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DESIGN AND CONSTRUCTION OF A SALTWATER ENVIRONMENT SIMULATOR



**FEDERAL WATER
QUALITY
ADMINISTRATION
NORTHWEST REGION**

**PACIFIC NORTHWEST
WATER LABORATORY**

CORVALLIS, OREGON

DESIGN AND CONSTRUCTION OF A
SALTWATER ENVIRONMENT SIMULATOR

by

Waldemar A. DeBen

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United States Department of the Interior
Federal Water Pollution Control Administration, Northwest Region
Pacific Northwest Water Laboratory
200 Southwest Thirty-fifth Street
Corvallis, Oregon 97330

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FEDERAL WATER POLLUTION CONTROL ADMINISTRATION

NORTHWEST REGION, PORTLAND, OREGON

James L. Agee, Regional Director

PACIFIC NORTHWEST WATER LABORATORY

CORVALLIS, OREGON

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INTRODUCTION

Field studies complemented by laboratory work are essential in evaluating conditions which may be detrimental to a biotic community. Water quality criteria must take into account long-term, sublethal exposure effects on test organisms.

Laboratory efforts to provide information on factors that influence organism abundance and state of health require simulation and control of natural environmental parameters. Most systems in current use involve static water bioassays or recirculating systems. Burke and Ferguson (1968) state that:

...objectionable features of static tests include a decline in concentration of toxicants during the exposure period by uptake by the experimental organisms, its adsorption onto the container or other surfaces, and its chemical alteration....Furthermore, accumulation of waste products, reduction of dissolved oxygen supply, and growth of microbial populations may produce an undesirable test environment.

Poole (1966), while rearing Cancer magister zoeae under static water conditions, encountered heavy mortality due to microbes. Burdick (1967) believed that continuous-flow assays are ideal for either long or short-term tests and that this type of system may be used to establish the test animal's physiological limits of resistance.

Described in this report is an experimental, continuous-flow bioassay apparatus designed to utilize and stabilize salt water derived from an estuarine source. The prototype was constructed

to provide a single-test supply of up to eight liters of test water per minute at four levels of selected temperature and a near-constant concentration of any selected salinity or dissolved oxygen. Provision for the introduction of measured quantities of specific materials for test completed the basic system.

This apparatus, called the "Saltwater Environment Simulator" (or "Simulator"), was intended for use in evaluating the effects of pollutants and natural environmental changes on marine and estuarine animals. It was constructed by personnel of the National Coastal Pollution Research Program of the Federal Water Pollution Control Administration located in the Marine Science Center at Newport, Oregon.

SIMULATOR

Description

The Saltwater Environment Simulator consists of three basic units: (1) a water storage tank to ensure the availability of a selected high salinity water during operation, plus a filter to eliminate suspended material from entering the rest of the system; (2) a tower to support a salt-freshwater mixing tank, a head tank, and aeration, degassing, and heat exchange equipment; and (3) a table for heating chambers, flow meters, animal holding tanks, and temperature monitoring thermocouples. A side view diagram of the tank, tower, and table is shown in Figure 1 and a top view diagram in Figure 2.

The saltwater supply for the Marine Science Center is pumped from an intake located about two feet off the bottom of Yaquina Bay at depths of approximately 20 to 30 feet, depending on tidal stage, and distributed throughout the laboratory in polyvinyl chloride (PVC) pipe. High salinity water enters the 600-gallon epoxy-painted metal storage tank from the laboratory line via a motor valve. This quantity of water is sufficient to sustain the system over any ordinary low salinity period during a single tidal cycle. Suspended material settling in the tank is removed periodically by using a small centrifugal pump attached to a piece of suction hose and pipe.



