

AN ABSTRACT OF THE THESIS OF Ronald D. Strong for the Master of Science in Biology presented April 17, 1975.

Title: Distribution, Morphometry, and Thermal Stress Studies on Two Forms of Linckia (Asteroidea), on Guam

Approved:

Masashi Yamaguchi
MASASHI YAMAGUCHI, Chairman, Thesis Committee

Two similar forms of the asteroid genus Linckia were surveyed by a timed swimming count at eight locations around Guam. Linckia laevigata, cobalt blue in color, was found mainly on the reef flats but larger specimens occurred as deep as ten meters, whereas Linckia species, brown in color, usually occurred between depths of ten and 20 meters. Linckia species tends to have longer, more slender arms than L. laevigata. The rate of oxygen uptake increased between 25° and 34° C in Linckia species. It also increased between 25° and 31° C, but leveled off between 31° and 34° C in L. laevigata. At 36° C, the rate of oxygen consumption in both forms of starfish decreased, indicating a disturbance in metabolic activity because of increased temperature. Linckia laevigata was more tolerant to short exposures at 36° C and survived for longer periods of time when maintained at 34° and 36° C than did Linckia species. Thus, L. laevigata seems better adapted for living in a reef flat environment than does Linckia species. Phylogenetic relationships between the two forms remain unclear.

TO THE GRADUATE SCHOOL:

The members of the Committee approve the thesis of RONALD D.
STRONG presented April 17, 1975.

Masashi Yamaguchi
MASASHI YAMAGUCHI, Chairman

Lucius G. Eldredge
LUCIUS G. ELDREDGE

Lynn Raulerson
C. LYNN RAULERSON

Don K. Worsencroft
DON K. WORSENCROFT

ACCEPTED:

James A. McDonough
JAMES A. MCDONOUGH
Dean of the Graduate School

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INTRODUCTION

Previous experiments have shown that many coral reef invertebrates have a narrow upper tolerance for temperature (Mayer, 1914; Cary, 1931; Edmondson, 1928, 1946; Rupp, 1973; Yamaguchi, 1974). Above this limit, feeding and respiration (metabolic activity) are seriously disturbed, with death eventually occurring above a critical temperature. Normal temperatures experienced on the reef and reef front areas around Guam vary from 26° to 29° C year round, although on shallow reef flat pools during low spring tides and intense solar radiation temperatures may exceed 36° C. A man-induced source of increased water temperatures on coral reefs is the thermoelectric power plant such as the one located at Tanguisson Point, Guam. This power plant uses seawater for condenser cooling and then releases the effluent directly onto the reef flat. Studies at Tanguisson have shown the discharge temperature varies from 32.8° to 37.8° C, with a mean of 33.8° C (Jones and Randall, 1973).

Very little work has been done on the thermal tolerance of tropical echinoderms. Temperature effects on respiration of temperate echinoids have been investigated by Farmanfarmaian and Giese (1963), Johansen and Vadas (1967), McPherson (1968), and Ulbricht and Pritchard (1972). Rupp (1973) tested the effects of temperature on the fertilization and early cleavage of some tropical echinoids and asteroids, including Linckia laevigata. Yamaguchi (1974) studied the effect of temperature on the metabolic activity of Acanthaster planci.

The asteroid genus Linckia is a very conspicuous and common component of the reef fauna, occurring throughout the tropical Indo-West Pacific area. When one examines the various areas along the reef, an apparent zonation of the forms of Linckia is evident. Linckia laevigata is commonly found only in a shallow, reef-flat habitat, whereas Linckia sp. is generally found only in the deeper areas of the reef front and reef terrace (Yamaguchi, in press).

The reef flat is an unique environment and organisms living there must be able to withstand rather severe environmental changes. These stresses include increased water temperatures, occasional exposure and reduced salinities caused by fresh water runoff during periods of high rainfall. An asteroid such as Linckia laevigata which inhabits the reef flat would be better adapted to these stresses than other starfish. Prior work (Yamaguchi, 1974), on thermal stress of Acanthaster planci which is rarely found on the reef flat, suggested a comparison of Acanthaster with L. laevigata. One might find greater resistance to thermal stress in L. laevigata than in A. planci.

Another large form of Linckia common on the reef is Linckia sp. Like Acanthaster, this starfish is rare in the reef flat area. In addition, Linckia sp. may be an eco-form of L. laevigata, as they appear almost identical in form, differing only in color and certain minor characteristics in morphology. A comparison of the thermal tolerance of L. laevigata and Linckia sp. is made to test the hypothesis of thermal stress as a limiting factor of a local distribution pattern. Some comparative comments are made with reference to similar studies on A. planci made by Yamaguchi (1974).

MATERIALS AND METHODS

A. Field

In the field, observations of reef areas were made to determine the habitats, depths and apparent zonation of Linckia laevigata and Linckia sp. In all, eight locations were surveyed using Scuba (see Fig. 1). The locations were chosen to give a variety of habitats and depths, and included one station (#8) on the windward (eastern) side of Guam. Preliminary surveys had indicated a lack of asteroids on the windward coast.

Preliminary field work had shown that Linckia individuals are generally aggregated, and these aggregations are randomly distributed. This type of distribution would rule out the point-quarter and random pairs sampling techniques. A 100 m transect line was tried, where the whole length was searched out 10 m to either side. This method was discontinued as it yielded a very low number (less than 10) of starfish per transect.

Therefore, a timed swimming-search approach was used. Search times of 30 minutes were used and the following data were recorded on an underwater slate: the time of sighting a starfish, species, arm radii (minimum and maximum), depth, substrate and any unusual morphological features. On a reef slope or submarine terrace, a zig-zag pattern of search was used allowing even coverage to all depths down to 30 meters. Although not as accurate as quadrat sampling, the swimming count method enabled the diver to count adequate numbers of starfish and to search a greater area.

