AN ABSTRACT OF THE THESIS OF William James Tobias for the Master of Science Degree in Biology presented June 15, 1976.

Title: Ecology of <u>Siganus argenteus</u> (Pisces: Siganidae) in Relation to its Mariculture Potential on Guam

Thairman, Thesis Approved:

To assess the mariculture potential of <u>Siganus argenteus</u> on Guam, a study was conducted to integrate the known information on <u>S. argenteus</u>, to provide relevant information of the general biology (i.e., habitat preference, feeding habits, behavior, growth rate, and diseases) of sub-adult and adult fish and to determine their tolerance to environmental parameters (i.e., temperature, salinity, and oxygen), as encountered on Guam's fringing reefs.

Field observations have shown that <u>S</u>. <u>argenteus</u> juveniles migrate from the reef flat to spend their sub-adult and adult lives feeding diurnally on the algal turf of the submarine terrace and seaward slope (3-40 m in depth).

Analysis of stomach contents from 20 sub-adult/adult specimens has shown <u>Tolypiocladia glomerulata</u> (Importance Value=47.8), <u>Halimeda</u> <u>discoidea</u> (IV=24.1), <u>Dictyota</u> sp. (IV=19.3), and <u>Galaxaura marginata</u> (IV=10.4) to be the most important algae consumed in the field. Comparison of stomach content analysis with quantitative field analysis of the dominant macro-algae present indicates that the algal species ingested directly reflect the algal availability of that specific area. S. argenteus exhibits no active food selection, with the exception of possible avoidance of the blue-green alga <u>Schizothrix calcicola</u>.

The growth rate of <u>S</u>. <u>argenteus</u> is faster than <u>S</u>. <u>canaliculatus</u> and <u>S</u>. <u>spinus</u>. Fork length and weight measurements of similar initial size and weight <u>S</u>. <u>argenteus</u>, <u>S</u>. <u>spinus</u>, and <u>S</u>. <u>canaliculatus</u> grown under similar laboratory conditions after seven months were 187 mm/114 g, 124 mm/29 g, and 158 mm/59 g, respectively. Lengthweight regression line slopes for <u>S</u>. <u>argenteus</u>, <u>S</u>. <u>canaliculatus</u>, and <u>S</u>. <u>spinus</u> were not significantly different. <u>S</u>. <u>argenteus</u> was sexually mature in 11 months at 201 mm fork length and approximately 150 g.

Some laboratory reared <u>S</u>. <u>argenteus</u> developed symptoms similar to those caused by deficiencies of B-complex vitamins. <u>S</u>. <u>argenteus</u> juveniles develop exopthalmia in water with a mean temperature of  $33.2^{\circ}C \pm 1.3^{\circ}C$  and 6.68 ppm dissolved oxygen concentration.

The survival rates of fish subjected to water temperatures of 28, 30, and 32°C were 100, 94, and 79%, respectively, over 14 days. Fish in 34°C water had 50% mortality in 2.7 days, 96% mortality in 5 days, and 100% mortality in 8 days. <u>S. argenteus</u> juveniles are tolerant to reduced salinities and oxygen concentrations. The lower lethal salinity limit range was  $4-7^{\circ}/_{\circ\circ}$ . The survival of <u>S. argenteus</u> is reduced to 50% after two days at  $4^{\circ}/_{\circ\circ}$  salinity. The growth rate of <u>S. argenteus</u> was not significantly different at salinities of 10, 20, and  $30^{\circ}/_{\circ\circ}$  over a 1-month period.

The 24-hour lower dissolved oxygen concentration limit for <u>S. argenteus</u> held in 48-liter aquaria was 1.0-2.0 ppm. The average fish survival time was 1.5 and 4.6 hours, respectively, for the .5 and 1.0 ppm oxygen experiments; 100% survival was recorded for fish maintained in 2.0 and 3.0 ppm dissolved oxygen concentrations. An oxygen consumption rate of .1 mg  $1^{-1}$  hr<sup>-1</sup> was recorded for fish maintained in 2.0 and 3.0 ppm dissolved oxygen concentrations.

ECOLOGY OF <u>SIGANUS ARGENTEUS</u> (PISCES: SIGANIDAE) IN RELATION TO ITS MARICULTURE POTENTIAL ON GUAM

by

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## INTRODUCTION

Eight species of siganids (rabbitfish) are reported from Guam; however, only six have been observed recently (Kami and Ikehara, In Press). Of these, only <u>Siganus spinus</u> and <u>S. argenteus</u> support heavy fishing demand. Although both species have high local demand (Bryan, 1975; Kami and Ikehara, In Press) and have juveniles equally available during certain times of the year (Tsuda and Bryan, 1973), the faster growth rate (based on preliminary observations in holding tanks) and the larger attainable size of adult <u>S. argenteus</u> favor this species as the more desirable siganid for mariculture on Guam.

<u>Siganus argenteus</u> (Quoy and Gaimard) is the most widely distributed of the siganid species, occurring from the Tuamotu Islands to the Red Sea (Herre and Montalban, 1928; Fowler and Bean, 1929; Schultz et al., 1953; Hiatt and Strasburg, 1960; Smith and Smith, 1963; Ben-Tuvia et al., 1973; Popper and Gundermann, 1975; and D. J. Woodland, personal communication).

<u>S. argenteus</u> juveniles (mean fork length range: 54-64 mm) school onto the reef flats of Guam at the same time as <u>S. spinus</u> juveniles (two days plus or minus the last quarter moon during the months of April, May, and occasionally in June and October) and begin to feed on filamentous and smaller fleshy algae (Tsuda and Bryan, 1973). The number of juveniles entering the reef flat areas varies yearly as indicated by creel census figures collected over a 13-year period by the Guam Division of Fish and Wildlife (Kami and Ikehara, In Press). Statistical analysis of creel census information shows no pattern or cycle to these fluctuations.

Little information is available on the general biology of subadult and adult fish of this species on the reefs. Some data are available on its habitat preference (Schultz et al., 1953), food habits (Hiatt and Strasburg, 1960), and behavior (Popper and Gundermann, 1975); however, this information is at best fragmentary.

Growth rate experiments have been conducted on a variety of siganids to determine their mariculture potential - <u>S</u>. <u>rivulatus</u> (Ben-Tuvia et al., 1973; Popper and Gundermann, 1975), <u>S</u>. <u>canaliculatus</u> (Lavina and Alcala, 1973; May et al., 1974; Westernhagen and Rosenthal, 1975), <u>S</u>. <u>spinus</u> (Westernhagen, 1974), <u>S</u>. <u>guttatus</u> (Westernhagen and Rosenthal, 1975), and <u>S</u>. <u>luridus</u> (Popper and Gundermann, 1975). However, there is no published literature on the growth rate of <u>S</u>. <u>argenteus</u>. No information is available on the tolerance and growth of <u>S</u>. <u>argenteus</u> at elevated temperatures or varying salinities. Oxygen data relevant to siganid culture, particularly tolerance to low dissolved oxygen concentrations often encountered in reef flat environments, are nonexistent.

The purpose of this study is to integrate the known information on <u>S. argenteus</u>, to provide relevant information on the general biology (i.e., habitat preference, feeding habits, behavior, growth rate, and diseases) of sub-adult and adult fish, and to determine their range of tolerance to environmental parameters (i.e., temperature, salinity, and oxygen), as encountered on Guam's fringing reefs. The results of the data obtained are of significance in assessing the mariculture potential of Siganus argenteus on Guam.

#### MATERIALS AND METHODS

#### Field Observations

Field observations of the behavior and feeding associations of sub-adult and adult <u>Siganus argenteus</u> were made with the aid of scuba gear on the submarine terrace and seaward slope reef zones off the Tanguisson Power Plant, which is located between Tanguisson Point and Amantes Point on the northwest coast of Guam. <u>Siganus argenteus</u> is abundant in this area and provides adequate opportunity for observation.

A description of the submarine terrace and seaward slope reef zones off the Tanguisson Power Plant has been provided by Jones et al., (1976). The first submarine terrace is approximately 110 m in width and slopes gradually from 3-15 m in depth to the seaward slope. The seaward slope, approximately 70 m in width, slopes sharply from 15-40 m in depth to the second submarine terrace.