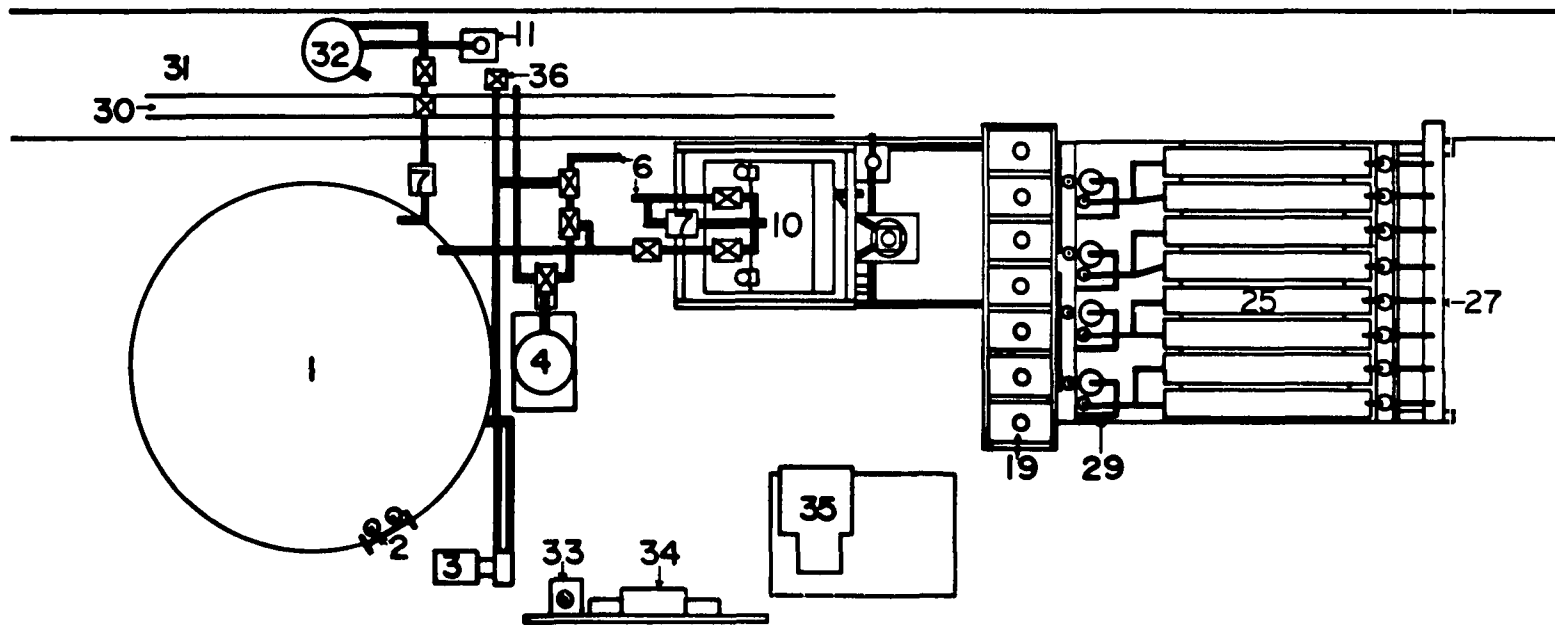


Fig 2. Top view of water storage tank, tank tower, and test table.

KEY TO NUMBERS FOR FIGURES 1 AND 2

<u>Number</u>	<u>Item(s)</u>
1	Saltwater storage tank
2	Float switch panel
3	Flex-liner water pump
4	Saltwater filter
5	Freshwater filter
6	Freshwater line
7	Electric valve actuator
8	Saltwater valve
9	Stirring motor
10	Salinity adjustment tank
11	Salinity sensor
12	Wastewater bucket
13	Head tank
14	Saltwater return line
15	Gas extraction tube
16	Immersion cooling unit
17	Antifreeze-water bath
18	Centrifugal pumps
19	5-gallon plastic containers
20	Water manifold
21	Line to manifold
22	Heating chamber
23	Heater and thermoregulator
24	Flow meter
25	Animal test tank
26	Thermocouple container
27	Plastic drain pipe
28	Test table
29	Oxygen sampling tube
30	Laboratory saltwater line
31	Wastewater trough
32	Constant head bucket
33	Timing motor
34	Circuit breaker box and electrical panel
35	Temperature strip chart recorder
36	Pressure relief valve