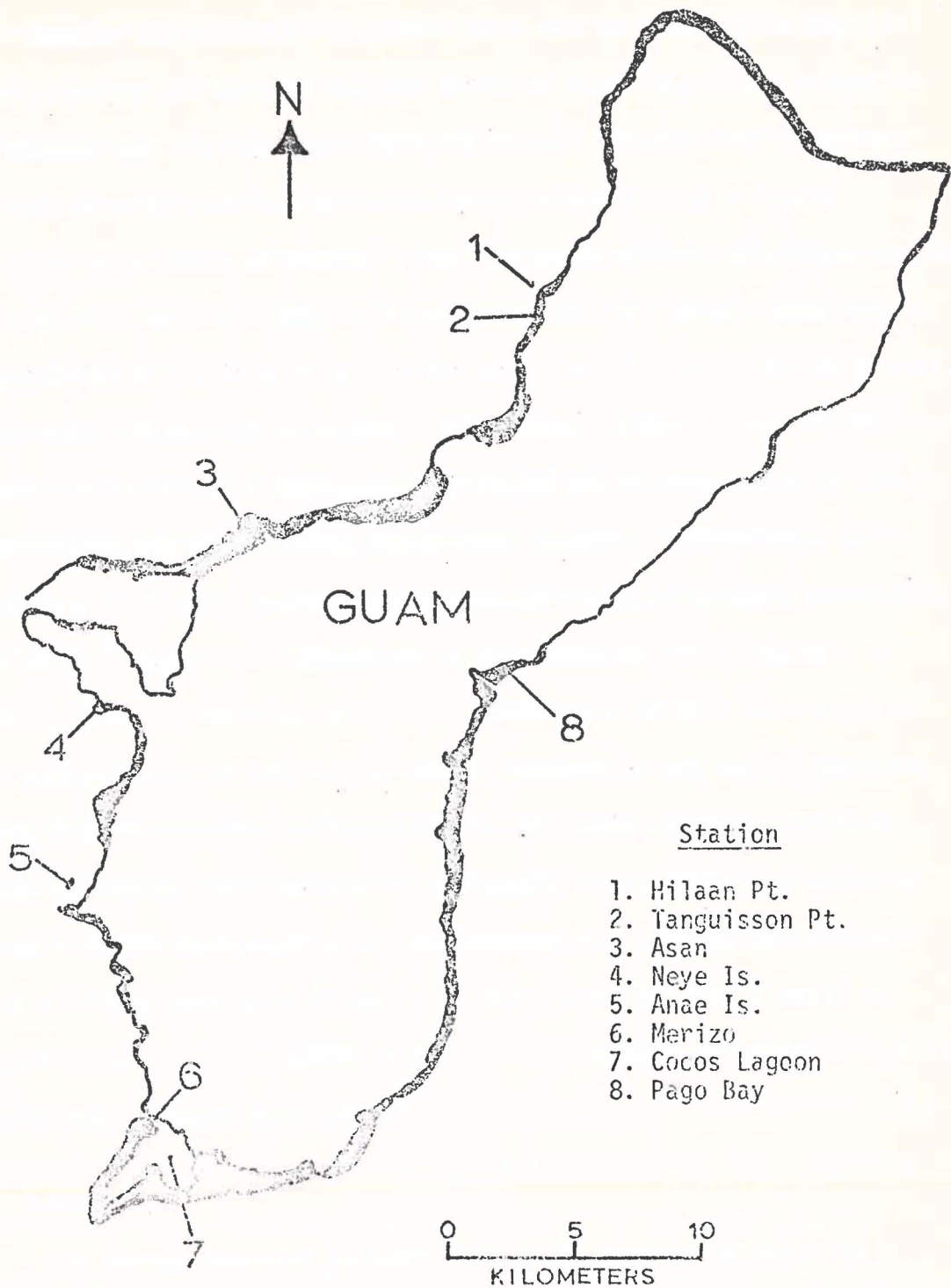


Figure 1. Eight Linckia survey locations. The shaded areas indicate fringing reefs.

Another objective of the field work was to obtain some physical and morphological data on the two forms of starfish, since a species difference has not been clearly established. Twenty-five specimens of each form of starfish were collected, representing a nearly complete size range of adults. The following weights and measurements were taken: each arm length (from center of oral disc), each arm diameter (out 1 cm from edge of disc), disc radius (between each arm), underwater weight and wet weight. Measurements were taken to the nearest 0.1 mm with a caliper. Underwater weight was taken to the nearest 0.1 g with an Ohaus Dial-O-Gram balance, supporting a perforated plexiglass platform which was submerged in a large round tank filled with seawater. Measurements were made at 28° C (water temperature) and 35 ‰ salinity. Underwater weights were used as the permeability of body walls causes inaccurate wet weight measurements (see discussion by Yamaguchi, 1974). Wet weights were also taken by drying the starfish as thoroughly as possible and weighing it on a triple beam balance to the nearest 0.1 g. Ten specimens of each starfish were also dried in a drying oven at 110° C until no significant change in weight occurred. Dry weight was established as drying for 24 hours at 110° C.

B. Laboratory

The energy expended by many animals in expressing relative fitness or adaptation to a given habitat or stress is reflected in their basal metabolism rate. Thus the energy expenditure caused by changing temperature can be reflected by oxygen uptake in an animal at rest, since the rate of oxygen uptake is directly related to

temperature in poikilotherms (see review, Vernberg and Vernberg, 1970). Thermal stress studies were conducted on Linckia laevigata and Linckia sp. using oxygen consumption as an index. All work was done at the University of Guam Marine Laboratory.

Collections of both forms of Linckia were made weekly from February 1974 to September 1974. The reasons for obtaining specimens fresh from the field were twofold. First, the need for well-fed animals has been shown, since starved animals show a drastic reduction in oxygen consumption (Farmanfarmaian, 1966; Yamaguchi, 1974). Since the feeding preferences of Linckia have not been established, it is not possible to keep the starfish in holding tanks for long periods of time to insure their nutritional state. Secondly, a type of infectious growth often appears on starfish which are held in tanks. This appears as a slimy, milky-white growth (possibly connective tissue) which spreads rapidly over the surface of Linckia. It has been observed to occur as soon as 24 hours after field specimens are brought to the laboratory, but sometimes may not appear even after several weeks.

The starfish were placed in large holding tanks approximately 1.2 meters in diameter which featured an enclosed filtering and water circulation system. Prior to an oxygen determination run, starfish were placed in two holding aquaria to adjust to the experimental temperature for a period of 12 hours. These were 40-liter rectangular plastic aquaria with a double air-lift driven sand-bed filter system. The temperature was maintained in these acclimation tanks by aquarium immersion heaters regulated by a thermostat to the desired temperature.

Two respiration chambers were placed inside an aquarium identical to the holding aquaria. Each respiration chamber consisted of a cylindrical four-liter Lexan container fitted with a water-tight plexiglass lid held in place by latex tubing (see Fig. 2). Three holes were drilled in the top. A glass shaft with a three-bladed plastic propeller connected to a motor of about 120 rpm was placed in the center hole. The other two holes at opposite edges of the lid held plastic tubing which allowed for water circulation between the chamber and the holding aquarium. Circulation was accomplished by a Cole-Parmer masterflex tubing pump at a rate of approximately 1.5 liters/min.

Standard metabolic activity of Linckia laevigata and Linckia sp. was determined at 25, 28, 31, 34 and 36° C. Experimental temperatures were maintained by using a Versa-Therm electronic temperature controller in conjunction with a mercury thermoregulator and heating elements. These units provided very precise and constant temperatures ($\pm 0.05^\circ$ C) necessary for this work.

After the starfish had been adjusted to the experimental temperature for 12 hours, each starfish was transferred to a respiration chamber and the cover sealed. The propeller and water circulation pumps were activated to provide both water circulation and agitation within the chambers. This was continued for 60 minutes to allow the water to thoroughly mix and to enable the starfish to maintain a stable position of rest (no movement). It was necessary for the animals to remain quiescent during the experimental period (usually six hours) to properly measure the metabolic rate. Generally, the starfish would affix themselves inverted on the cover of the

