LARVAL SETTING AND SURVIVAL OF YOUNG OYSTERS, *Ostrea lurida* Carp., UNDER LABORATORY CONDITIONS

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INTRODUCTION

Studies concerning the setting of free swimming oyster larvae, the mortalities and the growth of young artificially reared oysters under controlled conditions were conducted at the Yaquina Bay Fisheries Laboratory from June 1954 to January 1955. This was a continuation of previous investigations performed at the laboratory directed toward the successful artificial propagation of larvae from the native oyster, *Ostrea lurida* Carpenter.

The need for production of large quantities of "seed", or young oysters, has long been realized by oyster culturists and biologists. The case is particularly well presented at Yaquina Bay, Oregon, where early exploitàtions followed by excessive siltation and the lack of adequate cultch have resulted in insufficient annual spatfall of seed oysters necessary to sustain a profitable native oyster fishery. Because of its fine edible qualities there has been a much greater demand for this oyster from Yaquina Bay than has been produced for many years. As a result larval rearing studies have been instituted for the past several years at the Yaquina Bay Fisheries Laboratory of Oregon State College.

Professor Roland E. Dimick studied the status of the native oyster at Yaquina Bay from 1939 to 1946, seeking possibilities for rehabilitation of the exhausted natural beds and founding the groundwork for future investigations. Robert W. Morris began laboratory research in 1946, developing a culture media for the successful rearing of larval food organisms and devising a filter for changing water in rearing containers without loss of larvae. These efforts were succeeded by those of Eugene P. Haydu in 1949 and 1950 who developed further rearing techniques and first raised oyster larvae to the setting stage at the laboratory. In 1951, Gerald M. Watson made preliminary studies of some marine bacteria as oyster larval foods and Charles E. Warren examined possibilities of the flagellate, Bodo lens (O. F. Muller), as food for oyster larvae, each obtaining limited spatfalls from their efforts. Wilbur P. Breese described the total processes of the developed rearing techniques in 1953 while Nicholas Pasquale, in a companion thesis, studied the varied effects of concrete and wooden tanks as rearing containers. These two latter workers obtained further success in rearing larvae to the seed stage, the possibilities of producing commercial quantities of spat being clearly indicated from their investigations.

The material herein is therefore an addition to previous investigations at the Yaquina Bay Oyster Laboratory. With the rearing of larvae to the spat stage in 250 gallon tanks under controlled conditions proving an adequate reality, the selection of a suitable spat collecting material for research purposes was first needed. These experimental spat collectors or "cultch" should provide uniform areas of surface for larval attachment, be of common physical composition, and also contribute possibilities for experimentation with various combinations of surface textures and surface positions. Glass plates cut into four inch by twelve inch rectangles were chosen. Upon attachment of the free mature swimming larvae, notations were made as to location of spat upon the cultch, types of surfaces selected, relative abundance of settlement at all levels in the tank and the set per unit area. The spat thus obtained were subjected to various processes designed to discover methods of holding and conditioning prior to placement in Yaquina Bay. Growth, survivals and mortalities were closely observed and factors detrimental or advantageous to spat development were noted.

It is not necessary to report in detail the previous investigations mentioned in the introduction since the reports are on file at the Department of Fish and Game Management at Oregon State College. Consequently, when a detailed explanation concerning a rearing process is needed, the reader will be referred to the appropriate work.

BIOLOGY OF THE NATIVE OYSTER

A condensed summary of the biology of the native oyster, _Ostrea lurida_ Carpenter, is considered desirable here as the knowledge of certain unique features is indispensable to the understanding of this paper. This oyster can be readily identified from exotic species of oysters now on the Pacific Coast by the smaller size of the adult and a clearly outlined, uncolored muscle scar on the inner surface of each valve (5, p.42). A comparison of the ostia or gill perforations of the native oyster and those of introduced species will show those of the native to be somewhat larger in size.

The native oyster inhabits several protected harbor areas along the Pacific Coast of North America from Queen Charlotte Sound, British Columbia to the Tiajuana estuary, south of San Diego Bay, California. At the present time this species is found in Oregon at Yaquina Bay, Lincoln County, and in limited numbers at Netarts Bay, Tillamook County.

A span of three to five years is necessary for this oyster to reach a market size of $1\frac{1}{2}$ to 2 inches; however sexual maturity is often reached in one year. Being a protandric hermaphrodite, the young adult first possesses male sexual organs and alternates for the rest of its life as male and female.

During the female phase the eggs pass into the exhalent chamber, through the gill ostia, and into the inhalent chember where they are fertilized by sperm mixed with sea water. "Spawning" refers to the release of the eggs from the ovaries and sperm from the gonads with the resulting fertilization. The native oyster retains its developing eggs within the inhalent chamber for a period of incubation, hence it is called larviparous. Each female produces between 250,000 and 300,000 eggs which are capable of nearly 100 per cent fertilization (10, p.614). The larvae develop for a period of 9 or 10 days (8, p.536), reaching a reported length of 180-185 microns (8, p.470). At this stage expulsion into the bay occurs and the larvae become free swimming, a process called "swarming".

In the male phase clusters of sperm or "sperm balls" are discharged from the gonads. These pass into the exhalent chamber and hence to the surrounding waters. The matrix enclosing the sperm ball then disintegrates and releases approximately 250 to 2,000 or more individual sperm.

Spawning in Yaquina Bay, while beginning as early as April in some years, usually commences in late May or June and continues until August in varying degrees of intensity. A few gravid or reproductively ripe oysters are found in late September. The variation in lengths of the spawning season and abundance of gravid adults depends primarily upon prevailing temperatures. Thirty days is generally the length of the free swimming period in Yaquina Bay, depending upon both water temperatures and available food organisms which aid or hinder the development of the larvae. During this stage the larvae grow to a length of 260 to 320 microns, coming to rest if a suitable hard surface is available (8, pp.470-471). This process of attachment is known as "setting", the settled larvae is called an oyster "spat", and the object to which the larvae is cemented is "cultch". Here the spat undergoes a metamorphosis and adapts itself to a sedentary life for growth into an adult. "Seed" oysters are spat of a size suitable for transplanting operations.

Growth of the spat is dependent primarily upon the presence of adequate food substances in the seawater. During feeding the action of cilia on the gills cause a flow of water through the inhalent chamber. These cilia select suitable food particles, engulf them in mucous, and pass them toward the mouth. It is generally believed that oysters benefit from but a few of the many micro-organisms available in sea water as food.

From the time of swarming, oyster larvae are subject to high mortalities caused by a variety of environmental conditions such as tidal currents, varying salinities, lack of food, inimical temperatures, etc. Small sized spat are subjected to losses from the effects of siltation and other undetermined causes. Consequently, only a small number of free swimming oyster larvae eventually reach market size.

REVIEW OF PREVIOUS WORK

Throughout the history of oyster culture investigators have collected data concerning spatfall, growth, and mortality of oysters under natural conditions. However, only in recent years has information been made available on the artificial rearing of oyster larvae under controlled conditions. Since the natural habits of oyster larvae and spat may show a direct relationship to laboratory studies they are thus mentioned here.

Spatfall

Spatfalls occur following the free swimming period. The larvae, reaching a certain size and encountering suitable environmental conditions, will come to rest upon a clean stationary object. Types of cultch in use for obtaining commercial quantities of spat vary widely throughout the world. Included are brush and crates of tile in France, bundles of rope in Italy, bamboo stocks in Japan, piles of rock in Australia, limed tile in England, and containers filled with shell in the United States (6, pp.203-204). These spat collectors present surfaces of wide diversity in texture and chemical composition, in addition to being held in numerous positions in relation to current flow.

In 1913 Stafford used pieces of window glass to obtain spat of the Eastern oyster. These smooth glass strips were placed in crocks, a dozen per crock, and planted adjacent to oyster beds. In spite of a heavy growth of marine organisms some attachment of spat occurred.

The work of A. E. Hopkins (1935) concerning natural attachment of native oyster larvae in Puget Sound is noteble. Using a spat collector of cardboard covered with concrete, he found that about 115 times as many spat settled on lower horizontal surfaces than on upper horizontal surfaces. This was presumably because larvae seem to attach most abundantly to lower surfaces and not to the possibility that survival on such surfaces may have been greater. On plane glass Hopkins observed about 100 times as many spat caught on under horizontal as on vertical surfaces, while almost none were attaching to upper horizontal surfaces. This investigator also stated, "...spat settle most abundantly on rough surfaces, ---plates parallel to tidal flow catch more spat than those perpendicular to the current (more larval bearing water passing the surface),---and where the water is highly turbulent the larvae would frequently be turned over and have more opportunity to attach to upper surfaces".

Bonnot (1937) also believed that native oyster larvae under normal conditions set more abundantly on the under side of horizontal surfaces. Yet he found, using cement coated boards as collectors in Humboldt Bay, California, that 73 per cent of attached native oyster spat settled on the upper surface. This was explained as resulting from swirling water which carried larvae into all positions, the pull of gravity causing them to fall on the surface below. Concerning the density of natural spatfall, Bonnot found an average of 98 spat per square inch on upper surfaces and 36 on lower surfaces, averaging 67 spat per square inch.