Stored seawater is forced through a filter for removal of the remaining suspended matter and to the salinity adjustment tank with a seal-less, neoprene flex-liner pump (5 gpm rating). A safety float switch stops the pump when water level drops within a few inches above the storage tank water outlet. Salt water enters the salinity mixing tank at the rate of one gallon per minute. PVC ball valves and a valve actuator control the rate of saltwater flow and that of the fresh water used for dilution and introduced through an auxiliary line. Both are discharged through a common line near the bottom of the tank. Excess water is returned to storage via an overflow bypass line.

The mixing tank has a 29-gallon capacity and is of polyethylene construction. Two variable-speed motor stirrers with three-bladed polyethylene stirring rods attached accomplish mixing. Water temperature is held nearly constant in the tank with a mercury thermoregulator-glass jacketed immersion heater combination. Two discharge pipes are located at the brim of the tank - one to transport water to a salinity sensor and to a 24-gallon head tank; the other to serve as the overflow line for returning excess diluted water to the storage tank.

Salinity-adjusted water passes by gravity flow from the head tank into the top of a glass oxygen "scrubbing" tube through a distribution pipe at the bottom. At this point the flow is divided. One portion is pumped through a stainless steel heat exchanger for

cooling, and the other is pumped to heating chambers. Water to be heated and the cooled water are directed into identical heating chambers through separate manifolds at the rear of the test table. Hand-adjusted PVC valves are used to regulate final flow rates through the chambers and to the animal test tanks. Temperature-adjusted water leaving the chambers passes through double-ball flow meters to facilitate flow regulation. A flow rate of 500 ml/min/chamber usually was used during "checkout" procedures. Maximum capacity is 1 L/min/chamber.

Water leaving the test chambers flows into eight-ounce polyethylene jars, each of which contains a thermocouple for the purpose of obtaining a continuous record of experimental temperatures. All discharge water is piped to a drain trough in the floor.

Salinity

Accurate control of salinity is one of the most important functions in the development of a flowing seawater system. Laboratory animals subjected to fluctuating salinities undergo ionic, osmoregulatory, or other physiological changes that can influence experimental results. Fluctuating tidal levels and freshwater inflows complicate the control problem.

A salinity sensing device composed of a hydrometer, photocell-relay, and electric valve actuator was fabricated to continuously monitor the incoming estuarine water and to regulate the flow of

a desired salinity into the storage tank. The device is designed to accept all water of selected or higher salinities and to reject or close the intake valve to water of lesser specific gravity. A float switch controls the opening and closing of the valve when a continuous supply of high-salinity water is available.

A cut-away view of the salinity sensing unit, less hydrometer, is shown in Figure 3. Water coming from the seawater system enters near the base of the sensor through PVC pipe. The body and tubing arrangement is fabricated from PVC pipe and plastic hose. Two openings (windows) are located in the upper portion of the unit and are enclosed in a movable machined plastic collar which functions as a water spillway. Mounted above the "windows" in two short pieces of pipe is a grid-type cadmium sulfide photocell-relay and a 2.5 volt flashlight bulb. A small volume of incoming seawater is introduced near the base of the sensing unit and allowed to circulate around the hydrometer to the overflow spillway.

Construction methods and materials for the hydrometer are based on the formula used by Thayer and Redmond (1969) to calculate hydrometer stem diameter relative to body displacement. Discarded standard seawater ampules are used to construct the hydrometer body. A black plastic vane is attached to the stem of the hydrometer to prevent light from energizing the photocell during periods of salinity of lower than desired level.