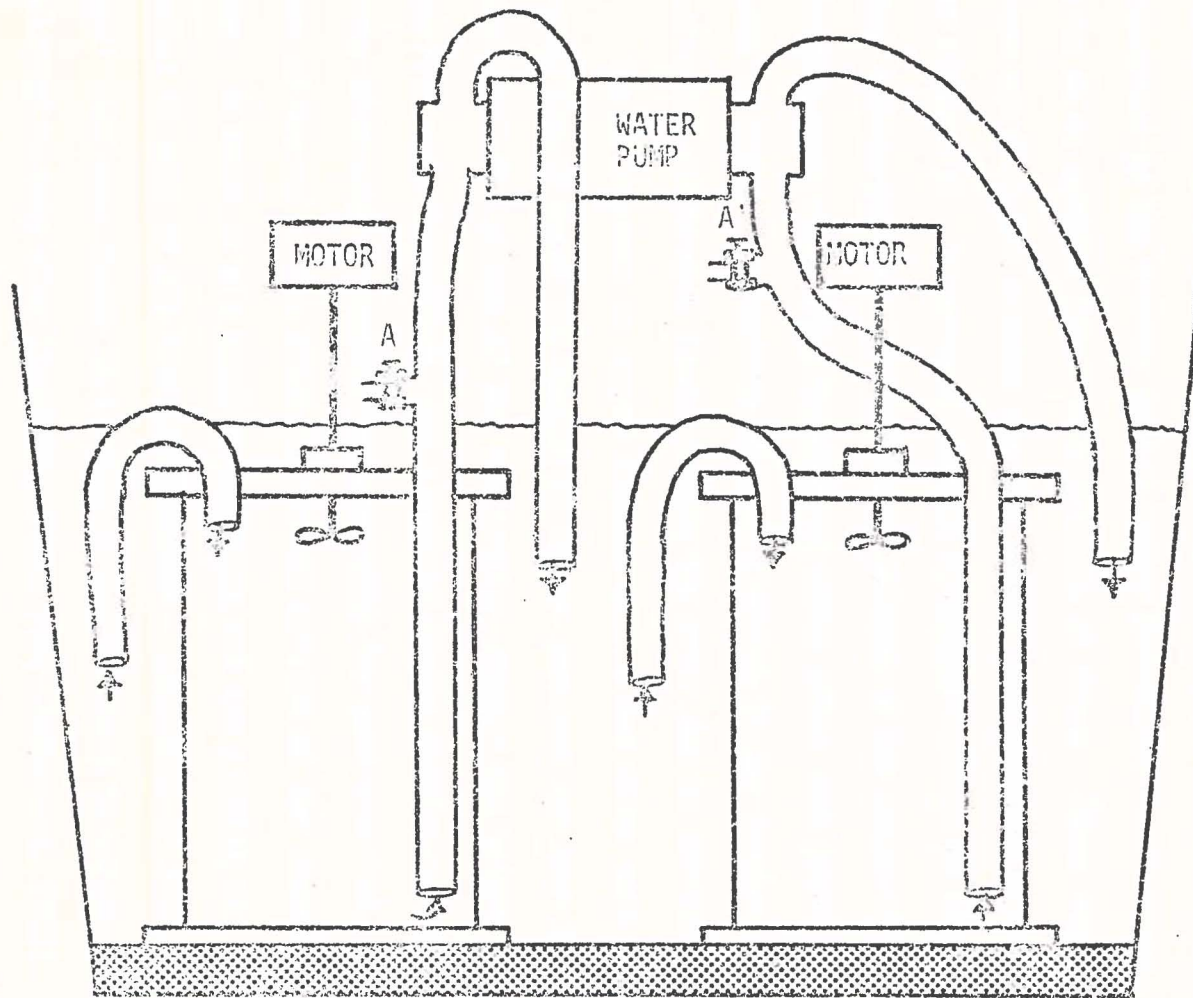


Figure 2. Respiration chambers situated in the thermally controlled aquarium. The arrows indicate water movement. The points where water samples were siphoned from the chambers are indicated by A and A'.

chamber and remain there for the duration of the experiment. After one hour of water circulation, the circulating pumps were stopped, isolating the water in the respiration chamber. Mixing of water within the chamber was continued by the propeller. Oxygen concentration of the water at the beginning and end of a 60-minute run was determined by the Winkler method (after Strickland and Parsons, 1968), using 60 ml samples taken in replicates of three. These samples were drawn out from the chamber via the siphon tube equipped with a shut-off (Figure 2). To prevent changes in oxygen because of algae and other microorganisms, the interior surfaces of the respiration chambers were thoroughly cleaned and dried after each day's use. Tests of oxygen consumption with no starfish in the chamber showed a negligible difference (smaller than the error due to titration).

The difference in oxygen content before and after each 60-minute period should equal the amount of oxygen consumed per hour by the starfish. The volume of the respiration chamber and weight of the starfish were determined, and a rate of oxygen uptake was obtained. Three 60-minute determinations were made for each starfish, with one hour of mixing of the water inside and outside of the chamber between runs. A total of six Linckia laevigata and six Linckia sp. were tested at each of the five temperatures. Specimens of approximately equal size were chosen to minimize the difference in oxygen consumption relative to body size (Farmanfarmaian, 1966; McPherson, 1968). Underwater weights were taken (see Field section) immediately prior to each experiment.

Since rate of oxygen uptake in poikilotherms is affected by factors other than temperature, all other parameters were closely

controlled. Salinity was maintained at 35.00 ± 0.20 ‰ by the addition of distilled water when evaporation occurred. The pH varied between 8.0 and 8.2.

RESULTS AND DISCUSSION

A. Field

Time swimming counts were conducted at eight locations around Guam (see Fig. 1). The Hilaan Point area surveyed is a large mound approximately 300 meters from shore which rises to within five meters of the surface. The sides are steeply sloped and drop off to over 30 meters on the northern edge and to about 15 to 20 meters on the southern and seaward slopes. Coral cover is rather barren except for Millepora and a few encrusting coral species. There is often a strong current present. The area contains numerous holes, crevices and depressions which provide good hiding places for starfish. This area supports the largest and most dense population of Linckia sp. of any area the author has seen in almost three years of diving Guam's reefs (see Table 1).

Tanguisson Point, the site of the Tanguisson Power Plant, is about 0.8 km to the south of Hilaan Point. Thermal discharge has killed most of the reef life on the reef flat and reef margin directly in front of the discharge channel. This area is characterized by a wide submarine terrace (up to 100 m wide) gently sloping from a depth of about five meters to 15 meters where the seaward slope drops sharply to a second submarine terrace at about 30 meters deep. This area has numerous low relief mounds, pinnacles, and rocks, with occasional channels which provide numerous holes and cracks for hiding. Coral cover has increased since the Acanthaster infestation of 1968-69 left over 95 per cent of the hermatypic coral dead in

Table 1. Results of Linckia surveys at eight locations on Guam.

| Station and Number of 30- minute counts | <u>Linckia laevigata</u> | | | | <u>Linckia sp.</u> | | | |
|---|-----------------------------------|---------------------------------|---------------------------|--------------------------|-----------------------------------|---------------------------------|---------------------------|--------------------------|
| | Number and Percent of Total | Density per Diver Hour | Mean Depth (Meters) | Median Radius (mm) | Number and Percent of Total | Density per Diver Hour | Mean Depth (Meters) | Median Radius (mm) |
| 1. Hilaan Pt. (6) | 0 (0) | 0 | -- | -- | 174 (100) | 58 | 12.5 | 178 |
| 2. Tanguisson Pt. (6) | 1 (1) | 1 | 9.0 | 150 | 67 (99) | 22 | 11.0 | 185 |
| 3. Asan (2) | 58 (100) | 58 | 3 | 98 | 0 (0) | 0 | -- | -- |
| 4. Neye Is. (5) | 72 (82) | 29 | 7.1 | 161 | 16 (18) | 6 | 10.7 | 186 |
| 5. Anae Is. (4) | 91 (100) | 45 | 3 | 113 | 0 (0) | 0 | -- | -- |
| 6. Merizo (2) | 4 (100) | 4 | 3 | 140 | 0 (0) | 0 | -- | -- |
| 7. Cocos patch reef (2) | 5 (100) | 5 | 5.3 | 139 | 0 (0) | 0 | -- | -- |
| 8. Page Bay (4) | 0 (0) | 0 | -- | -- | 0 (0) | 0 | -- | -- |

this area (Randall, 1973). However, the coral relief is low with many areas still barren of live coral. This however seems to be an ideal habitat for Linckia sp., since this area also supports a large population of this starfish (Table 1).

The Asan location was surveyed from shore along the extensive reef flat (approximately 200 m wide) down to the second submarine terrace at a depth of about 30 meters. No specimens were located beyond the reef-flat zone, even though the topography and coral cover are very similar to that of the Tanguisson area. The population on the reef flat is extensive but composed of small Linckia laevigata (see Table 1). The mean arm radius of the individuals of this population, which has been monitored over a period of 12 months (Yamaguchi, personal communication), has not increased significantly, averaging about 100 mm in radius. No individuals were located in deeper water out from the reef flat as was the case at Anae Island.

Neye Island, located at the north of Agat Bay, is connected to the shore by a very shallow, narrow reef flat. This flat extends to the east of Neye Island and drops off slowly to a depth of 12 meters. The slope along the southern and western sides of Neye Island is steeper and slopes rather quickly to a depth of 18 meters. The bottom around Neye Island is similar to Tanguisson with numerous rocks, mounds, and gullies and rather sparse coral cover. This location supports populations of both L. laevigata and Linckia sp.