Prytherch (1928) says of the setting of the Eastern oyster, "The heaviest setting was found near the bottom and on the lee side of the collectors, presumably because of the eddies created by them".

An explanatory description showing the relationship between the normal swimming position of the native oyster larvae and its position of attachment was given by Hopkins, (7 , p.86):

> "The foot, by means of which the larvae holds on to a surface before cementing itself, is adjacent to the velum at the open borders of the valves. The velum is a rather flattened, ciliated swimming organ and must support the weight of the body proper and the larval valves. In this case the velum would naturally be maintained above the body, and the foot would be pointing upward. As one watches the larvae swimming in a dish it is noticeable that they characteristically swim in this position, though swaying back and forth. Presumably in

> nature, as the larvae are carried along with the current, the foot may cling to a surface with which it comes in contact. This, most frequently, would be an under surface, and the more nearly horizontal the surface the more likely the contact."

Records at Yaquina Bay by Dimick and Long (1931) constantly indicated that natural spatfall on strings of Japanese and Eastern oyster shell occurred most abundantly on the lower surfaces. Using 144 shell in their experiments and counting only smooth surfaces, an average of 3.77 spat per shell were obtained. These shells were suspended in the bay near the oyster beds approximately one month prior to examination.

Growth

It is generally believed that a wide variation in growth of spat exists and that this variation is controlled by environmental conditions. With larvae being liberated over a period of three or four months, a certain variation at the end of the first growing season is expected. However, on neighboring beds spat often show striking differences in rate of development and reach marketable sizes at different ages.

According to Stafford (1939) the shell of the young native spat is longer than high, resembling the larvae. He stated also:

> "At first the addition of spat shell to the anterior and posterior angles is liable to be greater than that of the rounded part below, but when about 1 mm. in height the proportions become reversed and from this time forward the shell grows faster below and at the posterior-inferior angle".

Growth records from Yaquina Bay by Dimick, Eglund, and Long (1941) indicated that natural spat grow faster in upper portions of the bay than in lower regions. Average lengths of samples from three stations were 11.25, 6.38, and 5.62 mm. These measurements were taken approximately two months after the termination of the spatting season.

Mortality

A single native oyster is capable of producing 250,000 to 300,000 young (8, p.459). This high potential is probably related to the destruction encountered during development.

Orton (1935), in discussing mortality of the European oyster, stated:

> "By far the greatest mortality occurs in the larval stage. There is also considerable loss among spat with a decreasing percentage in brood and later ages to the adult stage. Enemies, mud accumulation, and other unfavorable environmental conditions are probably the most frequent causes of death in spat and brood, although much loss from non-apparent causes also occurs. The amount of mortality in spat and brood from all causes is very variable but may range very high."

Obtaining an average set of 67 native oyster spat per square inch, Bonnot (1937) found that in eight months the set was reduced to 14.5 per square inch. This was a mortality of 78 per cent. The greatest losses occurred on upper sides of the spat collectors, for while the initial population was greatest on upper surfaces the unequal mortalities resulted in the number of surviving spat on all surfaces being approximately the same.

Dimick, Eglund, and Long (1941) obtained a comparable mortality of 79 per cent on spat from Yaquina Bay between August 1939 and May 1940. These investigators also checked three wire bags filled with shell under more controlled conditions. Two bags, checked at monthly intervals and cleaned free of silt, showed a mortality of 29.7 per cent and 38.3 per cent. A third bag, permitted to be covered with mud, showed a mortality of 74.1 per cent. These bags were in the bay for approximately seven months.

With the Eastern oyster in Long Island Sound, Loosanoff and Engle (1940) found a high mortality of spat on wire bag collectors. This mortality ranged from 86 to 100 per cent for a seven week period.

EXPERIMENTS WITH SPATFALLS

Comparison of Spatfall on Old and New Shell

Shells of the Pacific oyster, *Crassostrea gigas* (Thunberg), were utilized as cultch in many of the investigations reported here. The shell was fairly large in size and easily obtained from piles located on the banks of Yaquina Bay near Oysterville. Having been discarded many years ago, these piles were of undetermined age. During this time the action of the elements have subjected the exposed shells to a continuous weathering process.

Orton (1935) stated that shell is composed largely of calcium carbonate, with a small percentage of organic matter, and that there are three distinct layers. The outer layer or periostracum, is composed of conchyolin, the inner layer of prismatic crystalline calcareous material, and the middle layers of pearly macre or calciteostracum. Since weathering processes may have affected the old shells by rendering them less attractive to mature oyster larvae, it was therefore desirable to ascertain any differences which might have existed between old and new shells as cultch.

Shells for the experiments were selected for uniformity in shape and surface area. The new shells were recent discards, having been placed on the mud flats near the laboratory one and a half months previously. All shells were washed and wire brushed prior to introduction into a wooden rearing tank for attachment of larvae. Non-toxic nylon leader material was used to suspend the shells individually, with the smooth side down, several inches below the surface of the water in the rearing tanks.

Two tanks were used for obtaining spat on old and new shell. In the first series, twenty shells, ten old and ten new, were placed in tank 3 on August 26. The suspended shells were arranged in horizontal line with the old shells in one group followed by new shells. Bubbles from an aeration device were directed so as to rise close to the new shell, which may have lessened settlement thereon due to the creation of minor water currents. On September 20, ten old shells and ten new shells were placed in tank 5, the twenty shells being staggered, first one old shell and then one new, with no air bubbles rising in close proximity. This second series was designed to correct a possible error resulting from aeration close to the new shell in the first experiment. All shell were removed from each tank after seven days and the spat enumerated, table 1.

In tank 3, by far the greatest majority of spat were attached to old shells, with 90.2 per cent compared to but

Table 1. Comparison of spatfalls obtained on old and new oyster shell, September, 1954.

	Old Shell		New Shell	
	Tank no. 3	Tank no. 5	Tank no. 3	Tank no. 5
	3,775	44	14	20
	1,813	12	20	37
	125	49	114	24
	3,035	29	74	158
	9,720	6	175	38
	1,365	58	52	7
	183	11	150	169
	557	19	544	19
	6,129	14	44	3
	1,737	65	1,902	32
Totals	28,439	307	3,089	507
Per Cent	90.2	37.7	9.8	62.3
Average	3,844	31	309	51

9.8 per cent on new shells. On the other hand, in tank 5, the new shells had 62.3 and old shells 37.7 per cent of the total spat.

Consideration must be given to the fact that tank 3 presented the highest total spatfall of all rearing attempts during the summer of 1954, while tank 5 had only a moderate spatfall and most of the set perished following attachment. Aeration may have decreased spatfall on new shell in tank 3 as nearby rising air bubbles sent out minor currents in the water. However, some protection was provided by the concave inner surfaces of the shells, which may have minimized this hypothetical disturbance.

A wide variation in numbers of spat per individual shell was noted from a range of 9,820 to 6 on old shell and 1,902 to 3 on new shell. This variation indicated that free swimming larvae found certain shells more attractive than others. Why some shells were chosen and others ignored by the larvae could not definitely be determined. Because of the conflicting data and a wide variation in settlement upon individual shells, no definite conclusions could be drawn concerning the value of old shell in comparison to new shell as cultch in rearing containers.

Graphite Mortality

In order to locate individual spat later in the investigations, some were encircled on the glass slides by graphite pencil markings. In addition other graphite markings were made on the cultch. These marks retained definition in water for satisfactory periods of time. To avoid error it was essential to determine if graphite particles had a detrimental effect upon the young spat.

Early in the summer several clean shells of the Pacific oyster were suspended individually, smooth side down, in a wooden larval rearing tank. Once an adequate set was obtained five shells were selected for the experiment. Each shell was divided into halves by a scratched line down the center, this line being made by a diamond point glass etching pencil. Spat on one side of the line were encircled by scratching while those opposite were graphite encircled. A total of 240 spat were thus identified, 120 by graphite and 120 by scratching. The shells were then returned to the tank from which they had originated for a nine-day period. Using a binocular microscope, the spat were checked for indications of mortality as shown in table 2.

Table 2. Data on mortalities of spat encircled by graphite and etched markings during a nine-day period, July 27 to August 5, 1954.

Shell Designation	Graphite Encircled		Scratched Encircled	
	No. Spat Per Shell	Mortality	No. Spat Per Shell	Mortality
A	39	4	26	5
B	2	2	16	0
C	14	0	42	4
D	22	4	8	1
E	41	4	28	1
Totals	120	14	120	11

Chi-square value: .0232 Critical range: X2 3.84

Graphite encircled spat and scratch encircled spat appeared to sustain approximately the same amount of mortality. At a 5 per cent significance level and with 1 degree of freedom, the chi-square value of .0232 was considerably smaller than 3.84. Statistically, evidence was insufficient to prove a definite difference in mortality between graphite encircled spat and scratch encircled spat. Young spat often extended their shell across the graphite and scratched lines, showing no apparent regard for either type of markings. The mortality shown was considered to be the expected occurrence of deaths since a certain percentage of spat usually perished soon after settlement, as shown in early growth-mortality studies.