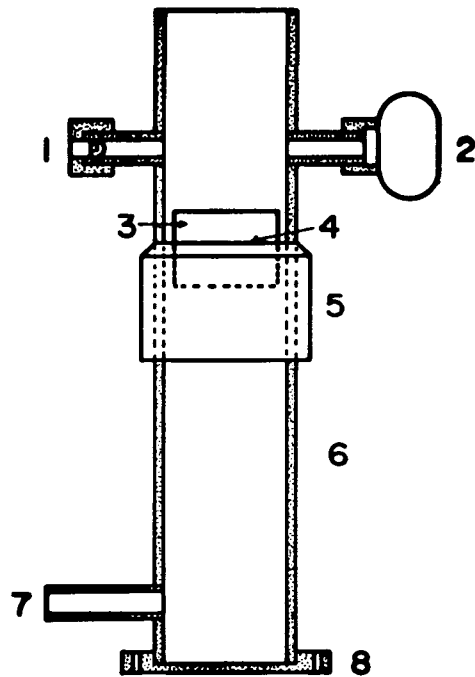


Fig.3 Cut-away view of salinity sensor.

KEY TO NUMBERS FOR FIGURE 3

<u>Number</u>	<u>Item(s)</u>
1	Flashlight bulb
2	Photo cell-relay unit
3	"Window"
4	Water spillway
5	Movable plastic collar
6	Cell body
7	Water entrance pipe
8	Cell base

Five precalibrated hydrometers were made for salinities of 5, 10, 15, 20, and 25 o/oo. Seawater of greater specific gravity than the control hydrometer displaces the hydrometer upward to raise the vane out of the light path, thereby energizing the photocell and valve actuator. A 25 o/oo hydrometer was used for test operation of simulator systems. A diagram of the electrical system which controls the entrance of high-salinity water into the storage tank is shown in Figure 4.

Parallel platinum wire guides, mounted only in the mixing tank sensor, are used to limit lateral movement of the hydrometer and keep it in the light path at all times. The sensor fabricated for the salinity adjustment tank is similar to the previously-mentioned unit except that a Clairex CL-704* cadmium sulfide photocell with a narrow (about 1/16-inch) light-sensitive band is used. By using this type of photocell, slight changes in salinity will activate or deactivate the motor valve circuit, therefore maintaining a near-constant salinity.

Dilution and salinity control are accomplished with one of two adjustment procedures. A test salinity in the 5 to 15 o/oo range is rapidly obtained by manual adjustment of the freshwater ball valve until a concentration 2 o/oo higher than desired is reached. The valve actuator is allowed to automatically add the additional fresh water required and to maintain the selected concentration. Dilution to the 15 to 20 o/oo range is efficiently achieved without

* Mention of product or company name does not constitute endorsement by the Federal Water Pollution Control Administration.

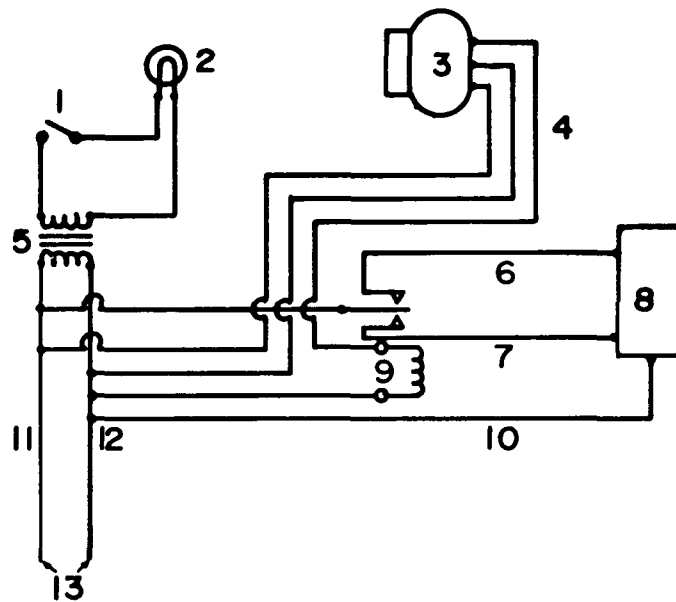


Fig. 4. Electrical circuit for control of sea water storage.