Anae Island is located in the southern part of Agat Bay, and is situated 1,000 m off shore. The shoreward side of the island displays an extensive, shallow reef flat approximately 400 m long and 200 m wide which supports a large population of small L. laevigata on the reef

flat, as well as a few larger specimens in deeper water (Table 1). The areas surrounding the island slope off rather gently (except for the seaward slope) and are characterized by high relief and numerous mounds. Coral cover here is much more extensive than at stations 1-4.

In Cocos Lagoon, two areas were surveyed. One was located directly off the village of Merizo, approximately 200 m southeast of the entrance to Mamaon Channel. This area is a narrow (50 m) reef flat which slopes into the main channel. Visibility is low and Acropora is the dominant coral. Only a few L. laevigata were found in this area (Table 1). The other location was inside the lagoon proper on one of the patch reefs located in 4-6 meters of water. These patch reefs are almost entirely Acropora formosa surrounded by a sandy bottom. Only a few L. laevigata were seen in this area.

Pago Bay, situated on the eastern side of Guam, is subject to heavy surf action most of the year. There is a reef flat about 100 m wide, a narrow first submarine terrace and a fairly steep slope down to a second submarine terrace at about 25 meters deep. The coral cover is rich and diverse with the buttress system well developed. Four surveys (as well as numerous pleasure dives) failed to reveal a single specimen of Linckia.

The results of the surveys made at the eight stations are summarized in Table 1. This table shows the number and percentage of each starfish at each location, its density per hour of search (one diver), and the mean depth and size of each population. The calculated densities are most likely conservative estimates, for not all the animals could be located in the area surveyed. Being cryptic, some starfish, especially Linckia sp., may be totally or partially

hidden from sight within a coral head, in holes or under ledges. An effort was made to search such areas, and starfish were counted even if not retrievable for measurement.

Figure 3 shows the starfish composition at each depth class, based on a total of 547 starfish counted. The number of starfish at each depth is given by the number within each bar. Figure 4 shows how starfish are distributed among the five depth classes. From the above two figures it is clear that Linckia laevigata occurs mostly in the shallow areas, especially the reef flat, whereas Linckia sp. is not found on the reef flat, preferring deeper water. There is considerable overlap of the two forms in the 3.3-6.2 meter and 6.3-9.2 meter zone. This cohabitation was most evident at Neye Island where similar size animals were found well mixed between six and 12 meters of water. However, in water shallower than six meters and deeper than nine meters, the two Linckia seem to be well zoned. No Linckia were located at depths greater than 21 meters.

The results of the comparison of wet weight to underwater weight and dry weight to underwater weight for the two starfish are shown in Figure 5. Corresponding regression line formulas and high correlation coefficients indicate a good fit for the data. There was no significant difference ($p \leq 0.05$) between L. laevigata and Linckia sp. in either relationship. Underwater weight of Linckia was approximately one-half (0.480) the dry weight and one-sixth (0.162) the wet weight.

Since the two forms of Linckia being considered appear to be very similar morphologically but well zoned in their habitats, it was decided to look at some of their physical characteristics. An attempt was made to establish a relationship between some easily measurable

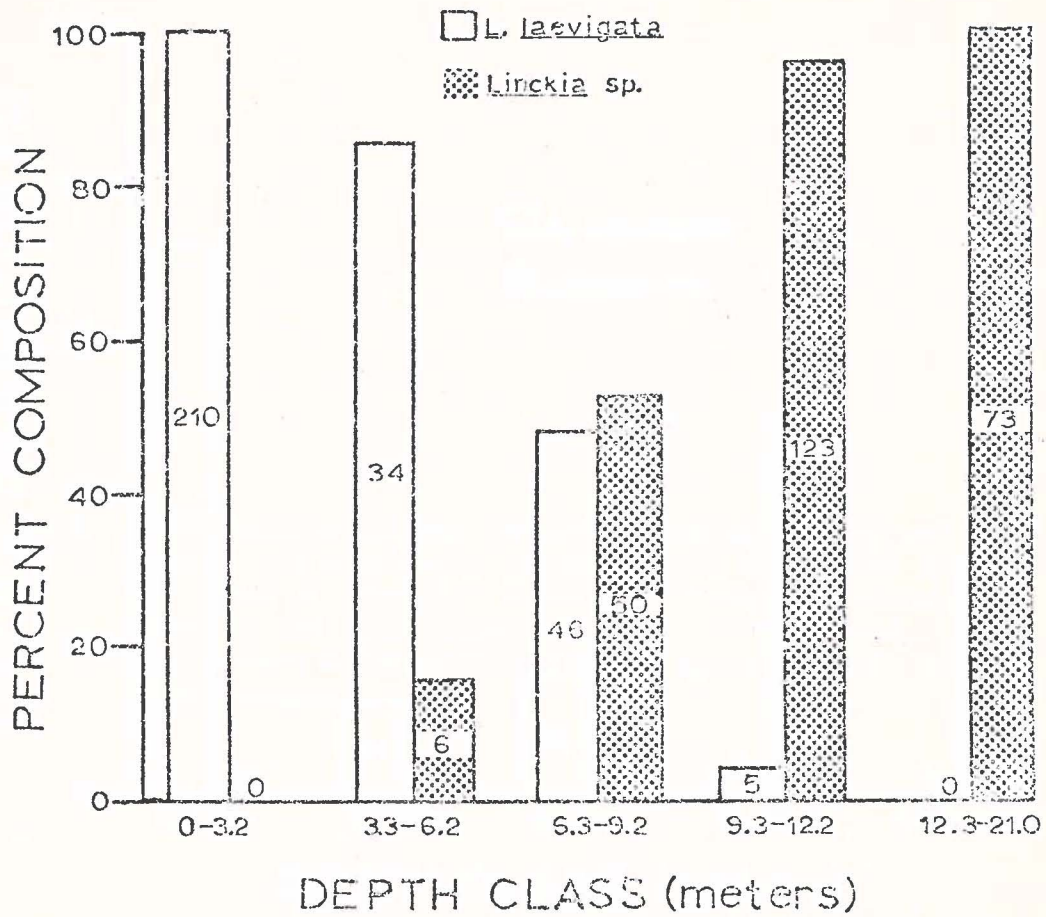


Figure 3. Percent composition of two *Linckia* forms in various depth classes. Numbers inside the bars represent the actual number of individuals counted.

