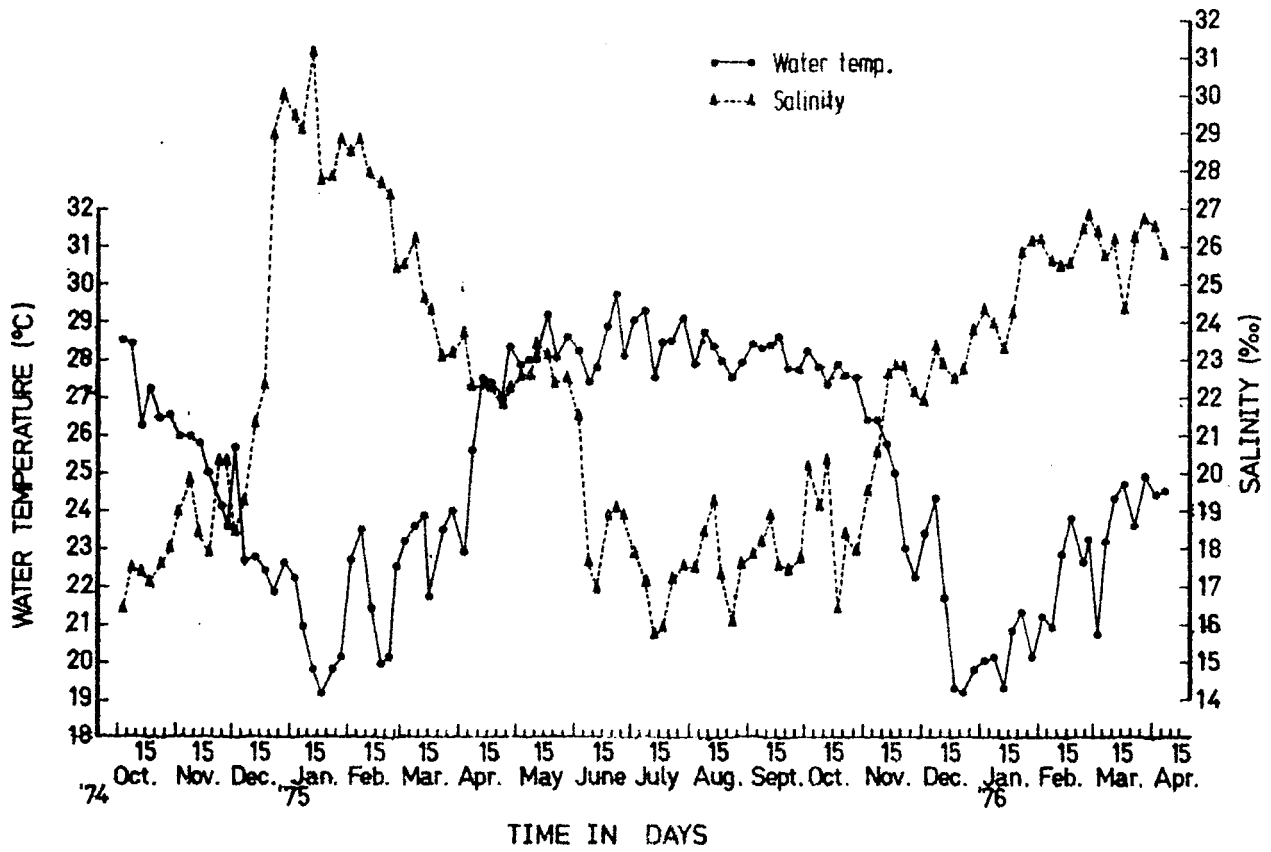


Fig. 1 The fluctuation of average water temperature and salinity of rearing water in late one and a half year.

Two pairs of milkfish were harvested respectively on 13 May 1975 and 11 April 1976 (Plate 2-1). The external morphology and genital pores were carefully observed to distinguish the male from the female. They were then sacrificed. Measurement of body weight and body length was made before dissection and gonadal weight was also determined. The gonad was fixed in Bouin's fluid and stained with Delafield's hematoxylin and eosin for histological study.

Results

The sex of milkfish was fairly hard to be distinguished by their external characteristics as reported by Tampi (1957). Both during 1975 and 1976 a pair of milkfish could be selected with great difficulty and confirmation made only after the dissection.

The ovaries or testes of milkfish are more or less symmetrically developed, being suspended in coelomic cavity by mesovarium or mesorchium which connects their dorsal inner side with the peritoneal wall.

The ovary consists of numerous ovigerous lamellae where ovarian oocytes are present. The inner side of ovaries is surrounded with fibrous ovarian wall whereas ovigerous lamellae on outer side are exposed in coelom. As shown in Table 2 and Plates I-3 and I-5, the ovary of adult milkfish aged five years weighed 6.99 g with the GSI of 0.23 and most of the ovarian oocytes observed from the histological section of the ovary were found at the primary stage. Only 0.54% of the ovarian oocytes in the present study were at oil droplet (yolk vesicle) stage (Yamamoto et al., 1965; Hayashi, 1975) and the rest (99.46% were still at peri-nucleolus stage or even at the chromatin-nucleolus stage. The ovary of the tank-reared milkfish aged six years weighed 21.20 g with the GSI of 0.66. The ovary was yellowish orange in color with distinctly separate eggs. The biggest ones measured 0.4 mm in diameter. Histological study of ovarian section showed that 8.41% of the ovarian oocytes were at oil droplet (yolk vesicle) stage and with some extra small eosinophilic granules while the rest (91.59%) were at the peri-nucleolus stage or even at the earlier stage. In other words, the ovary of milkfish aged six years is found comparatively more mature than that of five-year old female.

Table 2. Gonadal development of tank-reared adult milkfish.

<u>Fish No.</u>	<u>Date</u>	<u>Sex</u>	<u>Age (yr)</u>	<u>B.W. (kg)</u>	<u>T.L. (cm)</u>	<u>G.W. (g)</u>	<u>GSI</u>	<u>Gonadal development</u>
1	13-5-75	♀	5	2.99	71.95	6.99	0.23	Immature; 99.46% of oocytes at earlier and peri-nucleolus stage; 0.54% at oil droplet (yolk vesicle) stage.
2	13-5-75	♂	5	2.34	64.55	0.94	0.04	Mature; spermatogenesis in process with spermatocytes at various stages.
3	11-4-75	♀	6	3.20	70.04	21.20	0.66	Immature; 91.59% of oocytes at earlier and peri-nucleolus stage; 8.41% at oil droplet (yolk vesicle) stage.
4	11-4-76	♂	6	3.89	72.67	4.63	0.12	Mature; spermatogenesis in process with spermatocytes at various stages.

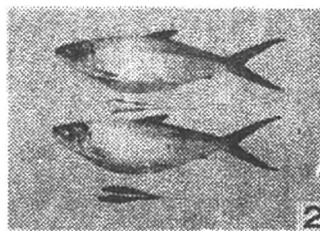
The testes were covered with a layer of tunica forming the smooth surface. The testes appeared tough, flat and greyish white in color. Each testes consists of numerous testicular lobules that have lumen inside and have spermatogonia in peripheral zone. As shown in Plates I-4 and I-6, there were mature spermatozoa and many spermatocytes at various stages of development in testicular lobules of milkfish aged five or six years. In fact, there was no distinct difference in the stages of maturity of testes between five years and six years old milkfish. The only difference was that the testes of the latter weighed heavier than those of the former.

Discussion

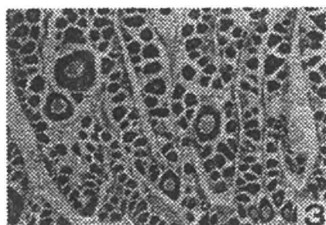
Tampi (1957) was the first scientist to report on this subject. He made a detailed and clear description of the structure of milkfish gonad. Comparing with the more popular classification of gonadal maturity stages as described by Yamamoto et al. (1965) and Hayashi (1972), the one



The round cement tank for rearing the adult milkfish (8.25 m in diameter, 1.5 m in depth)



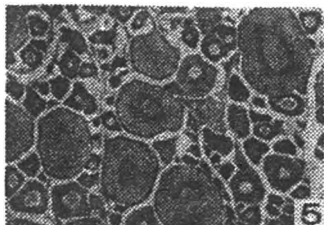
The adult tank-reared milkfish (above, male; below, female) and their gonads. (Fish No. 3 and 4 in Table 2)



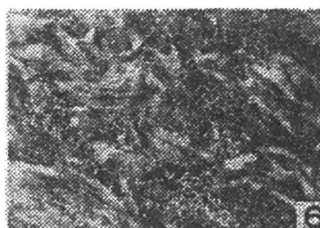
Section of the ovary of Fish no. 1 in Table 2 showing the small oocytes at primary stage.



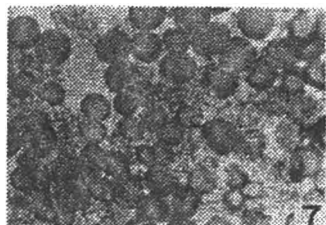
Section of the testis of Fish no. 4 in Table 2.



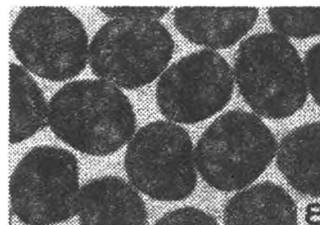
Oocytes at oil droplet (yolk vesicle) stage and peri-nucleolus stage in the ovary of Fish no. 3 in Table 2.



Section of the testis from Fish no. 2 in Table 2.



Immature eggs (0.3 - 0.4 mm in diameter) of Fish no. 3 in Table 2.



Nearly mature eggs (0.89 x 0.98 - 1.32 x 1.50 mm in diameter) from Fish no. 1 in Table 3.

described by Tampi (1957) is somewhat broad; he classified it into four stages depending on the degree of development, for his convenience. Tampi's description is not complete probably because he did not come across the intermediate stages. Since there is no other paper that gives more confirmative and complete information on the gonadal development of milkfish, the classification of Yamamoto et al. and Hayashi has been followed in the present study.

Ovary of milkfish is of "gynovarian type" but not "cystovarian type" as in most teleosts, while its testes is just like that of other teleosts,

The biggest ovarian oocytes of fish aged six years measured 0.4 mm in diameter and were found already at oil droplet (yolk vesicle) stage when vitellogenesis was ongoing as the important step towards attaining maturity. As shown in Table 2, the gonadal development of the six-year old female milkfish was found more advanced than that of five-year old one. This observation, supported by the GSI study by Kuo et al. (1975), suggests the likelihood of the oocytes reaching secondary yolk stage or tertiary yolk stage if they are reared for several more months in captivity. By that time, hormone treatment can be given based on the information obtained in an induced breeding of grey mullet done in this Laboratory. Kuo et al. (1975) mentioned that receptive females should have the oocytes of the tertiary yolk stage between 0.7 and 0.8 mm in diameter. For inducing breeding of milkfish by hormone treatment, six milkfish of both sexes aged six years are being treated with hormone*. The feasibility of raising tank-reared spawners will be fully ascertained if these treated females reach fully ripe condition. The success will be a big breakthrough in artificial propagation of milkfish.

Liao (1971) observed that in coastal area of southern Taiwan, wild mature spawners of milkfish are captured during the period from April to June. However, there is no record so far of capturing them in the fall months. According to the occurrence of wild milkfish fry, there should be two spawning seasons, one in spring and the other in fall. Tampi (1957) reported that each milkfish normally spawns only once a year. Therefore, it might be conjectured that oocytes at oil droplet (yolk vesicle) stage in April of the six-year old tank-reared milkfish would reach secondary yolk stage or even tertiary yolk stage in fall of the same year. It is important to investigate the monthly gonadal development of milkfish reared in captivity.

*No significant response was observed after the fifth injection on 24 April 1976. Fish are healthy and are continued to be treated.

Tampi (1957) pointed out that the age of wild female milkfish at first maturity is roughly between four and five years. However, our unpublished data on the gonadal development of tank-reared milkfish at different age, showed that the ovarian oocytes of fish under five years developed to peri-nucleolus stage only and oocytes at oil droplet (yolk vesicle) stage started to appear at the age of five years. As noted previously by Liao (1971), the minimum age of wild female milkfish at first maturity was five years in Taiwan. Age of tank-reared milkfish can be correctly ascertained based on the date of stocking. However, age of captured fish was determined from scale rings although some parts are not clear enough for reading. Therefore, further study in this respect is needed.

Tampi (1957) observed that the size of captured female fish at its sexual maturity is about 110 cm total length and body weight about 11.0 kg, which is larger than that observed in wild spawner in Taiwan (Table 3). Liao (1971) attributed the reason that probably different spawning stocks are there in Mandapam area and Taiwan area. As listed in Tables 2 and 3, the measurements of tank-reared spawners were smaller than those of wild ones. These data correspond more to that recorded by Kuo *et al.* (1975). As listed in Table 4, the fish being reared in a big pond for two to three years measured almost the same in total length and body weight as the fish of five years of the present study, but gonadal development of the former was far behind that of the latter. It is concluded that age of fish is the more important factor to sexual maturity than its size.

Table 3. Gonad weight and GSI of four adult milkfish captured from the coast of Southern Taiwan.

<u>Fish No.</u>	<u>Date</u>	<u>Sex</u>	<u>Age (yr)</u>	<u>B.W. (kg)</u>	<u>T.L. (cm)</u>	<u>G.W. (g)</u>	<u>GSI</u>	<u>Remarks</u>
1	5-6-70	♀	6	8.20	104.0	950.0	11.59	Almost mature with eggs 0.89 x 0.98 / / 1.32 x 1.50 mm in diameter, captured from Heng-Chun.
2	22-4-71	♀	5	6.03	87.0	201.2	3.34	Immature with eggs 0.54 x 0.59 mm in average diameter, captured from Heng-Chun.
3	9-6-72	♂	6	7.69	98.7	482.1	6.27	Mature, captured from Nan-Jen Wan.
4	10-4-74	♂	6	9.50	100.0	416.5	4.38	Mature, captured from Nan-Jen Wan.

Table 4. Gonad weight and GSI of four milkfish cultured in pond near Tungkang.

<u>Fish No.</u>	<u>Date</u>	<u>Sex</u>	<u>Age (yr)</u>	<u>B.W. (kg)</u>	<u>T.L. (cm)</u>	<u>G.W. (g)</u>	<u>GSI</u>
1	3-7-75	♀	2	2.62	68.2	1.23	0.05
2	2-8-75	♀	3	2.18	-	1.97	0.09
3	2-9-75	♂	3	2.91	71.34	0.47	0.02
4	2-9-75	♂	2	2.42	-	0.44	0.02

Both measurement of body length, body weight and gonad weight and gonadal development of tank-reared spawners (Table 2) were less than that of wild milkfish of same age (Table 3). Development of the ovarian oocytes in the same tank-reared spawner were rather diverse just as in captive grey mullet (Pien, 1975) and this may vary the egg production. It might be caused by either ecological or physiological factors. Further study on the reproductive ecology of wild milkfish is also needed in order to improve the rearing techniques to accelerate their growth and gonadal development.

As shown in Table 2, both the male milkfish aged five years and six years had mature sperms, but little milt was obtained on stripping the testes of the latter only. However, their GSI was much lower than that of wild spawner (Table 3) and there was very limited amount of milt probably even not enough for the eggs of one female. Consequently, hormone treatment is needed to induce proper development as well as maturation of gonad that has reached the developmental stage as the experimental fish in the present study.

In recapitulation, male tank-reared milkfish is mature at the age above five years and may be stripped for milt through the help of hormone treatment. As for female milkfish of six years age, they are not ripe enough to be stripped but the gonadal development is encouraging. The smaller size of tank-reared spawners than the wild ones may cause some effect on their fecundity but is advantageous to handle and operate in artificial propagation. In addition, the domestication of tank-reared spawners is also an advantage in the process of hormone treatment. On the contrary, wild adult milkfish are highly excitable and extremely difficult to capture and handle without causing stress and shock which usually result in injury and death. It is concluded that the idea to start raising milkfish in captivity six years ago is of significance. There still exist many

unsolved problems such as improving the composition of compound feed lest the milkfish should be too fatty and finding out a dependable method to distinguish the male from the female adult milkfish. It is hoped to establish and complete the process of artificial propagation of the economically important milkfish through continuous effort in the near future.

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A SIMPLE METHOD FOR MONITORING
THE SPAWNING ACTIVITY OF FISH IN NET ENCLOSURES

by

R.C. May, G.S. Akiyama, and M.T. Santerre*

Abstract

A simple method is described for monitoring the spawning activity of fish held in suspended net enclosures. The method, which involves an airlift pump, has been used successfully with the threadfin, Polydactylus sexfilis, and has revealed important aspects of the daily, monthly and yearly spawning rhythms of this species. It is suggested that this approach may be useful in studies of Chanos chanos.

Introduction

Information on the spawning characteristics of a fish species, particularly the natural spawning rhythm, is extremely useful when methods are being developed for controlled reproduction. Such information, however, has traditionally been difficult to obtain. Catches of reproductively active fish in nature are usually dependent upon chance particularly for species which spawn at sea. Rarely has it been possible to gather enough samples in this way to reveal the small-scale rhythmicity of spawning, such as the exact time of day or time of month when spawning occurs. Confinement of fish in tanks may be useful for small, hardy species, but in the case of fish such as Chanos chanos which are very large when sexually mature, confinement in tanks may be extremely stressful, or may require such large tanks as not to be feasible in many instances. Furthermore, certain natural stimuli such as tides are missing in tanks, and restricted or highly directional water flow and the presence of walls may inhibit normal activities.

A large suspended net enclosure is a more natural situation than a tank. Although the latter permits observation of the fish, a net maintains the fish in a natural body of water and allows the free exchange of ambient water. We have found that the threadfin, Polydactylus sexfilis (Cuvier and Valenciennes), exhibits its natural spawning behavior when confined in net enclosures. This led to the discovery of a well-defined lunar spawning rhythm in this species and has enabled us to define with some precision the time of day when

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spawning occurs (May, 1976; May et al, MS). To gather this information, we developed a simple airlift egg collector which may be equally applicable to other species such as *Chanos chanos* which produce pelagic eggs. Without such an apparatus, it may be impossible to tell whether fish kept in nets are spawning, unless frequent biopsies entailing much handling and a good deal of stress are made. While the system described here is certainly not an answer to the problem of controlled spawning, it is a simple means by which the reproductive activity of fish can be monitored in a semi-natural situation. It may also reveal aspects of natural spawning such as lunar rhythmicity, which may be extremely useful in the development of more controlled spawning procedures.

Design and Operating Characteristics

The egg collector consists of two major components: a collecting basket which retains the eggs, and an airlift pump which creates a continuous flow of water from the net enclosure into the basket (Fig. 1). The collecting basket is a commercially available, circular plastic pan with the bottom cut off and replaced with Nitex nylon mesh. Obviously, the mesh size of the Nitex should be smaller than the diameter of the eggs to be collected, but not so small that clogging becomes a problem. In the case of *P. sexfilis*, which has eggs of 800-825 μ m diameter, we use a 500 μ m mesh in the collector. The collector is checked once each day by rinsing the contents into a bucket and examining them closely with a microscope at the laboratory. The mesh is cleaned with a brush after each collection to minimize or eliminate clogging. A new collecting basket is installed every few weeks and algae which has accumulated in the meshes of the old basket is removed by soaking in a hypochlorite bath and cleaning with a high-pressure stream of water.

The airlift pump -- which, along with the collecting basket, is buoyed up by a styrofoam float surrounding the latter -- consists of a PVC pipe of 4 cm diameter, fitted with two PVC elbows at the top so that it feeds water directly into the basket. We generally use an intake pipe which is 90 cm long (about one third the depth of the net), but the length can be varied in accordance with the depth of water at which sampling is desired (this may vary with the fish species and its spawning behavior). Air is supplied by either a compressor or a blower and is introduced through an airstone near the bottom of the intake pipe. We ordinarily use an air source, a compressor connected to an air storage tank which produces a certain cycling in the rate of airflow and hence the waterflow. The latter varies, in our instance, between 10 and 20 liters/minute with a mean of approximately 15 liters/minute.

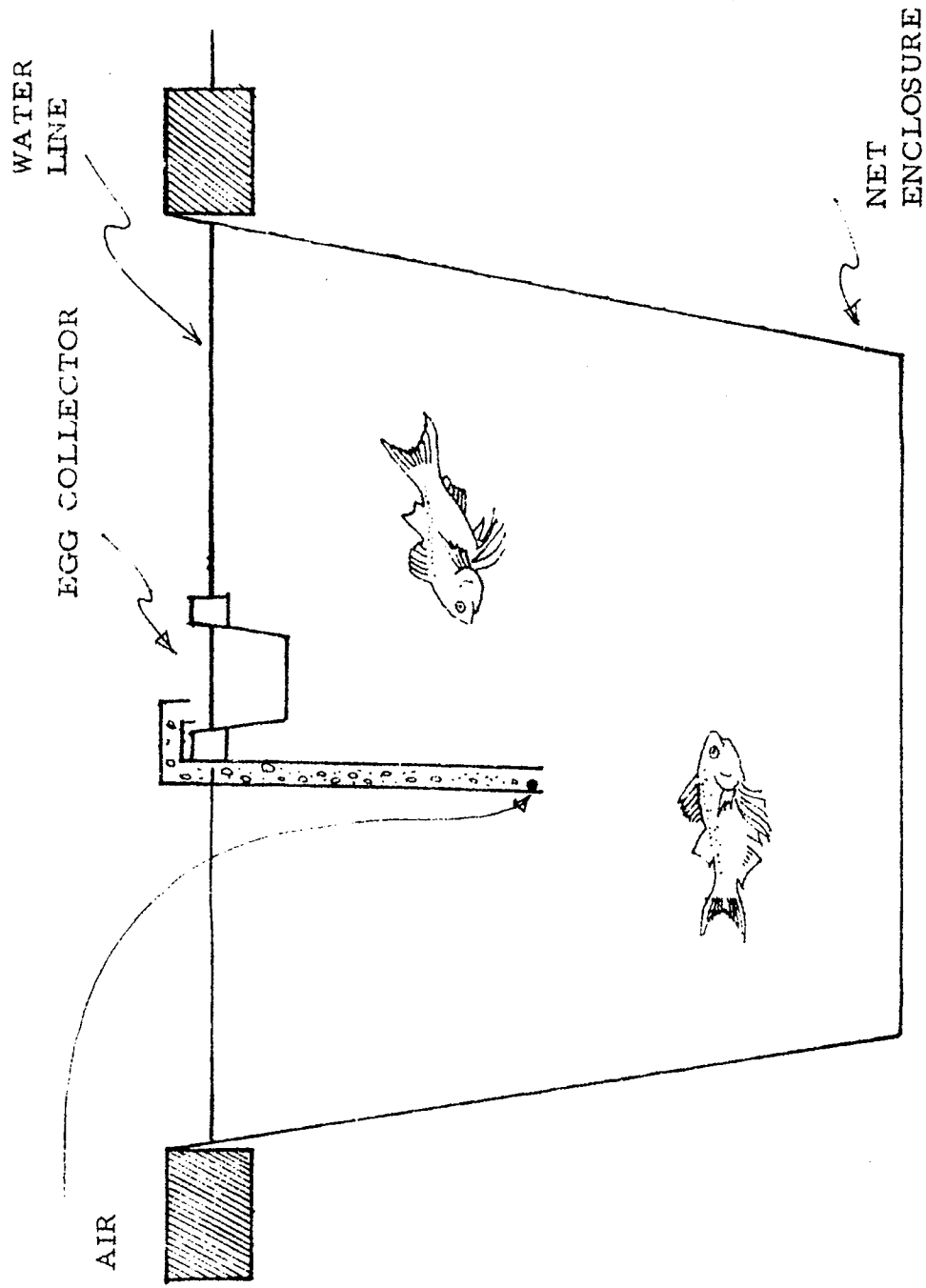


Fig. 1

After having established the time of month when P. sexfilis spawns (May, 1976; May, Akiyama, and Santerre, MS) we looked for a method of determining the exact time of day when spawning took place. For this purpose, we installed a hose in the net enclosure close to the intake of the airlift pump and connected it to a centrifugal pump on board a barge anchored next to the enclosure. In this way, we could pump water from the enclosure into a small collecting basket on board the barge where our activities, shielded from the enclosure by the super-structure of the barge, did not disturb the fish. Continuous visual monitoring of the basket enabled us to pinpoint almost exactly the time of spawning since eggs in the basket were readily apparent to the observer. By employing two observers with synchronized watches, one monitoring the collector and the other observing the fish in the net, it was possible to correlate the production of eggs with certain behavior patterns which were therefore taken to be the spawning behavior of the species (May, Akiyama, and Santerre, in preparation). It should be stressed that, whereas the airlift pump may damage eggs slightly, it nonetheless allows the majority to survive (May, Akiyama, and Santerre, MS). The centrifugal pump kills all eggs and hence is useful only in establishing the time of spawning.

Since the enclosure for the fish is made of netting, only a small sample of spawned eggs is obtained by the airlift collector, the vast majority being lost through the meshes by diffusion and turbulence. The average number of eggs collected by the airlift on peak spawning days was about 20,000 and estimates of the number of eggs released by single P. sexfilis females range from 70,000 to 500,000 (Kanayama, MS; May and Akiyama, unpublished data). Hence, if only one female spawns, the collection efficiency would be between 4% and 29%; if more females spawn on a given day, these figures would be even lower. Based on behavioral observations, we believe that on some spawning days only single females release eggs in our situation but that on other days two or more females may release eggs.

The pelagic eggs of extraneous fish species are occasionally caught by the collector. Usually these are present only in very small numbers, while eggs produced by the fish in the net are obtained by the collector in much greater abundance. Eggs of the species under study can also be distinguished from extraneous eggs by diameter and other morphological characteristics which in many cases are recorded in the literature.

Conclusions

The airlift egg collector here described has proved useful in initial work on controlled breeding in Polydactylus sexfilis. It has revealed the monthly spawning rhythm of this species, and a modification involving a centrifugal pump has given precise data on the time of day when spawning occurs. There is every indication that the pattern of spawning displayed by fish maintained in the net enclosure is the same as that of fish living in nature (May, Akiyama, and Santerre, MS). In addition, the airlift collector has provided eggs which were subsequently hatched and used in studies of larval rearing methods. We suggest that a similar approach may prove valuable in the early stages of investigating controlled breeding in other fishes such as Chanos chanos which produce pelagic eggs.

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PRELIMINARY CAPTURE, HUSBANDRY AND INDUCED
BREEDING RESULTS WITH THE MILKFISH,
Chanos chanos (FORSKAL)

by

Colin E. Nash and Ching-Ming Kuo*

Abstract

The program objective is to breed the milkfish, Chanos chanos (Forsk.) in captivity and to raise the fry from the egg, undertaking any research and development which may be necessary to attain that goal.

In this, the first year of the project, eight very mature fish were captured and deemed ready for final spawning inducement. One fish (with eggs 0.818 mm in diameter) hydrated and was partially ovulated by two injections of 25 mg salmon gonadotrophin SG-G100. The eggs were not fertilized. The injection procedure or excessive handling and sampling of three other fish resulted in early atresia (reabsorption) of the oocytes. Two fish died from the sampling and handling procedures, and two proved to be too immature with eggs below 0.6 mm in diameter.

Preliminary results indicate that oocytes of 0.8 mm and above are at a critical stage (or beyond) at which immediate hypophysation is needed. Injection cannot be delayed. Excessive handling or stress at this stage causes rapid atresia of the oocytes. It appears that oocytes of about 0.7 mm are more suitable for reacting positively to injections. The size of an ovulated egg is about 1.2 mm in diameter.

The level and dose rate of SG-G100 used for mullet (20 µg/g body weight) appears too high for the milkfish. A dose of between 12 and 15 µg/g body weight is suggested at present.

A resident population of adults of varying ages, numbering over 50 fish, has been assembled. Some were brought from the island of Hawaii to Oahu by land and sea involving 18 hours of travel. All survived the journey. Consequently a small operating satellite field center has been established on Hawaii.

A total of 179 dead adult fish have been used for future compilation of age, weight/length, GSI, scale and otolith data. All of the samples

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are stored. Determination of the CSI for Hawaiian fish indicated a rapid maturation of oocytes and spermatids in June, with a peak spawning period in July and early August.

Adult fish have been placed under photoperiod and temperature-regulated conditions to promote maturation out of season.

General husbandry methods have been developed for adults in captivity, and a diet formulated which appears to be acceptable.

The work on health care is proving to be very informative. Safe handling systems have been developed using ice, and hypotheses made for the results of this treatment and general conditions of stress. Autopsies on dead fish have revealed growths and evidence of heart attack and gastritis. Techniques using commercially available human clinical test kits are proving useful indicators of stress; for example, the presence of hemoglobin and ketones in the mucus increases with stress.

Eye lens protein analyses are being undertaken to determine any different racial origins of milkfish.

Brookstock Collection and Husbandry

Wild Fish

Although the milkfish, Chanos chanos, is an important component and subsistence product of the brackishwater coastal pond culture systems of the Philippines, Indonesia and Taiwan, the indigenous fish to Hawaii generates little commercial interest. It is, however, abundant and is caught opportunistically with other species by the local fishermen and goes for consumption predominantly by the Hawaiians and Filipinos.

Commercial fishing equipment and methods are therefore not conducive to the capture and safe handling of potential brookstock fish. As a result new techniques and methods have had to be developed to give the fish the greatest chance for survival following capture.

