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FLOW-THROUGH MICROCOSMS FOR SIMULATION OF MARINE ECOSYSTEMS:

DEVELOPMENT AND INTERCOMPARISON OF OPEN COAST AND BAY FACILITIES

by

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October 1976

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ADMINISTRATIVE STATEMENT

Funds for the 1973–74 design and construction phases of the Ulupau microcosm facility were provided by the Naval Facilities Engineering Command, Code 032B, under Project Order PO-4-0016(PM-03), and by the Environmental Protection Agency (EPA) under Project R800906. The final construction, instrumentation, and calibration of the facility (late 1974 through mid-1975) were funded primarily by the Office of Naval Research, Code 443, on Contract N0001475-WR50121, and by the Environmental Protection Agency under Project R800906. The EPA-supported programs were conducted by investigators at the Hawaii Institute of Marine Biology (HIMB) of the University of Hawaii. Input by HIMB–EPA to the development of the Ulupau facility consisted largely of technical advice, labor (related primarily to construction of the seawater system), sample processing, and data analysis (related to the intercomparison study).

Navy-derived funds were assigned to the Marine Environmental Management Office (MEMO), Undersea Sciences Department, Naval Undersea Center (NUC), Hawaii Laboratory. Personnel from that office and from other departments of NUC were responsible for the construction, planning, and coordination, and also provided most of the construction labor. This report was reviewed for technical accuracy by Dr. John E. Bardach and Paul L. Jokiel of HIMB and by Gerald S. Key of Computer Sciences Corporation.

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A low-nutrient flow-through seawater facility is constructed on an open coast of Oahu, Hawaii. The facility complements a system of similar design existing in the high-nutrient environs of Kaneohe Bay. Present at each site are 12 tanks, each having a capacity of 160 gallons (600 liters) and each of which can be adjusted for flow-through rates of 0.1 to 15.0 lpm. Biotic communities of varying complexity can be established by the natural recruitment or transplantation of organisms. Results are presented for a calibration test conducted from March through June 1975 to determine interfacility differences in source water chemistry, organism recruitment, and growth of biota in the tanks.

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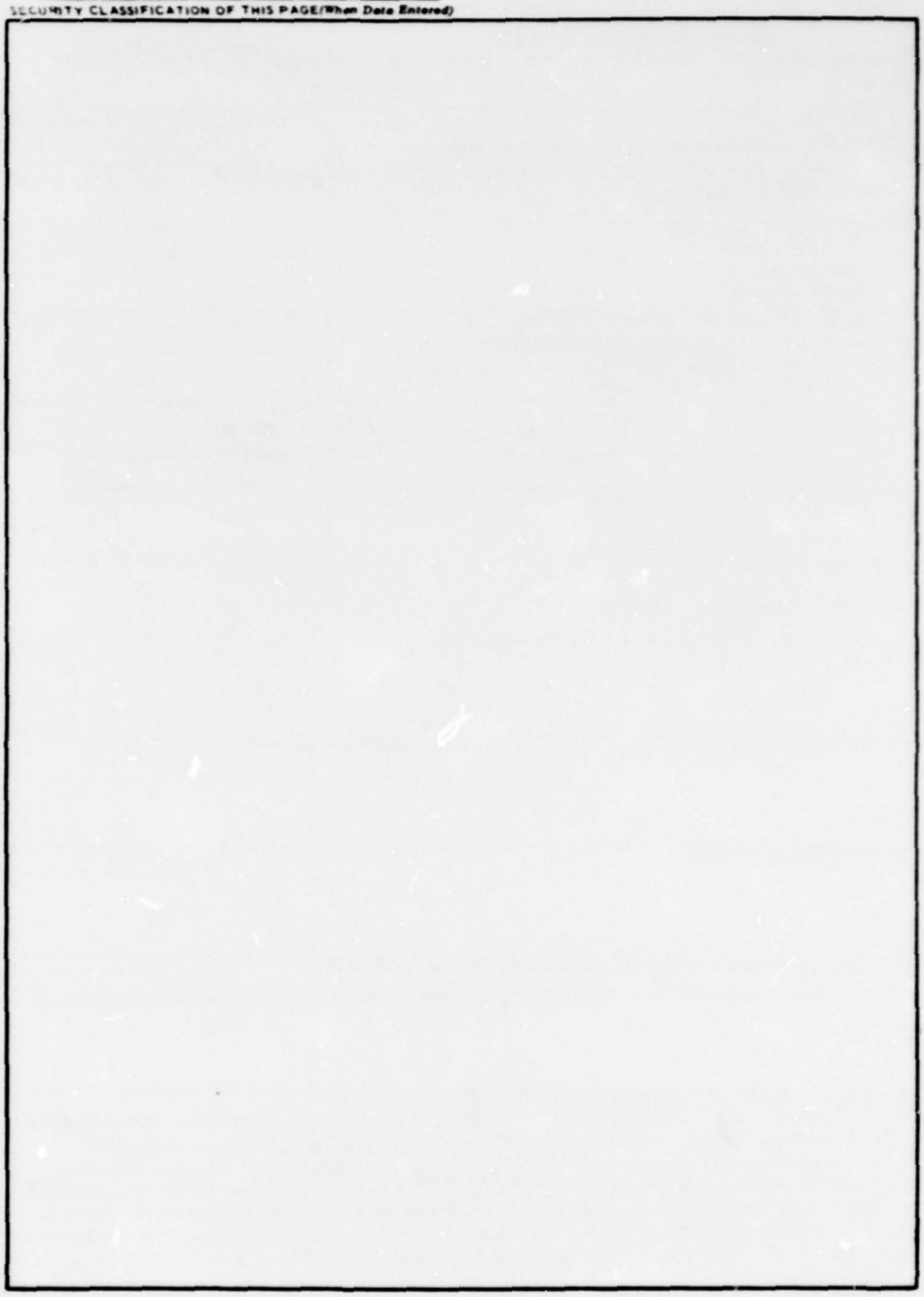
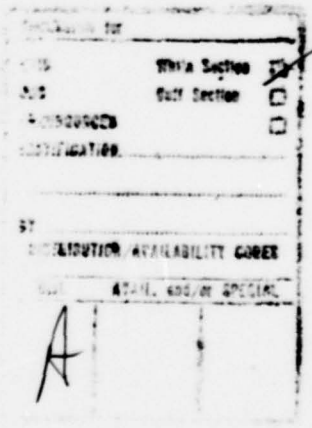


SUMMARY

A low-nutrient flow-through seawater facility was constructed on the seaward side of Mokapu Peninsula (Ulupau Head), Oahu, Hawaii. This facility complements a system of similar design existing in the nearby high-nutrient environs of Kaneohe Bay. The double-plumbing innovation introduced at the newer facility has been very successful in reducing antifouling maintenance. Twelve outdoor tanks, each having a capacity of 160 gallons (600 liters) and identical dimensions, are present at each site. Flow rates are adjustable from 0.1 to 15.0 lpm.

A calibration test was performed to determine interfacility differences in source water chemistry, organism recruitment, and growth of biota in the tanks. At both locales simple microcosms consisting of sorted coral rubble, sand, and varying numbers of herbivorous fish, *Acanthurus triostegus*, were subjected to 6 months of flow-through at 10 lpm. The bay source water generated a diverse, near-climax community within 60 days. At the oceanic facility succession was much slower and community productivity was significantly lower than in the bay microcosms. *A. triostegus* at Ulupau invariably lost weight and died; whereas, at the bay site they experienced slow increases in weight.

The principal difference in source water macronutrients between the two facilities was the PO_4 content, the bay values being about double those of the oceanic source. NH_3 and NO_3 contents were nearly the same, but showed large fluctuations which correlated with tide and surf conditions.



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Frontispiece: View from Ulupau Head. The Ulupau Microcosm Facility is seen slightly right of lower center. North Beach and western Mokapu Peninsula (Kaneohe Marine Corps Air Station) extend across mid-photo. At the foot of the distant Koolau Mountains is central Kaneohe Bay. The HIMB Microcosm Facility is located on Coconut Island (Moku o Loe Is.) at upper left.

INTRODUCTION

Congress, through the National Environmental Policy Act of 1970, directed the U.S. Navy to develop sensitive means of determining and predicting the effects of its operations on the environment, including marine communities. Early in 1971, the Assistant Secretary of the Navy (R&D) established the Navy Environmental Protection Program. In April of 1971, the Deputy Chief of Naval Material (Development) assigned primary responsibility for RDT&E in marine biology to the Naval Undersea Center. To meet this responsibility, the Center formed the Marine Environmental Management Office (MEMO) to utilize the techniques of applied biology to improve the Navy's capability for effective marine environmental management.

Because the Navy operates extensively in a number of U.S. and foreign ports, the principal thrust of the effort has been directed toward obtaining a better understanding of the condition and dynamics of these important ecosystems. This concentration of effort is based upon several considerations. First, harbors are usually situated in estuarine environments, and estuaries are among the most biologically productive areas in the ocean. Many estuarine harbors support, directly or indirectly, extensive and commercially important fisheries. Second, the majority of estuarine systems are already under severe, but poorly understood, environmental stress. Prompt and accurate corrective action is required if any of these systems are to be restored to their natural and highly productive state. Third, if appropriate assessment and management techniques can be developed for estuaries, these same procedures can certainly be adapted for effective application elsewhere in the ocean. Fourth, the Navy is a readily visible user of ports where general public interest in environmental issues tends to be high, yet the proportion of environmental stress due to Navy operations, as opposed to other activities, is entirely unknown. Fifth, since the Navy must operate under a wide variety of environmental laws and public attitudes, some unifying principle is urgently needed. An intercomparison of ports, each with an appropriate reference area, offers the best and possibly the only chance of developing such principles both for impact assessment and subsequent protective management.*

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*The findings of the Pearl Harbor Biological Survey (Evans, 1974, pp. 5.0–29ff) strongly suggest that the intercomparison of marine communities from very different locations is best done by first noting similarities between heavily stressed communities at each location and next noting local community response as the stresses are relaxed. The concept is the reverse of the usual notion of biological control.

In order to obtain a better quantitative understanding of the environmental condition and natural dynamics of ports, the Marine Environmental Biology (MEB) Program was developed. It is fully described in the MEB 5-year plan (Evans, 1974). Briefly, this Program represents a three-pronged approach to the problem of effective marine environmental assessment and management, namely: (1) development of accurate short-term survey techniques suitable for determining general marine environmental status, and also suitable for the intercomparison of field results from very different marine communities; (2) development of reliable environmental indicators (physicochemical or biological) in order to increase the speed and reliability of the field survey procedures; and (3) development of sophisticated analytical procedures using current computer technology so that (a) trends or patterns in environmental data are readily discernible, (b) results of different surveys can be quantitatively compared, and (c) a basis can be established for predicting future changes in environmental status or community responses to specific management strategems. Although the MEB Program is only completing its second year, considerable progress has been made along all three of these objectives. A rapid way has been found for dividing a harbor into three zones: a zone highly influenced by shipping and shipyard activities, a transitional zone, and a zone relatively unaffected by ships and shipyards. At least one organism has been found suitable for the intercomparison of environmental stress in such widely different locations as Hawaii, the Southern California Bight, and Puget Sound. Factor analytical and ordination techniques have been combined so that the normal redundancy in biological field data is reduced, yet patterns and trends remain clearly visible. During this past year, the experimental approach to environmental monitoring and management has been greatly augmented through the development and application of microcosms.

The concept of microcosms as experimental tools is certainly not new. The idea first took the form of the chemostat,* developed by Monod (1950) and independently by Novick and Szilard (1950). Experimental uses of microcosms have been expanded by Odum and Hoskins (1957), Odum *et al.* (1963), and many others (McIntire *et al.*, 1961; Abbott, 1966; Abbott, 1967; Hall *et al.*, 1971; Warren and Davis, 1971; Ryther *et al.*, 1972; Cooper and Copeland, 1973; and Mullin and Evans, 1974). In brief, a microcosm is simply a contained and usually simplified marine system** which can be experimentally manipulated in various ways. It is the experimental design which distinguishes it from a simple aquarium. Microcosms can be separated into two basic categories; namely, closed systems, either static or recirculating, and open or flow-through systems. The

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* Chemostat = a continuous-flow growth apparatus in which a population of microorganisms can be maintained always in the growth phase and at a constant level over an indefinite period of time.

** Microcosms other than marine are, of course, possible. Terrestrial and freshwater microcosms do exist. Discussion here is restricted to marine microcosms.

latter may be further subdivided on the basis of whether the supply water is filtered or unfiltered. The microcosms discussed here are the flow-through, unfiltered type, and as such, may be considered as extensions of the natural world they are presumed to mimic.

The addition of microcosms to the program of field surveys enables the attainment of a true and necessary midpoint between the precision of laboratory studies and the realism of *in situ* surveys. Simplified-to-complex communities representative of various parts of the marine ecosystem under survey can be replicated as a series of microcosms. These replicates are maintained under normal diel variations in ambient conditions, subsist on the normal (or near-normal) food webs, are subjected to normal seasonal variations in recruitment, and thus can experience quasi-natural seral development. Furthermore, some of the replicates can be subjected to controlled environmental stresses and their metabolic and structural responses compared with other replicates maintained as controls. This powerful combination of microcosm experimentation and field survey can be utilized to:

1. Quantify functional interrelationships between organisms
2. Determine aggregate response (antagonistic and synergistic) of a community to a given environmental stress or to a combination of stresses
3. Identify those members of a given community exhibiting a usable scalar response to a range of intensities for a given stress, thus expediting the search for bioindicators
4. Develop those aggregate and scalar responses into procedures suitable for field use
5. Check, under controlled conditions, situations observed in the field and thought to represent natural responses to specific stresses
6. Test and quantify (by ganging two or more microcosms) the biological detoxification capacities of given marine communities

Many environmental stresses can be tested in microcosm systems, including: waste heat, freshwater dilution, silt, elevated or depressed nutrient status, biologically induced stresses, heavy metals, oil, detergents, and pesticides. The intensity and variability of stress can be carefully controlled. Lethal and sublethal levels can be effectively used since long-term observation is possible. Because, as mentioned above, a number of microcosm replicates can be ganged (connected in series), both the detoxification capacity of a given community and the combined effect of detoxification and dilution can be determined. Various members of a microcosm community can be used for bioassay as well as bioindicators. Electrochemical probes and automatic recording systems can also be added to the microcosms, as indeed is planned for the system described here. In short, microcosms offer great research flexibility, as well as being powerful experimental

adjuncts to the field survey. The Environmental Protection Agency (EPA) has also recognized the value of the microcosm approach and has installed similar systems at both its east coast and its west coast laboratories. Thus, research performed by the Navy using these systems can easily be duplicated by EPA and thus is more likely to be accepted by that regulatory agency.

This report presents details of the construction of the Navy microcosm facility at Ulupau. The Ulupau facility was modeled after a system developed and built by the Hawaii Institute of Marine Biology (HIMB) as part of an EPA-supported project (Jokiel *et al.*, in press). Much of the basic theory and design of the Navy system is the work of Paul Jokiel, Eric Guinther, and Gerald Key of that project. The following sections present first a brief outline of future uses of the facilities together with a recapitulation of the theory of microcosm application. Next described are the planning and site selection prior to construction, and the design and construction of the Navy facility. Following is the description of an initial interfacility calibration test, conducted because both the Navy and the HIMB installations are to be used as part of a joint Navy–HIMB program. Finally, a conclusions section completes the report.

FUTURE USES AND THEORY OF APPLICATION

Before future uses of the microcosms are discussed and the theory of their application to the MEB Program is explored at greater length, it is appropriate to emphasize again that both the Navy and the HIMB systems are the unfiltered, flow-through type. Such systems have certain disadvantages and advantages:

Disadvantages

1. Nearly continuous pump operation is required; thus there are higher maintenance costs and greater experimental risks due to breakdown. Backup equipment and proper facility design can, of course, significantly reduce these risks.
2. Biological recruitment into the system is largely uncontrolled. Some control can be achieved by adjusting the depth from which supply water is drawn; also filters can be used to exclude undesired plankton.
3. There are constraints on certain variables (for example, salinities higher than ambient are possible but difficult) and also on the size of the microcosm. Although size limits have not been thoroughly investigated, not all components of a given community (large carnivores, for example) can be accommodated.
4. It is also difficult (though possible) to simulate such variables as water motion and tidal variations, as well as truly planktonic conditions.

Advantages

1. Microcosms are extensions of the natural world. Normal diel variations in ambient conditions can be duplicated. Natural foods and recruitment can be provided. Normal seasonal variations can be followed and natural seral development is allowed.
2. Resident fauna can subsist on their normal (or near-normal) diet.
3. Wastes, biogenic toxins, hormones and pheromones can be kept at near-natural concentrations.
4. Microcosms represent an excellent middle stage between controlled laboratory experiments and *in situ* field observations.
5. Microcosms offer great research flexibility. Units can be connected in series; thus progressive detoxification or stripping processes may be studied. Conversely, a single unit may be simultaneously used for a number of nondestructive studies. Microcosms may be used as a biological analogue to statistical sorting by factor analysis for the identification of potential bioindicators. They may also be used for bioassay or for detoxification.
6. Various perturbants can be introduced into a number of replicate microcosms under controlled conditions. Sublethal levels may be used since long-term experiments are quite feasible; such experiments also lend themselves to chain-response studies in complex communities.
7. Electrochemical probes and automatic recording systems can be added to the microcosms, thus facilitating detailed and continuous observation. The link between such additions and automatic data processing is obvious.
8. Because microcosms can be more precisely controlled and monitored than the natural systems they are used to simulate, they are more amenable to the validation of mathematical ecosystem models. Accurate computer models of natural systems are, ultimately, a major objective of the environmental sciences, for only with the high-speed simulation of ecological processes can a proper evaluation be made of the likely long-term effects on the environment of a given management decision.

The advantages just enumerated will be used to expand microcosm research considerably beyond past usage, which has tended to be limited to studies within the microcosm itself. In theory, microcosm experimentation and field surveys are considered as two complementary parts of a larger program to understand the environmental condition and the natural dynamics of harbors and ports. This understanding will be applied in the design and testing of management stratagems which optimize both the practical utilization and the environmental health

of these water bodies. Microcosms are not, therefore, considered only as tools for restricted studies but rather as an integral part of a larger experimental system employing field survey, testing, and evaluation. Through automatic data processing, a further expansion to the formation of computer networks facilitating the intercomparison of harbors is definitely contemplated.

The 6-week interfacility comparison study herein reported has shown some striking differences between the communities establishing themselves at HIMB and at Ulupau. These differences appear principally to be due to differences in available phosphate and in recruitment populations in the supply water. Since both facilities will be used in ongoing studies of the nutrient effects in Kaneohe Bay, further detail is needed on these differences. An experimental series is currently underway in which known amounts of nutrients in the form of ammonia and phosphate are being added to the supply water of three microcosms established during the interfacility comparison study. Types and rates of changes resulting from these additions are being followed. Another planned investigation will involve a detailed study of community responses to copper. Interest in copper arose as a direct result of the analysis of field survey data. Factor analysis of the heavy-metal burdens of sediments from various harbors has indicated that copper content is a good indicator of ships and shipyard activity. The common bay mussel *Mytilus edulis* has also exhibited high body burdens of this metal in areas experiencing heavy usage by shipping. Consequently, it is important to use microcosms not only to search for further suitable bioindicators of the presence of copper, but also to determine the exact community responses to elevated copper ion in the water. Particular attention will be paid to tunicates since their reaction to copper is strongly mediated by sunlight, copper being much more toxic to them in the light (Chesher, 1971). This property could be used as a sort of internal standard on field surveys by noting the conditions of tunicates in shaded versus exposed situations at a given location. Experiments with copper will be followed by a similar series with lead. Field surveys have also shown lead to be associated with stressed conditions in a number of harbors. Lead, however, is more probably associated with general urbanization around a harbor rather than with ships and shipyard activity. Since one of the important sources is tetraethyl lead, both aeolian and fluvial transport can be presumed important entry routes into harbor waters, in contrast to copper, which probably enters directly from the hulls of ships and heat exchangers. Thus, not only differences in geographic distribution (and hence associated pollutants) can be expected for these two metals, but also significant differences in chemical or particulate form.

