## UNITED STATES DEPARTMENT OF THE INTERIOR, Stewart L. Udall, Secretary FISH AND WILDLIFE SERVICE, Clarence F. Pautzke, Commissioner BUREAU OF COMMERCIAL FISHERIES, Donald L. McKernan, Director

# TANK CULTURE OF TILAPIA

BY RICHARD N. UCHIDA AND JOSEPH E. KING



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#### ABSTRACT

This study evaluated the feasibility of producing bait-size tilapia by the tank-culture method. Two facilities were used: a pilot plant constructed on the grounds of the Biological Laboratory at Honolulu and a second and more elaborate plant constructed at Kewalo Basin, Honolulu.

Study of some of the factors associated with reproductive rates revealed that (1) only a slight increase in water temperature was necessary to increase spawning frequency during winter months, (2) prolonged high temperatures seemed to have a detrimental effect on spawning fish, (3) a sex ratio of  $39:1\delta$  resulted in the highest reproductive rate, (4) a concentration of brood stock that allowed 4.0 square feet of bottom area per male and 1.0 square foot per individual provided optimum conditions for courtship and spawning, (5) brood stocks fed a high-quality feed had a higher reproductive rate than those maintained on a low-quality feed, and (6) brood stocks maintained in brackish water of about 10 °/<sub>00</sub> had significantly higher fry production than those in fresh water.

Crowding affected growth rate of young as did quality of the food and salinity of water.

The major causes of mortality among the adults were handling, disease, asphyxiation, and possibly hydrogen sulfide poisoning. High mortality rates among the young were caused by an infectious disease and infestation by ectoparasitic protozoans.

Experiments on cannibalism indicated that juvenile tilapia averaging 20.4 mm. killed or consumed fry up to 10.0 mm. in length, while juveniles averaging 64.4 mm. in length were able to kill or consume smaller juveniles up to a maximum size of 24.5 mm. Starved juveniles were more aggressive than well-fed juveniles.

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## TANK CULTURE OF TILAPIA

## By RICHARD N. UCHIDA and JOSEPH E. KING, Fishery Research Biologists

#### BUREAU OF COMMERCIAL FISHERIES

Until about two decades ago, the cichlid fish Tilapia mossambica Peters had received only minor attention as a food and game fish in its native East African environment. No intensive cultivation of tilapia was carried on in Africa, and it was not until this fish mysteriously appeared in East Java in 1939 that anyone recognized that it possessed many of the desirable characteristics of a pondfish and that it was readily adaptable to culture (Atz, 1954). The potentialities of various species of *Tilapia* were demonstrated by W. H. Schuster before a gathering of inland fisheries experts held at Surabaja in 1939 (Vaas and Hofstede, 1952). Since that time, tilapia have been successfully introduced into many southeastern Asian countries where they have become an important source of protein food.

In recent years, many scientists in various parts of the world have studied the biology of tilapia and its culture in ponds and rice paddies. The work of Vaas and Hofstede (1952), Chen (1953), Panikkar and Tampi (1954), and Swingle (1960) is particularly noteworthy. Chimits (1955, 1957) has published excellent reviews of tilapia culture and his bibliographies bring together a wealth of information on these fishes. Baerends and Baerends-Van Roon (1950) should be mentioned for their contributions to knowledge of the behavior of the cichlids. Other sources of information on various aspects of tilapia culture and biology are Brock (1954), on spawning in salt water; Fish (1955) and Le Roux (1956), on feeding habits; and Lowe (1955), on fecundity. Brock and Takata (1955) and King and Wilson (1957) reported on use of young tilapia as supplementary tuna bait, and Hida et al. (1961) on the tank culture of bait-size tilapia.

Swingle (1957) was the first to recognize that species of *Tilapia* have potentialities as a freshwater game fish in the United States. Other investigators have found several of the many species of *Tilapia* to be suitable laboratory animals for many types of physiological research.

The decision to introduce *T. mossambica* to Hawaii was based primarily on two major considerations: the usefulness of the fish for clearing aquatic vegetation from irrigation ditches and canals and the possibility that the young could be used as bait fish in the Hawaiian skipjack fishery (Brock and Takata, 1955). Since its introduction to Hawaii in 1951, tilapia has received widespread attention and is now well established in many private and commercial ponds throughout the major Hawaiian islands.

The pole-and-line fishery for skipjack (Katsuwonus pelamis) is the largest commercial fishery in Hawaii. Descriptions of this fishery and the associated live-bait fishery have been published by June (1951), Brock and Takata (1955), and Yamashita (1958). All of these investigators cite the shortage of bait fish as the principal factor limiting production of this pole-and-line fishery.

Because of this critical demand for bait, attention was focused on tilapia as a possible supplement to natural bait supplies. Tester et al. (1954), experimenting with artificial materials (both edible and inedible) to attract tuna to the stern of a fishing vessel, reported generally negative or inconclusive results, which gave added impetus to the search for a suitable substitute live bait. In the summer of 1954, Brock and Takata (1955) initiated the first sea trials to evaluate tilapia as live bait and in a number of the sea tests they obtained encouraging results. King and Wilson (1957: p. 8) made further sea tests during the summer and fall of 1956 and concluded that the young of *Tilapia mossambica* are an adequate bait fish for catching skipjack. They further pointed out that although tilapia in some respects was in-

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ferior to nehu (*Stolephorus purpureus*), the principal bait used in the skipjack fishery, it was on the other hand, hardier than nehu and could tolerate a wider range of salinity and lower oxygen concentrations.

In view of the reported success in using young tilapia as a tuna bait fish, consideration was given to devising rearing methods that would be economically and biologically feasible for producing adequate numbers of fish of proper size. Two immediate possibilities presented themselves: pond culture, whereby existing ponds on the islands would be utilized with some modifications; and tank culture, with separate spawning tanks and fry-rearing or nursery facilities.

King and Wilson (1957: p. 8) utilized bait-size tilapia obtained from private ponds and reservoirs for their sea trials and after a number of bait-seining operations concluded that—

it does not appear that the rearing of tilapia for bait purposes can be done most effectively in water reservoirs and natural ponds with little control over spawning, cannibalistic traits of the species, and predation, and with the difficulty of harvesting the fish efficiently at an optimum size.

It was anticipated that tank culture of the fish under controlled conditions might prove to be a more efficient and economical way to produce baitsize tilapia. As a result, a study of tank culture of tilapia on a pilot-plant scale was initiated at the Bureau of Commercial Fisheries Biological Laboratory at Honolulu. The primary objectives of this study were to determine the physical and biological problems associated with tank culture and the potentialities of producing bait-size tilapia in sufficient quantities under controlled conditions in a hatchery-type operation.

The pilot plant was constructed on the grounds of the laboratory in October 1956 and experiments were carried on there until July 1958, after which the facilities were transferred to more spacious grounds adjacent to the laboratory's new docksite building at Kewalo Basin, Honolulu. Studies continued at the Kewalo plant until September 1959. While the purpose of the pilot plant was primarily to examine the general problems associated with production of bait-size tilapia, the Kewalo plant studies were designed to obtain a more detailed and quantitative evaluation of tank culture and to assess the various factors associated with variations in reproductive, survival, and growth rates.

The purposes of this report are to discuss our efforts in establishing operational procedures and basic requirements that would be applicable to a commercially operated tilapia hatchery, to present our observations and conclusions on reproduction and growth, and to describe the cannibalism, predation, and diseases of tilapia observed during the experiments.

As work progressed, the need for information on other rearing methods became evident and a study of pond culture of tilapia under controlled conditions was begun by the Hawaii Division of Fish and Game at Kaneohe, Oahu, under contract with the (Territorial) Economic Planning and Coordination Authority (EPCA) and the Bureau of Commercial Fisheries. Production of baitsize tilapia in ponds under uncontrolled conditions was investigated to some extent by King and Wilson (1957) during the summer of 1956.

The Maui Fisheries and Marine Products Co., Ltd., expressed an interest in establishing a tilapia hatchery on a semicommercial basis, following our initial success in producing bait-size tilapia at the pilot plant. A contract was signed with Maui Fisheries, the Hawaii Division of Fish and Game, and the Bureau of Commercial Fisheries, as principals, late in December 1957, for the operation of a tilapia-rearing plant at Paia, Maui. A fishery biologist, supplied by the Bureau, was placed in charge of the plant to obtain detailed records of the production and of operational costs. The Paia hatchery was operated for 2 years and the results have been reported by Hida et al. (1961).

In frequent references throughout this report to the various stages of development of tilapia, we have tried to conform to generally accepted terminology, but in some instances we found combinations of categories more suitable and in other ways we have diverged from ordinary usage. The terminology that we have employed is defined as follows:

Fry.—Includes both the prolarval (yolk-bearing) and postlarval (nonyolk-bearing) stages. Includes tilapia up to 19.0 mm. (0.75 in.) in length.

Juvenile.—Stages between fry and adult, with a range in size from about 20 to 100 mm. (0.75 to 4 in.).

Bait size.—Juveniles suitable for skipjack bait range in size from about 38 to 51 mm. (1.5 to 2 in.); however, on several occasions juveniles falling outside this length range have been used for bait.

Young.—A general category that includes fry and juveniles.

Adult.—Mature fish that are potential breeders and are distinguished by the display of coloration, especially by the males in the reproductive phase. Larger than 100 mm. (4 in.) in length.

The length measurement used in this report is fork length; that is, the length from tip of the snout to ends of the middle caudal rays. Body measurements (length and weight) were made in both English and metric units during the early phases of the investigation. However, all original measurements in English units have been converted to their equivalent in metric units, and where applicable or when appropriate the English units are given in parentheses. Reference to plant facilities is either to the pilot plant constructed on the grounds of the Bureau's Honolulu Laboratory or to the Kewalo plant located at the Kewalo Basin docksite.

We gratefully acknowledge the advice and suggestions given us by the Bait-fish Research Coordinating Committee composed of biologists of the Hawaii Division of Fish and Game, the University of Hawaii, and the Bureau of Commercial Fisheries, Honolulu, and representatives of the Hawaiian Tuna Boat Owners Association and the Hawaiian Tuna Packers, Ltd.

## CULTURE METHODS REARING FACILITIES

#### **Pilot Piant**

Three redwood raceway-type tanks, each 5 feet wide by 20 feet long and 3 feet deep, with a capacity of 1,400 gallons, were constructed on the grounds of the laboratory in October 1956 (fig. 1). The floors sloped downward toward the outflow end of the tank at the rate of 1 inch for each 10 feet of length. The outflow, located in one corner of the tank, consisted of a removable standpipe that slipped into a hole in the floor of the tank and connected with a gate valve and drainpipe on the outside. Baffle boards and a brass screen enclosed a triangular area occupied by the standpipe. The baffle boards were raised about



FIGURE 1.-A redwood raceway tank at the pilot plant.

2 inches above the floor. Thus, the outflowing water was drawn from the floor of the tank and aided in removing detritus from the tanks. The brass screen prevented the fry from being carried out through the drain. The inflow, situated on the opposite end of the tank, was a faucet from which fresh water from the Honolulu water supply was dripped into the tank at the rate of onehalf to 1 gallon a minute. The tanks were given a coat of aluminum paint before being used.

Initially, these three tanks were used as brood or spawning tanks. With the onset of fry production in December 1956, it was necessary to convert one of the tanks to a fry-rearing tank. Continued fry production created a need for more rearing space. In June 1957, a fourth redwood raceway-type tank, 30 inches wide by 30 feet long and 30 inches deep, with a capacity of about 1,250 gallons, was constructed adjacent to the other tanks. Screens of fine-meshed Monel stretched over square wooden frames were used to partition the tank into six approximately equal compartments. Each compartment had a siphon-type drain of plastic tubing. A standpipe drain was placed at one end of the tank in the event that the siphons clogged or failed. Each compartment was served by a fresh-water tap.

In June 1957 we acquired a number of surplus assault boats from the U.S. Army, four of which were installed at the laboratory and used as brood and fry-rearing tanks. These undecked plywood boats measured approximately 4 by 12 feet on the bottom and each had a capacity of about 840 Although of very light construction, gallons. they were fairly watertight for more than a year. Water flowed in through a hose attached to the stern; the drain consisted of a plastic siphon attached to the bow. Several 1-inch holes were drilled in one side of the square bow just above the water line for controlled overflow if the siphon failed. Figure 2 shows an assault boat converted into a fish-holding tank, while figure 3 illustrates the plan of the pilot plant.

A filter system, consisting of a sand filter box and a pump, was installed experimentally on tank 2. It soon became apparent that the filter box was not adequate and that beach sand was inappropriate as a filtering medium. Cleaning and backflushing of the filter was necessary two or three times a week, requiring considerable time and effort. The sand was eventually replaced with several layers of fine-meshed Monel screen, which was not a very effective filter but did remove large amounts of fecal matter and other detritus. The turbulence and the splash on the surface caused by the water as it was returned to the tank under pressure also increased the oxygen concentration.

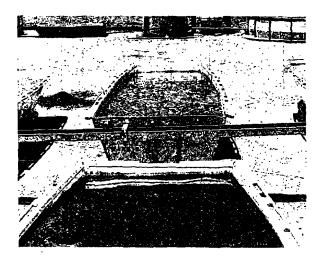


FIGURE 2.—An assault boat converted into a fish-holding tank, Kewalo plant.

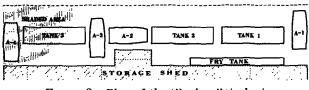


FIGURE 3.---Plan of the tilapia pilot plant.

Other minor modifications in the pilot plant included installation of an aeration system and floodlights on tank 3. The aeration system consisted of an air compressor, a rubber air hose, and a pipe (drilled with holes) that ran crosswise of the floor in the center of the tank. Air was pumped through this pipe and slowly bubbled through the water in the tank.

Two 150-watt projector floodlights were installed on tank 3, one at each end, approximately 5 feet above the surface of the water. It was hoped, by day-and-night illumination, to increase the algal content of the tank, which was very low, and also possibly increase the production of young. No changes were detected, however, and the floodlights were removed after 3 months.

#### **Kewalo** Plant

In July 1958, the redwood tanks (three brood tanks and one fry tank) at the pilot plant were dismantled and reassembled at the Kewalo Basin docksite. A filter system (fig. 4), consisting of a sand filter box 24 inches wide by 26 inches long and 18 inches deep and a pump, was attached to each of the three brood tanks. The fry tank was modified by increasing the width 10 inches and by partitioning it with plywood separators (instead of screens) into six compartments. Essentially, each compartment was a separate tank with its own drain and tap. A filter box, 30 inches on the sides and 18 inches deep, and a pump were installed to filter and to recirculate the water.