Exposure Mortality

During the course of the investigations it was necessary to subject newly attached spat to considerable handling. Enumeration by means of a microscope or hand lens necessitated that the young oysters be retained from the tanks for varying periods of time, resulting in a possible mortality from exposure. The main factor to be considered was evaporation of moisture from the spat. This mortality could be avoided by determining the length of time which spat could safely be removed from water.

Six clean Pacific oyster shells were suspended individually, smooth side down, in wooden larval rearing tank until a minimum of 50 spat had attached to each smooth inner surface. Identification during future examinations was assured by encircling each young spat with a graphite pencil mark. The shells were then exposed

Table 3. Mortality data of air exposed oyster spat at 24.5° C., July 28, 1954.

Shell	No. of Spat	Exposure Period (Minutes)	Mortality at one week	Mortality at five weeks	Per Cent Mortality at five weeks
C	50	10	2	11	22.0
D	50	15	5	11	22.0
E	50	20	10	12	24.0
F	50	30	13	16	32.0
G	50	45	28	41	82.0
H	50	60	33	34	68.0

to a constant air temperature of 24.5° C. for varying periods of time as indicated in table 3.

When the young spat were first checked for mortalities at the end of one week some dead individuals undoubtedly escaped detection as growth occurring in all cases was slight. At the termination of five weeks, all spat surviving exposure had showed definite growth while those perishing showed no growth and, in many instances, had disappeared entirely from the experimental shells.

An expected amount of natural mortality of 22 to 32 per cent was shown by shells C, D, E and F. The living spat on shells G and H were in a "pocket" formed by the concave inner surface. This pocket remained moist longer than the slanting sides of the shell, thus resulting in a protected area.

From this experiment the indications were that a thirty minute exposure period was apparently a fairly safe limit for removal of spat from the rearing tanks. Young spat were capable of retaining moisture for some time after the surrounding surface had become dry. Older spat could withstand longer periods of exposure than younger spat. This experiment was conducted in an enclosed room protected from the drying effects of wind and sun, the shell being handled carefully to minimize crushing.

SPATFALL

Native oyster larvae have been successfully reared to the setting stage at the Yaquina Bay Fisheries Laboratory by previous investigators. A full account of the techniques developed and employed at this station was given by Breese (1953) in a thesis entitled, "Rearing of the native Pacific Coast oyster larvae, *Ostrea lurida* Carp., under controlled laboratory conditions". In general, identical techniques were utilized in obtaining spatfall for the following experimental data.

Cultch used in these investigations were plates of common window glass and weathered shells of the Pacific oyster. Glass plates were primarily used to facilitate accurate spat counts per unit of area on either rough or smooth surfaces while oyster shells were employed for comparison purposes. In previous years shells were mainly utilized in rearing attempts at the laboratory.

Preparation of Glass

Sheets of glass, of varied sizes and shapes, were first cut into rectangles of 4 inches by 12 inches. "Frosting" on one side was accomplished by a hand-made sand blaster, constructed from a box, and an air hose attached to a compressor. The box was assembled with 12 inch by 12 inch sections of one quarter inch plywood.

The bottom of the box had a small hole into which was inserted a three inch piece of one fourth inch copper tubing, which, in turn, was perforated with four small holes. A three and one half inch square opening was cut into the top and the box partially filled with coarse sand.

In operation the nozzle of the hose from the compressor was inserted into the copper tubing from below. The glass slide was placed over the hole on top of the box. By turning on the compressed air and impelling it upward into the box, grains of sand trickling through the punctured tubing were caught and hurled upward with sufficient force to chip the glass. While this method of frosting the glass proved satisfactory, the process was somewhat time consuming. Numerous halts were needed, permitting the compressor to build up an efficient 100 to 150 pounds of air pressure. No glass slides were broken by the force of the sand blasts.

By placing the frosted glass slides in vertical columns which separated each slide and by alternating the upper surfaces, information was obtained on the numbers of larvae attaching to upper and lower surfaces, ground and smooth surfaces, and to the approximate levels in the tank at which various degrees of spatfall occurred. Four combinations of surfaces was thus obtained: upper smooth, lower ground, lower smooth and upper ground.

For holding the glass slides in position when the free swimming larvae were attaching, "ladders" of one quarter inch plastic were designed. These ladders were capable, when filled, of retaining seventeen glass slides 2 inches apart, figure 1. Two strips of plastic, 4 inches wide and 36 inches long were used for the sides of the ladder. Alternating grooves, first on one side and then on the other, were cut to receive the glass slides. Four pieces of plastic, 4 inches wide and 10 inches long were used for holding the two sides parallel to each other. These were fastened together with plastic cement, which held firmly throughout the experiments conducted.

Factors Influencing Spatfall

Two concrete and four wooden tanks with an approximate capacity of 250 gallons were used to rear the larvae to the setting stage. The wooden tanks were designated as tanks 1, 2, 3 and 4, the concrete tanks as 5 and 6. Pasquale (1953) described the factors to be considered and the problems encountered by the use of these tanks in a thesis entitled "Rearing of the native oyster larvae, *Ostrea lurida* Carp., in concrete and wooden tanks under controlled conditions".

Having been used in past rearing experiments, the

Figure 1. Plastic ladders with inserted glass slides partially prepared for insertion into rearing tank.

tanks were "conditioned", i.e., harmful substances believed leached from the concrete and wood, and judged satisfactory for larval rearing. Notable is the fact that the wooden tanks appeared more dependable than the concrete tanks for obtaining adequate supplies of healthy spat.

Under controlled laboratory conditions the degree of success obtained in rearing larvae to the setting stage varied widely with each rearing attempt. While one factor may have been primarily responsible for the limited success attained at times, it is probable that many factors contributed. Some of these factors are briefly discussed for they may be essential or inimical to obtaining adequate spatfall upon glass slides or shell. Failures in obtaining spatfalls of sufficient magnitude have occurred and the causes of failure have been difficult to interpret in many cases.

Sea water used in rearing containers was first pumped from the bay at the laboratory into large storage tanks where the temperature rises several degrees and the debris settles. Prior to pouring into the rearing tanks, the water was filtered to remove larger organisms, which may have proved detrimental to the free swimming oyster larvae. A vacuum type filter was used, being described in detail by Breese (1953), and only a brief outline of its operation is given here. The water was passed from the outside storage tank into the laboratory through a plastic line. This flow was regulated into an earthernware crock containing several inches of coarse beach sand. A quart bottle without a bottom and containing glass wool was imbedded in the sand. Glass tubing was inserted through a rubber stopper in the neck of the bottle and connected to several holding jars by short sections of glass and rubber tubes. A suction was created by attaching a water jet vacuum pump to the line, pulling the water through the filter and into the retaining jars.

A water temperature near 20° Centigrade was maintained in the rearing tanks since this appeared favorable for the growth of <u>Ostrea lurida</u> larvae. Larvae reared at this temperature completed the free swimming period 9 to 5 days sooner than the 30 days expected in nature (2, p.20). The water temperature was kept quite constant in the rearing rooms by insulation of the walls and thermostat controlled electric heaters. Lower temperatures appeared to slow down the growth of larvae (3, p.111), while the higher temperatures were not generally favorable for increased growth. The duration of the free swimming stage, under laboratory conditions as in nature, depended primarily upon prevailing water temperatures.

The salinity of the water used in obtaining spatfall tended to vary from tank to tank and within each tank with evaporation and water changes. In all experiments the salt concentrations were kept between 24 and 32 parts per thousand since this had proven most satisfactory for rearing Ostrea lurida larvae (2, p.35). Salt water from Yaquina Bay increased in salinity throughout the summer and by fall reached 34 parts per thousand. Adding fresh water to the tanks lowered the salinity, but this method has rarely proven favorable for the free swimming larvae. Consequently obtaining spatfalls late in the spawning season became increasingly difficult when the salinity was high.

Varying concentrations of 400,000 to 600,000 larvae were planted in each 250 gallon rearing tank. These larvae were obtained from adults tonged earlier from the natural beds and were held overnight in a five gallon glass jar while being examined for indications of unhealthiness. To prevent obtaining an entire group of poor larvae from one oyster, young were taken from several oysters and mixed. The large concentrations of larvae used in the rearing tanks did not seem to retard either the group's rate of development or its intensity of setting. Loosanoff (1954) believed that the danger of overcrowding bivalve larvae was not too acute and Breese (1953), in his experiments at Yaquina, did not feel that maximum concentrations of larvae were ever reached.

Possible reasons for failures of larvae to reach the setting stage, as discussed by Loosanoff (1954), included the presence or absence of unknown factors which affected the larvae's ability to utilize food, the use of eggs and larvae from bivalves that did not possess normal vitality, and possible mortalities caused by pathogenic bacteria and virus.

Feeding of the microscopic free-swimming oyster larvae within the rearing tanks has presented a problem to many investigators. The food organisms should measure 7 to 9 microns or less to be ingested; they should be acceptable to the larvae; they should provide the needed nutritive substances beneficial to the larvae's development and they should be non-toxic. The culture media used at the laboratory, having been developed by Morris in 1949, was composed of:

500 milliliters unfiltered salt water
500 milliliters fresh water
1.5 grams starch, soluble, purified powder
.15 grams potassium nitrate
.3 grams disoldium phosphate

A culture, containing micro-organisms such as the naked flagellate protozoa Bodo lens (O.F.Muller), other unidentified microscopic protozoans and bacteria, was used to innoculate the media. One liter of this innoculated culture media was then introduced into the rearing tanks on alternate days. This "feeding" was continued well into the spat stage, concluding when the young oysters were removed from the rearing tank.