A survey of the local fishery, fishpond operators, organizations and private citizens who capture the milkfish was made to determine the best locations of adults and juveniles during the seasons, and the collecting gear and methods used. Figure 1 illustrates the prime locations for adults around Cahu.

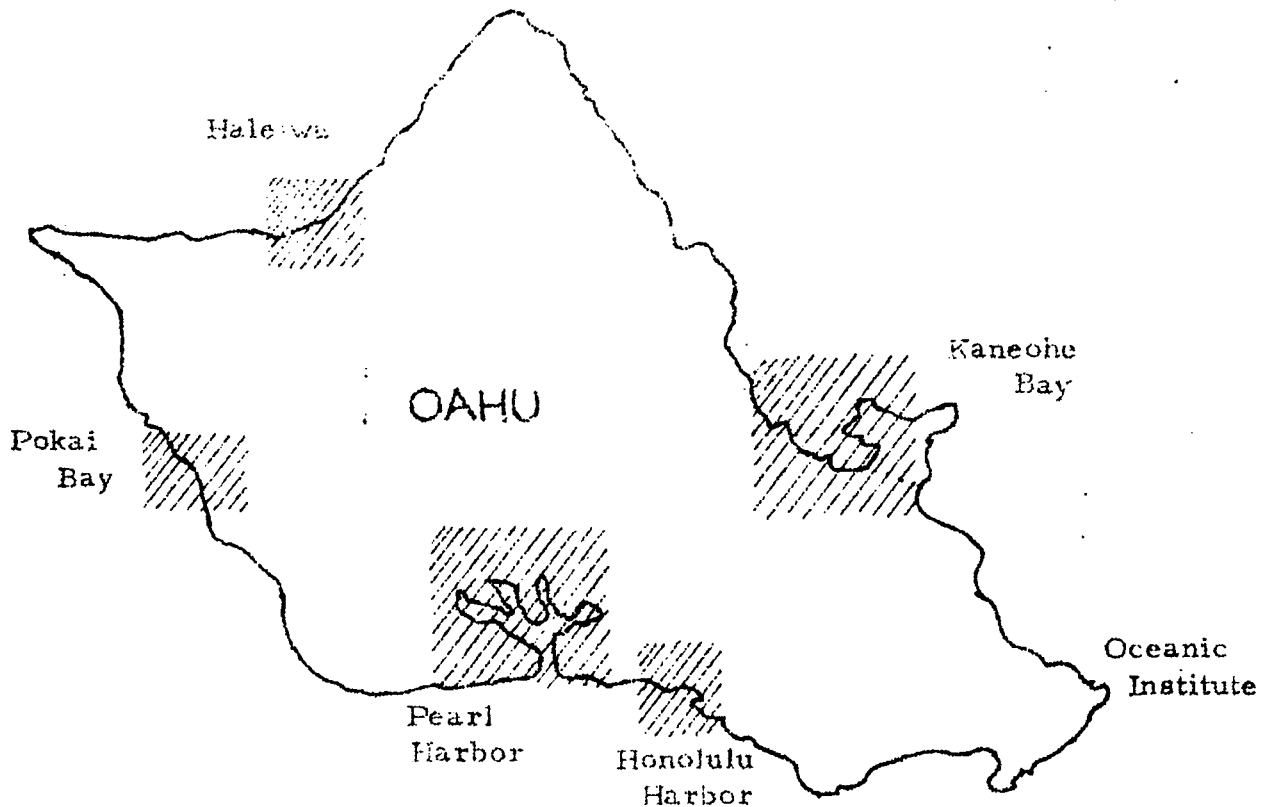


Fig. 1. Adult milkfish fishing locations in Oahu

Sources of information have included the National Marine Fisheries Service (Honolulu); the State of Hawaii Department of Land and Natural Resources, Division of Fish and Game; past and present commercial fishermen; sports-fishermen; pond owners and operators; and individual fishermen who still use the traditional Hawaiian throw-nets. Outside the State, contact has been made with individuals in the Philippines, Fiji and Tonga for information on their native techniques, fish behavior and life history. Data received includes migration of adults at spawning, location and arrival of juvenile schools, food preferences and requirements, and natural behavior. Fishing methods, gear and anecdotes have all been described.

A review of the survey indicates several significant factors which comply with our own observations and experiences. Primarily there was the universal comment that all the milkfish were not susceptible to handling, and that stress caused rapid death particularly if the fish had been captured with gill nets. Furthermore, the large size and strength of the fish prevented convenient handling and particular containers for transportation were necessary.

Notwithstanding these comments, the proximity of the natural spawning season in Hawaii (June through August) compelled an immediate intensive fishing effort to capture adults with existing boats and gear, but using the experience gained from many years of capturing the grey mullet. Although large adult milkfish are reported to frequent the offshore waters of Hawaii, the depths are beyond the design of the fishing gear available, and therefore a concentrated effort was made within the more sheltered waters of Pearl Harbor and Kaneohe Bay.

At present over 50 live adult milkfish have been collected from the sea and are held as potential broodstock. The largest individuals weigh between 10 and 12 kg, and measure 1 -1.2 m in length. There were, in addition, a further 20 live adults which were lost one night due to storm damage to the tank's life support systems. The Institute has also collected about 125 large juveniles (1-2 kg) as a future broodstock being domesticated.

Successful capture of the large adults has been due to the careful procedures which have been practised, modifying the methods used for mullet capture. The procedure utilizes two small 6 m boats and between 3 and 4 men. The net is a 200 x 2 m deep gill net of 6.6 cm mesh. Success with this monofilament net is related to water turbidity. In turbid waters, for example in Pearl Harbor, the net is invisible and the captured fish do not struggle much. In clear waters the net is readily visible and avoided by the fish. If driven into the net the fish struggle hard, often break the net and either escape or damage themselves beyond recall. However, sometimes good fish are taken if the net is brought in quickly.

The fish are transferred by dip-net or pail to a large circular 750 liter holding tank on one boat. No more than five adults can be handled at once and the boat is driven quickly to shore. A flow-through water system pumps water into the holding tank and exhausts over the side. On shore the fish are transferred to a 1,200 liter circular tank with a firm cover, located on the back of a truck.

The strongly aerated water in the tank is reduced in salinity and temperature immediately to about 12 ‰ and 20 °C, respectively by the addition of a large ice block. This treatment, together with the darkened tank, helps to counter, we believe, the physiological symptoms of stress. These, we hypothesize, are contraction of the blood vessels and reduction of oxygen to the brain.

A final transfer to a shore-based tank of 16,000 liters is made, and newly captured fish are retained there for two weeks before removal to the permanent facilities. Treatment for injury is made at this time. A water-soluble antibiotic is used for treatment at a rate of 200 g/l until lesions or abrasions are healed. Reduced infection from bacteria can be accomplished by keeping the fish in lower salinity water.

This effective but laborious technique has enabled the staff and contracted fishermen to capture in good condition fish up to 12 kg in weight. An estimate of the current success rate is about 50% of all fish captured. Most mortalities appear to be the direct result of physiological stress and not injury. All fish which do not survive capture or die later in captivity are autopsied, measured and weighed. Their gonads are preserved for histological examination and scales removed for growth and age studies. Preliminary analysis of the available data suggests that milkfish will be ready for spawning at about 4 years of age, and that natural growth in open waters is about 15- 20 cm per annum.

Pond Fish

Among the many constructed and natural ponds which occur along the coastlines of the Hawaiian Islands, a small cluster of natural anchialine ponds was made available to us through a large development corporation. The ponds cover more than 5 acres in area. Biologically and geologically they are unique, occurring exclusively in recent lava flows and harboring both marine and brackishwater biota. The salinity varies but can drop to about 7 ‰ because of subsurface springs.

The site of Lahuipuaa on the Kona coast contains five irregular ponds, two of which are connected to the ocean of sluice gates. The others are not connected to the sea but at times join each other. Historically these particular ponds have been used for holding and raising fish, principally mullet, milkfish, moi and other euryhaline species.

A first survey of the ponds was made in December 1974 to determine the abundance and distribution of fish in these ponds. The results showed a substantial stock of adult milkfish. Following discussions with the pond owners the stocks were safeguarded until a second visit was made prior to the milkfish breeding season in summer.

In June 1975 a census of the main Hopeaia Pond was made. Forty large milkfish were caught with seine nets and handled. A total of 34 large fish were marked for reference with Floy tags, anaesthetized with quinaldine sulfate, weighed and measured. A sample of gonad tissue was removed through the oviduct with a polyethylene catheter using the standard sampling procedure developed for grey mullet. An additional three fish were killed and blood samples, eye lenses and other tissues sampled for future examination.

All fish were returned to the ponds, except 24 which were relocated in a small pond for subsequent transportation to the Institute.

Hopeaia Pond is an irregular 0.5 ha pond not connected to the ocean. The maximum depth is about 1.5 m but many parts are shallow. The water is clear and slightly brackish (7 ‰) and seawater penetrates through the ground into the pond at high tide. The bottom of the pond, which is mud, is thickly carpeted with the vascular plant, Ruppia maritima, which appears to be the principal food for both the mullet and milkfish which have been stocked in the pond in the past.

Although milkfish have been recorded in Hawaii and elsewhere as large as 30 kg and almost 2 m in length, little data is available as to the size and age at first maturity. Many of the adult fish in the Hopeaia Pond, while only 3-5 kg in weight and less than 65 cm in length, were in fact maturing. Many males were ripe with active spermatozoa, and the females in general showed advanced stage II and III oocytes-- that is the yolk vesicle and yolk globule stages. While spawning is not anticipated in such low salinity waters, the ability of fish to mature under captive and pond conditions and at a size manageable in the laboratory is an important point for all future breeding work.

A third trip was made to the ponds in July to construct a small laboratory within an existing building on site. A small 1.5 kw generator was installed to run lights, water pumps and air compressors. A plastic swimming pool was installed and filled continuously with oceanic water, discharging into the nearby pond.

Twenty additional large fish were collected, marked with tags as before and weighed and measured. All fish not showing advanced signs of maturity were relocated in one pond for next year. Six mature fish were relocated in the swimming pool. The most advanced female was subsequently injected with SC-C100 to induce the final stages of spawning. Hydrating eggs were later released by this fish slowly over a long period, and these were not fertilized. Slow release following hypophysation in the mullet is an indication of premature spawning.

A fourth trip was made in August as part of a planned relocation of the larger tagged fish back to the Institute. The visit was timed to coincide with the shakedown cruise of the National Marine Fisheries Service vessel, the Townsend Cromwell, through the cooperation of the local laboratory Director. The fish were transported ten miles by road to the pier at Kawaihae in the 1,200 liter tanks. They were then transferred to the 3,800 liter fish transport tanks designed and used by the NMFS for tuna, and were connected to the ship's pumps for large-volume water exchange. The ship then sailed back immediately to Kewalo Basin in Honolulu and the fish were subsequently transported to the Institute by truck. The total time for transportation was 13 hours and not one fish was lost.

In addition to the volume of experience that has been gained in the capture and transportation of the larger milkfish, the availability of these resources has made it possible to undertake a great deal of laboratory work without jeopardizing future breeding adults. One invaluable result has been the better understanding of the female reproductive system. Autopsy of mature gravid females has shown that the eggs are released through a funnel rather than through the oviduct. This fact has demanded some change to the established sampling procedures developed for grey mullet, and a satisfactory technique has not yet been developed. The use of a cystoscope was contemplated to avoid actual sampling, but the dimensions of the available instruments were too big. This anatomical feature answers the questions of previous sampling difficulties experienced by ourselves and other workers.

In addition to the Lahuipuaa Ponds, further contact and informational exchanges were made with fishpond operators throughout the State. Some well advanced fish were obtained from the owner of the Molii Pond in Kaneohe Bay, about ten miles from the Institute. These fish were also safely transported back to the Institute tanks by truck.

A great deal of assistance was provided by private individuals in the State, particularly from the Hawaiians who still take milkfish by the traditional throw-nets. The lack of a mobile force to take advantage of their captured animals made this source of adults impractical to follow.

Apart from those fish taken by the Institute staff, all other fish, both alive and subsequently dead, were paid for at an advantageous rate to the collector. However, certain restrictions were made as to what was acceptable.

Juvenile Fish

Post-larval and juvenile milkfish were collected at various locations around Oahu for data on growth, behavior, nutritional requirements and disease, and also for trade with the pond operators in exchange for their larger fish. These juveniles should then become the resources several years from now.

The post-larvae, 12-16 mm in length, are almost transparent with pigment confined to the head and gut regions. They are usually found in the upper reaches of streams in warm shallow waters feeding on diatoms, organic sediments and algae. Pigmentation of the body is complete after about three weeks of inshore life and the juveniles between 25 - 30 mm in length move downstream into deeper waters. During this period the fish can be captured easily with fine nets and transported with little loss of life. This is a distinct contrast to the handling of the older fish.

Juvenile recruitment has always been an indicator of the numbers and reproductive success of the adults at sea. Observations by Institute staff through the years indicate that the fish have only one spawning period in the year (June to August), and that the local population, while not large, is probably sufficient to produce progeny annually to maintain this population.

Evidence indicates that actual spawning is cyclic within the spawning period and related to lunar periodicity. This is most apparent for observations of pond fish where there is access to ocean waters. Throughout the year, for example, the large fish in the Molii Pond in Kaneohe Bay are not seen or caught near the pond gates. However, coincidental with two consecutive high tides (and thus new moons), the largest fish moved through to the gates in an attempt to go to sea, presumably to spawn. Although these particular fish did not get out to sea, the first post-larvae were recorded in the area about four weeks later.

Light and temperature data of the environment where spawning is believed to occur are being collected to establish the controlled conditions for breeding fish on a year-round basis.

A final collection of juveniles was made in November. Many of these juveniles were released into ponds for future use in the project. A great deal of assistance has been obtained from the owners of the ponds around Oahu and other islands, for both fish and the behavioral observations.

Husbandry of Captive Stock

A vital importance is the maintenance of a healthy stock in captivity if the fish are expected to breed. Little is known of the qualitative and quantitative requirements of the milkfish diet.

In nature, the milkfish is accepted to be a herbivore feeding on diatoms, organic material and various algae. In certain farm ponds in Southeast Asia and around Hawaii, the milkfish are known to feed directly on raw sewage. Large schools do congregate around the sewage outfalls. These areas are well known fishing locales but the fish are caught on rod and line. The locations are unsuitable for netting gear.

The more practical feeds used for the herbivorous captive fish are modifications of the pelletized feeds used for trout and carfish. Further, less proteinaceous diets have been developed by the Institute for such fish.

All broodstock fish are usually retained in the larger swimming pool tanks or open dirt ponds lined with a butyl rubber liner. The pools and tanks are undisturbed to allow a natural growth of vegetations for the fish to browse on. This diet is supplemented with a prepared feed made in the laboratory. One such diet supplement being developed consists of the following ingredients.

Wheat middlings	55%
Cottonseed meal	14%
Soy bean meal	14%
Tuna fish meal	14%
Propylene glycol	1.4%
Visorbin (Vitamin E Complex)	1.4%
Vitamin pre-mix	0.2%

The ingredients are milled to 0.5 mm size and stored dry. When needed, the feed is mixed to a dough with water and fed at a rate of about 0.75 kg per day per 25 large fish. The approximate cost of the feed is 35 cents per kg.

Environmental Regulation of Captive Stock

A section of the Environmental Control Laboratory has been isolated for a number of adult fish to be subjected to controlled temperature and photoperiod regulation to encourage egg maturation out of season. The controls for the period have been set at 26°C and 13L/6D photoperiod. A total of six fish are being maintained under these conditions and fed daily with the prepared diet.

Induced Breeding

Data

During the year a month-by-month fishing effort was maintained for the capture of brookstock and for the accumulation of statistical data on the breeding cycle of milkfish in Hawaiian waters. The summary data is detailed in Table 1.

Table 1. Monthly Gonado-Somatic Index (GSI)

Month	Males				Females				
	GSI	sd	mean	sd	n	GSI	sd	mean	sd
Jan	0.193	0.049	0.005	0.3	0.25	0.119	0.205	3	
Feb	0.20	0	0	1	0.115	0.085	0.120	2	
Mar	0.03	0	0	1	0.435	0.076	0.152	4	
Apr	0.046	0.009	0.02	5	0.306	0.060	0.158	7	
May	0.120	0.04	0.057	3	0.385	0.045	0.064	2	
June	0.038	0.005	0.012	15	0.171	0.020	0.079	15	
July	3.225	0.095	0.134	2	3.13	0	0	1	
Aug*				0	1.345	0.299	0.597	4	
Aug**	0.043	0.009	0.028	9	0.175	0.044	0.138	10	
Sep	0.091	0.027	0.119	19	0.369	0.055	0.258	22	
Cct	0.08	0.041	0.135	11	0.250	0.021	0.098	21	
Nov	0.068	0.031	0.008	6	0.212	0.114	0.362	10	
Dec	0.004	0	0	1	0.307	0.153	0.265	3	
				(75)				(104)	

*Early

**Late

The GSI is the most practical measure of maturity development by fishery biologists and can be expressed by:

$$\text{GSI} = \frac{w \text{ (weight in g for both gonads)} \times 100}{W \text{ (body weight in g)}}$$

The GSI described in graphic form (Fig. 2) clearly indicates the natural season in Hawaiian coastal waters commencing in June and terminating toward the end of August. July and early August are unquestionably the peaks of spawning activity in Hawaii. Although two individual spawning periods have been attributed to milkfish in the waters of Indonesia (or there are two separate races), and long single spawning periods are attributed to fish of India, the Philippines and Southeast Asia, in Hawaii and other Pacific Islands, for example Fiji, there is only one clearly defined and short period of two months.

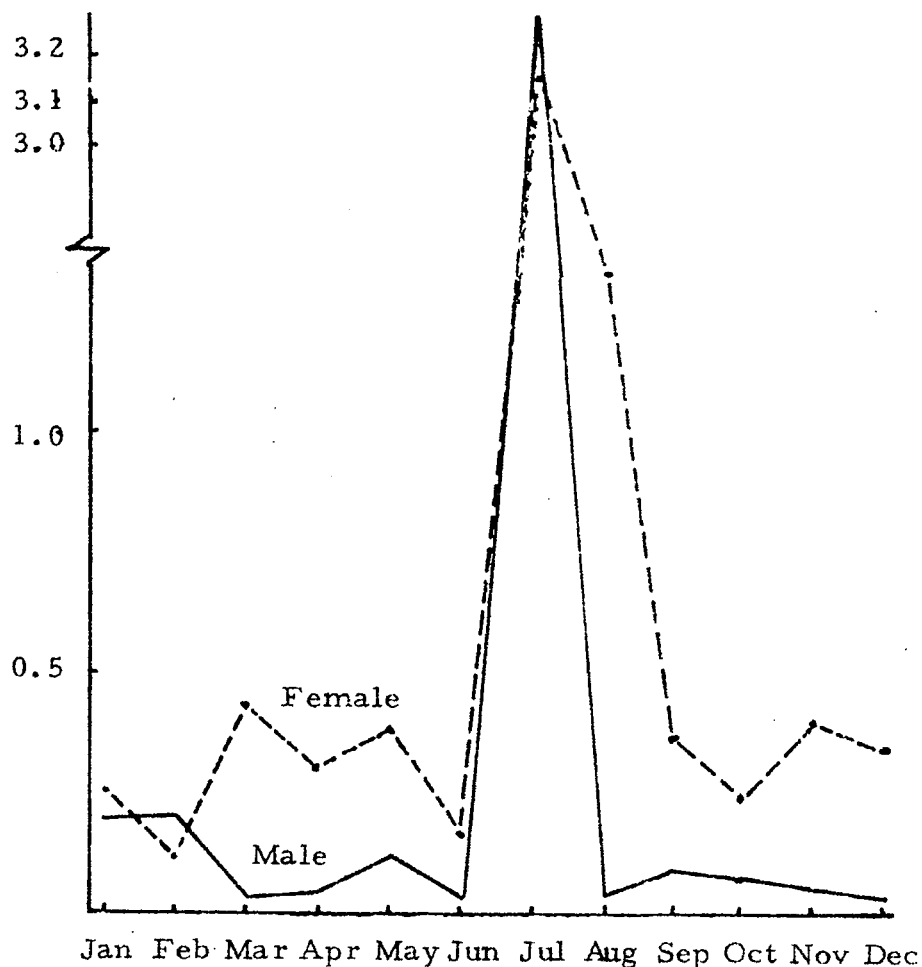


Fig. 2. Monthly gonado-somatic index of the milkfish

Observations of fish within local coastal fishponds in May and June indicate the strongest desire of the fish to escape to sea. Large fish move toward the outlets of the pond where they can be readily caught in the traps at the gates. These fish in fact proved to be an important contribution to mature broodstock.

The weight of the fish showing signs of maturation proved interesting. Historical records in the literature indicate that the smallest mature females are about 70 cm in length (about 4,500 in weight, or 10 lb). Mature female fish taken in Hawaii proved to be much smaller, even as small as 60 cm in length and weighing only 2,500 g. The scales of the fish have been taken but data on actual age has still to be produced.

Pond fish seem to mature slightly later than non-captives in the sea. This was certainly true for the grey mullet species.

Egg Sampling

The induced breeding methods developed by the Institute have always demanded two conditions. These are (1) an accurate knowledge of the exact stage of development of the eggs of fish about to be induced, and (2) the use of a standardized hormone. Sampling and histological studies have, over the years, enabled an accurate development picture to be produced for the oocytes of the grey mullet. This development sequence has then enabled gross observed parameters to be used as the indicators for the level, dose rate, and time sequence of the standard hormone to be used.

The approach for the milkfish must be identical. Consequently the gonads of the broodstock female fish are sampled on a regular basis using the traditional techniques. A fine polyethylene cannula is inserted into the oviduct and a sample of oocytes is withdrawn by oral suction.

Unfortunately, sampling milkfish does not prove to be simple. The genital anatomy of the milkfish differs from that of the grey mullet and approaches more than that of the salmon. The oocytes are released through a funnel from the gonad, enter the oviduct close to the genital pore, and are then expelled through the cloaca. As a consequence, the funnel does not provide the easy sampling passageway which is true of grey mullet, and egg sampling has proved difficult.

Egg samples can be obtained from the milkfish gonads, but are difficult to obtain. The fine cannula gets lost in the body, can rupture membranes and body organs, and many attempts may have to be made to obtain an egg sample -- sometimes resulting in death of the fish.

It was hoped that the use of a cystoscope would make sampling unnecessary, and that an internal examination would prove sufficient. However, a fine enough cystoscope is not yet available.

Eggs that were sampled readily were measured, preserved, and examined histologically using standard procedures to determine vitellogenesis and to begin an assembly of an 'atlas' of egg development.

Egg diameter distributions of some fish before injection are illustrated in Fig. 3.

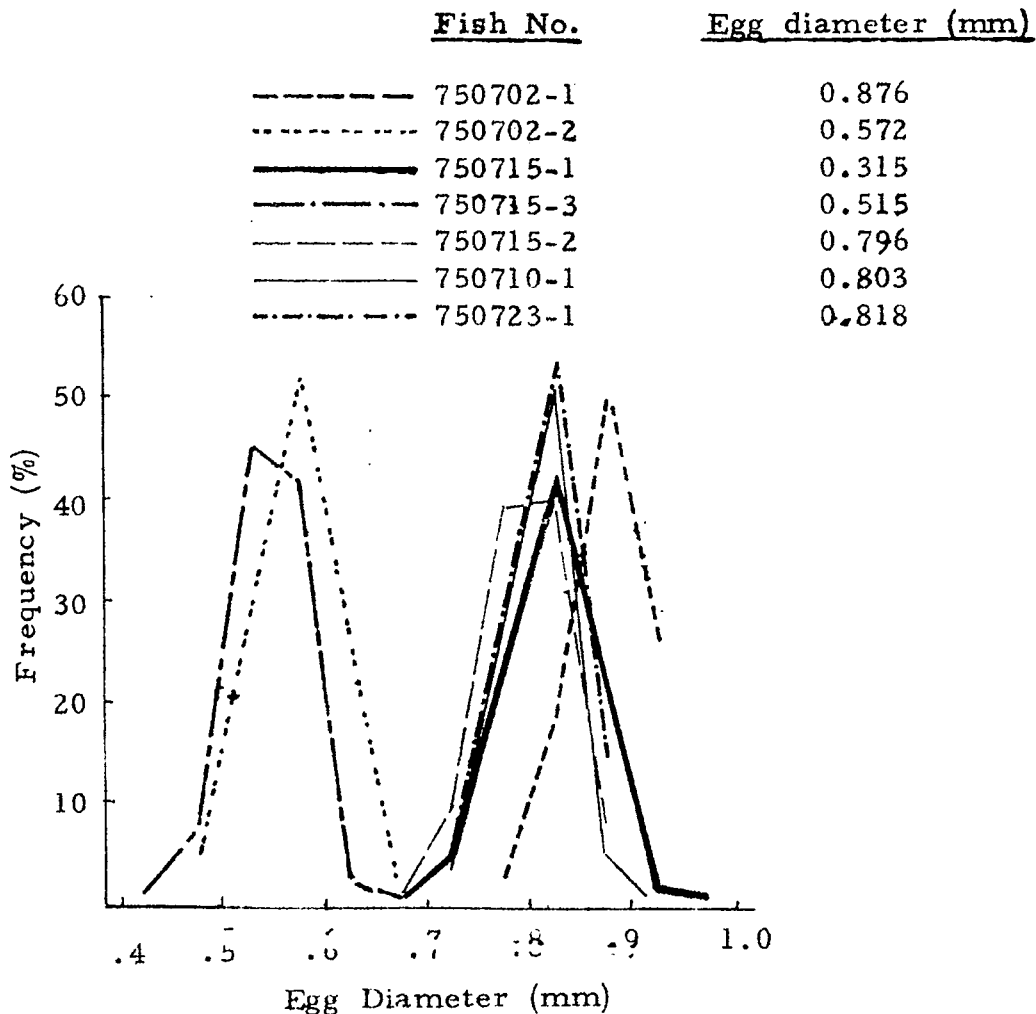


Fig. 3. Egg diameter distribution

Work on Mature Female Fish

Eight fish were deemed ready for final induced treatment in the summer. Background data of the fish is described in Table 2.

Table 2. Mature Female Fish

Fish number	Date	Egg diameter mm	Stage	Weight g
750702-1	7:8:75	0.876	IIIc	4,000
2	7:8:75	0.572	III	3,500
750715-1	7:17:75	0.815	IIIc	5,000
2	7:17:75	0.796	IIIc	4,750
3	7:17:75	0.545	IIIb	
750710-1	7:10:75	0.803	IIIc	6,750
2	7:10:75	0.545	IIIb	4,500
750723-1	7:23:75	0.812	IIIc	2,500

The stages of development used are those described by the Institute for the grey mullet. Stage IIIc is therefore the tertiary yolk globule stage, and IIIb the secondary yolk globule stage.

The standard hormone used is salmon gonadotropin SG-G100, prepared exclusively by the University Laboratories in British Columbia and designated ECR#1.

Data on the induced spawning tests is as follows in Table 3.

Table 3. Injection Dose, Sequence, and Result

Fish Number	Date	Egg diameter mm	Dose mg	Stage	Observation
750710-1	7:10	0.803	0	III	Mature
	7:14		15		
	7:15		30		
	7:16		40		
	7:17		0		
750725-1	7:17	0.815	0	III	Mature
	7:21		0		Atretic
750715-2	7:17	0.796	0	III	Mature
	7:21		0		Atretic
750723-1	7:23	0.812	25	III	Mature
	7:24		25		Hydrated
	7:25		0		Partially ovulated

Of the remaining four fish, two (750702-2 and 750710-2) proved to be too immature to be injected (egg diameters less than 0.6 mm); and two fish (750702-1 and 750715-3) died from repeated handling while trying to obtain egg samples.

The two fish detailed above (750715-1 and 750715-2) immediately went atretic, or began reabsorption of their eggs, following initial handling; and the other fish (750710-1) went atretic during the injection sequence.

The best result was with the small female (750723-1) which was induced to hydrate and ovulate with two injections of 15 mg of hormone 24 hours apart.

From these preliminary tests it seems certain that eggs of 0.8 mm and above are at the critical stage or beyond, at which immediate hypophysation is needed. Injection cannot be delayed. Excessive handling or stress at this stage causes the eggs to become atretic, or reabsorbed. It appears that eggs of about 0.77 mm are more suitable for reacting positively to injections. The size of an ovulated egg is 1.2 mm in diameter.

The first dose of injection was made at the same level and dose rate as that developed for the grey mullet, namely about 20 µg/g body weight of the recipient. The milkfish which did hydrate (750723-1), underwent a very rapid late development. This suggests that the dose rate established for mullet may be too high for milkfish, and that about 12 - 15 µg/g body weight may be optimum. This has some obvious economic advantages. The cost of injecting a mullet for spawning is about \$80 - 120 per fish. At the same rate milkfish will be well over \$500 per fish, because of their large size. The lower dose will give significant savings.

Health Care of Captive Fish

Stock Identification

The work in stock identification consisted of the collection and electrophoretic processing of eye lense nuclei from different fishes. The protein patterns that result from the processing may be used in a number of ways. One major use is to identify breeding populations of fishes. For aquaculture, the identification of breeding populations having characteristics such as stress resistance would be important. These populations would be likely to thrive better in captivity.