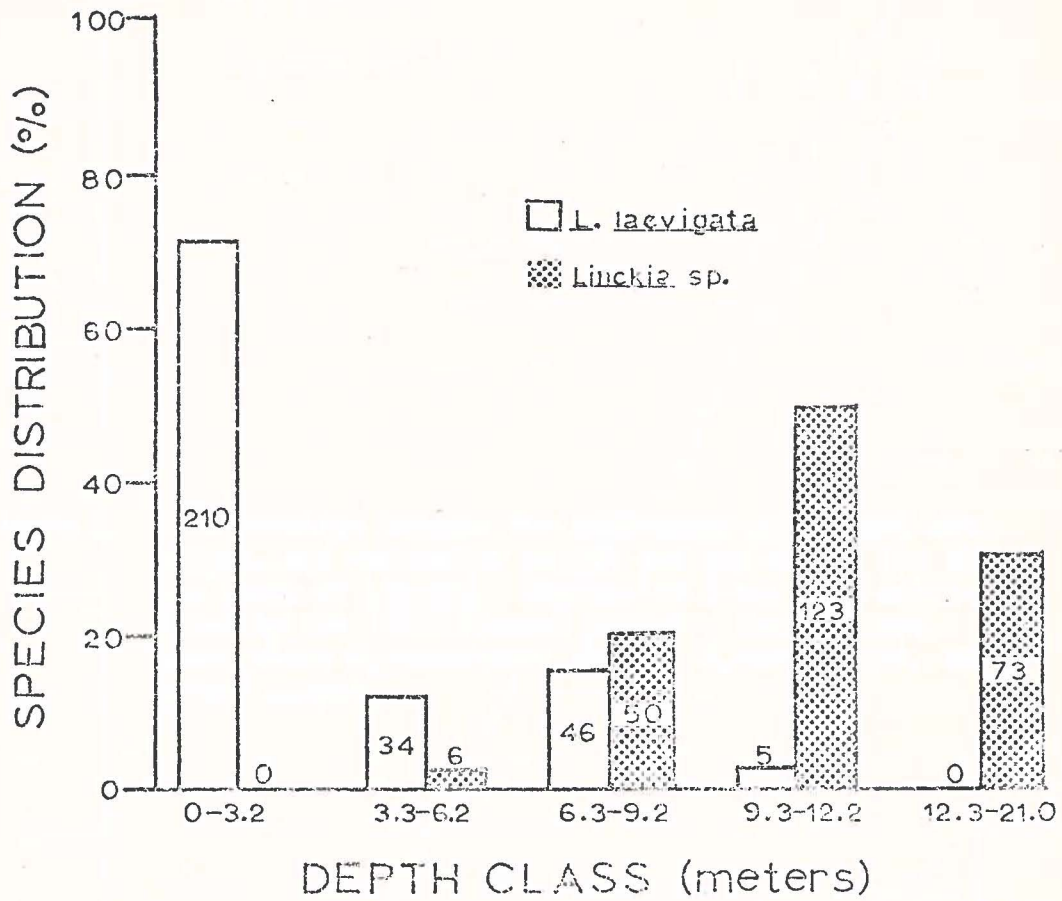
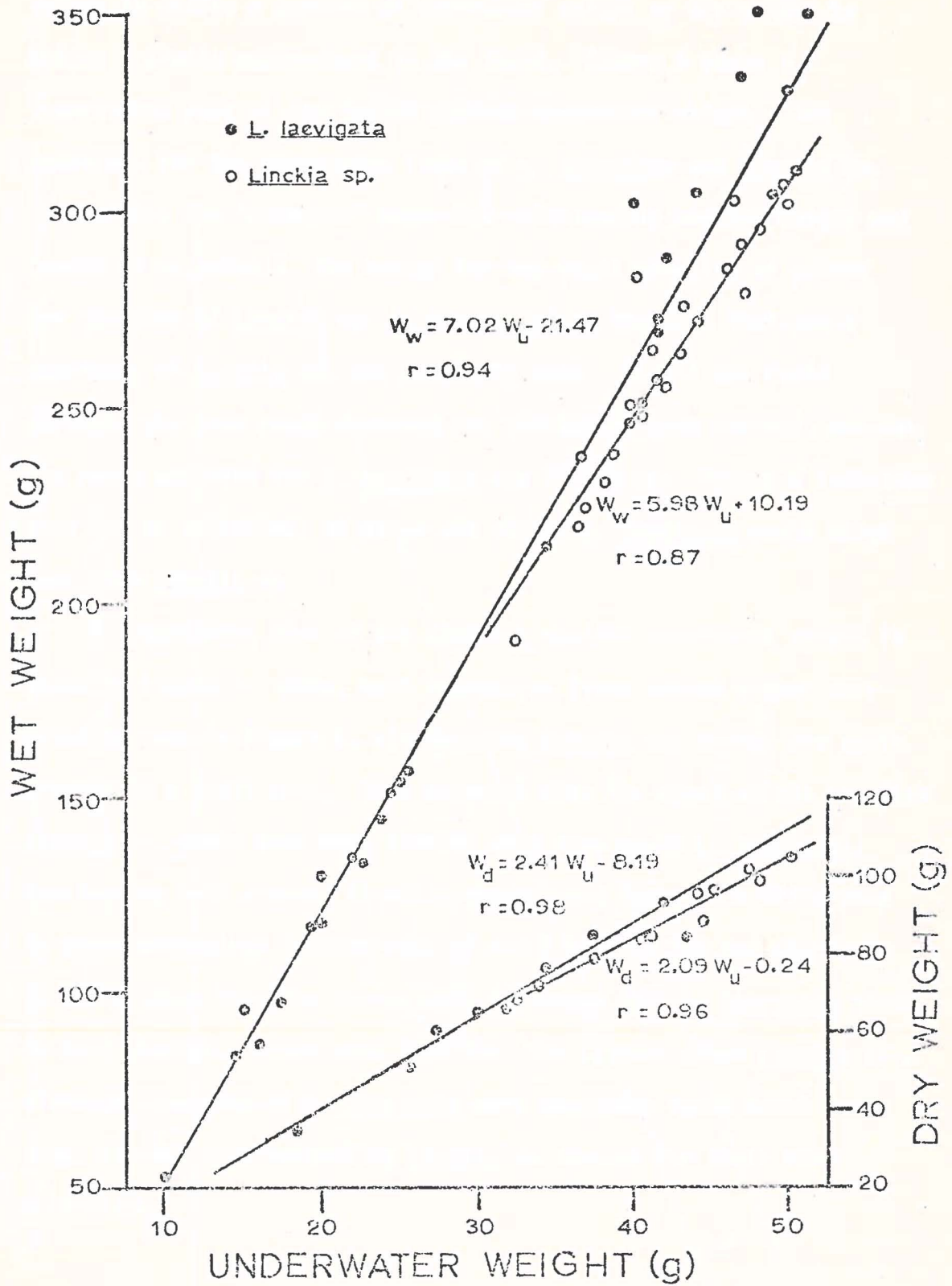


Figure 4. Species distribution of two *Linckia* forms in various depth classes. Numbers inside the bars represent the actual number of individuals counted.

Figure 5. Relationships between the underwater weight and wet weight, and underwater weight and dry weight for the two forms of Linckia. Regression lines were calculated by the least-squares method. Formulas are shown where W_U =underwater weight, W_W =wet weight, and W_D =dry weight.



parameter and body weight for Linckia. This would also enable a person to obtain a measure of underwater weight or dry weight by making a simple measurement in the field. Figure 6 shows a logarithmic plot of arm length against underwater weight. The equations for the regression lines of L. laevigata and Linckia sp. fall within the "expected" power-law relationship between weight and length of an animal. The reason for the small grouping of points for the plot of Linckia sp. is attributed to the fact that small specimens of Linckia sp. are extremely rare. In all the field surveys, the size range observed was 150-230 mm (arm radius), whereas the range observed for L. laevigata was 70-200 mm. Figure 6 indicates that for an individual of given arm radius L. laevigata would weigh more than Linckia sp.

A logarithmic plot of arm diameter against underwater weight is shown in Figure 7. Here, both regression lines showed higher correlation than in Figure 6, although the Linckia sp. points are again grouped in a small area. This suggests that for equal weight starfish Linckia sp. would have more slender arms than would L. laevigata. Considering both graphs, it would appear that there is a difference in gross morphology (slenderness of arms) between the two starfish. It was evident from the survey work performed that L. laevigata tends to have shorter, fatter arms than does the slimmer-armed Linckia sp., although specimens of L. laevigata were sometimes found which had long, slender arms resembling Linckia sp. except for their blue coloration.