When the swimming larvae were being reared, approximately 75 gallons of water in the rearing tanks were changed every eight days. An apparatus similar to the salt water filter (2, p.14) was used to remove the water from the tanks without loss of the larvae. This also consisted of a crock partially filled with coarse beach sand, a bottle minus the bottom and containing glass wool, a rubber stopper, a glass tube and a long section of rubber tubing. The crock was placed in the rearing tank, the bottle inserted in the sand, the stopper and glass tube fitted into the neck of the bottle and the rubber tubing extended over the rim of the tank from the glass tube to the outside. To accomplish the water change, a suction was applied to the rubber tubing and the water slowly siphoned out. Filtered bay water with a temperature near that of the water remaining in the tank was then added.

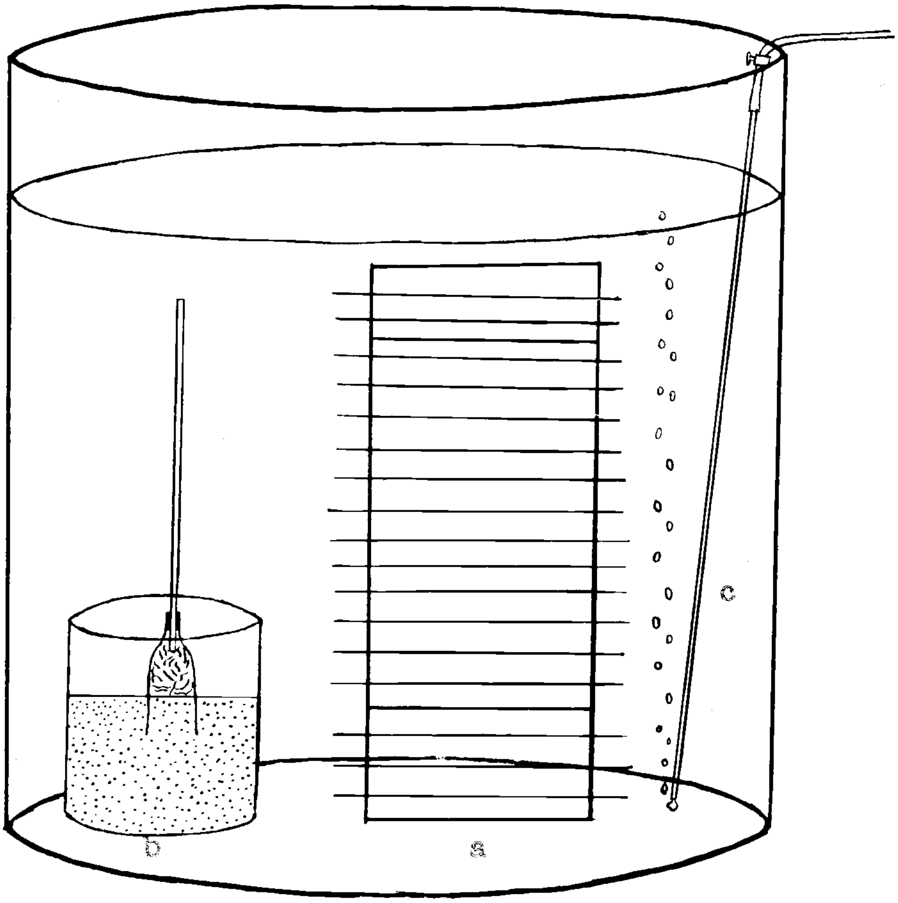
The time at which glass slides and shell were introduced into a larval rearing tank may possibly have influenced spatfall through "fouling". This was due to an organic growth that appeared on the cultch several days after immersion and which interfered with larval fixation. Korringa (1954) stated that, while perfectly clean surfaces were less attractive to oyster larvae than so-called prefouled collectors, cultch placed too early and covered with too many organisms appeared to be a "death trap" for the larvae.

Placement of the cultch occurred when the largest larvae measured about 250 microns. If none of the larvae reached that size in 21 days, cultch was introduced as setting will probably occur before the end of 30 days (2, p.42).

The slides were removed from the tanks for enumeration at the termination of the setting period. A beaker of water from the rearing tank held to the light easily indicated the presence or absence of larvae. Setting was considered complete if only a few free swimming larvae were observed and microscope slides or shell placed in the tank for a 24 hour period received no spatfall.

Spatfalls on Glass

Seven groups of larvae were successfully reared to the setting stage during the summer. These larvae attached to glass slides arranged in ladders and placed in the rearing tanks at the commencement of the setting periods. Figure 2 shows a diagrammatic view of a wooden rearing tank, the inserted plastic ladder with slides, the water change filter and aerator. Each slide was frosted on one side and left smooth on the other, the rough surfaces



a. plastic ladder with inserted glass slides

b. water change filter

c. aeration apparatus

Figure 2. Diagrammatic view of wooden rearing tank containing plastic ladder, inserted slides, wster change filter and aeration apparatus.

being divided into square inches for enumeration by graphite lines. These slides were then arranged in the ladders so as to alternate the position of smooth and ground sides on upper and lower horizontal surfaces. Each individual slide was marked for identification by a diamond point glass etching pencil. Capital letters A, B, C, D, E, and F were used as series designation while numbers 1, 2, 3, 4, etc. were used for slides within a given group. Thus slides placed in ladder "A" were identified A-1, A-2, A-3, etc., with A-1 being the top slide of the series. In all rearing experiments, the top slide was submerged 2 to 4 inches below the surface of the water.

Because of the small size of the newly attached spat, initial enumerations were made by means of a hand lens and a binocular microscope. A convenient holder was devised for holding the slides by the corners while examining. This holder permitted light to enter from below, aiding microscopic observations and minimizing the crushing of spat resulting from handling, figure 3.

Dead and unhealthy larvae along with debris frequently made exact enumeration of spat difficult; therefore, each slide was washed gently in the tank before enumeration to remove organisms caught in the surface film or loosely attached to the glass. Particles on the

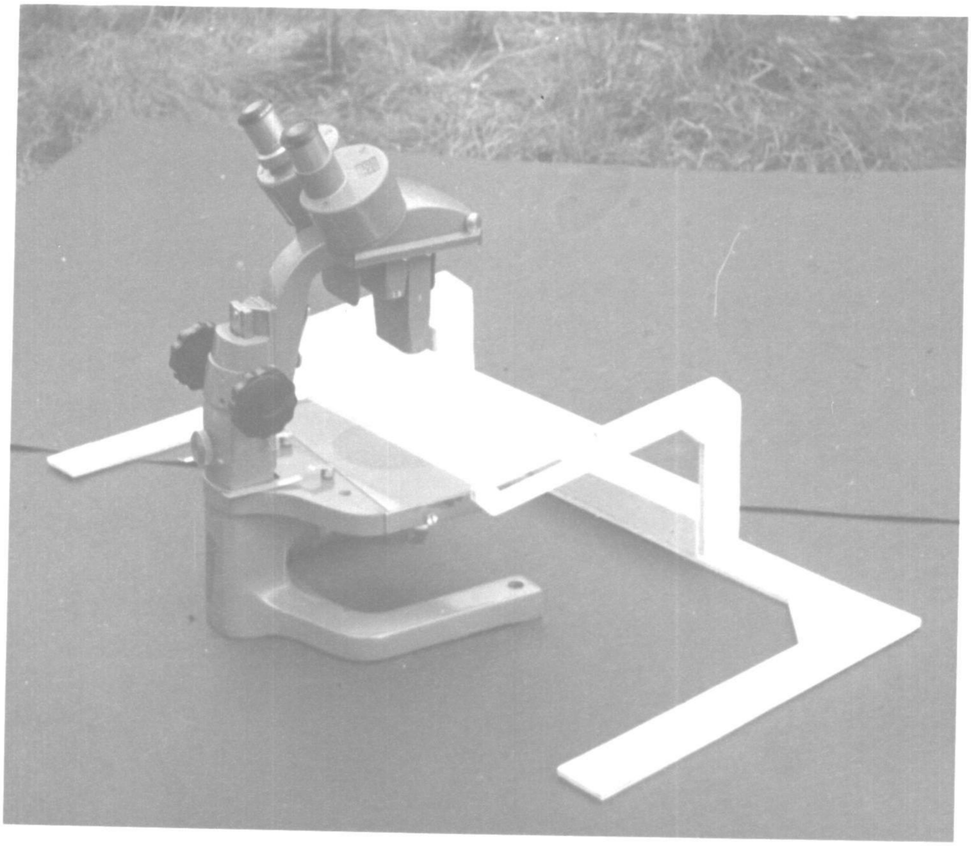


Figure 3. Slide holder and binocular microscope for enumeration of spat on glass.

surface film of ground slides were not as readily removed as those on smooth slides during the rinsing.