In addition to a series of experiments centered on specific metallic contaminants, the microcosms will be used to determine the environmental role played by silts in harbors. Field surveys strongly suggest that fine bottom material resuspended by the action of ships' propellers can have beneficial environmental effects (Evans, in preparation; Evans, 1974). Furthermore, the introduction of silts into harbor waters by streams or by harbor mainte-

nance and dredging operations is a common phenomenon, the environmental effects of which are not yet fully understood. The Ulupau microcosms will be modified so that controlled exposures to various amounts of suspended silt will be possible. Experiments will then be conducted to determine the response of microcosms to silt as an inert perturbant, and also to silt as a carrier and exchanger of metals and other pollutants. This work will represent an extension of work already performed by Corps of Engineers investigators (Peddicord *et al.*, 1975). Elutriators modified to operate on the principle of microcosms will be used to determine the exchange rates for various biologically important materials into and out of bottom sediments. In these elutriator experiments, established microcosms will be employed downstream of the elutriator in both a bioassay and a detoxification mode. Another series of experiments will use the microcosms to test and evaluate various bioindicator systems currently being used in field survey work. Such indicator systems include the use of fouling panels, the byssal thread formation rate in mussels, amino acid ratios and condition index in oysters, and growth ring formation in various mollusca and corals. Such questions concerning these bioindicators as limiting or controlling factors, specificity, and purging rates can be most efficiently worked out through the use of microcosms. Findings from these experiments can immediately be applied in ongoing field surveys involving various harbors in Hawaii and on the west coast.

PLANNING AND SITE SELECTION

In early 1973 the basic requirements were formulated for an "oceanic" water source flow-through microcosm facility which would complement and be modeled after the HIMB microcosm system (Henderson and Evans, 1973). These requirements were as follows:

1. A source of low-nutrient, normal-salinity seawater.
2. An unshaded level-graded site of at least 650m^2 at an elevation preferably of less than 15 meters. This area is to be located as close to the seawater source as possible.
3. Access for vehicles, power and water, and reasonable security.
4. An air-conditioned laboratory–office building of approximately 60m^2 This structure will house electronic monitoring apparatus and provide dry lab and office space for two or three scientists.
5. A covered wet-lab space of 20 to 30m^2 with at least one seawater spigot and sink.
6. A 2.4-meter-high deck of 46m^2 to hold twelve 160-gallon (600 liter) microcosm tanks complete with inlet and outlet flow control boxes.

7. An intake, pumping, and distribution system capable of supplying a continuous flow of seawater at a total volume of at least 100 gpm (380 lpm).
8. A 360-gallon (1400 liter) capacity, 1.2-meter-high receiving tank on a 3.7-meter-high support structure. This tank will serve as a reservoir to supply water at a constant gravity head to the rest of the flow-through system.

An elevated deck was chosen as a support structure for the microcosm tanks for a number of reasons. First, it was deemed desirable to locate the microcosm tanks as close as possible to the wet-lab and lab–office space so as to avoid problems caused by excessive lengths of hose and piping which would carry sampled seawater from tanks to monitoring devices. Elevation of the tanks to the eave height (2.4 meters) of the lab buildings would eliminate any potential shading problems caused by structures adjacent to the tanks.

Secondly, all seawater plumbing was to be installed in duplicate to enable the "stagnation" cleaning of pipes. It was decided that such an array of pipes and hoses would be much more serviceable if located at an overhead level with walk-under access. Additionally, much of the under-deck space could be utilized for weatherized experimental gear or for storage space.

In the initial stages of searching for potential areas to locate the facility, the entire perimeter of the island of Oahu (Fig. 1) was examined. Only those sections of shoreline owned by the federal government or controlled by oceanographic research institutions were considered because of cost, logistic support, and security. All sites along the southern shore were excluded because of varying degrees of sewage or runoff contamination (as from Barbers Point, Pearl Harbor, and Honolulu city and harbor). Other potential sites on the northern and western shores also were excluded for various reasons, such as remoteness, extreme rough water conditions, or wide nearshore fringing reefs (which would have required long intake lines to reach "oceanic" water).

Apart from the seaward side of Mokapu Peninsula, the only other area which appeared feasible as a site for the project was the Makai Range pier complex near Makapuu Point (about 24 kilometers southwest of Mokapu). Security and access to deck, office, and lab space were available at nominal cost; however, installation of a 150-meter intake line would have been required to draw water of acceptable quality. The other obvious shortcoming of the pier location was its distance from the Naval Undersea Center, the Hawaii Institute of Marine Biology, and Kaneohe Bay, a water body to be studied jointly by the two laboratories.

When all factors were weighed and considered, Mokapu Peninsula (Kaneohe Marine Corps Air Station) remained an obvious first choice for the new microcosm facility. However, some variations in water quality were expected in the

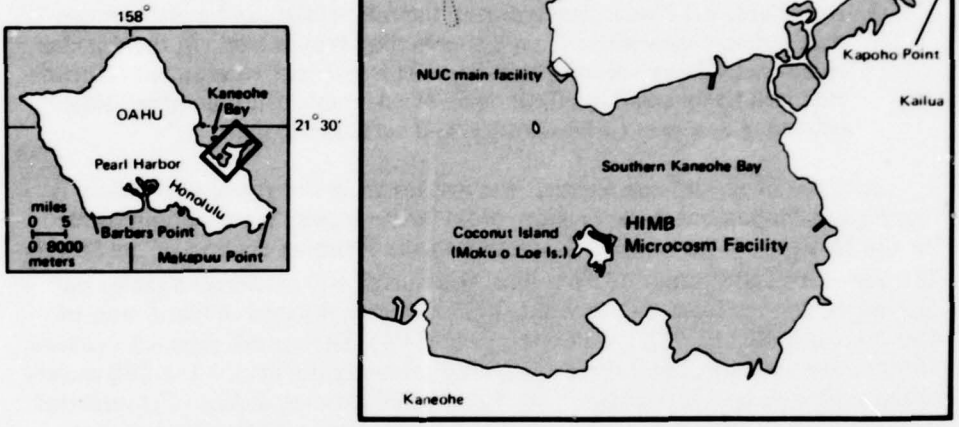

Figure 1. Map of shoreline of Oahu, with enlarged inset showing Kaneohe Bay and Mokapu Peninsula.

waters offshore of Mokapu because of changes in seasonal currents and the discharge from nearby medium-level nutrient sources. Therefore, a study was conducted to gather and evaluate data from past oceanographic studies in the vicinity of Mokapu Peninsula. Some water samples were also collected at locations of special interest. The water samples were analyzed for nitrate–nitrogen, since it had been noted from past studies that nitrate concentrations generally are indicative of levels of other nutrients (such as phosphate, total nitrogen, and nitrite–nitrogen). The summertime analyses for areas offshore of Mokapu showed values ranging from 0.05 to 0.24 μg-atom/l NO_3–N. These values were considerably lower than the year-average figures of 0.40 to 0.60 μg-atom/l obtained in approximately the same locations by the Kailua Bay Monitoring Study (Bathen, 1972). These differences were presumably attributable to the higher winter values included in the annual averages and to differences in analytical techniques. However, all values from waters off the northern Mokapu shore areas were at least an order of magnitude lower than the average values reported for inner Kaneohe Bay water.

Other water quality characteristics pertinent to offshore Mokapu which were noted in the Kailua Bay Monitoring Study are given below:

1. The Kailua Bay sewage outflow at Kapoho Point is an area of high nutrient concentration. This water is transported towards Mokapu Point during flooding tide and to the south–southeast during ebbing tide.
2. Mokumanu Island is an area of moderate nutrient output, frequently affecting water quality in areas off North Beach during northwest current flow and areas off eastern Mokapu during south–southeast current flow. The source of the Mokumanu nutrients appears to be local and probably results from fecal material added to the area by the large bird populations on that island.
3. In the area off North Beach during the fall, winter, and early spring season, nutrient-rich water from Kaneohe Bay may intrude in the 4-meter to 16-meter layer via southeast flow. This nutrient enrichment is, however, relatively small in effect, with considerable dilution apparently occurring as a result of horizontal and vertical mixing.

A factor of recent origin which was not noted in the above-cited study is that Flyer's Monument Point (eastern Mokapu Peninsula) is the chosen locale for the Mokapu sewage outfall. According to the Planning Section of the Sewers Division, City and County of Honolulu, this outfall will discharge a major portion of the sewage from the Kaneohe–Kailua area (estimated effluent rates of 480 l/sec initially and 935 l/sec by the year 1993). The outfall pipe is to extend 1500 meters in a northwest direction to the 50-meter depth; the last 290 meters of the pipe is to be a diffuser section. The present estimated date of completion of the sewage facility is mid 1977. Although engineering studies predict that most of the effluent will be carried seaward to deeper water via offshore-heading bottom currents, it is assumed that from time to time abnormal oceanographic conditions may cause nutrient-laden waters to move either south into Kailua Bay or northeast toward Mokumanu Island and North Beach.

Five sites along the Mokapu coast were compared in a final evaluation in which points were tallied for six factors. Water quality–current effects and potential intake line stability and maintainability (as determined by estimated intake pipe length and surf zone characteristics) were the factors which were most heavily weighed. As a result of this evaluation, Pyramid Rock and Ulupau were established as the first and second choice sites. In later negotiations with the air station administration, it was discovered that Pyramid Rock Peninsula was planned as an area for recreational cottages. Therefore, Ulupau became the only available site.

Water quality at the Ulupau site was considered to be the best relative to all areas off of Mokapu Peninsula because of its distance from both the Kailua Bay–

Mokapu outfall and Kaneohe Bay nutrient sources. The major costs to be incurred at the Ulupau site would be for the construction of a 370-meter access road and the installation of power and water utilities. The specific area chosen for the laboratory was a barren patch of level ground on the eastern slope of Ulupau cone at an elevation of 12 meters. Along the shoreline an elevated (2- to 7-meter elevation) shelf of coralline beach rock averaging 15 meters in width extends nearly 400 meters in either direction. Below this shelf is an intertidal* shelf of coralline rock averaging 20 meters in width. This shelf is essentially of flat relief, with shallow potholes and gullies occurring over approximately 50% of its surface. Below the project site, at the seaward edge of the intertidal shelf, is a tidepool with about a 4-meter diameter and a 1.2 meter depth. This pool is well flushed by surf action, even at low tide, and was chosen as a convenient feature in which to locate an intake screen housing (where the main seawater suction pipes would terminate). From engineering and maintenance standpoints, such a location was desirable since it would be readily accessible during low tide and low surf conditions; it would also afford protection from high winter surf. Such a configuration for the intake piping meant that it would not be necessary to place excessive lengths of pipe in the area beyond the intertidal shelf, thus avoiding costly diving and work-boat operations.

DESIGN AND CONTRUCTION

Construction of the Ulupau microcosm facility began in February 1974. By late July the main laboratory structures (lab–office building, wet-lab, tank deck, and receiving tank tower) had been erected (Fig. 2). On a spring low tide in late July, a 600-pound (270-kg) concrete sewer elbow was placed, via helicopter lift, into the tide pool and was anchored with over three-hundred 18-kg bags of dry-mixed concrete. This elbow was to be used as an intake screen housing and was outfitted with a stainless-steel shelf, plastic screens, and a plastic access hatch (Fig. 3).

Installation of the 24-meter lengths of twin 4-inch (10.2 cm) suction pipes which were to lay on the intertidal shelf was scheduled for the next spring low-tide period in August. Because of occasional 5-meter winter surf on this exposed section of coast line, it was imperative that the intake pipes be sheathed in a protective covering that would be held securely to the shelf surface. The short duration of low-tide intervals precluded the building of concrete forms in the intertidal zone, so it was decided that a concrete "pillow sandwich" be used. For this

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* Actually supratidal; the mean elevation of the shelf is approximately 0.3 meter above MLLW. However, under normal surf conditions of 0.6 to 1.2 meters in height, the shelf is awash and essentially intertidal in appearance.

Figure 2. Ulupau microcosm facility. Seawater is pumped from the reef flat visible in the right-center of the photo. The two main seawater pipes can be seen rising to the receiving tank. The twelve microcosm tanks and their inlet and outlet boxes are situated on top of the deck. Wet-lab space is located in the white enclosure located behind the staircase. Partially hidden from view, on ground level in front of the deck, are the six auxiliary-holding tanks.

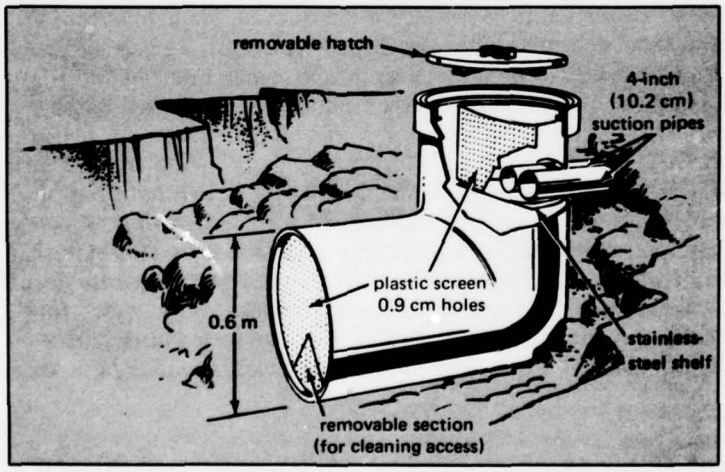

Figure 3. Cutaway view of the intake screen housing. Except for the access hatch and screen face, this structure is buried in several layers of concrete-filled bags.

purpose, 35 closed burlap bags (10-ounce material), each 0.9 meter by 2.2 meters, were fabricated. A patch of reinforced fabric was sewn in the center of a flat side of each bag, and a 20-cm-long slit was cut through each patch area.

On the selected low-tide morning, four hundred 9-kilogram bags filled with dry-mix concrete were placed in the small potholes and ravines along the planned pipeline route. Next, on top of the path leveled by the smaller bags, a row of the larger burlap bags was laid down and pumped with concrete to a 15- to 20-cm thickness (Fig. 4). The dual lengths of 4-inch (10.2 cm) PVC pipe were then coupled and put in position on top of the pillows, and a second row of pillows was laid over the pipeline. Finally, short lengths of steel reinforcing rod were driven vertically through the pillow sandwich and, where existing, into the smaller underlying bags (Fig. 5). The job was completed in 5 hours, at which time the concrete had set to a point where it could withstand mild surf action.

The pipeline has now been in place for 1 year and has withstood a winter of periodic heavy surf with no signs of movement. There is apparently strong bonding between the individual bags and the limestone stratum of the shelf. The coarse texture of burlap and muslin bag fabrics allowed extrusion and intermixing of the finer cement components. Also, it appears that the strong alkaline output of the wet concrete served to clean much of the organic material off the underlying limestone, thus promoting more effective bonding between those surfaces.

Figure 4. Installation of the suction pipes across the intertidal reef shelf. The pillows in the foreground have just been filled. Workers are readying other pipe and pillow sections for emplacement.

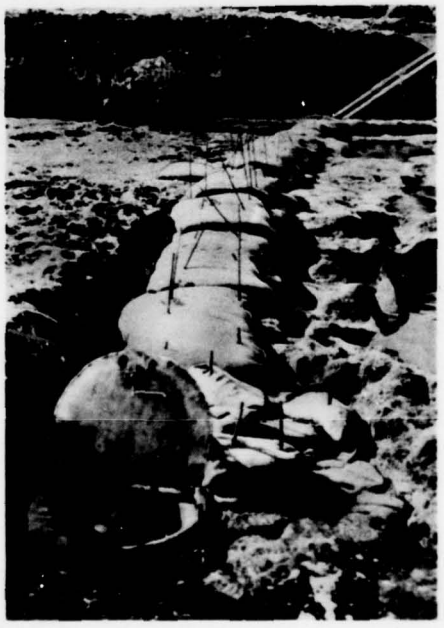

Figure 5. Completed suction pipe structure. The protruding reinforcing bars were later trimmed and bent over. In foreground is the intake screen housing, with the temporary metal hatch opened to show the suction pipe ends.

At the landward edge of the intertidal shelf a Technocheck® flapper-type check valve was placed in-line on both intake pipes. The check valves were placed in this position relatively far upstream to ensure access to them for servicing even under moderately high surf conditions. The water volume in the 21 meters of line seaward of either check valve has been lost on only one occasion. This occurred on a spring low tide when dead algae blocked the inner intake screen, and the intake housing compartment was momentarily pumped dry. Now that the reliability of the check valves has been proven and it has been found that they are not noticeably affected by biological fouling, these valves will eventually be relocated at the intake termini.

Immediately upstream of the check valves, the suction lines rise at a 45-degree incline to the top of the 2.7-meter-elevation beach rock shelf. Situated 6 meters landward of this incline is a 2.7-meter by 2.7-meter by 2.1-meter-high pumphouse of splash-proof hollow tile construction. In the pumphouse are two Sethco* SFA 3 by 4 by 10 model 2310-30 centrifugal pumps (Fig. 6). The impeller casings, covers, impellers and impeller nuts on these pumps are made entirely of filament-wound glass impregnated with a thermoset vinyl-ester resin.

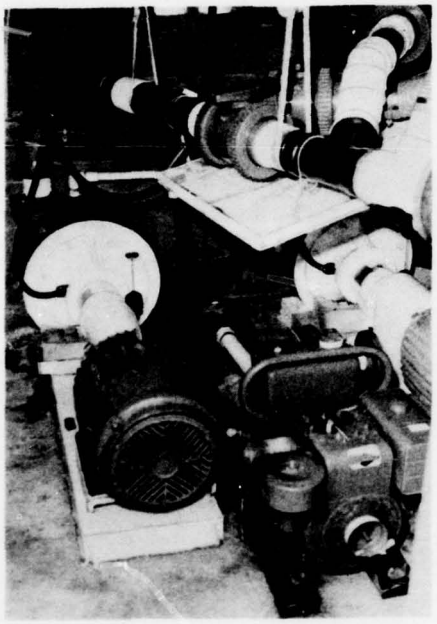

Figure 6. Pumphouse view. An electrically driven Sethco pump is shown on the left and the gasoline-driven back-up pump at lower right. The other electric pump is visible at extreme right. Above the pumps is the 3-inch (7.6-cm) discharge piping and crossover valving.

* Manufactured by Sethco Pump Co., 235 Township Line Road, Hatfield, Pennsylvania 19440.

The pump shafts are enclosed in titanium sleeves, and the seal packing used is of graphite filament. A low flow of seawater, which is obtained from the pump's main discharge line, is filtered through an in-line strainer and is directed to the packing for additional cooling and for a back-flushing flow which assists in excluding sand from the seal.

The pumps are driven by 25-hp, 1750-rpm, 230-v, U. S. Electric Marine motors. Each pump is capable of delivering 180 gpm (680 lpm) of seawater to the top of the receiving tank at an elevation of 17 meters above sea level at 19.5 meters total dynamic head. Under normal operating flow the discharge valves are throttled to 140-gpm (530 lpm) delivery. Actually, 10-hp motors would have been sufficient to provide the required pumping rate (at least 100 gpm; 380 lpm); however, at the time the pumps were being procured, the smaller motors were available only on back-order periods of more than 1 year.

To date each pump has been operated for about 7000 hours over a period of 18 months. During this period the performance of each pump has been flawless, and there are no obvious signs of wear on any pump components. Only one pump is in operation at any one time, allowing routine cleaning and servicing of the inactive pump. Switch-over between pumps is usually made once a week.