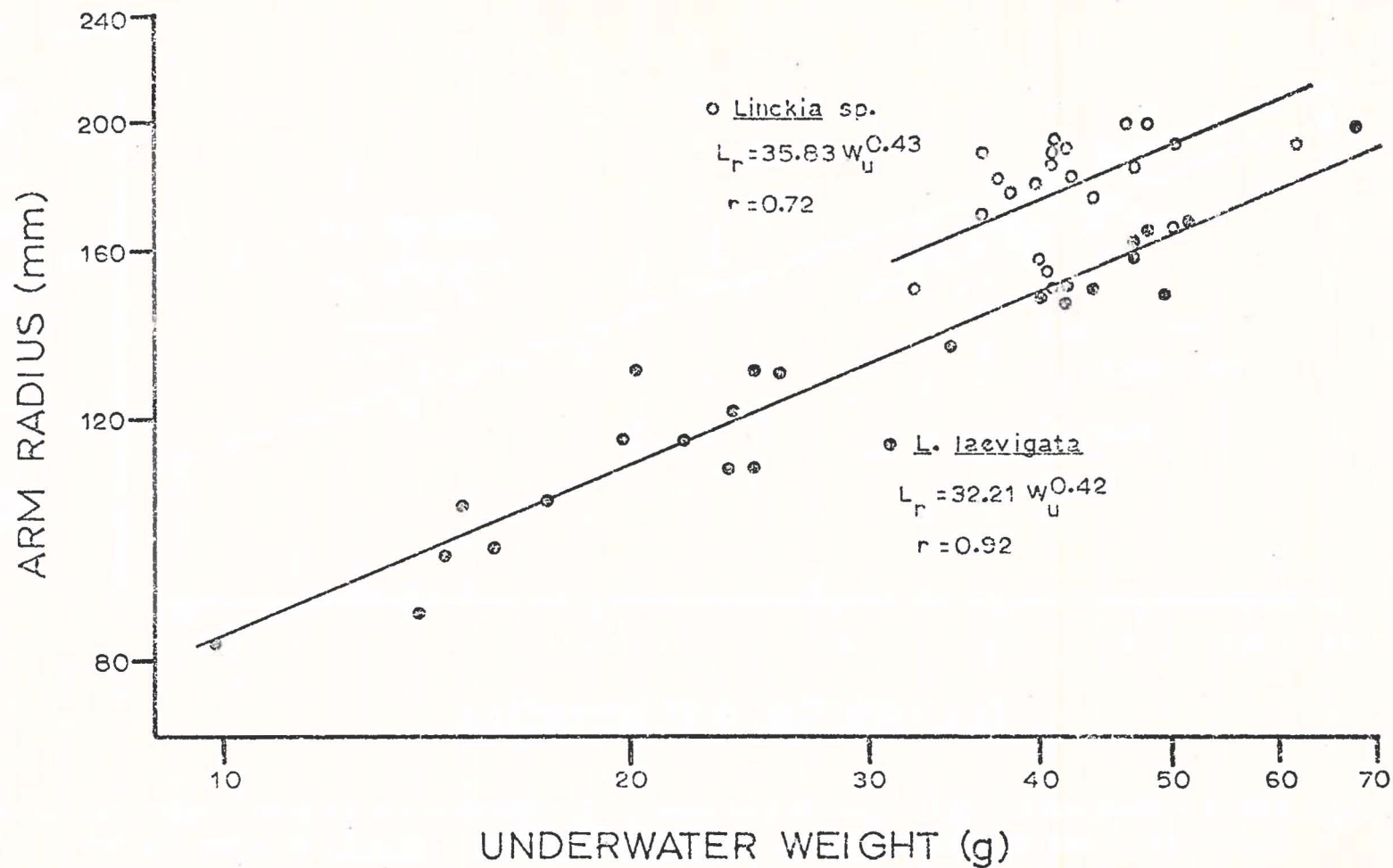


Figure 6. Logarithmic plot of the relationship between arm radius (L_r) and underwater weight (W_u) for both forms of *Linckia*. The regression lines were determined by the least-squares method.

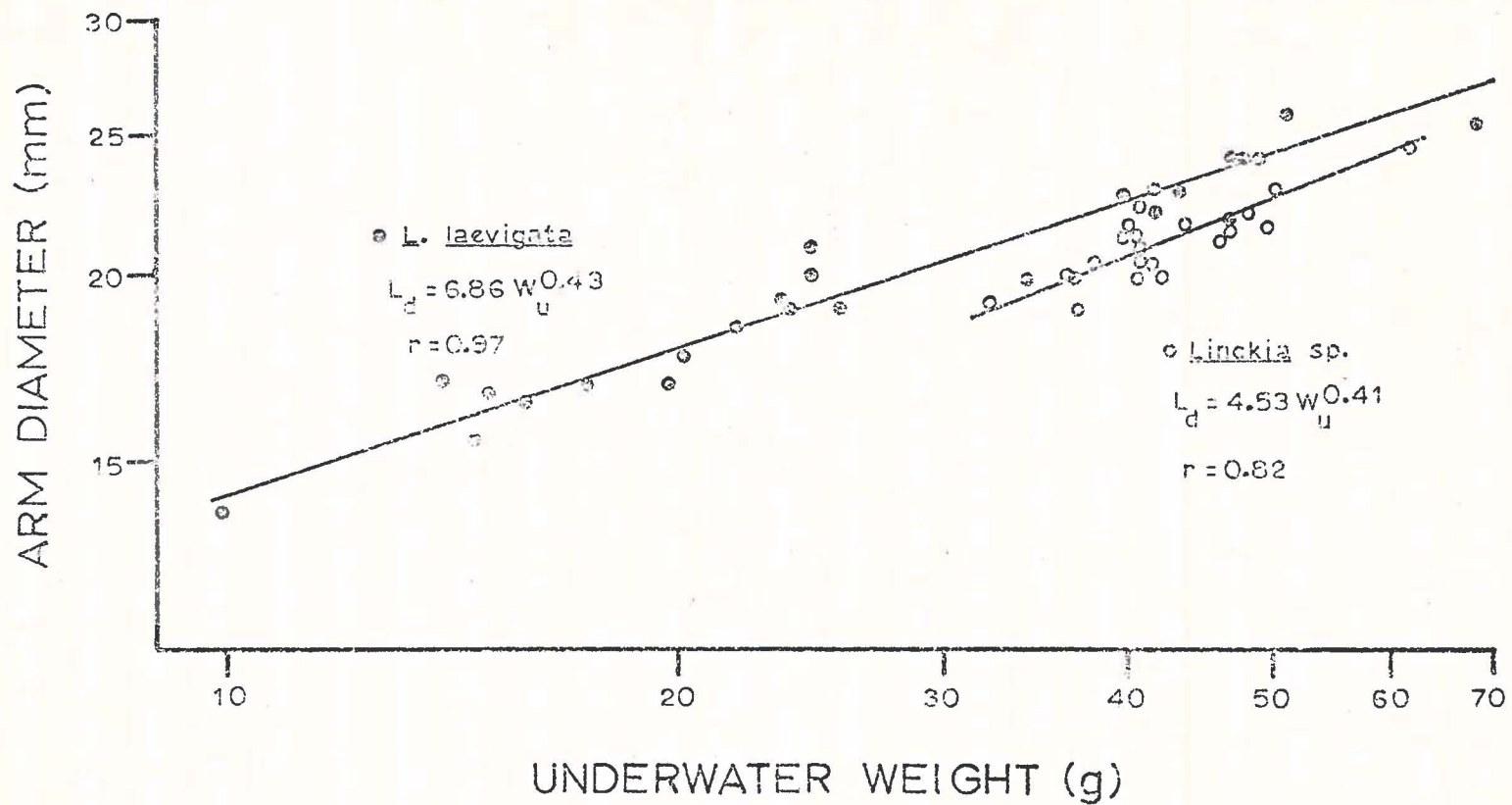


Figure 7. Logarithmic plot of the relationship between arm diameter (L_d) and underwater weight (W_u) for two forms of *Linckia*. The regression lines were determined by the least-squares method.