To date, about 100 pairs of lenses have been collected from milkfish and other different species, and processing is underway. For the purpose of visualizing the information from these patterns, a map is being planned. It will attempt to show the range, distribution, seasonal variations, migratory movements, etc., of fish population as they are reflected from the patterns. The patterns themselves will be stored in a book for easy display.

Studies on Stress

(1) Occult hemoglobin in skin mucus of fish

The results of experiments showed that when fish are left out of the water, their skin mucus produces positive reactions to a commercial test ('stix') for free hemoglobin. Other studies confirmed that the hemoglobin appeared in response to stress and not other factors, e.g. new mucus production. These studies also demonstrated the specificity of these tests to hemoglobin, and not to other substances in the mucus.

(2) Ketone bodies in skin mucus

The experiments on free hemoglobin suggested the possibility that skin mucus could also reveal specific sources of stress such as starvation. An experiment was performed in which 'stix' were again used, but this time to determine the presence of ketone bodies in the skin mucus of starving mullet (as all milkfish are being retained for breeding). These mullet did show positive reactions for ketone bodies as the starvation period lengthened. After feeding was resumed, ketone body reactions decreased.

These experiments on stress demonstrate the value of using skin mucus of fishes to quickly assess their health. They also show the case of the technique of testing and reveal the promise of 'stix' in diagnosing general and specific forms of stress.

Future work on stress may include the following: (1) working out the time relationships between the first detection of stress and subsequent appearance of pathological consequences. These consequences may include lipoplepherities in the milkfish, abnormal behavior, or mortality itself; (2) investigating what effects certain conditions of containment may have in producing stress; and (3) examining other fish species for stress- and starvation-associated free hemoglobin and ketone body production, respectively. In all cases, stress studies can not be performed using a sensitive and quantifiable measure--

the color change on a plastic strip ('stix')--without resorting to mortality and the waste of animals that it entails.

Survey of Naturally Occurring Pathology

Autopsies for naturally occurring diseases were routinely performed on fishes as their lenses were being collected. About 50 autopsies have already been performed on milkfish and other species.

Milkfish have been found to have a high incidence of a grossly and histologically identifiable gastritis. Of 60 individuals autopsied, this condition was found in 67%. With respect to location of capture, the percentage of afflicted fish ranges from 38% to 100%. Other parameters for example fish size, sex, etc., are currently being evaluated for possible relationship to the stomach condition.

The well known 'nervousness' of the milkfish may be a manifestation of the stomach irritation, or its cause. The role of this condition in the long-term health and survival of the animal is currently being evaluated. For example, does the stomach lining sometimes bleed, causing, as it does in humans, anemia or even mortality if enough blood is lost to lead to hypovolemic shock? A possible preventive is also being considered.

There is evidence that a wide variety of conditions--e.g. stress, microbial disease, trauma, malignancy, etc.-- can lead to clot formation in vivo. It has been speculated that should such clots form, from whatever cause, and lodge in blood vessels to vital organs, for example the heart, it could result in the death of fish.

To explore this possibility in milkfish, the hearts and ventral aortas from four individuals were autopsied. The ventral aorta from one of three of these fish, all of which had been caught by hook and line and stored frozen prior to autopsy, revealed several 'old', organized clots. Because of their size, number and location, they almost certainly interfered to some extent with the outflow of blood from the heart. Most dramatic, however, were the hearts from the remaining two fish in this group and a fourth fish, which died of unknown causes after thrashing about in a pond for approximately one hour. These hearts each contained a fairly fresh clot that was solidly impacted in the conus arteriosus. There was no question that this clot completely sealed the outflow tract from the heart, and that these (or any other) fish could not survive such an event.

Studies continuing on more animals are strengthening the hypothesis of in vivo clotting from a variety of conditions, as mentioned, resulting in at least morbidity (first milkfish above) or the final cause of death (remaining three milkfish above). The conditions that initiated clot development in the first and fourth milkfish of this study are not clear; but hypoxia-induced ventricular fibrillation, resulting in blood stasis in the heart, is probably the cause of the clots in the other two ocean-caught fish, and may also have been involved in forming the clot in the pond fish. In many cases, the conditions initiating clot development may not be evident, but they may be considered of academic concern because the clots once formed become the principal problem. Therefore, effort is being directed at evaluating tests from human clinical laboratory medicine to detect clotting in its earliest stages, and of ways to stop its further propagation in the live animal.

Immune Status of Milkfish

A possible quick and easy method, adapted from human clinical laboratory medicine, was tested for assaying the presence and concentration of protective antibodies in milkfish. The method, known as radial immunodiffusion (R.I.D.), consists of applying samples of extract--in this case, from milkfish lens nuclei (an easily obtained and stored source)--on a gel containing antiserum against antibodies. Positive results are produced in the form of rings of precipitate around the sites of sample application. In the milkfish, some variation in concentration of precipitate among individuals was observed, but results were all clearly positive. They suggest that R.I.D. can be applied as practicable method in milkfish culture for:

- a) Following the chemical immune response of individuals in both health and disease (possibly using blood or skin mucus for this), and
- b) Differentiating milkfish stocks with varying degrees of antibody production. Those with the highest titers may be most useful for pond culture.

Hypersensitivity Responses to Milkfish Serum

The common Hawaiian sea cucumber, Holothuria cinerascens, responds with severe contractions of its body wall musculature to contact with diluted milkfish serum. This response appears to be immunologic in nature, and desensitization is possible leading to improved health and longevity. Follow up studies are attempting to evaluate the sea cucumber/milkfish serum system for investigating immunologic responses, stress resistance, and longevity in an invertebrate generally considered ancestral to fishes and other vertebrates, including man. Application and benefits to the latter group are anticipated from this basic research.

Tag Development and Tracking

The conventional acoustic fish tag, a pinger which periodically emits a short pulse of high-frequency sound, has been in use for many years, with considerable success in lakes, rivers and estuaries. For example, pingers facilitated much of what is known about the habits of salmon in our waterways.

The simple pinger fails in the open sea, however, because it provides only one dimension of the required tracking information, direction leaving distance and depth almost impossible to estimate.

A less-known but very effective device, the acoustic transponder Fig. 4, is able to provide these missing dimensions, while at the same time making more efficient use of its batteries. Briefly, the transponder contains a sensitive acoustic receiver, and only emits its pulses upon hearing a coded signal from the tracking equipment. Range is derived from the round-trip travel time of the interrogating and responding signals. The responding signal may be coded to provide depth or other additional information.

Tracking requirements of one km for range and 300 m for depth in the open ocean, and lesser values in coastal waters, are adequate for navigation of small craft to keep a migrating milkfish in sight on the sonar screen. These requirements can be met by a commercially available high-resolution sonar set (Wesmar 200AB, Western Marine Electronics, Seattle, Washington), suitably modified, in conjunction with a transponding tag which can deliver one watt of acoustic power.

Adult milkfish dissected show large, elongated swim bladders, extending over about half their body length. The acoustical target strength of such animals is 20 to 25 db for an effective source level of 1,200 watts, with directivity index of 18 db with respect to an isotropic source. Taking the active sonar/passive target case, and allowing an echo signal-to-noise ratio of $\neq 6$ db, the maximum allowable transmission loss is 87 db for tropical oceanic conditions. This much loss will occur in somewhat over 100 yards (90.77m). For a school of, say, 100 fish, the range about doubles. Thus an untagged individual (or school) can be reliably detected only if it is less than 100 yards (90.77 m) or 200 yards (181.54 m) away.

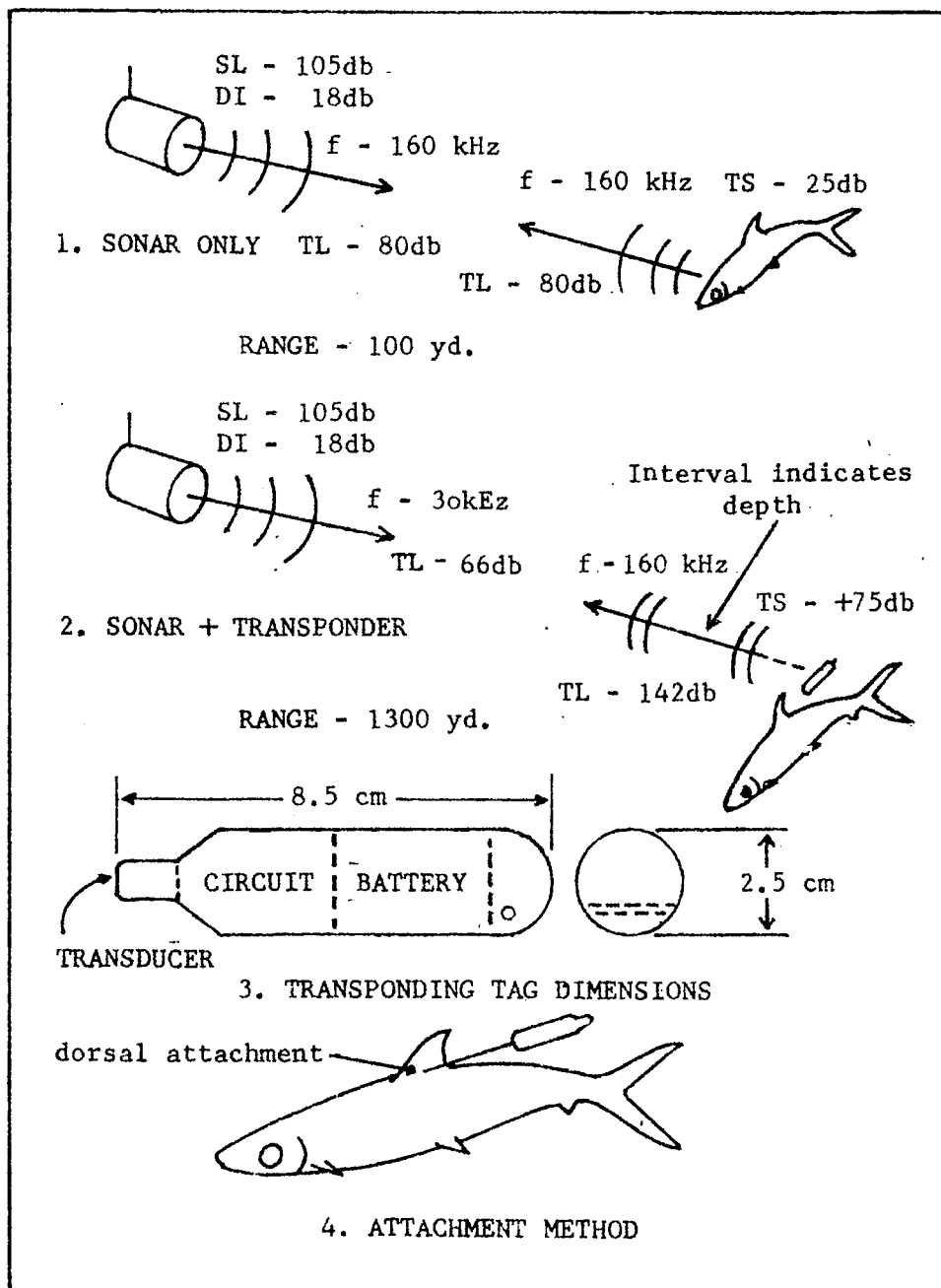


Fig. 4 Tagging and tracking information

Introducing a 1-watt transponder has the effect of increasing the apparent target strength of a tagged fish to /75 db, an overall increase of 100 db. Allowing the same echo signal-to-noise ratio as before, the maximum detection range is now 1,300 yards (1,180 m), which matches well with the 1 km display range of the Wesmar sonar.

The Wesmar sonar has been installed and modifications designed and tested to provide an interrogation signal. A prototype transponder has been designed and dummy tags fitted to live adults.

No attempt has been made yet to miniaturize the transponder circuit, as this will require tooling that is circuit-dependent. However, no problems in this regard are anticipated and the transponder's battery will be its largest component.

Acknowledgements

In addition to the two authors of this report to the Conference, other senior scientific staff involved in the program are Guy N. Rothwell, Jr. (electronic engineering) and Albert C. Smith, Ph.D., M.D. (pathology). The team is backed by the scientific and technical aquaculture staff of the Oceanic Institute in Hawaii.

MILKFISH CULTIVATION IN SARAWAK

by

Ong Kee Bian

Abstract

The paper reports the results of experimental rearing of milkfish fry in both fresh and brackishwater ponds in Sarawak. Data on the growth and survival of these imported fry are also given.

Introduction

Adult milkfish has been reported to occur in Kuala Lawas in the Fifth Division (Ong, 1968). The cultivation of this fish was first experimented on when a batch of 5000 milkfish fry were imported from Thailand in 1971. Of these, 4,000 fry were stocked in both the brackish and freshwater ponds in the Coastal Aquaculture Station, Semariang Batu, while the rest were stocked in the freshwater ponds of Semongok. At Semariang Batu, it was observed that the fish stocked in brackishwater ponds grew much faster than those stocked in the freshwater ponds. The yield is considered remarkably high with our present method of culture. No explanation could be given except that organic manure/poultry dropping was added directly from the poultry shed built over the ponds. No special nursery ponds were used. The fry were stocked directly into the rearing ponds.

Experiments and Results

Experiment 1

Fry of 15-20 mm were stocked in a one-half acre (0.2 hectare) brackishwater pond at a stocking density of 2,000 fry per acre (approx. 1 fry/2 sq m). The salinity of water ranged from 3 to 18‰. Unlike the ponds used in Indonesia, the water level could not be kept at a low level of 30.5 to 61 cm (1-2ft), but at a depth of about 91.5 cm (3 ft). Poultry dropping was used to fertilize the pond and fine rice bran was used as feed. There was little or no algal felt found on the bottom of the pond.

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Sampling done when transferring the fish temporarily to another pond to allow repair work to be carried out showed that they grew to 140 mm in 80 days.

After stocking for 15 months the fish were harvested. They grew to an average size of 368.3 mm (14.5") and to an average weight of 688 g (approx. 1-1/2 lb). The recovery rates were 77.2% after 80 days and about 50% after 1-1/4 years of stocking.

The general growth of fry when food and stocking conditions were satisfactory was about 50-70 mm in 1 month, 120-150 mm in 2 months and 400 mm in 1 year (Hora and Pillay, 1962).

Experiment 2

When other factors remained the same, with reduced stocking rate of 1200 fry per acre (3,000 fry/hectare or approx. 1 fry/.83 sq m, the growth was 40.674 cm (16") and 890.2 g over the same growing period. The recovery was 81.8% in 3 months and 65.2% in 1-1/2 years stocking.

However, in ponds where inorganic fertilizer was used, the fish attained the length of 30.5 cm (12") in one and a half years but only with an average weight of 460.0 g (slightly over 1 lb). Experiments indicate that poultry manure is superior to inorganic fertilizer.

The second batch of fry imported from Thailand on October 22, 1973 was in poor condition. Practically, all of the fry died on the second day after the arrival.

The third batch of 64000fry with sizes ranging from 15 to 20 mm was received on June 8, 1975 from Indonesia. These were kept in an enclosure made of coarse material (cloth) for two months before they were sent to Pueh, Sematan and Punang. The fry grew to 102 mm (about 4") in 2 months. The feed consisted of rice bran.

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NOTES ON FOOD AND FEEDING HABITS
OF MILKFISH (CHANOS CHANOS) FROM THE SEA

by

Alie Poernomo*

Abstract

Study on the natural food of adult milkfish was made from specimens caught from the open sea. The contents of guts from 15 specimens were qualitatively analysed, and both planktonic as well as benthic forms were encountered. The relationship between the gut length and body length has been worked out. Ten out of 15 milkfish examined were also found to be infected with Acanthocephalan parasites confined mainly to the anterior part of the intestine.

Introduction

Under the UNDP/FAO project on Brackishwater Shrimp and Milkfish Culture Applied Research and Training, operating from the Shrimp Culture Research Centre, Jepara, an intensive program of maturation and spawning of milkfish has been initiated. One important aspect of this program is attempting hypophysation and spawning of milkfish, utilising spawners caught from the open sea. Spawners collected under the above program are also being utilised for a continuing study of the natural food of large milkfish, as our information on this aspect at present is far from complete. The salient features of this study are briefly reported in this note.

Material and Method

During the course of the study 24 specimens ranging from 52 cm/1.15kg to 101cm/9.0 kg have been examined, and the contents of 15 guts were qualitatively analysed. The coils of the guts were carefully unravelled and the length of each gut recorded. Analysis of the gut contents was made section-wise separately from oesophagus, gizzard, duodenum and intestine, to gain some idea of the extent of digestion of different items of the ingested matter.

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Observations

Length of gut in relation to the total body length of the specimen ranged from 3.32 to 7.1, but no definite correlation could be established.

Out of the 15 guts analysed, two were empty. In most of the specimens examined the anterior region of the alimentary canal from the oesophagus to the gizzard was empty. Only in three specimens this region contained some food. State of fullness of the gut was low in all specimens, though generally, guts of smaller specimens contained more food than the larger specimens. However, the limited number of guts analysed does not permit any inference about feeding habits in relation to sex and maturity of the species.

In almost all specimens the gut contents included appreciable quantities of a slimy, semi-solid substance, as observed by earlier workers also. Indistinguishable, organic detritus accounted for the bulk of the gut contents in almost all specimens. Among the distinguishable items the three dominant forms are listed in order of abundance in Table 1.

Though diatoms were represented by several species, and usually in bulk, they constituted only a small proportion of the contents. In specimen No. 13, however, Coscinodiscus formed an appreciable part of the contents. In specimens No. 10 and 12 the dominant item of food was Lucifer which were packed in the posterior end of oesophagus adjacent to gizzard and were discernible in different stages of digestion throughout the gut (Plate 1). However, in specimens No. 2 and 11 bottom invertebrates such as gastropods, lamellibranchs and their broken shells, and foraminifera constituted the bulk of the feed (Plates 2 and 3). Both planktonic as well as benthic organisms were encountered in the gut contents.

Parasitism

Ten out of the 15 specimens examined were infected with Acanthocephalan parasites (Plates 4, 5, and 6). These parasites were mainly confined to the anterior part of the intestine, though two of these extended also into the duodenum and the gizzard. All stages of the parasites were present, the eggs (40-110 microns), the young ones (10-50 mm) and the adults measuring 50-80 mm in length. The infection was heavy and the number of parasites encountered varied from 45 to 658. In almost all cases the intestinal wall at the infected area was thicker compared to the uninfected portion of the gut. This appears to be the first report of Acanthocephalan parasites infecting milkfish. Hematodes were also present, though in a limited number.

Table 1: Dominant items of food in the guts of Chanos spawners

Specimen No.	Total Length (cm)	Weight (kg)	Sex/Maturity	Length of gut (cm)	Dominant items of food	Location from where caught
1	99	6.20	indistinguishable	535	Plant tissue, Copepods	Legon Gede
2	59	1.46	-	291	Broken shell of Foraminifera, Gastropods	Legon Gede
3	100	7.80	female-spent	706.5	Empty	Katang
4	96	6.30	male-mature, oozing	349	Plant tissue, Pleurosigma, Copepods	Menco
5	52	1.15	-	201.7	None	Legon Pinggir
6	93.5	6.50	male-mature, oozing	338	Plant tissue	Menco
7	92	7.30	indistinguishable	556.5	Nitzschia, Phormidium, Copepods	Gresik
8	101	9.00	female-developing	505.5	Pleurosigma, Rhizosolenia, Nitzschia,	Menco
9	91.3	7.20	male-developing	502	None	Menco
10	90.5	5.50	male-mature	341.5	Lucifer, Copepods Ostracods	Legon Bajak
11	91	5.75	female-developing	303.5	Broken shell of Foraminifera, Gastropods	Legon Bajak
12	87.5	4.75	female-developing	434	Lucifer, Copepods, Coscinodiscus	Legon Bajak
13	72.5	2.75	--	361.5	Coscinodiscus, Hyperiidan, Amphipods Filamentous algae	Legon Bajak
14	90.5	5.30	male-developing	367.5	Plant tissue, Ostracods, Copepods	Legon Bajak
15	88	5.20	male-mature	328	Empty	Legon Bajak

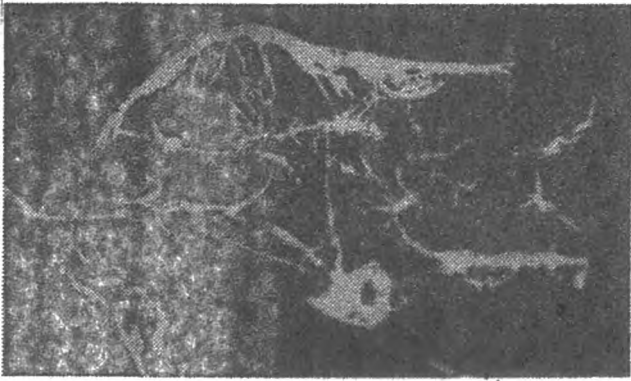


Plate 1. Lucifer.

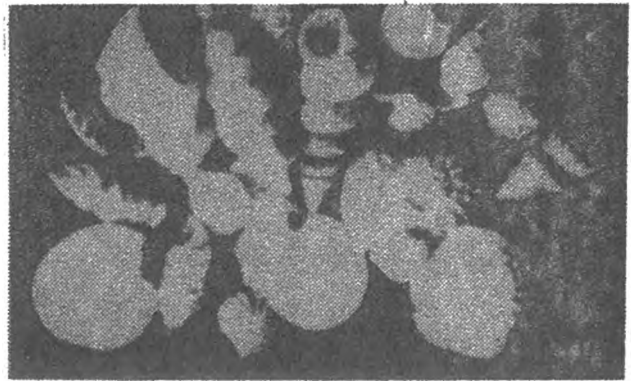


Plate 2. Foraminifera, Gastropods
and other Molluscs

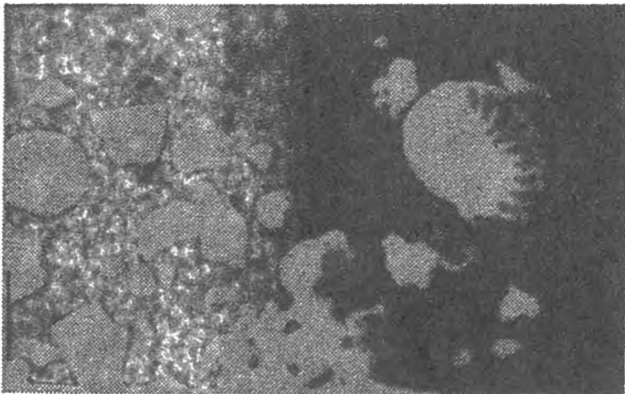


Plate 3. Foraminifera and
broken shell of Molluscs

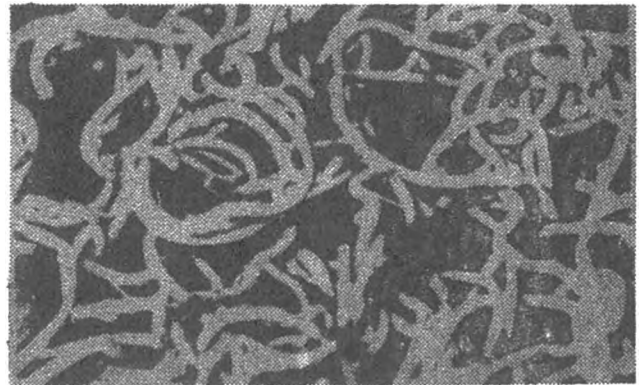


Plate 4. Acanthocephalid worms

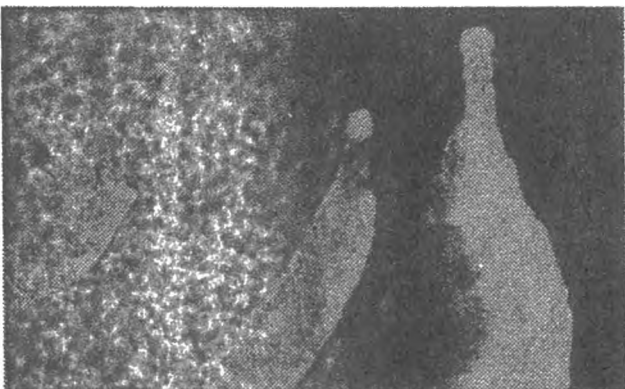


Plate 5. Proboscis of
Acanthocephalid worms

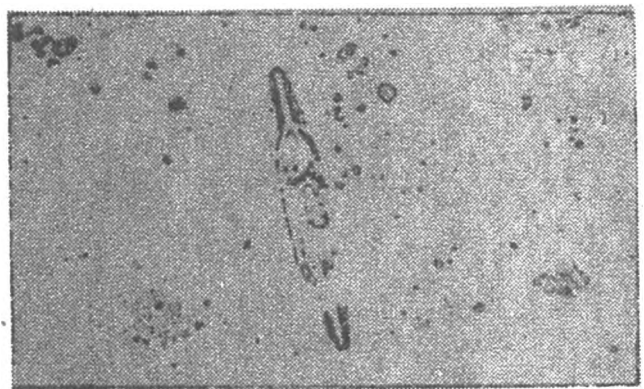


Plate 6. Egg of Acanthocephalid
worms.

Discussion

Divergent opinions have been expressed about the food and feeding habits of Chanos. LeMare 1950; cited by Tampi, 1958 assumed Chanos to be a selective feeder and Sunier classified it as a benthic feeder (1922), while Chacko (1949) after studying over 300 specimens concluded that it is a plankton feeder. That Chanos is a plankton feeder has been reported from Taiwan and Philippines and this view has been supported by the high production obtained from the deep tambaks and fish pens in Laguna de Bay in the Philippines, where the predominant food is planktonic. The gill rakers of Chanos, though small, are numerous and joined together into an effective fine sieve that could retain even fine food particles.

Data presented in this note are of interest since both planktonic as well as benthic organisms were encountered in the gut contents. However, no inference about the feeding habits of milkfish could be drawn from the few specimens examined in the present study.

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OCCURRENCE OF MILKFISH EGGS IN THE
ADJACENT WATERS OF PANAY ISLAND, PHILIPPINES

by

Tetsushi Senta, Shigeru Kumagai and Leo Ver*

Abstract

Location of spawning grounds of milkfish is one of the most important steps towards gaining knowledge on the spawning habits of the fish as well as the early life history and nature of its eggs and larvae. The present study is an attempt towards this objective. Surveys were made in selected areas in the sea around the Panay Island and milkfish eggs were collected on several occasions from surface to 20 m depth water by towing with larval nets. The eggs floated in the water in a glass jar. The eggs and newly hatched larvae had the same characteristics as described by Delsman (1929). A comparative study has been made to distinguish milkfish eggs from other more or less similar size pelagic eggs of fishes occurring in the same waters at the same time.

Introduction

The Philippines is situated in the center of distribution of milkfish, Chanos chanos (Forsk.) Filipino fishermen offer scientists a lot of information on spawning activities and spawning sites of adult milkfish. Milkfish with ripe gonads and others with spent gonads are often caught with gill nets, set nets and hook-and-line in various coastal waters. However, so far nobody has yet succeeded in collecting milkfish eggs from Philippine waters.

Aiming at locating the spawning grounds of milkfish and obtaining knowledge about early life history of milkfish in the wild, the present authors started larval net collections in the waters around Panay Island, Philippines, in early April, 1976. During the period from 4 April to 13 May, 1976, a total of 109 tows with a larval net at various layers ranging from the surface to 30 m deep was made in several areas around Panay Island. Milkfish eggs were found in 18 tows out of the 109, and the total number of eggs amounted to 88. No milkfish larva

*Dr. Senta is Technical Adviser to the Aquaculture Department, SEAFDEC, sent by the Japan International Cooperation Agency (JICA) for a period of three months, from 15 March to 14 June, 1976. Mr. Shigeru Kumagai and Mr. Leo Ver are researcher and research aide, respectively of the Aquaculture Department, SEAFDEC.

was encountered in our larval net samples. On two occasions, a few of the milkfish eggs collected were brought back alive to the laboratory and hatched out successfully in a small glass container. One of the larvae survived for 102 hours after hatching.

In any biological phenomenon, e.g., spawning of fish, distribution of eggs, etc., daily seasonal-, and annual fluctuations may occur. We may therefore, need year-round surveys for at least three years covering a wide area before we come to a conclusion with any certainty regarding, among other things, where and when milkfish mainly spawn, or in which area their eggs are most abundantly distributed. The present authors would like to request the readers of this report not to come to a hasty conclusion regarding the spawning grounds of milkfish.

Materials and Methods

The larval net used in the present study has a mouth-opening of 70.5 cm in diameter or 0.4 m² in area, and is made of nylon grid gauze of GG 38, about 500 micron in mesh size (Fig. 1). A flow-meter, TSK 4-hands type, was attached to the center of the mouth-opening of the net. At each station and layer, the net was horizontally towed for 10 minutes at about 1.5 knots.

In a surface tow, the net was kept apart from the side of the boat by using the boom so that the net was towed outside the wake (Fig. 2a). In a subsurface tow, two nets for two layers were simultaneously towed, using one of the wire warps for trawl net. A 15-kg depressor was fitted at the end of the warp, and the first net was attached 1.5 m above it. Farther, 15 m above from the first net, one end of a 14-m rope for the second net was attached (Fig. 2b). The two nets were connected with each other by a rope, 5 m in length. The wire was extended to a length three times the desired depth for the first net. Two plastic floats, 10 cm in diameter, were tied to the mouth frame of each of the nets to give a weak bouyancy to the nets. This is to prevent the net from scooping bottom mud when the boat stops to haul the net, and a precaution not to lose the net even if bridles are broken.