#### Stomach Content Analysis

A quantitative analysis of stomach contents was conducted on 20 specimens of <u>S</u>. <u>argenteus</u> speared on the submarine terrace and seaward slope reef zones off the Tanguisson Power Plant in water 6-40 m deep. The stomach contents were preserved in 70% alcohol for later identification under the microscope. A modified point method was used to quantify the food items (Jones, 1968a; Bryan, 1975). Relative abundance (percent composition by species), relative frequency (percentage of occurrence), and importance values (relative abundance + relative frequency) were calculated for the items ingested.

The electivity index,  $E = \frac{r_i - p_i}{r_i + p_i}$ , formulated by Ivlev (1961)

to determine the degree of choice a predator shows in its feeding behavior, was used to determine the electivity for certain algae exhibited by <u>S</u>. <u>argenteus</u> in its feeding behavior (E = electivity,  $r_i$  = percent composition of an alga in the stomach contents, and  $p_i$  = percent composition of that alga in the field). Values of -1, 0, and +1, respectively, indicate complete avoidance, no active selection, and complete selection.

#### Growth Rates

Holding Tanks

Juvenile <u>S</u>. <u>argenteus</u> and <u>S</u>. <u>spinus</u>, netted from the reef flats of Guam in May 1974, were transported to the Marine Laboratory and reared in 1000-liter circular polyethylene tanks, with 50 fish per tank. Each tank was equipped with a recirculating seawater system. Seawater was continuously pumped from the rearing tank by a submersible magnetic pump (Iwaki Pump Co., Model MDS-15) up through an external gravel filter box (64 cm<sup>2</sup>; 200 liters), containing crushed coral separated by plastic-coated fiber window screening. Aeration was supplied via a 1/3-hp dri-air pump (Conde Pumps, Model No. 2MMRB).

A diet consisting of the green alga <u>Enteromorpha clathrata</u> and Purina Trout Chow was fed twice daily (0800 and 1700 hours) to groups of <u>S. argenteus</u> and <u>S. spinus</u> (50 fish each) from May to December 1974. <u>Enteromorpha</u> has been shown to be the most preferred food of juvenile siganids (Tsuda and Bryan, 1973). Previous growth experiments with S. canaliculatus (July 1973-February 1974), reared under identical

laboratory conditions, revealed that the addition of trout chow with <u>Enteromorpha</u> yielded a faster growth rate in fish (Tsuda et al., unpublished report).

Enteromorpha and trout chow were fed in excess but replenished with a fresh supply twice daily. Excess dry food and fecal material were removed with a siphon hose prior to each feeding. Ten fish were selected at random from each tank and weighed in a preweighed container of seawater (wet weight) on a torsion balance (Torbal, Model PL-12) and measured (fork length) every two weeks during the 7-month period.

#### Floating Pens

Juvenile <u>S</u>. <u>argenteus</u> were also raised in three floating fish pens from May to December 1974. The fish pens, measuring 1.5 x 1.5 x 1.2 m, were constructed of a synthetic mesh net (.7 cm mesh) held rigid by a 2.5-cm PVC frame and supported by pre-cast foam floats on each corner and in the center. The nets were tied across an intake channel leading to the Piti Power Plant and stocked with approximately 500 fish per net. This group of fish simulated a "wild" population, existing under conditions similar to those in a natural environment, but without predators. This site was chosen because of its good water flow qualities and because it provided protection from possible outside interference.

The fish in each pen were fed approximately 9 kg (20 lbs) of <u>Enteromorpha</u> twice a week; however, due to the occasional unavailability of this alga, it was necessary to alter the feeding pattern to one heavy feeding per week. A natural algal growth, predominantly of the blue-greens <u>Schizothrix calcicola</u> and <u>Microcoleus lyngbyaceus</u>, and the brown algae <u>Dictyota bartayresii</u> and <u>Sphacelaria tribuloides</u>, covered the nets, thus providing an additional food source.

Ten fish selected at random from each of the three nets were weighed (wet weight) on a triple beam balance (O'Haus, Model 700) and measured (fork length) monthly over a 7-month period.

#### Diseases

An important consideration in evaluating the mariculture potential of an organism is its susceptibility to diseases under controlled rearing conditions. The diseases encountered during the comparative growth experiments of <u>S</u>. argenteus, <u>S</u>. spinus, and <u>S</u>. canaliculatus are described and causes discussed.

<u>S. argenteus</u> and <u>S. canaliculatus</u>, fed a diet of trout chow and <u>Enteromorpha clathrata</u>, developed symptoms similar to those caused by deficiencies of B-complex vitamins in salmon, trout, carp, and catfish (Lagler et al., 1962; Halver, 1972). To determine if this condition was caused by a vitamin B-complex deficiency, a preliminary vitamin enrichment experiment was conducted for one week with <u>S. argenteus</u> juveniles having various degrees of the symptoms. Nine groups of fish, four fish per group, were given intramuscular injections (.01-.03 mg) of combinations of thiamine (vitamin B<sub>1</sub>), riboflavin (vitamin B<sub>2</sub>) and pyridoxine (vitamin B<sub>6</sub>) at ten times the minimum daily requirement (.15-.68 mg/kg body wt/day) for salmonid fish (Phillip and Brockway, 1957). Information on the vitamin requirements of herbivorous fish is fragmentary. The dosage of ten times that required for salmonids was chosen because the vitamin requirements of herbivores are believed to

be greater than those of carnivores (Halver, 1972). Two control groups (four fish per group), one group injected with 1 cc of sterile water and one group not injected, were also monitored.

## Environmental Parameters

### Thermal Tolerance

Mass mortality of <u>S</u>. <u>argenteus</u> juveniles maintained in laboratory holding tanks has been observed during extreme low spring tides where tidal range is about 1 m and mid-day air temperatures are 29.5-30.4°C on the reef flat. Reef flat water, ranging in temperature from 26.2-32.0°C, exits from the reef flat through a shallow intake channel from which the laboratory draws its seawater supply.

In an effort to determine the cause of the fish kills, the effects of thermal stress (tolerance and growth) on <u>S</u>. <u>argenteus</u> were studied in a thermal simulator apparatus (Jones et al., 1976) consisting of four series of 79 x 59 x 50 cm polyethylene tanks (Series A, B, C, and D), three tanks per series, equipped with standpipes to hold the water level at 39 cm (182 liters). All tanks were set up with an open seawater system and flow rates were regulated at 7.5 liters/min for a tank turnover time of 2.5/hr. Series A tanks received ambient temperature seawater ( $\approx 28^{\circ}$ C). The water, before entering Series B, C, and D tanks, was regulated at temperatures of +2 ( $\approx 30^{\circ}$ C), +4 ( $\approx 32^{\circ}$ C), and +6°C ( $\approx 34^{\circ}$ C), respectively, in 200-liter holding tanks with a Pac-Tronics temperature controller (Model 1442) and Clepco quartz immersion heaters (Model No. 6-2215-V). The controllers automatically adjust the experimental temperatures to 2-degree increments above the existing ambient temperature of about 28°C. Stratification of heated water in the holding tanks was prevented by vigorous aeration through air stones in the corners. The water temperature in Series A, B, C, and D tanks was continuously monitored via temperature probes with a scanning telethermometer (Yellow Springs Instrument Co., Model 47) and recorder (Yellow Springs Instrument Co., Model 80A). Dissolved oxygen (ppm) was measured at about 0900 hours daily with an oxygen meter (Yellow Springs Instrument Co., Model 51A).

Four sub-adult <u>S</u>. <u>argenteus</u> per tank were measured (fork length) and weighed (wet weight) before and after each temperature experiment, the duration of which was two weeks. The mean range of fish size and weight for the four thermal experiments was 114-119 mm and 24-28 g, respectively. Growth was recorded for fish in each temperature series after the 2-week period.

The fish to be placed in the +2, +4, and +6°C temperature series tanks were acclimated in 16-liter plastic containers, half filled with ambient temperature seawater. An individual container was then placed into the 182-liter tank containing the heated water. When the water temperature inside the smaller container warmed up to within .5°C of the temperature of the larger tank, the fish were released into the larger tank. Acclimation times were less than two hours.

All fish were fed <u>Enteromorpha clathrata</u> and trout chow, a high protein supplement, twice daily (0800 and 1700 hours) during the experiments.