Slides A

An estimated 400,000 oyster larvae, averaging 235.6 microns in length, were placed in wooden tank number 2 on June 24. These had an average daily growth of 5.2 microns the first 7 to 8 days. The larvae commenced setting about July 3 and a plastic ladder containing slides A-1 to A-17 was placed in the tank. On July 20, seventeen days later, setting was completed and the slides were removed from the rearing tank. The enumeration of the spat is presented in table 4.

Slides in series A received a total set of 784 spat of .46 spat per square inch. This averaged 46.3 spat per slide, with the greatest spatfall generally occurring near the center of the rearing tank. A marked exception was found on slide A-2 which had a concentrated area of spat on its lower ground surface. Lower ground surfaces received the greatest spatfall with 41.7 per cent, upper ground followed closely with 37.6 per cent, upper smooth had 17.1 per cent and lower smooth received the lowest spatfall with 3.6 per cent. However, if it were not for slide A-2, upper ground surfaces would have rated higher than lower ground.

Table 4. Numbers of spat setting on upper, lower, smooth and ground glass surfaces, slides A-1 to A-17 in tank no. 2, July 3 to July 20, 1954.

Slide	Upper Smooth	Lower Ground	Upper Ground	Lower Smooth	Spat Per Slide
A-1			29	6	35
A-2	7	134			141
A-3			38	7	45
A-4	11	33			44
A-5			31	3	34
A-6	33	21			54
A-7			28	4	32
A-8	23	34			57
A-9			45	3	48
A-10	26	56			82
A-11			43	3	46
A-12	15	27			42
A-13			32	2	34
A-14	15	18			33
A-15			29	0	29
A-16	4	4			8
A-17			20	0	20
Totals	134	327	295	28	784
Percentages	17.1	41.7	37.6	3.6	100.0
Averages	16.75	40.9	32.8	3.1	46.3

Ground surfaces, with 79.5 per cent of the total set, attracted more spat than did smooth surfaces with 20.5 per cent. There appeared to be little difference between upper and lower surfaces, with 54.7 per cent and 45.3 per cent respectively, for attracting larvae.

Slides B

An estimated 500,000 oyster larvae, averaging 235.6 microns in length, were placed in wooden tank number 4 on June 28. Displaying an average daily growth of 7.3 microns the first 7 to 8 days, the larvae commenced setting about July 5 and a plastic ladder containing slides B-1 to B-17 was immersed in the tank.

On July 26, twenty-one days later, setting was completed, the slides were removed from the rearing tank and the spat were enumerated. However, this count was proven to be incorrect when the spat were checked at the conclusion of the first two week holding period. The error was apparently due to a heavy film that had coated the slides, preventing the removal of perished larvae when the slides were rinsed. As a result, the initial enumeration on July 26 was discarded and the second enumeration made on August 9 was used for the spatfall counts, table 5.

The second accurate enumeration showed that slides in series B had a total of 1,163 attached spat, a set of .71 spat per square inch. This averaged 68.4 spat per slide with the set being quite uniform at all levels in the rearing tank. Upper ground surfaces had 47.1 per cent of the total set, upper smooth 43.1 per cent, lower ground 33.2 per cent and lower smooth 12.5 per cent.

Ground surfaces, with 58.1 per cent of the set, attracted more spat than smooth surfaces, with but 41.9 per cent. Upper surfaces showed more attached spat than lower surfaces with 65.8 and 34.2 per cent.

Table 5. Numbers of spat setting on upper, lower, smooth, and ground glass surfaces, slides B-1 to B-17 in tank no. 4, July 5 to 26, 1954.*

Slide	Upper Smooth	Lower Ground	Upper Ground	Lower Smooth	Spat Per Slide
B-1	30	30			60
B-2			61	13	74
B-3	42	43			85
B-4			39	17	56
B-5	44	28			72
B-6			48	18	66
B-7	54	35			89
B-8			42	10	52
B-9	46	21			67
B-10			56	16	72
B-11	46	40			86
B-12			47	8	55
B-13	24	41			65
B-14			34	9	43
B-15	59	35			94
B-16			50	8	58
B-17	43	26			69
Totals	388	299	377	99	1,163
Percentages	33.4	25.7	32.4	8.5	100.0
Averages	43.1	33.2	47.1	12.5	68.4

* Determined after a two week period in the inside holding tank.

Slides C

An estimated 500,000 oyster larvae, averaging 239.1 microns in length, were placed in concrete tank number 5 on June 24. Exhibiting an average daily growth of 7.7 microns the first 7 to 8 days, the larvae commenced setting about July 5 and slides C-1 to C-11 were placed in the tank. Since the concrete tanks were shallower than wooden tanks, but with equal capacity, only 11 slides could be accommodated. On July 27, twenty-two days later, setting terminated and the slides were taken from the rearing tanks for enumeration, table 6.

Table 6. Numbers of spat setting on upper, lower, smooth and ground glass surfaces, slides C-1 to C-11 in tank no. 5, July 5 to 27, 1954.

Slide	Upper Smooth	Lower Ground	Upper Ground	Lower Smooth	Spat Per Slide
C-1			624	25	649
C-2	313	68			381
C-3			418	23	441
C-4	201	121			322
C-5			497	26	523
C-6	180	226			406
C-7			643	22	665
C-8	212	66			278
C-9			277	22	299
C-10	171	39			210
C-11			215	20	235
Totals	1,077	520	2,674	138	4,409
Percentages	24.4	11.8	60.65	3.1	100.0
Averages	215.4	104.0	445.7	33.0	400.8

Slide series C received a total set of 4,409 spat or 4.2 spat per square inch. This averaged 400.8 spat per slide with the spatfall being slightly greater in the upper portion of the rearing tank. Upper ground surfaces received 60.65 per cent of the spat and upper smooth 24.4 per cent, as compared to lower ground with 11.8 per cent and lower smooth with 4.1 per cent. Concentrated areas of spat were found on lower ground surfaces of slides C-4 and C-6.

Ground surfaces, with 72.4 per cent of the total set, attracted more spat than did smooth surfaces, with 27.55 per cent. Upper surfaces showed a much greater spatfall than lower surfaces, with 85.2 per cent and 14.8 per cent. This indicated that the larvae were probably settling downward with gravity influencing attachment.

<u>Slides D</u>

An estimated 500,000 oyster larvae, averaging 249.8 microns in length, were placed in concrete tank number 5 on July 30. With an average daily growth of 2.3 microns the first 7 to 8 days, the larvae began setting about August 16 and slides D-1 to D-11 were placed in the rearing tank. On August 27, eleven days later, setting terminated and the slides were removed for enumeration, table 7. This was the second spatfall obtained from

Table 7. Numbers of spat setting on upper, lower, smooth and ground glass surfaces, slides D-1 to D-11 in tank no. 5, August 16 to 27, 1954.

Slide	Upper Smooth	Lower Ground	Upper Ground	Lower Smooth	Spat Per Slide
D-1	43	7			50
D-2			29	3	32
D-3	31	10			41
D-4			21	21	42
D-5	12	56			68
D-6			120	14	134
D-7	61	15			76
D-8			18	84	102
D-9	79	34			113
D-10			102	12	114
D-11	44	2			46
Totals	270	124	290	134	818
Percentages	33.0	15.2	35.4	16.4	100.0
Averages	45.0	20.7	58.0	26.8	74.4

tank 5 during the summer's rearing attemots.

Slide series D received a total set of 818 spat or .77 spat per square inch. This averaged 74.4 spat per slide with spatfall generally better in the lower half of the tank. Upper ground surfaces received 35.4 per cent of the spat, upper smooth 33.0 per cent, lower smooth 16.4 per cent and lower ground 15.2 per cent.

Ground surfaces, with 50.6 per cent, and smooth surfaces, with 49.4 per cent received approximately the same amount of spatfall. Upper surfaces showed a greater spatfall than lower surfaces with 68.5 per cent and 31.5

per cent. Areas of spat concentration were noted on slides D-6 and D-10.

Slides E

An estimated 400,000 oyster larvae, averaging 239.1 microns in length, were placed in wooden tank number 1 on July 30. With an average daily growth of 3.1 microns the first 7 to 8 days, the larvae commenced setting about August 16 and slides E-1 to E-17 were submerged within the tank. On September 6, twenty days later, setting was completed and the slides were removed for enumeration, table 8.

Slide series E received a total set of 2,238 spat or 1.37 spat per square inch. This was an average of 131.6 spat per slide with the heaviest spatfall near the center of the rearing tank. Lower ground surfaces received half of the total spatfall with 51.2 per cent, upper ground received 30.5 per cent, upper smooth 15.1 per cent, and lower smooth received 3.2 per cent. The lower ground surface of slide E-12 received 402 spat which tends to distort the averages.

Ground surfaces attracted 81.7 per cent of the total set, far surpassing smooth surfaces with but 18.3 per cent. Lower surfaces received a set of 53.3 per cent compared to upper surfaces with 46.7 per cent.

Table 8. Numbers of spat setting on upper, lower, smooth and ground glass surfaces, slides E-1 to E-17 in tank no. 1, August 16 to September 6, 1954.