Each larval net collection was usually kept in a specimen bottle, 300 ml in capacity, and preserved in 5% formalin solution in situ. The specimens were examined under a stereo-microscope in the laboratory to sort out milkfish eggs. A trial was carried out with specimens from 16 tows made on 4 and 5 May to pick up milkfish eggs alive in situ aboard the boat. The remaining specimens, after sorting, were preserved in 5% formalin and later they were examined in the laboratory for any milkfish eggs which we might have missed picking out alive on the spot.

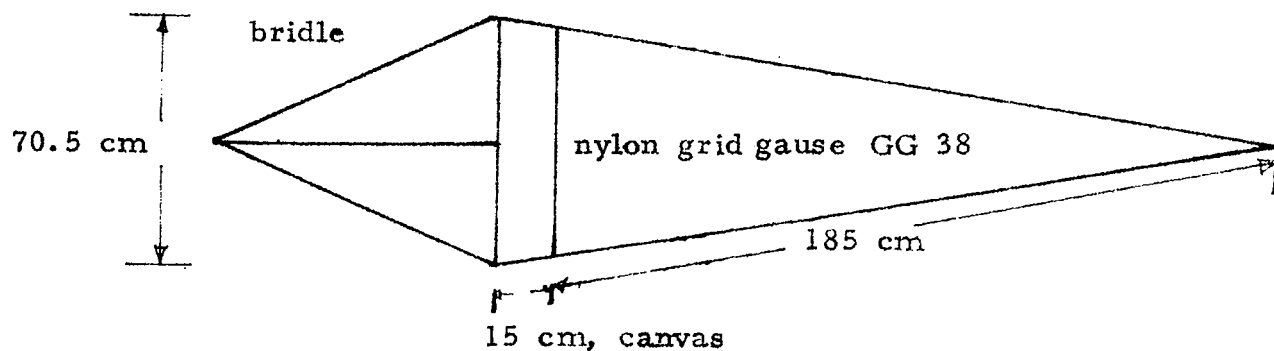


Fig. 1. A larval net used in the surveys of milkfish eggs around Panay Island in April and May, 1976. The net is conical, and made of nylon grid gauze GG 38 (mesh about 500 micron). A flow meter was attached at the center of the mouth opening.

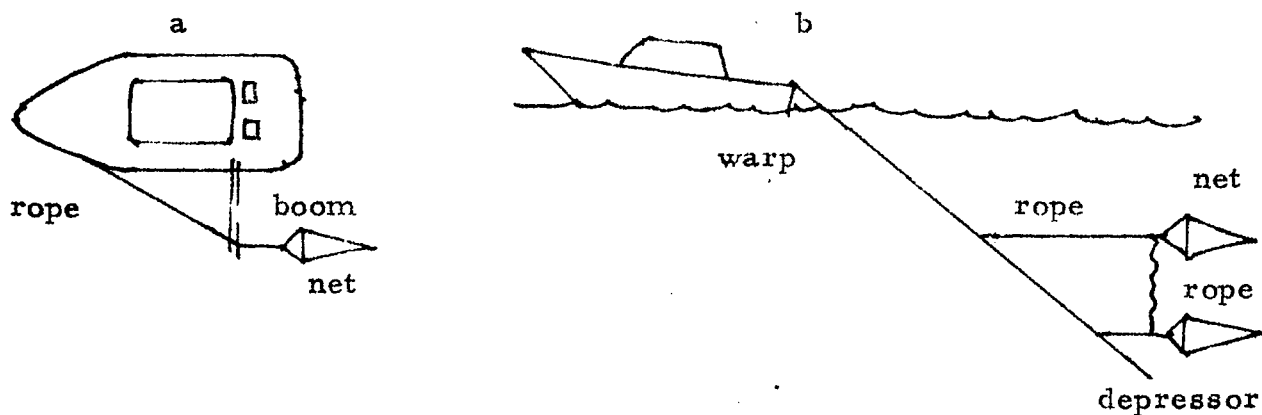


Fig. 2. Methods of towing net at surface (a) and at two layers of subsurface (b)

With the specimens collected in the Antique waters (Antique is one of the four provinces of Panay Island) in early April, not only milkfish eggs and larvae (though no milkfish larva was actually found), but also eggs and larvae of other fishes were sorted and counted. An effort was made to identify, as detailed as possible, these eggs and larvae. Only milkfish eggs and larvae were looked for in the collected samples due to lack of time. Further analysis of these samples will be done during "off season" in the future.

Areas Surveyed

For convenience's sake, the areas covered by the present survey were divided into three regions: 1. Antique waters, 2. Panay Gulf, and 3. Estancia waters. (Fig. 3).

1. Antique waters

Antique Province, one of the four provinces of Panay Island, extends along the entire west coast of the island, facing Cuyo East Pass. Larval net collections were made in three areas in this region: in Panday Bay, around Batbatan Island and off Hamtik.

a) Pandan Bay

Pandan is located at the northern end of Antique coast, where the coast line suddenly turns to the west to form a bay, Pandan Bay. Many fish corrals (traditional fish traps made of bamboo fences) are set in shallow waters along the coast, and four otoshi-amis (large size fish trap made of net)* are set along the northern coast of the bay. Adult milkfish, 70 to 90 cm in fork length and 4.5 to 7.7 kg in weight, some with mature gonads and others either immature or with spent gonads, are often caught in fish corrals and otoshi-ami from April to June, but particularly abundant in May. In 1975, 106 adult milkfish were captured in the otoshi-ami during the period from 10 May to 16 June (Anon., 1976).

A total of 13 tows of larval net collection were made at various layers from surface to 20 m deep in early April and early May. The depth of the sea ranged from 10 to 33 m.

*The word "otoshi-ami" is originally a Japanese word. Local fishermen call the net also "otoshi-ami" because the nets were donated by the Japanese Government and some Japanese experts taught the fishermen how to operate these nets.

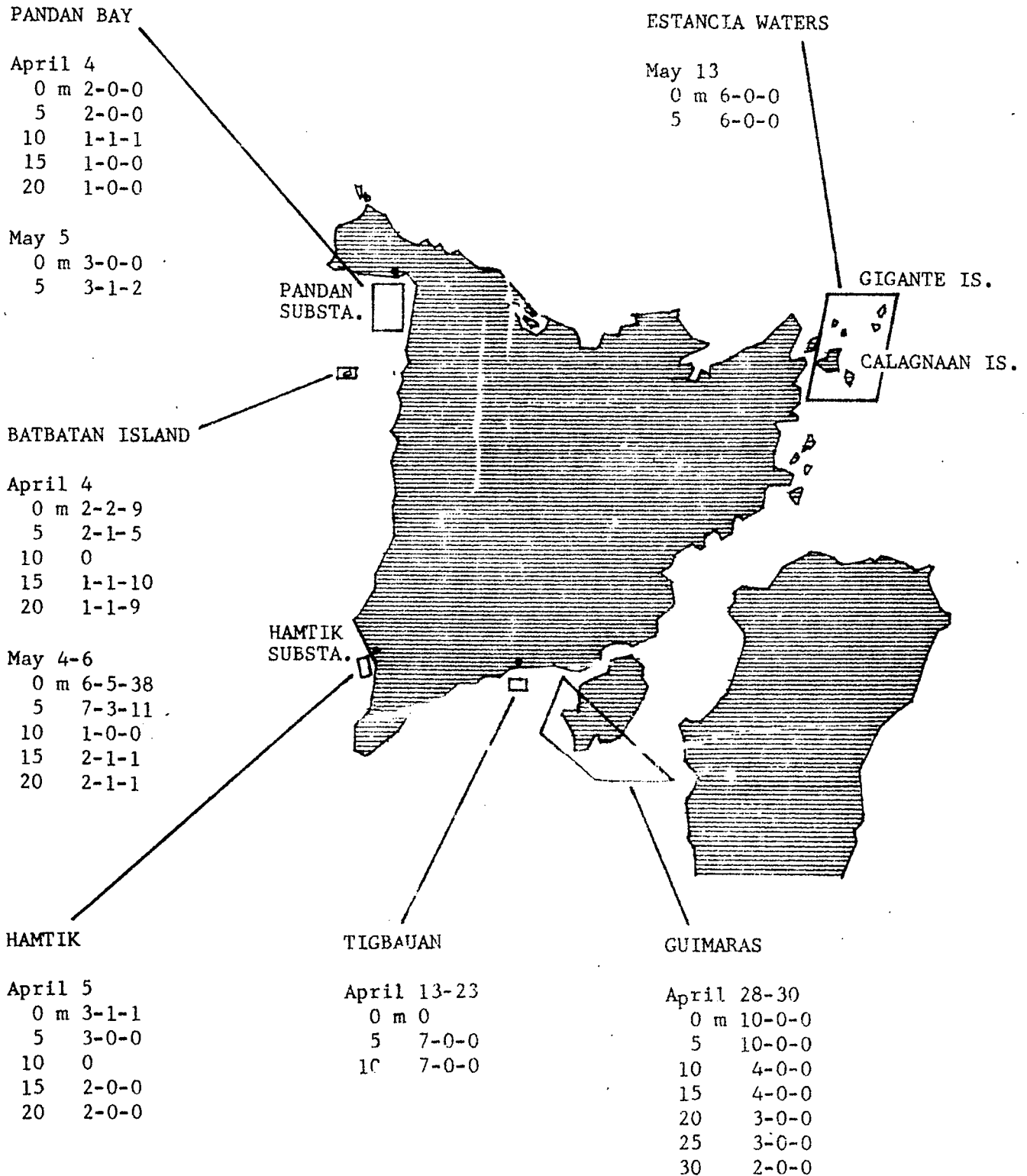


Fig. 3. Record of collections of milkfish eggs around Panay Island in April and May, 1976. Figures below the date, from left to right, show: a) the depth of the layer in which a larval net was towed; b) the number of tows made; c) the number of tows which yielded milkfish eggs; d) the total number of milkfish eggs collected.

b) Batbatan Island

Batbatan Island is a small island, about 4.4 km from east to west and about 1.5 km from north to south, situated about seven nautical miles off Lipata Point, the northern part of Antique coast, and is surrounded by a deep sea of over 200 meters deep. Near the eastern end of the island, a sandy shallow area extends some 100 m or so toward south. A sand-bar develops along the eastern margin of the shallow area to form a kind of cove which opens toward south. Outside, the sandy shallow area is hemmed by a rocky bottom of about 10 to 40 m deep and the sea becomes suddenly deeper farther outside it. According to Pandan fishermen who are engaged in gill net fishery around Batbatan Island, they had often seen milkfish jump near the above-mentioned cove during moonlit nights as well as dark nights in April and May. The same information was also obtained from the fishermen of the island.

A total of 24 tows with larval nets were made at various layers ranging from surface to 20 m deep close to the island, within 750 m from the coast, and around the above-mentioned cove. The depth of the sea ranged from 10 to 70 m.

c) Hamtik

Situated in the southern part of Antique coast, Hamtik beach forms one of the best beaches for milkfish fry collection in the Philippines.

A total of 10 tows with larval nets made at various layers from surface to 20 m deep in early April. The depth of the water ranged from 5 to 10 m.

2. Panay Gulf

Panay Gulf is surrounded by Panay Island on the north and by Negros Island on the east, with Guimaras Island between them, and opens to Sulu Sea on the south-west.

Adult milkfish are often caught in fish corrals and with gill nets along the southern coast of Panay Island and the west and south coasts of Guimaras Island. In April and in the first half of May, 1976, the Aquaculture Department, SEAFDEC, was able to collect some 20 live adult milkfish from the local fishermen.

This region is arbitrarily divided into two areas in the present report: off Tigbauan and around Guimaras Island.

a) Tigbauan

Tigbauan is located on the southern coast of Panay Island which makes a good fry ground for milkfish. A total of 14 tows of larval net collection at 5 m and 10 m layers were made off Tigbauan, the depth of water ranging from 10 m to 20 m, during the period from 13 to 23 April.

b) Guimaras Island

A total of 36 tows with larval nets at various layers from surface to 30 m deep were made in late April. The depth of the water was from 10 m to 126 m.

3. Estancia waters

Estancia is located at the north-eastern corner of Panay Island, facing the Visayan Sea. According to local fishermen, adult milkfish are often caught with gill nets and hook-and-line in the shallow waters of the Visayan Sea, especially in Asid Bay and the west coast of Masbate Island, around Zapatos Island, near Gigante Islands, and Sicogon Channel. Local people also consider that two bays (of Calagnaan Island) Apad and Balili Bays, as well as the sea around Bayas Island, a small island lying south of Calagnaan Island, are spawning grounds of milkfish.

A total of 12 tows of larval net collection at surface and 5 layers were made around Calagnaan, Gigante, Sicogon, and Bayas Islands on 13 May. The depth of the water ranged from 9 to 10 m.

Occurrence of Milkfish Eggs

Fig. 3 summarizes the results of collections made in April and May, 1976. Explanations from several aspects are given below.

1. Occurrence of eggs by areas

Milkfish eggs were collected only in Antique waters. Not a single egg was obtained either in Panay Gulf or in Estancia waters during the period of the present study.

a) Pandan Bay

In two out of 13 tows made, milkfish eggs were obtained, although the total number of eggs collected were only three. One of them was from the bottom layer at a station 10 m in depth.

The other two were from 5 m layer at a station 33 m in depth. At the latter station, an additional milkfish egg was obtained by a vertical haul from the bottom to the surface with a plankton net, 45 cm in diameter. Water temperature and salinity at the station were 31.0°C and 33.94‰ at surface, and 30.8°C and 33.83‰ at 5 m layer, respectively.

b) Batbatan Island

A total of 84 milkfish eggs were obtained by 15 out of 24 tows made in this area. The largest number of milkfish eggs obtained from a single tow, 29 eggs occurred at surface layer of a station about 750 m north-east from the eastern end of Batbatan Island. The depth of the water at this station was about 70 m, and water temperature and salinity were 30.0°C and 33.97‰ at surface, and 29.9°C and 33.95‰ at 5 m layer, respectively. The depth of the water at the other stations where milkfish eggs were collected ranged from 10 m to 40 m.

c) Hamtik

Only a single milkfish egg was collected with larval nets in this area. It was from surface layer of a station with a depth of 20 m. Surface water temperature was 28.1°C, and salinity 33.96‰.

An experimental collection of milkfish fry has been conducted daily between 0900 h and 1000 h by the SEAFDEC staff of the Hamtik Substation along the beach in front of the substation. At the end of each haul of sagap (net), fry are gathered in a small amount of water, and a collector scoops the fry (with a basin) together with water. On the land, using a small plate, the collector scoops up the fry one by one. During the period from 29 March to 6 May 1976, the water in the basin, after scooping the milkfish fry, was filtered by a plankton net and anything in the water was put into a specimen bottle and preserved with 5% formalin. Such samples were sent to the authors who examined them, looking for milkfish eggs. From the sample of 23 April, a milkfish egg was found.

d) Panay Gulf and Estancia waters

No milkfish egg was obtained either from Panay Gulf or from Estancia waters. Water temperature and salinity in both regions during the survey period were:

In Panay Gulf, 27.8-33.4°C and 32.70-34.44‰ salinity at surface layer, and 27.8-33.0°C and 33.95-34.55‰ at 10 m layer, respectively.

In Estancia waters, 29.5-30.2°C and 33.36-34.17‰ salinity at surface layer, and 29.1-30.1°C and 34.07-34.23‰ at 5 m layer, respectively.

2. Vertical distribution

As the milkfish eggs were most frequently and abundantly obtained in Batbatan waters, collection record was studied from the aspect of vertical distribution of milkfish eggs.

As seen in Fig. 3, milkfish eggs were obtained from any layers from surface down to 20 m layer, the deepest layer at which the larval net was towed. There was not much difference in number of milkfish eggs per tow by layer on 4 April, suggesting that the eggs were almost evenly distributed vertically, at least from the surface to 20 m deep. In the trip made in early May, more eggs were collected from the upper layers. On 5 May, as many as 29 eggs were obtained by a single tow at surface layer of one station, while only three eggs were collected from the 5 m layer of the station. No larval net tow was made at other layers in this station.

In our experiments on hatching and rearing of milkfish eggs, it was observed that live milkfish eggs usually floated to the surface of the water in a glass jar.

3. Daily fluctuation in catch

Within a small area close to Batbatan Island, we conducted larval net sampling on four separate days: one day in early April and three consecutive days in early May. The date indicated in the lunar calendar in parenthesis, the number of tows made at every layer, and the total number of milkfish eggs collected are as follows:

4 April 1976	5 March	6 tows	33 eggs
4 May 1976	6 April	6 tows	no egg
5 May 1976	7 April	3 tows	42 eggs
6 May 1976	8 April	4 tows	9 eggs

4. Developmental stages by time of collection

So far a total of 90 milkfish eggs were collected by larval net sampling, plankton net sampling and from the specimens of fry collection. Among them, 6 eggs were kept alive for rearing experiments and 84 eggs were preserved in formalin in situ. Below is a grouping of these 84 eggs by time of collection and by developmental stages.

<u>Time of collection</u>	<u>Developmental stage*</u>		
	<u>Aa-Ac</u>	<u>Bb</u>	<u>Ca</u>
0800-1000 h	2	1	
1000-1200	1	38	
1200-1400			
1400-1600			23
1600-1800			19

*Aa-Ac: early stages of development up to yolk invasion half completed. As yolk is easily damaged by shock of collection and preserving, it is usually difficult to determine more in detail the developmental stage with preserved specimens.

Bb: embryonic streak reaches its maximum length on the yolk, but its posterior end is flat. (In Bc, the posterior end of the embryo is vertical to the surface of yolk).

Cc: The tip of the tail of the embryo is free from the yolk, but not quite elongated.

Among six eggs brought back to the laboratory, two eggs hatched at 1900 h and two at 1915 h on the day of collection, and one at 0600 h on the following day. The remaining egg died before hatching.

5. Abundance of milkfish eggs relative to eggs of other fishes.

As shown in Fig. 3, a total of 35 milkfish eggs were collected in the Antique waters in early April 1976. The number of eggs of other fishes obtained during the same trip amounted to 10,531. The percentage of milkfish eggs was about 0.33% of total number of eggs collected. Total number of fish larvae collected during the trip amounted to 1,725. We did not get any milkfish larva, except for one doubtful specimen.

Discussion

From the results given above, we know that milkfish eggs are distributed in Antique waters. The eggs were especially abundant in Batbatan waters, suggesting that a spawning ground of milkfish is near the island. However, this does not mean that the majority of fry collected along Antique coast originate from Batbatan waters. Batbatan waters must be just one of the best, or good, spawning grounds of the fish. Further investigation in other waters, especially in the waters of Cuyo Islands, is needed.

Although no milkfish egg was obtained in Guimaras waters and Estancia waters, it is too early to conclude that there are no spawning grounds in these areas. As earlier stated, we did not get any milkfish egg in Batbatan waters on 4 May. This suggests to us the existence of daily fluctuation in the spawning activity of the milkfish. Also at present, we do not know if the spawning activity of milkfish is related to the lunar periodicity (tidal phase) or not. It was on the fifth to the eighth days of the month in the lunar calendar when the milkfish eggs were collected in Batbatan waters, whereas larval net samplings in Estancia waters were made on 15 April in the lunar calendar, that is, on a full moon day. It may be possible that the difference in lunar periodicities might have affected the results.

The live milkfish eggs floated to the water surface in a glass jar. Also, more milkfish eggs were collected at surface layer than at 5 m layer in Batbatan waters on 5 May. On the other hand, a fairly good number of eggs were obtained from deeper layers, at least up to 20 m layer. This may be attributed to the vertical movement of water in the sea. As our data regarding vertical distribution of the milkfish eggs are still very limited, more extensive and systematic studies are needed.

All the milkfish eggs collected at the same time in a day were almost in the same developmental stage. This may suggest that spawning of the milkfish takes place at a certain time of a day, probably in the evening as Delsman and Hardenberg (1931) stated. All the eggs collected by Delsman (1929) hatched late in the evening, some at 2100 h and some slightly later than 2200 h, although one egg which he collected earlier hatched early in the morning (Delsman, 1926). Most of the eggs we collected also hatched in the evening, at about 1900 h; one egg hatched early the following morning. To determine the exact incubation period in the wild, we need a 24-hour sampling.

There is one serious problem for us, however. Although in the earlier portions of this report, the words "milkfish eggs" were repeatedly used, the authors are not actually hundred percent sure whether they are truly milkfish eggs. There is no doubt that the authors are dealing with the same eggs as Delsman (1929) considered milkfish eggs because every characteristic of eggs and hatched larvae agreed with what he described. We will know the truth only when we succeed in rearing hatched larvae up to size of about seven millimeters, the identifiable stage or when we succeed in artificial insemination of milkfish.

Identification of Milkfish Eggs Preserved in Formalin

1. Characteristics of the milkfish egg

Delsman (1929) described the milkfish egg as follows: "The egg is 1.2 mm in diameter, having no oil-globule. No special structure in egg membrane. The yolk of the egg is segmented, but the segmentation is considerably fine so that according to him, he might have overlooked it with some of the eggs. The yolk has a yellow tinge."

Chacko (1950) gave a similar description, except that the diameter of the egg ranged from 1.0 to 1.2 mm.

2. Differentiation of milkfish egg from similar eggs.

Delsman (1921-1939) described about 60 species of pelagic fish eggs, while Mito (1960) gave the keys to 246 species of pelagic eggs.

According to the above-mentioned authors, a majority of fish species have eggs with single or multiple oil globules. Fortunately for us, only a limited number of fishes have been reported, so far, to produce eggs without oil globules - similar to the milkfish egg.

Among the pelagic eggs without any oil globules, we must be careful with the following five kinds of eggs, as the diameter is more or less the same as that of the milkfish egg and many of them frequently occur in the same waters.

Etrumeus microps: Yolk segmented, diameter of egg is 1.23-1.32 mm, and myotomes in newly hatched larva more than 50.

Saurida elongata: Yolk not segmented, diameter of egg is 1.20-1.33 mm, and myotomes in newly hatched larva 57-60.

Scorpaenidae type: Yolk not segmented, diameter of egg is 1.0-1.1 mm, and myotomes of embryo less than 30.

Saurida type: Yolk not segmented, diameter of egg is 1.0-1.1 mm, and myotomes of embryo more than 30.

Unknown fish 'A': Yolk not segmented, diameter of egg is 1.2 mm, and yolk probably with a yellow tinge.

Although it is usually difficult to know whether the yolk is segmented or not with preserved specimens, yolk in eggs of Saurida elongata, Scorpaenidae type, and Saurida type in formalin look more transparent and colorless compared to the yolk of milkfish egg. At first sight, egg of unown fish 'A' is almost similar to milkfish egg in size, shape of embryo, and color. However, small melanophores are more densely distributed on embryo and myotomes cannot be recognized clearly with preserved specimens, while in milkfish egg myotomes are marked when viewed from the side. Furthermore, we can ascertain the segmentation of the yolk by tearing the egg membrane and puncturing the yolk with pins.

Differentiation of milkfish eggs from those of Etrumeus microps (Family Dussumieridae) seems to be rather difficult, especially in the early stage of development, although the yolk of the milkfish egg is more finely segmented, about 0.04 to 0.05 mm, than that of Etrumeus microps, which is about 0.06 mm. In later stages of development, we can estimate the number of trunk myotomes of the embryo and the melanophores in the embryo is much thinner in the milkfish egg than in Etrumeus microps.

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REVIEW OF WORK PROGRAMMES CARRIED OUT IN INDIA
ON THE BIOLOGY OF THE MILKFISH CHANOS CHANOS

by

P. R. S. Tampi and P. Bensam*

Abstract

This account is a brief review of the investigation carried out in India on the various aspects of the biology of the milkfish. It covers the records of observations on the distribution of Chanos fry in the coastal areas, food and feeding of the fish, its growth, maturity, fecundity and spawning, and also certain experimental results on the physiological adaptation of the fish together with the histological structures of its kidney, pituitary and thyroid. While emphasising the paucity of data on the adult milkfish in the wild, the priority areas where information is lacking especially on the spawning ground, reliable identifying characters of the egg as well as on the possible existence of different racial stocks have been discussed.

Introduction

The milkfish occurs in the seas around India and in limited numbers the fish are caught near the coast of Orissa, Andhra Pradesh and Tamil Nadu on the east and Kerala and Karnataka on the west coast. The fish is also being captured near the islands of the Andaman-Nicobar group in the Bay of Bengal and around the Laccadives in the Arabian Sea. The fish in all cases are invariably obtained in the nets operated by local fishermen within about 8 km. from the coast. One fact remains that in place in this country does the milkfish constitute a significant commercial fishery; its position in this country's fish catch statistics being almost nil. In certain backwaters, estuaries and lagoons there is a seasonal fishery of some magnitude and this is restricted to the juveniles whose size rarely exceeds 500 mm. Thus, the status of this fish in the present commercial fish production of India is comparatively low and insignificant.

While its natural fishery is poor, it is noteworthy that there is a fairly abundant occurrence of the fry and fingerlings of the fish in many tidal creeks, estuaries and backwaters along the coast of peninsular India

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and also in tidal inlets of the bay islands. An assessment of this valuable resource or its proper utilization is yet to be carried out. This is largely because salt water fish farming in an organized and scientific manner has been initiated in this country only in recent times and the efforts in this direction so far have been only on experimental basis. For this reason, biological studies on the milkfish, Chanos chanos also have been limited. This paper presents a brief review of the various items of research carried out on the biology of the fish (culture excluded), together with identification of the major gaps in our existing knowledge.

Distribution and Abundance of Fry

In view of its utility in brackishwater culture operations, investigations on the availability and abundance of the fry and fingerlings were initiated since the early thirties (Raj, 1931; Chacko, 1942; Job and Chacko, 1947; Ganapati et al, 1950; Panikkar et al, 1952; Chacko and Mahadevan, 1956; Rao, 1972). In the zone extending from Orissa State in the east to Karnataka in the west, the State of Tamil Nadu is found to furnish the maximum yield of milkfish fry and fingerlings, with the Ramanathapuram-Tirunelveli districts constituting the most important region. It was further pointed out (Tampi, 1973) that intensive collection of data on the occurrence and abundance of Chanos fry has not been made so far. Neither any particular effort has been taken nor any adequate survey methods have been followed till now. It was estimated that with greater efforts and by extending the period of collection throughout the fry season all along the Tamil Nadu coast alone, at least ten million fry could be collected every year. And, a rough estimate for the whole of the peninsular India would take us to the order of nearly twenty millions of fry and/or fingerlings per annum. It is obvious that the collection of data on the milkfish fry resources remains an important piece of work to be taken up.

Chanos fry occurring in the shallow lagoon adjoining the Palk Bay were first observed in 1952. The fry find their way through the temporary connections that are established between the sea and the lagoon in certain years and continue to remain in the lagoon during the season as long as the sand bars remain open. In the adjacent areas along the coast itself, however, it has not been possible to locate any fry. Some experiments were conducted to see if the fry could be made to enter other areas. For this purpose shallow channels were excavated at strategic places which led to small pools. The experiments were rewarding in that we were able to make collections of fry in such artificial channels. During the fry seasons in subsequent years also similar experiments were repeated with success. It thus appears possible to make fry collections

even from places which may not normally constitute a collection centre, provided suitable environmental conditions are created to induce the fry to swarm in (unpublished observations of Tampi and S.V. Job). It is perhaps common knowledge that shallow bays, lagoons and tidal creeks are excellent nursery grounds for the young ones of a variety of fish and crustaceans. Many of these in the early stages of their life history seek entry into the calm sheltered waters of the coast where benthic food organisms and detritus are abundant and where ecological conditions are quite favourable for their life and growth. The fry of a number of marine forms such as the mullets and shrimps exhibit the same type of behaviour. It is thus felt that the behavioural characteristics of the fry could form an important aspect of study and the results could open up possibilities for improving the existing collection techniques.

Food and Feeding

Brief references to the food and feeding of Chanos were made by Chacko (1945, 1949) and Chacko and Mahadevan (1956) that the larvae feed entirely on planktonic organisms like diatoms and copepods found in the shore areas; fingerlings feed on diatoms, copepods and to a great extent on algae in the shore regions; and the adolescents as well as adults feed on diatoms, copepods, larval bivalves, Lucifer, Mysis, etc. indicating indiscriminate feeding in their movements between neritic and oceanic areas.

Tampi (1958) made a more detailed analysis of the food and feeding of milkfish and reviewed the data published in the light of his own observations. For purposes of study, the growth stages were divided into three categories, (i) fry and fingerlings, including all sizes from 14 mm to 100 mm in total length; (ii) juveniles ranging from 100 mm to sexually immature specimens; and (iii) adults which have attained sexual maturity. The fry and fingerlings had fed mostly on diatoms such as Chroococcus, Fleurosigma, Diploneis, etc., blue-green algae, detritus and occasionally on nematodes and crustacean larvae, usually found at the bottom of the shallow swampy areas. Juveniles were found to feed more on algae, particularly blue-green and green algae and less on diatoms. A few fish collected from the sea ranging from 250 to 300 mm showed mainly Microcoleus, Phormidium, Lyngbia, etc. in their stomachs. Specimens above 300 mm in length showed small quantities of mucoid substance, with the alimentary canal collapsed and empty in many cases. The condition of the alimentary canal gave the impression that the fish had been starving for some time before their capture. A few fish collected from culture ponds had shown mostly mud and decaying organic matter with a few diatoms from the pond bottom.