#### Salinity Tolerance

Salinity tolerance studies were conducted to determine the lower lethal salinity limit and the effects of reduced salinity on the survival and growth of S. argenteus.

The first salinity study was conducted to determine the range of the lower salinity limit. Three resin-coated wood tanks (143 x 84 x 21 cm; 252 liters), two experimental and one control, were filled with seawater of 31.6°/oo salinity. Ten fish were selected at random and placed in each tank. Aeration was added via a diaphram pump (Cole-Palmer, Model P-200) and the tanks were half covered to prevent outside stimuli from exciting the fish.

The salinity was reduced in the two experimental tanks by  $10^{\circ}/^{\circ}$  daily to about  $12^{\circ}/^{\circ}$  and reduced again the following day to  $8^{\circ}/^{\circ}$  by the addition of tapwater that was vigorously aerated for 24 hours to drive off residual chlorine (American Public Health Association, 1971). Prior salinity experiments had shown that <u>S</u>. <u>argenteus</u> will tolerate daily salinity reductions of about  $10^{\circ}/^{\circ}$  from 30 to  $8^{\circ}/^{\circ}$ ; behavior and feeding habits remained normal. The salinity was further reduced from  $8^{\circ}/^{\circ}$  in the experimental tanks by  $1^{\circ}/^{\circ}$  daily and percent mortality was recorded.

Daily temperature and oxygen measurements were made at about 0900 hours with a calibrated mercury thermometer (Yoshino, No. 467361) and oxygen meter. Salinity was determined with a refractometer (American Optical Corporation, Model 10402). All fish were fed <u>Enteromorpha</u> <u>clathrata</u> and trout chow daily for the duration of the experiment (10-12 days). Four replicates of the experiment were conducted.

A second salinity tolerance study was conducted to narrow the boundaries of the lower lethal salinity limit and to determine the short term effects of reduced salinity on <u>S</u>. <u>argenteus</u>. The salinity of seawater in a circular 1000-liter polyethylene tank containing 60 S. argenteus (mean fork length: 85 mm; mean weight: 10 g) was reduced from 31.6 to  $10^{\circ}/_{\circ\circ}$  by the method described above. A series of 200-liter tanks (Series A, B, C, and D), three tanks per series, were set up in which the salinity of water in each series had been reduced to 10, 8, 6, and  $4^{\circ}/_{\circ\circ}$ , respectively. Aeration was supplied to each tank.

Twelve fish were selected at random from the 1000-liter tank and four fish were placed in each of the three tanks of each series to test for survival rates. Temperature and dissolved oxygen were recorded daily and pH samples were taken before and after the 96-hour experiment. All fish were fed Enteromorpha daily.

Quantitative observations were made on the fish's response to tactile and light stimuli, equilibrium, coloration, and feeding ability in the reduced salinity water. Normal responses to tactile and light stimuli are to avoid touch contact and to back out of the path of light when disturbed at night by a flashlight beam. Normal equilibrium and coloration responses are exhibited by the fish's ability to maintain a horizontal swimming position and to adapt to background shading. Feeding observations are reported for tank group response.

A third study was conducted to determine the effects of reduced salinity on the growth rate and survival of <u>S</u>. <u>argenteus</u> over a 1-month period. The salinity of seawater in three 1000-liter circular polyethylene tanks was reduced to 10, 20, and 30°/oo, respectively, as described above. Each tank was equipped with a submersible magnetic pump which recirculated the water through an external gravel filter box (64 cm<sup>2</sup>; 200 liters). Aeration was supplied via a 1/3-hp dri-air pump.

The fish used in the experiment had been previously held in a tank identical to the experimental tanks. The salinity was reduced to the experimental level by  $10^{\circ}/_{\circ\circ}$  daily (from 30 to 20 to  $10^{\circ}/_{\circ\circ}$ ) and the fish selected at random and placed into the corresponding experimental salinity tank. The mean length and weight of fish in the 10, 20, and  $30^{\circ}/_{\circ\circ}$  salinity tanks were 88 mm/10 g (n=26), 90 mm/12 g (n=16), and 90 mm/12 g (n=16), respectively. A greater sample number of fish was inadvertently used in the  $10^{\circ}/_{\circ\circ}$  salinity group; however, crowding was definitely not a problem.

The fish were fed <u>Enteromorpha</u> and a combination of chicken starter crumbles and rabbit feed twice daily, due to the unavailability of trout chow.

#### Oxygen Tolerance

A series of four oxygen experiments with fish held in water of dissolved oxygen concentrations of .5, 1.0, 2.0, and 3.0 ppm were conducted to determine the lower lethal oxygen concentration limit of <u>S. argenteus</u>. Oxygen consumption rates for <u>S. argenteus</u> were calculated at each level of treatment.

The respirometry tanks consisted of six covered 40-liter aquaria, four experimental and two control tanks. A total of four oxygen experiments were run; one each at dissolved oxygen concentrations of .5, 1.0, 2.0, and 3.0 ppm. These dissolved oxygen levels were selected from a series of pre-trial runs which indicated that the fish's lower lethal oxygen concentration was within these limits.

The dissolved oxygen concentration in the experimental tanks was reduced to the desired level by the addition of nitrogen gas bubbled

through air stones placed at opposite ends of the tank. Graham (1949) and Carpenter and Cargo (1957) employed a similar technique to gradually reduce the oxygen content of seawater. Shelford and Allee (1913) studied the behavior of 16 species of fish to gradients of oxygen, carbon dioxide, nitrogen, and ammonia. They concluded that there was no significant response to the nitrogen gradient and the concentration of that gas is of no importance as long as it is not present in such excess as to cause "gas bubble" disease (exopthalmia).

To assure even dispersion of the nitrogen gas and subsequent reduction of oxygen in the seawater, a plastic impeller connected by a glass rod to a small electric motor was used to gently stir the water without disturbing the air-seawater interface. The motor, located above the tank, rotated the impeller at a rate of 110 rpm. The dissolved oxygen concentration was read from a continuously reading oxygen meter (Yellow Springs Instrument Co., Model 54). Czaplewski and Parker (1973) reported the accuracy of the oxygen probe equivalent to or better than the Winkler method. The oxygen probe was placed close to the impeller blades to allow a continuous flow of water past the probe membrane. The uneven rotation of the impeller also imparted a gentle back and forth movement to the oxygen probe. Two small holes were drilled in each of the aquaria lids to allow access for the oxygen probe and impeller-motor apparatus.

Opaque partitions were placed between and at the ends of all tanks containing fish. This eliminated inter-tank stimuli of fish and subsequent hyperactive states which may result in abnormal metabolic increases.

The rabbitfish were randomly selected and transferred from 1000liter holding tanks (temperature - 28°C; dissolved oxygen - 6.4 ppm; salinity - 33.3°/oo) to a 200-liter tank at 2100 hours while the fish were in a semi-quiescent state. Fish of the same cohort (mean fork length: 116 mm; mean weight: 29 g) were removed and placed directly into the deoxygenated water in the experimental tanks, one fish per tank including the first control tank. Upon addition of the fish into the control tank, the aeration was removed and the fish allowed to slowly deplete the oxygen supply for the duration of the experiment. A second control tank series with no fish, but with the dissolved oxygen reduced to that of the experimental tanks, was monitored separately. The change in oxygen concentration of the water due to diffusion of oxygen from the tank atmosphere across the water interface was recorded at hourly intervals.

If the dissolved oxygen concentration varied more than  $\pm$  .2 ppm from the previously set concentration, nitrogen or oxygen was added to adjust the oxygen concentration back to the desired level.

The duration of each oxygen experiment was 24 hours or until death of all the fish. Dissolved oxygen readings were taken at hourly intervals for the .5 and 1.0 ppm experiment, 2-hour intervals for the 2.0 ppm experiment, and at approximately 4-hour intervals for the 3.0 ppm experiment. The fish were not fed during the experiment.

Salinity was measured with a hand-held refractometer. Samples for pH determination were taken at the beginning and end of each experiment and measured with a Beckman Expandomatic SS-2 pH meter

(Beckman Instrument Co., Model 76). Temperatures were recorded with a calibrated mercury thermometer.