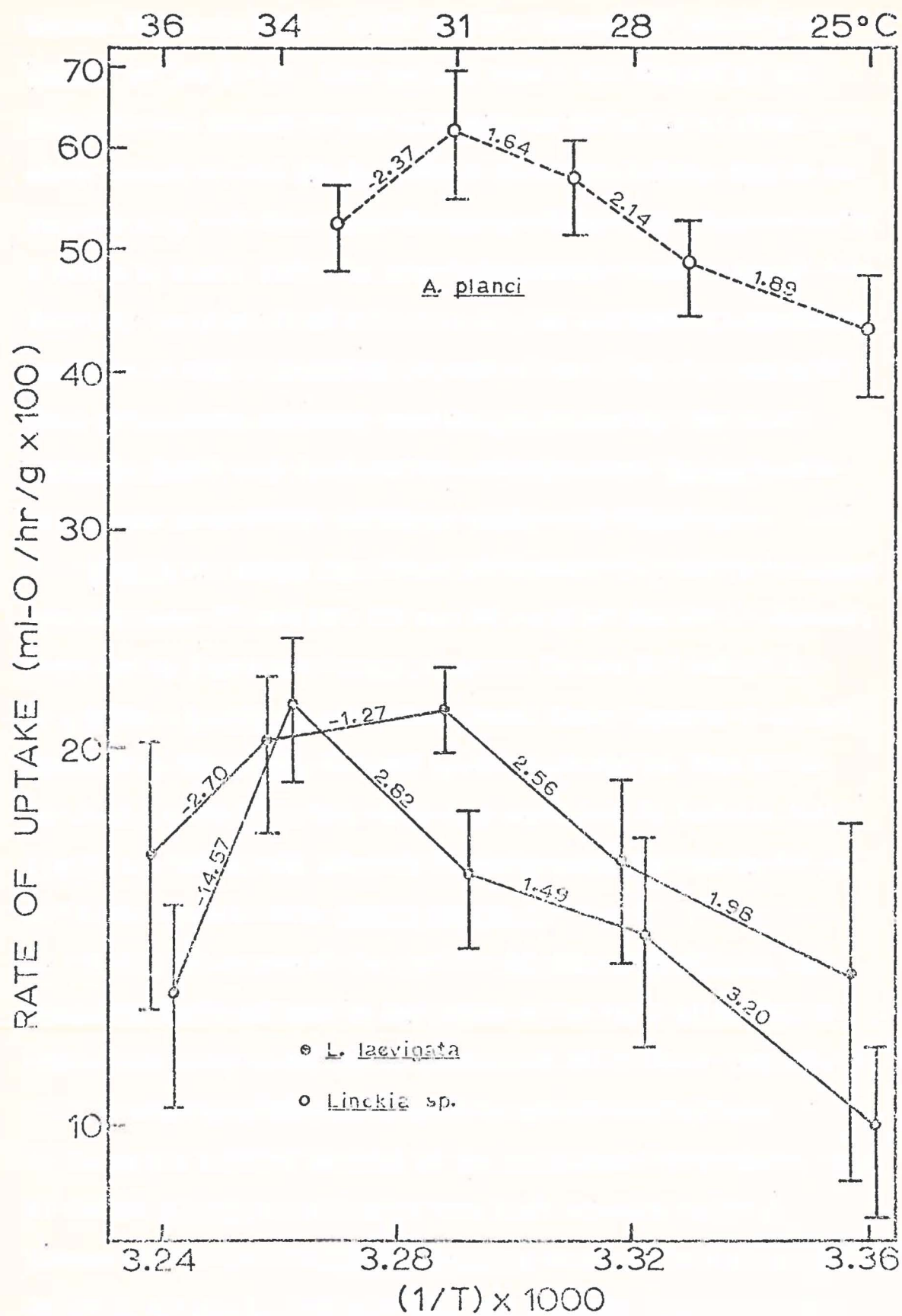
B. Laboratory

Results of the thermal experiments are shown by the Arrhenius plot (after Farrell and Rose, 1967), using the log of rate of oxygen uptake against the inverse of the absolute temperature (Figure 8).

The means were calculated from the consumption rates of six starfish, with each starfish being used for three, 60-minute runs at each temperature. The bars represent the 95 per cent (two standard deviations) confidence limits of each mean uptake. For comparison, the plot of Acanthaster planci is included (after Yamaguchi, 1974). The Q_{10} values are shown for each slope. For 34° and 36° C, metabolic rates were also measured on starfish that were not adjusted to the experimental temperature for 12 hours. Results of the oxygen uptake by both forms of starfish yielded significantly higher ($p \leq 0.001$, t-test) rates at both 34° and 36° C. These results were similar to those obtained by Farmanfarmaian (1966). He concluded that when the temperature is abruptly changed, most poikilotherms show an initial overshoot (shock reaction) that lasts for seconds or minutes, but there then follows a stabilized rate which usually lasts for several hours. Similar findings were also reported on the littoral sea urchin Eucidaris (McPherson, 1968). McPherson found that urchins placed in higher or lower temperature tanks resulted in a marked increase in oxygen consumption during the first 20-30 minutes, followed by a decrease and stabilization after about 40 minutes. For this reason he used a one hour acclimation period in his experiment, as did Ulbricht and Pritchard (1972).

Analysis of the rate of oxygen uptake against temperature for Linckia laevigata, revealed no significant difference ($p \leq 0.05$)

Figure 8. Arrhenius plot of the relationship between the rate of oxygen uptake and the inverse of the absolute temperature for the two forms of Linckia. For comparison, values are shown for Acanthaster planci (after Yamaguchi, 1974). Vertical bars indicate the 95% confidence limits (two standard deviations). The Q_{10} values are shown for each line.



between the oxygen uptake at 25° and 28°, between 31° and 34°, or between 34° and 36° C. This tends to show a leveling off of the metabolic rate between 31° and 34°, with an indication of stress somewhere between 34° and 36° C, although not a very intense drop in consumption ($Q_{10} = -2.70$). This flattening of the curve was observed in a review by Newell (1970) in several intertidal animals. Newell felt that this leveling off of the rate indicated an important homeostatic mechanism in a poikilotherm which would allow relatively constant rates of metabolic reactions despite fluctuations in temperature. McPherson (1968) also concluded that the sea urchin Eucidaris maintained some control over its oxygen uptake with temperature.

Linckia sp. showed significant differences ($p \leq 0.001$) in oxygen uptake between 25° and 28°, 31° and 34°, and 34° and 36° C. However, there was no significant change in uptake between 28° and 31° C ($p \leq 0.05$). Linckia sp. showed an almost linear increase in oxygen uptake from 25° to 34°, followed by an abrupt decline from 34° to 36° C ($Q_{10} = -14.57$). This sharp drop would tend to indicate that Linckia sp. is severely stressed thermally somewhere between 34° and 36° C, much more so than Linckia laevigata.

During the course of the six-hour determination, Linckia sp. frequently would collapse on the bottom of the respiration chamber rather than cling to the top or sides as it did at lower temperatures.

When the starfish were returned to ambient holding tanks following the total of 18 hours at the experimental temperature, none of the six Linckia sp. recovered from their exposure to 36° C, whereas four of the six Linckia laevigata recovered from the exposure. At 34° C, all six L. laevigata recovered from the 18-hour exposure,

but two of the Linckia sp. died. These observations would tend to support the belief that an intertidal animal such as L. laevigata is better adapted to survive the fluctuating environmental conditions of the reef flat than an animal which inhabits the deeper water.

Attempts to determine the thermal death point resulted in highly variable results. Death was considered when the starfish was no longer able to right itself when inverted in the testing aquarium. The critical temperature would seem to be somewhere between 34° and 36° C based on the metabolic rate temperature curves. At 36° C, L. laevigata survived for a minimum of 12 hours up to a maximum of 30 hours, with a mean survival time of approximately 22 hours. The other starfish, Linckia sp., had a survival time that varied from one hour to a maximum of 20 hours, with a mean of approximately eight hours. Survival of Linckia sp. was even more variable than L. laevigata, but the trend indicates that L. laevigata could survive the elevated temperatures of the reef flat for longer periods of time than could Linckia sp.

Rupp (1973) tested the effects of temperature on the fertilization success and early cleavage of several tropical echinoderms. He found that fertilization success in Acanthaster planci was reduced at 34.0° C, but that it remained unaffected in L. laevigata, while both species were blocked almost equally at 36° C. These results suggested an upper thermal tolerance limit of 31° to 34° C for A. planci, and 34° to 36° C for L. laevigata. The range of 34°-36° C for the upper limit for fertilization of L. laevigata is in good agreement with the results of this study, and the range of 31°-34° C for A. planci agrees well with the critical temperature

of 31°-33° C found by Yamaguchi (1974). It would be interesting to determine the effect on fertilization and early cleavage for Linckia sp. to see whether or not they are affected by temperatures lower than 36° C. It has been reported that L. laevigata shows continuous spawning throughout the year near the Equator (Pearse, 1968). Therefore it is likely that the gametes and fertilized eggs would be subjected to the elevated temperatures of the reef flat for at least short periods of time, if spawning occurred during the day. Perhaps fewer larvae of Linckia sp. which find their way onto the reef flats are able to survive.