In the event that an operating pump should lose either suction or discharge pressure or that a power failure should occur, an automatic cut-off system has been installed (Fig. 7). Basically, this system consists of a perforated bucket suspended from a cord; the cord is attached above to the unhinged end of a spring-loaded lever arm. Underneath the lever arm at a position between the cord attachment and the fulcrum–hinge is a push-button microswitch. At normal

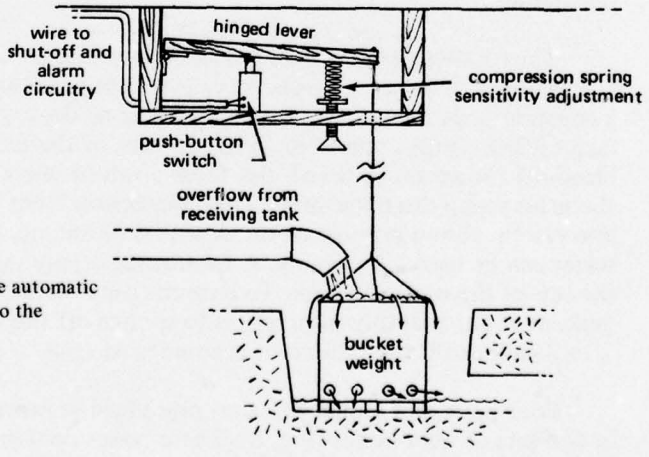

Figure 7. Schematic diagram of the automatic shut-off and alarm system linked to the seawater pumps.

pumping volume (140 gpm; 530 lpm), overflow from the receiving tank is discharged into the bucket at a rate which keeps the bucket overflowing. A 30–40-gpm (115–150 lpm) decrease in flow for a period of over 5–10 seconds is enough to allow the bucket to run dry, whereupon the spring pressure pushes the lever arm up, releasing the microswitch. The microswitch activates a series of relays which turn off the pump motors, start an alarm in the main building, and activate an automatic telephone dialer. A prerecorded tape in the dialer unit repeats an alert message to the homes of several laboratory workers and to an answering service. Thus, the pumps will remain off until an operator can troubleshoot the malfunction and ensure that the suction intake pipe is primed prior to restarting the pump.

If a prolonged power outage occurs or if both electric pumps are inoperative, a gasoline-powered 6-hp Marlow Model 233 cast iron pump is available in the pumphouse. This pump utilizes the existing suction and discharge lines of one of the electric pumps and is capable of delivering 140 gpm (530 lpm) of seawater to the receiving tank. This gasoline-powered system is intended for use only as a back-up water supply to avoid the damaging effects on the microcosm communities due to elevated temperature, rainwater dilution, lowered oxygen, and buildup of metabolic wastes. During daylight hours under full sunlight, 4 hours of no-flow is the approximate length of time at which temperatures begin to approach damaging levels. Somewhat longer periods of no-flow can be tolerated during night hours.

Seawater is pumped from the pumphouse to the receiving tank, a distance of 60 meters, via either of two buried 3-inch (7.6 cm) PVC pipes. In the pumphouse, cross-over valving allows either pump to discharge seawater through either pipe. This feature ensures that a back-up discharge line can always be left stagnant for a sufficient length of time (approximately 10 days) to free it of biological fouling.

The receiving tank is made of fiberglass and has a capacity of 360 gallons (1400 liters). A 3-inch (7.6-cm) overflow pipe near the top of the tank maintains a constant head of seawater for gravity feed to the experimental tanks. A 1¼-inch (3.2 cm) pipe connected to the bottom of the tank serves as an additional bleed-off for excess flow and also feeds a valved line which connects to one of the main pump discharge lines. This connection from the tank to the discharge line system allows priming of the downhill plumbing. Either salt water or freshwater can be used for priming. A freshwater supply pipe and valve are located at the top of the receiving tank. To date the only maintenance required on the tank, at about monthly intervals, is to sponge off the interior walls and remove 2 to 3 gallons (8 to 12 liters) of accumulated sand.

Four pairs of 1¼-inch (3.2 cm) pipes feed seawater from the receiving tank to two sets of inlet boxes (Fig. 8). These boxes contain two adjustable-level overflow standpipes to maintain constant water levels. Into six pairs of ports in the

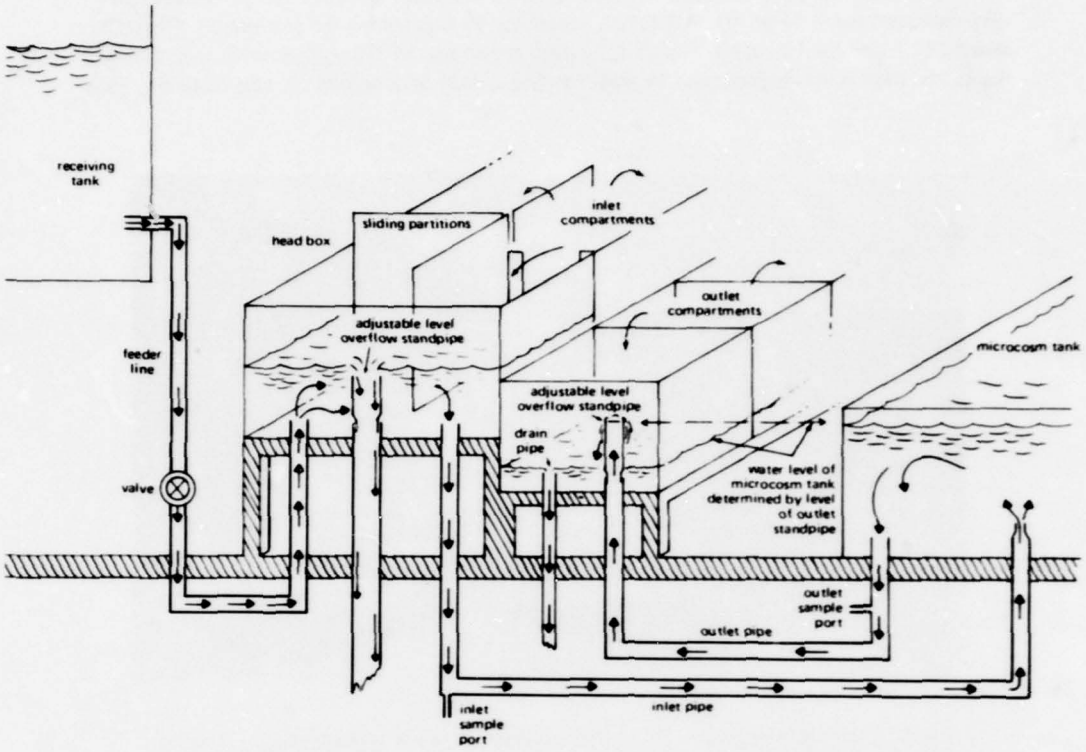

Figure 8. Schematic section showing inlet and outlet plumbing for one microcosm tank.

bottoms of the inlet boxes, water is fed to six microcosm tanks; another identical inlet box feeds the other six microcosm tanks. To maintain flow rates of 2½ to 3 gpm (10–12 lpm)*, the water level in the inlet boxes is set 3 to 4 inches (7.6 to 10.2 cm) higher than the water level in the experimental tanks. To obtain higher or lower flow rates (from 0.1 to 15.0 lpm), the standpipes can be adjusted up or down. Vertical compartment walls with sliding partitions enable the inlet ports to be grouped into many different combinations which can receive isolated water supplies. This arrangement allows different experimental additives (such as silt, nutrients, and heavy metals) to be fed directly to the appropriate inlet compartments.

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* This flow rate range had been selected by the HIMB staff to produce an approximate water residence time of 1 hour in the 600-liter microcosm tanks.

The 12 microcosm tanks are arranged in a square around the perimeter of the elevated deck (Fig. 9). All tanks measure 46 inches by 46 inches by 18 inches deep (117 cm by 117cm by 45.7 cm) and are made of fiberglass with gel-coated surfaces which are pigmented brown on the inside and white on the outside. The

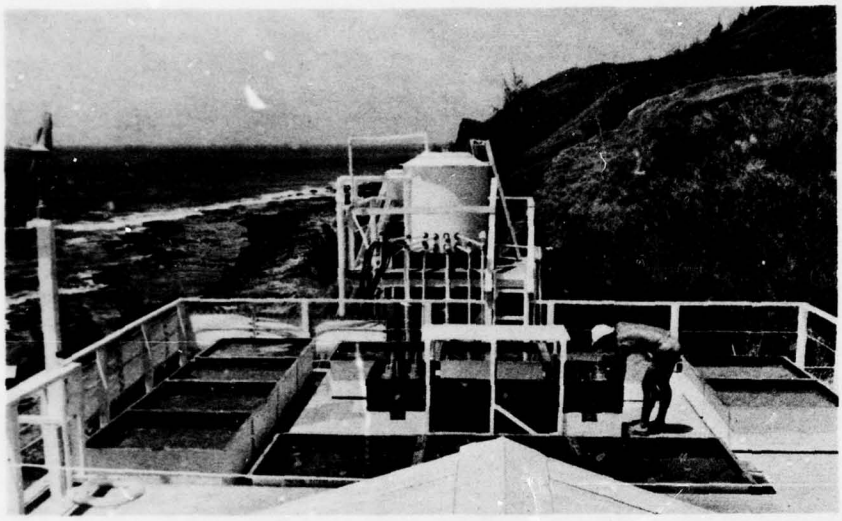

Figure 9. Deck view of microcosm tanks. The large-diameter overflow pipe extending from the top left side of the receiving tank maintains a constant gravity head in that tank. Smaller-diameter feed lines that connect to the inlet boxes protrude from the lower-front of the receiving tank. Inlet and outlet boxes are located in the center of the deck. The operator is adjusting a valve on one of the four feeder lines.

tanks rest directly on the deck surface and all inlet–outlet plumbing for the tanks is located under the deck. A pair of outlet pipes extend from each tank to an outlet compartment in the center of the deck. In this compartment, water exits the outlet pipes through adjustable standpipes; the overflow levels of these standpipes are set to the levels of water desired in the microcosm tanks. Also located in each outlet compartment is a single drain pipe which carries the outlet water into the master drain system. These drain pipes are fitted with 2.2cm diameter automatic siphons which allow the water level in the compartments to rise to nearly the top height of the overflowing outlet standpipes before siphoning action is started. At a flow rate of 2½ gpm (10 lpm), each outlet compartment fills in about 60 seconds, while siphon action, when initiated, empties the compartment in less than 15 seconds. This rhythmic siphoning action can be used as a means of automatically monitoring flow rates. In the near future, a pair of sensing electrodes will be placed within the lower and upper

limits of the water-level travel. An automatic data logging system is presently being installed. One function of this system will be to use the time intervals between water contact on the two electrodes to periodically compute the filling rates (flow rates) of the compartments. The logging system then records these rates on magnetic tape for later analysis.

Six 100-gallon (400 liter) wooden tanks with fiberglassed interiors are positioned alongside the deck at ground level. These tanks are used to hold and acclimate organisms which are later used in the microcosm tanks. Water is provided to these tanks through plastic spigots which branch from a single 1¼-inch (3.2 cm) pipe. Outlet water from these tanks passes directly into a drain system via centrally located standpipes. Flow rates through the holding tanks can be maintained at 1 to 5 gpm (4–20 lpm).

All of the inlet piping from the receiving tank to the microcosm tanks and the outlet piping from the tanks to the outlet boxes is double-plumbed. This scheme allows one-half of the twin system to be capped and valved shut, causing the contained seawater to become anoxic, thereby killing any accumulated fouling organisms. Once a week the changeover is made between active and inactive systems. The entire process takes about 30 minutes. In a 1-week period little fouling occurs, and a very small amount of detritus is flushed into the tanks from the anoxic lines.

If required, mechanical cleaning of the pipes is facilitated by crosses and tees installed wherever space permitted on all 90-degree bends or at branch points. In the course of 5 months of continuous operation, mechanical cleaning has not been required on any of the water supply systems except for the sun-exposed inlet and outlet boxes, which are brushed and siphoned free of algal buildup and debris every few days. An important factor in the low maintenance experience for the system is that the supply water is low in nutrients and oceanic biota. Whereas barnacles, oysters, tunicates, serpulid worms, and anemones are very common fouling organisms in Kaneohe Bay water, just 6.4 km away at Ulupau these animals are rare or, in the case of barnacles, have yet to be observed in the Ulupau fouling community.

Presently undergoing fabrication at the Ulupau facility is an automated system which will be capable of shunting samples of inlet and outlet water from each experimental tank through a sensing loop where several parameters, such as oxygen, temperature, and pH, will be measured. A full sampling cycle will require about 60 minutes for completion. Sensor instruments will provide inputs to an analog-to-digital data logger where all information will be recorded on tape. A prototype of this automatic sampling and data l ging system has been under development and test at the HIMB microcosm facil.ty for the last 2 years. During this test period, it was noted that a number of changes in the basic layout of the tanks and their associated plumbing would benefit greatly the interfacing of the

sampling system with the rest of the flow-through system. For example, placing the tanks at Ulupau in a square configuration on top of a deck avoided potential problems related to changes in the flowing water. Sample hoses which tap water outlet pipes directly beneath the tanks were made uniform in length, thus avoiding differences in water chemistry caused by differing residence times in the sample hoses. The central solenoid valve manifold, to which all the sample lines connect, will be located under the center of the deck. By placing all the inlet and outlet boxes in one central cluster, the problem of varying lengths of sample hoses from the inlet ports will also be resolved. Additionally, the lengths of the inlet sample hoses would be minimal because of the closeness of the solenoid valve manifold. The central location of the inlet and outlet boxes also establishes that lengths of inlet piping from the inlet boxes to the tanks will be similar.

All materials used in the seawater distribution system are constructed of plastic (excluding the titanium pumpshaft sleeves). Piping and fittings are of PVC (polyvinyl-chloride) plastic, except for four ABS (acrylonitrile-butadiene-styrene) plastic 3-inch "wye" fittings in the main supply pipes. PVC ball valves are used at most of the flow control points. PVC gate valves, ¾ to 1¼ inches (1.9 to 3.2 cm), have been used in a few places. These proved generally unsatisfactory because of brittle components. On the low-pressure sample hose system, over fifty ½-inch (1.3 cm) "twist-pinch" PVC K-valves* were used and have been found to be a reliable, low-cost alternative to ball valves for low-use application.

Various data for the seawater supply systems of both facilities are given in Table 1. These characteristics have a direct effect on the occurrence and survival rates of organisms entrained in the seawater supply piping. The hole sizes in the suction intake screens and the size of maximum flow-through gaps in the pumps govern the average size of organisms which can pass intact through the system. The speed at which the impellers are rotating, the velocities of water in the pipes, and the lengths of the pipes are other factors which affect entrained organisms. Small invertebrates and sub-adult fish are commonly passed through the Ulupau system intact, although a substantial amount of these motile animals rapidly leave the tanks through the drain system. The largest undamaged animal to pass through the Ulupau pumps was a 1.3 cm diameter by 30 cm long moray eel.

At HIMB, the higher pump motor speed and pumping velocities, in conjunction with smaller intake screen and impeller gaps, serve to filter out all but the smaller planktonic organisms. However, the survival rates of those organisms are at least high enough to support the recruitment and food requirements of the productive HIMB microcosms.

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*Ryan Herco Products Corp., 1311 West Magnolia Blvd., Burbank, California 91506.

Table 1. Seawater supply system data for Ulupau and HIMB facilities.

	Ulupau	*HIMB*
Intake screen holes diameter	1.3 cm	0.6 cm
Impeller-to-casing clearance	1.6 cm	0.6 cm
Pump operating speed	1750 rpm	3500 rpm
Normal pumping flow	140 gpm (530 lpm)	80 gpm (300 lpm)
Suction line diameter/length	10.2 cm/24.4 m	3.8 cm/9.1 m
Discharge line diameter/length	7.6 cm/54.9 m	3.2 cm/18.3 m
Maximum pumping velocity	1.9 m/s	5.2 m/s

INTERFACILITY CALIBRATION TEST

Design of Calibration Test

The Ulupau and HIMB facilities provide a series of environmental contrasts which produce a considerable spectrum of conditions suitable for the study of microcosm community responses. The most conspicuous potential differences between the sites are in their water qualities and in the quality and quantity of contained planktonic larvae. An intercomparison test was conducted to evaluate these noted differences. Another conspicuous difference not evaluated in this test, but to be assessed in the future, is the difference in solar radiation between the two areas. Effects such as variable temperature and salinity at both facilities appear to be so small as to be considered trivial.

The following intercomparison design was adopted to evaluate the consequences of these differences. A total of 12 tanks, 6 at Ulupau and 6 at HIMB were dedicated to the test. All tanks were fully exposed to sunlight, with special care taken to avoid shading. Treatments were duplicated at both facilitites. To each tank (Fig. 10) common starting substrata were added: 2-cm particle-size quarried coral limestone approximately 5 cm deep and, centered on the south side of each tank, a 30-cm-diameter by 5-cm-deep plastic pan filled with 0.7mm particle-size sand. The tank walls provided a third (hard-bottom) substratum, and terracotta and asbestos panels served as fourth and fifth substratum types.

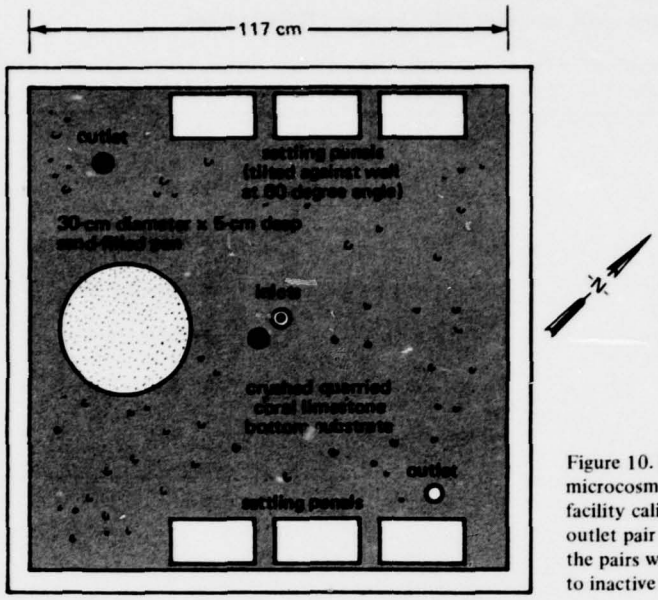


Figure 10. Plan view diagram of typical microcosm tank as configured for inter-facility calibration test. Only one inlet-outlet pair was in use at a given time; the pairs were switched from active to inactive once a week.

Unfiltered seawater was supplied to the 500-liter tanks at approximately 10 lpm, yielding approximate 50-minute residence times for the water in the microcosms. After 14 days fish (*Acanthurus triostegus)* were added to the tanks: two fish in one treatment pair at each facility, one fish in a second treatment pair, and no fish in a third pair. Thus, for common available substrata the effects of varying the grazing pressure in the two water quality regimes could be evaluated.

At each facility the water quality was examined over a 3-month period. Two questions were to be asked of the water quality: (1) What is the chemical environment imposed upon the biota? (2) What biotic activity can be inferred from that water quality? The biota was examined *per se* for evidence of the effects of water quality, the effects of larval differences, and the effects of varying grazing pressure. Biological observations were made on a routine basis so that the successional stages of the settling organisms could be documented for both sites. Substratum effects, which might also be examined, have so far been given only very preliminary consideration.

Meteorological measurements were made at both sites to enable comparison of those variables, and those data will be reported at a later date when a full year of data are available.

Methods

Salinity. Through the period 20 March to 17 April, water samples were taken twice daily (near 1000 and 1500 hours) from the inlet boxes at both sites. These samples were measured for salinity with a Plessey Environmental Systems model 6230N laboratory conductivity salinometer. The meter was standardized against secondary offshore seawater samples, which were in turn standardized against "Copenhagen Water," the oceanographically accepted primary salinity standard.

Nutrients. Nutrient samples were collected from the inlet boxes of both sites concurrently with salinity samples (usually twice daily near 1000 and 1500 hours). The samples were filtered through Millipore® type AA 0.8-μm 47-mm-diameter membrane filters into 10-cc polystyrene vials capped with polyethylene snap lids. The samples were immediately frozen and processed later using standard techniques (Strickland and Parsons, 1968) with a Technicon II Autoanalyzer.