The four assault-boat tanks at the pilot plant, after being in use for about a year, were not worth salvaging. Thirteen assault boats were removed from storage and converted into 12 brood tanks and 1 filter tank at the Kewalo plant. Drain water from the tanks was carried down a flume to the sand-filter tank and pumped back to each tank. All filter boxes contained a bottom layer of crushed rock and a top layer of coarse black sand (volcanic cinders), which was found to be much more effective than the fine beach sand used initially.

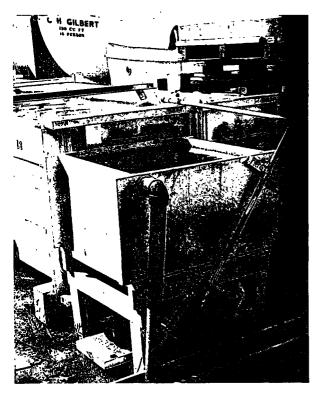


FIGURE 4.—Filter and pump on a raceway tank.

The general arrangement of the assault-boat tanks and the redwood tanks is shown in figure 5.

#### STOCKING THE BROOD TANKS

One of the first problems to be considered in the operation of the pilot plant was what sex ratio should be used when stocking the tanks. Chen (1953: p. 6), working on tilapia in Taiwan, stated that the proper sex ratio for propagation purposes is one male to a female. Other investigators, however, reported that in mouth brooders, which group includes T. mossambica, the female visits the spawning grounds only briefly to extrude her ova and collect the fertilized eggs, and then moves away or often is chased away by the male. The male remains on the spawning ground to guard the nest and immediately begins to court other females. He is, thus, available for and presumably capable of fertilizing ova from a succession of ripe females (Baerends and Baerends-Van Roon, 1950; Lowe, 1955). Lowe (p. 48), with respect to the mouth brooders, concluded that as male fish can continue fertilizing over a long period, the number of eggs ferti-

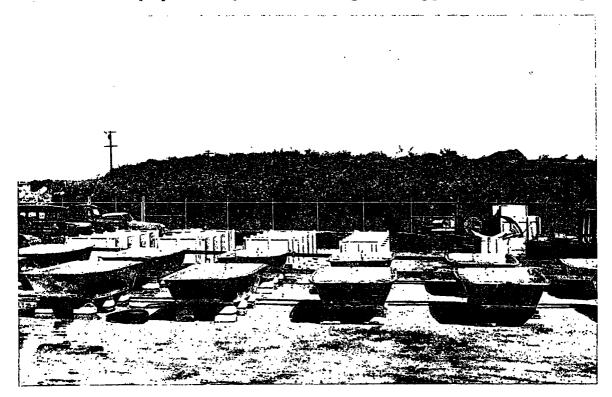


FIGURE 5.—Tilapia tanks at the Kewalo plant after a heavy rain.

lized appears to be determined more by the number of ripe females than by the number of males.

Taking advantage of this behavior to realize a maximum production of young with a minimum of brood stock, a ratio of  $2 \ 9 \ :1 \ s$  was tried initially, with results that were considered to be successful. A later experiment at the pilot plant using a ratio of  $5 \ 9 \ :1 \ s$  yielded less-successful results, as will be described later.

Another important consideration was the density of the brood stock or carrying capacity of the brood tanks. Originally, it was decided to try a total of 48 fish with about 1.9 square feet per individual in one tank, and 96 fish or approximately 0.9 square foot per individual in the other two tanks, each with a sex ratio of 29:13(table 1).

TABLE 1.—Number of adults of each sex and space allowance per male and per individual, in brood tanks at the pilot plant

Tank No.	Number	Number		er— (sq. ft.)	Volume of water per individual
	. males	females	Male	Individual	(cu. ft.)
1 2 3	32 32 16	64 64 32	2, 8 2, 8 5, 6	0.9 .9 1.9	1.9 1.9 3.8

On October 12, 1956, the three tanks at the pilot plant were stocked with adult fish averaging 20 cm. (8 in.) in length and 150 g. (5.3 oz.) in weight. These fish were supplied by Hawaiian Tuna Packers, Ltd., and obtained from Ewa plantation pond No. 6. The initial mortality among the brood stock was very low, amounting to only three males and three females the first 15 days after stocking. All casualties were replaced. The exact number of adult fish in each of the tanks varied during the course of the test, however, owing to unobserved mortalities, vandalism, and the recruitment of juveniles that escaped dipnetting and grew to adult size in the brood tanks.

The stocking of the Kewalo plant followed an experimental design and will be discussed under factors affecting reproduction, page 37. Stock for the Kewalo plant was also obtained from Ewa pond No. 6. The males averaged 142 g. (5 oz.) and the females about 113 g. (4 oz.) in weight. No length measurements were made.

#### FEEDING

Many investigators have reported on the feeding habits of the various species of tilapia. Generally, *T. mossambica* is considered to be omnivorous (Schuster, 1952; Chen, 1953; Atz, 1954; Panikkar and Tampi, 1954; Brock and Takata, 1955; Van Pel, 1955). Vaas and Hofstede (1952: p. 35) stated that tilapia is herbivorous, but will feed on planktonic Crustacea "if such kind of food is more plentiful than vegetable food" and will show a preference for vegetable food when a mixture of the two is present.

Feeding of the brood stock at the pilot plant was started immediately after stocking was completed. Various types of commercial feed, such as rice bran (powdered), millrun (powdered), chicken starter mash (powdered and granules), alfalfa pellets (%2 in. in diameter), pelletized rabbit feed  $(\frac{5}{2})$  in. in diameter), and a pelletized pond-fish feed  $(\frac{5}{2})$  in. in diameter), were tried to determine which were most acceptable to the fish. With a daily feeding rate of about 2 percent of the fish weight, we observed that finely divided, unpelletized feed, such as rice bran, millrun, and chicken mash, was not efficiently utilized because of the small particle size. The leftover feed contributed to fouling of the tanks. The alfalfa pellets and rabbit feed were also found to be undesirable because of their high content of indigestible fiber, which collected on the bottom of the tanks and also caused fouling.

Early observations on the feeding habits of the newly emerged fry indicated that they did not respond to supplementary feed until about 2 weeks after they were placed in the fry tank. The fry grade of pondfish feed (granules) was further ground to a flourlike consistency to accommodate the very young. The larger fry and juveniles reared at the pilot plant were fed pondfish feed in the crumble grade, which was slightly larger than the fry-grade granules.

Except in feeding experiments, the brood stock at the Kewalo plant was fed almost exclusively on a prepared trout feed (developer grade, about  $\frac{1}{4}$  in. in diameter), which in a smaller particle size was also fed to the fry (starter grade, powdered; fry grade, granules) and juveniles (small fingerling grade, crumbles). Table 2 gives the composition, supplied by the manufacturer, of the various feeds that were used.

Feed	Pro- tein <sup>1</sup>	Fat 1	Fiber <sup>2</sup>	Ash :	Added min- erals <sup>2</sup>	Nitro- genfree ex- tract <sup>1</sup>	
Rice bran Chicken mash Alfalfa pellets Pondfish feed Trout feed: Developer Starter. fry. and small fingerling Wheat white middlings Millrun Rabbit feed	9.0 20.0 15.0 30.0 25.0 40.0 13.5 12.0 16.0	4.0 2.0 4.5 4.0 2.5 3.0 3.0 2.5	4.5 28.0 6.0 3.5 5.5 4.0 10.0 21.0	7.5 12.0 9.0 5.0 6.0 10.0	3.0	35.0 28.0 44.0 44.0	12, 0 18, 0 14, 0

TABLE 2.—Guaranteed analyses (percent) of ingredients in the feeds used at the pilot plant and the Kewalo plant

The brood stock at both plants was fed once a day, usually at midday. There were periods during the operation of both plants when the adults did not feed readily, and during these times smaller portions were supplied to the fish. This apparent lack of interest in feeding is probably associated with the mouth-brooding habit of the female.

The amounts of the various kinds of feed that were supplied to the adults and young during operation of the pilot plant and the Kewalo plant are given in tables 3 and 4. The fry were usually fed twice a day during weekdays, once in the morning and once just after midday, and once a

TABLE 3.—Amount (in pounds) of the feeds used at the pilot plant

	Rice	Chicken		ish feed	Alf	Rah-	
Month	bran	mash	Pellets	Crum- bles	Pellets	Meal	bit feed
956:							
October	2.5	2.5	25.0				
November		8.8	50.8				
December	22.0	16.0					
1957:		1			1		
January	23.3		11.0	3.5			
February March			26.0	8.9			
March			31.0	11.6			
April		- <b></b>	30.0	12.2			
May			31.0	15.0			
June			31.5	18.9			
July			30.0	19.6			
August September October November		8.0	30.0	13.1 14.8			
September		15.2	31.5	14.8	1.0		
November		14.0	36.8	14.0			
December		[	46.0	14.0			
1958:			30.0				
January	l	l	50.0	0.1	0.5		ŀ
January February			38.5	0. Î	0.0		
March	1		42.6	4 8			
April May June		1	40.4	6.0			
May			1.4	3.5			44.
June				2.3			45.
July		·		1.2			25.
Number of pounds. Average cost per	47.8	64.5	613.5	182.5	16.5	11. 4	114.
pound.	\$0.04	\$0.05	\$0, 17	\$0.27	\$0.04	\$0.04	\$0.0

TABLE 4.—Amount (in pounds) of the feeds used at the Kewalo plant

	Trou	t feed			Wheat
Month	Developer	Fry and small fingerling	Rabbit feed	Mill- run	white middlings
1958:	•				
August.	57.1		7.0	7.0	
September	111.6	0.2	14.8	14.8	
October	107.6	1.6	14.0	14.0	
November	93.1	2.5	13.0	13.0	
December		1.5			
1959:					
January	40.5	1.7			
February	42.0	1.7			
March	44.2	1.7	2.2		
April	22.5	5, 2	22.5		
May		9.7			0.4
June	45.0	20.8			1.6
July	51.2	21. 2			5.3
August		22.4			3. 1
September	22.0	29.5			
Number of pounds	751.8	119.7	73.5	48.8	10.4
Average cost per pound.	\$0.13	\$0.13	\$0.06	\$0.04	\$0.0

day on weekends. The amount of feed given to the young varied with the number and size of fry and juveniles.

#### FRY PRODUCTION

T. mossambica reportedly first spawns at the age of 2 to 3 months and at a length of 8 to 9 centimeters. The frequency of spawning varies considerably, depending on environmental factors, and ranges from 6 to 16 times a year (Chen, 1953; Panikkar and Tampi, 1954; Chimits, 1955). Chen reported that a spawning fish of about 8 cm. in length produces from 100 to 150 ova at each spawning, although at the first spawning it may produce less than 100 ova. He stated further that the number of ova spawned increases with successive spawnings, so that a fish more than 6 months of age may produce in excess of 1,000 ova per spawning.

The developmental period of the ova is likewise variable but, generally, the ova hatch after 2 to 5 days and the young are carried in the mouth of the female for another 5 to 8 days before they are released (Chen, 1953; Panikkar and Tampi, 1954; Chimits, 1955).

From the time of the initial stocking at the pilot plant, careful daily observations were made on each tank to determine if young were being produced. As no signs of young fish were seen for about 6 weeks, the three tanks were partially drained and cleaned on November 28-29, 1956. Before the water level was restored in each tank, the detritus on the bottom of the tank was ex-

<sup>&</sup>lt;sup>1</sup> Not less than. <sup>2</sup> Not more than.

amined for signs of ova and young. The females were also examined to see if ova or young were being carried in the mouth. It was noted at this time that all the brood stock appeared in excellent condition but there were no signs of spawning.

On December 5, 1956, one-half cubic yard of white beach sand was placed in two of the brood tanks (tanks 1 and 3), covering the bottom to a depth of about 3 inches. It was our original intention to determine if the tilapia would spawn on the bare floor of the tanks, which seemed probable in view of Chen's (1953: p. 7) observation that tilapia were seen spawning successfully in a garden pond with a concrete bottom.

Good evidence of excavating and nest-building was noted the next morning following placing of the sand, and the nests continued to increase in number during the next few days until they occupied at least two-thirds of the sandy bottom. Two weeks after the sand was placed in the tanks, the first young were noticed on the surface of tank 1. The young were removed and subsequently placed in tank 2, which was drained and converted into a fry tank. Six days later, the fish in tank 3 started to produce young.

During the draining of tank 2 in preparation for conversion into a fry tank, one large male was observed carrying six yolk-sac fry in its mouth cavity, although in this species the female is supposed to brood the young. This behavior was also observed by Vaas and Hofstede (1952), who reported that the male incubated the ova in exceptional cases. Further evidence of spawning was found in tank 2, where many ova and yolksac fry were seen widely scattered on the tank floor. It was our supposition that the adults had become excited as the water level dropped and ejected the ova and fry.

At the Kewalo plant, where all the brood tanks were supplied with sand, fry production started 11 days after the tanks were stocked. Further data on fry production at the Kewalo plant will be presented later in this report, together with the results of the various experiments.

#### FRY COLLECTION

Newly hatched fry of T. mossambica measure 5 millimeters in length, 5.8 mm. on the second day, and 8.0 mm. at the end of the fifth day. On about the fifth day, they begin to spend less time in the

mouth of the female or leave it altogether and swim about in a tight school near the surface of the water, feeding on tiny food particles (Panikkar and Tampi, 1954). This behavior of the fry made their capture by dipnetting a relatively simple process. Another behavior pattern that became evident to us through daily observation was the tendency for the fry to congregate along the walls of the tank, especially in the corners and, at times, directly under the inflowing water.

At both plants, the young emerging each day were captured with a fine-meshed dipnet and counted as they were released into the fry tank. Although our daily netting effort varied somewhat in efficiency, as indicated by the differentsized young netted, the method proved to be quite effective as evidenced by the few juveniles that were removed from the tanks when they were drained.

Early in the operation of the pilot plant, the displacement and weight methods for assessing the daily production of young were considered, but both had major drawbacks for enumerating the very small tilapia fry. We concluded that only by an actual count could we obtain the accuracy desired.

In August 1957, a shelf-collection method of capturing young was investigated at the pilot plant. This method was originally the idea of biologists of the Hawaii Division of Fish and Game who observed that tilapia fry tended to congregate in shallow water near the edge of the ponds. Consequently, it was hypothesized that if the fry had shallow water available to them along the walls of the brood tank, perhaps their capture would be simplified.

A redwood shelf, running the length of the inside wall just below the waterline, was installed in tank 1. The water level in the tank was maintained so that the outer edge or lip of the shelf was ordinarily about one-half inch below the surface. The water over the shelf could be drained through a hole in the wall of the tank and the young fish caught in a net.