## RESULTS AND DISCUSSION

## Feeding Habits

#### Foraging Behavior

Field observations show that after approximately 4-8 weeks of feeding on the reef flat, <u>S</u>. <u>argenteus</u> juveniles (<105 mm fork length) migrate from the reef flat community to spend their sub-adult (105 to <170 mm fork length) and adult (sexual maturity, >170 mm fork length) lives in deeper water of the submarine terrace and seaward slope. Surge channels and reef flat areas are less frequently used as feeding grounds.

Tusda and Bryan (1973) concluded after examining 331 dead juvenile <u>S</u>. <u>spinus</u> from seven locations around Guam, 74% of which had empty guts, that <u>S</u>. <u>argenteus</u> outcompeted <u>S</u>. <u>spinus</u> for food. My observations indicate that <u>S</u>. <u>argenteus</u> did not outcompete <u>S</u>. <u>spinus</u> for food, but simply had migrated off the reef flat. Attempts to net juvenile <u>S</u>. <u>argenteus</u> were fruitless and none were observed in reef flat areas where they were caught in abundance several weeks before. Groups of juvenile and sub-adult <u>S</u>. <u>spinus</u> (10-200 individuals) were observed in the same area feeding on benthic algae.

Observations made after a fish kill, believed caused by aerial application of the organophosphate pesticide Malathion on a portion of the reef flat on the northwest coast of Guam (May 23, 1975), further supports this hypothesis. An estimated 65,000-85,000 fish were reportedly killed in an area of  $360 \text{ m}^2$  (Kami, Division of Fish and Wild-life); approximately 40% of the fish were S. spinus juveniles and

sub-adults and 60% were species other than siganids. No juvenile or sub-adult <u>S</u>. <u>argenteus</u> were found. Juveniles of both species first appeared in this area during the first week of April 1975. By the date of the spraying, it is believed that <u>S</u>. <u>argenteus</u> juveniles had migrated from the reef flat to the submarine terrace and seaward slope and therefore were not affected by the pesticide in the shallow reef flat waters.

<u>S. argenteus</u> is predominantly a schooling rabbitfish, occasionally occurring in pairs but most often in aggregates of less than 100 individuals, feeding diurnally on the algal turf of the submarine terrace and seaward slope (3-40 m in depth). These results are derived from 50 hours of underwater observation at the Tanguisson station location. Hobson (1972) found that during daylight hours, the behavior of Hawaiian reef fish is dominated by feeding, and twilight and dark periods are dominated by measures to enhance security. As observed by Popper and Gunderman (1975) for <u>S. rivulatus</u>, the schools tend to move randomly over larger portions of the submarine terrace and seaward slope.

External body morphology (i.e., fusiform shape, pointed shout, low fins, narrow caudal peduncle, and deeply incised caudal fin) and countershading coloration (blue dorsally overlain with yellow dots and commas with lighter sides) indicate that <u>S. argenteus</u> is a free-swimming, open water fish (Herre and Montalban, 1928; Schultz et al., 1953; D. J. Woodland, personal communication). Jones and Randall (1971) commonly found <u>S. argenteus</u> on the seaward slope in the vicinity of the Agana outfall. Schultz et al. (1953) stated that this species enters shallow reef flat areas and frequently becomes trapped in pools during low tide; 103 of 106 specimens collected were from Marshall Islands atolls and only three were from Guam. It is believed that his observations reflect those areas where specimens available for study were most abundant. As mentioned by Bryan (1975) for <u>S. spinus</u>, it is possible that due to the flighty or nervous behavior of <u>S. argenteus</u> and increased recreational use of the reef flat areas of Guam, sub-adult and adult <u>S. argenteus</u> less frequently enter shallow reef flat waters to feed.

Two types of feeding associations commonly occur, conspecific feeding and mixed aggregate feeding. The latter feeding association consists of occasional congeners and fish from the families Scaridae, Labridae, Acanthuridae, Mullidae, and Chaetodontidae. <u>S. argenteus</u> occupies a relatively higher position in the water column than would normally be expected for a herbivore.

<u>S. argenteus</u> is the last species of fish to enter the mixed feeding aggregates and the first to leave when approached by a diver. <u>S. argenteus</u> in the field approaches the turf substratum at an angle of  $45-90^{\circ}$  and with one or more quick bites crops the algal turf close to the bottom substratum, occasionally removing calcareous sediments with the turf. When disturbed, the siganids reschool and head for deeper, more open waters. Similar observations have been made by Woodland (personal communication) for this species.

<u>S. argenteus</u> reared in 1000 and 7000-liter tanks under laboratory conditions (both open and closed seawater systems) and fed a diet of <u>Enteromorpha</u> and trout chow exhibit schooling behavior similar to wild populations (swimming and feeding together). Occasionally, one or two fish per tank exhibited aggressive displays (i.e., raised dorsal, pelvic and anal fins; altered color pattern different from that

exhibited for fright; and lowered head swimming position) and became territorial, defending the area where food was placed in the tank. Similar conditions believed caused by improper diets, were observed by Lam (1974) and Westernhagen and Rosenthal (1975) for <u>S. canaliculatus</u> reared under laboratory conditions. I found that <u>S. argenteus</u> fed eagerly on pieces of shrimp and fish when dried food or <u>Enteromorpha</u> was not available; however, aggressive behavior became more prominent.

## Stomach Content Analysis

Stomach content analysis (Table 1) of 20 sub-adult and adult specimens of S. argenteus collected during 1974-1976, ranging from 133-256 mm fork length and 38-284 g in weight, indicate a more diverse diet than the juveniles and reflect the ability of larger fish to bite and ingest a greater variety of fleshy and calcareous algae. Dictyota, Gelidium, Bryopsis, Jania, and Codium, avoided by the juveniles, were consumed by the larger fish (respective importance values of 19.3, 6.1, 5.8, 4.5, and 1.6). The red alga Tolypiocladia glomerulata had the highest importance value of 47.8; the highly calcified green alga Halimeda discoidea, rejected by juvenile S. argenteus, was the second most important food item in the stomach analysis (IV=24.1). Galaxaura marginata, a calcareous red alga, had an importance value of 10.4. The highly calcified green alga Neomeris annulata (IV=5.0) was also common. The rank order of representation by algal divisions of the stomach contents is 42% reds, 27% greens, 21% browns, and 10% bluegreens.

Of particular interest is the relatively high frequency (RF=6.8) and importance value (IV=9.8) for the blue-green alga <u>Microcoleus</u> lyngbyaccus. The absence of blue-green algae in the diets of

Table 1. Relative abundance (RA), relative frequency (RF), and importance value (IV) of stomach contents from <u>Siganus argenteus</u> (n=20), speared from the submarine terrace and seaward slope reef zones off the Tanguisson Power Plant, 1974-1976, compared to the RA, RF, and IV of algae quantified in the same area by R. T. Tsuda, July 13, 1970. Food items are arranged in the order of importance value. A modified point method (Tsuda, 1972) was employed to quantify the benthic algae by placing a 25 cm<sup>2</sup> quadrat frame, subdivided into 5 cm<sup>2</sup>, five times around designated transect stations so that overlapping occurred.