KEY TO NUMBERS FOR FIGURE 4

<u>Number</u>	<u>Item(s)</u>
1	Float switch
2	Sensor flashlight bulb
3	Grid-type photocell-relay
4	Switched line
5	2.5 volt filament transformer
6	Opening lead
7	Closing lead
8	Motor valve
9	Relay coil
10	Common line
11	Energized line
12	Neutral line
13	110-volt AC line

manual assistance. A separate hydrometer, calibrated to the desired salinity, is required for each experiment. Hydrometers were constructed for each 5 o/oo interval.

The electrical diagram for the adjustment tank salinity sensor and monitoring unit is shown in Figure 5. The salinity sensor activates the valve actuator to allow only small increments of fresh water to enter the mixing tank at any one time. This system maintains limits within ± 1 o/oo of the desired salinity as measured by an RS5-3 electrodeless induction salinometer (Industrial Instruments) mounted in the head tank. This salinity control is accomplished by using the following components:

(1) Modified actuator valve. A small change in salinity (the photocell responds to 0.3 o/oo difference), with subsequent movement of the hydrometer, will cause activation or deactivation of the photocell, which ultimately controls the direction of motor valve rotation. Modification of rotation direction on the electric valve actuator is accomplished by using two limit switches, one which controls clockwise and the other counterclockwise rotation. In the 90 degree rotation of the ball valve there are 12 stopping points from open to closed positions. Both limit switches are under the control of the photocell timer circuit.

(2) Photocell timer circuit. This circuit controls the quantity and duration of freshwater inflow into the salinity adjustment tank. A one rpm (1/250 hp) timing motor drives a circular cam on

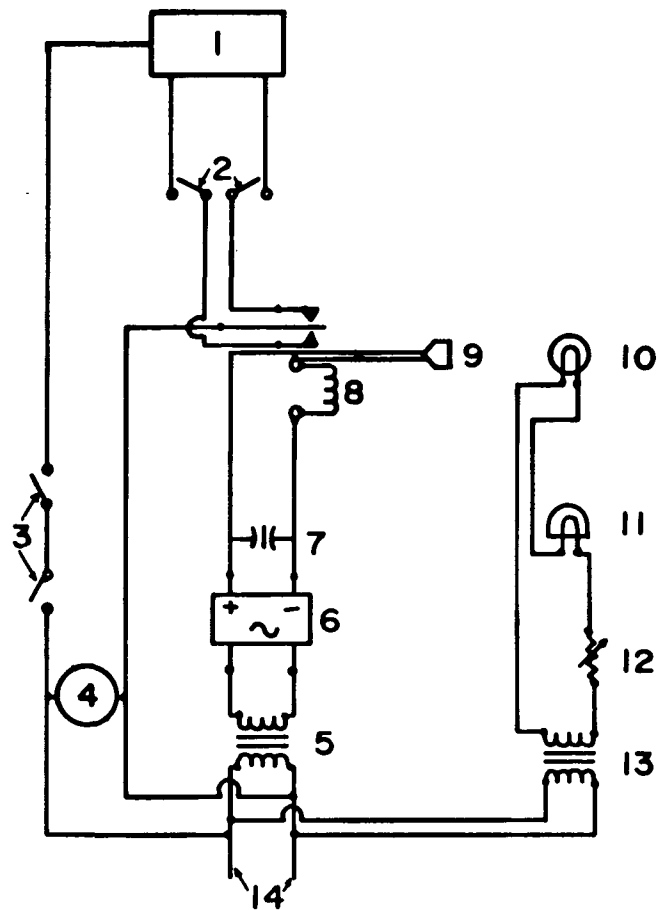


Fig. 5 Electrical circuit for salinity adjustment tank sensor and monitor.