Comparison of the number of fry collected from the shelf with that collected elsewhere in the tank by dipnetting indicated that the shelf was effective in the removal of only about 25 percent of the fry that emerged daily. Further, many of the fry congregated under the shelf, rather than over it. In view of the results that were obtained, the shelf collection method was abandoned.

#### FRY REARING

As fry production increased, it became a serious problem to provide sufficient space for the young fish. Initially, the fry were placed in one large fry tank. After a few weeks, cannibalism became widespread: the juveniles readily devoured the newly collected fry when they were transferred to the fry tank. As a remedy, removable frames covered with fine-meshed Monel screen were installed in the tank dividing it into three compartments. The fry and juveniles could then be segregated by age and size.

With the construction of a new fry tank with six compartments and the acquisition of four assault-boat tanks at the pilot plant, additional space for rearing the young was available. The procedure became standard to place the fry in the fry tank for about 4 to 5 weeks, after which time they were transferred to assault-boat tanks.

When fry were removed from the brood tanks, they usually ranged in size from 7.8 to 13.6 mm., with the average about 10.8 mm. Of a total of 154 fry ranging from 7.8 to 10.3 mm., about 26 percent carried remnants of the yolk sac. The newly collected fry, as mentioned earlier, generally paid little or no attention to the prepared feed when first offered it and only took it after about 2 weeks in the fry tank.

Vaas and Hofstede (1952) observed that young tilapia fed on diatoms, unicellular green algae, small Crustacea, and periphyton. Varying numbers of these organisms were present in the tanks and undoubtedly constituted a major portion of the diet of the fry during their first few weeks of life after absorption of the yolk sac. The feces of the young fry were usually bright green, indicating that algae were a \*major constituent of their diet.

The necessity of utilizing all fry-rearing space available during periods of heavy fry production occasionally forced us to overcrowd the fry tank and frequently resulted in an outbreak of disease. A criterion for determining when an overcrowded condition existed was difficult to formulate, but through experience we arrived at what we considered an optimum stocking density. The general plan was to stock the fry-tank compartments with approximately 200 fry per square foot of surface area and crop each compartment at frequent intervals, removing the larger, faster-growing individuals; i.e., fish that were 20 mm. (0.75 in.) or larger. In this manner, cannibalism and disease were kept to a minimum. Also, through the necessity of occasionally overcrowding the juveniles in the assault-boat tanks, we learned that they were much more tolerant to crowding than were the fry and also that they were afflicted less frequently with ectoparasites.

The most critical period for the fry appears to be the first 4 to 5 weeks of life, for it is in this period that the fry are most susceptible to infectious diseases and ectoparasitic infestation. Proper sanitary conditions in the tanks and adherence to an optimum stocking density for a given area seem to be the two most important factors determining survival and subsequent health of the fry.

#### LENGTH-WEIGHT RELATION OF YOUNG

During the operation of both plants, it was frequently necessary to estimate the size and quantity of young fish on hand. In order to have a convenient means of converting length into weight and vice versa, the length-weight relation for tilapia 17 to 73 mm. in length was determined from body measurements of 109 fish. The logarithmic formula for the expression of this relationship is

 $\log W = 3.088 \log L - 4.8935$ 

where W is the weight in grams and L is the length in millimeters. A curve demonstrating the length-weight relation is shown in figure 6.

#### WATER-QUALITY DETERMINATIONS

Some chemical determinations were made routinely once each week. Only oxygen measurements were made at the pilot plant; but at the Kewalo plant, additional properties, such as free carbon dioxide, bicarbonates, normal carbonates, and hydrogen-ion concentration were also measured weekly in selected tanks.

#### **Oxygen Content**

Determinations of dissolved oxygen in the brood and fry tanks were made by the modified Winkler method. Analyses were made once a week at

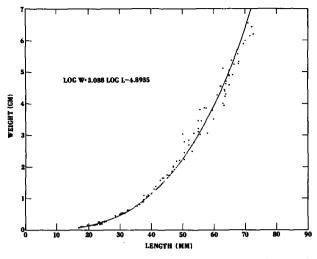


FIGURE 6.—Length-weight relation for tilapia 17 to 73 mm. in length.

about the same hour each time to obtain a general record of the variation in oxygen content of the water (appendix tables 1, 3, 4, and 5). In a 24hour series (with sampling at hourly intervals) obtained at the pilot plant November 12-13, 1957, we found a marked diurnal change in the concentration of oxygen in the tanks, the minimum concentration occurring at about daybreak and the maximum concentration at about midafternoon (fig. 7; appendix table 2). The maximum concentration was certainly the result of photosynthesis by the algae in the tanks, and the minimum was caused by respiration of both algae and Water temperatures in the tanks showed fish. a similar diurnal variation, with the occurrence of

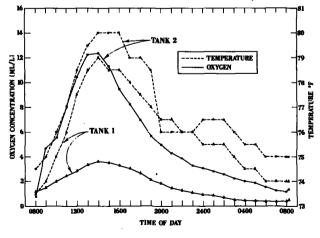


FIGURE 7.—Diurnal variation in oxygen concentration and temperature in tanks 1 and 2 at the pilot plant.

maximum and minimum temperatures corresponding very closely to the maximum and minimum concentrations of oxygen.

Vaas and Hofstede (1952) stated that T. mossambica has a high metabolic rate compared with carp, and that oxygen is consumed and carbon dioxide liberated in large quantities within a short time by the species. They also found that when oxygen tension is low, especially during the morning, the fish concentrate at the surface of the water suspended in a diagonal position, sucking in the well-aerated water of the surface layer with wideopen mouths.

It is suspected that some of the mortalities among the adults at both plants were associated with an insufficient amount of oxygen. In tank 3 at the pilot plant, deaths occurred periodically for about 2 months in the summer of 1958. During this period, the oxygen minima occurring in the early morning were consistently low, averaging only 0.38 ml./l. Of the 10 adults that died during this period, 9 were large males averaging 29 cm., and the single female was 23 cm., perhaps indicating the greater susceptibility of the larger tilapia to low oxygen concentration.

Since our purpose in sampling oxygen was primarily to monitor environmental suitability, we have not attempted to relate the data to fry production, except in the heating experiment, where low oxygen concentrations were believed to be associated with the poor fry production. From the data it appears that, in most instances, the minimum or near-minimum oxygen concentration in the sampled tanks was sufficient to sustain the tilapia.

#### Hydrogen-Ion Concentration

Determination of hydrogen-ion concentration was made by the colorimetric method at the time the oxygen samples were drawn. The pH values of the tanks that were sampled (appendix tables 6-8) ranged from 7.3 to values exceeding 8.8 (the color standard used at first could measure pH only to 8.8). It was expected that the pH in our tanks would remain on the alkaline side, since city water (pH range, 7.9 to 8.3) was used and, with the exception of the fry tank, the tanks contained a 3- to 4-inch layer of calcareous beach sand. The hydrogen-ion concentration and its significance to aquatic organisms has been relegated for some time to a minor position by many investigators (Welch, 1935; Odum, 1959). It may possibly be a limiting factor to some organisms, however.

#### Free Carbon Dioxide

Analysis of the amount of free carbon dioxide in the tanks was started at the Kewalo plant in February 1959. The samples were drawn immediately after those for the oxygen determination had been drawn and fixed. The amount of free carbon dioxide present was approximated by titration of a 100-ml. sample to the phenolphthalein endpoint with 0.02 N sodium hydroxide.

Welch (1935: p. 175) stated that carbon dioxide is one of the most important substances in the life of organisms, but that it should be present only under suitable circumstances and in proper amounts. He stated further that a small amount of carbon dioxide appears to be essential for aquatic animals. Doudoroff (1957) regarded free carbon dioxide concentrations between 100 and 200 p.p.m. as fatal to moderately susceptible freshwater fishes; also, that exposure to concentrations between 50 and 100 p.p.m. causes immediate distress and may be lethal if the exposure is prolonged. He noted that even in polluted waters free carbon dioxide concentrations rarely exceed 20 p.p.m.

Throughout the period of sampling at the Kewale plant, the free carbon dioxide concentration (appendix tables 9 and 10) was never found to be in excess of 16.2 p.p.m. No attempts were made to determine a relation between carbon dioxide concentrations and fry production. Our routine observations were made primarily to detect excessive amounts of free carbon dioxide in the brood and fry tanks.

#### Alkalinity

Chemical analyses of the water in the tanks at the Kewalo plant included determinations of total bicarbonate and carbonate alkalinity. A 100-ml. sample was titrated with 0.02 N sulfuric acid against the phenolphthalein and methyl orange endpoints.

Alkalinity directly influences the biological productivity of a body of water. The carbonates and bicarbonates, which are in close chemical combination with carbon dioxide, are utilized by algae (Welch, 1935), and also act as buffers by keeping the hydrogen-ion concentration close to the neutral point (Odum, 1959).

Our alkalinity determinations (appendix tables 8 and 10) were made primarily to gain a general knowledge of the type of water present in the tanks, as soft water has a smaller supply of these ions and, therefore, is less productive.

#### MORTALITIES

#### Among the Adults

Observed mortalities among the brood stock amounted to 31 males and 11 females at the pilot plant and 55 males and 56 females at the Kewalo plant. The dead fish were carefully examined and deaths were usually attributable to one of the following factors: Rough handling, disease, asphyxiation and, possibly, hydrogen sulfide poisoning.

While it might have been possible to reduce mortalities by using more care in handling fish and by proper treatment of diseased fish, it was not always possible to prevent mortalities caused by asphyxiation, since many factors contributed to the oxygen concentration in the tanks.

The heaviest mortality to occur in a single day among the adults was experienced at the Kewalo plant. A total of 7 males and 29 females was found dead in the two brood tanks containing brackish water (salinity-spawning experiment) and the deaths were believed to have been caused by either asphysiation or hydrogen sulfide. Faint odors of hydrogen sulfide gas were detected in the brackish-water tanks for several days before the mortalities occurred, and, undoubtedly, this gas was one of the contributing factors, if not the determining factor in the deaths. The tanks were not drained and cleaned at the time hydrogen sulfide was first detected in the tanks, however, because the experiment was to be terminated within a few days.

#### Among the Young

Many factors contributed to the loss of young fish, chief of which was disease, with deaths from handling judged to be of secondary importance. At the pilot plant, the observed mortality in the fry tank amounted to 34,784 fish, while juvenile deaths totalled 4,523 fish. Although the total observed mortality among the young amounted to

15.5 percent of the 253,548 fish produced, the unobserved mortality (difference between net production and observed distribution and losses) was much higher, amounting to an estimated 42,900 fish or about 17 percent of the total production. The causes for these unobserved losses were difficult to assess, but presumably can be attributed to a variety of factors: vandals entering the plant and removing unknown quantities of fish; loss of fish through the drain; predation by dragonfly nymphs; and cannibalism. Davis (1946: p. 9) in reporting on "uncounted mortality" in trout hatcheries stated that this was due "either to improper construction of the raceways, so that many of the fish are able to escape; to the attacks of enemies, such as fish-eating birds; or to cannibalism."

At the Kewalo plant, observed mortalities among the young amounted to 44,600 fish or about 13 percent of the 347,700 fish produced.

#### DISEASE PREVENTION AND CONTROL

Prevention and control of fish diseases and infestation of parasites are important factors in the success of any type of fish-rearing program. Fish, like other animals, are subject to a wide variety of infectious diseases and parasites and seem particularly susceptible in an unnatural or artificial environment.

Several outbreaks of disease or parasitic infestation were observed among tilapia at both the pilot plant and the Kewalo plant. All dead fish were examined microscopically for signs of unnatural conditions, such as mucous film or irregular blotches on the body and unusual blisters or swelling. When sick fish were observed in the tanks, similar examinations were made on their external surfaces, gill region and, on occasion, the gastrointestinal tract. It was not always possible to distinguish between losses from diseases and from other causes, although in many instances of high mortality rates parasites were seen and identified to genus. Several exceptions occurred when the cause of high mortality rates among the fry could not be determined by isolation of any causative organism. By careful observation of symptoms, it was possible on a few occasions to restrict the probable cause of death to a virus infection. In general, the fry, juveniles, and adults were susceptible to infection in that order.

#### Trichodina spp.

Infestation by the ectoparastic trichodinids, considered one of the most highly specialized protozoans, was the most common malady among adult and young tilapia, with infestation along the dorsal fins, dorsal region of the caudal peduncle, and gill region being most prevalent. Tilapia with trichodiniasis were sluggish, showed loss of appetite, had a reddish tinge on the skin in the caudal-peduncle region, and, in some instances, in and around the region of the dorsal fins, they had white blotches accompanied by a fraying of these fins.

According to Davis (1953: p. 220), Trichodina is very easy to control. We used several recommended treatments such as salt (1.23 percent), acetic acid (1:500), formalin (1:4,000), pyridylmercuric acetate or PMA (2 p.p.m.), copper sulfate (0.5 p.p.m.), and potassium permanganate (3 p.p.m.). All of these reagents proved effective in controlling the disease; however, as more experience was gained, we found that potassium permanganate was the most suitable, since it was exhausted after a period of time and the treated tank did not need to be flushed, as was required with the other chemicals.

#### Chilodon spp.

Only a few outbreaks of disease were attributable to this protozoan, a common ectoparasite of warm-water fishes. Davis (1953) states that this organism may be very destructive to fish crowded in small holding tanks or ponds and has been known to cause serious losses among trout fingerlings.

On a number of occasions, both adult and young tilapia were found to be infested with this parasite. Areas infested were usually the dorsal and caudal fins and along the dorsal surface of the fish, especially near the base of the dorsal fins. Very frequently, trichodinids were also present.

Treatment to rid the fish of this parasite was usually with potassium permanganate or with PMA, although the former was used more extensively because of its ease of application.

#### **Infectious Pancreatic Necrosis**

Infectious pancreatic necrosis, until recently known as acute catarrhal enteritis (Lagler, 1956; Snieszko and Wolf, 1958), was the most serious affliction of young tilapia. Outbreaks of the disease caused high mortality rates among fry about 2 to 3 weeks old, soon after they had started supplementary feeding. Symptoms characteristic of this disease were violent whirling or corkscrewing accompanied by rapid breathing. The afflicted fish usually exhibited a series of these whirling movements, then sank to the bottom of the tank and stopped feeding. Cessation of feeding caused many of the fry to have a "pinhead" appearance, that is, a large head and shrunken body.

These symptoms are similar to those described for octomitiasis, commonly called whirling disease or pinhead condition (Lagler, 1956; Davis, 1953; Snieszko and Wolf, 1958). Octomitiasis is caused by the protozoan *Octomitus salmonis*, which occurs in the intestine either in the flagellated form, when the condition is chronic, or in an intracellular stage, when the disease is acute. The etiology of infectious pancreatic necrosis, on the other hand, is still in doubt, although it has been reported as probably caused by a virus (Lagler, 1956; Snieszko and Wolf, 1958).