Settling Panels and Biological Observations. Two types of panels, terracotta (red clay) and asbestos, 20 cm by 30 cm, were placed on opposite (north and south) sides of the tanks. The panels were assigned at random to six mid-side positions and were tilted against the tank sides at approximately 30 degrees off vertical. Figure 11 shows the five exposure intervals used in the study. One pair of panels (terracotta and asbestos) was removed from each tank at four different time exposures. Asbestos panels are presently being used by NUC for field studies of fouling communities, and the terracotta panels have been used in

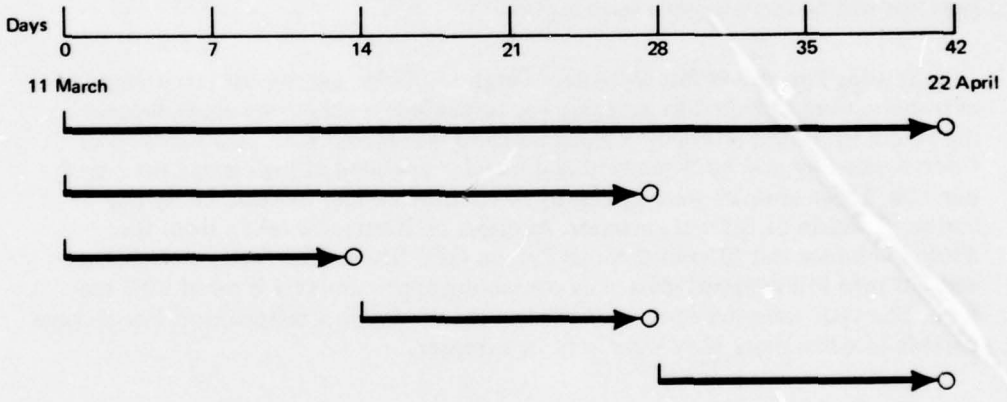


Figure 11. Settling panel exposures.

a number of field and laboratory microcosm studies by HIMB researchers. For the calibration period it was decided to use both panel types so that suspected differences in settlement on the two surfaces could be studied.

Upon removal for sampling, both sides of the panels were inspected for types, sizes, and abundances of all visible algae and invertebrates. At approximately the same frequency, these observations were made in a less quantitative fashion on the walls and bottom of each tank.

Settling Panels and Weight Determination. For weight determinations, the attached biota on the lower 20-cm by 20-cm (400 cm^2) area of each panel was scraped off with a razor blade into tared crucibles. Those samples were weighed for wet weight and then dried to dryness in an oven at 100°C. After being weighed for dry weight, the samples were ground to a fine homogenous powder in an automatic grinder and then stored in a dessicator.

For ash weight determinations, weighed subsamples of 50 to 100 mg were placed in small-volume crucibles and combusted in a muffle furnace for 4 hours at 500°C. After being cooled in a dessicator, the samples were then reweighed to yield ash weights. The percent ash figures derived from those analyses were applied to the 400-cm^2 dry weights to obtain ash-free dry weights (presented in a later section).

From each panel scraping a dry weight subsample of 3 to 5 mg was also analyzed for carbon–hydrogen–nitrogen content using an F and M Model 185 CHN analyzer. Samples were combusted at 1100°C and cyclohexanane-2, 4-dinotrophenolhydrozone was used as a standard. These data are not reported here but will be included in a later report.

Settling Panels and Phytopigment Content. Biota samples for phytopigment extraction were obtained by scraping the upper-left panel corner areas, measuring 10 cm by 10 cm (100 cm^2). Each scraping was mixed with 50 to 200 ml of filtered seawater and homogenized in a blender operated at high speed for 1 to 2 minutes. These samples were increased to volumes of 150 to 1000 ml by the further addition of filtered seawater. Aliquots of 10ml were taken from the diluted volumes and filtered through 2.4-cm GFC filters. The filters were folded and put into foil-wrapped glass vials containing approximately 8 ml of 90% acetone. The vials were stored for periods less than 1 day in a refrigerator. For storage periods of a few days, they were kept in a freezer.

The final stages of analysis followed the techniques and calculations specified in Strickland and Parsons (1968).

Fish Growth. Fish *(Acanthurus triostegus)* were put into the tanks after 2 weeks of flow-through to allow for a period of time in which algal growth could accumulate for food. Young adults of 20- to 50-g size were used because of the ready availability and hardiness of this size range of individuals. All fish were obtained by trapping from inner Kaneohe Bay (around Coconut Island) and were weighed and measured for standard length before being introduced into the tanks. Thereafter, they were reweighed at bi-weekly intervals. Fish that died or disappeared were replaced as soon as was practical. Individuals that disappeared had either jumped out of the tanks and subsequently been taken away by small night-foraging animals or, in the case of small individuals, had died and were eaten by crabs.

Oxygen. Dissolved oxygen measurements were made at both microcosm locales with Yellow Springs Instruments Model 54 oxygen meters. At each facility, waters from inlet boxes and tank outlets were sampled through fittings located at those points and were channeled through hoses and valve manifolds to a central sampling loop. Hose lengths and sampling flow rates were similar at both sites, and oxygen probes were placed in the sampling loops with the sensing faces parallel to flow directions. At Ulupau, oxygen readings were taken manually near 0800, 1200, and 1600 hours on most working days. At HIMB, the automatic sampling and data logging system was operational for much of the intercomparison period, and measurements were recorded by that system approximately every 20 minutes. The HIMB data-logged information has not yet been processed; however, data are available (and are presented in a later section) for a 2-week period in April, when readings were recorded manually.

Results

Water Quality. The HIMB facility is located approximately 2.5 km from the Kaneohe municipal sewage treatment plant outfall and is subjected to relatively high nutrient levels. The Ulupau facility is on an open and dry coastline and is apparently not ordinarily subjected to any significant terrestrial pollution. Nutrient levels were judged to be the water quality parameters most likely to vary from the one location to the other. Therefore, nutrient samples were collected from the inlet water usually twice daily (mid-morning and mid-afternoon) between 12 March and 7 April. In addition, oxygen was monitored at the inlet and outlet of one tank at each site to establish the community effect of metabolic

activity in the tanks. Salinity and temperature were also monitored; those parameters showed variations and between-facility differences which are regarded as inconsequential to the microcosm communities (Table 2).

Table 2. Summary of temperature and salinity data for Ulupau and HIMB. Values are for inlet water.

	Ulupau	*HIMB*
April 1975 mean temperature	23.3°C	data unavailable
May 1975 mean temperature	23.5°C	data unavailable
Mean temperature for 10-day period in April 1975	23.6°C	23.9°C
Maximum diurnal variation	1.5°C	1.5°C
Maximum heating/cooling effect (above or below ambient) of pipes and microcosm tanks	1.8–2.0°C	1.8–2.0°C
Mean salinity March–April 1975	34.99 ppt	34.69 ppt
Maximum/minimum salinity March–April 1975	35.60/34.99 ppt	35.09/33.71 ppt

Table 3 summarizes the inlet nutrient data; and Fig. 12 through 14 present the data in the form of frequency histograms. The PO_4 values differed dramatically as expected; HIMB levels were about twice those found at Ulupau (means of 0.46 versus 0.22 μg-atom PO_4-P/l). On the other hand, both NO_3 and NH_3 provided unexpected results. The NH_3 values did not differ significantly from HIMB to Ulupau (means of 1.61 versus 1.96 μg-atoms/l), while the HIMB NO_3 values were substantially lower than those at Ulupau (means of 0.58 versus 2.64 μg-atoms/l).

A comparison of the histograms demonstrates the nature of the greatly differing NO_3 values. Both localities have very similar NO_3 distributions below 3 μg-atoms/l, with modal values of about 0.4. However, the Ulupau samples show a strong secondary mode of about 10 μg-atoms/l. In summary, PO_4 showed the expected relation of high values at HIMB and low values at Ulupau; both NO_3 and NH_3 were clearly higher at Ulupau than was anticipated.

It has been demonstrated (Webb *et al.*, 1975; Wiebe *et al.*, 1975) that coral reef communities can fix large amounts of atmospheric nitrogen and can subsequently export that fixed nitrogen through the marine environment. Thus it

Table 3. Inlet nutrient data, summary statistics for 12 March through 7 April 1975.

Values are mean and standard errors in μg-atoms/l, and in parentheses, number of analyses.

Ulupau	*HIMB*	*HIMB–Ulupau Difference*	*Significant at 95% ?**
PO_4 0.22 ± 0.032 (62)	0.46 ± 0.027 (57)	+0.24 ± 0.042	yes
NO_3 2.64 ± 0.657 (61)	0.58 ± 0.060 (56)	−2.06 ± 0.660	yes
NH_3 1.96 ± 0.567 (61)	1.61 ± 0.148 (57)	−0.35 ± 0.586	no

* Twice the standard error of the difference is regarded as the 95% confidence interval on the difference. This is not strictly true, but the difference between such a calculation and a standard t-test for large-sample statistics is small.

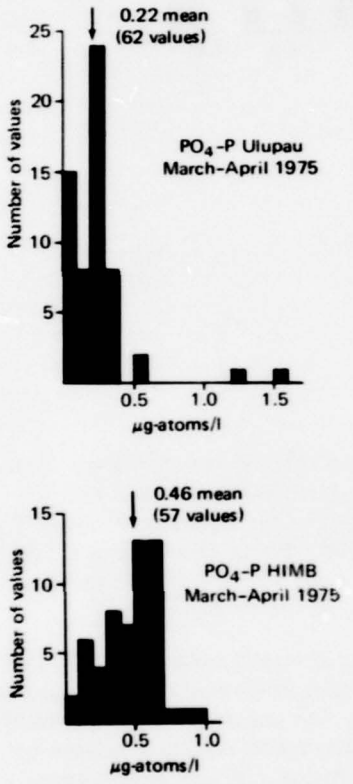


Figure 12. Frequency histograms of PO_4-P concentrations for Ulupau and HIMB inlet water, March–April 1975.

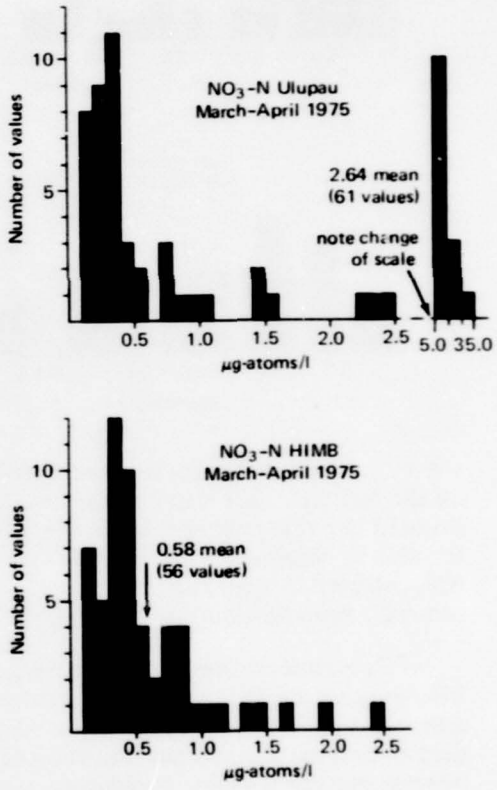


Figure 13. Frequency histograms of NO_3-N concentrations for Ulupau and HIMB inlet waters, March–April 1975.

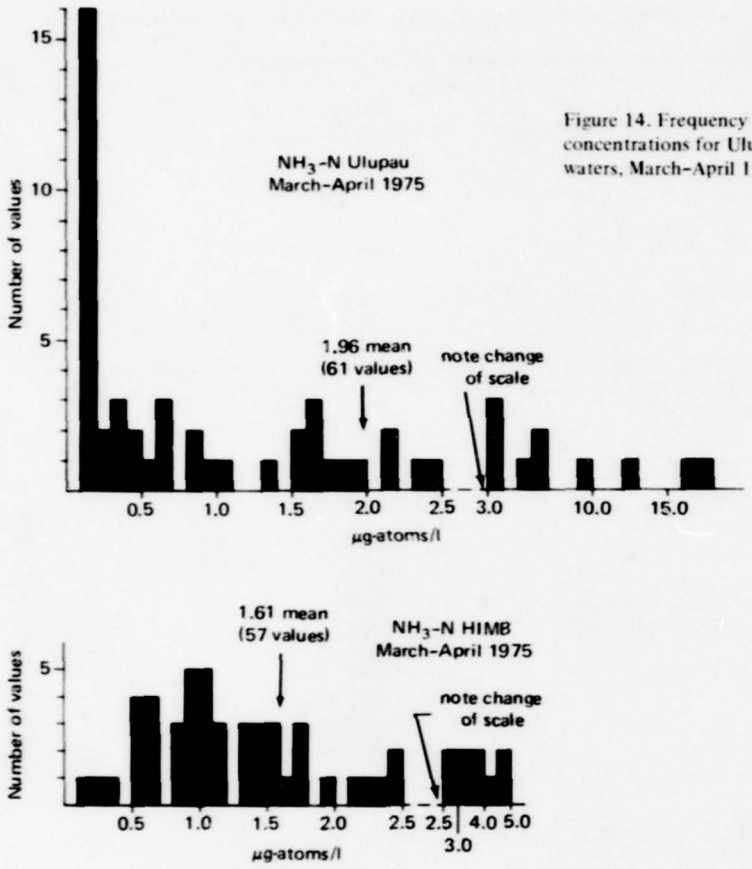


Figure 14. Frequency histograms of NH_3-N concentrations for Ulupau and HIMB inlet waters, March–April 1975.

was hypothesized that the high nitrogen values might represent nitrogen fixation on the reef flat. That water would then be pumped from the intake sump (located in a tide pool at the reef flat outer edge) to the microcosm tanks. To test this hypothesis, water was sampled on the reef flat at Ulupau and in inlet water at both Ulupau and HIMB on 12 and 13 June 1975. Table 4 provides summary statistics and Fig. 15 through 20 present the data graphically.

PO_4 values remained lower at Ulupau than at HIMB, while the NO_3 and NH_3 patterns shifted. Ammonia values were higher at Ulupau over this 30-hour series than in the March–April series. Also, NO_3 was somewhat higher at HIMB than at Ulupau; nevertheless, the Ulupau nitrogen values remained relatively close to the HIMB values. Examination of the Ulupau nutrient data demonstrated that virtually all of the variability in NH_3, NO_3, and PO_4 at Ulupau occurs at mid-tides (either rising or falling through levels of 0.2 – 0.4 m). The following hypothesis was constructed.

Table 4. Inlet nutrient data, summary statistics for 12–13 June 1975.

Values are mean and standard errors in μg-atoms/l, and in parentheses, number of samples.

Ulupau	*HIMB*	*HIMB–Ulupau Difference*	*Significant at 95% ?**
PO_4 0.19 ± 0.017 (28)	0.43 ± 0.067 (28)	+0.24 ± 0.069	yes
NO_3 0.62 ± 0.069 (28)	0.85 ± 0.075 (28)	+0.23 ± 0.102	yes, barely
NH_3 2.37 ± 0.190 (34)	1.56 ± 0.106 (29)	−0.81 ± 0.218	yes

*Twice the standard error of the difference is regarded as the 95% confidence interval on the difference. This is not strictly true, but the difference between such a calculation and a standard t-test for large-sample statistics is small.

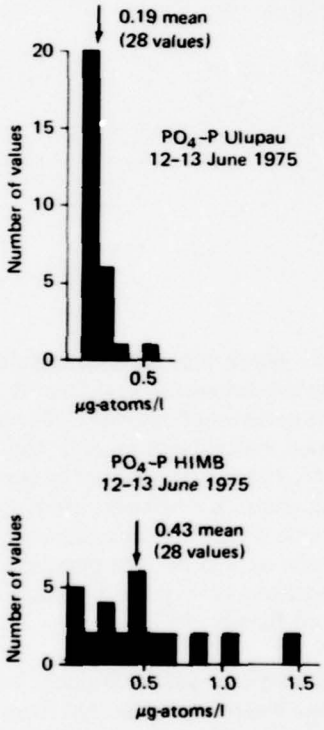


Figure 15. Frequency histograms of PO_4-P concentrations for Ulupau and HIMB inlet waters, 12–13 June 1975.

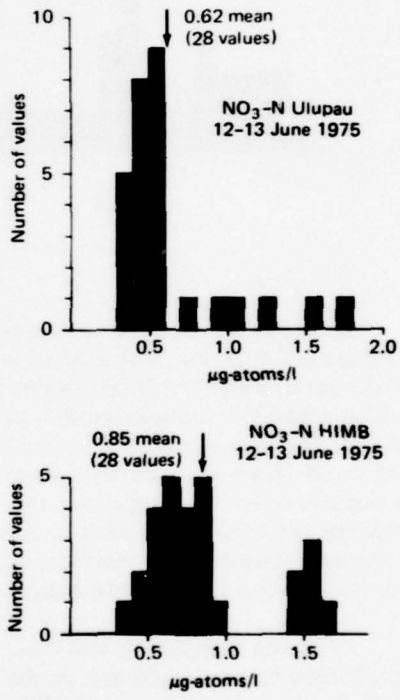


Figure 16. Frequency histograms of NO_3-N concentrations for Ulupau and HIMB inlet waters, 12–13 June 1975.

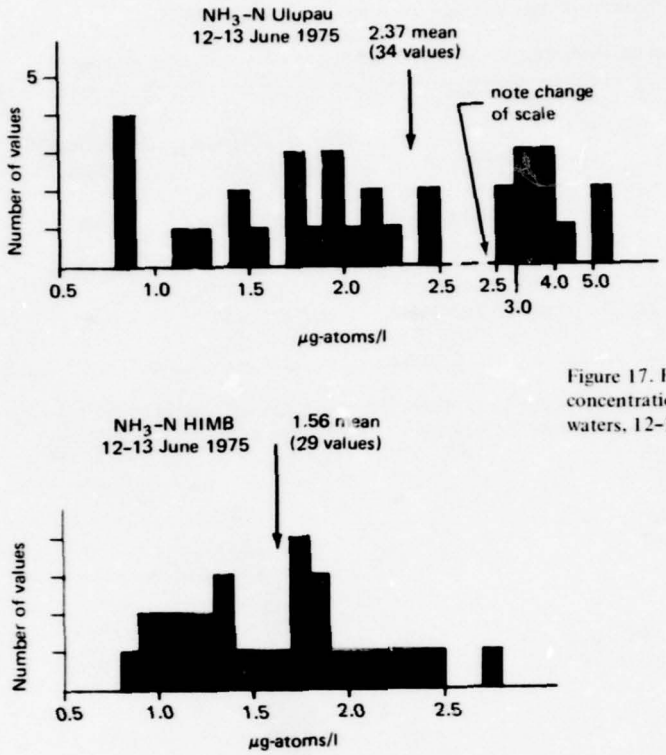


Figure 17. Frequency histograms of NH_3-N concentrations for Ulupau and HIMB inlet waters, 12-13 June 1975.

At low tides under the low-surf conditions which prevailed during the 30-hour sampling period, virtually no water is washed across the reef flat into the intake sump pool. The intake water is nearly oceanic in composition because it is drawn into the sump through a subtidal tunnel which penetrates to the fore-reef. As the tide rises under low-surf conditions, water surges onto the reef flat and washes high-nitrogen water into the intake sump. Eventually, the volume of oceanic water washing across the reef increases with the rising tide and dilutes the reef effects. As the tide falls, the reverse of this pattern prevails. There is not an equivalent PO_4 pattern because phosphorus is very tightly retained and recycled by the reef community (Pilson and Betzer, 1973).

Sampling designed to test this hypothesis provided partial support but was insufficient to prove or disprove the hypothesis. Water was collected from tide pools on the reef crest and reef flat over a tide cycle. There is a suggestion that the NO_3 and NH_3 levels increase at mid-tides, whereas PO_4 is relatively constant (Fig. 18). This pattern is very close to that observed in the inlet water (Fig. 19).