KEY TO NUMBERS FOR FIGURE 5

<u>Number</u>	<u>Item(s)</u>
1	Motor valve
2	Limit switches
3	Roller switches
4	1 RPM timing motor
5	45-volt transformer
6	Full wave rectifier
7	50 mfd capacitor
8	10,000 ohm relay coil
9	Clairex photocell
10	Photocell activator light
11	Pilot light
12	250 ohm potentiometer
13	2.5 volt filament transformer
14	110-volt AC line

which ride two roller microswitches. Current passing between the microswitches energizes the valve actuator, only if needed to maintain desired salinity, for about 0.5 second every one-half minute and either opens or closes the valve slightly, depending upon the salinity.

All components except the valve actuator, photocell activator light, and photocell are mounted on an electrical panel near the base of the tower.

Temperature

Stable water temperatures within any laboratory system are a necessity, since temperature variations alter the oxygen consumption rate, growth, enzyme activity, toxicant uptake, and other metabolic functions of aquatic animals. The temperature control capacity of the simulator is designed to produce water temperatures similar to those found in the environment, i.e., in areas of heated effluent discharge or under the cool conditions of winter and upwelling.

The heat exchanger assembled for cooling consists of a 50-foot length of 1/2-inch ID, 20 ga., #316 stainless steel tubing with a surface area of approximately six square feet. It is formed into a series of bends (resembling a trombone slide) and immersed into a water-tight plywood box (21" x 17" x 25 1/4") covered with a sheet of glass wool--aluminum foil insulation. This inner box, which contains coolant, fits into a slightly-larger box to reduce

heat transfer from the surrounding air. Thirty-five gallons of a water-ethylene glycol mixture (4:1) is used in cooling.

Inserted through the top cover of the plywood cooling tank is a 1/3 horsepower immersion cooling unit with a cold adjustment range of from 2 to 20°C. This is adequate to lower the temperature of the tank liquid to within a few degrees of the lower limit. The coolant mixture is circulated at the rate of 4,500 gallons per hour throughout the tank and over the compressor's stainless steel cooling coils by a 1/12 horsepower circulator motor mounted in the lower section of the cooling unit. Test water, cooled in circulating through the 50-foot stainless steel coil, is transported to two of the four heat adjustment chambers in insulated (glass wool-aluminum foil) polyethylene tubing.

The four heating chambers, mounted in the rear of the testing table, are constructed of four-inch PVC pipe 24 inches long, fitted with threaded caps. The caps are drilled to accommodate rubber stoppers which hold a one-half-inch bottom entrance pipe and a similar size discharge pipe, mercury thermoregulator, and immersion heater at the top. The chambers are covered with a layer of glass wool-aluminum foil insulation to restrict the loss of heat. A pre-set mercury thermoregulator (sensitivity to $\pm 0.05^{\circ}\text{F}$ temperature change), heavy duty mercury plunger relay, and an interchangeable 750 or 1000 watt glass-jacketed immersion heater make up the temperature control circuit for each chamber. Water temperatures

remained within $\pm 0.3^{\circ}\text{C}$ of the desired setting for the duration of checkout procedures as recorded on a twelve-channel strip chart temperature recorder.

This recorder is sensitive to temperature ranges of from 0 to 60°C . The strip chart is graduated in 0.5°C divisions and hourly intervals for easy reading. Each of the units' 12 thermocouples is compacted ceramic insulated and sheathed in stainless steel. Temperatures monitored, in addition to the animal test chamber water, included air temperature over these test tanks, water in the salinity adjustment and water cooling tanks, and water in a tank used to acclimate animals to laboratory conditions.