All of the symptoms noted here were observed in one particular outbreak of disease at the pilot plant. Dissection and examination of the stomach and anterior intestine of six afflicted fish revealed that only one fish had a protozoan in the intestine. All of the others appeared normal internally. On the assumption that this disease was probably octomitiasis, we started immediate treatment with Carbarsone (p-Ureidobenzene arsonic acid) at the rate of 1 gram per pound of food (Davis, 1953). The mortality rate decreased appreciably in the next few days following treatment and by the end of the tenth day had been reduced to a low level. Although Carbarsone seemingly effected a cure, we are not certain that Octomitus was the causative organism, since the protozoan was not positively identified. It might possibly have been infectious pancreatic necrosis, or an acute infection of octomitiasis caused by the intracellular stage of the flagellate, or a combination of the two. Snieszko and Wolf (1958) state that many cases diagnosed as octomitiasis are in reality infectious pancreatic Careful microscopic examination is necrosis. necessary for correct diagnosis.

Subsequent periodic outbreaks of infectious pancreatic necrosis were definitely identified by examination of the stomach and anterior intestine of diseased individuals. In most instances these organs were distended and filled with a colorless, opaque fluid, indicating a stoppage of bile flow. As there is no known effective therapy for this disease, the usual procedure was to treat the fish with potassium permanganate at a concentration of 3 p.p.m. to prevent secondary infection of the weakened fish with ectoparasites and to observe strict sanitation measures. In this way, we believe that most of the disease outbreaks were kept localized.

Mortality rates among the fry were usually highest during the first week after an outbreak and gradually subsided during the following 2 to 3 weeks. The most serious outbreak of disease occurred at the Kewalo plant soon after it was in operation. A succession of infections that spread among the newly emerged fry caused an estimated loss of about 80 percent of the 146,776 fry produced in a 3-month period. The disease was controlled after 10 weeks.

#### **Prophylactic Measures**

Despite knowledge that the density of fish is extremely important in relation to the outbreak and spread of disease, we tended to overcrowd our tanks on occasion. Since the amount of fryrearing facilities was limited, it was impossible to avoid overcrowding during periods of heavy fry production unless we discarded some of the young fish. Our records indicate that several of the disease outbreaks were directly associated with periods of high production. Whenever an overcrowded condition existed, we made every effort to prevent disease outbreaks by increasing the rate of the inflowing water, by cropping the frytank compartments frequently, and by observing strict sanitation.

Effective prophylactic measures are probably of greater importance than control measures in successful fish culture. Potassium permanganate was periodically added to the water as a prophylactic measure and, when conditions permitted, tanks which held any diseased fish were thoroughly scrubbed with a brush, refilled with water, and copper sulfate or formalin was added in high concentration. The environment thus created was believed unfavorable to whatever ectoparasites may have remained in the tank after the scrubbing.

#### FACTORS AFFECTING FRY PRODUCTION

In commercial bait-rearing operations, it is necessary to obtain maximum production and survival of young from a minimum-sized brood stock. Insofar as possible, optimum conditions of temperature, salinity, food, sex ratio, and brood-stock density are maintained. Some preliminary information was obtained at the pilot plant and more detailed information at the Kewalo plant on the importance of these factors.

#### **TEMPERATURE AND SPAWNING**

Fry production started in December 1956 at the pilot plant, and by mid-January 1957 it was apparent that the brood stock in tank 3 was not as productive as that in tank 1.

Environmental conditions in the two tanks differed in several respects. The water temperature in tank 3 averaged 1° to 2° F. cooler than in tank 1. This difference probably arose from the fact that tank 3 was shaded by trees and a storage shed most of the day, whereas tank 1 was situated in more open surroundings and received more hours of direct sunlight. In an attempt to eliminate the temperature difference, the water entering tank 3 was piped through a 100-foot length of black, three-fourths-inch, garden hose that was stretched across the roof of the storage shed. On sunny days, the temperature of the water flowing into tank 3 was raised as much as 5° to 10° F. and the temperature of the water in the tank to about the same level as in tank 1 (table 5).

A second major difference between the tanks was the consistently lower oxygen concentration in tank 3. This condition probably accounted for the dead yolk-sac fry that were frequently found in the detritus siphoned from the floor of the tank. Aeration brought about a significant increase in the concentration of oxygen in tank 3 (appendix table 1).

The pattern of water circulation was also different in the two tanks. The flow of water from inlet to outlet in tank 3 opposed the prevailing wind while that in tank 1 was in the direction of the prevailing wind. The resulting circulation pattern kept tank 1 relatively clean and the water usually green with algae, while tank 3 had much detritus and generally stagnant water. The color of the water in tank 3 varied from clear to

 TABLE 5.—Average minimum and maximum temperatures
 (° F.) in tanks 1 and 3 at the pilot plant

	Та	1k 1	Таг	1k 3
Month	A verage to	emperature	Average te	emperature
	Minimum	Maximum	Minimum	Maximum
1957:				
January	70.8	· 75.6	70.1	75. 5
February	69.8	75.2	68.6	75. 2
March		78.0	69.8	78.2
April	71.3	78.0	71.2	78.9
May June	72. 8 74. 7	81.3 82.3	73.2 75.6	81.3 82.5
July		83.3	76.5	82.2
August	75.7	82.6	75.6	81.1
September	(1)		76.0	81.9
September October	(1) (1)	(1) (1)	75.2	80.3
November	72.8	77.0	74.2	78.2
December	70. 3	74.5	70.7	74.4
1958:				
January		74.9 76.7	70.2 70.7	74.0
February March	70.6	77.3	70.8	75.7
April		79.6	71.6	78.1
May		80.1	72.8	77.8
June		82.9	73.8	79.5
July	75.4	82.6	73.9	79.2

<sup>1</sup> No data; thermograph out of order.

light green to dark brown. Improvement in water circulation in tank 3, resulting from aeration and slight heating of the water, was sufficient to produce a significant increase in fry production during March 1957. Tables 6 and 7 give the production per female per month for these brood tanks (tank 2, which was later converted to a brood tank in August 1957, was similar in all respects to tank 1).

 TABLE 6.—Fry produced in tank 1, per female and month, at the pilot plant, December 1956–July 1958

Month	Broo	d stock		er of fry luced	Feed		
	Males	Females	Total	Per female 1			
1956: December.	ecember. 32		578	9.0	Chicken mash and ric bran.		
1957:							
January	32	64	4,498	70.3	Rice bran.		
February	32	64	2, 107	32.9	Pondfish feed.		
March	32	64	6, 433	100.5	<u>D</u> o.		
April	32	64	4,050	63.3	Do.		
May	32	64	20, 297	317.1	Do.		
June	31	64	6,146	96.0	Do.		
July	31	64	20,910	326.7	Do.		
August	31	64	18, 520	289.4	Chicken mash.		
September.	31	64	7,743	121.0	Do. Do.		
October	31 29	64 64	4,159	65.0	Alfalfa pellets an		
November	29	04	188	2.9	pondfish feed.		
December	31	64	409	6.4	Do.		
1958:	91	01	409	0.4	D0.		
January	19	59	0	0.0	Pondfish feed.		
February	22	61	143	2.3	Do.		
March	19	58	48	0.8	Do.		
April	19	58	1,418	24.4	Do.		
May	17	58	10,701	184.5	Rabbit pellets.		
June	17	58	9,158	157.9	Do.		
July	12	58	6,172	106.4	Do.		

<sup>1</sup> Because of mortalities and transfer of fish, the number of females used to calculate the production per female is based on the number of females that were present in the tank for more than 2 weeks.

Month	Broo	d stock	Numb prod	er of fry luced	Feed
	Males	Females	Total	Per female 1	
1956: December.	16	32	69	2. 2	Chicken mash and rice bran.
1957: January	55	93	0	.0	Rice bran and pondfish feed.
February	31	. 63	374	5.9	Pondfish feed.
March	31	63	1,278	20.3	Do.
April	31	62	8, 322	134.2	Do.
May	31	62	13, 685	220.7	Do.
June	31 31 31	62	9, 290	149.8	Do
July	31	62	14, 197	229.0	Do.
August	31	62	9,327	150.4	Do.
September	31	62	2, 621	42.3	Do.
October	31	62	1,769	28.5	Do.
November	31	62	1, 294	20.9	Do.
December	31	62	274	4.4	Do.
1958:		1 1			
January	31	62	32	.5	Do.
February	30	61	147	2,4	Do.
March	29	61	213	3.5	Do.
April	29	61	· 566	9.3	Do.
May	29 28	61	3, 205	52.5	Rabbit pellets.
June	28	61	<b>78</b>	1.3	Do.
July	20	60	92	1.5	Do.

 TABLE 7.—Fry produced in tank 3, per female and month, at the pilot plant, December 1956–July 1958

<sup>1</sup> Because of mortalities and transfer of fish, the number of females used to calculate the production per female is based on the number of females that were present in the tank for more than 2 weeks.

A second experiment in which the effect of temperature on production of young was examined was conducted at the Kewalo plant from January to August 1959.

For tilapia to be most useful as a supplementary skipjack bait, there must be a stock of bait-size fish on hand in May or June, at the beginning of the main fishing season in Hawaiian waters. To achieve this, heavy fry production must be under way by late winter. Brock and Takata (1955: p. 24) reported that tilapia spawn throughout the year in Hawaiian waters, but that the spawning is less intense during the winter months. Consequently, an experiment was conducted at the Kewalo plant to determine if raising water temperatures would induce tilapia to spawn at a high rate during the winter months. Three redwood tanks (tanks 13, 14, and 15) were arranged as follows for the experiment:

1. Tank 13 was not modified in any way and served as the control.

2. The water in tank 14 was artificially heated with a 60-foot, lead-sheathed heating cable rated 3.63A-115V, that produced 400 watts, or 6.7 watts per foot. A thermostat with a capillary tube was placed in the tank to control the temperature. A cover made of sisal-glaze, a clear, longlasting plastic, was placed over the tank (fig. 8) to prevent excessive heat loss, especially at night.

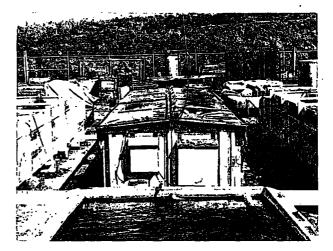


FIGURE 8.--Plastic cover over artificially heated tank 14.

3. The water in tank 15 was also artificially heated with the same type of heating cable used in tank 14, but the tank was not covered to prevent heat loss.

Each tank was stocked with 32 males and 64 females and fed trout feed. Emerging fry were collected and counted daily. The daily variation in water temperature was recorded by thermographs and minimum and maximum thermometers.

The production per female per month and the average and range of minimum and maximum temperatures for each of the three tanks are recorded in table 8 and shown graphically in figure 9. In January, tank 14, which was modified with heating cable and cover, had relatively better production than the other two tanks. In February, artificial heating was started in tank 15, and resulted in a marked increase in production comparable to that in tank 14. Production was still low in the control tank. In March, with rising air temperatures the water warmed in all three tanks and production increased in all; however, the increase in the slightly warmed tank 15 greatly exceeded that in the other tanks. Temperatures remained about the same in April, but production dropped, particularly in tanks 14 and 15. With higher temperatures in May, the control tank maintained its slight lead over tank 15, and during the last 3 months of the experiment, it outproduced the two artificially heated tanks by a significant margin. Over the course of the experiment, total fry production in tank 13 (control) was about twice that in the other two tanks.

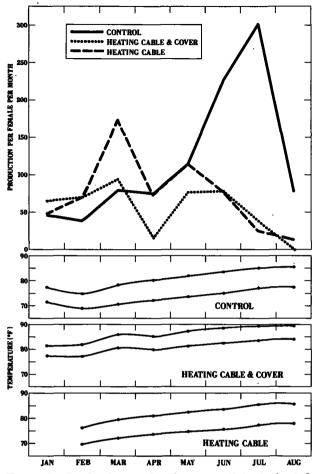


FIGURE 9.—Production per female per month in the heated tanks and in the control tank, and average minimum and maximum water temperatures in the tanks.

Analysis of the data indicates that although some increase in spawning was induced during the winter months by raising the water temperature, the increase in production was not great. In addition, only a slight rise in water temperature apparently produced the same results as a marked increase in temperature. The results also indicate that prolonged or constant high temperatures may detrimental to spawning. Innes (1951) be pointed out that at constant high temperatures the oxygen content of the water becomes diminished and this seems to have a weakening effect on fish. Doudoroff (1957: p. 415) stated-

that persistent nonlethal deficiency of dissolved oxygen undoubtedly can adversely influence the activities of fishes and have serious detrimental effects on fish populations in their natural environments.

The oxygen record (appendix table 4) discloses that throughout the period of the experiment, tank 14 had generally lower concentrations of oxygen, which may account for the decrease in fry production during the summer months.

Although tanks 13 and 15 had similar temperatures, fry production in these two tanks differed significantly for the months of June to August. While it is true that tank 15 was heated artificially (thermostat set at 80° F.), much of the heat was lost through convection. Thus, tank 15 remained only slightly warmer than the control tank. The average temperatures in both tanks fell within the optimum temperature range for propagation, which is from 20° to 35° C. (68° to 95° F.) according to Chen (1953), while the weekly oxygen determinations showed very little difference between the two tanks. We are unable to advance a satisfactory explanation for the poor fry production in tank 15.

 TABLE 8.—Summary: Fry production and temperatures of brood tanks used in the heating experiment, January– August, 1959

	[Ten	nperature	° F.]			
Month	Numbe prod	er of fry úced		e tem- ture	Temperar	erature 1ge
	Total	Per . female	Mini- mum	Maxi- mum	Mini- mum	Maxi- mum
Tank 13 (control): January February March April May June July August	2, 967 2, 492 5, 032 4, 739 7, 365 14, 456 19, 319 5, 010	46. 4 38. 9 78. 6 74. 0 115. 1 225. 9 301. 8 78. 3	71. <b>4</b> 67. 9 70. 6 72. 1 73. 5 75. 0 77. 0 77. 3	77.4 74.8 78.2 80.1 81.8 83.6 85.1 85.4	63.0 64.2 67.7 67.1 70.1 72.6 74.3 72.3	83. 4 78. 5 81. 9 84. 6 86. 4 86. 2 90. 4 90. 3
Total Tank 14 (with heating cable and cover): January February March March May June June	61, 380 4, 064 4, 448 5, 922 992 4, 804 4, 935 2, 376	63. 5 69. 5 92. 5 15. 5 75. 1 77. 1 37. 1	77.3 77.2 80.4 79.8 81.2 82.5 83.5	81. 4 81. 8 85. 9 85. 2 87. 2 88. 6 88. 6 89. 4	72.1 74.8 78.0 76.0 76.8 81.0 79.0	88.3 84.6 88.0 90.7 90.2 90.6 90.9
August Total Tank 15 (with heating cable):	0 27, 541	0.0	84.1	89.5	79.8	<u>90.9</u>
January February March April June July August	3,046 4,424 11,166 4,660 7,269 4,905 1,555 822	47.6 69.1 174.5 72.8 113.6 76.6 24.3 12.8	69.6 72.1 73.6 74.8 75.7 77.3 77.9	76.0 79.6 81.0 82.6 83.7 85.4 85.4 85.6	65.0 69.0 69.0 72.0 73.6 74.0 72.8	78.0 83.0 85.0 87.0 85.4 89.9 89.4

37.847

Total.....