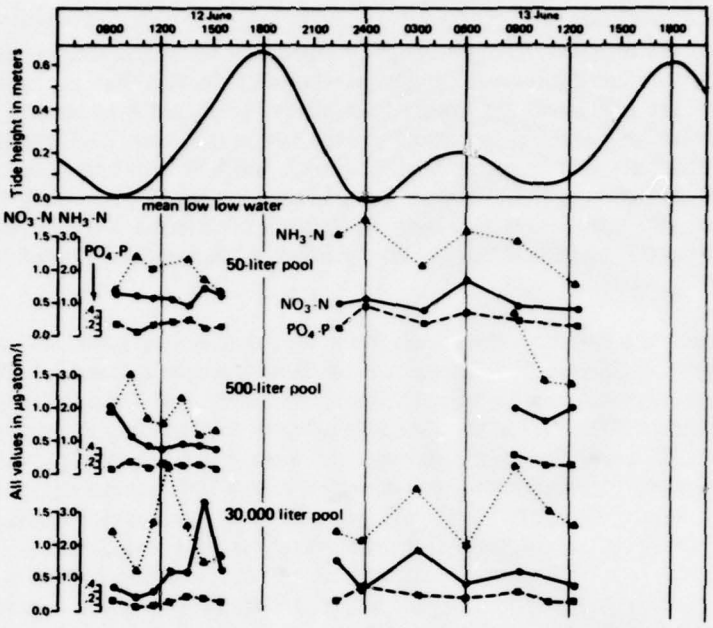


Figure 18. Concentrations of PO_4-P, NO_3-N, and NH_3-N versus time and tide for three reef flat pools at Ulupau, 12–13 June 1975.

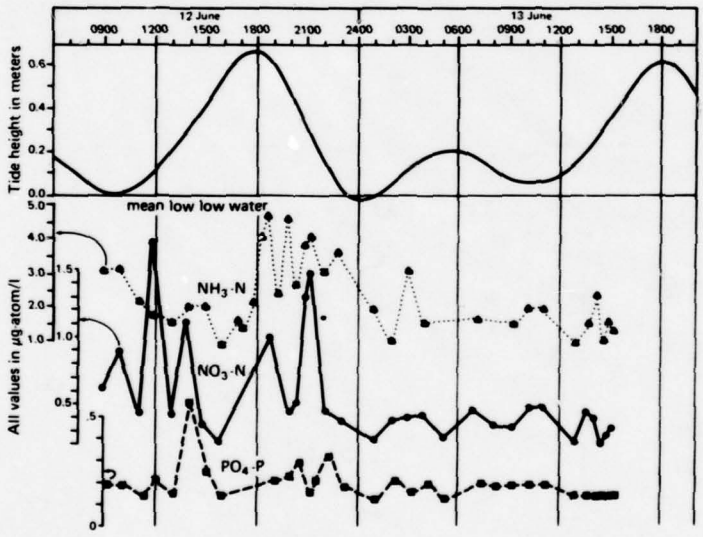


Figure 19. Concentrations of PO_4-P, NO_3-N, and NH_3-N versus time and tide for Ulupau inlet water, 12–13 June 1975.

Again, an hypothesis can be offered. L. Smith and S. Smith (unpublished) have noted that the nitrogen-fixing blue-green algae *Calothrix* and *Hormothamnion* are much more common on the higher portions of the flat than in the tide pools. Therefore, the tide pools are similar to sump pools in that they receive nitrogenous water only when it is washed off the flat at mid-tide. Unfortunately, by the time this pattern was recognized, L. Smith and S. Smith had noted a decline (probably seasonal) in the nitrogen-fixing blue-green algae. From July through September 1975, there has also been an absence of extreme NO_3-N values (greater than 0.4 μg-atom/l) in the inlet waters at Ulupau (L. Franzisket, unpublished data).

Figure 20 shows that the HIMB pattern in nutrient variation differs markedly from that of Ulupau. NH_3 shows no significant temporal variation. Both NO_3 and PO_4 decrease on a rising tide, then increase as the tide falls. One sample on a low-high tide differs from this trend. It appears that the major component of variation is tidal and that this variation can be attributed to periodic dilution of high-nutrient bay water with ocean water. In the NH_3 record this pattern is absent, apparently because NH_3 levels away from the immediate vicinity of the Kaneohe sewer outfall do not differ dramatically from oceanic values. Because NH_3 is the preferred nitrogen compound for phytoplankton utilization in the bay, it is quickly reduced to ambient levels (J. Caperon, personal communication, and Bienfang, 1975).

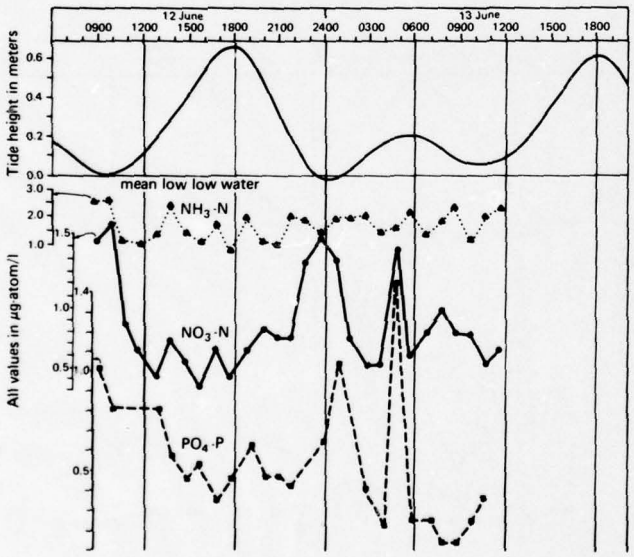


Figure 20. Concentrations of PO_4-P, NO_3-N, and NH_3-N versus time and tide for HIMB inlet water, 12-13 June 1975.

The oxygen content of inlet water at both Ulupau and HIMB ordinarily does not differ markedly from saturation levels (about 6 to 7 mg O_2 /l at the temperature and salinity of the typical inlet waters). Figure 21 illustrates the Ulupau O_2 pattern on 12–13 June. There is a small diel oscillation attributable in large part to biological activity. Biological activity in the tanks greatly alters the O_2 levels of those waters. In fact, the differences in oxygen level between the inlet water and tank outlet water is a measure of the biological activity in the tank microcosms; concentration differences times flow rates yield metabolic rates. There can also be gas exchange between the atmosphere and the water in the tanks, although this flux is normally small in relation to the biologically induced O_2 differences.

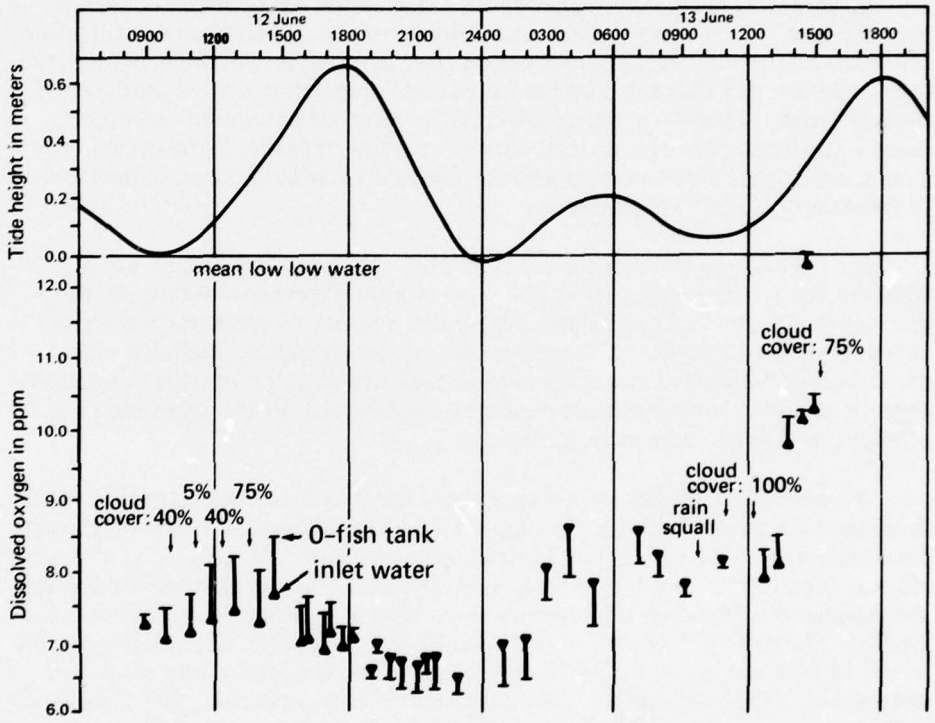


Figure 21. Dissolved oxygen concentrations versus time and tide for inlet water and a zero-fish microcosm tank at Ulupau, 12-13 June 1975.

Figure 22 illustrates the noontime outlet minus inlet O_2 values at Ulupau and HIMB for a tank without fish at each location. All the Ulupau tanks exhibited O_2 production for about the first 3 weeks of the experiment, but data for the following 3 weeks showed apparent net O_2 consumption at noontime. Only after about 3 months did the Ulupau tanks consistently produce O_2 at noontime. In contrast, the HIMB tanks showed higher O_2 production after 3 weeks than were shown anywhere over the 5-month course of measurement at Ulupau. This contrast between the productivity of the two sites is consistent with the hypothesis that benthos metabolism at Ulupau may be limited by the availability of one or more materials dissolved in the water. This limitation initially manifests itself in primary production (that is the production of O_2, hence organic carbon). The reduction of autotrophic fixation of food is then transmitted up the food chain to the herbivores, carnivores, and detritivores.

By monitoring net oxygen production during the daytime and consumption at night, it is possible to derive an estimate of the net fixation or utilization of organic carbon by the tank microcosm communities. As will be demonstrated below, the Ulupau tank selected for this initial comparison is a net producer of organic carbon. Therefore, the community is either accumulating carbon (as increasing biomass or organic detritus) or exporting material. If the community were a net organic carbon consumer, there would necessarily be an importation of fixed carbon as a food resource.

On 12–13 June 1975, the outlet and inlet values in Ulupau tank no. 3 (without fish) were monitored at 30- to 60-minute intervals to determine the diel oxygen (hence carbon) balance. A parallel automated experimental series at HIMB was lost because of equipment failure. In the future, such diel experiments will be conducted routinely at both facilities to determine the community organic carbon production, storage, import, and export. In the meantime, Fig. 23 provides the Ulupau data as an exemplar.

Numerical integration of the area above the x-axis demonstrates that on 12 June the tank community fixed a net of 3.32 g O_2. During the succeeding nighttime period (to 0800 on 13 June) respiration expended 3.16 g O_2. The nighttime respiration rate was about 4.4 mg O_2/min. If this rate is assumed to be constant throughout the day, then the community gross production rate was about 6.5 g O_2/day. The difference between the daytime net production and the respiration over 24 hours was about 0.16 g O_2. In other words, the community produced only about 2.5% more oxygen than it consumed. If it is assumed that 1 mole of carbon is produced or consumed for each mole of oxygen production or consumption, then the carbon production was about 0.06 g/day.

Although data for 12–13 June are unavailable for HIMB, net carbon production rates in those tanks are generally above 1 g C/day (G.S. Key *et al.*, unpublished). This substantial difference between Ulupau and HIMB production

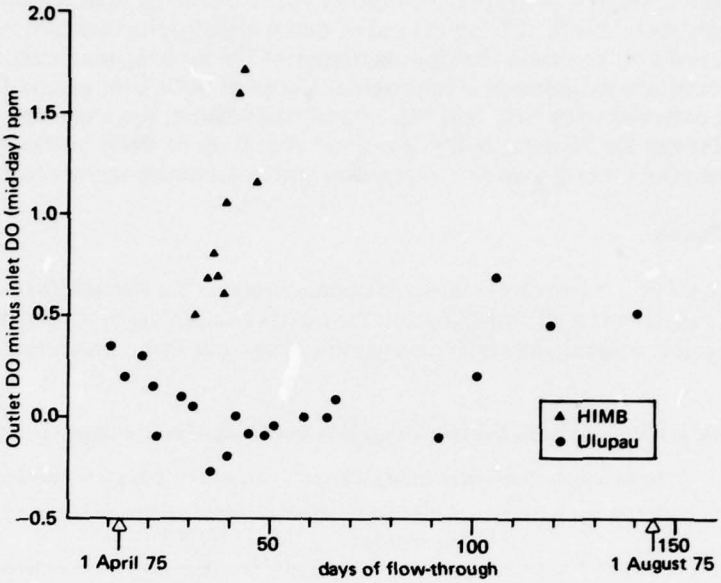


Figure 22. Mid-day concentrations of outlet water dissolved oxygen minus inlet dissolved oxygen versus time (days of flow-through) for a zero-fish tank at Ulupau and HIMB.

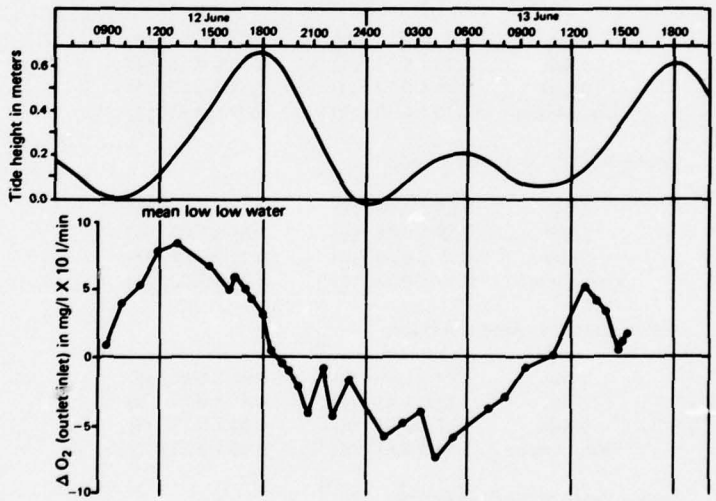


Figure 23. Oxygen production or consumption (as *outlet dissolved oxygen minus inlet dissolved oxygen* times 10 l/min average flow rate) versus time for a zero-fish tank at Ulupau, 12–13 June 1975.

rate is again consistent with the assumption that a dissolved material limits community metabolism at Ulupau (and in many oligotrophic benthic ecosystems). In order to ascertain the characteristics of the limiting materials, a nutrient addition experiment is in progress. Tanks at both Ulupau and HIMB are being enriched with NH_3 and PO_4. As of this writing, the experiment has been underway for 30 days, and it is evident that both of these nutrients have significant effect on biological composition and community metabolism.

Settling Panels

Tables 5 and 6 present means and standard errors for dry weights and ash-free dry weights from the asbestos and terracotta panel scrapings. The dry weights include organic material, inorganic $CaCO_3$ and SiO_2, and scrapings from

Table 5. Dry weight for all panel exposure groups and fish treatments at Ulupau and HIMB.

Means and standard errors are in g/400 cm^2, number of samples is in parentheses.

	Ulupau	*HIMB*
2-week panels removed at 2 weeks (no fish in tanks during this exposure)		
Grand mean	1.22 ± 0.09 (12)	0.79 ± 0.05 (12)
2-week panels removed at 4 weeks		
2 fish	0.81 ± 0.03 (2)	0.34 ± 0.11 (2)
1 fish	1.20 ± 0.22 (4)	0.50 ± 0.11 (4)
0 fish	1.41 ± 0.13 (4)	0.62 ± 0.17 (4)
Grand mean	1.20 ± 0.12 (10)	0.49 ± 0.08 (12)
2-week panels removed at 6 weeks		
2 fish	0.23 ± 0.03 (4)	0.18 ± 0.05 (4)
1 fish	0.18 ± 0.08 (4)	0.30 ± 0.05 (4)
0 fish	0.17 ± 0.08 (4)	0.29 ± 0.07 (4)
Grand mean	0.19 ± 0.04 (12)	0.25 ± 0.03 (12)
4-week panels removed at 4 weeks		
2 fish	2.54 ± 1.05 (3)	0.45 ± 0.05 (4)
1 fish	3.45 ± 0.45 (4)	0.49 ± 0.07 (4)
0 fish	4.17 ± 0.43 (4)	1.03 ± 0.13 (4)
Grand mean	3.46 ± 0.41 (11)	0.66 ± 0.13 (12)
6-week panels removed at 6 weeks		
2 fish	0.41 ± 0.25 (4)	0.46 ± 0.08 (4)
1 fish	0.30 ± 0.17 (4)	0.46 ± 0.21 (4)
0 fish	0.23 ± 0.16 (4)	0.74 ± 0.10 (4)
Grand mean	0.31 ± 0.10 (12)	0.63 ± 0.08 (12)

Table 6. Ash-free dry weight for all panel exposure groups and fish treatments at Ulupau and HIMB.

Means and standard errors are in g/400 cm^2, number of samples is in parentheses.

	Ulupau	*HIMB*
2-week panels removed at 2 weeks (no fish in tanks during this exposure)		
Grand mean	0.31 ± 0.03 (12)	0.27 ± 0.03 (12)
2-week panels removed at 4 weeks		
2 fish	0.29 ± --- (1)	0.07 ± 0.03 (3)
1 fish	0.49 ± 0.11 (2)	0.12 ± 0.01 (3)
0 fish	0.46 ± 0.03 (2)	0.13 ± 0.02 (4)
Grand mean	0.44 ± 0.05 (5)	0.11 ± 0.01 (10)
2-week panels removed at 6 weeks		
2 fish	0.05 ± 0.02 (3)	0.09 ± 0.01 (3)
1 fish	0.03 ± 0.01 (4)	0.10 ± 0.02 (4)
0 fish	0.01 ± --- (1)	0.11 ± 0.02 (4)
Grand mean	0.04 ± 0.01 (8)	0.10 ± 0.01 (11)
4-week panels removed at 4 weeks		
2 fish	0.96 ± 0.48 (3)	0.13 ± 0.02 (4)
1 fish	1.14 ± 0.24 (4)	0.12 ± 0.02 (4)
0 fish	1.58 ± 0.16 (2)	0.33 ± 0.02 (4)
Grand mean	1.18 ± 0.19 (9)	0.19 ± 0.03 (12)
6-week panels removed at 6 weeks		
2 fish	0.09 ± 0.03 (3)	0.18 ± 0.01 (4)
1 fish	0.06 ± --- (1)	0.23 ± 0.05 (4)
0 fish	0.11 ± 0.05 (3)	0.27 ± 0.01 (4)
Grand mean	0.09 ± 0.02 (7)	0,23 ± 0.02 (12)

the panels (at least for asbestos panels, which tended to lose asbestos material when scraped). Consequently, the ash-free dry weights are judged a more satisfactory weight measure for intercomparisons. Inspection of Tables 5 and 6 reveals consistently higher scraping weights at HIMB for the panels not exposed to fish as compared to those exposed to fish. At Ulupau, however, those differences are not as consistent. Apparently the fish-grazing effects (tending to reduce panel biomass) were overwhelmed in the Ulupau tanks by the growth of abundant populations of grazing sea hares. Because of this confounding effect on the Ulupau fish treatment data, the weight and phytopigment parameters discussed later in the text are pooled by duration of exposure at each facility.

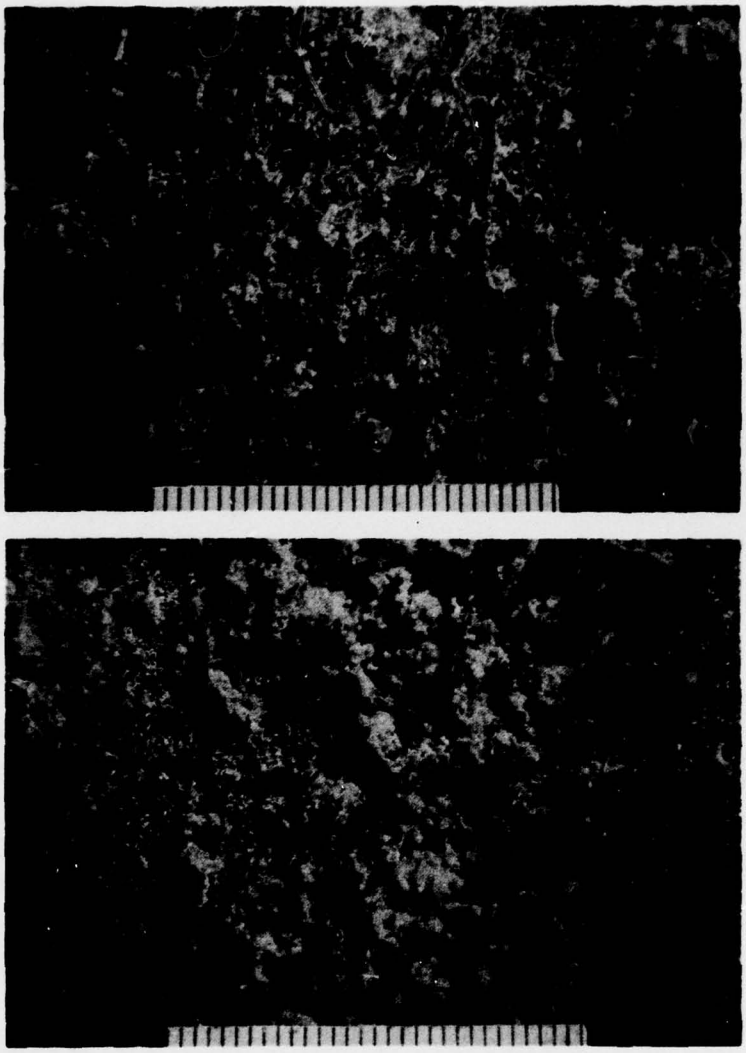
At Ulupau the ash-free dry weights of 0.31, 1.18, and 0.09 g/400 cm^2 at 2-weeks, 4 weeks, and 6 weeks, respectively, reflect the buildup of an algal mat over the first 4 weeks and the subsequent removal of that growth by a "bloom" of grazing invertebrates through weeks 5 and 6. Herbivorous fish *(Acanthurus triostegus)* were added to the tanks at the end of the first 2 weeks, and approximately 10 days later the first sea hares *(Stylocheilus longicauda)* were noted in the tanks. By the end of the third week the combined effect of the grazing by the fish and the increased population of sea hares had stripped all obvious algae from the tank walls, panels, and substrata.