It has been reported by several workers (Farmanfarmaian, 1966; Newell, 1970) that rate of oxygen uptake is directly related to the amount of dissolved oxygen in asteroids and it would seem that Linckia would react similarly. Since the solubility of oxygen in seawater decreases with increasing water temperature, the amount of dissolved oxygen available to Linckia was less at the higher experimental temperatures. Comparing the amounts of dissolved oxygen at the various temperatures to the theoretical concentration at full saturation (Green, 1965), the saturation level varied from 83 per cent at 36° C to 92 per cent at 25° C with the animal in the chamber. When comparing oxygen uptake and concentration of oxygen (oxygen tension) with three species of the urchin Strongylocentrotus, Johansen and Vadas (1967) found steady levels of oxygen consumption until the pressure of oxygen dropped below 60-70 mm Hg (less than 50 per cent saturation). Because of the small amount of oxygen consumed by Linckia during a 60-minute run, it is unlikely that the concentration of dissolved oxygen influenced results significantly.

Possibly if the 100 per cent saturation level of oxygen at 36° C had been maintained for all other experimental temperatures, the Q_{10} values (slope) might have been greater than those observed.

CONCLUSIONS

Linckia laevigata is a common inhabitant of the reef flat zone, whereas Linckia sp. is rarely found in this area. There exists considerable overlapping of the two forms between the depths of three and nine meters. Linckia laevigata is rarely found in depths greater than nine meters. One reason for this zonation of forms might be the degree to which these asteroids respond to elevated temperatures.

Morphologically slight but consistent differences exist between the two forms. Linckia laevigata tends to have shorter but fatter arms than does the more slender-rayed Linckia sp.

Within a geographic region, such as around Guam, the two starfish can be distinguished by color. Linckia laevigata is rich cobalt blue whereas Linckia sp. is brown in color. However, Marsh (1974) recorded one brown L. laevigata from Tahiti. Three small brown L. laevigata were also found in Fiji (Marsh, personal communication). Occasionally, L. laevigata has an orange oral surface on an otherwise blue body, as reported from the Philippines (Domantay and Roxas, 1933) and Palau (Hayashi, 1938). The author has collected two L. laevigata from Guam which had orange actinal surfaces.

Thermal work has indicated that Linckia laevigata might exhibit a limited degree of control over its metabolic activity at 31° to 34° C. Linckia laevigata is affected less severely by 36° C temperatures than is Linckia sp. Preliminary data suggest that L. laevigata can better survive short exposures of high temperature water such as

that encountered at low tides during the day than can Linckia sp. This agrees well with the habitat zonation shown in this paper.

Rupp (1973), showed fertilization of eggs in Linckia laevigata to be unaffected up to 34° C, and concluded that this asteroid is better suited to an intertidal existence than Acanthaster planci. When compared to A. planci, studied by Yamaguchi (1974), both Linckia seem to have a greater thermal resistance and higher critical temperature (about 3° C higher).

The problem of what to call Linckia sp. still remains. Taken together with the differences in metabolic activity, morphometry, habitat (zonation) and coloration, it is clear that a phenotypic difference exists between L. laevigata and Linckia sp. Although this is a difference at a species level in the classical sense of taxonomy, more work needs to be done to demonstrate whether Linckia sp. is a distinct species or an eco-form of L. laevigata. Results of cross fertilization between L. laevigata and Linckia sp. resulted in 100 per cent fertilization and normal development to the advanced bipinnaria stage (Yamaguchi, personal communication). It would be interesting to investigate further the phylogenetic relationships between the two forms of Linckia.

LITERATURE CITED

- Cary, L. R. 1931. Studies on the coral reefs of Tutuila, American Samoa with special reference to the Alcyonaria. Carnegie Inst. Washington. No. 413:53-98
- Domantay, J. S., and H. A. Roxas. 1938. The littoral Asteroidea of Puerto Galera Bay and adjacent waters. Philipp. J. Sci. 65:203-237.
- Edmondson, C. H. 1928. The ecology of an Hawaiian coral reef. B. P. Bishop Mus. Bull. 45:1-64.
- _____. 1946. Behavior of coral planulae under altered saline and thermal conditions. Occasional Paper B. P. Bishop Mus. 18:283-304.
- Farmanfarmaian, A. 1966. The respiratory physiology of echinoderms. In R. A. Boolootian (Ed.), Physiology of Echinodermata. Interscience, New York. pp. 245-265.
- Farmanfarmaian, A., and A. C. Giese. 1963. Thermal tolerance and acclimation in the western purple sea urchin Strongylocentrotus purpuratus. Physiol. Zool. 36:237-243.
- Farrell, J., and A. H. Rose. 1967. Temperature effects on microorganisms. In A. H. Rose (Ed.), Thermobiology. Academic Press, New York. pp. 147-218.
- Green, E. J. 1965. Solubility of oxygen in sea water. In R. A. Horne, Marine Chemistry. 1969. John Wiley and Sons, Interscience, New York. p. 198.
- Hayashi, R. 1938. Sea-stars of the Caroline Islands. Palao Trop. Biol. Station Studies No. 3. pp. 418-449.
- Johansen, K., and R. L. Vadas. 1967. Oxygen uptake and responses to respiratory stress in sea urchins. Biol. Bull. 132:16-22.
- Jones, R. S., and R. H. Randall. 1973. A study of biological impact caused by natural and man-induced changes on a tropical reef. Interim report submitted to Office of Research and Monitoring, U. S. Environmental Protection Agency, Washington, D. C. 184 p.
- Marsh, L. M. 1974. Shallow-water Asterozoans of southeastern Polynesia. I. Asteroidea. Micronesica 10(1):65-104.

- Mayer, A. G. 1914. The effects of temperature upon tropical marine animals. *Publs. Carnegie Inst. Wash.* No. 183:3-21.
- McPherson, B. F. 1968. Feeding and oxygen uptake of the tropical sea urchin *Eucidaris tribuloides* (Lamarck). *Biol. Bull.* 135:308-321.
- Newell, R. C. 1970. *Biology of Intertidal Animals*. American Elsevier Publishing Co., Inc. New York. 555 pp.
- Pearse, J. S. 1968. Patterns of reproductive periodicities in four species of Indo-Pacific echinoderms. *Proc. Indian Acad. Sci.*, B6:247-279.
- Randall, R. H. 1973. Distribution of corals after *Acanthaster planci* (L.) infestation at Tanguisson Point, Guam. *Micronesica* 9(2):211-222.
- Rupp, J. H. 1973. Effects of temperature on fertilization and early cleavage of some tropical echinoderms, with emphasis on *Echinometra mathaei*. *Mar. Biol.* 23:183-189.
- Strickland, J. D.H., and T. R. Parsons. 1968. *A Practical Handbook of Seawater Analysis*. Bull. No. 167 of the Fisheries Research Board of Canada. Ottawa. 311 pp.
- Ulbricht, R. J., and A. W. Pritchard. 1972. Effect of temperature on the metabolic rate of sea urchins. *Biol. Bull.* 142:178-185.
- Vernberg, F. J., and W. B. Vernberg. 1970. *The Animal and the Environment*. Holt, Rinehart & Winston, Inc. New York. 398 pp.
- Yamaguchi, M. 1974. Effect of elevated temperature on the metabolic activity of the coral reef asteroid, *Acanthaster planci* (L.) *Pac. Sci.* 28:139-146.
- _____. In Press. Coral reef asteroids of Guam. *Biotropica*.