#### SEX RATIO AND BROOD-STOCK DENSITY

A ratio of  $2 \circ 1 \delta$  was used initially at the pilot plant and judged to yield satisfactory results. The best production resulting from this sex ratio was realized during July 1957 at the pilot plant, when production in tank 1 amounted to 20,910 fry or 327 fry per female (table 6). A second grouping of  $5 \circ 1 \delta$  (total stock, 72) was also tried in tank 2 from August to November, 1957. The production per female per month is given in table 9. The best production for that period and grouping occurred in August, when fry production amounted to 2,532, or 42 fry per female.

 TABLE 9.—Production of fry per female per month in tank 2

 at the pilot plant, August 1957–July 1958

Month	Brood	l stock		er of fry luced	Feed	
	Males	Females	Total	Per female <sup>1</sup>		
1957:				·]		
August	12	60	2,532	42.2	Pond-fish feed.	
September	12	60	2, 383	39.7	Do.	
October	12	60	1,535	25.6	Do.	
November	12	60	Ý 0	0.0	Do.	
December	16	56	107	1.9	Do.	
1958:						
January	16	56	3	0.1	Do	
February	16	56	2,891	51.6	D0.	
March	16	56	7, 691	137.3	Do.	
April	16	56	6,660	118.9	Do.	
May	31	72	11,637	161.5	Rabbit pellets.	
June	31	72	19, 826	275.4	Do,	
July	29	72	7,781	108.1	Do.	

<sup>1</sup> Because of mortalities and transfer of fish, the number of females used to calculate the production per female is based on the number of females that were present in the tank for more than 2 weeks.

A more detailed experiment designed to determine the ideal sex ratio for maximal yield of fry was conducted at the Kewalo plant from September to November 1958. The effect of broodstock density on spawning was simultaneously investigated in the same experiment.

The 12 assault boats mentioned earlier were stocked with various sex ratios and concentrations of fish. The design of this partially confounded factorial experiment is shown in table 10. The brood stock was fed dry trout feed at the rate of 2 percent of its weight daily. It was assumed that any differences in temperature or oxygen concentrations which might occur among the tanks would not bias the results.

TABLE 10.—Sex ratios and concentrations of tilapia used to stock the 12 assault-boat tanks, September-November 1958

Sex ratio			Sex ra	tio in cor	ncentrati	on of		
(२:४)	30 1	fish	50	fish	70	fish	90 fish	
Ç 0"	Ŷ	ď	ę	ð	ę	ď		
2:1 3:1 4:1	20 23	10	33 38	17 12	47	23	68	22
4:1 6:1	24	6	43	7	56 60	14 10	68 72 77	18 13

The experiment was terminated after 13 weeks. An analysis of variance of the data (table 11) indicated significant differences (F=5.73, P< 0.01) between fry production with respect to the sex ratios used in the experiment. (A probability level of 5 percent is considered the maximum value for a conclusion of significance in this report.) The most productive ratio was 3  $\mathfrak{P}$  : 1  $\mathfrak{F}$  (tanks 4 and 5). Tanks 1 and 2 with a ratio of  $2\mathfrak{P}$  : 1  $\mathfrak{F}$ were almost equally high in production for the first 2 months of the experiment (fig. 10).

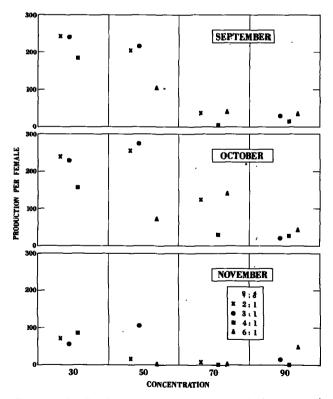


FIGURE 10.—Production per female in relation to sex ratios and concentrations of brood stock.

The relation between total production per female and bottom area (square feet) per male is shown in figure 11, while the total production per female in relation to bottom area per individual is shown in figure 12. From table 11 and

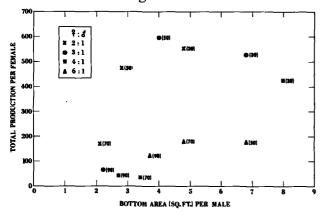


FIGURE 11.—Total production per female in relation to bottom area (sq. ft.) per male. (Total stock in parentheses.)

figures 10 to 12, it may be seen that the tanks with 50 fish or less (except tank 10) had a much better production than those with more, while an allowance of 4 square feet of bottom area per male and 1 square foot per individual gave the highest production per female (see tank 5, table 11).

The analysis of variance showed that the differences in fry production with respect to the concentrations, 30, 50, 70, and 90 fish, used in the experiment were significant (F = 11.44, P < 0.01). There was also a significant interaction between sex ratios and brood-stock density, indicating that the production resulting from any specific ratio did not vary in a uniform manner with respect to availability of space.

#### DIET AND REPRODUCTION

Early attempts to assess the qualities of different types of feed in relation to fry production at the pilot plant proved inconclusive, owing to dissimilar conditions in the brood tanks (tanks 1 and 3). However, we were able to observe the general acceptability of the five types of feed that were used.

Finely ground feed, such as rice bran and chicken mash, was found to be unsuitable for the adults as they cannot strain small particles from the water. Much of the feed was wasted and

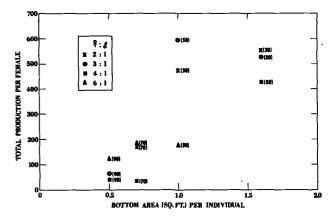


FIGURE 12.—Total production per female in relation to bottom area (sq. ft.) per individual. (Total stock in parentheses.)

TABLE 11.—Sex ratio and concentration of brood stock and production of fry, September-November, 1958

Item						Tank	No. —					
	1	2	3	4	5	6	7	8	9	10	11	12
Sex ratio (9: 7) Number of fish:	2:1	2:1	2:1	3:1	3:1	3:1	4:1	4:1	4:1	6:1	6:1	6:
Females	20 10	33 17	47 23	23 7	38 12	68 22	24 6	56 14	72 18	43 7	60 10	7 13
Total fish	30	50	70	30	50	90	30	70	90	50	70	
Bottom area (sq. ft.): Per male Per fish Production:	4.8 1.6	2.8 1.0	2. 1 0. 7	6.8 1.6	4.0 1.0	2.2 0.5	8.0 1.6	3.4 0.7	2.7 0.5	6.8 1.0	4. 8 0. 7	3. 0.
September: Fry produced Fry per female October:	4, 872 243. 6	6, 762 204. 9	1, 7 <b>23</b> 36. 6	5, 567 242. 0	8, 209 216. 0	2, 077 30. 5	4, 426 184, 4	272 4.8	1, 043 14. 5	4, 459 103. 7	2, 471 41. 2	2, 62 34. 1
Fry produced Fry per female November:	4, 775 238. 8	8, 402 254. 6	5. 891 125. 3	5, 292 230. 1	10, 443 274. 8	1, 396 20. 5	3, 766 156. 9	1, 620 28. 9	1, 955 27. 2	3, 099 72. 1	8, 376 139. 6	3, 08 40.
Fry produced Fry per female Total:	1, 441 72. 0	504 15. 3	. 334 7.1	1, 287 56. 0	4, 036 106, 2	921 13. 5	2, 054 85. 6	20 0.4	37 <sup>.</sup> 0. 5	28 0.6	36 0.6	3, 56 46.
Fry produced Fry per female	11, 088 554. 4	15, 668 474, 8	7, 948 169, 1	12, 146 528. 1	22, 688 597. 0	4, 394 64. 6	10, 246 426. 9	1, 912 34. 1	3, 035 42. 2	7, 586 176. 4	10, 883 181. 4	9, 28 120.

tended to foul the tanks. On the other hand, pelletized feed, such as pondfish and trout feed, was consumed by the adult fish with little wastage. Rabbit feed had a high percentage of crude fiber, which seemed to pass through the fish undigested and left much residue in the tank. Alfalfa pellets were somewhat less acceptable than the other feeds mentioned, probably because of their large size and their high fiber content.

An experiment to evaluate the effects of different types of feed in relation to fry production was conducted at the Kewalo plant in September-November, 1958. For this experiment, the three redwood tanks (tanks 13, 14, and 15) were each stocked with 32 males and 64 females. The adults in tank 13 were fed trout feed, which we believed to be high in nutritional value and which was relatively expensive. Those in tank 14 were fed rabbit feed. As stated earlier, this feed was acceptable to the tilapia and was also much cheaper than trout feed. Preliminary trials at the pilot plant indicated that relatively good fry production was possible with this feed. The fish in tank 15 were fed millrun, which was locally produced and the least expensive of the feeds tested.

Table 12 gives the production per female for the 3 months that the experiment was conducted. A plot of the number of fry per female per month is shown in figure 13. It is evident that the fish that were fed trout feed (tank 13) produced the greatest number of fry per female, while the fish that were fed rabbit feed (tank 14) and millrun (tank 15) had very low fry production.

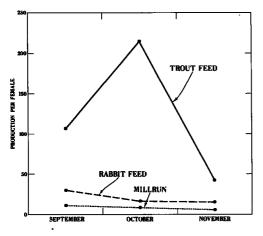


FIGURE 13.—Effect of type of feed given brood stock on production per female.

Table	12.—Production	per j	female	in	three	brood	tanks,	by
	type of feed, S	epten	nber–N	ove	mber,	1958		-

	Tank 13 (trout feed)	Tank 14 (rabbit feed)	Tank 15 (millrun)
September:			
Fry produced	6, 863	1,874	751
Fry per female	107.2	29.3	11.7
October:			
Fry produced	13, 769	1,074	529
Fry per female	215.1	16.8	8.3
November:	0.007	000	
Fry produced	2,695	982	394
Fry per female	42.1	15.8	6.2
Total:	00.007	0.000	1 074
Fry produced	28, 327	3,930	1,674
Fry per female	364.5	61.4	26.2

Temperature was not considered a factor in fry production in this experiment, since there was very little difference in water temperatures among the tanks. Assuming that other environmental factors were similar among the tanks, it follows from this experiment that the use of a nutritionally balanced feed is highly important in obtaining good fry production. We are not certain, however, of the long-range effect of such high-protein feeds on tilapia, which are principally herbivorous. These trout feeds are manufactured primarily for trout culturists who intend to market the fish rather than use them as brood stock-they may possibly be detrimental to spawning fish. Schaeperclaus (1933: p. 98) stated that the ovaries of troutunder intensive artificial feeding-degenerate and produce few usable eggs. It is likely that spawning stocks of tilapia held for extended periods of time should be fed a diet more in keeping with their natural food or be replaced after a year or two by a new stock of brood fish grown under more natural conditions.

#### SALINITY AND SPAWNING

During the summer of 1959, the Hawaii Division of Fish and Game started plans for construction of a tilapia hatchery in an area with free accesss to brackish water (about  $10 \, {}^{\circ}/_{\infty}$ ). At the Division's request, experiments dealing with spawning and growth of young tilapia in brackish water were initiated at the Kewalo plant.

Several investigators have observed and reported that tilapia will spawn in a saline environment. Vaas and Hofstede (1952: p. 11, 16) reported that spawning occurred in a period during which the salinity of the water ranged from 3 to 4.8 percent. They pointed out, however, that "According to subsequent findings of the Extension Service in Indonesia, good growth is limited by a salinity of 4 percent and spawning by one of 3 percent." Brock (1954) reported tilapia spawning in sea water of a chlorinity of  $19.29 \circ/_{00}$ (equal to a salinity of about  $34.85 \circ/_{00}$ ) and a pH of 7.95.

Four tanks were used in this experiment: two test tanks contained brackish water and two controls contained fresh water. The experiment was started in August 1959 and terminated during the latter part of September when heavy mortalities among the adults caused by either asphyxiation or hydrogen sulfide occurred in both brackishwater tanks.' Each tank was stocked with 36 females and 12 males, that is, with a 3:1 sex ratio, and at a concentration of 1 square foot per individual.

The yield of fry for the 2 months of the experiment is given in table 13. The total production was almost three times as great from the treatment tanks (brackish water) as from the controls. Analysis of variance indicated a significant difference (F=472.65, P<0.05) between fry production with respect to brackish- and fresh-water methods of culture, but no significant difference between replicates.

Exactly what influence the saline environment. exerts in bringing about this increased fry production is not known, but the results of this experiment emphasize the desirability and possibilities of tilapia culture in brackish water.

 
 TABLE 13.—Fry production in brackish-water and freshwater tanks, 1959

	August	September	Total
BRACKISH WATER			
Salinity (°/): Minimum			
Minimum Maximum	8.9 12.3	9.4 15.2	
Average Tank 1:	10.9	13. 2	
Fry produced Fry per female	11, 845 329, 0	15, 519 431, 1	27, 364
Tank 3: Fry produced Fry per female	17, 319 481. 1	11, 745 326, 2	29,064
FRESH WATER			
Tank 5: Fry produced Fry per female Tank 12:	6, 479 180. 0	1, 169 32. 5	7, 648
Fry produced Fry per female	8, 910 247. 5	2, 172 60. 3	11,082

#### FACTORS AFFECTING GROWTH OF YOUNG

The effects of environmental factors on growth of young tilapia were studied experimentally at the Kewalo plant for 12 weeks, beginning in April 1959. The effects of space or density of the fish, diet, and salinity were examined during the experiment.

We tried to vary one factor at a time, keeping the others constant, so that the single factor under observation could be evaluated with some degree of precision. We did not attempt to control the temperature, since differences among the tanks were not significant. Other factors, such as volume of water in the tank and the rate of water flow, were held as uniform as possible. In each experiment, the fish were fed at the same rate per fish regardless of lot size. An excellent quality trout feed was fed to the fish twice a day, except weekends, at the regular feeding times.