Another manner in which this effect may be seen is in the comparison of 2-week panels which were introduced and removed successively later in the tank histories. The first two such sets of panels developed relatively heavy growths in 2 weeks (0.31 and 0.44 g/400 cm^2, respectively), while the third set developed almost no growth in 2 weeks (0.04 g/400 cm^2).

In the weeks following the intercomparison period, the *Stylocheilus* were greatly reduced in numbers, apparently because of predation by crustaceans which had grown to young adult size and because of a lack of sufficient food (algae) in the tanks. Concurrent with the decrease in the sea hare population was an increase in algal growth in the tanks. Also, sessile invertebrates (such as *Spirorbis* spp., *Hydroides* spp., and *Aiptasia diaphana*) colonized all substrata at a more rapid rate than was observed in the first 6 weeks of flow-through. Figure 24 presents a group of photographs of various substrata in the tanks of both facilities after 7 months of exposure.

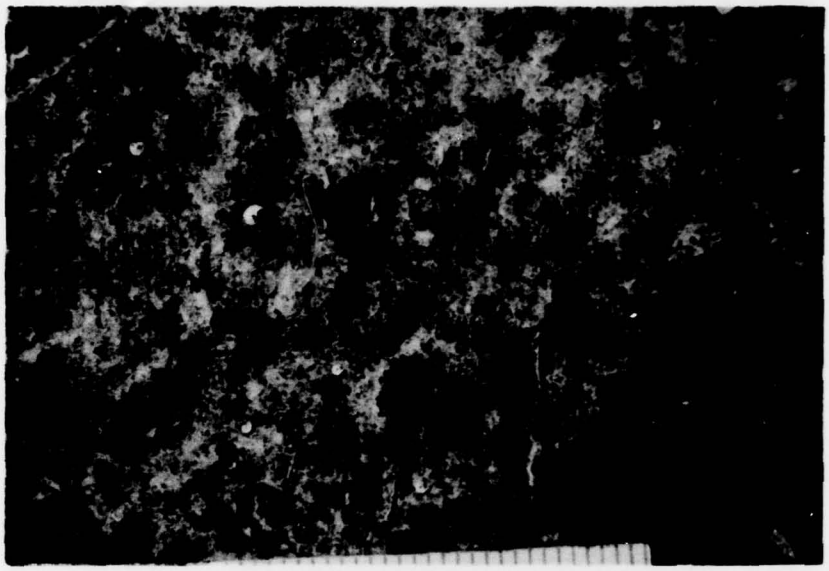
At HIMB a more diverse assemblage of algae and sessile invertebrates settled in the tanks. Diatoms and other algal forms dominated in the first 2 weeks of flow-through. However, tunicates, barnacles, oysters, anemones, and vermetid and polychaete worms, which had settled earlier in larval stages, grew rapidly and by the fourth week were the more obvious organisms on the panels. The ash-free means of 0.11, 0.19, and 0.23 g/400 cm^2 for the 2-, 4-, and 6-week exposures, respectively, are illustrative of the relatively stable level of absolute biomass on the HIMB panels and tank walls.

The difference in diversity between the two systems may be variously interpreted. It is frequently stated that stressed communities are of low diversity. One might conclude, therefore, that the HIMB microcosm communites are less stressed than those at Ulupau. A more reasonable evaluation would consider the biotic structures of the communities in terms of resource (nutrients and then food) availability. The "stress" imposed by the nutrients in Kaneohe Bay might be more appropriately regarded as a selective stimulus which may provide certain algae a competitive advantage over other, probably more slowly growing, algae. The rapidly replenished and abundant standing crop of algae is then available as food and may actually increase the number of niches available, and hence the diversity of the heterotrophic community.

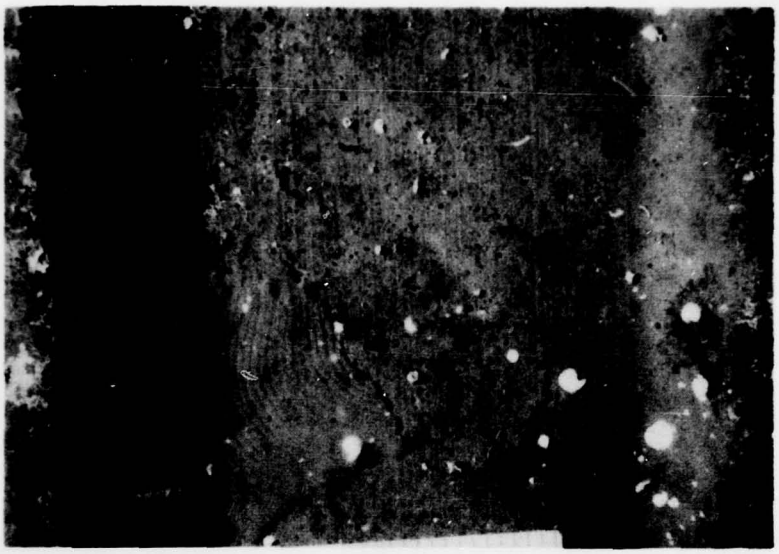


(a) Front of asbestos panels. After this length of exposure, invertebrate growth such as tunicates, oysters, annelids, and anemones had decreased markedly on the HIMB panels. Most of the coverage on both panels is an algal mat of diatoms and blue-green and green algae. At the left in the HIMB photo is a barnacle (*Balanus* sp.). Dimensions of each photo are 6.4 cm x 4.4 cm.

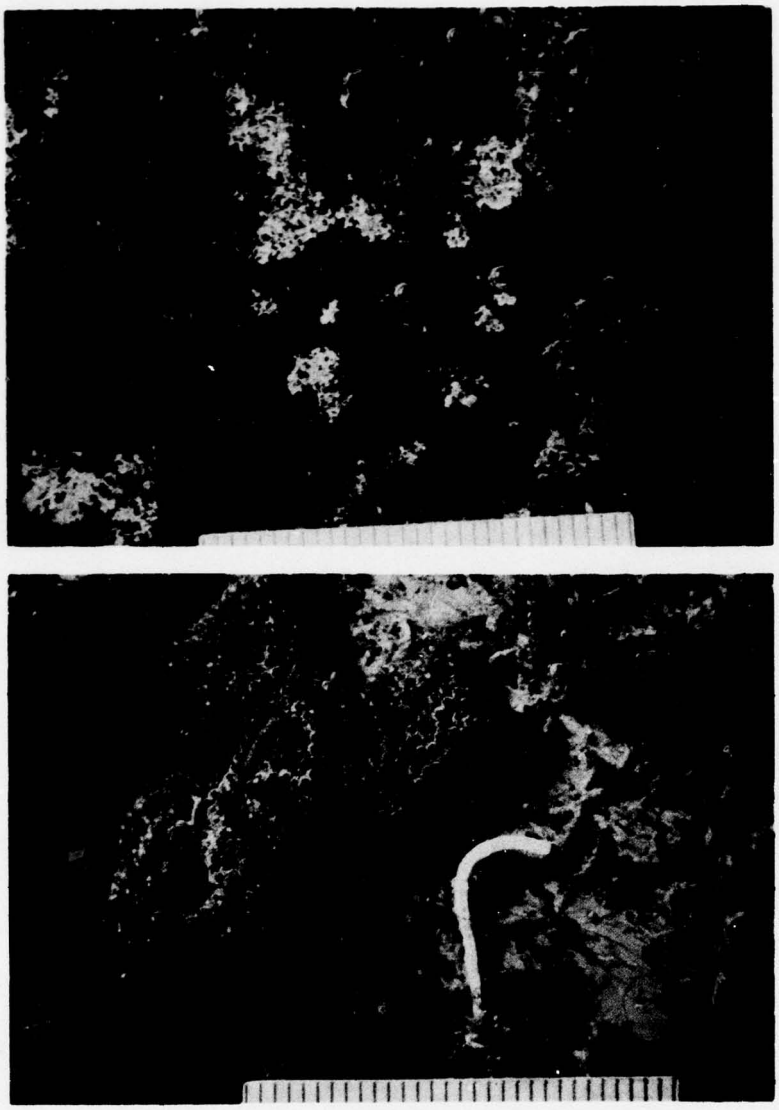
Figure 24. Various substrata in the tanks at Ulupau and HIMB after 7 weeks of exposure. Ulupau substrata are shown in top photos, HIMB substrata in bottom photos.



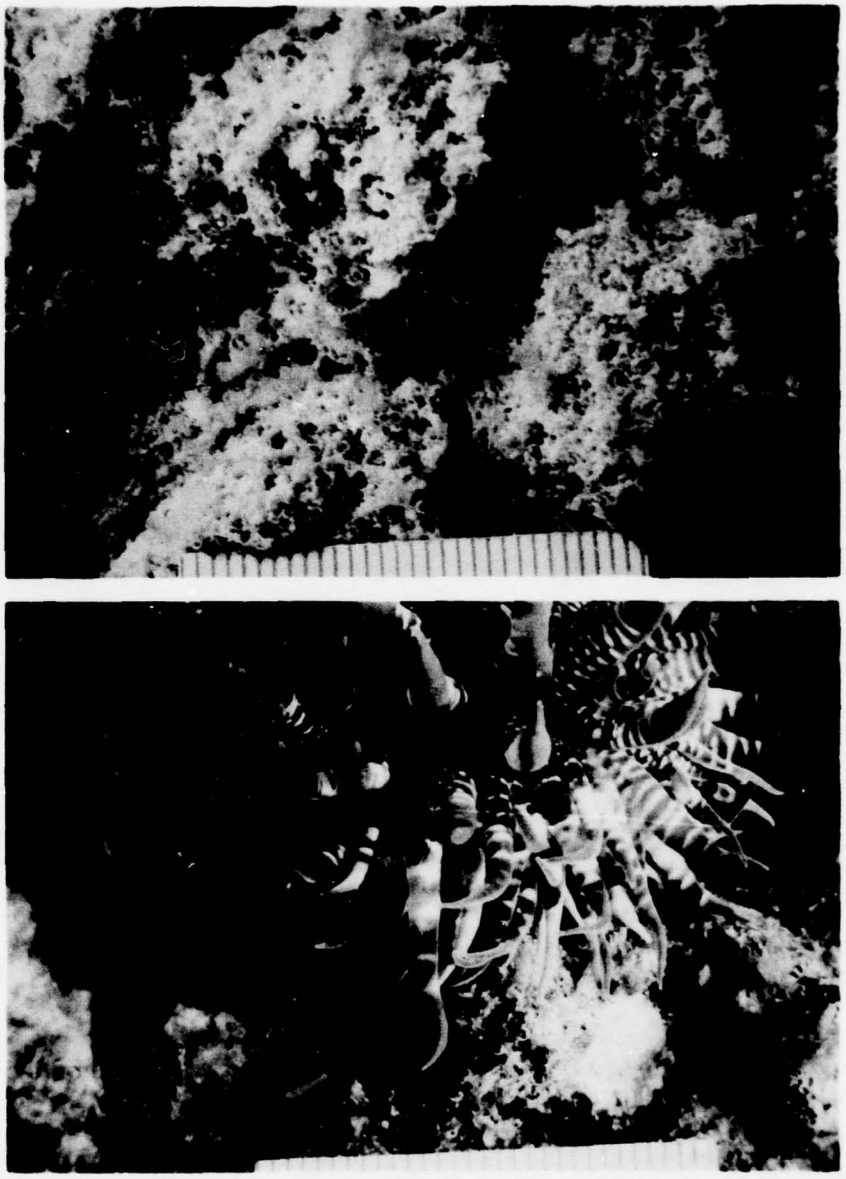
(b) Back sides of asbestos panels. Note the sparcity of algal growth. The smaller white tubes on both panels are largely *Spirorbis* spp. and *Hydroides* spp. The long tubes on the HIMB panel are serpulid worms. Also visible on the HIMB panel are colonial tunicates (*Polyclinum constellatum*) and barnacles (*Balanus reticulatus*). Dimensions of each photo are 6.4 cm x 4.4 cm.



(c) Back sides of terracotta panels. Conspicuous algal growth on the Ulupau panel is calcareous algae (nondescript pink and white blotches). Larger pink-colored mass near lower-left of Ulupau panel is a *Dolabrifera* sp. egg coil. On both panels the shells of small *Spirorbis* spp. and *Hydroides* spp. are common. Other organisms on the HIMB panel are *Crucibulum spinosum* (lower left), *Ostrea* sp. (upper center), colonial tunicate (lower right), and serpulid worms and solitary tunicates (upper left and right). Dimensions of each photo are 6.4 cm x 4.4 cm.



(d) Northwest tank wall at 10-cm depth. The darker blotches on the Ulupau panel are largely blue-green algae; white areas are calcareous algae. Some small shells of *Spirorbis* spp. and *Hydroides* spp. are barely visible. On the HIMB panel the brown string-like structures in lower right are tubes of tube-dwelling amphipods (Corophiidae). The red-orange colonial tunicate is *Botryloides* sp. growing around a barnacle. A serpulid worm tube is in lower center. Other growth is primary filamentous algae and diatoms. Dimensions of each photo are 6.4 cm x 4.4 cm.



(e) Crushed coral limestone bottom substratum. Dominant growth at both sites is a mat of diatoms and filamentous algae. At HIMB, larger invertebrates such as anemones (*Boloceroides mcmurrichi* in photo), tunicates, annelids, oysters, sponges, and vermetid and serpulid worms are very common. Dimensions of each photo are 6.4 cm x 4.4 cm.

Analyses for phytopigment concentrations were performed to obtain approximate measures of the relative quantities of plant material (diatoms and other algae) in the panel samples. Table 7 lists the principal algal groups noted in the microcosm tanks and shows the phytopigments found in those groups (Dawson, 1966). Chlorophyll *a* and carotenoids are common to all algal groups and are thus roughly indicative of total algal quantities, whereas chlorophylls *b* and *c* can be used as general indicators of the presence of green algae, diatoms, and brown algae.

Table 7. Distribution of chlorophylls and carotenoids among principal algal groups found in microcosm tanks (modified from Dawson, 1966).

	Chlorophylls			
	a	b	c	*Carotenoids*
Rhodophyta (red algae)	+			+
Cyanophyta (blue-green algae)	+			+
Phaeophyta (brown algae)	+		+	+
Bacillariophyceae (diatoms)	+		+	+
Chlorophyta (green algae)	+	+		+

Means and standard errors for phytopigment values of exposure groupings all panel samples are given in Table 8; included also are the ash-free dry weight statistics so that the phytopigment levels can be compared with total biomass values. Ratios of the amounts of chlorophyll *c*, carotenoids, and ash-free dry weights are tabulated in Table 9. Chlorophyll *b* data are also presented in this table, but only as numbers of samples in which a measurable amount of chlorophyll *b* was found. Because chlorophyll *b* occurred in relatively few samples, and then only in very small amounts, mean values would have been of no interpretive value.

As would be expected, chlorophyll *a* and carotenoids show a relatively constant relationship to each other (in an approximate ratio of 2:1). In turn, those parameters vary in relation to ash-free dry weight means presumably as a function of total algal content of the samples. At Ulupau chlorophyll *a* and carotenoids increase in the first 4 weeks and then show a marked decrease at the 6-week sampling (undoubtedly as a result of the intensive grazing by the fish and sea hares). The same pigments at HIMB show mean concentrations of 0.98 and 0.91 $\mu g/cm^2$ for 4- and 6-week exposures, respectively. Those values and the ash weight means for the same exposures indicate again that the HIMB algal communities were capable of producing more organic material than the grazers in the microcosms utilized.

Table 8. Ash-free dry weights and phytopigments for the various panel exposures at Ulupau and HIMB.

Values are mean, standard error, and in parentheses, number of samples.

	Ash-free Dry Weight ($g/400\ cm^2$)	*Chlorophyll a* ($\mu g/cm^2$)	*Chlorophyll c* ($\mu g/cm^2$)	*Carotenoids* ($\mu g/cm^2$)
2-week panels removed at 2 weeks				
Ulupau	0.31 ± 0.03 (12)	0.37 ± 0.04 (10)	0.08 ± 0.02 (10)	0.22 ± 0.02 (10)
HIMB	0.27 ± 0.03 (12)	0.30 ± 0.04 (12)	0.13 ± 0.02 (12)	0.16 ± 0.02 (12)
Significant difference ?*	no	no	no	no
2-week panels removed at 4 weeks				
Ulupau	0.44 ± 0.05 (12)	0.44 ± 0.08 (7)	0.22 ± 0.05 (7)	0.27 ± 0.05 (7)
HIMB	0.11 ± 0.01 (10)	0.48 ± 0.07 (10)	0.17 ± 0.02 (10)	0.24 ± 0.02 (10)
Significant difference ?*	yes	no	no	no
2-week panels removed at 6 weeks				
Ulupau	0.04 ± 0.01 (8)	0.06 ± 0.01 (8)	0.05 ± 0.01 (8)	0.02 ± 0.01 (8)
HIMB	0.10 ± 0.01 (11)	0.34 ± 0.06 (11)	0.11 ± 0.03 (11)	0.18 ± 0.03 (11)
Significant difference ?*	yes	yes	no	yes
4-week panels removed at 4 weeks				
Ulupau	1.18 ± 0.19 (9)	2.85 ± 0.49 (9)	0.77 ± 0.13 (9)	1.37 ± 0.23 (9)
HIMB	0.19 ± 0.03 (12)	0.98 ± 0.20 (12)	0.31 ± 0.07 (12)	0.45 ± 0.09 (12)
Significant difference ?*	yes	yes	yes	yes
6-week panels removed at 6 weeks				
Ulupau	0.09 ± 0.02 (7)	0.20 ± 0.04 (12)	0.09 ± 0.02 (12)	0.11 ± 0.03 (12)
HIMB	0.23 ± 0.02 (12)	0.91 ± 0.20 (12)	0.21 ± 0.02 (12)	0.48 ± 0.10 (12)
Significant difference ?*	yes	yes	yes	yes

* Twice the standard error of the difference is regarded as the 95% confidence interval on the difference. This is not strictly true, but the difference between such a calculation and a standard t-test for large-sample statistics is small.

For Ulupau data, the ratio of chlorophyll *c* to chlorophyll *a* is 0.22 for the first 2-week exposure and 0.27 for the 4-week exposure. The 6-week ratio increases abruptly to 0.45, reflecting the dominance of diatoms (and possibly some brown algae) on the overgrazed surfaces. The same ratios for HIMB are higher on the 2-week panels (0.43) and decrease on the 4- and 6-week exposures (0.32 and 0.23, respectively) as the settling communities experience a succession which produces more diverse growth of invertebrates and green, blue-green, and red algae.

Table 9. Ratios of amounts of chlorophyll *c*, carotenoids, and ash-free dry weights to chlorophyll *a* for the various panel exposure groups at Ulupau and HIMB. (Also included are the numbers of samples for which there was measurable chlorophyll *b*.)