The adult fish caught from the sea revealed the presence of Hypnea and Gracilaria in large quantities, but the most significant feature was the presence of lamellibranch spats in the stomach contents, sometimes in gorging proportions. Based on the fact that these spats are usually found attached to algae, Tampi (1958) was of the opinion that the fish had resorted to browsing among the algae growing near the bottom along the coastal regions where they were caught in shore-seines, and that the fish was able to crush such organisms in the highly muscular gizzard. It has also been inferred from these studies that the adult fish is not as much predominantly vegetarian as had been considered earlier, but that in the later stages of their growth they appeared to be capable of consuming and digesting a variety of animal matter also along with plant material from the surroundings. The observations of Kapoor (1954) on the morphology and histology of the pharyngeal organ in Chanos would be of relevance while interpreting the food and feeding of the fish.

Growth and Age

Our knowledge on the growth and age of Chanos in India is based chiefly on the observations on the fish cultured in marine, brackish or freshwater ponds and very little information is available on the growth and age of the fish from its natural habitats. Chacko and Mahadevan (1956), while reviewing the Chanos culture experiments carried out at Krusadai Island, found that the growth in the first six months was much rapid, (160-170 mm) and considerably slower in the succeeding six months. The fry measuring 50 mm and 85 mm stocked in June had grown to 235 mm and 260 mm by next June. These authors had stated that this limit might be considered to represent the normal growth of the fish in the neritic regions of the sea during its first year. These authors had further stated that the growth of Chanos stocked in certain brackish-water ponds was 320 mm to 425 mm in the first year, 500 mm in the second year and 550 to 600 mm in the third year. This is said to be much faster growth than the condition observed in the sea or in the marine fish farm. These authors had also found that the growth was even more rapid in freshwater tanks, up to 380 mm in the first six months, 600 mm in one year and 750-950 mm in two years. Based on a few specimens collected from the sea, Tampi (1957) had tentatively concluded that the age of females at first maturity, measuring about 1,100 mm could be 4 to 5 years. It was also observed that the females about 600 mm long could be about two years old and those of 900 mm are about three years. In the culture experiment carried out at Mandapan, Tampi (1960) observed a growth increment of 220 mm by the end of one year, the growth being more rapid in the first few months than in subsequent period. Observations on growth checks on fin spines, vertebrae or scales were less helpful in making reliable estimates of age.

While the opercular bones and the fin spines did show some definite rings on them, their utility in correlating with age remains questionable. The scales, on the other hand, were found to be too thick and opaque as the fish advances in age and conventional processings had not yielded any satisfactory results.

Maturity

Tampi (1957) found that Chanos below 500 mm in total length generally remain indeterminates. Differentiation of the gonad into testis or ovary under the microscope was possible only in specimens above this length. Two stages of maturity in males were collected, one in a very early stage and the other in an advanced condition. The absence of mitotic figures or dividing spermatogonia in males below 830 mm indicated that spermatogenesis took place in later stages only. In mature males measuring 1,080 to 1,140 mm total length, sperms were observed in spermatic tubules. In the female specimens four maturity stages were recognizable: (i) the earliest one in which differentiation as ovary was possible; (ii) the immature stage with uniform development of ova; (iii) the nearly mature condition with ripe intra-ovarian eggs; and (iv) the spent and recovering stages. The ripe ovaries were bright yellow in colour owing to the yolk-laden eggs. The ova were arranged closely in the ovigerous lamellae in a transverse manner. They were exposed, facing the coelomic wall, showing a gymnovarian condition. The ripe ova were spherical and translucent, without an oil globule, contained finely divided yolk and measured an average diameter of 0.8 mm. The spent ovaries contained bodies resembling stages in the formation of a "corpus luteum".

Although sufficient data and material have not been available on these aspects, the studies of Tampi (1957) have indicated that the ovary starts active development with the formation of oocytes when the fish is more than about 500 mm in total length, but in males the spermatocytes begin to appear only after the fish reaches a size of more than 900 mm. The size ranges of males and females of corresponding maturity stages have shown that the maturation of the testes proceeds faster than the ovaries and that the males are smaller than the females at the same maturity condition. The histological studies of gravid females did not reveal the possibility of the fish having spawned earlier; and, based on this it was assumed that the first spawning takes place when the fish is about 1,100 mm long and 11 kg in weight.

Fecundity and Spawning

Chacko and Mahadevan (1956) gave the fecundity of Chanos as 1.9 to 2 million eggs; but, the studies of Tampi (1957) have shown that mature eggs in gravid females varied between two million and five million in number. It was also found that the number of eggs present was proportional to the length of the fish.

The ova diameter frequency studies undertaken by Tampi (1957) on mature fish have shown the absence of multiple model curves, but the presence of only two widely separated groups of immature and mature eggs, indicating that the fish has only one spawning season in a year. However, the presence of indistinct minor modes and the wide range of size seemed to indicate some sort of fractional spawning within the season.

Based on the observations on the occurrence and availability of the fry, it is inferred that the fish usually spawns during February to May period. Panikkar et al (1952) once recorded the fry during November and December in the Mandapam area when spent females also were collected. Therefore, they had drawn attention to the possibility of a subsidiary spawning during October-December. Rao (1972), based on the continuous occurrence of the fry from February-March to October, is of the opinion that individual fish may be spawning independently of the others in the population.

As in the case of spawning season, the extent of the spawning grounds of Chanos in the Indian seas is to be inferred only from the occurrence and abundance of the fry. The Gulf of Mannar side on the southeast coast is undoubtedly one of the areas which show the most abundant concentration of Chanos fry. Similarly, many places around Point Calimere, Madras, Masulipatnam and Chilka farther towards the north are good collection centers for early fry. Judging by the size of these fry and the time required for their migration to the coast, it may be contended that the spawning grounds are not far off from the coast. But concrete evidences are lacking.

Based on the relative abundance of the fry during new moon period than during full moon period (Chacko and Mahadevan, 1956; Rao, 1972), it appears that the spawning intensity is greater during the former period. The observations of Malupillay and Chacko (1959) and Rao (1972) that the peaks of occurrence of the fry were preceded by rainfall, indicate that the spawning process takes place during or soon after the rains. Many marine species of fish are known to become sexually active on the onset of the first rains.

Eggs and Larvae

The only report on the collection of the eggs of Chanos from Indian waters is that of Chacko (1950) from the Gulf of Mannar. Their identification was based on the descriptions given by Delsman (1926, 1929) of Chanos eggs collected from the Jave coast. Detailed descriptions and figures of these eggs and the larvae hatched out of them have not been published so far. Subsequently, Chacko and Mahadevan (1956) had collected from the Nellore coast eggs resembling those of Chanos, but these were said to be smaller in diameter. One of us (P.R.S.T.) had isolated large numbers of eggs from the surface plankton collections of the Gulf of Mannar during January-March 1951, i.e. the months immediately preceding the appearance of fry along this coast and what could be the probable spawning season. These eggs closely agreed with the known descriptions of Chanos eggs. However, the larvae hatched out from them, especially of the second and third days, failed to show the pattern of arrangement of the muscle fibers in the myotomes or the pigmentation said to be characteristic of Chanos larvae. Similar collections were made in subsequent years also from these localities. Difficulties in rearing the hatchings beyond the critical phase and to take them to an identifiable stage have been the main bottleneck in settling the identity of the eggs and larvae.

Physiological Adaptation

Some studies were made of the adaptive responses of the fish to variations in salinity, temperature (Panikkar, et al, 1953) and dissolved oxygen contents of water (Viswanathan and Tampi, 1952). It was found that the fingerlings maintained their blood serum chloride fairly constant in a wide range of external salinity variations. In external salinities corresponding to a chloride content of 5 ppm and less, the serum chloride values were suggestive of a breakdown of osmoregulatory mechanism of the fish. However, the presence of calcium (144 ppm) in the external medium aided the fish in the retention of blood salts thus pointing to the significance of calcium ions in osmoregulatory functions. The salinity of the water did not appear to influence to any significant extent such metabolic characteristics of the fish as heat-death temperature, oxygen consumption, resistance to asphyxiation, etc. Discussing the significance of the data, the authors pointed out that any rational system of fish culture requires as an essential prerequisite information on the tolerance limits of salinity and temperature, the influence of ions in the water and of the size of the fish. The relationships between size and metabolic activity of the fish were found useful in the calculation of the rate of variation of the number of viable fish under transport with the size of the fish and in providing a convenient measure for the comparison of the viability of the fish under different environmental conditions.

Pituitary, Thyroid and Kidney

In view of the importance of the excretory organs and endocrine system in the maintenance of a steady state of internal environment of the fish, a study of the kidney, pituitary and thyroid in different stages of the fish was made by Tampi (1951, 1953 and 1959). The kidney in the adult fish has the conventional histological structure, composed of glomeruli followed by a ciliated neck, a comparatively narrow proximal convoluted segment and a broader distal segment. The kidney in the larval stages in the 15 mm larva (unpublished data of P.R.S.T.) is a much simple structure consisting of 15-20 glomeruli, each with a short, apparently undifferentiated tubule, all of which are embedded in a highly vascular haemopoietic tissue surrounding the main dorsal blood vessel. At this stage the urine is conducted to the outside by means of a common ureter. The significance of the finer structure of the rural tubules, if properly understood through experiments, would be of immense benefit in understanding the osmoregulatory powers of the fish. Structure peculiarities in the pituitary of the early fish were observed although their functional significance is not known. Similarly, there is a fairly good account available on the nature of the thyroid cells of the larval fish under different salinities, temperature and under the influence of certain thyroid inhibiting drugs (unpublished data of P.R.S.T.).

General Remarks on Future Lines of Investigations

The paucity of data on the adult milkfish has been a general constraint and the need to have a better understanding of the biology of the fish has been widely stressed. As the distribution of the fish covers a very large region spread over the Indo-Pacific, it is essential that the various countries concerned collaborate and obtain as much reliable information as possible in this respect. Several agencies in these countries are at present actively engaged in studies on the different aspects, most of which perhaps lay greater stress on the practical problems related to its culture. While a cooperative approach could yield quick and useful results in unravelling the problems on the life of such a fish with extensive distribution, it may be possible to tackle some of the studies on a regional or localized basis and depending on the facilities and interests of the specific region. In this manner the existing gaps could be covered so that a complete account could be built up if a planned approach is taken.

When viewed in this background and priorities are drawn up, studies on artificial inducement of spawning of the milkfish may perhaps gain the first place. It is gratifying to note that certain

organizations are seized of this problem and experiments are underway to make the fish breed artificially through the technique of administering pituitary hormones. This technique, as we know today, is applicable only to make sexually mature fish release the gonadal products so that external fertilization of eggs could be carried out. In the case of the milkfish, obtaining sexually ripe fish either in nature or in ponds remains a problem by itself. Therefore, efforts will have to be made to see how the fish could be reared to a stage when pituitary hormone administration could be effective. In this connection some fundamental studies on the physiology of the fish, covering the whole gamut of reproductive physiology together with endocrinology as well as adaptational physiology and metabolism, would be helpful.

The next item where a cooperative effort would be called for is the location of the spawning grounds as well as identification of the eggs and early larvae from the plankton. It is time that the question of identification of the eggs is reviewed in greater detail and the identity established so that the workers in the different regions could have a positive guide in this respect. For this purpose, horizontal and vertical plankton collections during appropriate seasons should be intensified in the different regions and efforts should be made to hatch and rear the eggs to an identifiable stage so that the reliable characters could be delineated. This would also help in the location of spawning grounds of the fish.

A critical study of the spawning season reveals certain interesting features. By and large, the main fry and fingerling season in many regions of the Indo-Pacific coast is during the summer months of April-July. However, in many places fry have been reported to occur in other months; this has led us to believe that there is more than one breeding season, or that individual fish may exhibit variations besides the species as a whole showing protracted spawning behaviors. Apart from the possibility of existence of different racial stocks with different spawning characteristics, one factor which appears to be involved and which does not seem to have been given due consideration is the pattern of migratory behaviour of the fry and fingerlings. For, if the exact time of appearance of fry along any particular coast is carefully analyzed, especially in relation to the size, some definite pattern is likely to emerge. At least, it is so in the case of the occurrence of *Chanos* fry along the east coast of India where the fry are first reported from the Gulf of Mannar area and gradually as the season advances they show up in the northern centres towards the later months. There is some increase in their average size also as they occur in these northern centres. While this is only a general observation which needs more critical analysis and a basis determined, from our unpublished observations along the Indian coast there appears to be a

need to study this aspect carefully. It is not clear if hydrographic factors have any influence on this type of behaviour. Side by side with such studies, it would be advantageous to conduct investigations on factors that tend to attract the fry to specific localities, any periodicity or cyclic occurrence and also their migration back to the open seas when once they pass the fingerling stage. Some tagging operations on fingerlings at selected centres would prove beneficial in unravelling some of the less known problems of behaviour of the fish in this early phase.

For such of those regions which are primarily concerned with this fish as the principal species for brackishwater culture and where potentials exist, it is imperative that a proper survey and estimation of the natural fry resources are completed without further delay. At the same time, those regions indulging in fry collection as a trade would do well to watch the trend of collections from year to year so that any possible effect on the ultimate stock could be studied, although the removal of a few million fry every year from any locality is unlikely to result in any serious adverse effect in the case of a species whose fecundity is phenomenal.

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MORPHOLOGICAL MEASUREMENTS, GONADAL DEVELOPMENT
AND ESTIMATED AGE OF ADULT MILKFISH, CHANOS CHANOS
CAPTURED IN PANDAN BAY FROM 10 MAY - 16 JUNE, 1975*

by

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Abstract

From 10 May to 16 June, 1975, 106 adult milkfish were captured in an otoshi-ami 500 meters offshore. Of the 106 specimens, 37 were females in various stages of gonadal development or spent and 69 were males of which 41 were sexually mature. The age of these fishes were estimated to be between 3 and 5 years.

Introduction

From 10 to May 16 June, 1975, the SEAFDEC study team which was stationed in Pandan, Antique Province, Philippines (Kumagai et al., 1976) obtained 106 adult milkfish, Chanos chanos from the otoshi-ami adjacent to the laboratory site at Mag-aba, Pandan. Information on the length, weight, gonadal development, gonado-somatic indices of the captured milkfish together with records on salinity and temperature of water at the otoshi-ami have been detailed in this report. Attempts have also been made to determine age of these sabalo by study of their scales.

Materials and Methods

The daily routine of the commercial fishermen working on the otoshi-ami at Mag-aba included lifting the bag of the net and collecting the fish catch at about 0400, 1200 and 1600 hours. Members of the study team accompanied the fishermen daily and obtained water samples from depths of 5, 15 and 30 meters during the 1200 hour fishing operation. Water temperatures were recorded immediately and the salinity of each sample was determined at the laboratory. Whenever adult milkfish (sabalo) were caught in any of the three daily catches

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they were usually killed at the otoshi-ami and transported to the laboratory within one hour of removal from the net. Prior to arrival of the study team at Mag-aba on 13 May, the commercial fishermen captured 19 sabalo on 10 May. These specimens were immediately placed in a holding pen constructed of 2-inch (5 cm) stretch mesh knotted nylon net 12 m x 13 m x 15 m deep adjacent to the otoshi-ami. On 14 May these fish were killed and designated as sample Nos. 1-19 by the study team. Fourteen of the sabalo captured on 18 May were also placed in the holding pen and 2 of these (Nos. 64 and 66) were killed on 24 May while the remaining 12 (Nos. 76-87) were killed on 27 May. All specimens which were held in the holding pen sustained multiple injuries and had opaque adipose eyelids at the time of sampling.

After recording the length and weight of each sample at the laboratory, gonads were weighed and preserved in 10% seawater formalin. Similarly, the anterior spiral portion of the oesophagus was preserved in 10% seawater formalin for qualitative analysis of its contents as reported by Villaluz et al. (1976).

Two specimens of female sabalo containing almost sexually mature ovaries were captured in a fish corral located at Hamtik, Antique, 100 km south of Pandan on 8 April and 8 May. Fecundity of these samples and of Mag-aba sample No. 62 was determined gravimetrically (FAO 1958).

Five scales were removed from each of the following four regions of each sabalo sampled: (1) midway between the lateral line and the dorsal fin; (2) immediately posterior to the ventral fin; (3) directly above the pelvic fin; and, (4) around the anus. The scales were air-dried and approximately two months later were cleaned in warm water to remove attached mucus and re-dried. The cleaned and flattened scales were examined at a magnification of 400 times and the age of each fish estimated by the method followed by Chiu (1965). This method, developed for age study of silver carp, consists of counting the number of sets of discontinuous lines as illustrated in Fig. 1. Scales collected from above the pelvic fin did not contain clear sets of continuous lines and were discarded. Scales from the other three regions on each fish contained the same line patterns and were used for estimating the age.

Results and Discussion

The total daily catch of adult milkfish from the Mag-aba otoshi-ami between 10 May and 16 June, 1975, together with those obtained from the operator of the otoshi-ami during January to 4 June, 1974 and from January to 9 May, 1976 were recorded and data analyzed.

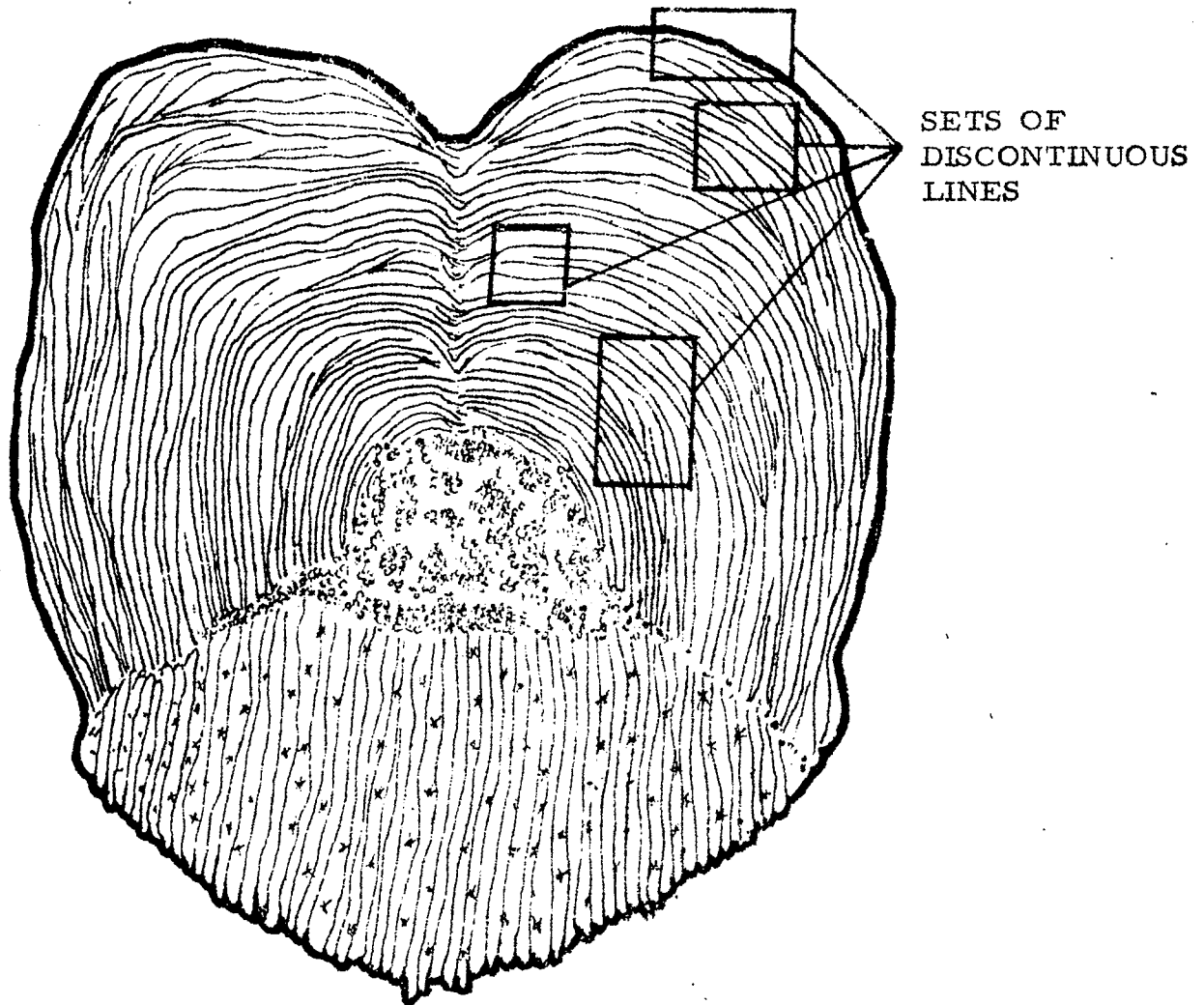


Fig. 1. Diagram of a typical dorsal scale from a Sabalo with an estimated age of four years.

The temperature and salinity data obtained from the operator of the net and also collected by the study team is presented in Table 1.

Although the otoshi-ami was in operation from December to late June during the 1973/74 and 1974/75 fishing seasons, no sabalo was caught prior to late April 1974 or early March 1975. Of the 106 specimens obtained by the study team during 1975 (Tables 2 and 3) 37 were female in varying stages of gonadal development or spent, and 69 were males of which 41 were either sexually mature, or partially spent with freely flowing milt. Five or more scales from each of the 81 of the 106 sabalo obtained from the Mag-aba otoshi-ami contained distinct sets of discontinuous lines. The age of these 81 fish were estimated to be between 3 and 5 years. From preliminary observations, Tampi (1957) suggested that the age of female milkfish at first maturity may be between 4 and 5 years and Liao (1971) estimated the age of 9 adult milkfish caught in the colder waters of Taiwan, to be between 5 and 7 years.

Sets of discontinuous lines were absent from the scales obtained from several six to eight months old pond-reared milkfish. However, two sets of discontinuous lines were present in the scales of two 2-year-old female fish obtained from the Bureau of Fisheries fishpond at Naujan, Mindoro. These two fish weighed 1.2 and 1.5 kg with fork lengths 43 and 45 cm and total lengths 53 and 57 cm respectively. Histological examination of the ovaries from these two fish showed the beginning of ova development which is in agreement with the findings of Tampi (1957).

Ovaries with developing ova were light yellow or pale orange in color while the ovaries from spawned fish were dark reddish-brown. The total number and diameters of ovarian eggs from Mag-aba Sample 62 and the 2 samples from Hamtik are presented in Table 4 together with data from other investigators.

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Table 1. Range and mean monthly water temperature ($^{\circ}\text{C}$) and salinities ($^{\circ}/\text{oo}$) at depths of 5, 15 and 30 meters during the 1974-75 fishing season at the otoshi-ami located at Mag-aba, Pandan, Antique.

	Dec. '74	Jan. '75	Feb. '75	Mar. '75	Apr. '75	May '75	June '75
Depth (m)							
Temperature ($^{\circ}\text{C}$)							
5	26.0-27.8 (26.5)	24.5-26.9 (25.8)	23.5-27.0 (25.5)	26.0-28.0 (26.8)	26.0-28.5 (27.5)	28.0-30.1 (29.5)	28.5-30.4 (29.1)
15	26.0-28.0 (26.5)	24.9-26.8 (25.9)	23.5-27.0 (25.5)	26.0-28.0 (26.8)	26.0-28.5 (27.5)	27.7-29.8 (29.5)	27.3-29.5 (28.7)
30	26.0-28.0 (26.5)	25.0-26.9 (25.9)	23.5-27.0 (25.5)	26.0-27.0 (26.8)	26.0-28.5 (27.6)	27.7-29.6 (29.0)	27.0-28.8 (28.2)
Salinity ($^{\circ}/\text{oo}$)							
5						May 15-30 33.4-35.0 (34.2)	June 1-17 31.9-34.4 (33.3)
15						33.2-35.2 (34.6)	32.6-34.4 (33.4)
30						34.0-36.4 (35.0)	32.9-35.4 (34.2)

Table 2. Fork Length, body weight, ovary weight and estimated age of adult female milkfish

Date of Catch & Fish No.	Fork Length (cm)	Fork Weight (kg)	Ovary Weight (gm)	Gonadal Development	Gonado-somatic Index	Estimated Age
May 10/75						
1	87.5	7.70	50	Spent	0.65	-
2	76.0	5.32	40	"	0.75	3
4	75.0	5.20	25	"	0.48	3
7	79.0	6.50	60	"	0.92	-
8	83.5	6.10	20	"	1.31	-
9	75.0	6.45	20	"	0.31	-
12	85.5	7.00	80	"	1.14	4
13	72.5	5.60	70	"	1.25	3
14	76.0	5.90	75	"	1.27	4
15	89.4	6.15	95	"	1.54	-
16	81.6	6.80	90	"	1.32	4
18	81.6	7.27	70	Developing	9.32	4
19	77.0	5.75	95	Spent	1.65	3
	(85.5)	(6.28)				
May 18/75						
24	84.5	6.37	190	Spent	2.98	5
64	77.3	6.25	60	"	0.16	3
76	78.5	5.80	66	"	1.14	4
78	76.7	5.80	59	"	1.02	4
80	81.0	6.65	119	Developing	1.79	4
83	74.5	5.15	48	Spent	0.93	4
86	72.8	5.13	136	"	2.65	-
87	76.1	6.54	50	"	0.76	4
	(77.7)	(5.96)				

Figures in parentheses represent mean length and body weight of fish caught on the same day.

Table 3.. Fork length, body weight, testes weight and estimated age of adult male milkfish

Date of Catch & Fish No.	Fork Length (cm)	Body Weight (kg)	Testes Weight (gm)	Gonadal Development	Gonado-somatic Index	Estimated Age
May 10/75						
3	77.0	5.47	20	Spent	0.36	3
5	78.0	4.50	10	"	0.22	3
6	85.0	4.57	10	"	0.22	-
10	75.0	5.35	20	"	0.37	-
11	80.5	5.90	40	"	0.68	-
17	76.0	6.20	50	"	0.81	4
	(78.6)	(5.33)				
May 17/75						
20	83.0	6.35	80	Spent	1.26	-
May 18/75						
21	77.0	6.22	265	Mature	4.26	3
22	76.5	6.52	315	Mature	4.83	4
23	74.0	4.95	105	Mature	2.12	4
66	72.2	4.95	80	Spent	1.62	-
77	80.5	6.00	20	Spent	0.33	4
79	74.6	4.85	14	Spent	0.29	4
81	74.7	5.55	20	Spent	0.36	4
82	70.5	4.10	8	Spent	0.19	-
84	73.9	5.55	19	Spent	0.34	4
85	75.9	5.70	21	Spent	0.37	4
	(75.0)	(5.44)				
May 19/75						
25	85.0	7.15	230	Mature	3.21	-
26	73.5	6.15	99	Spent	1.61	4
30	76.5	6.25	250	Mature	4.00	3
31	77.5	5.75	60	Spent	1.04	4
	(78.1)	(6.33)				

Figures in parentheses represent mean length and body weight of fish caught on the same day.

Table 3. (cont'd.)

Date of Catch & Fish No.	Fork Length (cm)	Body Weight (kg)	Testes Weight (gm)	Gonadal Development	Gonado-somatic Index	Estimated Age
May 24/75						
63	71.1	5.70	210	Mature	3.68	3
65	74.8	6.70	410	Mature	6.12	3
	(73.0)	(6.20)				
Mat 25/75						
69	73.3	6.81	345	Mature	5.07	4
70	74.2	6.75	180	"	2.67	4
71	72.5	5.50	130	"	2.36	4
72	68.7	4.70	174	"	3.70	4
73	74.2	5.85	237	"	4.05	5
74	76.2	6.34	123	"	1.94	4
	(73.2)	(5.99)				
May 26/75						
75	77.0	7.40	365	Mature	4.93	4
May 28/75						
88	79.1	6.65	100	Spent	1.50	4
89	72.1	5.65	300	Mature	5.30	4
90	74.9	5.40	110	"	2.04	4
92	75.6	6.23	230	"	3.70	4
93	71.2	5.30	100	Spent	1.80	4
94	78.0	6.60	110	Mature	1.66	5
	(75.2)	(5.97)				
May 29/75						
95	71.4	5.10	127	Mature	2.49	4
96	73.2	5.80	224	"	3.86	4
97	75.4	5.93	290	"	4.89	-
98	75.0	5.80	200	"	3.45	4
99	77.0	5.40	18	Spent	0.33	4
	(74.4)	(5.60)				

Table 3. (cont'd.)

Date of Catch & Fish No.	Fork Length (cm)	Body Weight (kg)	Testes Weight (gm)	Gonadal Develop- ment	Gonado- somatic Index	Esti- mated Age
June 15/75						
103	72.1	5.85	326	Mature	5.57	4
June 16/75						
104	77.5	6.60	280	Mature	4.24	3
105	67.4	4.60	150	"	3.76	3
106	78.0 (74.3)	5.30 (5.50)	460	"	8.68	3

Table 4: Fecundity and size of milkfish eggs.