Slide	Upper Smooth	Lower Ground	Upper Ground	Lower Smooth	Spat Per Slide
E-1			62	3	65
E-2	36	85			121
E-3			61	9	70
E-4	28	55			83
E-5			62	5	67
E-6	64	92			156
E-7			117	14	131
E-8	35	176			211
E-9			99	9	108
E-10	44	106			150
E-11			69	8	77
E-12	71	402			473
E-13			65	8	73
E-14	15	153			168
E-15			108	8	116
E-16	45	76			121
E-17			41	7	48
Totals	338	1,145	684	71	2,238
Percentages	15.1	51.2	30.5	3.2	100.0
Averages	42.5	143.1	76.0	8.0	131.6

<u>Slides F</u>

An estimated 600,000 oyster larvae, averaging 639.1 microns in length, were placed in wooden tank number 3 on July 6. With an average daily growth of 7.4 microns the first 7 to 8 days the larvae commenced setting about July 16 and slides F-1 to F-17 were immersed in the rearing tank. On August 6, twenty-one days later, setting was completed and the slides were removed for enumeration,

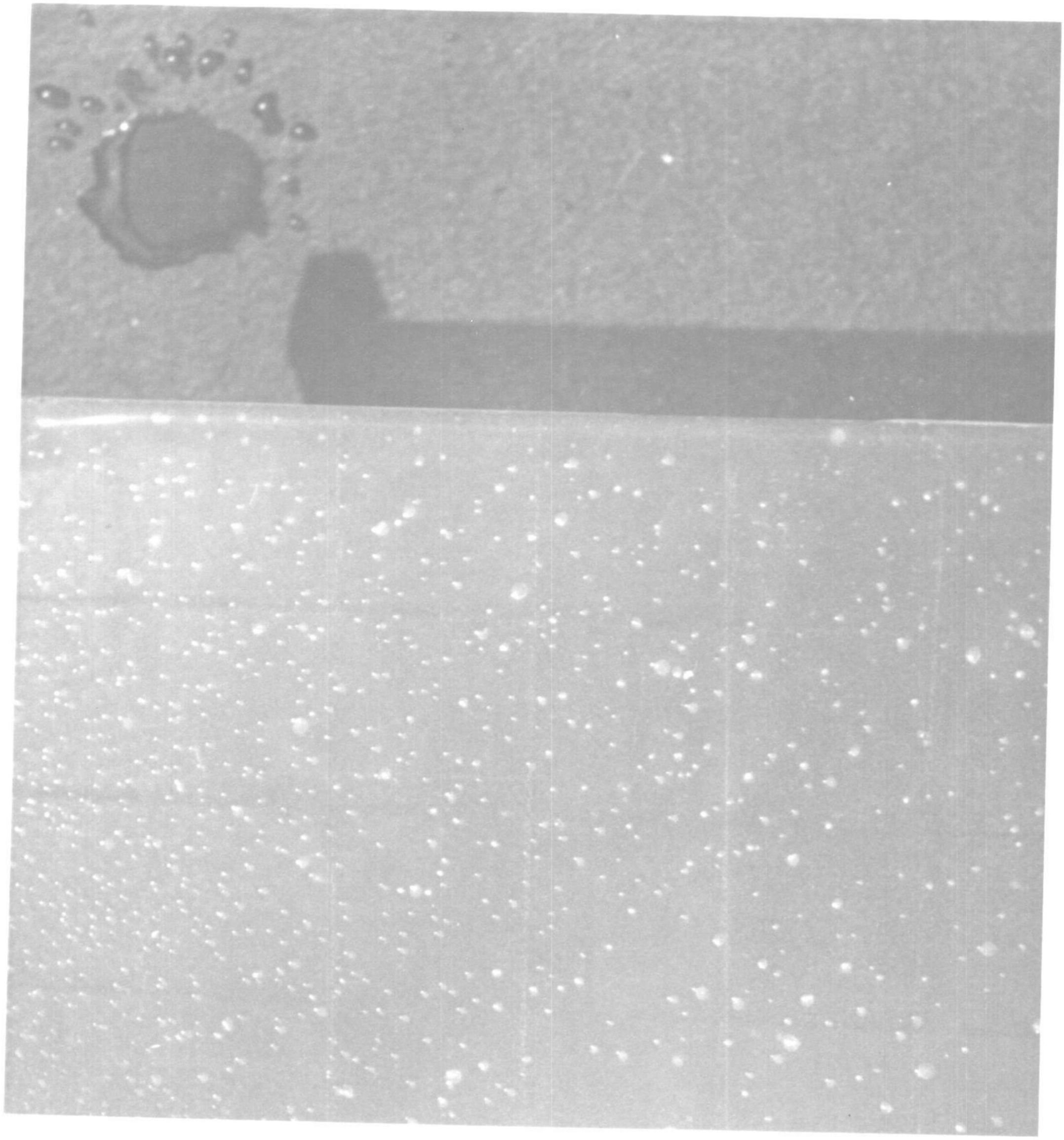


Figure 4. An abundant set upon a glass slide from tank no. 3, showing the surface of the glass marked into square inches and the minute size of the young spat.

Table 9. Numbers of spat setting on upper, lower, smooth and ground glass surfaces, slides F-1 to F-17 in tank no. 3, July 16 to August 6, 1954.

Slide	Upper Smooth	Lower Ground	Upper Ground	Lower Smooth	Spat Per Slide
F-1	189	1,903			2,092
F-2			275	105	
F-3	225	2,042			2,267
F-4			253	138	391
F-5	181	1,975			2,156
F-6			263	124	387
F-7	210	2,895			3,105
F-8			325	170	495
F-9	270	2,132			2,402
F-10			312	141	453
F-11	330	960			1,290
F-12			1,362	155	1,517
F-13	261	1,037			1,298
F-14			402	151	553
F-15	413	441			854
F-16			146	94	240
F-17	322	417			739
Totals	2,401	13,802	3,338	1,078	20,619
Percentages	11.7	66.9	16.2	5.2	100.0
Averages	226.8	1,533.5	417.2	134.7	1,212.9

table 9.

Slide series F received a total set of 20,619 spat, an average of 12.6 spat per square inch. The heaviest concentrations were found upon lower ground surfaces with 31.9 spat per square inch. This was the most abundant set attained during the summer's rearing experiments upon glass slides, the inside walls of the tank also attracting numerous spat. Figure 4 shows the abundant set upon one of these slides and reveals the small sizes of the young oysters. Each slide averaged 1,213 spat, with the heaviest sets occurring in the upper two thirds of the rearing tank upon lower ground surfaces. A deviation was found on slide F-12, which received 1,362 spat on its upper ground surface, tending to distort the average for that column. Lower ground surfaces received the greatest spatfall with 66.9 per cent, followed by upper ground, upper smooth and lower smooth with 16.2, 11.7 and 5.2 per cents. However, upper smooth and upper ground surfaces would have been nearly equal but for slide F-12.

Ground surfaces, with 83.1 per cent of the total set, attracted more spat than did smooth surfaces, with but 16.9 per cent. Lower surfaces received 72.2 per cent and upper surfaces 27.9 per cent. Lower ground surfaces, with an exceedingly high abundance of spat, dominated the entire set.

Slides G

An estimated 600,000 oyster larvae, averaging 252.5 microns in length, were placed in tank number 3 on August 8. With an average daily growth of 1.3 microns the first 7 to 8 days, the larvae commenced setting about August 25 and slides G-1 to G-17 were immersed in the rearing tank. On September 4, nine days later with

Table 10. Numbers of spat setting on upper, lower, smooth and ground glass surfaces, slides G-1 to G-17 in tank no. 3, August 25 to September 4, 1954.

Slide	Upper Smooth	Lower Ground	Upper Ground	Lower Smooth	Spat Per Slide
G-1			20	16	36
G-2	3	36			39
G-3			13	7	20
G-4	4	39			43
G-5			11	5	16
G-6	11	17			28
G-7			16	9	25
G-8	5	26			31
G-9			11	8	19
G-10	9	19			28
G-11			11	5	16
G-12	3	12			15
G-13			10	7	17
G-14	2	37			39
G-15			5	1	6
G-16	7	5			12
G-17			6	2	8
Totals	44	191	103	60	398
Percentages	11.05	48.0	25.0	15.1	100.0
Averages	5.5	23.9	11.4	6.6	23.4

setting neared completion, the slides were removed and the spat were counted, table 10. This was the second successful rearing attempt of the summer from tank 3.

Slide series G received a total set of 398 spat, an average of .24 spat per square inch. This was the least abundant spatfall received during the summer's rearing experiments. Each slide averaged 23.4 spat with the distribution of abundance scattered throughout the upper levels of the tank. Lower ground surfaces received 48.0 per cent of the spatfall, followed by upper ground, lower smooth and upper smooth with 25.9, 15.1 and 11.05 per cents.

Ground surfaces with 73.9 per cent of the total set attracted more spat than did smooth surfaces with but 26.1 per cent. Lower surfaces with 63.1 per cent exceeded upper surfaces with 36.9 per cent. Because of insufficient numbers of spat, the slides were removed from further experimentation.

Discussion

The total numbers of free-swimming larvae attaching to glass slides varied widely with the seven experimental rearings from which spatfalls were obtained. Data from these rearings, table 11, discloses that the number of spat obtained was probably independent of the number of free-swimming larvae placed in the tank. Because of their microscopic size and lack of sufficient current in the rearing containers, many larvae may have failed to contact a surface when prepared to attach.