	Chlorophyll c: *Chlorophyll* a	*Carotenoids:* *Chlorophyll* a	*Ash-free Dry Weight:* *Chlorophyll* a	*Number of Samples with* *Chlorophyll* b
2-week panels removed at 2 weeks				
Ulupau	0.22	0.59	2094	1
HIMB	0.43	0.53	2250	3
2-week panels removed at 4 weeks				
Ulupau	0.50	0.61	2500	4
HIMB	0.35	0.50	573	0
2-week panels removed at 6 weeks				
Ulupau	0.83	0.33	1667	4
HIMB	0.32	0.32	735	1
4-week panels removed at 4 weeks				
Ulupau	0.27	0.48	1035	0
HIMB	0.32	0.46	485	0
6-week panels removed at 6 weeks				
Ulupau	0.45	0.55	1125	7
HIMB	0.23	0.53	632	0

Chlorophyll *b* occurs most frequently (13 times) in 2-week exposure samples. None was measured in any 4-week exposure sample and, on the low-biomass 6-week exposures of Ulupau, 7 occurrences were recorded. Thus, in these data, chlorophyll *b* shows an association with low-biomass or early succession situations where competition for free substrata is low. Additionally, the high occurrence of this pigment on the heavily grazed 6-week exposure Ulupau panels suggests that the green algae species involved may be grazing-resistant forms.

Fish Growth

In previous experiments in the HIMB microcosms (Jokiel and Coles, 1974; Jokiel *et al.*, in press; Key, in preparation) grazing by fish has been noted as a process which is of great importance in determining overall community structure. For example, successful settlement of coral larvae in microcosm tanks was increased appreciably by the introduction of one or more herbivorous fish. Apparently, the grazing activity provides a greater amount of hard substratum which is free of detritus and living organisms and is thus preferred by the settling larvae. Grazing also favors the dominance of grazing-resistant forms of sessile organisms and may even result in the total exclusion of some species heavily

sought as food by grazers. In natural coral reef communities a variety of fish, crustaceans, and molluscs feed directly upon algal growth. At least a partial introduction of invertebrate grazers is insured in the microcosms because of the large number of planktonic life-stage forms that are provided from the source seawater. On the other hand, fish are very rarely recruited to the tanks via flow-through addition because of the specfic food requirements and fragility of the larvae and juveniles. At Ulupau 15 to 20 small (1 to 6 cm long) gobiids and eleotrids have been introduced to the tanks through the seawater system; most are still alive and growing normally. Flow-through introduction of fish to the HIMB tanks is a rare event because of the smaller sizes of screen openings and pump clearances.

To produce a range of grazing pressures on the settling surfaces of the microcosms, either 1 or 2 convict surgeon fish (*Acanthurus triostegus*) were added to each of two pairs of tanks after 2 weeks of flow-through. Two tanks were maintained without fish as controls.

Figure 25 shows weight curves for the fish at Ulupau. All fish in the Ulupau microcosms lost weight at a fairly constant rate. Five of six introduced fish died within 35 days. The most obvious cause of death was starvation. Availability of benthic algae, aside from the algae contained in the diatom–detritus mat, was very low at Ulupau, especially in the first 5 weeks of flow-through. This scarcity of food was attributed largely to the heavy influx of grazing sea hares in the 3-week through 6-week period.

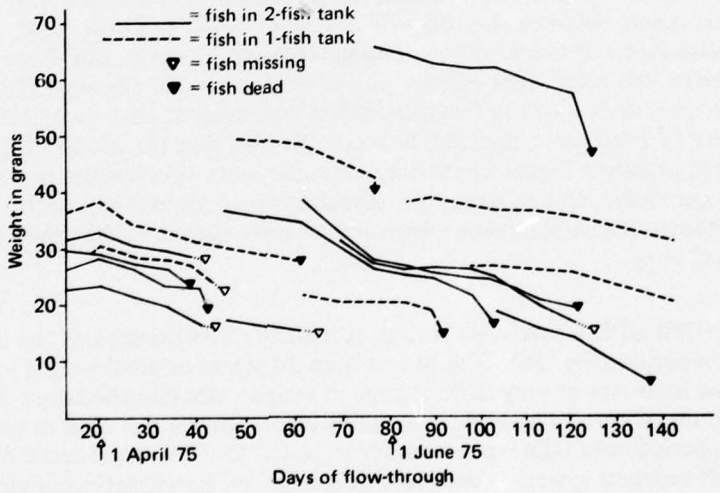


Figure 25. Weight of fish *(Acanthurus triostegus)* added to Ulupau tanks versus time.

A. triostegus is classed as a browser that bites and tears off bits of multicellular benthic algae without (or at least rarely) ingesting any of the inorganic substratum (Jones, 1968). This habit was verified in the Ulupau tank fish; the guts of three individuals were examined, and the only material found was an occasional small (1 to 2 mm) blade or spherule of brown or green algae. No fine sediment or diatoms were found, as would have been expected if they had been grazing on the diatom mat. Additionally, a substantial amount of plant material that the *A. triostegus* fed upon was apparently brought into the tanks in the supply water as small bits of free-floating macroalgae that were broken free from the algae-rich reef flat. In the tanks, the fish frequently could be seen swimming in and near the inlet water jet, picking out suspended algal pieces.

In general, fish added to the Ulupau tanks after 45 days of flow-through continued to lose weight at rapid rates, although compared to the group added first, they survived longer and lost greater amounts of weight (up to 70% of original weight) before dying. Through this period (from 45 to 130 days of flow-through) a new community of very close-cropped, film-like algal forms developed. Approximately 30% of the forms were crustose coralline red algae. Continued grazing by sea hares and fish (in the with-fish tanks) all but eliminated any filamentous or leafy algae, and very little diatom growth was observed. The longer survival periods for fish added to Ulupau tanks after 45 days may have been related to a slightly higher availability of macroalgal forms (possibly even the crustose algae) upon which the fish browsed.

For the 14-day to 45-day period, no significant differences in rate of weight loss were evident between the fish which were in 1-fish or 2-fish tanks. Three fish in the 1-fish tanks of the 45-day to 145-day period, however, did show slightly lower weight loss rates. This slightly slower weight loss for the single fish and the overall longer survival of the fish introduced later suggest that a small increase in availability of food was occurring. It is notable also that the group of fish introduced first showed a higher incidence of disease, with skin lesions, patches of hemorrhagic tissue, and skin pigment discolorations occurring in nearly 30% of the individuals. These symptoms were almost nonexistent in the group of fish introduced later.

At HIMB all fish survived through at least the first 60 days of the intercalibration period (Fig. 26). Fish of less than 30 grams original weight experienced slow increases or very little change in weight, whereas the larger fish (49 and 55 grams) showed steady decreases in weight. Only 4 fish died in the 84-day reporting period, and HIMB personnel have noted that the usual cause of death in the high-nutrient system is disease, not starvation. Examination of gut contents of *A. triostegus* maintained in the HIMB tanks has revealed moderate quantities of several species of macroalgae that grow in the tanks (Key, in preparation).

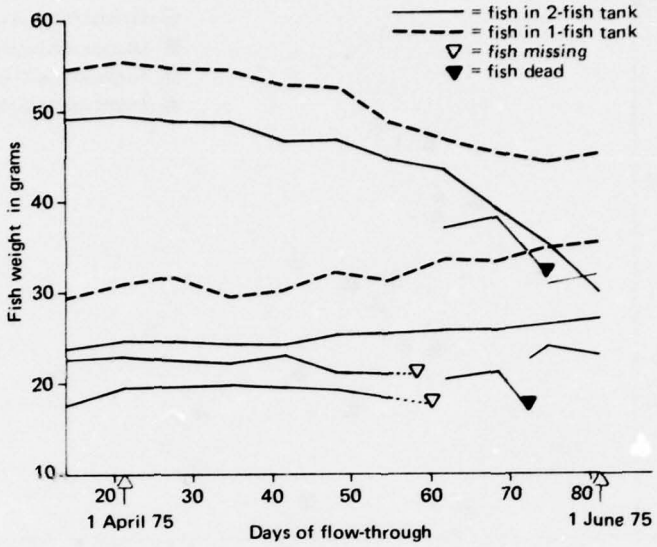


Figure 26. Weight of fish *(Acanthurus triostegus)* added to HIMB tanks versus time.

In Fig. 27, slopes (rates of weight change) for linear regression lines fitted to the data in Fig. 25 and 26 are plotted versus starting weights of fish. As expected, no discernible trends are present in data from Ulupau, where all fish lost weight at rapid rates regardless of starting weight or treatment (1 fish or 2 fish). However, as noted above, the HIMB fish of higher starting weights lost weight at higher rates than smaller individuals. Moreover, the two fish which were in 1-fish treatments fared better in their lower-competition environments, as shown by their higher (more positive) weight-change rates as compared to smaller individuals maintained in 2-fish treatments. Thus it is apparent that for Ulupau, where no fish gained weight through the first 140 days of flow-through, there was an insufficient quantity of food alga present in the microcosm biota to support even solitary *A. triostegus.** Gosline and Brock (1960) have reported field growth rates of approximately 2g/month for 10-cm-long *A. triostegus.* Although weight gains of 2.2 and 1.3 g/month were seen in two HIMB fish of 29.6 grams (10 cm long) and 22.5 grams (9.4 cm long), respectively, fish of larger size lost weight at rapid rates. For sustenance of solitary *A. triostegus,* the HIMB microcosms were marginally adequate in algal abundance.

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* This was also found to be true in more recent experiments where *A. triostegus* continued to lose weight in Ulupau tanks with over 7 months of accumulated algal growth.

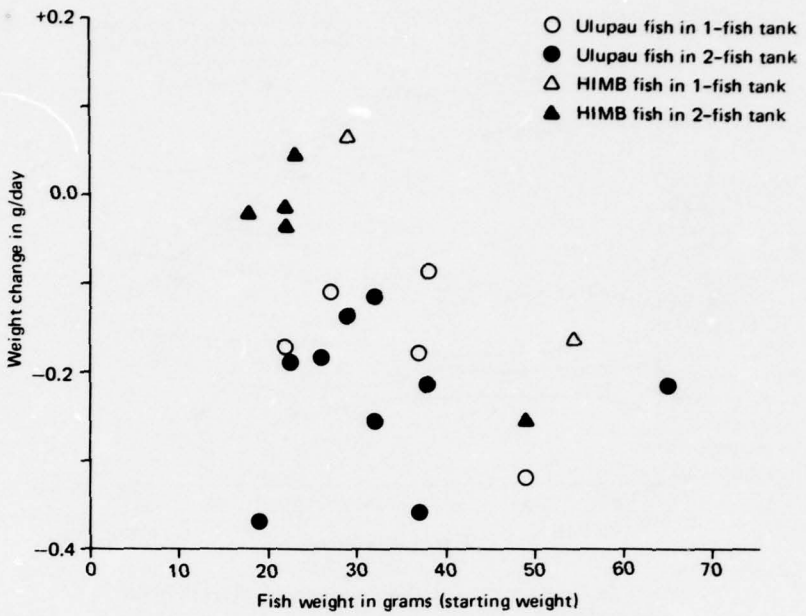

Figure 27. Rates of weight change versus starting weight for Ulupau and HIMB fish *(Acanthurus triostegus)*.

A species of parrot fish, *Scarus sordidus*, has been maintained in a set of six tanks (other than those involved in the intercalibration study) at Ulupau. In each of these tanks there are two *Scarus sordidus* (average weight 20.0 grams), two *A. triostegus* (average weight 13.3 grams), and one *Dasyllus albisella* (average weight 7.3 grams). Also present are two *Thalamita edwardsii* (portunid swimming crabs, average weight 51.9 grams), two *Holothuria atra* (sea cucumbers, not weighed), and 10 small colonies of the two coral species *Pocillopora damicornis* and *Montipora verrucosa.* The crabs in these tanks have preyed on and eliminated all sea hares. Without the grazing effect of the sea hares, the walls and bottoms of the tanks accumulate a 2- to 3- mm-thick diatom filamentous algae "fuzz". *A. triostegus* lose weight rapidly in these tanks, whereas over an average period of 60 days, the *S. sordidus* showed an average decline in weight of only 0.3 gram. In its natural habitat, *S. sordidus* is a herbivore which feeds mostly by scraping fine benthic algae that have overgrown the surfaces of dead corals. Hobson (1974) made this deduction after examining the stomach contents of several specimens from the island of Hawaii; the guts were "full of bits of algae, mixed with calcareous powder, organic slurry, and sand (proportions undetermined, but the algae constituted less than 20%)". Apparently *S. sordidus* is an opportunistic grazer that can utilize the diatom–detritus mat for at least a portion of its total diet.

The damsel fish, *D. albisella*, fared even better, showing average weight increases of 0.7 gram for three individuals which were in tanks for 60 days. This species is a planktivore that, according to Hobson, eats primarily larvaceans and copepods. In the tanks *D. albisella* very rarely pick food from the walls or bottom substrate; they were usually observed picking minute bits of food from the in-flowing water.

Unless macroalgae become more abundant in the tanks, *A. triostegus* will not be used in further Ulupau experiments. At present *S. sordidus* appears to be a herbivore more suited to the oceanic flora that develop in the Ulupau microcosms. *D. albisella* and other planktivorous and omnivorous species will be studied further as potential adaptable species to be used in investigations of other food chains.

Recruited Organisms

Some of the species and groups of organisms that have been observed in the two microcosm locales have been mentioned in previous sections. In general, the establishing communities approached the biological composition of the reef flats from which their supply waters are drawn. A particularly striking difference between the two systems was obvious in their rates of accumulating biomass, especially macroorganisms. A diverse assemblage of algae and sessile and motile invertebrates developed very rapidly in the HIMB tanks; this community was relatively unchanged after about 1 month of flow-through, with no appreciable changes in the proportions of dominant organisms. In contrast, the lower-diversity and lower-biomass microcosms at Ulupau were much slower in overall succession. Community structure at Ulupau was prone to change in "catastrophic" steps when irruptions of organisms such as sea hares and crabs occurred. The abnormal population growth and dominance by such species in the "low nutrient" microcosms were apparently a result of the low species diversity and lack of both intraspecific competition and normal predator–prey chains.

Table 10 lists organisms which have entered the microcosms of both facilities through the seawater systems. The species shown were recruited in larval or juvenile forms and have grown to at least young adult size in the tanks. Lists for both locales are far from complete, since identification of many species is still in process. At Ulupau, where the tanks are still in early succession, many more species will certainly appear as the communities mature. For HIMB the organism listing combines biological observations from previous long-term studies (Jokiel *et al.*, in press) as well as from the intercalibration run. The great abundance of species (estimated at several hundred) that occur in the HIMB system and the difficulties involved in the detailed taxonomic sorting of such communities have required that many organisms be grouped into higher phylogenic divisions. For future cost-effective analysis of such biological samples, as collected in the

course of microcosm and field studies, a Processing Center is being established at NUC/MEMO-Hawaii which will have the expertise and equipment necessary to accomplish the needed taxonomic analyses.

Table 10. Partial list of species or taxa of organisms which have entered tanks at both locales through the seawater systems and have grown to young adult size. Ulupau data from March through July 1975 only; HIMB data from late 1970 through July 1975.

Ulupau	*Hawaii Coastal Zone Data Bank I.D. Number**	*HIMB*
CHLOROPHYTA		CHLOROPHYTA
Chlorophyceae		Chlorophyceae
Ulotrichales		Ulotrichales
Ulvaceae		Ulvaceae
Enteromorpha sp.	0413160100	*Enteromorpha* sp.
	0413160201	*Ulva fasciata* Delile
Cladophorales		
Chadophoracea		
Cladophora sp.	0418010100	
		Siphonales
		Bryopsidaceae
	0421040100	*Bryopsis* spp.
	042104XXXX	*Pseudobryopsis oahuensis (?)*
CHRYSOPHYTA		CHRYSOPHYTA
Frustula sp.	07XXXXXXXX	
Streptothecia sp.	07XXXXXXXX	
Bacillariophyceae		Bacillariophyceae
Centrales		Centrales
Pennales		Pennales
Fragilaraceae		
Synedra sp.	0742010500	
Naviculaceae		Naviculaceae
Navicula sp.	0742070100	*Navicula* sp.
Amphiprora sp.	0742070900	
Nitzshiaceae		
Nitzchia sp.	0742080100	
Surirellaceae		
Surirella sp.	0742110100	
CYANOPHYTA		CYANOPHYTA
Schizophyceae		Schizophyceae
Oscillatoriales		Oscillatoriaceae
Oscillatoriales		Oscillatoraceae
Oscillatoraceae sp.	0213010000	Oscillatoraceae spp.
Lyngbya sp.	0213010200	
Oscillatoria sp.	0213010400	
Schizothrix sp.	0213010600	

* University of Hawaii computer listing. "X's" indicate that numbers have not yet been assig..ed; final "0's" indicate the level to which taxonomic identification has been performed.

(Contd)

Table 10. (Contd)

Ulupau	*Hawaii Coastal Zone Data Bank I.D. Number**	*HIMB*
Nostocaceae		
Nostoc sp.	0213030200	
Rivulariaceae		
Rivularia sp.	0213050400	
PHAEOPHYTA		PHAEOPHYTA
Phaeophyceae		Phaeophyceae
	1110000000	Phaeophyceae spp.
Ectocarpales		
Ectocarpaceae		
Ectocarpus sp.	1111010100	
Dictyotales		
Dictyotalaceae		
Dictyota sp.	1115010200	
Padina sp.	1115010500	
Zonaria sp.	1115010300	
Dictyosiphonales		Dictyosiphonales
Punctariaceae		Punctariaceae
Chnoospora sp.	1119040100	
	1119040400	*Rosenvingia* sp.
Sctyosiphonaceae		
Colpomenia sp.	1119050100	
Hydroclathrus sp.	1119050300	*Hydroclathrus* sp.
RHODOPHYTA		RHODOPHYTA
Rhodophyceae		Rhodophyceae
Nemalionales		
Chaetangiaceae		
Galaxaura sp.	1321050100	
Cryponemiales		Cryponemiales
Corallínaceae		Corallinaceae
Corallinaceae spp.	1323070000	
Porolithon onkodes Heydrich	1323071501	*Porolithon onkodes* Heydrich
Gracilariaceae		
Gracilaria sp.	1324150100	
Ceramiales		Ceramiales
Ceramiaceae		Ceramiaceae
	1326010700	*Spyridia* sp.
Centroceros sp.	1326011100	
Rhodomelaceae		Rhodomelaceae
Acanthophora spicifera (Vahl)	1326040101	*Acanthophora spicifera* (Vahl)
Laurencia sp.	1326040500	
Polysiphonia sp.	1326040700	*Polysiphonia* sp.

(Contd)

Table 10. (Contd)

Ulupau	*Hawaii Coastal Zone Data Bank I.D. Number*	*HIMB*
FORAMINIFERA		FORAMINIFERA
Foraminifera spp.	1852000000	Foraminifera spp.
		PORIFERA
		Demospongia
	3520000000	Demospongia spp.
		Poecilosclerida
		Amphilectidae
	3524030000	Amphilectidae sp.
		Hadromerida
		Suberitidae
	3527010100	*Terpios* sp.
	3527010102	*Terpios granulosa* Berquist
CNIDERIA		CNIDERIA
Anthozoa		Anthozoa
Actinaria		Actinaria
	3742000000	Actinaria sp.
		Boloceroididae
	3742010101	*Boloceroides mcmurrichi* (Kwietniewski)
	3742010201	*Bunadeopsis medusoides* (Fowler)
		Stoichactinidae
	3742200101	*Radianthus cookei* (Verrill)
Aiptasiidae		Aiptasiidae
Aiptasia pulchella (Carlgren)	3742340101	*Aiptasia pulchella* (Carlgren)
		Madreporaria
		Seritoporidae
	3746020101	*Pocillopora damicornis* (Linnaeus)
		Poritidae
	3747110110	*Porites compressa* (Dana)
CTENOPHORA		CTENOPHORA
Tentacula		Tentacula
Tentacula spp.	3910000000	
		Platyctenea
	3912010100	*Coeloplana* sp.
PLATHYHELMINTHES		PLATYHELMINTHES
		Turbellaria
	4130000000	Turbellaria sp.