Once each week, length and weight measurements were made on a randomly collected subsample from each tank under observation. All of the fish in the subsample were returned to their respective tanks after measurements were completed. The number of fish in a subsample varied with each experiment.

We realize that, ideally, all phases of the experiments should have been conducted simultaneously. This was not possible, however, because of a lack of sufficient quantities of fry. As a result, different phases of the experiments were commenced as fry became available in adequate amounts to stock the tanks.

#### CONCENTRATION OF FISH AND GROWTH

A series of tests to determine the rate of growth of young tilapia in relation to their concentration, or the amount of space available for growth, was started at the Kewalo plant in April 1959. Five lots of fry, ranging in number from 1,000 to 6,000, were placed in the assault-boat tanks; however, all tanks were not stocked simultaneously.

The fish were fed trout feed (starter, fry, and small fingerling grades) for 12 weeks. The particle size and amount of feed was gradually increased as growth progressed. The fish were fed twice daily except on weekends, when they were fed once a day. Twenty-five fish were measured from each of the lots of 1,000, 2,000, and 3,000 fish, 40 from the lot of 4,000 fish, and 50 from the lot of 6,000 fish. Table 14 gives the weekly averages of length and weight of fish in the randomly collected subsamples, while figure 14 shows the regression lines fitted to the growth data.

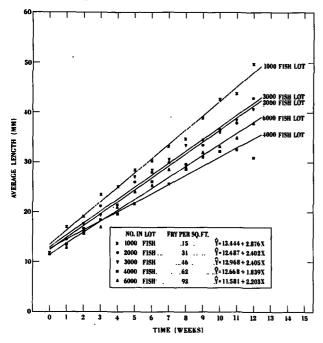


FIGURE 14.—Regression of length on time for five lots of tilapia reared in various concentrations of fish in tanks.

Examination of the regression coefficient from each lot indicated that young in the tank stocked with 1,000 fish had the best growth rate (2.9 mm. per week) during the 12-week period, and that growth rates were somewhat slower with an increasing number of fish per lot. Fish in tanks stocked with 2,000 and 3,000 fry had almost identical growth rates (2.4 mm. per week) but less favorable growth than the 1,000-fish lot. The lot of 6,000 fish, for some unknown reason, had a better growth rate than the lot containing 4,000 fish (2.2 and 1.8 mm., respectively, per week).

Statistically, the differences in growth among the five lots of fish were significant (F=41.92, P<0.01). From the results of the experiment, it follows that the ideal stocking density for nursery waters (fry tanks) would be 9 fry per cubic foot or 15 fry per square foot of surface area (1,000-

 TABLE 14.—Weekly average length (mm.) and weight (g.) and average absolute growth rates in random subsamples of tilapia from various concentrations of fry, 1959

	-				
	1,000 fry	2,000 fry	3,000 fry	4,000 fry	6,000 fry
Experiment began	May 29	May 29	Apr. 9	May 22	Apr. 9
Experiment ended	Aug. 21	Aug. 21	July 2	Aug. 14	July 2
Initial length	12.4	12.0	11.9	11.8	11.9
Initial weight	0.024	0.012	0.024	0.012	0. 024
1st week:					
Length	17.0	14.5	13.6	13.5	12.8
Weight	0.072	0.040	0.040	0.030	0.040
2d week:					
Length	19.2	16.7	17.6	16.0	15.7
Weight	0.116	0.068	0.096	0.052	0.062
3d week:		0.000	0.000	0.00-	
Length	23.5	21.2	19.3	18.3	16.8
Weight	0.168	0.148	0.094	0.092	0.062
4th week · í					
Length	25.1	21.5	25.0	19.7	21.1
Weight	0.284	0.176	0.272	0.115	0.148
5th week:					
Length	28.3	26.1	27.2	21.7	24.0
Weight	0.400	0.320	0, 308	0.152	0. 224
6th week:			0.000		
Length	30.3	28.0	28.4	26.2	25.3
Weight	0.480	0.400	0.416	0.312	0.264
7th week:	0. 200	0.100	0.110	0.012	0. 20.
Length	33.2	29.9	30.6	25.7	28.4
Weight	0.640	0.492	0.500	0.300	0.432
8th week:	0.010		0.000		
Length	34.7	29.6	33.5	28.6	29.2
Weight	0.792	0,500	0.705	0.412	0.484
9th week:			0.100	0	
Length	39.1	33.4	34.5	31.1	31.9
Weight	1.116	0.692	0.780	0.578	0. 628
10th week:		0.001	0.100		
Length	42.7	36.6	36.2	32.2	33.2
Weight.	1.448	0,960	0.844	0.605	0. 670
11th week:		0.000	0.011	{ 0.000 }	0.011
Length	43.8	37.8	38.4	32.5	34.8
. Weight	1.596	0.964	0.964	0.615	0.70
12th week:	21000	0.001	0.001	0.010	
Length	49.8	42.8	40.7	30.8	37.8
Weight	2.408	1. 528	1.280	0.612	0.972
•		<sup></sup>			
Average absolute growth			)	1	
rate	3.1 mm.	2.6 mm.	2.4 mm.	1.6 mm.	2.2 mm

fish lot). Some lesser concentration, as yet undetermined, might prove even better. However, from a practical standpoint, these very low stocking rates might not be the most feasible economically, as the space requirements and construction costs for fry-rearing tanks would be enormous in a commercial operation.

Earlier, we mentioned that newly emerged fry almost always were captured near the surface of the water. Upon transfer to nursery waters, the fry continue to exhibit this behavior. Therefore, we believe that stocking density should be related to surface area rather than to volume of water.

#### DIET AND GROWTH

An experiment to compare the growth rates obtained on an inexpensive, commercially available feed and the more expensive, but highly nutritious, trout feed was initiated in April 1959 simultaneously with the experiment on relation of space to growth at the Kewalo plant. An assault-boat tank was stocked with 3,000 fry and fed exclusively with wheat white middlings, a locally available livestock feed, while the lot of 3,000 fry in the space-growth experiment was fed trout feed.

The weekly average lengths and weights of a random subsample of 25 fish from each of the two lots, for the 12-week period of the experiment, are presented in table 15. Regression lines were fitted

**TABLE 15.**—Weekly average length (mm.) and weight (g.) and average absolute growth rates in random subsamples of tilapia from 2 lots of 3,000 fry reared on different diets, 1959

Item	Trout feed	Wheat white mid- dlings
Experiment began Experiment ended Initial length Initial weight	Apr. 9 July 2 11.9 0.024	May 15 Aug. 7 12.1 0.014
1st week: Length Weight 2d week:	13.6 0.040	13.7 0.032
Weight	17.6 0.096	16.9 0.064
Ueight	19.3 0.094	18.9 0.104
Weight	25. 0 0. 272	21.0 0.160
Length	27. 2 0. 308	21.5 0.144
Length	28.4 0.416	23.5 0.424
Length Weight 8th week:	30.6 0.500	27.1 0.392
Length Weight 9th week:	33.5 0.705	28.6 0.380
Length Weight 10th_week:	84.5 0.780	30.3 0.520
Length Weight 11th_week:	36.2 0.844	31.7 0.620
Length Weight 12th week:	38.4 0.964	34.0 0.672
Length	40.7 1.280	33.9 0.768
Average absolute growth rate	2.4 mm.	1.8 mm.

to the growth obtained in these tanks. The regression coefficient for the lot fed trout feed was 2.4 mm. per week while that for the lot fed wheat white middlings was 1.9 mm. per week (fig. 15). An analysis of variance of the results disclosed a significant difference in growth rates between the fish in the two tanks (F=31.09, P<0.01), indicating that quality of the feed is highly important where fast growth rates are desired.

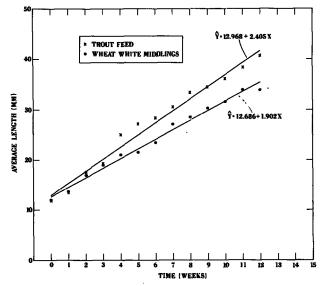


FIGURE 15.—Regression of length on time for two lots of fish reared on different diets.

#### SALINITY AND GROWTH

It has been reported by Vaas and Hofstede (1952) and Brock (1954) that *T. mossambica* will spawn and the young will grow in salt water. To determine growth rates under varying degrees of salinity, preliminary experiments were conducted at the Kewalo plant laboratory using 30-gallon aquariums.

Five 30-gallon aquariums were used in the experiment: two containing fresh water, two brackish water (salinity of about  $16 \,^{\circ}/_{oo}$ , and one sea water. Each aquarium was stocked with 200 fry. Those fish to be tested in brackish water and in sea water were acclimatized to sea water for a period of 24 hours before being placed in the aquariums. The fish in one tank in each pair of the fresh-water and brackish-water aquariums were fed wheat white middlings, while fish in the other two tanks and in the sea-water tank were fed trout feed (starter).

From the first week, a large number of deaths occurred in the brackish- and sea-water tanks and the deaths continued to occur for several weeks. At first, the dead fish in each of the tanks were removed and counted each morning and replaced with an equal number of individuals of similar size from a reserve stock which was held in sea water in another aquarium. Because of the high mortality rate and the low growth rate in all the

 TABLE 16.—Differences in average length (mm.), weight
 (g.) and average absolute growth rates of 5 lots of 200 fish, in relation to types of water and feed

Item	Aquarium No								
	1	2	3	4	5				
Type of water Type of feed	Fresh Trout	Fresh Wheat white mid- dlings.	Brackish_ Trout	Brackish. Wheat white mid- dlings.	Sea. Trout.				
Initial length of of fry	12.8	13. 3	13. 3	13. 2	13.0.				
Initial weight of fry Length at 5 weeks_ Weight at 5 weeks_	0. 03 17. 9 0. 12	0. 03 18. 3 0. 12	0. 03 17. 8 <sup>1</sup> 0. 10	0. 03 18. 0 0. 12	0. 03. 18.2. 0.14.				
Average absolute growth rate Total deaths	1.02 mm 66	1.00 mm 95	0,90 mm 122	0.96 mm 208	1.04 mm 333.				

<sup>1</sup>These figures are based on length-weight measurements at end of the fourth week.

experimental tanks, the experiment was terminated at the end of 5 weeks.

A random subsample of 10 fish was collected from each aquarium and the fish measured once each week during the 5-week period (table 16). The results were not suitable for statistical analysis. A general summary (table 16) shows that mortality was lowest in fresh water and highest in sea water. Differences in growth rate were slight and most likely of no biological significance.

A second experiment, which dealt with the effects of salinity on growth of the young, was conducted simultaneously with the experiment on

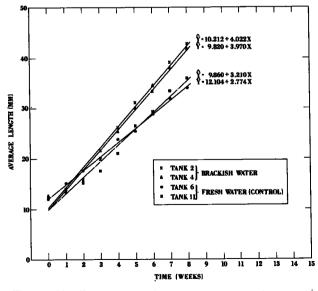


FIGURE 16.—Regression of length on time for young tilapia reared in brackish and in fresh water.

the effect of salinity on the rate of reproduction.

Four assault-boat tanks, two containing brackish water with a salinity of about 10  $^{\circ}/_{\infty}$  and two with fresh water serving as controls, were stocked simultaneously with 4,000 fry. The fish were fed trout feed twice a day. Salinity determinations (by hydrometer) of the brackish-water tanks were made daily (table 13 lists average and range of salinities for the 2 months of the experiment). Length and weight measurements were made weekly for 8 weeks on a random subsample of 40 fish from each tank. The results of the experiment are recorded in table 17, while the regression lines describing the growth in each tank are shown in figure 16.

It is evident from the regression coefficients that the brackish-water environment had a pronounced influence on the growth rate of the young fish. Here again, we are not certain of the effect of the saline environment on young tilapia, but presumably it alters metabolic processes enough to affect growth considerably. An analysis of variance indicated a significant difference in growth between treatments (F=39.94, P<0.01), and no significant difference in growth between replicates.

Another interesting aspect of this experiment was that the growth rates of the fish in the fresh-

TABLE 17.—Average length (mm.) and weight (g.) and average absolute growth rates of random subsamples of tilapia from 4 lots of 4,000 fry reared in brackish and fresh water

. Item	Brackis	h water	Fresb	water
	Tank 2	Tank 4	Tank 6	Tank 11
Initial length Initial weight	12.4 0.028	12.6 0.030	12. 0 0. 025	12. 1 0. 025
lst week: Length Weight	14. 1 0. 042	13.5 0.040	15. 2 0. 055	13.5 0.042
2d week: Length Weight 3d week:	15.8 0.070	15.5 0.072	17.6 0.080	15, 2 0, 065
Length Weight: 4th week:	21.6 0.178	19.8 0.145	20.2 0.265	17.5 0.225
Length Weight 5th week: Length	26. 2 0. 295 31. 2	25.3 0.280 29.9	23.8 0.242 25.4	2!.0 0.145 26.5
Weight 6th week: Length	0.548	0.500 34.5	0.300 28.8	0.380
Weight 7th week:	0.772 39.2	0.892 38.1	0. 490 32. 0	0. 512 33. 4
Length Weight 8th week:	1.200	1.100	0.632	0.712 36.0
Length Weight	42.8 1.412	41.8 1.378 3.6 mm.	34. 1 0. 682 2.8 mm.	36.0 0.870 3.0 mm

water control tanks compared favorably with the growth rates of the 1,000-fish lot in the spacegrowth experiment. We expected that the growth rates in the control tanks would be somewhat similar to those experienced in the 4,000-fish lot of the space-growth experiment. The growth rates in the two control tanks were 3.2 and 2.8 mm. per week over an 8-week period, while the growth rate of the 4,000-fish lot was 2.1 mm.

The fish were raised under seemingly identical conditions, except for the time of year that the experiment was conducted. The 4,000-fish lot was reared from mid-May to mid-August, while the control-tank lots were reared in August and September. Although there are no temperature records for these growth experiments, the temperature record of the heating experiment (table 8) discloses a difference of about  $4^{\circ}$  F between May and August in control tank 13. Presumably, this difference also applied to the assault-boat tanks. Therefore, it is quite reasonable to conclude that water temperature is important in obtaining fast growth; however, the optimum temperature has not been determined.

Some other factor, possibly environmental, chemical, or genetic, acting singly or in combination with temperature may also have contributed to this discrepancy in growth rates. The end results of the salinity-growth experiment paralleled the results of the salinity-spawning experiment, however, indicating that a commercial rearing plant can, and should be operated on a brackishwater system, thereby reducing or eliminating the high cost of using fresh water.