On certain slides, small areas of intense spat concentration was found. While these may have been caused by minor water currents within the tanks, gregariousness of the oyster larvae may also have contributed to the phenomenon.

Table 11. Data from seven experimental rearings, slide series A to G, June to September, 1954.

Slide Series	No. of Larvae	Growth* of larvae (microns)	Setting Period (days)	No. of Spat	Spat per Square Inch
A	400,000	5.2	17	784	.46
B	500,000	7.3	21	1,163	.71
C	500,000	7.7	22	4,409	4.2
D	500,000	2.3	11	818	.77
E	400,000	3.1	20	2,238	1.37
F	600,000	7.4	21	20,619	12.6
G	600,000	1.3	9	398	.24

* From measurements taken parallel to the hinge, 1 to 8 days after placement in rearing tank.

A comparison of the percentages of set upon the four glass surface combinations from the experimental rearings was made in figure 5. While lower smooth surfaces usually attracted the least amount of larvae, a wide variation was indicated between upper smooth, lower ground, and upper ground surfaces. However, in four of the seven sets, lower ground surfaces received the greatest spatfall while upper ground was highest in two, indicating that the larvae prefer ground surfaces.

That larvae preferred ground surfaces for attachment was also indicated by a comparison of total set on ground and smooth surfaces, figure 6. In all rearings, spatfall was least on smooth surfaces.

No conclusion could be reached concerning the value of upper and lower surfaces for larval affixture. The

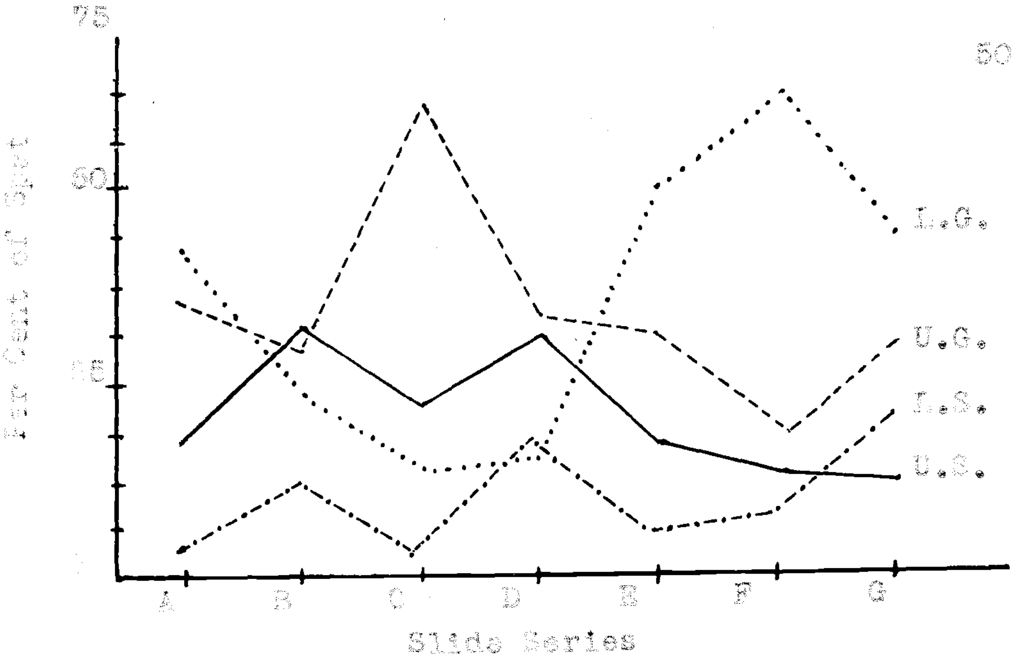


Figure 5. Comparison of spatfall on upper smooth, lower ground, upper ground and lower smooth glass surfaces, slide series A to G, June to September, 1954.

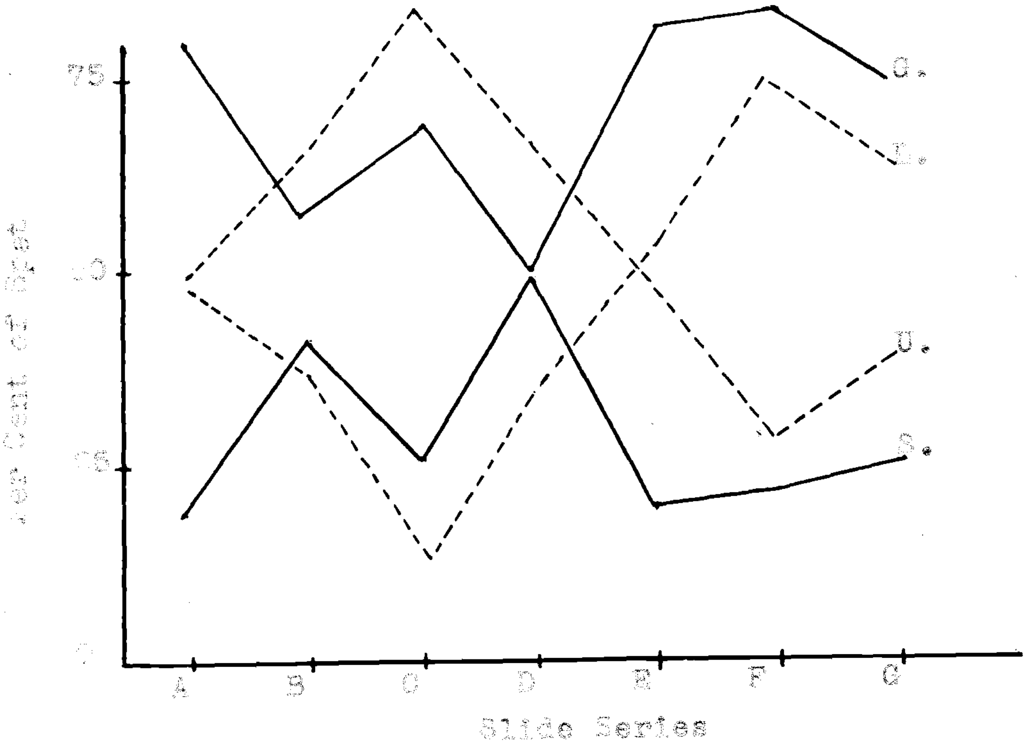


Figure 6. Comparison of spatfall on ground and smooth, upper and lower glass surfaces, slide series A to G, June to September, 1954.

first four rearings showed a greater set on upper surfaces while the last three rearings received a greater set on lower surfaces.

Whether or not the greatest spatfall occurred on upper or lower surfaces may have depended upon the physical condition of each group of larvae as determined by environmental conditions present in each rearing tank, larvae of poor health tending to settle downward under the influence of gravity. Series D had a greater percentage of spat upon upper surfaces, most of which appeared in poor condition through little growth following attachment. In contrast, the numerous spat on the lower ground surfaces of series F appeared to be quite healthy, beginning growth immediately following fixation.

The amount of set occurring at various levels in the rearing tanks tended to fluctuate widely with each group of larvae, figures 7, 8, 9 and 10. Because of alternating surface combinations from the top to the bottom of the tank, an extreme variation between alternate slides was found. This is particularly noted from series F where the larvae exhibited definite selection of lower ground surfaces.

In general, the spatfalls indicated a greater set in the top three quarters of the rearing tanks than in the lower quarter. This may be the result of insufficient

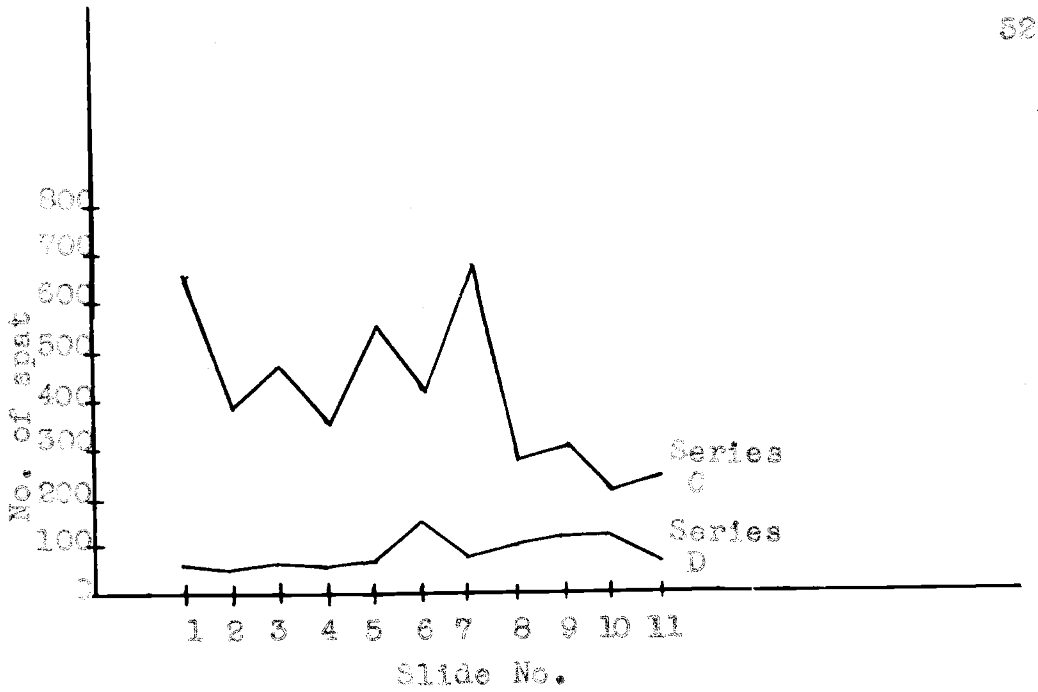


Figure 7. Comparison of set, slide series C and D, at various levels in concrete tank no. 5, July 27 and August 27, 1954; slide 1 being near the surface of the tank and slide 11 near the bottom.