Oxygen

Water is aerated to saturation or near-saturation with compressed air in the head tank before it passes to the top of a glass degassing column (3 1/2" x 4'). A constant head of water is maintained in the column by means of an overflow pipe inserted in a machined PVC collar fitted at the top. As the water flows downward the dissolved oxygen is displaced to the degree desired by a rising shower of controlled nitrogen bubbles. Bottled nitrogen gas is dispensed through a two-stage regulator attached by tubing to a fritted dispersion disc placed in the bottom of the degassing column. In tests using the diffuser stone alone, bubbles tended to rise along one side of the column. A circular sieve plate was

constructed from 1/4-inch plexiglass and several inches of glass Raschig rings were placed on the sieve to eliminate this problem. This dispersed the bubbles more or less evenly and removed the dissolved oxygen more efficiently. Preliminary tests indicate that dissolved oxygen levels can be maintained within limits of ± 0.3 mg. dissolved oxygen per liter.

Water samples for dissolved oxygen determinations were taken from a tube inserted in the line between the flowmeters and the animal test chambers. The azide modification of the Winkler method was used.

Water Filtration System

Salt Water

Since different species of test animals have to be held for different lengths of time during a test period, provisions were made to ensure that adequate food could be supplied. Fish and crustaceans were held in filtered flowing water with food introduced into the test chambers. Non-filtered water could be routed around the filter and pumped directly to the salinity adjustment tank for filter feeding bivalve mollusks.

A cut-away view of the saltwater filter showing piping, valves, and direction of saltwater flow (as indicated by arrows) is shown in Figure 6. Filter body construction is based on a formula developed by Neptune Microfloc, Inc., and filled with filter media consisting

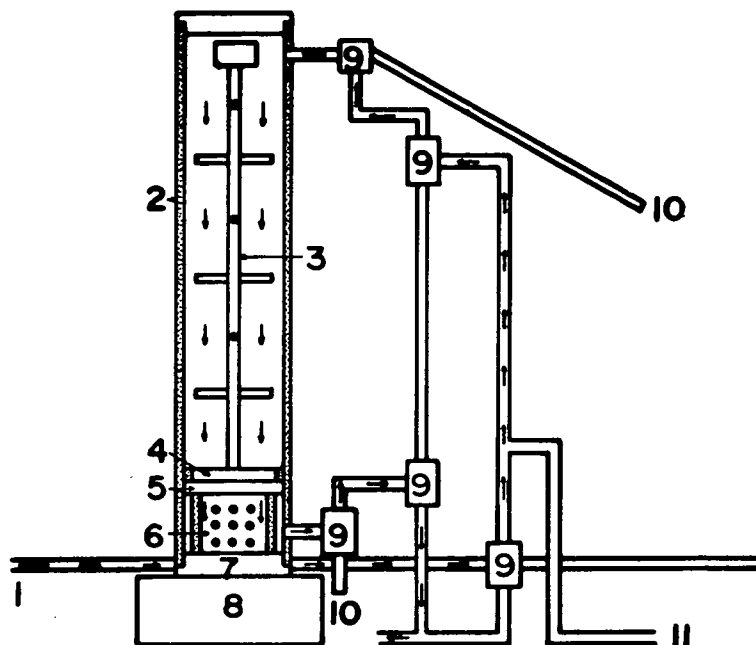


Fig. 6. Cut-away view of salt water filter body. Arrows show path of salt water flow.

KEY TO NUMBERS FOR FIGURE 6

<u>Number</u>	<u>Item(s)</u>
1	Saltwater line from pump
2	Filter body
3	Media retainer
4	Support ring
5	Carborundum stone
6	Spacer ring
7	Plexiglass end piece
8	Support stand
9	Multiport ball valve
10	Wastewater discharge pipe
11	Freshwater line

of fine and coarse garnet, ground carborundum, and anthracite coal. Analysis of particle size using a Coulter Counter revealed no particles larger than 10 microns in water leaving the filter. The filter can maintain water of this quality at a pumping rate of 5 gallons per minute.