## CANNIBALISM AND PREDATION

Cannibalism and predation are important factors in the successful rearing of young tilapia. Our initial plan to rear tilapia fry to bait-fish size in a single large fry tank met with a major setback when about 2 months after production started we discovered juveniles, ranging in length from 25 to 38 mm. (1 to 1.5 in.), chasing and consuming newly emerged fry that were being released into the tank. This situation was remedied by installing Monel-screen partitions to separate the tank into three compartments and segregate the young according to size. Cannibalism among tilapia was also reported by Chen (1953). The results of his observations are summarized in table 18.

#### TABLE 18.—Size relation between intraspecific predator and prey in T. mossambica

[Data from Chen (1953)]

Size of predator	Size of prey
0.75 inch	Up to 0.38 inch.
7 inches	Up to 1.25 inches.

In order to extend the findings of Chen and further define this intraspecific, predator-prey size relation, the following experiments were conducted in 35-gallon aquariums. Eight juvenile groups (predator) of different average lengths were selected. The average length of the groups ranged from 20.4 to 64.4 mm. (about 0.75 to 2.5 in.). A predator group consisting of 10 juveniles was first measured and placed in an aquarium. A second group of 20 to 30 young (prey), all fry or fry and juveniles, was also measured and placed in the same aquarium. Each of the eight experiments was carried out for a period of 72 hours, after which time the remaining prey were removed and measured. The fish did not receive any supplementary feeding during the period of the experiment. By comparing the lengths of the remaining young with the lengths of the young that were put into the aquarium, we were able to determine the maximum size of the young that were killed or consumed by each size group of The results are given in table 19 and predators. figure 17.

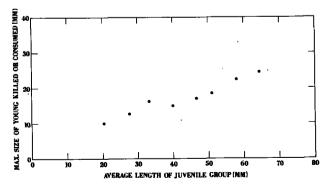


FIGURE 17.—Relation between size of juvenile groups (predators) and maximum size of young killed or consumed.

Length of group (1		Length of prey group (mm.)			
Range	Average	Range	Maximum size killed		
18.3-23.7 24.2-29.6	20. 4 27. 6	9. 7–12. 5 10. 2–15. 6	10.0 12.7		
30. 4-36. 4	33.2	9.8-19.2	16.2		
36.9-42.3	40.0	10.3-19.2	15.0		
43.2-48.7	46.7	9.8-20.0	17.0		
49.4-54.8	51.0	9.0-21.2	18.5		
55.7-61.4 62.0-67.8	58.0 64.4*	9.0-29.0 9.2-29.0	22.5 24.5		

 TABLE 19.—Size of young (prey) killed and consumed by different juvenile groups (predator)

[A juvenile group consisted of 10 fish of approximately equal size]

Other experiments with starved and well-fed juveniles showed that the degree of cannibalism increases when the fish are starved. One particular group of 50 juveniles, averaging 31.8 mm. (1.25 in.) in length, when well fed consumed 9 fry averaging 11.1 mm. (0.44 in.) in 15 minutes. This same group, when starved for a period of 3 days, killed or consumed 17 fry averaging 14.3 mm. (0.56 in.) in 12 minutes. When starved, the juveniles were aggressive upon introduction of the fry, whereas when well fed they were not particularly excited by appearance of the fry and generally remained near the bottom of the tank. An aggressive response by well-fed juveniles was noticeable only when a single fry or group of fry approached closely.

These experiments indicated the importance of keeping each compartment of the fry tank stocked with young fish of uniform size.

Another source of attrition, although not considered so important as cannibalism, was predation by dragonfly nymphs. These highly predacious larvae, which occurred commonly in the fry and assault-boat tanks, usually preyed on the smaller fry. Considerable effort was made to remove these nymphs. Chemical means of control (salt, potassium permanganate, and pyridylmercuric acetate, commonly called PMA) proved ineffective; dipnetting them individually seemed the most effective method.

#### SUMMARY AND CONCLUSIONS

This study evaluates the physical and biological feasibility of producing bait-size tilapia in tanks. Two facilities were used during the experiments. The first facility, or pilot plant, constructed on the grounds of the Bureau of Commercial Fisheries Biological Laboratory at Honolulu, Hawaii, was used from October 1956 to July 1958; the second, at the Kewalo Basin docksite, Honolulu, was used from August 1958 until September 1959. Results obtained at the pilot plant were of a preliminary and general nature, but aided in planning the more detailed experiments designed to examine factors associated with variations in reproduction and growth at the Kewalo plant.

Brood tanks at the pilot plant were stocked at the rate of 0.9 and 1.9 square feet of floor area per individual and with a sex ratio of  $2 \circleon$  : 1 \$\varsis\$. Stocking of the Kewalo tanks varied from 0.5 to 1.6 square feet of bottom area per individual, and the sex ratios ( $\circleon$  : \$\varsis\$) were 2:1, 3:1, 4:1, and 6:1.

Supplementary feeding of the tilapia included rice bran, chicken mash, alfalfa pellets, rabbit feed, and pelletized pond-fish feed and trout feed. Generally, for the adults pelletized feeds were much more satisfactory than finely divided mash and bran. Young fish were fed finely ground pondfish and trout feed.

Chemical analyses to determine concentrations of oxygen, free carbon dioxide, total bicarbonate and carbonate alkalinity, and hydrogen-ion were made routinely at weekly intervals to follow gross changes in the environment within the tanks.

Fry production at the pilot plant started in December 1956, approximately 9 weeks after the initial stocking. Nest-building activity was noted only after 3 to 4 inches of calcareous beach sand was placed in two of the brood tanks. Evidence of spawning on the bare floor, however, was noted when the one tank without sand was drained.

At both rearing plants, as the young emerged each day they were captured with a fine-meshed dipnet and counted. They were then placed in a fry tank and segregated by size in different compartments to prevent cannibalism.

Mortalities among the adults were attributable to factors such as handling, disease, asphyxiation, and possibly hydrogen sulfide poisoning. Highest mortality rates among the young were attributed to outbreaks of infectious disease and infestation by ectoparasites. Infestation by protozoan ectoparasites such as trichodinids and *Chilodon* spp. was rather easily controlled by chemical treatment. Outbreaks of infectious pancreatic necrosis, believed to be caused by a virus, were not controlled by any of the methods tried. Strict sanitation, prevention of overcrowding in the fry-tank compartments, and periodic prophylactic treatments were found to be good control measures for preventing outbreaks of disease.

Several factors were found to affect fry production. The spawning rate was increased during the winter months by artificially raising the temperature of the water. Only a slight rise was necessary to increase spawning frequency, but prolonged constant high temperature seemed to have a detrimental effect.

A sex ratio of  $3 \ controls 2 \ controls 1 \ controls 2 \ controls 2$ 

The growth of young fish was influenced by environmental factors. Significantly faster growth rates were found among young reared in less crowded tanks than in crowded tanks. Young reared on high-quality feed also evinced a much faster growth rate than those fed low-quality feed. The growth of young fish in brackish water of a salinity of about  $10 \circ/_{\infty}$  was remarkably fast.

An investigation of cannibalism indicated that juveniles averaging 20.4 mm. (about 0.75 in.) can kill or consume fry up to 10 mm. (about 0.38 in.) in length, and juveniles averaging 64.4 mm. (about 2.5 in.) in length are able to kill or consume smaller juveniles up to a maximum size of 24.5 mm. (about 1 in.). Starved juveniles evinced a much more aggressive response than well-fed juveniles to fry introduced into their tanks.

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### APPENDIX

APPENDIX TABLE 1.—Oxygen concentrations (ml./l.) at the pilot plant, determined weekly, April 2, 1957–July 8, 1958

Date	Zone time		Tank No			Assault-boat	<sup>1</sup> tank No. –	-	Fry tank	Tap wat
		1	2	3	1	2	3	4	_	l
57:										
Apr. 2	0900	0.75	3.87	4.61						4.
9	1000	4.75	6.75	5.71						6.
16	1000	2.67	5.80	3.81			<b>-</b>			6.
23	1000	4.88	5.56	3.90			<b>-</b>	]		5
30 May 7	1000 1100	3. 72 2. 94	5. 30 5. 25	4.80 3.75						56
14.	1000	0.77	2.56	2.87						6
21	1000	3.36	2.59	2.70						
28	1000	3. 33	3.64	3. 34						è
June 4	1000	1.68	2.03	2.42						l 6
11	1000	3.16	2.78	2.70						
19	1000	<u> 0. 63</u>	0.54	1.10			·			6
26	1000	1.12	0.54	1.56						6
July 2	1000	2.68	0.96	4.22 3.76						9
9 16	1000 ( 0830 )	3.58 1.78	3.30 1.73	3.42		5.40	2.40	3. 53		{
23	0830	3.78	3.34	5.40		5. 40 5. 19	1.90	3.69	1.05	
31	1000	5.60	6.96	6.54	4.38	4.34	3.25	4.30-	5.14	
Aug. 6-	0880	1.34	0.00	2. 51	3.10	4.02	3. 77	2.46	4.37	
13	0830	1.30	2.76	1.40		3,66	3. 59		3. 57	
22	1000	5.22	4.33	2.59	6, 51	3.43	5.49		4.11	[ .
28	0830	0.62	1.95	1.40	5.14	4.12	4.84	4. 52	4.64	1.
Sept. 5 10	0830	1.81	3.04	3. 33	5.64	4.00	5.72	4.70	4.11	
	0830	0.74	1.23	2.75	4.86	4.27	4.77	4. 53	3.54	
18	0830	1.18	0.92	1.39	4.86		2.96		2.77	
25	0830 0830	0.85	1.17 1.31	1.65 1.41	5. 52	2.69 4.42	2.63		3.36	
Oct. 3	0830	0.87 0.56	0.85	3, 13	5.79	4. 42 3. 85	2.03		4.20	
16	0830	0.58	0.72	1.49	5.48	4.18	1.90	2.46	4.75	
23	0830	0.85	1.35	1. 55	4.57	1.10	1.56	1.05	3, 38	
30	0830	2.68	1.37	2.41	4.44	3, 67	0.95	0.93	5.00	(
Nov. 6	0830	0.90	6.42	3.97	6.31	5.66	2.66	2.06	6.81	
12	0800	1. 17	0.78	1.84	4.36	2.77	0.74	0.73	3.00	ļ
20	0845	1. 44	2.57	2.45	5.27	3.84	1.69	0.61	3.08	j '
Dec. 8	1000	2.40	3.75	2.80	5.48	5.02	4.64	j	5.44	j .
10	1000	4.23	6.34	2.14	5.72	5.18	4.54		5.82	( ·
20 26	1100 0900	3.74 0.40	6.16 1.86	2.36 1.83	4.79			5. 54		
20 8:	0000	0.40	1.00	1. 60	4. / 9			0.04		'
o. Jan. 9	1300	3, 24	7.93	2.46	1	5.95	]		6.97	] (
16	1000	0.56	0.26	2.18		6.23			6.48	
24	0815	0.38	0.90	2.42		5,77			5.76	
Feb. 6	1300	4.20	3.13	5.21					6.96	1 0
14	1000 ]	0.51	1.07	1.07					5.85	; I
28	1300	5.65	6.74	2.42					7.04	
May 26 June 12	0800	0.64	0.69	0.34					5.66	
June 12	0800	0.46	0.19	0.28						
17	0830 0815	1.60	0.97	0.38 0.35						
24 July 2	0815	0.64 1.26	0.10 1.15	0.35					5.45	1 '
8	0800	1. 15	0.91	0.85					5.46	
V	1 0000	1.10	0.01	0.00					1	1

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<sup>1</sup> Acquired June 1957.

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	Tan	k11	Tan	k 2 ²		
Zone time	Oxygen (ml./l.)	Temper- ature (° F.)	Oxygen (ml./l.)	Temper- ature (° F.)	Cloud cover	Comments
800	$\begin{array}{c} 2.00\\ 2.46\\ 2.83\\ 3.32\\ 3.59\\ 3.49\\ 3.265\\ 2.12\\ 1.82\\ 1.24\\ 1.08\\ 0.94\\ 0.94\\ 0.86\\ 0.50\\ 0.43\\ 0.37$	$\begin{array}{c} 73.5\\ 74.0\\ 75.0\\ 75.0\\ 77.5\\ 78.5\\ 78.5\\ 78.5\\ 78.5\\ 78.5\\ 78.0\\ 77.5\\ 78.5\\ 78.5\\ 78.0\\ 77.5\\ 78.5\\ 76.0\\ 75.5\\ 76.5\\ 75.5\\ 75.5\\ 75.5\\ 75.5\\ 74.5\\ 74.0\\$	$\begin{array}{c} 0.78\\ 4.71\\ 5.57\\ 8.08\\ 10.46\\ 12.24\\ 12.36\\ 11.31\\ 6.98\\ 5.96\\ 4.30\\ 3.26\\ 2.80\\ 2.26\\ 2.26\\ 2.26\\ 2.26\\ 2.26\\ 2.26\\ 2.26\\ 1.54\\ 1.23\\ 1.17\\ 1.33\\ 1.17\\ 1.17\\ 1.33\\ 1.17\\ 1.17\\ 1.33\\ 1.17\\ 1.17\\ 1.13\\ 1.17\\ 1.17\\ 1.17\\ 1.17\\ 1.17\\ 1.13\\ 1.17\\ 1.17\\ 1.13\\ 1.17\\ 1.13\\ 1.17\\ 1.13\\ 1.17\\ 1.13\\ 1.17\\ 1.13\\ 1.17\\ 1.13\\ 1.17\\ 1.13\\ 1.17\\ 1.13\\ 1.17\\ 1.13\\ 1.17\\ 1.13\\ 1.17\\ 1.13\\ 1.17\\ 1.13\\ 1.17\\ 1.13\\ 1.17\\ 1.13\\ 1.17\\ 1.13\\ 1.17\\ 1.17\\ 1.13\\ 1.13\\ 1.13\\ 1$	74. 5 75. 0 76. 0 77. 0 78. 5 79. 5 80. 0 80. 0 80. 0 80. 0 80. 0 80. 0 79. 0 79. 0 79. 0 79. 0 79. 0 78. 5 76. 0 76. 0 79. 0 76. 5 76. 0 76. 0 76. 0 76. 0 76. 0 76. 0 76. 0 76. 0 76. 5 76. 0 76. 0 76. 0 76. 5 76. 5 75. 5 75. 5 75. 5 75. 5	6/8	Sun clouded over. Tanks partly exposed to sunlight. Tanks fully exposed to sunlight. Do. Tanks 1/2 shaded from sunlight. Tanks 1/2 shaded from sunlight. Tanks 1/2 shaded from sunlight. Tanks completely shaded from sunlight. Do. Do. Do. Do. Do. Do. Do. Do

APPENDIX TABLE 2.—Oxygen concentrations in tanks 1 and 2 sampled hourly for 24 hours, at the pilot plant, November 12–13 1957

<sup>1</sup> Tank 1 was cleaned on Nov. 4, 1957 and the water was still relatively low in algae on Nov. 12 and 13. <sup>2</sup> Tank 2 had a high concentration of algaé (*Chlorella*).