Reference	Sample Number	Gonado-somatic Index	Number of Eggs	Egg Diameter (mm)
1. Ovarian eggs in late stages of maturation:				
Sunier 1922			5,700	
Reijntjes 1923 ²			4,200	
Herre 1929 ²			3,060	
Adams et al 1932 ¹			3,000	
Tampi 1957	1	6.9	2,118	0.5 - 0.8
	2	8.6	3,433	0.5 - 0.7
	3	10.3	4,896	0.5 - 0.8
Liao 1971	2	11.6	3,180	0.9 x 1.0 1.3 x 1.5
	5	3.3	1,319	0.5 x 0.6
	8	24.9	3,288	1.1 x 1.2
Present study	62	7.7	2,113	0.7
Hamtik	8/4	4.2	3,717	0.6
	8/5	11.5	3,968	0.8
2. Eggs undergoing resorption:				
Tampi 1957				0.2
Present study	68			1.5
3. Fertilized eggs				
Delsman 1929				1.2

¹ Cited by Tampi 1957

² Cited by Schuster 1960

CAPTURE, TRANSPORT AND DOMESTICATION OF
ADULT MILKFISH, CHANOS CHANOS*

by

W.E. Vanstone, A.C. Villaluz, P.E. Bombeo, and R.B. Belicano**

Abstract

Methods used in the capture, transport and domestication of adult milkfish are described and illustrated.

Introduction

The difficulties of domestication of adult milkfish, Chanos chanos, and the importance of overcoming this obstacle before achieving large-scale spawning and fry production from captive animals, have been documented (Anon., 1974; Oceanic Foundation, 1975; and Schmittou, 1975). This problem was solved in Hawaii in 1975 (Madden et al., 1976 and has now been successfully overcome in the Philippines.

A description of our methods of capture, transport and domestication of adult milkfish, locally called sabalo, from the wild, forms the basis of this report.

Methods and Results

During the present fishing season, November, 1975 to June, 1976, four otoshi-ami were installed along the northwest coast of Antique Province (Kumagai et al., 1976). One of these was located adjacent to the laboratory site at Mag-aba; one at Bulanao, 10 km west of Mag-aba; one at Libertad, 17 km west of Mag-aba, and one at Bagaas, 4 km west of Libertad. In addition, two fish corrals were located at Hamtik, 135 km south of Mag-aba. The otoshi-ami and the type of fish corral installed at Hamtik have been described (Manacop, 1976).

*This work has been partially supported through a grant to SEAFDEC by the International Development Research Centre under Project No. 3-P-74-0146.

**Dr. Vanstone is a scientist with the SEAFDEC-IDRC Milkfish Project. Mr. Villaluz is a researcher and Messrs. Bombeo and Belicano are fisheries technicians of the SEAFDEC Aquaculture Department.

After capture in an otoshi-ami or fish corral (Plates 1, 2, 3 and 4) the sabalo were transferred by large dip net, into the holding-transport cage which was in an upright position (plates 5 and 6). The open draw-string end of the cage was then secured. The frame of the 10' x 5' (3.5m x 1.52m x 1.52m) holding-transport cages (plate 7) was constructed of 2" (5.08cm) PVC pipe and fittings with knotted 1/2" or 3/4" (1.27 or 1.9cm), stretch nylon netting and its buoyancy could be adjusted by filling it with water through a stoppered 2" (5.08cm), PVC Tee.

After the sabalo were in the cage, the cage was rotated 90° and lashed to the frame of the outrigger of the pumpboat (plate 6) and brought close to the shore at a speed of 1.5 knots. It was then hand-carried to water of approximately 50 cm deep.

When the sabalo catch was in the otoshi-ami at Mag-aba the cage was brought to the shore near the laboratory (plate 9). One team member then entered the cage carrying a 1.5 m long plastic bag made of 0.6 mm plastic tubing 81.3cm in circumference knotted at one end (plate 10). The plastic bag was then filled with water, the fish guided into it and the bag with its open end held securely closed, floated out of the cage on a hammock (plates 11 and 12). Two men then carried the hammock while a third man kept the open end of the bag securely closed (plates 13 and 14). The hammock was then placed beside the holding tank. The bag with fish and water was then lifted and passed to two men waiting inside the tank, who released the fish (plates 15, 16, 17, 18 and 19).

When the sabalo catch was at one of the other 3 sites, the fish was transferred from the holding-transport cage to a water-proof canvas tank on a pick-up truck (plate 20). A flow of oxygen was maintained in the tank during transport to Mag-aba, where the fish was placed in a plastic bag with water and transferred to the experimental or holding tank.

The holding facilities at the laboratory site consist of 2, 4 and 12 m diameter tanks 1.5 m deep and a lagoon of which about 0.3 ha have been fenced in. The tanks are modified backyard swimming pools fabricated from water-proof canvas with one 4" (10.16 cm), drain centered in the bottom and 2" (5.08 cm) outlets, 6" (15.24 cm) above the bottom, on each side of the tank.* The tanks were supplied with continuously flowing water and aeration. Water levels are maintained at 50 to 75 cm.

*Manufactured by Basic Indusia, Inc. Pasong Tamo St., Makati, Rizal.

Results

From 3 November 1975 to the present, 12 May 1976, a total of 259 sabalo were captured in the three otoshi-ami located at Bagaas, Libertad and Mag-aba and the two fish corrals at Hamtik. No sabalo were captured in the otoshi-ami located at Bulanao. The catch statistics together with mean weekly temperatures of the water from a depth of 5m at the Mag-aba otoshi-ami are illustrated in Fig. 1. Salinities of the water at 5m at Mag-aba ranged between 33.2 and 35^o/oo from 1 December 1975 to 12 May 1976.

While sabalo were captured occasionally between November and March they were not captured in large numbers until early April at which time water temperatures rose above 26.5^oC. This is in agreement with the findings of Kuronuma and Yamashita (1962) who showed that milkfish fry occurred in the coastal waters of Eastern Vietnam only after the water temperature rose above 27^oC.

Of the 259 captured sabalo, 100 were released from the otoshi-ami and 18 died during capture (Table 1). Fifteen fish were badly injured and descaled during capture, four of these fish died during truck transport and nine died later during domestication. No fish were lost during cage transport to the shoreline. During the catch at Bagaas on 6 May, 23 fish were placed in the cage and transported in excellent condition to the shore. On being carried into the shallow shore waters the approaching cage was greeted by 100-125 cheering children and the fish became excited. The children were fascinated by the excited fish and commenced rhythmic shouting; the fish were examined and placed in the holding tank. Two of these fish had broken jaws, one had a ripped operculum, two had deep gashes in the top of their heads and ten were badly descaled. Thirteen of these 23 fish died within three days of capture.

During the first truck transport on 27 March from Hamtik, diluted sea water (20^o/oo chilled to 18^oC was used. The two fish being transported died within half an hour of loading into the tank on the truck. On the next fish transport from Hamtik on 4 April, diluted sea water (25^o/oo) chilled to 24^oC was used and the transport was successful. Diluted sea water (18-26^o/oo) with no chilling was used in all truck transports until 15 April. The temperature of the water ranged between 29 and 32^oC and there were no mortalities. Non-diluted sea water (34^o/oo) at ambient temperatures (29-32^oC) was used in the truck transport on 22 April and all subsequent transports. Three fish were lost during the truck transport from Bagaas on 9 May due to a malfunction in the oxygen regulator.

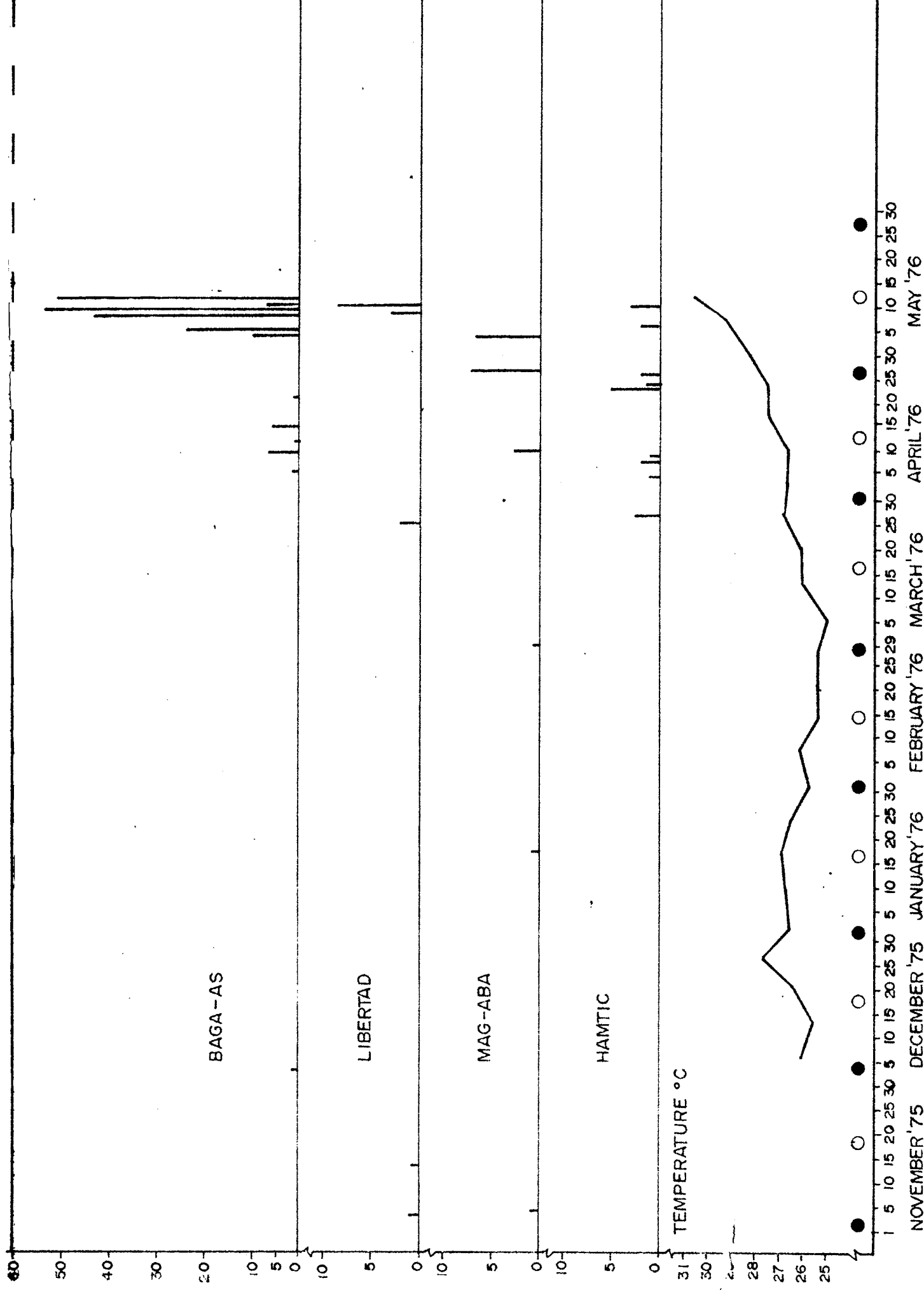


Figure 1. Adult milkfish captured from Otoshi-ami and two fish corals in Antique Province November 1975 to May 1976.

Table 1. Balance sheet of the 259 adult milkfish captured in the three otoshi-ami located at Bagaas, Libertad and Magaba and two fish corrals located at Hamtik, Antique, from 3 November 1975 to 12 May 1976.

	Number of fish		
	Released or still alive in which died	Released or still alive in tanks	Experimental fish transferred to lagoon
Died during capture	18	-	-
Released to sea immediately after capture	-	100	-
Died during transportation in truck	9	-	-
Badly injured during capture and cage transport	22	12	-
Domestication experiments - tanks	1	13	21
Domestication experiments - lagoon	10*	-	44 - 10
Spawning experiments	6	4	6
Domesticated fish released to sea by tagging team	-	3	-
			71 - 10
	66	132	61

*Specific origin of these 10 dead fish unknown - refer to text.

The first live sabalo was obtained on 4 November 1975 and due to lack of facilities at that time it was released into the enclosed portion of the lagoon. This occurred during the rainy season at which time the salinity of the top 50 cm was 32-34^o/oo. This halocline was maintained throughout all tidal changes. Salinity of the lagoon had been previously determined daily for a week in late June, 1975. At that time there had been heavy rainfall and the salinity of the top 50-75 cm ranged from 5 to 14^o/oo while the salinity of the water below this ranged from 26 to 32^o/oo. This fish escaped from the lagoon in mid-March. The fish captured on 7 January 1976 was placed in a tank containing sea water, (34^o/oo). This fish developed the typical opaqueness of the translucent adipose eyecover (referred to by Schuster [1960] as eyelid) and died 11 January. Twenty-three of the 35 fish captured after this date for domestication in the holding tanks, were maintained in diluted sea water, 18-22^o/oo, for 10-16 days at which time the salinity was increased to 34^o/oo. The adipose eyecover of these fish became opaque 2-3 days after capture, but this opaqueness disappeared after 9 days in captivity. The fish which died during domestication by this method were those injured during capture and cage transport as discussed above. Twenty-one of the 35 fish intended for domestication were captured on 4 and 6 May and were placed in the domestication tanks until 8 May. These 21 fish were then transferred to the lagoon and their fate is not known, as discussed below.

As soon as their eyecover cleared the tank-domesticated fish were observed to actively feed on diatoms and protozoans forming a scum on the surface of the water. Live zooplankton collected from the lagoon was then added to the tank once daily. These feeds were supplemented 3 weeks after capture with a twice daily feeding of a mixture of chopped fish and rice bran (300 gm rice bran/kilo chopped fish).

During April, three fish which had been domesticated in the tanks were donated to a tagging team for their use. Due to shortage of tank space, and because of the survival in the lagoon of the fish captured on 4 November, 6 fish from the spawning experiments of 4 and 6 May, 21 fish which had been in 20^o/oo water since 4 and 6 May and 44 newly captured sabalo, which were believed to be spawned out females, were placed in the enclosed lagoon between 6 and 12 of May. Ten of these fish died between 9 and 12 of May and more have died since. It was determined on 12 May that the water salinity of the lagoon ranged from 29^o/oo at the surface to 33^o/oo at the bottom and no halocline was present. The unfortunate loss of these fish in the lagoon reconfirms the findings of investigators in Hawaii and those involved in the present study that brackishwater (15-22^o/oo) is necessary for survival of adult milkfish during their first 10 to 20 days in captivity.

Between 15 April and 6 May, 16 fish were used in the spawning experiments reported elsewhere (Vanstone et al., 1976). Six of these fish died or were killed during experimentation and 4 were returned to the holding tanks where they were successfully domesticated and 6 were released in the lagoon.

Acknowledgment

The success of this and other milkfish studies conducted at Mag-aba, Pandan, Antique, could not have been achieved without the cooperation of Atty. Enrique Zaldivar and Mr. Rene Lorenzana, operators of the four otoshi-ami. We are indebted to these gentlemen and also to their fishermen for their assistance.

The dedication and untiring assistance of our colleagues at Pandan Station and at the Hamtik Sub-Station are acknowledged.

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Explanation of Plates

- Plates 1 - 4 : Capture of sabalo in the otoshi-ami.
- Plates 5 and 6 : Transfer of sabalo from the otoshi-ami to the transport cage.
- Plate 7 : The sabalo transport cage.
- Plate 8 : Attachment of transport cage with sabalo to outrigger of pumpboat.
- Plates 9 - 19 : Transfer of sabalo from transport cage at the shore to the holding tanks.
- Plate 20 : Transport tank on to pickup truck used for transporting sabalo long distance by road.