	STO	MACH CONTI	ENTS	FIELD OCCURRENCE (61% algae, 31% rock, 7% live coral, 1% invertebrates)
SPECIES	RA	RF	IV	RA RF IV
Tolypiocladia glomerulata	34.8	13.0	47.8	36.3 18.9 55.2
Halimeda discoidea	17.9	6.2	24.1	13.0 20.8 33.8
Dictyota sp.	9.7	9.6	19.3	8.3 7.5 15.8
Galaxaura marginata	5.6	4.8	10.4	4.4 7.5 11.9
Microcoleus lyngbyaceus	3.0	6.8	9.8	2.0 1.9 3.9
Antithamnion sp.	4.5	4.1	8.6	
Ceramium sp.	1.6	6.8	8.4	3.9 1.9 5.8
Leveillea jugermannioides	2.6	5.5	8.1	
Gelidium sp.	2.0	4.1	6.1	
Bryopsis pennata	2.4	3.4	5.8	
Neomeris annulata	.9	4.1	5.0	
Sphacelaria furcigera	1.0	4.0	5.0	
Jania sp.	1.8	2.7	4.5	9.3 5.6 14.9
Feldmannia indica	2.1	2.0	4.1	
Cladophoropsis sp.	1.6	2.1	3.7	
Calothrix pilosa	1.7	1.4	3.1	
Rhodymenia sp.	.7	2.0	2.7	
Sargassum sp.	1.2	1.4	2.6	
Sponge/spicules	.6	2.0	2.6	
Polysiphonia sp.	1.0	1.4	2.4	

## Table 1. Continued

	STC	MACH CONTE	FIELD OCCURRENCE (61% algae, 31% rock, 7% li coral, 1% invertebrates)					
SPECIES	RA	RF	IV	RA RA	RF	IV		
<u>Chondria repens</u> <u>Schizothrix calcicola</u> <u>Codium edule</u> <u>Laurencia sp.</u> <u>Enteromorpha tubulosa</u> <u>Ectocarpus sp.</u> Benthic diatoms Fish scales <u>Acrochaetium sp.</u> <u>Anadyomene wrightii</u> <u>Padina (vaughaniella)</u> <u>Asterocystis ornata</u> <u>Annelid worm</u> Copepod Foraminifera	.3 .2 .9 .8 .1 .1 .1 .1 .1 .1 .1 .1 .1 1 1	1.4 1.4 .7 .7 1.4 .7 .7 .7 .7 .7 .7 .7 .7 .7	1.7 7.6 1.6 1.5 1.5 1.1 .8 .8 .7 .7 .7 .7 .7 .7	1.5	3.8	5.3		
Porolithon onkodes coralline sp. Halimeda opuntia Lobophora variegata Amphiroa sp. Lithophyllum kotschyanum Chlorodesmis fastigiata Hemitrema sp.				11.1 4.4 1.5 2.0 1.0 .5 .5	7.5 5.6 5.6 3.8 1.9 1.9 1.9 1.9	18.6 10.0 7.1 5.8 2.9 2.4 2.4 2.4		

herbivorous fish have been reported by Randall (1961) and Jones (1968b) for acanthurids and by Tsuda and Bryan (1973) and Bryan (1975) for juvenile <u>S. argenteus</u> and <u>S. spinus</u> and adult <u>S. spinus</u>.

It would appear from the present analysis that micro-invertebrates are not significant in the diet of <u>S</u>. <u>argenteus</u>; however, it has been suggested that micro-invertebrates may represent a substantial dietary supplement for S. spinus (Bryan, 1975).

Tsuda and Bryan (1973) determined that juvenile <u>Siganus argenteus</u> and <u>S. spinus</u> were highly selective in the type of algae ingested and exhibited some food preferences. <u>Chlorodesmis fastigiata</u> was avoided by <u>S. spinus</u> and devoured by <u>S. argenteus</u>, and <u>Polysiphonia</u> sp. was rejected by <u>S. argenteus</u> but devoured by <u>S. spinus</u>. As juveniles, both siganids prefer filamentous algae and avoid blue-green and calcareous algae.

Hiatt and Strasburg (1960) provide additional data on the food habits of sub-adult and adult <u>S</u>. argenteus, based on 15 specimens collected from the Marshall Islands. They report 60% calcareous material ingested with scraped algal filaments in the stomach contents. Although this species is described to be a typical browsing herbivore, they have observed adult <u>S</u>. argenteus in the vicinity of garbage dumps where they reportedly consume waste meat scraps. This indicates that it is not an obligatory herbivore and that its feeding and digestive apparatus can convert to a carnivorous diet on occasion.

A comparison of the most important algal species identified from the stomach content analysis agrees quite well with the important algal species found by R. T. Tsuda in July 1970 on the submarine terrace and seaward slope (Table 1) off the Tanguisson Power Plant.

<u>Tolypiocladia glomerulata</u>, <u>Halimeda discoidea</u>, <u>Dictyota</u> sp., and <u>Galaxaura marginata</u> had importance values of 47.8, 24.1, 19.3, and 10.4, respectively, from stomach content analysis and 55.2, 33.8, 15.8, and 11.9, respectively, from field analysis.

The electivity index of Ivlev indicates no active selection for <u>Tolypiocladia glomerulata</u> (-.02), <u>Halimeda discoidea</u> (+.16), <u>Dictyota sp. (+.08), Galaxaura marginata</u> (+.12), or <u>Microcoleus</u> <u>lyngbyaceus</u> (+.20). An electivity index of -.76 for the blue-green alga <u>Schizothrix calcicola</u> indicates possible avoidance. It would appear that the algal species ingested directly reflect the algal availability of that specific area.

## Morphological Adaption for Feeding

Table 2 presents proportional measurements and angles of possible significance in feeding of juvenile, sub-adult, and adult <u>S</u>. <u>argenteus</u> and <u>S</u>. <u>spinus</u>. The measurements used to determine morphological adaptations for feeding were those used by Jones (1968b) in comparing Hawaiian and Johnston Island Acanthuridae. <u>S</u>. <u>argenteus</u> is larger than its congener in all respective age classes as indicated by standard length, standard depth, and head length measurements. The increasing head length sets the eye back on the head allowing for a more horizontal line of sight. The ability of <u>S</u>. <u>argenteus</u> to see in a horizontal plane would also aid in predator recognition in the open waters of the submarine terrace and seaward slope. The smaller eye/ mouth angle of <u>S</u>. <u>argenteus</u> (as with <u>N</u>. <u>hexacanthus</u> and <u>N</u>. <u>brevirostris</u>) indicates that the mouth is located higher on the head more in line with the eye so the fish must look ahead for its food. Small eye/mouth

Table 2. Comparative measurements and angles of possible significance of <u>Siganus argenteus</u> and <u>S. spinus</u> which may indicate morphological adaptations for feeding. j=juvenile, s-a=sub-adult, a=adult; FL=fork length, SL=standard length, SD=standard depth, SNT=snout length, EYE=eye diameter, HL=head length, EYE/MOUTH=angle between eye and mouth. Each measurement represents the mean of 10 fish per age class. All measurements are in millimeters except the EYE/MOUTH angle which is measured in degrees.

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SPECIES		FL			SL			SD			SNT			EYE			НL		EY	E/MOUT	Н
	j	S։	a	j	s-a	a	j	s-a	a	j	s-a	a	j	5 <b>-</b> 2	a	ij	s-a	a	j	s-a	s
<u>Siganus</u> argenteus	53.8	105.0	222.6	49.5	95.1	203.3	15.4	32.0	74.2	4.9	9.3	19.1	3.8	7.4	14.9	12.3	24.9	48.7	22.6	31.8	31.7
<u>Sicanus</u> <u>spinus</u>	41.5	89.0	190.0	37.3	76.0	154.3	12.2	27.6	64.3	3.5	7.2	16.1	3.5	7.4	14.5	10.4	20.4	39.7	32.8	38.0	41.2

angles may be advantageous for certain herbivores in selecting individual algal thalli (Jones, 1968b).

Preliminary observations of <u>S</u>. <u>argenteus</u> for the presence of a notch and cusp on both upper and lower teeth, premaxillary overlap of dentary, and feeding behavior on filamentous algae in the laboratory are similar to those described by Bryan (1975) for S. spinus.

## Growth Rates

Figure 1 presents the growth rates of Siganus argenteus in comparison to S. spinus and S. canaliculatus fed diets of Enteromorpha and trout chow and held in similar holding tanks. S. canaliculatus, reared from July 1973-February 1974 were fed only trout chow for 15 weeks before the addition of Enteromorpha. Temperature, salinity, and oxygen varied from 26.7-29.8°C, 31-33°/00, and 4.8-5.8 mg/1, respectively, during the growth rate experiments. Nephelometric Turbidity Units and pH ranged from .27-.42 and 8.1-8.6, respectively. In seven months, the mean fork length and weight of S. argenteus was 187 mm and 114 g, compared to 124 mm and 29 g for S. spinus, and 158 mm and 59 g for S. canaliculatus. S. argenteus, raised in a 7000-liter circular tank with an open seawater system, and fed the same Enteromorpha and trout chow diet, measured 270 mm in fork length and weighed 390 g in 11 months. Similar growth rates were obtained for S. argenteus reared in floating cages with carangids in Tahiti (A. Michel, personal communication).