APPENDIX TABLE 3A.—Oxygen concentrations (ml./l.) observed in sex ratio-fish concentration experiment, August 27-November 26, 1958

Date	Zone time	Tank 1	Tank 2	Tank 3	Tank 4	Tank 5	Tank 6	Tank 7	Tank 8	Tank 9	Tank 10	Tank 11	Tank 12
Aug. 27 Sept. 4	0830 0830	5. 65 4. 04	1.68	5. 36	3. 18	3. 28	2. 63	4.90	3.80	3.62	2.24	4.69	3.66 4.18
12	0815	1.53						3.57					1.83
19	0810	3.00				[					2.84		1.10
Oct. 2	0815				2.48	[	3.65		0.36				
9	0815	1.75				}				) 2,72	1.25		
16	0815				1.98	0.43							í 1.00
23	0800				0.80	1.54							0.81
30	0800				2.74	0.76							1.82
Nov. 6	0800	2.45						2.22					2.03
13	0800	1.58			0.81				]	1.16		1	1
20	0800	3.90			0.01	0.30				1.54			{
26	0800	0.00				1.44				1.01	1.06		2.87
AU	0000					1. 11					1.00		2.01

APPENDIX TABLE 3B.—Oxygen concentrations (ml./l.) observed in feeding experiment, August 27-November 26, 1958

Date	Zone time	Tank 13	Tank 14	Tank 15	Tap water
Aug. 27 Sept. 4	0830	1.17 2.81	1. 94	1.04	5, 74
12	0815 0810	1.09 2.31			
Oct. 2	0815 0815 0815		• 0.99  1.15	1.68	5, 96 5, 56 5, 58
23 30	0800 0800		0.86	2.98	6.00 5.84
Nov. 6 13 20	0800 0800 0800		1.83	1.69 0.97	6.57 5.96 6.68
26	0800		1.65		6.30

Date	Zone time	Tank 13	Tank 14 (with heating cable and cover)	Tank 15 (with heating cable only)	Tap water
Jan, 16	0815	6. 69	0.80	4. 72	5. 9
23	0800	2.49	0.69	2.57	6.0
28	0800	1.51	0.78	0.83	5.9
7eb. 5	0800	3.11	3.04	4.94	5. 7
12	0800	0.91	0.95	1, 17	5.8
19	0815	3.97	3. 59	3.09	6.3
26	0800	5. 32	4.14	4, 39	- 6.5
Mar. 5	0805	5.06	1.86	5.09	6.0
12	0800	4.83	2.11	4.05	4.8
19	0800	4.49	3.56	4.84	5.9
26	0800	3.01	1.37	4.87	7.7
Apr. 2	0800	3.49	1.64	2,86	5.9
9	0800	2.96	5.58	2.04	6.3
16	0800	1.23	2.77	1.53	6. 3
23	0800	1.72	2.63	3.28	6.2
30	0810	1.76	2.84	2.27	6.2
Aay 7	0810	3. 32	2.27	2.98	5.9
14	0810	2.54	3.72	4.05	6. 1
21	0800 0810	3.01	2.95 3.54	3.01 4.46	6. ( 6. )
une 4	0815	4.20	4.43	4,40	6.1
une •	0815	4.45	4.36	4.46	5.
18	0810	3, 54	3.79	3.93	5.
25	0800	4.29	3.99	4,99	5. 1
uly 2	0800	5.45	2.83	5, 39	6.
uly 2	0800	4.35	2.40	2.92	5.
16	0800	4.20	1.31	3.55	6.
23	0800	3.06	2.63	3.34	5.
30	0800	2.53	1.86	3.40	5.
·····	0000	. 2.00		. 0.40	υ.

APPENDIX TABLE 4.—Oxygen concentrations (ml./l.) in tanks 13, 14, and 15 during heating experiment, January 16-August 27, 1959

APPENDIX TABLE 4.—Oxygen concentrations (ml./l.) in tanks 13, 14, and 15 during heating experiment, January 16-August 27, 1959—Continued

Date	Zone time	Tank 13	Tank 14 (with heating cable and cover)	Tank 15 (with heating cable only)	Tap water
Aug. 6	0800	2. 08	1.51	2. 69	6. 26
13	0800	4. 92	0.84	1. 60	5. 98
20	0800	4. 42	1.53	3. 00	5. 70
27	0800	3. 71	0.89	1. 18	5. 97

APPENDIX TABLE 5.—Oxygen concentrations (ml./l.) in brackish-water and fresh-water tanks, during salinityspawning experiment, August 6-September 24, 1959

Date	Zone time	Tank 1 (brackish water)	Tank 5 (fresh water)	Tap water
Aug. 6 13 20 27	0800 0800 0800	4. 95 4. 16 3. 21 3. 22	2.63 2.08 1.13	6. 26 5. 98 5. 70 5. 97
Sept. 3 10 17 24	0800 0800 0800 0815 0800	3.22 1.82 1.18 4.11 0.90	1.42 0.61 1.44 1.28 2.34	5. 97 7. 25 5. 56 6. 05 6. 34

APPENDIX TABLE 6A.—Hydrogen-ion (pH) values observed in sex ratio-fish concentration experiment, October 2-November 26, 1958

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Date	Zone time	Tank 1	Tank 2	Tank 3	Tank 4	Tank 5	Tank 6	Tank 7	Tank 8	Tank 9	Tank 10	Tank 11	Tank 12
Oct. 2 9 23 30 Nov. 6 20 20 20	0800 0806 0800 0800 0800 0800 0800 0800	7.7 7.9 8.8 7.9 >8.8 5.8 \$.8 8.7 8.6	8.1 7.7 8.1 7.4 	8.2 7.7 7.7 7.4 	7.9 8.5 8.7 7.4 	8.6 7.7. 7.4 7.8 8.4 7.9 7.4	7.7 7.3 7.7 7.4 >8.8 7.9	8.6 7.6 7.5 7.8 >8.8 >8.8 8.7	7.3 7.6 7.6 7.5 	7.7 7.7 7.3 7.3 7.9 8.7 7.7	8.3 7.5 7.6 7.5  8.5 7.6	7.9 7.4. 7.5 7.5 	7.9 7.7 7.3 8.1 8.0 8.1 8.7

APPENDIX TABLE 6B.—Hydrogen-ion (pH) values observed in feeding experiment, October 2-November 26, 1958

Date	Zone time	Tank 13	Tank 14	Tank 15	Tap water
Oct. 2	0800	8.6	7.4	7.7	8.0
16	0800	8.7 8.7 7.7	7.6	7.7 8.1 7.5	8.2 8.2 8.3
23 30 Nov. 6	0800		7.5 8.3		
20	0800 0800 0800	8.4 8.1	7.7 8.1 7.6	8.2 7.9	8, 1

Date	Zone time	Tank 13 (control)	Tank 14 (with heating cable and cover)	Tank 15 (with heating cable only)	Tap water
Jan. 16	0815	8.7	7.7	8.6	
23	0800	8.4	. 7.8	8.3	
28	0800	7.8	7.5	7.9	8.1
Feb. 5	0800	8.8	8.1	8.8	8.1
12	0800	7.4	7.4	7.6	8.1
19	0815	7.6	7.6	8.8	
26	0800	7.9	8.0	8.4	7.9
Mar. 5	0805	7.7	7.5	7.9	8.2
12	0800	8.0	7.5	8.0	8,1
19	0800	7.9	7.7	8.1	8, 1
26	0800	7.7	7.5	7.9	8.3
Apr. 2	0800	7.7	7.5	7.9	8.3
9	0800	7.7	7.9	8.1	8.1
16	0800		7.7		8.3
23	0800	7.7	7.4	7.7	8.1
30	0810	8.4	7.9	8.3	8.1
May 7	0810	7.9	7.8		8.1
14	0810	8.1	8.1	8.7	8.3
21	0800	7.9		8.4	8.1
28	0810	7.7	. 7.9	8.1	7.9
June 4	0815	7.9	8, 1	8.5	7.9
11]	0815	8.2	8.1	8.2	7.9
18	0810	8.1	8.1	8.1	7.9
25	0800	8.1	7.7	7.9	8.1
July 2	0800	7.8	7.5	7.7	7.9
9	0800	8.1	7.6	7.4	7.9
16	0800	8.3	7.4	7.5	7.9
23	0800	7.5	7.4	7.5	7.9
30	0800	l. 7.8	7.5	7.7	7.9

APPENDIX TABLE 7.—Hydrogen-ion (pH) values during heating experiment, determined weekly, January 16-August 27, 1959

APPENDIX	TABLE 7	-Hydrogen-io	n (pH)	values du	iring
		determined	weekly,	January	16–
August 2	27, <i>1959</i> —Co	ontinued		-	

Date	Zone time	Tank 13 (control)	Tank 14 (with heating cable and çover)	Tank 15 (with heating cable only)	Tap water
Aug. 6	0800	7.3	7.3	7.4	8.2
13	0800	7.7	7.3	7.3	7.9
20	0800	7.8	7.3	7.5	8.1
27	0800	7.6	7.3	7.3	7.9

APPENDIX TABLE 8.—Hydrogen-ion (pH) values in brackishand fresh-water tanks during salinity-spawning experiment, August 6-September 24, 1959

Date	Zone time	Tank i (brackish <sup>i</sup> water)	Tank 5 (fresh water)	Tap water
Aug. 6	0800	8.3	7.7	8.2
13	0800	8.2	8.3	7.9
20	0800 0800	8.0 7.7	5.2 8.1	8.1
Sept. 3	0800	7.7	7.7	7.9 7.9
10	0800	7.6	7.9	8.0
17	0815	7.5	8.1	8.0
24	0800	7.6	8.1	7.9

APPENDIX TABLE 9.—Free carbon dioxide, bicarbonate, and normal carbonate in control and heated tanks during heating experiment and in the fry tank, determined weekly, February 26-August 27, 1959

[In parts per million]

ļ	Zone	<b>Ta</b>	nk 13 (cont	rol)	Tank 14 (	heated and	1 covered)	Та	nk 15 (heat	ed)		Fry tank	
Date	time	Free CO2	Carbon- ate	Bicarbon- ate	Free CO2	Carbon- ate	Bicarbon- ate	Free CO2	Carbon- ate	Bicarbon- ate	Free CO:	Carbon- ate	Bicarbon ate
Feb. 26	0800		37.6	115.8									
Mar. 5	0805	0.0	40.8	56.4	0.0			0.0	63.8	15.2	0.0	42.6	23. (
12	0800	0.0	7.4	112.6				0.0	13.0	75.3	0.0	32.6	30.1
19	0800	0.0	14.6	111.4	3.8	0.0	136.2	0.0	3.0	93.6	0.0	19.4	37.4
26	-0800	8.0	0.0	154.1	6.9	0.0	122.3	5.8	0.0	115.8	0.0	11.4	52. 7
Apr. 2	0800	7.3	0.0	181.7	5.9	0.0	141.6	0.0	5.4	115.4	0.0	3.6	58.9
.9	0800	5.5	0.0	173.3	2.0	0.0	65.2	0.0	3.6	104.8	0.0	9.8	60.7
16	0800		0.0	146.4	6.7	0.0	83.4		0.0	123.7	0.0	38.8	23.7
23	0800	5.4	0.0	147.0	6.8	0.0	86.3						
30	0810	0.0	2.0	118.4	3.7	0.0	85.5	0.0		130.5	0.0	12.0	57.0
May 7	0810	2.9	0.0	111.0	3.8 2.7	0.0	92.0		0.0	79.3	0.0	7.6	56.7
14	0810	3.9	0.0	117.0	2.7	0.0	85.4	0.0	13.4	59.1	0.0	17.2	42.8
21	0810	5.7	0.0	131.5		0.0	81.0	0.0		82.3	0.0	7.8	55.7
28	0810	4.1	0.0	123.5	2.6	0.0	81.4	0.0		76.6	0.0	29.0	34.0
June 4	0815	3.8	0.0	115.9	0.0		79.1	0.0	12.2	57.2	0.0	2.4	62.6
11	0815	0.0			[	[	i	0.0	3.8	76.7	0.0	7.4	62.9
18 25	0810 0800	0.0	26.0	84.3	4.1		104.3	0.0	10.8	83.8 85.0	0.0 0.0	16.0 32.0	57.7
July 2	0800		0.0	73, 1	1.4	0.0	116.6	0.0 2.2	16.2	77.2	0.0	32.0	57. 1 46. 3
9	0800	1.2	0.0	86.5	4.9	0.0	108.0	4.7	0.0	111.7	0.0	32.2	58.3
16	0800	0.0	11.6	74.1	4.9	0.0	108.0	3.8	0.0	107.9	0.0	15.4	60.6
23	0800	1.7	0.0	83.5	5.5	0.0	128. 3	4.8	0.0	117.4	2.3	0.0	81.4
30	0800	0.0		72.8	0.0 7.5	0.0	124.7	3.4	0.0	109.2	2.3	36.1	48.8
Aug. 6	0800	6.5	24.8	180.5	7.5 8.5	0.0	117.3	5.2	0.0	115.9	1.2	0.0	40.0 76.5
13	0800	0.0	0.0	97.4	8.5	0.0	133.4	6.9	0.0	115.9	1.2 2.4	0.0	70.5
20		0.0	17.0		10.0	0.0	135.0	7.0	0.0	125.8	2.4	66.2	20.6
20	0800 0800		17.0	102.8	13.3	0.0	140.0		0.0		0.0	22.0	20.0
21	0800	0.0	19.9	97.3	13.3	0.0	140.0	12.7	0.0	149.1	0.0	22.0	ov. 2

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Appendix Table 10 normal carbonate i:				
salinity-spawning 1959	experiment,	-	6–September	24,

		Tank 1 (brackish water)		Tank 5 (fresh water)			
	Zone time	Free CO <sub>2</sub>	Carbon- ate	Bicarbon- ate	Free CO2	Carbon- ate	Bicarbon- ate
Aug. 6 13	0800 0800	1.7	0.0 19.1	94. 4 69. 3	3.5 0.0	0.0 30.6	111.8
20 27 Sept. 3 10	0800 0800 0800 0800	0.0 2.6 4.0 16.2	14.4 0.0 0.0 0.0	86.7 123.4 183.3 188.5	0.0 0.0 0.0 0.0	24. 2 39. 0 80. 6 32. 2	80. 1 71. 0 36. 3 81. 8
10 17 24	0800 0815 0800	8.0 13.6	0.0	188.1 214.0	0.0 0.0	40.0 54.4	76. 3 45. 3

[In parts per million]