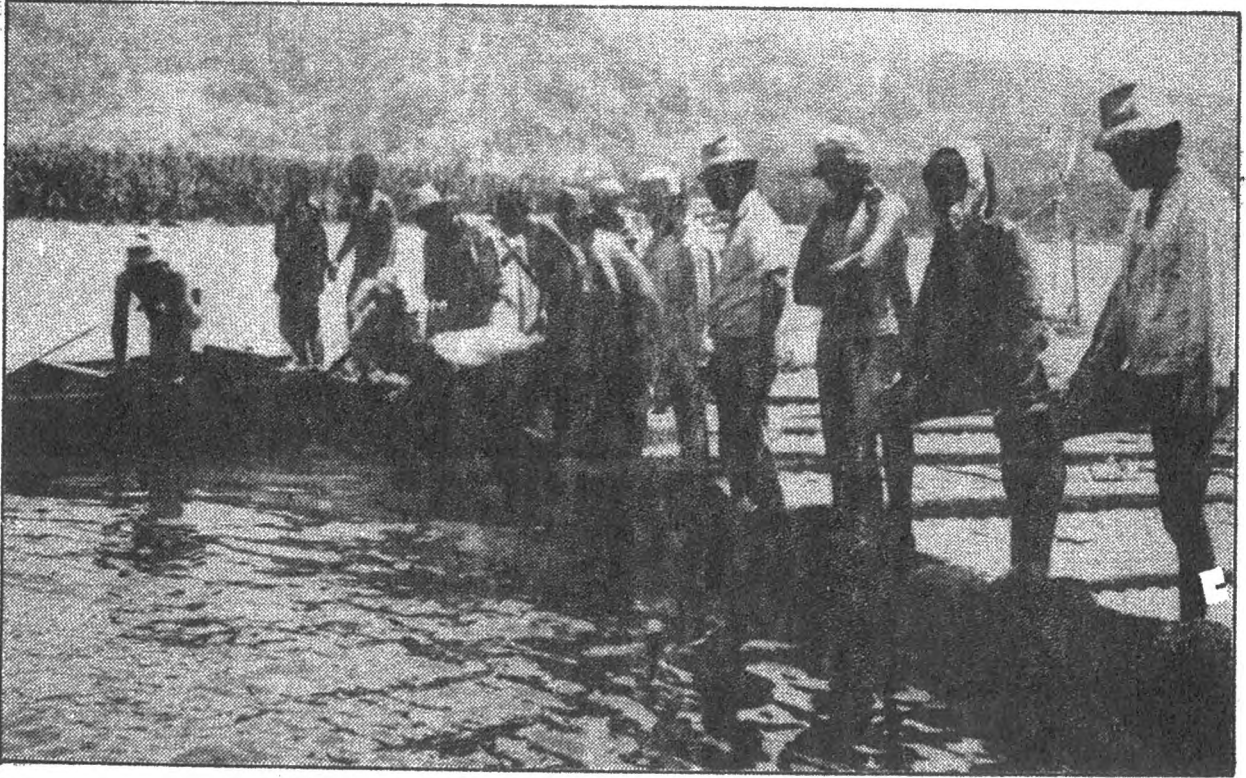


Plate 1

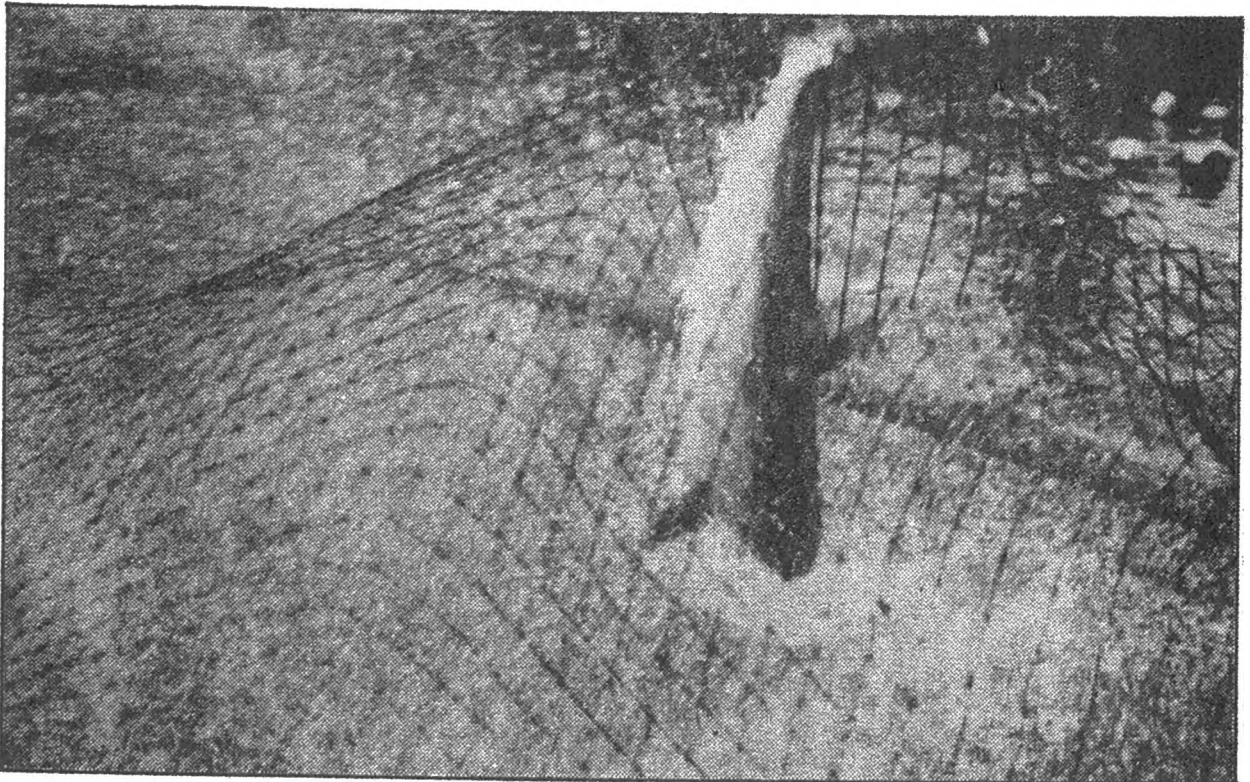


Plate 2

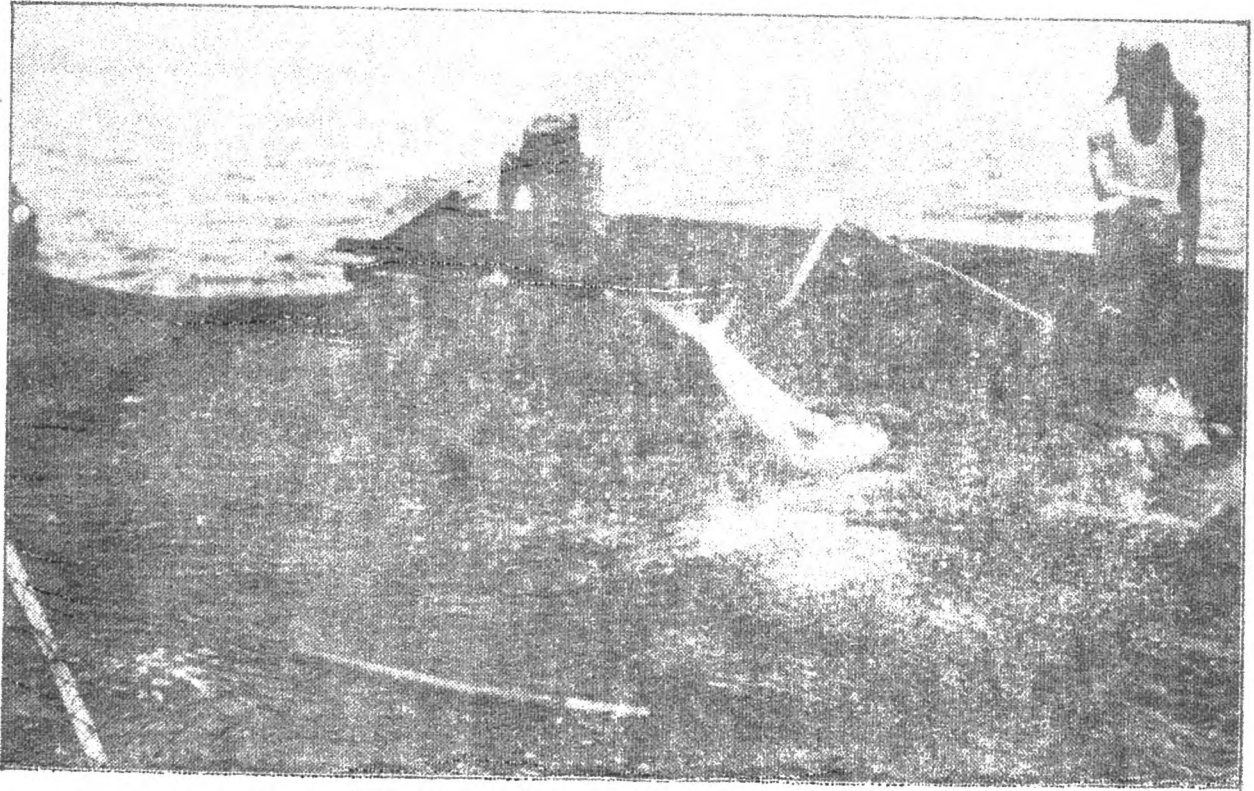


Plate 3

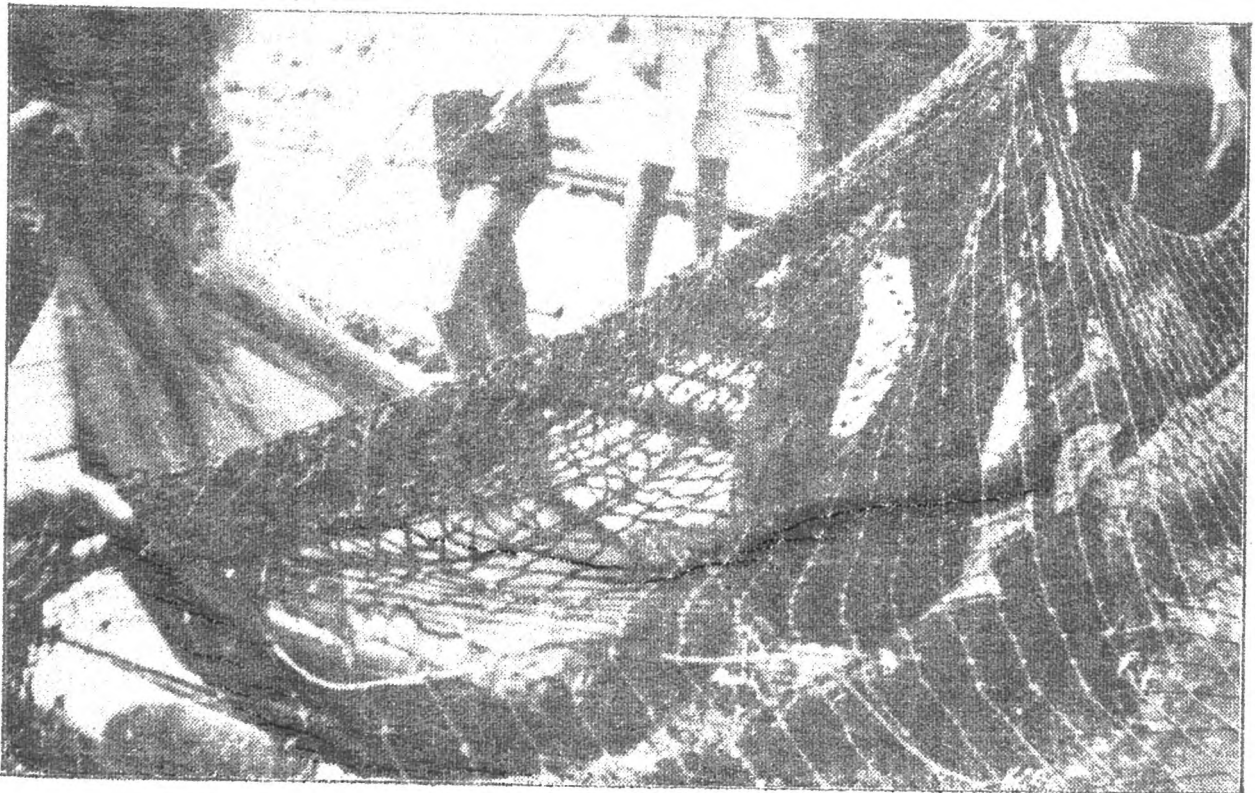


Plate 4

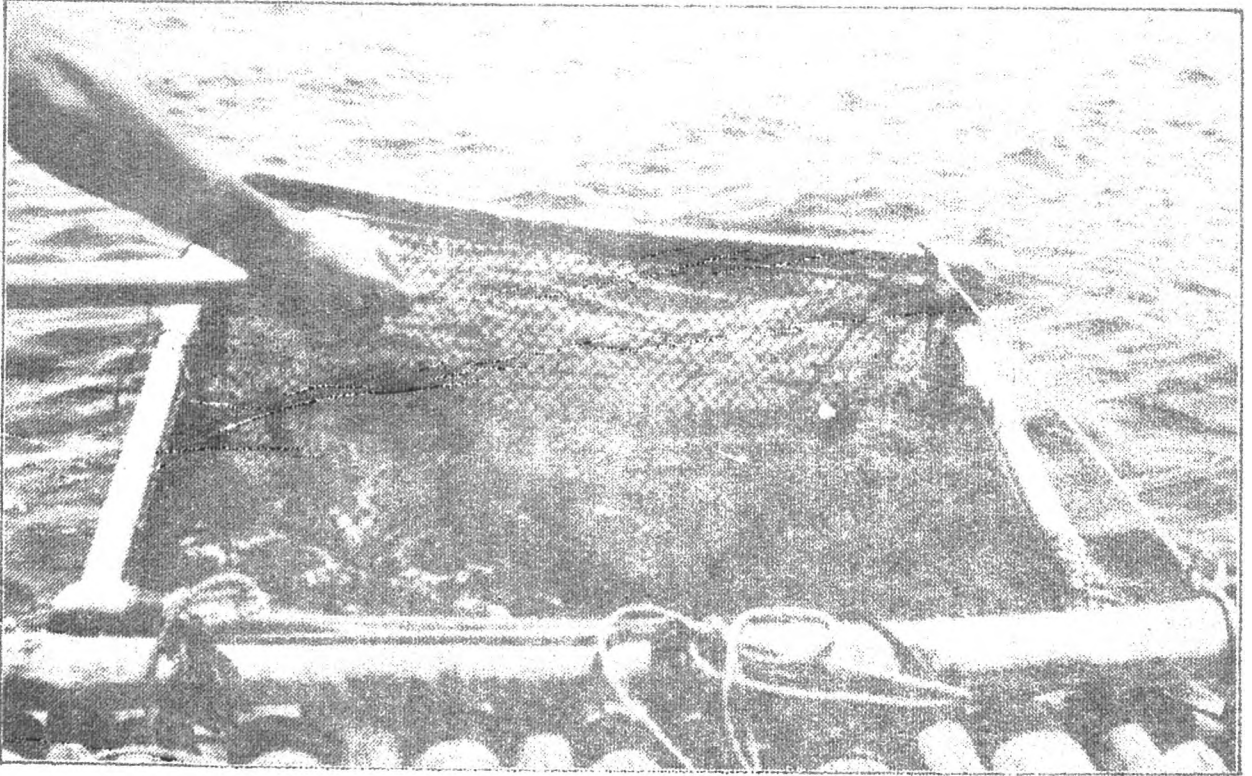


Plate 5



Plate 6

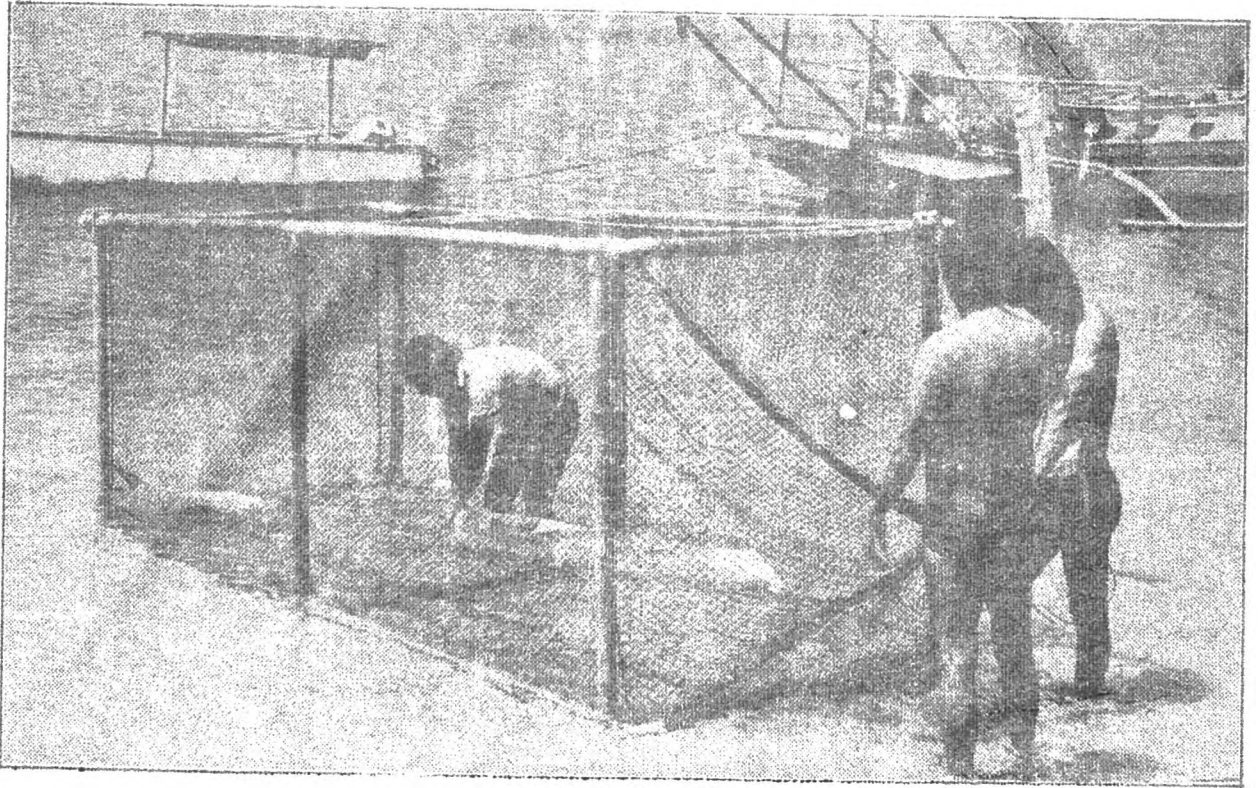


Plate 7

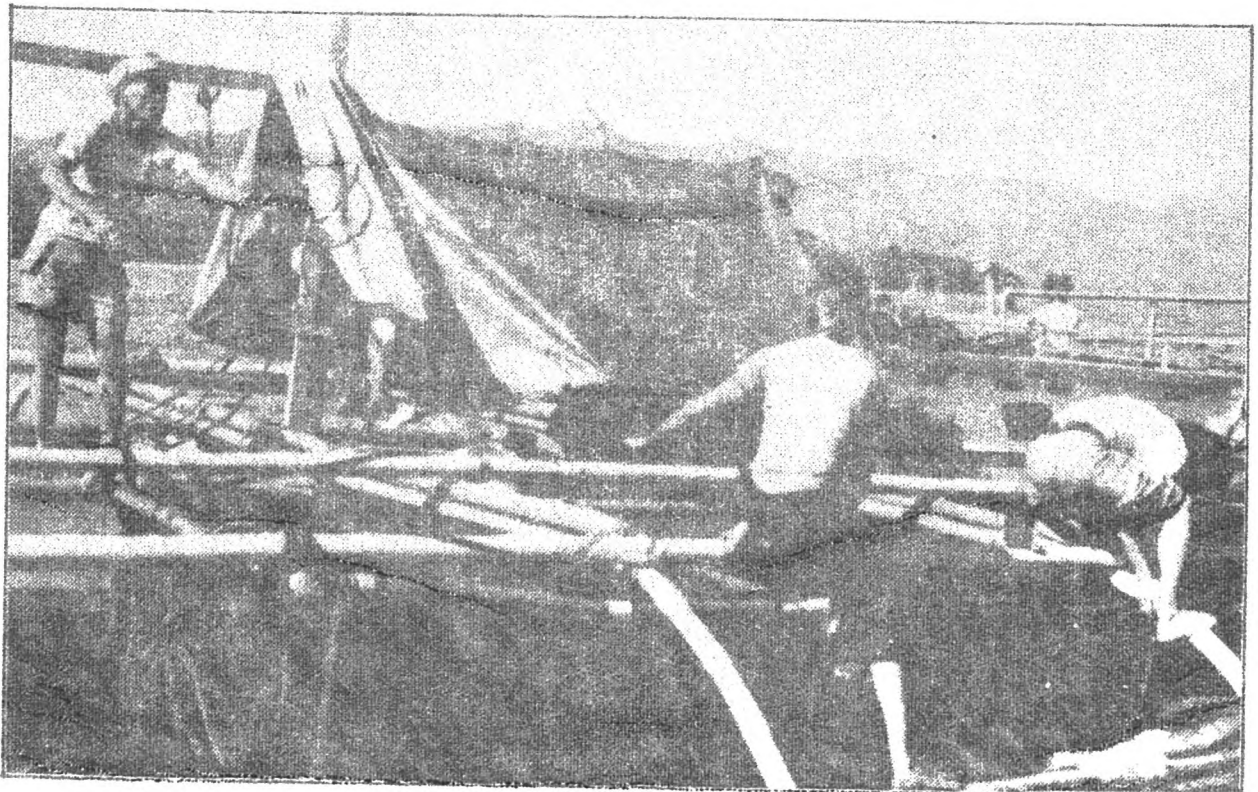


Plate 8

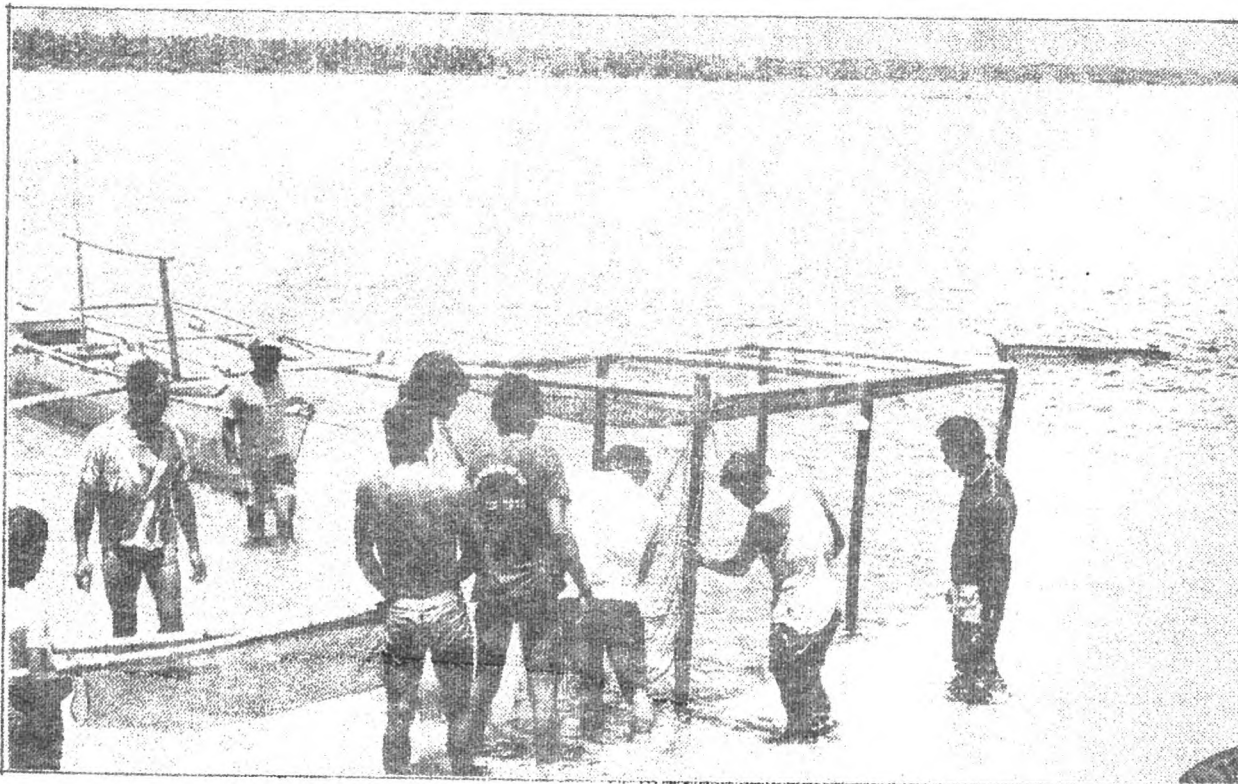


Plate 9

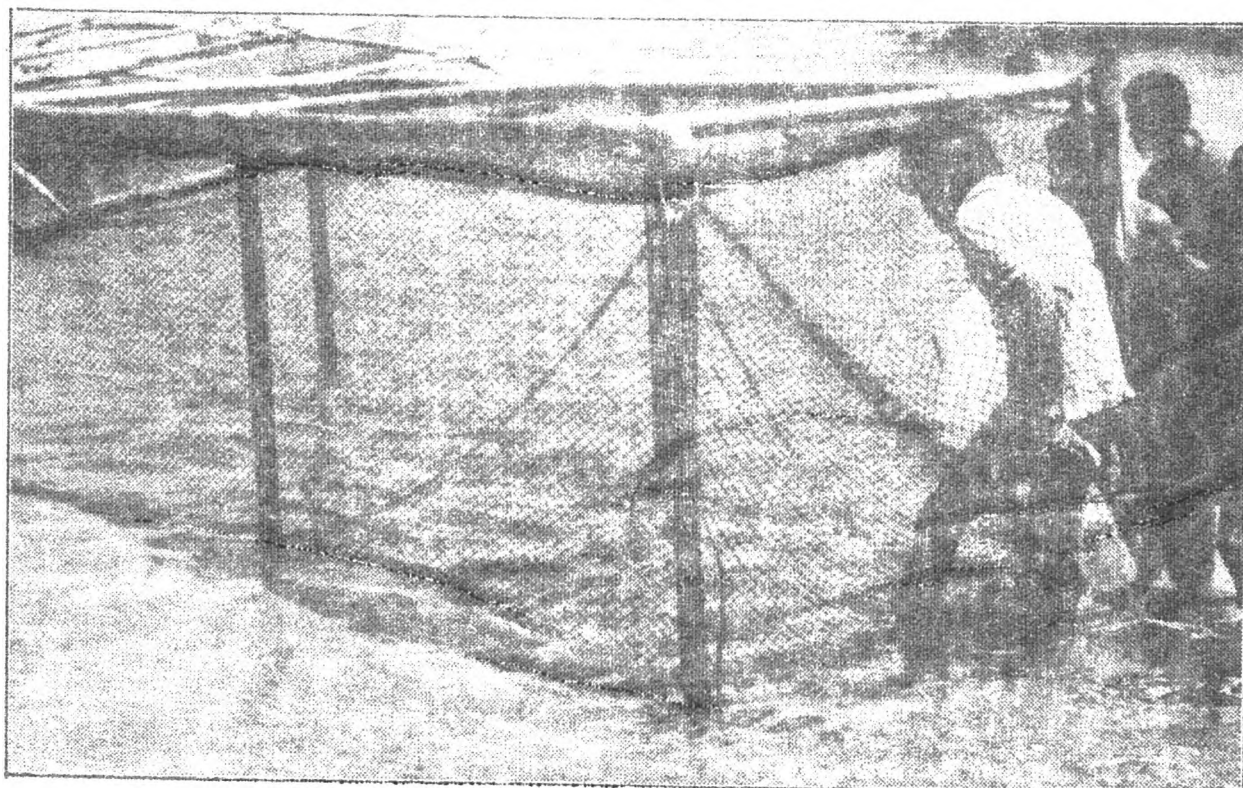


Plate 10



Plate 11



Plate 12

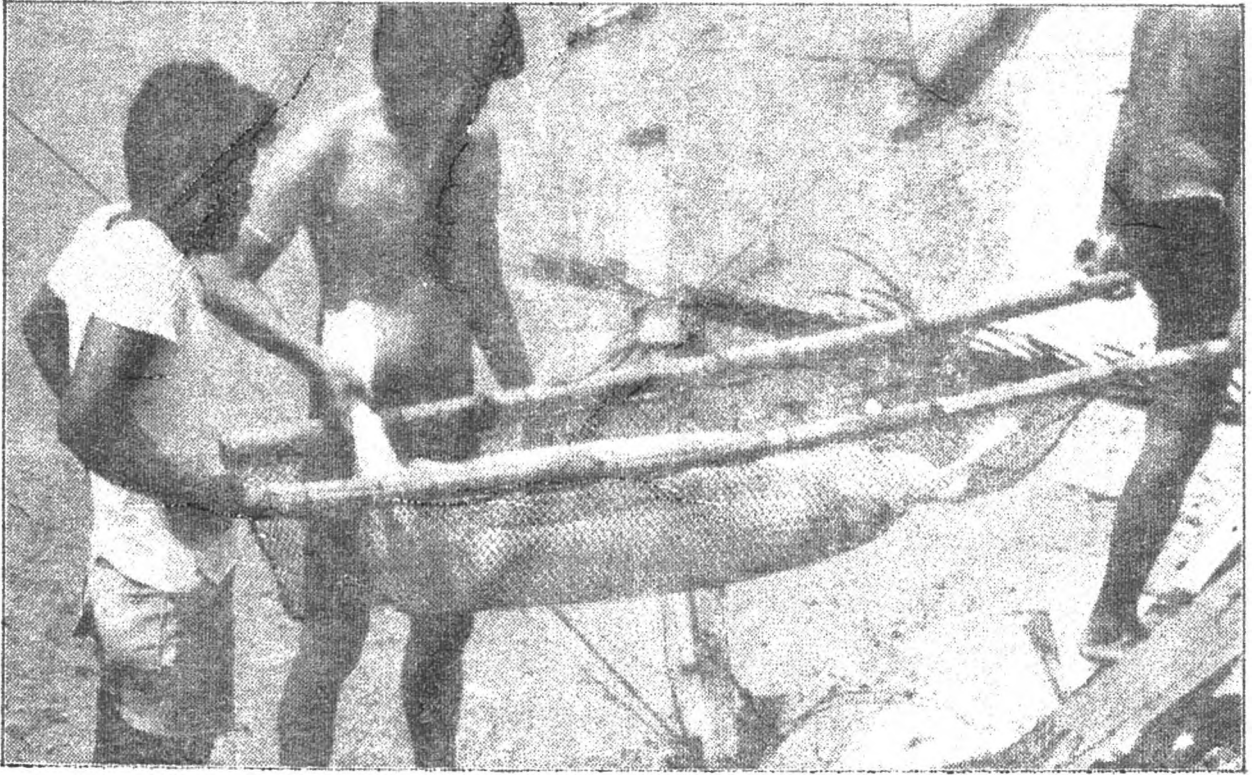


Plate 13



Plate 14



Plate 15

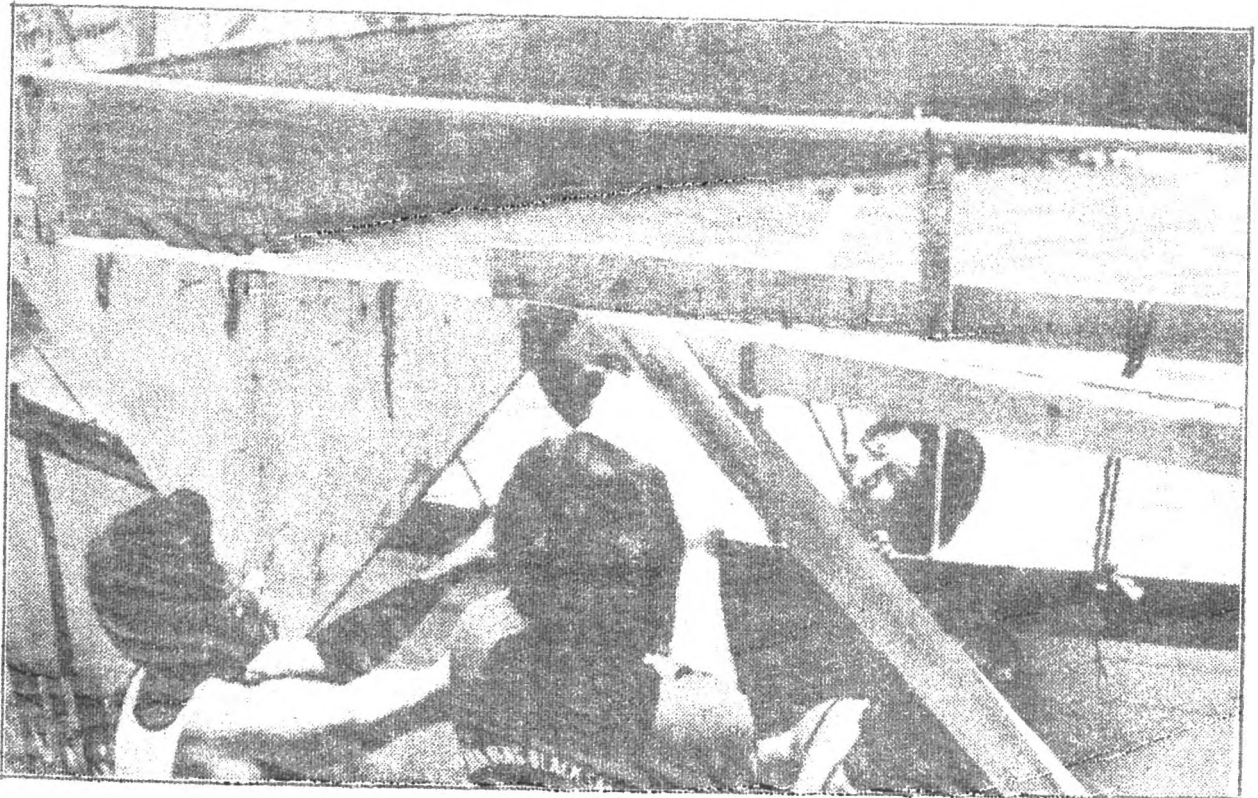


Plate 16

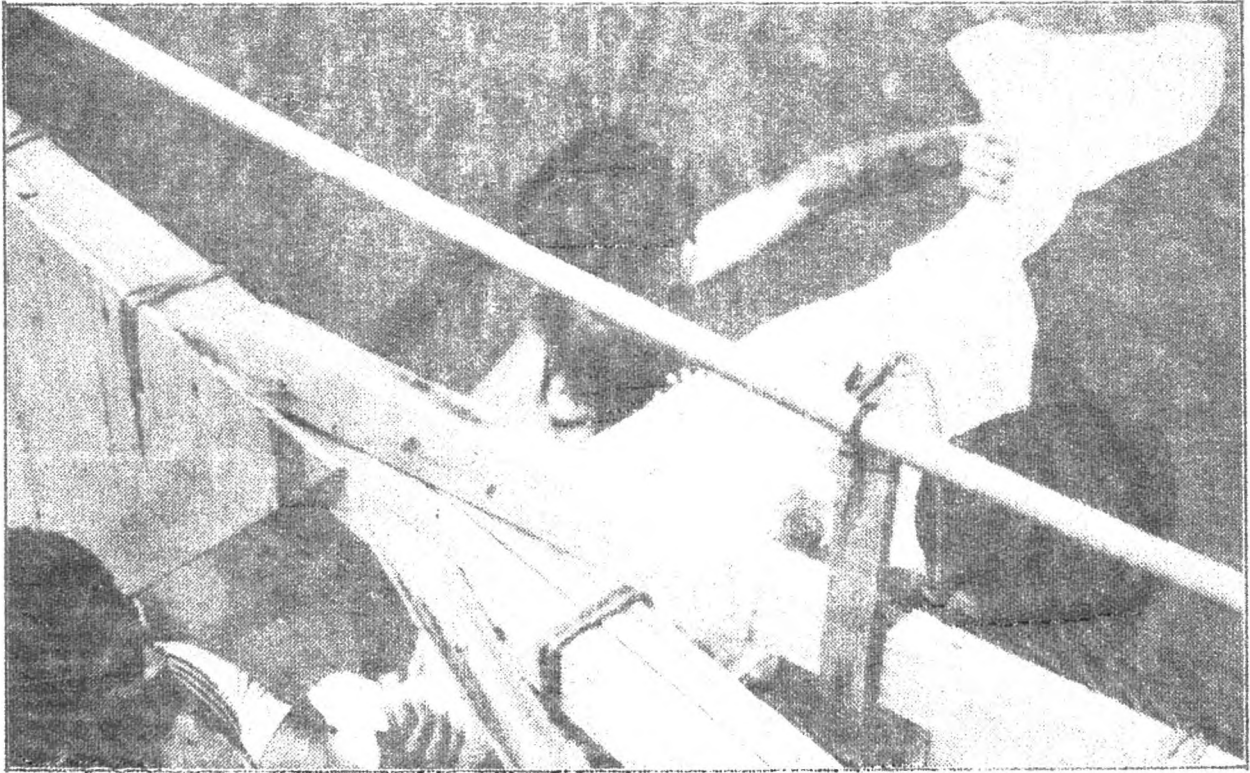


Plate 17

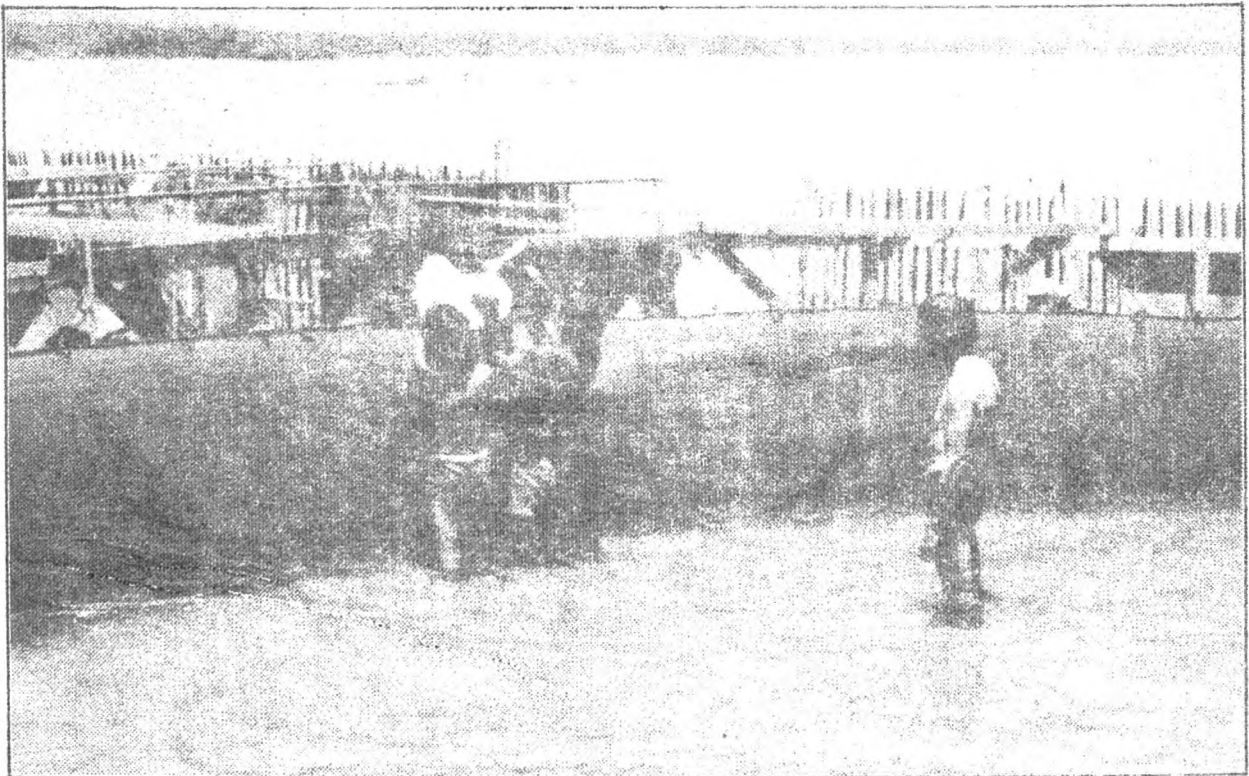


Plate 18



Plate 19



Plate 20

SPAWNING OF MILKFISH, CHANOS CHANOS,
IN CAPTIVITY*

by

W.E. Vanstone, A.C. Villaluz, and L.B. Tiro, Jr.**

Abstract

Newly captured milkfish released 0.8 mm in diameter non-hydrated eggs spontaneously in captivity. After injection with partially purified salmon gonadotropin (SG-G100), 1.2 mm in diameter hydrated eggs were released. These eggs, however, were not fertilized.

Introduction

The major initial objective of the SEAFDEC-IDRC Milkfish (Chanos chanos) Research Project is to ensure an adequate and dependable supply of milkfish seed, in addition to those collected from natural sources, and to extend and stabilize the period of fry availability throughout the year. This is to be mainly accomplished by breeding milkfish in captivity and by raising the fry from the egg. A major obstacle to this objective was the difficulty in capturing and domesticating adult milkfish captured in the wild. This problem has now been largely solved (Madden et al., 1976; Vanstone et al., 1976) and investigators can now concentrate on developing techniques designed to control the reproductive cycle of this species.

The present report is based on three spawning experiments that have been completed to date, 12 May 1976, during the present fishing season, November to June, 1975 along the Antique Coast of Panay Island, Central Philippines.

*This work has been partially supported through a grant to SEAFDEC by the International Development Research Centre under Project No. 3-P-76-0166.

**Dr. Vanstone is a scientist with the SEAFDEC-IDRC Milkfish Project. Mr. Villaluz is a researcher and Mr. Tiro is a research aide of the SEAFDEC Aquaculture Department.

Materials, Methods and Results

Methods of capture, transport and domestication of adult milkfish or sabalo together with catch statistics at each of the five capture sites have been reported elsewhere (Vanstone *et al.*, 1976). On 6 April 1976, a few non-hydrated eggs were found in the screened outflow from one of the 12 m diameter holding tanks which at that time contained three sabalo in diluted sea water of 20 ‰ salinity with a daily temperature range of 28 to 29°C. These almost spherical eggs had a mean diameter of 0.89 mm (range 0.80-1.07 mm) and were filled with granulated yolk but no oil globules (Plate 1). The salinity of the water in the tank was then increased to 34 ‰ over the next 12 hours. No further eggs were observed.