<u>S. argenteus</u> and <u>S. canaliculatus</u> remained parasite-free for the 7-month comparative growth-rate study. However, after three months,



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Figure 1. Growth rates of <u>Siganus argenteus</u> (@), <u>S. canaliculatus</u> (Δ), and <u>S. spinus</u> (o) raised in a closed system and fed <u>Enteromorpha clathrata</u> and trout chow (<u>S. canaliculatus</u> was fed only trout chow for 15 weeks before addition of <u>Enteromorpha</u>). Each data point represents the mean of 10 fish except as indicated for <u>S. spinus</u> (after 18 weeks, n=5).

the <u>S</u>. <u>spinus</u> group suffered heavy losses due to infestation of the fish's gills by a parasitic monogenic trematode and its growth rate was significantly reduced.

Figure 2 shows the length-weight relationship of <u>S</u>. <u>argenteus</u>, <u>S</u>. <u>canaliculatus</u>, and <u>S</u>. <u>spinus</u> reared under similar laboratory conditions in a closed seawater system. Regression line slopes for <u>S</u>. <u>argenteus</u>, <u>S</u>. <u>canaliculatus</u>, and <u>S</u>. <u>spinus</u> were not significantly different.

The growth rate of <u>S</u>. <u>argenteus</u> held in the three floating pens for the first four months was slow (72 mm/5 g final length and weight) compared to the laboratory group (141 mm/46 g). This is attributed to insufficient food supply, competition for food between siganids, competition for food by other herbivorous fish, and/or crowding.

When Enteromorpha was placed in the holding pens, a natural competition for food between siganids resulted, with the larger, more aggressive fish eating more algae than the smaller, slower growing fish. This was evident by the high standard deviation of the fish group in the floating pens ( $\bar{x}$ =47.3 g, S.D.=22.4 g, n=10) at the end of the 7-month period compared to similar size fish reared in the laboratory ( $\bar{x}$ =47.3 g, S.D.=13.9 g, n=10) for three months.

In constructing the holding pens, a net mesh size was chosen which would retain the juvenile fish and algae food and allow for good water flow through the net. The initial stocking density of 500 fish/ net (2.7 m<sup>3</sup>/fish) provided ample room for growth. However, algal thalli extending outside the net provided food for the resident herbivorous fish population in the intake channel, thus decreasing the



Figure 2. Logarithmic length-weight relationship of <u>Siganus</u> argenteus (\*), <u>S. canaliculatus</u> (△), and <u>S. spinus</u> (o) raised under similar conditions in a closed seawater system. Each point represents a mean of 10 fish (except <u>S. spinus</u> over 101 mm, n=5). Regression line equations are given (r=coefficient of correlation).

food source to the confined siganids. Fish of the families Scaridae, Pomacentridae, and Acanthuridae were observed feeding on <u>Enteromorpha</u> hanging through the net.

No sexually mature <u>S</u>. <u>argenteus</u> were observed at the end of the 7-month growth experiments. A second group of <u>S</u>. <u>argenteus</u> (n=41) reared in a 7000-liter tank from June 1975-May 1976 with an open seawater system and fed <u>Enteromorpha</u> and trout chow were sexually mature in 11 months. The mean fork length of these fish was 201 mm (approximately 150 g). Popper and Gundermann (1975) have reported that <u>S</u>. <u>argenteus</u> may reproduce at a larger size than other siganids, based on ripe field specimens collected in the Red Sea (450-550 g). However, <u>S</u>. <u>argenteus</u> in Tahiti showed maturation signs at 200 g and natural spawnings were observed during the months of September, October, November, and December. The eggs were pelagic, 700 microns in size. An induced spawning attempt with human chorionic gonadotropin hormone has also been successful.

#### Diseases

Behavioral observations made of fish suffering from possible vitamin B-complex deficiencies (approximately 25% of laboratory stock at times) showed a decrease in feeding efficiency, resulting in poor growth. In addition, the fish became dark in color and were unable to change their coloration pattern to blend in with background surroundings. Swimming activity decreased and the fish remained in one area of the tank. Eventually, all fish in this condition regressed to a continual swimming stage, in which the lens of the eye became

cloudy, and mono- or bilateral cataracts developed. The lens of the eye later became transparent and feeding ceased altogether. Some fish survived in this condition for several weeks until death resulted from apparent starvation.

During the growth experiments, it was noticed that the trout chow had become damp and partially moldy. Commercial feeds, such as trout chow will retain thiamine during dry storage; however, upon contact with moisture, thiaminase hydrolysis occurs and the thiamine moiety is destroyed (Halver, 1972). The coloration and activity of the fish did improve slowly over three months when they were fed fresh trout chow and <u>Enteromorpha</u>; however, no improvement was observed in fish with eye cataracts.

After seven days of vitamin injections, results from the preliminary vitamin enrichment experiment have shown that the activity and coloration of 8% of the vitamin-injected fish improved, but no other changes were observed. The incomplete recovery may indicate that a lack of B-complex vitamins was not the cause of the fish's condition. It is also possible that the vitamin dosages were too weak, the injections should have been continued for longer than seven days, or the fish may have been beyond recovery.

Halver (1972) reported that riboflavin deficiencies result in high salmonid mortality with eye opaqueness of most obvious external sign. The addition of riboflavin to the diet of salmonids will reduce the deficiency symptoms except when cataracts have developed and the protein crystal structure of the eye is lost (Halver, 1957). Westernhagen (personal communication) reported a similar condition affecting <u>S. canaliculatus</u> and <u>S. guttatus</u> when they were fed a diet of commercial chicken and rabbit feeds. He attributed this condition to a deficiency in folic acid in the feed.

During preliminary experiments in 200-liter holding tanks, <u>S. argenteus</u> juveniles developed exopthalmia in supersaturated water conditions (mean water temperature, 33.2°C ± 1.3°C; dissolved oxygen, 6.68 ppm, 111% saturation). This disease, resulting from gas embolism, is caused by the inability of the fish to maintain the internal partial pressure of oxygen in the blood and external partial pressure of oxygen in the water near equilibrium by normal gas diffusion across the gill filament surfaces (Duijn, 1971). As a result, nitrogen becomes supersaturated in the blood and gas bubbles accumulate between the cornea and lens of the eye.

The affected signaids recovered when aeration was added to increase gas diffusion in the water of the influent troughs. Exopthalmia was reported by Lam (1974) in culturing <u>S. canaliculatus</u>; however, the cause of the condition was not stated.

Parasitic trematodes have previously been described from siganids by Goto and Ozaki (1929), Yamaguti (1934, 1939), Young (1967), and Manter (1969). Several trematode species may be specific to siganids (Paperna, 1972). Although I have not found any trematodes on <u>S. argenteus</u>, a parasitic trematode, tentatively identified by D. I. Gibson as <u>Microcotyle mouwoi</u>, has been found on the gills of <u>S. spinus</u> from Guam and caused heavy losses to this species during growth studies. This microcotylid monogenean was originally described by Ishii and Sawada (1938) from the gills of <u>S. fuscescens</u> from Japan.

No symptoms of the disorder prior to the fish's death were noticed. Upon examination, the fish's gills appeared pale and were covered with

a heavy mucous secretion. This secretion, consisting of slime, presumably produced by the fish to rid the gill of parasites and destroyed epithelial cells, decreased the respiratory surface area on the gill filaments, causing the fish to suffocate (Davis, 1953). Lam (1974) described a similar infestation of monogenean trematodes in attempts to culture <u>S. canaliculatus</u>. Three field specimens of <u>S. argenteus</u> examined by Paperna (1972) were free from monogenea.

## Environmental Parameters

## Temperature Tolerances

The mean survival rates based on four runs of Series A (ambient), B (+2°C), and C (+4°C) fish were 100, 94, and 79%, respectively, for the 14-day experiments (Figure 3). Series D (+6°C) fish had 50% mortality in 2.7 days, 96% mortality in 5 days, and 100% mortality in 8 days. The upper tolerance limit, defined by Hoff and Westman (1966) as the temperature by and at which 50% of the experimental animals can no longer live for a designated exposure period, for <u>S. argenteus</u> is  $33.7°C \pm .3°C$  in 2.7 days. Earlier trial experiments with <u>S. argenteus</u> juveniles in Series D tanks resulted in 100% mortality in 20 hours when temperatures rose to 36.3°C.