Water enters through a multiport valve on the upper side of the filter, passes down through the filtering media, and a porous carborundum stone, which retains the filter media, and out of the filter through a multiport valve to the salinity adjustment tank. During periods of freshwater runoff, with its accompanying large load of suspended material, the filter media has to be cleaned about every twelve hours. As the volume of entrapped material increases, so does the amount of pressure exerted on the walls of the filter. To prevent rupture of the filter body, the intake pipe is fitted with a pressure relief valve, set to discharge water into a trough when the pressure reaches 30 pounds per square inch.

In the cleaning or "backflushing" procedure, fresh water enters the bottom of the filter body through a multiport valve, passes through the carborundum stone and resuspends the sediment and detritus trapped by the filter media. This dirty water then passes through a multiport valve on the upper side of the filter body and is discharged into a waste-water trough. The filter media is displaced upward but not discharged, due to the retentive action of the media retainer.

Fresh Water

Fresh water used in dilution is piped by the Seal Rock Water District to the Marine Science Center from various small streams. Analysis of water for free chlorine showed none present. Upon entering the building, the water passes through an activated charcoal filter with a filtering rate of 35 gallons per minute. A small charcoal filter is mounted in the waterline as a precautionary measure.

Additional Equipment Used in the Simulator

Eight animal test chambers were constructed for initial simulator trials. Each held approximately six gallons (40" x 6" x 9") and was made from 3/16-inch black plexiglass. Water entered near the bottom of one end and was discharged near the top of the other. The bottom is sloped to form a shallow "vee" to facilitate collection of feces and uneaten food. This waste material is flushed out by a drain in the bottom end of the tank. Black plexiglass was chosen to reduce any adverse effect in the behavior or physiology of test animals caused by external stimuli. Circulation of water through the tank, at a rate of 500 ml per minute, was studied by using neutral red dye. The result showed a well-mixed dye throughout the chamber, indicating that an introduced toxicant would probably follow this pattern. Temperature measurements within the animal tank showed no gradients present.

Test materials can be metered into the system at a point immediately before entering the test tanks. A ten-vein metering pump with a 0.12 to 5.25 ml per minute flow, depending on tubing size, was used for this purpose. Mounted above the metering pump are seven five-gallon plastic carboys, each connected to the pump by flexible plastic tubing. The carboys can contain several types and concentrations of test materials. The diluent water and test material were found to be adequately mixed in either a small plexi-glass chamber with a series of baffle plates or a small (250 ml) filter flask.

CONCLUSION AND SUMMARY

An experimental apparatus to control the environmental parameters of salinity, temperature, and dissolved oxygen was designed and constructed for the purpose of obtaining information on various physiological processes in test animals and to complement field studies carried out on estuarine fauna.

Salinity was controlled and adjusted by using a sensing unit employing a photocell-hydrometer-motor valve actuator system which maintained the salinity at ± 1 o/oo of the desired test salinity.

Temperature regulation was accomplished by using a stainless steel heat exchanger immersed in a cold antifreeze bath for cooling, while water was heated by using glass-jacketed immersion heaters working in conjunction with sensitive mercury thermoregulators. Water temperatures were maintained within $\pm 0.3^{\circ}\text{C}$ of the desired setting.

Oxygen levels were maintained by metering nitrogen gas through a two-stage regulator to a diffuser stone, mounted in the base of a large glass tube, which released an ascending shower of bubbles and decreased the amount of dissolved oxygen in the water, which entered at saturation or near-saturation levels at the top of the tube. Oxygen levels of ± 0.3 mg. per liter could be maintained in the system.

Preliminary tests have shown that this apparatus will provide controlled environmental conditions to better define some of the

physiological requirements of test animals living under estuarine conditions that can be related to ecological field studies. The capability of inducing conditions of "stress," either by changing one or more of the parameters or by introduction of a specific material, can help to establish water quality standards in the saltwater environment.

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