(Contd)

Table 10. (Contd)

Ulupau	*Hawaii Coastal Zone Data Bank I.D. Number*	*HIMB*
		Acoela
	4131000000	Acoela sp.
		Polycladida
		Stylochidae
	4137020000	Stylochidae sp.
	4137020200	*Stylochus* sp.
NEMERTINEA		NEMERTINEA
Nemertinea spp.	4400000000	Nemertinea spp.
ANNELIDA		ANNELIDA
Polychaeta		Polychaeta
Polychaeta spp.	5500000000	
Errantia		Errantia
		Aphroditidae
	5511012201	*Paralepidonotus ampulliferus* (Grube)
		Amphinomidae
	5511040201	*Eurythoe complanata* (Pallas)
		Hesionidae
	5511130300	*Ophiodromus* sp.
		Syllidae
	5511140000	Syllidae sp.
Nereidae		Nereidae
Nereidae spp.	5511160000	Nereidae spp.
	5511160101	*Ceratonereis mirabilis* Kinberg
	5511160603	*Platynereis massiliensis* (Moovin-Jandon)
	5511160804	*Perinereis cultrifera* (Grube)
Sedentaria		Sedentaria
		Cirratulidae
	5521040000	Cirratulidae spp.
Chaetopteridae		Chaetopteridae
Chaetopteridae sp.	5521070200	
	5521070201	*Chaetopterus variopedatus* Renier
		Terebellidae
	5521220000	Terebellidae sp.
Sabellidae		Sabellidae
Branchiomma cingulata (Grube)	5521230201	*Branchiomma cingulata* (Grube)
Serpulidae		Serpulidae
Spirobinae spp.	5521240000	Spirobinae spp.
	5521240100	*Spirorbis* spp.

(Contd)

Table 10. (Contd)

Ulupau	*Hawaii Coastal Zone Data Bank I.D. Number*	*HIMB*
Serpulinae		Serpulinae
Serpulinae spp.	5521244000	
	5521244200	*Hydroides norvegica* Gunnerus
	5521244201	*Hydroides crucigera* Morch
	5521244204	*Hydroides lunulifera* Claparede
	5521244701	*Pomatoleios kraussi* (Baird)
ARTHROPODA		ARTHROPODA
Insecta		
Coleoptera		
Hydrophilidae		
Tropisternus sp.	63XXXXXXXX	
Crustacea		Crustacea
		Thoracica
		Balanidae
	6451090100	*Balanus* sp.
	6451090102	*Balanus amphitrite* Darwin
	6451090103	*Balanus eburneus* Gould
	6451090106	*Balanus trigonus* Darwin
		Mysidacea
	6466120000	Mysidae sp.
	646612XXXX	*Anisomysis incisa* Tattersall
		Tanaidacea
		Apseudidae
	6468010100	*Apseudes* spp.
Isopoda		Isopoda
Isopoda spp.	6471000000	Isopoda spp.
Amphipoda		Amphipoda
Amphipoda spp.	6473000000	Amphipoda spp.
		Caprellidae
	6475010000	Caprellidae sp.
Decapoda: Caridea		Decapoda: Caridea
		Palaemonidae
	6483320000	Palaemonidae sp.
	6483320902	*Palaemonella tenuipes* Dana
	6483324100	*Palaemon* sp.
	6483324101	*Palaemon debilis* Dana

(Contd)

Table 10. (Contd)

Ulupau	*Hawaii Coastal Zone Data Bank I.D. Number*	*HIMB*
		Gnathophyllidae
	6483330301	*Gnathophyllum faciolatum* Stimpson
Alpheidae		Alpheidae
Alpheus spp.	6483411000	*Alpheus* spp.
	6483411001	*Alpheus lobidens polynesica* (Heller)
	6483411003	*Alpheus rapax* Fabricius
Alpheus pacificus Dana	6483411033	
	6483411200	*Synalpheus* sp.
	6483411201	*Synalpheus pachymeris* Coutiere
	6483411209	*Synalpheus paraneomeris* Coutiere
Hippolytidae		
Hippolysmata kukenthali (Deman)	6483430103	
		Processidae
	6483440101	*Processa processa* (Bate)
		Decapoda: Anomura
		Diogenidae
	6487130300	*Calcinus* spp.
	6487130302	*Calcinus latens* Randall
Decapoda: Brachyura		Decapoda: Brachyura
Dromiidae		Dromiidae
Dromiidae sp.	6488060000	Dromiidae sp.
Portunidae		Portunidae
Portunidae spp.	6488312000	
	6488313300	*Thalamita* sp.
	6488313301	*Thalamita integra* Dana
	6488313303	*Thalamita admete* (Herbst) Alcock
	6488313313	*Thalamita crenata* Latreille
	6488315101	*Podophthalmus vigil* (Fabricius)
Grapsidae		
Percnon planissimum (Herbst)	6488323103	
Xanthidae		Xanthidae
Xanthidae spp.	6488330000	Xanthidae spp.
	6488330801	*Madaeus simplex* A. Milne Edwards

(Contd)

Table 10. (Contd)

Ulupau	*Hawaii Coastal Zone Data Bank I.D. Number*	*HIMB*
	6488331404	*Carpilodes bellus* (Dana)
	6488331900	*Etisus* sp.
	6488331901	*Etisus electra* (Herbst)
	6488331905	*Etisus laevimanus* Randall
	6488332201	*Panopeus pacificus* Edmondson
	6488332301	*Phymodius ungulatus* (Milne Edwards)
	6488332400	*Chlorodiella* spp.
	6488335806	*Pilumnus oahuensis* (Edmondson)
Stomatopoda		Stomatopoda
Squillidae		Squillidae
Pseudosquilla sp.	6489010100	
	6489010201	*Gonodactylus falcatus* (Forsskal)
MOLLUSCA		MOLLUSCA
		Amphineura
		Ischnochitonidae
	7011010101	*Acanthochiton viridis* Pease
Gastropoda: Streptoneura		Gastropoda: Streptoneura
Streptoneura spp.	7020000000	Streptoneura spp.
Archaeogastropoda		Archaeogastropoda
		Fissurellidae
	7021050301	*Diodora octagona* (Reeve)
		Trochidae
	7021120101	*Trochus histrio* Reeve
Turbinidae		Turbinidae
Turbo sp.	7021140100	
	7021140101	*Turbo argyrostoma* Linnaeus
Neritidae		Neritidae
Nerita sp.	7021210100	
	7021210201	*Theodoxus neglectus* (Pease)
Mesogastropoda		Mesogastropoda
Vermetidae		Vermetidae
Vermetidae sp.	7022430000	Vermetidae spp.
	7022430100	*Dendropoma* sp.
Dendropoma platypus Morch	7022430101	
	7022430400	*Vermetus* sp.

(Contd)

Table 10. (Contd)

Ulupau	*Hawaii Coastal Zone Data Bank I.D. Number*	*HIMB*
Cerithiidae		Cerithiidae
	7022540100	*Cerithium* sp.
Cerithium nesioticum (Pilsberg & Vanatta)	7022540103	
	7022540202	*Bittum zebrum* (Kiener)
		Epitoniidae
	7022590100	*Epitonium* sp.
Strombiidae		
Strombus maculatus Sowerby	7022680101	
		Hipponicidae
	7022720101	*Hipponix pilosus* (Deshayes)
		Calyptraeidae
	7022750101	*Crucibulum spinosum* (Sowerby)
Gastropoda: Euthyneura		Gastropoda: Euthyneura
		Entomotaeniata
		Pyramidellidae
	7031010200	*Odostomia* sp.
	7031010202	*Odostomia pupu* Pilsberg
	7031010208	*Odostomia indica* (Melvill)
Cephalaspidea		Cephalaspidea
Bullidae		Bullidae
Bulla adamsi Menke	7033210101	*Bulla adamsi* Menke
Atyidae		Atyidae
	7033220100	*Atys* spp.
	7033220101	*Atys semistriata* Pease
Haminea sp.	7033220300	*Haminea* sp.
Aglajidae		
Chelidonura hirundundina (Quoy & Gaimard)	7033080402	
		Sacoglossa
		Elysiidae
	703906000	*Elysiidae* sp.
Anaspidea		Anaspidea
Aplysiidae		Aplysiidae
Aplysia parvula (Guilding)	7041020101	
Aplysia juliana (Quoy & Gaimard)	7041020105	*Aplysia juliana* (Quoy & Gaimard)
Stylocheilus longicauda (Quoy & Gaimard)	7041022101	*Stylocheilus longicauda* (Quoy & Gaimard)

(Contd)

Table 10. (Contd)

Ulupau	*Hawaii Coastal Zone Data Bank I.D. Number*	*HIMB*
Dolabrifera dolabrifera (Rang)	7041024101	*Dolabrifera dolabrifera* (Rang)
	7041026100	*Dolabella* sp.
Dolabella auricularia (Lightfoot)	7041026102	
Nudibranchia		Nudibranchia
Melibe sp.	7044XXXXXX	
Hexabranchus sp.	7044060100	
	7044680102	*Phestilla sibogae* Bergh
Bivalvia: Pteriomorpha		Bivalvia: Pteriomorpha
Mytiloida		Mytiloida
		Mytilidae
	7054010301	*Branchidontes cerebristriatus* (Conrad)
	7054010300	*Hormomya* sp.
Pinnidae		
Pinna muricata Linnaeus	7054050101	
		Pterioida
		Pteriidae
	7055010101	*Pinctada margaratifera* (Linnaeus)
		Ostreidae
	7055060100	*Crassostrea* sp.
	7055060201	*Ostrea sandvicensis* Sowerby
	7055060203	*Ostrea hanleyana* Sowerby
		Anomiidae
	7055200101	*Anomia nobilis* Reeve
		Bivalvia: Heterodonta
		Veneroida
		Veneridae
	7056530101	*Lioconcha hieroglyphica* (Conrad)
	7056530201	*Periglypta reticulata* (Linnaeus)
	7056530301	*Venerupis philippinarum* Adams & Reeve
		Tellinidae
	7056620201	*Quadrans palatam* (Iredale)
	7056620301	*Angulus hawaiensis* (Dall, Bartsch & Rehder)

(Contd)

Table 10. (Contd)

Ulupau	*Hawaii Coastal Zone Data Bank I.D. Number*	*HIMB*
		Myoida
		Hiatellidae
	7057100101	*Hiatella hawaiensis* (Dall, Bartsch & Rehder)
ECHINODERMATA		ECHINODERMATA
Asterozoa		Asterozoa
Gnathophiuroida		Gnathophiuroida
	7840000000	Gnathophiuroida sp.
Ophiacomidae		Ophiactidae
	7841010102	*Ophiactis savignyi* (Muller & Troschel)
Ophiocoma sp.	7842030100	
Echinoidea		
Diadematoida		
Diadematidae		
Echinothrix sp.	7852010100	
Echinometridae		
Echinometra sp.	7852240200	
Holothuroidea		Holothuroidea
Aspidochirota		Aspidochirota
Stichopodidae		
Stichopus horrens Selenka	7871020101	
Holothuridae		Holothuridae
Holothuridae spp.	7871030300	
Holothuria atra Jager	7871030302	
	7871030317	*Holothuria monocaria* (Lesson)
Apoda		Apoda
Synaptidae		Synaptidae
Synaptidae sp.	7878010000	
	7878010201	*Opheodesoma spectabilis* Fisher
		Chiridotidae
	7878020101	*Chiridota hawaiiensis* Fisher
CHORDATA: UROCHORDATA		CHORDATA: UROCHORDATA
Ascidiacea		Ascidiacea
	8310000000	Ascidiacea spp.
Enterogona		Enterogona
Phallusidae		Phallusidae

(Contd)

Table 10. (Contd)

Ulupau	*Hawaii Coastal Zone Data Bank I.D. Number*	*HIMB*
Ascidia spp.	8311040100	*Ascidia* spp.
	8311040101	*Ascidia sydneiensis* Stimson
	8311040102	*Ascidia interrupta* Heller
Ascidia corelloides (Van Name)	8311040103	
		Didemnidae
	8311120000	Didemnidae sp.
	8311120300	*Didemnum* sp.
		Polyclinidae
	8311130201	*Polyclinum constellatum* Savigny
		Botryllidae
	831201XXXX	Botryllidae sp.
	8312010000	*Botryloides* spp.
		Pleurogona
		Styelidae
	8312020100	*Styela* sp.
		Tethyidae
	8312030000	Tethyidae sp.
	8312030200	*Herdmania* sp.
CHORDATA: VERTEBRATA		CHORDATA: VERTEBRATA
Osteichthys		Osteichthys
Anguilliformes		
Muraenidae		
Gymnothorax petelli (Bleeker)	8522050611	
Scorpaeniformes		Scorpaeniformes
Blenniidae		Blenniidae
	8555340301	*Entomacrodus marmoratus* (Bennett)
Istiblennius zebra (Vaillant & Sauvage)	8555340401	
Gobiidae		Gobiidae
Bathygobius fuscus (Ruppell)	8555600802	
Eleotridae sp.	8555605100	
	8555605301	*Asterropteryx semipunctatus* Ruppell

The chronology of first occurrences of visible individuals of the more dominant invertebrates at Ulupau is given in Table 11. Included also are occurrences of major algae species; for microscopic algae the appearances of visible aggregations were used (such as filamentous tufts, globules, and mats). For invertebrates

Table 11. Chronology of first appearances for dominant invertebrates at Ulupau.

Weeks of Flow-Through	*Algae*	*Cnidarians*	*Crustaceans*	*Annelids*	*Molluscs*	*Echinoderms*
	diatoms					
1+	cyanophytes					
	filaments and tufts of algae					
2+						
	green tufts					
3+					*Stylocheilus longicauda*	
			amphipods			
4+						
					Cerithium nesioticum	
5+	calcareous algae					
		Aiptasia pulchella			bubble shells	
	brownish algae tufts				*Cheilidonura hirundindina*	
6+						Synaptidae sp.
					other nudibranchs	
					Aplysia parvula	
7+	*Ectocarpus*				*Aplysia juliana*	
	Valonia					
	Laurencia					
8+			portunids			
9+					*Dolabrifera dolabrifera*	
10+			*Percnon planissimum*			
11+					*Dolabella auricularia*	
					vermetids	
12+				*Hydroides* sp.		
			alpheids	spirorbids		
	Padina					
13+	*Hydroclathrus*					tunicates
	Colpomenia					(clear)
14+			*Hippolysmata kukenthali*		*Strombus maculatus*	
					Cerithium sinensis	
15+			*Gnathophyllum faciolatum*			
					Cypraea caputserpentis	
16+						
17+						*Echinothrix* sp.
						Stichopus horrens
18+						
					Pinna muricata	
19+						

at Ulupau the intervals to first occurrences are probably representative of the periods of time that it takes those species to grow from larval size to the young adult sizes at which they were seen. A similar plot for HIMB organisms has not been presented because of the rapid influx and growth rates of those complex communities. The occurrence of species in those tanks would probably be more heavily affected by competition and predation.

A number of common coral species obtained from both inshore (bay) and offshore (oceanic) areas have been maintained in the tanks at both sites. These species are:

Cyphastrea occellina
Fungia scuteria
Montipora flabellata
Montipora verrilli
Montipora verrocosa
Montipora patula
Pavona explanulata
Pavona varians
Pocillopora damicornis
Pocillopora meandrina
Porites compressa
Porites lobata
Psammocora stellata
Tubastrea aurea

At HIMB colonies of these corals have been maintained with no significant mortality for periods of more than 1 year. Moreover, several species have planulated and successfully reproduced in the tanks (Jokiel *et al.*, in press). A parasitic nudibranch, *Phestilla sibogae*, occasionally infests the *Porites* spp. coral heads in the bay tanks. This corallivore is prolific, and in a matter of weeks a group of individuals are capable of grazing all living polyps from several large *Porites* coral heads. If *Porites* specimens are inspected and picked clean of *Phestilla* when brought in from the field, and thereafter are maintained in uncrowded conditions in the tanks, then infestations of the parasite can usually be avoided. *Phestilla* have not been found on any *Porites* brought in from the offshore areas, nor have they infected any offshore corals supplied with oceanic (Ulupau) water.

The outlet boxes at Ulupau have developed thick growths of macroalgae which are higher in quantity and species composition than the algal growths occurring in the tanks. Six of these 30-cm-by-30-cm-area boxes contain automatic siphons and are filled to a depth of 20 cm and emptied to a depth of 5 cm every 100 seconds. The other six boxes have no siphons; incoming water spills out of 20-cm-high standpipes, and water levels are maintained at 5 cm by overflow drain fittings. Flow rates through all boxes are 10–12 l/min.

All algae listed in Table 10 were represented in the boxes, with *Padina, Acanthophora, Cladophora, Dictyota, Laurencia, Gracilaria, Chnoospora, Sargassum, Ulva,* and calcareous algae being the more conspicuous species. All of these species are common on the reef flat at Ulupau. Growth in the boxes without siphons was predominantly in masses of more fragile filamentous forms. In the boxes with siphons, where water motion was more intense, species of more durable form were prevalent.

Sea hares (*Stylocheilus*) colonized the six outlet boxes with siphons concurrently with the occurrence of sea hares in the six calibration microcosms which supplied water to those boxes. The sea hares grazed the walls and bottoms of the boxes, feeding mostly on diatom material and short filamentous algae. With light levels, water quality, and grazing being nearly the same in the tanks and the outlet boxes, water agitation appears to be the important factor which, in the outlet boxes, produced abundant growth of reef flat algae.

CONCLUSIONS

Completion of the Ulupau facility has produced a unique environmental research system consisting of 24 identical continuous flow-through microcosm tanks. Located at nearly opposite ends of an environmental gradient, the two sets of microcosms (of 12 tanks each) offer great potential for the study of interrelating factors that affect the chemical, physical, and biological character of coastal marine waters.

From a mechanical standpoint, the Ulupau facility is an outstanding success. The seawater distribution system, flow-rate control devices, and monitoring apparatus have shown high reliability with minimal maintenance requirements.

The Ulupau microcosm system exhibits different abiotic properties from HIMB, although these differences are not exactly as expected. The most consistent difference is in the PO_4 content of the inlet waters, with the HIMB mean levels of that nutrient being approximately twice those of Ulupau. Differences in NO_3 and NH_3 are neither as large nor as consistent as anticipated. Continued nutrient sampling will be necessary at both facilities to determine the nature and magnitudes of seasonal nutrient variations.

Biologically the two systems responded as expected. At HIMB the bay source water produced a diverse community of rapid succession which, relative to Ulupau, manifested itself in far higher productivities, production-to-respiration ratios, and plant-to-animal biomass ratios. Porifera, Actinaria, Bryozoa, Annelida, Pelecypoda, Cirripeda, and Tunicata were the major taxa that represented the bulk of the settling fauna at HIMB. At Ulupau organisms from those groups were uncommon or absent.

For the range of environmental conditions encountered at Ulupau and HIMB, biological responses on the panel surfaces over short periods are apparently too slow for the extensive seral development of fouling communities. Thus settling panels exposed for less than 4 weeks do not appear likely to be of great quantitative use as environmental indicators.

Quantitatively some of the panel parameters revealed meaningful patterns. Higher ratios of chlorophyll *a* to ash-free dry weights (suggesting relatively more plant biomass to animal biomass) were consistently linked with higher productivity-to-respiration ratios. Moreover, chlorophyll *a* values (hence, total plants)

were almost immediately stable at HIMB, reflecting the rapid growth of algae in that system. At Ulupau, chlorophyll *a* values (and total biomass) increased initially and then fell drastically, following closely the obvious events (increase of algal growth under conditions of no grazing followed by overgrazing) in those microcosms.

The growth rates of the herbivorous fish that were added to the microcosms were directly related to the levels of primary productivity (that is, the algal-standing crop) of the two facilities. *Acanthurus* at Ulupau invariably lost weight and most died, whereas at HIMB fish of smaller size (less than 40 grams) experienced slow increases in weight. In future experiments, herbivorous species of broader feeding habits (such as *Scarus*) will be used in the microcosms.

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