To maintain the desired temperatures (30, 32, and 34°C), adjustments with the thermal controller were made at 0800, 1200, 1700, and 2100 hours when necessary. Regulation of thermal Series C and D temperatures was most difficult during two of the runs because of heavy rains and frequent power outages. As a result, the lower mean temperatures may have increased the survival time of these fish. The



Figure 3. Mean survival rate of <u>Siganus argenteus</u> in four replicate thermal experiments at ambient  $(28^{\circ}C \pm .4^{\circ}C)$ , +2 (29.7°C  $\pm$  .4°C), +4 (31.8°C  $\pm$  1.4°C), and +6°C (33.7°C  $\pm$  .3°C) temperatures. Vertical lines indicate standard deviations.

dissolved oxygen concentrations in the thermal series tanks ranged from 6.01 to 6.68 ppm (94-113% saturation).

The fish in Series A, B, and C thermal tanks acted normally, either schooling or hiding along the tank bottom. The fish in Series B tanks consumed more food than those in Series C. Normal consumption occurred in Series A; no food was consumed by Series D fish. The behavior of Series D fish was noticeably different from that of the other fish. After schooling for two hours, the rabbitfish separated and remained in the more shaded corners of the tank. In 24 hours, the bodies of the fish appeared swollen. The coloration of the frontal head area of the fish became dark and the body turned pale gray with yellow outlines around the base of the dorsal and pelvic fin and caudal peduncle. Activity of the fish progressed from quiescent and sluggish swimming periods to erratic, spasmotic movements near death.

Previous growth measurements made on <u>S</u>. <u>argenteus</u> indicated substantial increments of growth in 14 days (see Figure 1). Analysis of covariance revealed no significant weight increase of or between Series A, B, or C fish (Sokal and Rohlf, 1969). Regressions of weight gain for each temperature series were not significantly different.

Drew (unpublished report) stated from field experiments that <u>S. canaliculatus</u> can tolerate temperature fluctuations of 23-36°C recorded in situ in the <u>Enhalus acoroides</u> beds in Palau. Lam (1974) also reported the ability of <u>S. canaliculatus</u> to tolerate temperature fluctuations of 25-34°C while maintained in laboratory holding tanks. <u>S. canaliculatus</u> and <u>S. guttetus</u> have also been reared by Westernhagen and Rosenthal (1975) in 25-30°C water. These reports indicate tolerance to brief temperature fluctuations but provide no information on the effects of sustained elevated temperatures in siganid culture.

Salinity Tolerances

While present on the reef flat areas around Guam, <u>S. argenteus</u> juveniles feed on the filamentous alga <u>Enteromorpha clathrata</u>, which grows luxuriantly in the intertidal zone (FitzGerald, 1976). Because of the structure of Guam's limestone aquifer and ground-water reservoir (Randall and Holloman, 1974), the salinity in this area is reduced, with exact levels depending on the amount of ground-water runoff. It is evident from the duration of time the fish spends feeding in this reef flat area that <u>S. argenteus</u> can tolerate some degree of salinity reduction.

The mean size and weight of fish used to determine the lower salinity limit range was 74 mm fork length and 6 g, respectively. Oxygen concentration and temperature in the experimental and control tanks varied from 6.1 to 7.0 ppm ( $\bar{x}$ =6.4 ppm) and 24.7-30.3°C ( $\bar{x}$ =26.1°C), respectively. A mean temperature change of .3°C occurred in the experimental tanks after the addition of dechlorinated tap-water.

Figure 4 shows the survival of <u>S</u>. <u>argenteus</u> (n=80) after daily salinity reductions of  $1^{\circ}/_{\circ\circ}$  from 8 to  $2^{\circ}/_{\circ\circ}$ . Survivorship ranged from 100% at 7°/ $_{\circ\circ}$  salinity to zero at 2°/ $_{\circ\circ}$  salinity. At salinities of 5, 5, 4, and 3°/ $_{\circ\circ}$ , survivorship was 99, 91, 36, and 6%, respectively. The lower salinity limit, defined as the interval from which survivorship is reduced from 100% to 50% as determined by this experiment, is between 4°/ $_{\circ\circ}$  and 7°/ $_{\circ\circ}$  salinity.





As the lower salinity limit was approached, the fish appeared light in color, unable to match their coloration with background surroundings. Their bodies were noticeably swollen with spines erect, and they were unable to maintain a horizontal, upright swimming position. Short erratic periods of activity continued until death. Apparently, the fish were unable to remain hypo-osmotic to the seawater environment, due to the increased ion and water absorption, and equilibrium was impaired.

During the second salinity study conducted for 96 hours, the mean water temperature was 30.6°C and the dissolved oxygen concentration and pH varied from 6.2 to 6.6 ppm and 8.11 (initial) to 8.78 (final), respectively. Although 100% survival was recorded for fish held in Series A  $(10^{\circ}/_{\circ\circ})$ , B  $(8^{\circ}/_{\circ\circ})$ , and C  $(6^{\circ}/_{\circ\circ})$ , the quantitative observations made of fish response to tactile and light stimuli, and effect of reduced salinity on coloration, equilibrium, and feeding indicate various degrees of metabolic inhibition (Table 3).

The first noticeable negative response to reduced salinity is a change in color from a light to dark phase. This is followed by a negative light response and reduced tactile (flight) and equilibrium response. In 72 hours, 25% of the fish in Series A ( $10^{\circ}/\circ \circ$ ) and B ( $8^{\circ}/\circ \circ$ ) tanks were dark in color and exhibited a negative response to light. In Series C ( $6^{\circ}/\circ \circ$ ), the same conditions occurred in 48 hours to 50% of the fish. Equilibrium was also impaired and tactile response reduced. Feeding ability was positive throughout the study for Series A and B fish and negative for Series D fish. Feeding ability, initially negative for Series C fish, became positive after

	<u> </u>		NUMB	ER OF F	ISH SHOWING N	ORMAL RESPON	SES
SERIES	HOURS	OF FISH	TACTILE	LIGHT	EQUILIBRIUM	COLORATION	FEEDING
А	24 48 72 96	12 12 12 12	12 12 12 12	12 12 9 8	12 12 12 12	11 10 9 8	+ + + +
В	24 48 72 96	12 12 12 12	12 12 12 12	10 10 9 8	12 12 12 12	10 9 9 8	+ + + +
С	24 48 72 96	12 12 12 12	12 12 11 9	5 5 5 6	10 10 10 9	7 7 5 6	- + +
D	24 48 72 96	8 6 5 3	2 2 2 2	2 1 1 1	2 2 2 2	2 1 2 2	- <u>a</u> / _a/

Table 3.	Response of Siganus argenteus to salinities of 10 (Series A)	,
	8 (Series B), 6 (Series C), and 4°/00 (Series D) for	
	96 hours. A (+) indicates a normal response for feeding.	

 $\frac{a}{Indicates}$  a positive response by one fish in one of the three series tanks.

24 hours for the remainder of the study. This may indicate partial acclimation at the  $6^{\circ}/_{\circ\circ}$  salinity level.

The survival of <u>S</u>. <u>argenteus</u> at  $4^{\circ}/_{\circ\circ}$  salinity is reduced to 50% after 48 hours (Figure 5). At 96 hours, survivorship was reduced to 25%.

Results from the third study to determine the effects of reduced salinity on the growth rate and survival of <u>S</u>. <u>argenteus</u> over a 1-month period have shown an increase in growth (weight) of 77, 59, and 51%, respectively, for fish held in 20, 10, and  $30^{\circ}/_{\circ\circ}$  salinity water. Analysis of covariance (Sokal and Rohlf, 1969) has shown no significant difference in growth rate for <u>S</u>. <u>argenteus</u> grown in the reduced salinity water. The survival rate was 94% for fish held in 20 and  $30^{\circ}/_{\circ\circ}$  salinity water and 81% for the  $10^{\circ}/_{\circ\circ}$  salinity fish.