Experiment No. 1:

Eight fish captured at Bagaas on 9 (Fish 9/4), 12 (Fish 12/4) and 15 April (Fish 15/4A-F) were distributed equally between two 4 m diameter x 1.5 m high experimental tanks containing 60-70 cm of the 20 ‰ diluted sea water. At 1630 hours on 15 April, non-hydrated eggs identical to those obtained on 6 April, were discovered in one of the tanks and at 1700 hours semi-purified chum salmon gonadotropin (SG-G100, prepared by B.C. Research Council, Vancouver, Canada and assayed on 1 July 1974) was injected into the dorsal musculature 2-3 cm below the dorsal fin in each of the eight fish. Each large fish, weighing approximately 7 to 8 kg, received 3 ml of 1.0% NaCl solution containing 10 mg SG-G100/ml and each smaller fish of approximately 6 to 7 kg received 2 ml. Following injection all fish were placed in a single 4 m diameter tank. The fresh water supply was then turned off and the salinity increased to 34 ‰ over the next eight hours. Beginning at approximately 1900 hours the fish became very active, swimming very fast, and jumping and circling each other. At 2020 hours on 15 April 1976, a sexually mature male (No. 15/4D weighing 6.0 kg) with freely flowing milt killed itself by jumping and hitting its head on horizontal bamboo poles located 50 cm above the top of the tank. At 2130 hours a female (No. 15/4A weighing 6.7 kg) with gonads at late stage III, killed itself in the same manner.

At 0830 hours on 16 April the abdomens of fish (15/4/E and F) were observed to be considerably enlarged and at 0945 hours hydrated eggs were observed in the tank. These eggs ranging in diameter from 1.07 to 1.36 mm and with a mean diameter of 1.18 mm (Plate 2) were spherical, without oil globules and contained yellow granulated yolk. Fish No. 15/4F died at 1600 hours. This fish

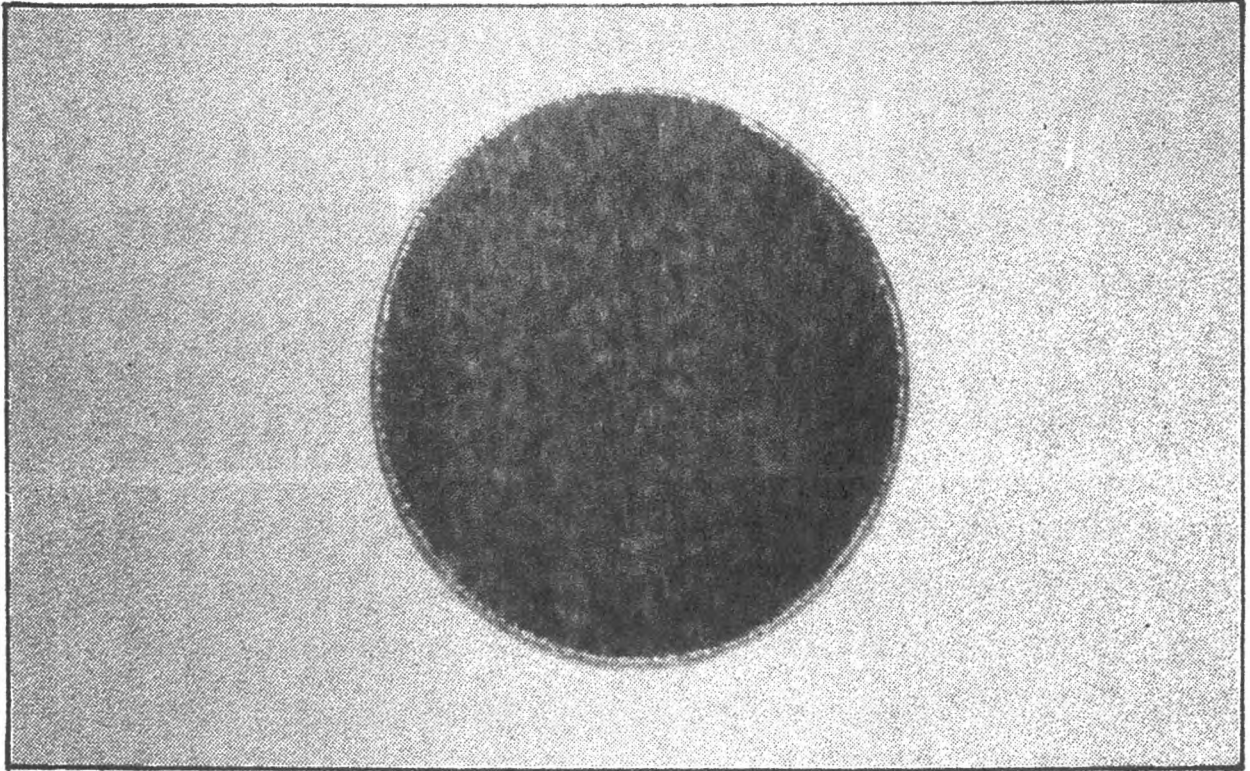


Plate 1. Non-hydrated milkfish egg.

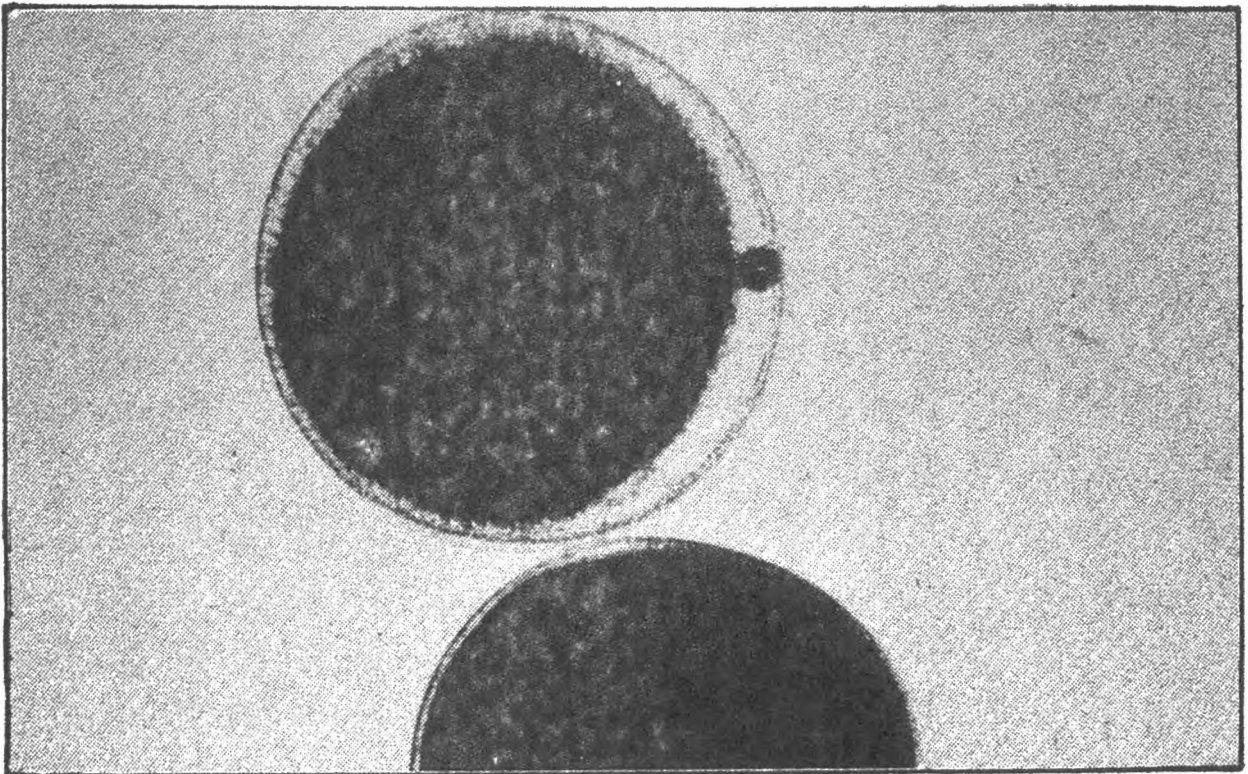


Plate 2. Hydrated milkfish eggs.

weighed 8.5 kg and its ovaries weighing 631 g were at late III state of development. At 1700 hours fish No. 15/4E appeared to be dying and was transferred to a second 4 m diameter tank where she died at 1900 hours. This fish weighed 7.8 kg and was partially spawned. At 1830 hours the four remaining fish in the experimental tank were again injected with 2 or 3 ml SG-G100.

On 18 April at 0900 hours hydrated eggs, ranging in diameter from 1.05 to 1.18 mm (mean - 1.11 mm), were found in the experimental tank. The four fish were then examined but were kept alive because of our desire to build up a brood stock for the 1977 spawning season. Fish numbers 9/4A, 12/4 and 15/4B were assessed to be sexually immature males or females and number 15/4/C was a spawned female. These four fish were transferred to the domestication tank.

Experiment No. 2:

During transport of the sabalo catch from Bagaas on 4 May, it was observed that fish number 4/5A, which had received several bad head wounds during capture, released non-hydrated eggs in the transfer cage at 1430 hours and also in the tank on the truck at 1530 hours. This fish together with two mature males, number 4/5B and C captured at the same time, was placed in a 2 m diameter experimental tank containing water at 34 ‰ salinity and 30°C at 1600 hours. The female fish was injected with 5 ml and the males with 2 ml SG-G100 solution at 1600 hours and non-hydrated eggs were released in the tank at 2000 hours. At 2330 hours the same day hydrated but unfertilized eggs were discovered in the tank.

This fish did not spawn again and died on 6 May. When examined, it was found that the fish was completely spent.

Experiment No. 3:

One mature male (No. HAM 6/5B) was obtained from Hamtik and two mature males (BAG 6/5N and T) together with a female (BAG 6/5S) which was believed to be at ovarian stage III were captured at Bagaas on 6 May. The males were injected with 1 ml and the female with 3 ml of SG-G100 solution when transferred from the truck to a 12 m diameter holding tank containing sea water at 34 ‰ at 30°C. The salinity of the water in the tank was lowered to 25 ‰ over the next 12 hours. On 7 May another sexually mature male (No. 7/5) captured at Bagaas, was injected with 1 ml SG-G100 solution, and placed in the same tank. The female fish was injected with 5 ml

SG-G100 solution, and placed in the same tank. The female fish was injected with 5 ml SG-G100 solution on 8 May at 1600 hours. On the possibility of obtaining ovulated eggs the female fish was killed at 1100 hours on 9 May. On examination it was discovered that the ovary contained a uniformly distributed mixture of I, II and early III stages of ova (Plate 3).

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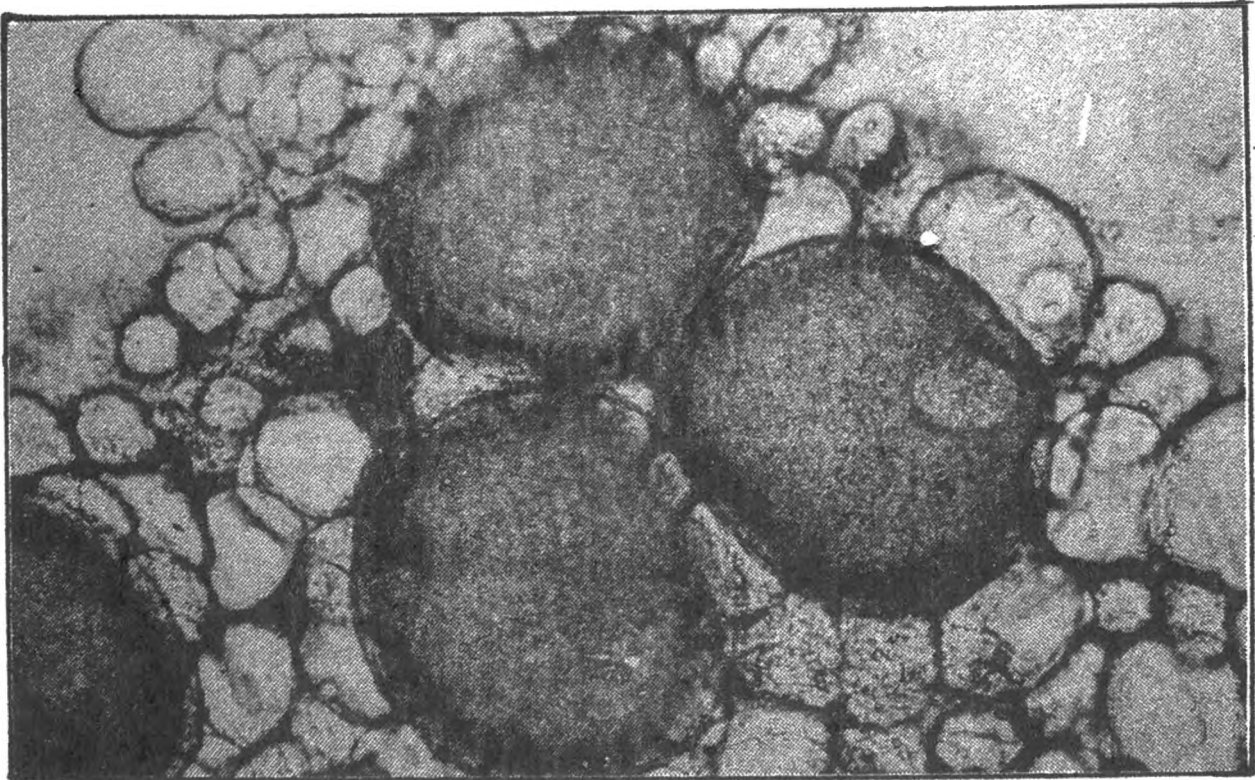


Plate 3. Ovarian tissue from sexually developing milkfish containing a mixture of I, II, and early III stage ova.

QUALITATIVE ANALYSIS OF THE CONTENTS
OF THE ANTERIOR PORTION OF THE OESOPHAGUS
FROM ADULT MILKFISH, CHANOS CHANOS,
CAPTURED IN PANDAN BAY FROM 10 MAY-JUNE 1975*

by

A. C. Villaluz, L. B. Tiro
L. M. Ver and W. E. Vanstone**

Abstract

Qualitative analysis of food items in the anterior spiral portion of the oesophagus suggests that adult milkfish feed on both benthic and planktonic materials.

Introduction

The little that is known of the food of milkfish in the wild has been reported by Sunier (1922), Reijntjes and Schuster (unpublished report cited by Schuster, 1949) and Schuster (1949) in Java and by Chacko (1949) in the Gulf of Mannar and Tampi (1953) also in Southern India. Others are of the opinion that milkfish in the sea are benthic feeders. While Chacko concluded that the milkfish are plankton feeders, Schuster (1960) has suggested that milkfish are facultative feeders.

Bridge and Boulanger (1910) has described the alimentary tract of milkfish, Chanos chanos, as follows: "The oesophagus, before making a real U-shaped bend in the gizzard section, follows a complete S-shaped curve. The interior surface of this part of the oesophagus displays parallel spiral folds with a great number of backward directed papillae. The lower part of the oesophagus is equipped with longitudinal folds, dwindling at the beginning of the gizzard, the so-called 'stomach' of Chanos."

*This work has been partially supported through a grant to SEAFDEC by the International Development Research Centre under Project No. 3-P-74-0146.

*Mr. Villaluz is a researcher, while Messrs. Tiro and Ver are research aide and fisheries technician, respectively, of the SEAFDEC Aquaculture Department and Dr. W. E. Vanstone is a Scientist with SEAFDEC-IDRC Milkfish Project.

A qualitative analysis of the contents of the anterior spiral portion of the oesophagus from 96 of the adult milkfish captured in Pandan Bay between 10 May and 16 June, 1975, as reported by Tiro et al. 1976. forms the basis of this report.

Materials and Methods

The methods used in obtaining and processing adult milkfish captured in the otoshi-ami located at Mag-aba, Pandan during the 1974-75 fishing season has been reported by Tiro et al 1976. Following dissection of these fish the spiral portion of the oesophagus was preserved in 10% seawater formalin and the contents analyzed at a later date. For analysis the spiral portion of the oesophagus was cut lengthwise and the contents rinsed into a specimen bottle with distilled water. After settling, the food particles were removed by pipet and placed on a microscope slide where tips of the papillae, which had been cut from the oesophagus, were removed. The particles were then covered with a cover glass and examined. The different food items found in the oesophagus were then identified (Esguerra, 1951 Raymont, 1963 Dawson, 1966 Shirota, 1966 Yamaji, 1966 and Barnes, 1968) and classified into four groups: diatoms, animal forms, algae and other items.

Results and Discussion

The contents of the oesophagus from the 96 specimens examined, consisted of both benthic and planktonic materials as summarized in Table I.

Table I. Oesophagus contents from 96 adult milkfish captured in Pandan Bay from 10 May-16 June 1975.

Food Items	Number of specimens containing each item	Percentage of specimens containing each item
<u>Diatoms</u>		
<u>Nitzchia</u> , <u>Thalassiothrix</u> , <u>Fragillaria</u> , <u>Rhizosolenia</u> , <u>Coscinodiscus</u> , <u>Deratium</u> , <u>Navicula</u> , <u>Chaetoceros</u> , <u>Eucampia</u> , <u>Ditylum</u> , <u>Melosira</u> , <u>Biddulphia</u>		
<u>Animal Forms</u>		
Vorticellid	1	1.0
Sarcodinan	1	1.0
Foraminifiran	1	1.0
Ciliophoran	2	2.1
Echinoderm larvae	1	1.0
Cladoceran	1	1.0
Copepods	83	86.5
Crustacean eggs and egg cases	9	9.2
Crustacean nauplii	2	2.1
Crustacean zoea	4	4.2
Crustacean juvenile	1	1.0
Crustacean shell and appendages	4	4.2
Fish eggs	14	14.6
Fish larvae	5	5.2
<u>Algae</u>		
Chlorophytes	39	40.6
Cyanophytes	37	38.5
Rhodophytes	28	29.0
<u>Other Items</u>		
Detritus	70	72.9
Sand	1	1.0

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PRELIMINARY OBSERVATION ON THE NUMBER OF VERTEBRAE
IN MILKFISH, Chanos chanos (Forsk.)

by

Tetsushi Senta-and Shigeru Kumagai*

Abstract

The existence of races and/or subpopulation in milkfish, a widely distributed species, is suggested by many workers. Comparison of mean numbers of vertebrae is one of the useful method of distinguishing subpopulations. The present paper reports the results of preliminary studies made on the vertebral counts of milkfish fry collected from several areas. The results obtained, though not conclusive, suggest the scope for further study in the line.

Introduction

The milkfish has a wide distribution extending from the east coast of Africa through the Southeast Asian waters to the Southwest coast of United States and Mexico. The existence of subpopulations in such a widely distributed species cannot be ruled out. It is also reported that in the Southern Philippines, as well as in Indonesia, there are two peaks in fry season while in the other regions, milkfish has only one spawning season. These information suggest the possibility of having more than one race in milkfish.

Since comparison of the mean numbers of vertebrae is often useful in distinguishing subpopulations, the present study was taken up with a view to determine subpopulation, if any. The data accumulated so far are very limited. The authors, however, intend to present this paper to the Milkfish Workshop Conference to stimulate interest in workers on this important aspect of milkfish study.

Materials and Methods

As a preparatory step, study was made to find out the progress of oosification in the vertebrae of different sizes of milkfish larvae.

*Dr. Senta is a technical adviser to SEAFDEC Aquaculture Department, sent by Japan International Cooperation Agency (JICA) for a period of three months, from March 15 to June 14, 1976, and Mr. Kumagai is a researcher of the SEAFDEC Aquaculture Department.

A few larvae collected at Pandan, Antique Province, Philippines, during 1975 were stained with alizarin red. It was observed that the calcified vertebrae alone had taken the dye.

In a larva of 8.6 mm in total length, only the dorsal half of the urostyle (the last vertebra) was well dyed, and the third to the 18th vertebrae were only partially dyed while the rest did not take the dye at all. Calcification of vertebrae, however, has progressed in a larva of 10.2 mm in total length, and all vertebrae excepting three anterior to the preceding urostyle were well dyed. In larvae of 11 mm in total length, all the vertebrae took the dye well and the number of vertebrae could be counted accurately, although the neural and hemal spines did not appear on the centrum of this stage. Neural and hemal spines start developing in about 15 mm long larvae, at first on the posterior vertebrae, and advance anteriorly as the fish grows.

The above findings confirmed that the dye could be effectively used for the study since total length of larvae caught by fry collectors in the beach, usually range from 10 to 15 mm and are old enough for the study of vertebral number.

The materials for the study were collected from three different stations: Station I: 188 fry (11.2-14.0 mm in total length) were caught by a fry collector along Tigbauan beach on the southern coast of Panay Island, Philippines, on April 21, 1976; Station II: 279 fry (10.1-15.0 mm in total length) were collected by the staff of Hamtik Substation, SEAFDEC Aquaculture Department, in Hamtik shoreline on the southwest coast of Panay Island, during March 29 to April 24, 1976 and Station III: 41 fry (15.0-28.0 mm in total length) were obtained from Taiwan through the courtesy of Dr. I-Chiu Liao of Tungking Marine Laboratory, Taiwan Fisheries Research Institute, on March 30, 1976.

The fry were dyed with alizarin red, and made transparent by following Hollister's method (Hollister, 1934).

Results and Discussion

Table 1 summarizes the results of the study giving the number of vertebrae present in specimens collected from three different stations together with the mean and some other statistics.

Table 1. Number of vertebrae present in milkfish fry collected from three different stations.

<u>Number of vertebrae</u>	<u>Stations</u>		
	<u>Tigbauan</u> (I)	<u>Hamtik</u> (II)	<u>Taiwan</u> (III)
40	--	1	--
41	--	4	--
42	2	3	2
43	100	145	19
44	86	119	20
45	--	7	--
Total number of specimens	188	279	41
Mean of vertebral number	43.447	43.427	43.439
Variance	0.2684	0.4465	0.3525
Standard deviation	0.5181	0.6682	0.5937
Standard error	0.0378	0.0400	0.0927

The majority of the specimens from each station had either 43 or 44 vertebrae. The highest value of the mean of the vertebral number was 43.447, observed in the Tigbauan specimens, and the lowest value of 43.427, in the Hamtik specimens. The difference in the means of vertebral numbers in the specimens from the three stations, is statistically insignificant. However, the number of vertebrae in the Hamtik specimens had a greater range (40-45) while that in the Tigbauan and Taiwan specimens were smaller (42 to 44). From this meager data, it will not be justified to conclude that the number of vertebrae in the Hamtik fry is more variable than the others. We need much more data for confirmation.

Table 2 shows the number of vertebrae of milkfish reported so far from several countries by various authors.

Table 2. Number of vertebrae in milkfish reported from several localities by various authors.

<u>Locality</u>	<u>Author</u>	<u>Number of vertebrae</u>
India	Gunther, 1868	44
Indonesia	Sunier, 1922	44-45
Indonesia	Delsman, 1923	43
Philippines	Herre & Mendoza, 1929	46
Hawaii	Jordan & Evermann, 1905	45

It is interesting to note that the vertebral numbers reported by some authors (Jordan and Evermann, 1905; Sunier, 1922; Herre and Mendoza 1929) are rather high compared to those observed in the present study. A detailed study based on larger number of specimens from remote areas of varied ecological and environmental conditions may throw some light on the existence of subpopulations of milkfish.

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INTERNATIONAL MILKFISH WORKSHOP-CONFERENCE
Tigbauan, Iloilo, Philippines
May 19-22, 1976

CONFERENCE CALENDAR

A. Conference Organization

1. Overall Program

Chairman - Dean Domiciano K. Villaluz
Co-Chairman - Dr. W. H. L. Allsopp

2. Steering Committee

Chairman - Mr. Porfirio R. Manacop
Co-Chairman - Dr. F. Brian Davy
Members - Dr. Hiralal Chaudhuri
- Dr. Cesar Villegas
- Atty. Jose A. Agbayani, Jr.

3. Discussion Leaders and Rapporteurs for each Session*

Session I: Behavior and Bio-ecology of Milkfish in the Wild

Discussion Leader - Mr. P. R. S. Tampi
Rapporteur - Dr. F. Brian Davy

Session II: Egg and Larval Surveys

Discussion Leader - Dr. Tetsushi Senta
Rapporteur - Dr. Deb Menasveta

Session III: Maturation and Reproduction of Wild and Captive Fish

Discussion Leader - Dr. William Vanstone
Rapporteur - Dr. Ching Ming Kuo

* Although all the topics in the Sessions are expected to be considered with the Conference Calendar in the order presented, the time allotments for each Session will be flexible, depending upon the importance, number of papers submitted and other data that may be presented during the Workshop-Conference.

Session IV: Egg Incubation and Larval Rearing

Discussion Leader • Dr. Robert May
Rapporteur - Mr. Chin Phui Kong

Session V: Parasites, Diseases and Physiological Stress

Discussion Leader - Dr. Arporna Sribhibhadh
Rapporteur - Dr. Colin Nash

Session VI: Research Network for Milkfish Research

Discussion Leader - Dr. H. Chaudhuri
Rapporteur - Mr. P. R. Manacop

4. Conference Secretariat

In-charge - Atty. Jose A. Agbayani, Jr.

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Tigbauan, Iloilo, Philippines
May 19-22, 1976

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ULTRASONIC TRACKING OF MILKFISH
IN THE PHILIPPINES*

ULTRASONIC TRACKING OF MILKFISH IN PHILIPPINES
STOP OBJECTIVE OF SEAFDEC - TO LOCALIZE SPAWNING
GROUND OF MILKFISH IN PHILIPPINES COASTAL WATERS
USING ULTRASONIC TRACKING TECHNIQUE STOP OBJECTIVES
CANADIAN TEAM: (1) TO DETERMINE FEASIBILITY OF USING
SONIC TRACKING TECHNIQUES TO FOLLOW MOVEMENT OF
MILKFISH IN COASTAL WATER DURING SPAWNING PERIODS;
(2) TO TEACH THIS TECHNIQUE TO A NUMBER OF FILIPINO
BIOLOGIST AND TECHNICIANS STOP FINDINGS - SUCCESSFULLY
TRACKED MILKFISH RESPECTIVELY FOR ONE 34 AND 24 HOURS
FOR A TOTAL DISTANCE OF 29 KMS. STOP ALL FISH WERE
CAUGHT IN TRAPS AT TIGBAUAN HAMTIC AND PANDAN STOP
TAGS OF 73KHZ WERE USED FOR FIRST 3 FISH AND 50KHZ FOR
24 HOURS TRACK STOP PULSE GRADES RANGE FROM TWO TO
FOUR PULSES PER SECOND STOP RECEPTION RANGE UNDER
TEST CONDITIONS FOR 73KHZ STOP WAS ABOUT 500 METERS
VERY EARLY MORNING REDUCING TO 250 METERS BY NOON
STOP 50KHZ TAG HAD ABOUT 50 TO 100 PER CENT MORE
RANGE STOP TAGS WERE ATTACHED BY FISH HOOK TO BASE

*Cabled Report by Groot Biological Station Nanaimo
to D.K. Villaluz/Q.F. Miravite.

OF ANAL FIN WHILE FISH WAS HELD IN A PLASTIC TUBE FILLED WITH SEA WATER STOP TRACKING WAS PERFORMED FROM BOAT SEAFDEC USING HYDROPHONES AND SONIC RECEIVERS BOTH POSITIONS WERE PLOTTED EVERY HALF HOUR BY RADAR STOP FISH WERE RELEASED ABOUT TWO KMS OFF SHORE AND ALL HEADED FOR SHORE SOON AFTER RELEASED STOP ALL RECORDED MOVEMENT OCCURRED PARALLEL TO SHORE LINE BETWEEN ONE TO TWO KMS FROM SHORE STOP SWIMMING SPEED DURING FIRST HOUR AFTER RELEASE WAS ABOUT TWO KMS PER HOUR AVERAGE SPEED THEREAFTER VARIED FROM ONE TO TWO KMS PER HOUR WITH MAXIMUM SPEED OF SIX KMS PER HOUR STOP FISH OFTEN SLOW DOWN AND HELD IN ONE POSITION FOR LONG PERIOD STOP DISTANCE BETWEEN BOAT AND FISH WAS ESTIMATED AT 150 TO 200 METERS FOR 73KHZ TAGS AND 200 TO 300M STOP PROBLEMS MILKFISH TEND TO SOUND AFTER RELEASE STOP HYDROPHONE MUST BE ABLE TO PAN AND TILT STOP BIOLOGICAL SOUND IN 73KHZ RANGE CREATES LISTENING PROBLEMS CLOSE TO SHORE STOP PROPELLER AND SHAFT NOISE WITH PULSE RATES SIMILAR TO TAG CREATE PROBLEMS AT 73KHZ RANGE STOP WARNING OF SERVICE WATERS WHICH RISING SUN REDUCE RATE OF TAG CONSIDERABLE METERS FOR 60KHZ STOP CONCLUSION AND RECOMMENDATION THAT FOLLOWING MOVEMENT OF MILKFISH IF POSSIBLE BY USING

SONIC TRACKING TECHNIQUE STOP 50KHZ TAG IS PREFERRED OVER 73KHZ STOP HYDROPHONE MUST BE ABLE TO PAN AND TILT TO FOLLOW SOUNDING THE FISH STOP BOTH SHOULD BE A SHALLOW DRAFT CHRIS CRAFT TYPE CRUISER OF 40 TO 50 FEET WITH TWIN DIESEL WESTMAR SONAR SS150 OR 200 TO OPERATE HYDROPHONE DEPTH SOUNDER RADAR MARINE RADIO BOAT TO BOAT COMMUNICATION OF VHF OR CITIZEN BAND AND SMALL RUBBER BOAT OF 12 FEET LONG PLUS OUTBOARD FOR PORT TO PORT STOP TRAINING-CANADIAN TEAM TRAINED ONE TECHNICIAN AND SEAFDEC CREW IN SONIC TRACKING TECHNIQUE.