Habitat observations of siganids indicate that they are subjected to wide fluctuations in salinity. Drew (unpublished report) stated that <u>S. canaliculatus</u> in Palau tolerates salinity fluctuations of  $17-37^{\circ}/\infty$ . Lam (1975) reported that <u>S. canaliculatus</u> can be gradually acclimated to  $5^{\circ}/\infty$  salinity and thrives in 33-50% seawater. Similar acclimation studies were conducted by Lavina and Alcala (1973), but they found <u>S. canaliculatus</u> unable to tolerate a medium of 25% seawater (8.75°/ $\infty$  salinity). <u>S. rivulatus</u> and <u>S. luridus</u> have established populations in the Mediterranean Sea by passing through the Suez Canal from the Red Sea (Ben-Tuvia, 1966). Popper and Gundermann (1975) concluded that <u>S. rivulatus</u> and <u>S. luridus</u> tolerate 20-50°/ $\infty$  salinity water; however, mortality increases and growth decreases at the extremes. They also found no significant difference





in growth rate for juvenile <u>S</u>. rivulatus and <u>S</u>. luridus reared in 20, 30, 40, and  $50^{\circ}/_{\circ\circ}$  salinity water for six months.

## Oxygen Tolerances

Temperatures in the experimental and control tanks ranged from 26.0-29.4°C. A maximum variation of 1.5°C occurred over 24 hours from insolation in the aquaria with 3.0 ppm dissolved oxygen. A general trend of decreasing pH, ranging from 8.39 to 8.04, was observed during each experiment with a maximum decrease of .30 in the 2.0 ppm dissolved oxygen aquaria. Salinity remained constant at 33°/oo throughout the experiments.

Table 4 shows the mean change in dissolved oxygen concentration in the experimental and diffusion control tanks. In the .5 and 1.0 ppm dissolved oxygen experiments, the diffusion of oxygen from the tank atmosphere into the water in the experimental tanks (+.1 mg  $1^{-1}$  hr<sup>-1</sup>) was appreximately the same as the control tank #2 diffusion rates (+.07 and +.06 mg  $1^{-1}$  hr<sup>-1</sup>). At dissolved oxygen concentrations of 2.0 and 3.0 ppm, the change in oxygen concentration from fish respiration (-.19 and -.13 mg  $1^{-1}$  hr<sup>-1</sup>, respectively) was greater than the diffusion rate of oxygen into the water (+.09 and +.04 mg  $1^{-1}$  hr<sup>-1</sup>, respectively).

Oxygen consumption rates, calculated from the mean change in oxygen concentration per hour in the experimental tanks minus the diffusion rate, were approximately .1 mg  $1^{-1}$  hr<sup>-1</sup> for fish maintained in dissolved oxygen concentrations of 2.0 and 3.0 ppm. Fish in control #1 tanks at an initial oxygen concentration of 5.4 ppm showed an average oxygen uptake of .15 mg  $1^{-1}$  hr<sup>-1</sup>. Table 4. Mean change in dissolved oxygen concentration in the .5, 1.0, 2.0, and 3.0 ppm experimental tanks (n=4) and control tanks. A (+) indicates diffusion of atmospheric oxygen into the water and a (-) indicates removal of the dissolved oxygen in the water by fish respiration.

INITIAL [02] ppm	∆ [0 <sub>2</sub> ] mg 1	EXPERIMENT			
Experimental tanks	Experimental Tanks (1 fish/tank)	Control <sub>a/</sub> #2 Tanks— (no fish)	DURATION (hr)		
.5	+.10	+.07	2.6		
1.0	+.10	+.06	6.5		
2.0	19 <sup>b/</sup>	+.09	24		
3.0	13 <sup>c/</sup>	+.04	24		

 $\frac{a}{Diffusion}$  rate across the tank atmosphere-seawater interface.  $\frac{b}{\Delta} [0_2] \text{ mg } 1^{-1} 2 \text{ hr}^{-1}$  divided by 2.  $\frac{c}{\Delta} [0_2] \text{ mg } 1^{-1} 4 \text{ hr}^{-1}$  divided by 4. Stress signs developed immediately after the fish were placed in the .5 and 1.0 ppm experimental tanks. All fish remained in the lower half of the tank, resting against the side of the tank or tank bottom, except for occasional erratic movements to the surface. A general lack of coordination and equilibrium was observed. The coloration of the fish became pale with dark areas around the head and sides, unlike the mottled fright pattern coloration. Opercular movements became more shallow and ventilation rates increased from 139/min to 300+/min for fish in the 1.0 and .5 ppm dissolved oxygen concentrations. The average survival time for <u>S. argenteus</u> in water of .5 and 1.0 ppm dissolved oxygen concentration was 1.5 and 4.6 hours, respectively.

The activity of <u>S</u>. <u>argenteus</u> held in water of 2.0 and 3.0 ppm dissolved oxygen concentration was characterized by periods of inactivity on the tank bottom, occasionally disrupted by periods of short swimming activity. Opercular movements appeared slower and deeper than in fish held in .5 or 1.0 ppm dissolved oxygen concentration water. Ventilation rate, monitored by opercular movements, averaged 270/min and 192/min for fish in the 2.0 and 3.0 ppm water, respectively. The experiments were terminated after 24 hours with 100% survival of fish.

Westernhagen and Rosenthal (1975) provide the only account of dissolved oxygen concentrations continuously monitored during growth experiments with <u>S. canaliculatus</u> and <u>S. guttatus</u> (3.81 ppm  $\pm$  .97 ppm). Lavina and Alcala (1973) and Lam (1974) both report that <u>S. canaliculatus</u> is sensitive to dissolved oxygen concentrations below 2.0 ppm.

## CONCLUSIONS

The mariculture potential of Siganus argenteus on Guam is encouraging. Juvenile stocks may be acquired locally during the months of April, May, June, and October. Despite being an open water, schooling herbivore, the fish adapts well to confinement in 1000 and 7000-liter holding tanks and accepts pelletized commercial food, which indicates its suitability for pond culture. The conditions which provide optimum growth of Enteromorpha clathrata, the preferred food of the juveniles, are known (FitzGerald, 1976) and may be utilized to provide an unlimited, inexpensive food source. S. argenteus has spawned naturally in captivity in Tahiti and initial induced spawning attempts have been successful (A. Michel, personal communication). Of the two most abundant siganids on Guam, S. argenteus has a faster growth rate and larger attainable size in seven months than S. spinus grown under similar laboratory conditions and fed a diet of Enteromorpha and trout chow (terminal lengths and weights of 187 mm/114 g and 124 mm/29 g, respectively).

<u>S. argenteus</u> juveniles are tolerant to increased water temperatures and reduced salinity and oxygen concentrations. Survival rates of fish subjected to water temperatures of 28, 30, and 32°C were 100, 94, and 79%, respectively, over 14 days. Fish held in water of 34.0°C had 50% mortality in 2.7 days, 96% mortality in 5 days, and 100% mortality in 8 days. The lower lethal salinity limit of <u>S. argenteus</u> was 4-7°/oo with survival reduced to 50% after two days at 4°/oo. Survival of <u>S. argenteus</u> for 24 hours at 2.0 and 3.0 ppm dissolved oxygen concentrations was 100%; however, at .5 and 1.0 ppm, survival times were reduced to 1.5 and 4.6 hours, respectively. Environmental extremes such as those tested on <u>S</u>. <u>argenteus</u> infrequently occur in mariculture operations; however, knowledge of adaptation to these extremes is essential to prevent complete loss of a mariculture stock during operation failures.

Major disadvantages must be minimized before the mariculture of <u>S. argenteus</u> on Guam will become a reality. Because of the yearly fluctuation in the number of juveniles appearing on the reef flat (Kami and Ikehara, In Press), induced spawning and larval rearing techniques must become routine to provide a continuous supply of juveniles. An inexpensive, high-protein dietary supplement must be found to replace expensive trout chow (\$.60 lb including shipping). Areas needing additional research, as implied above, include induced spawning and larval rearing, larvae-adult nutrition and food conversion, and diseases.

It is interesting to note that the local demand and market value of siganid juveniles (manahac) are higher than for the adults. Serious consideration should be given to rearing fish only to the juvenile stage for market in lieu of strict juvenile-to-adult culture. Advantages of the former would be decreased management costs, faster growth of fish to marketable stage, and several marketable crops per year. The juveniles may also represent an important source of baitfish which would be essential in developing Micronesia's tuna fishing industry. Juvenile-te-adult culture would be continued to provide a dependable source of breeding stock.

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