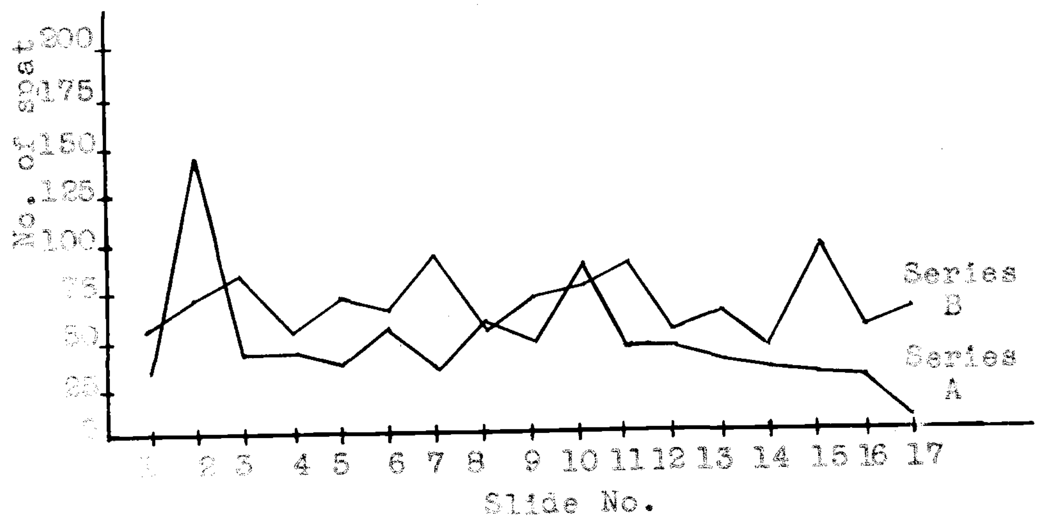


Figure 8. Comparison of set, slide series A and B, at various levels in wood tank no. 2 and 4, July 20 and 26, 1954; slide 1 being near the surface of the tank and slide 17 near the bottom.

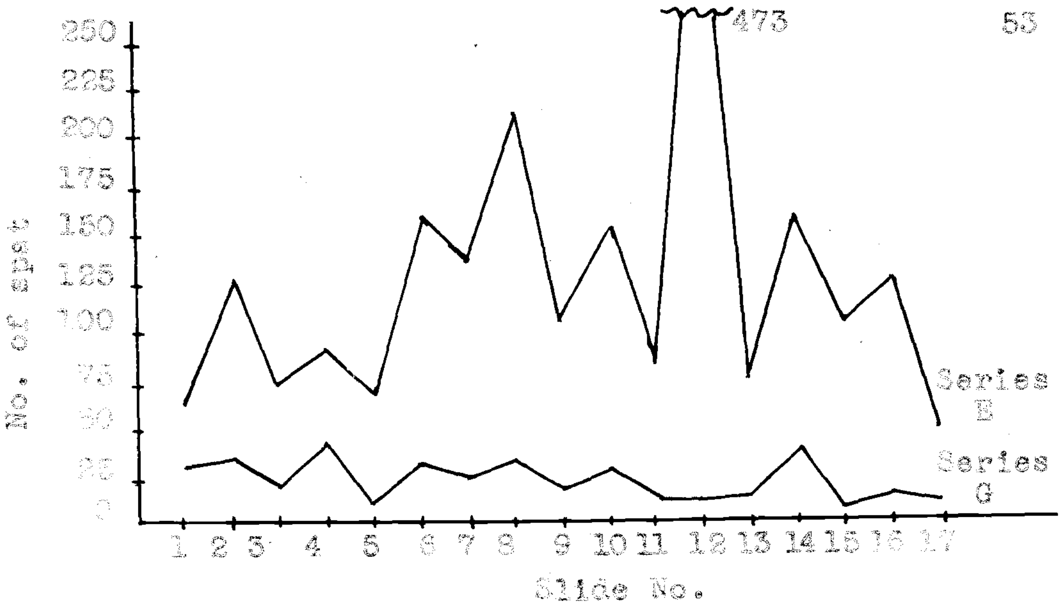


Figure 9. Comparison of set, slide series E and G, at various levels in wood tank no. 1 and 3, September [illegible] and 4, 1954; slide 1 being near the surface of the tank and slide 17 near the bottom.

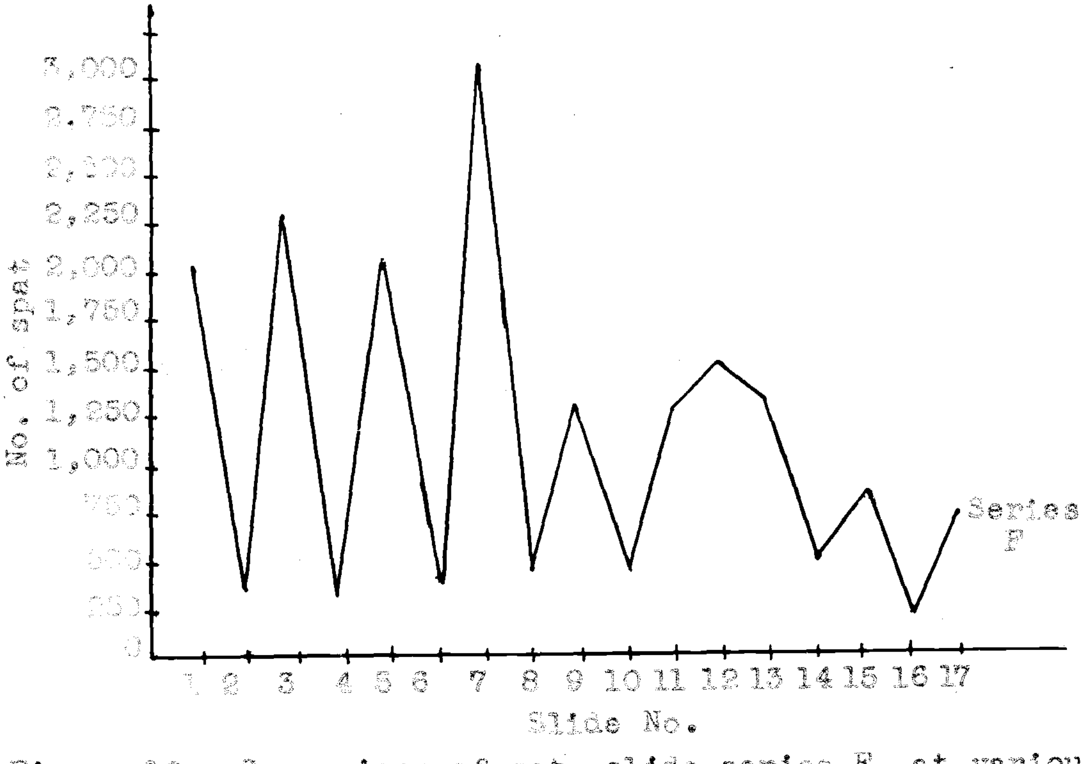


Figure 10. Comparison of set, slide series F, at various levels in wood tank no. 3, August 6, 1954; slide 1 being near the surface of the tank and slide 17 near the bottom.

water circulation and aeration in the lower areas. In certain rearings, a slight reduction in numbers of spat was noted on the top five slides which were examined weekly during the critical setting period.

GROWTH AND MORTALITY OF YOUNG SPAT

During the initial stages of spat growth in rearing tanks, immediately following fixation, the young oysters were presumed to be in a critical developmental period. At this time a metamorphosis adapting the spat to a permanent sedentary life was taking place and the formation of the adult shell was just beginning. The rates of growth and the amounts of mortality sustained by the spat while in the rearing tanks were important factors to be considered.

To collect spat for the following observations, shells of the Pacific oyster and the cockle, _Clinocardium nuttalli_ (Conrad), were utilized. Each shell was washed and wire brushed to remove foreign objects and prevent contamination of the rearing tanks. After cleaning two holes were drilled through each shell for stringing with nylon leader material. The shells were then suspended individually, smooth side down, in various rearing tanks for larval attachment. In these tanks, simultaneously, were the plastic ladders containing series of glass slides, so the larvae attaching to the shell were representative of those attaching to the glass.

Early Growth Stages

Young spat completing metamorphosis showed two distinct shells, the inner or old larval shell that was present prior to metamorphosis and the new shell formed by the growing oyster following metamorphosis. These two shells were readily observed in the young spat through a microscope.

To facilitate observations during this period, it was necessary to establish five arbitrary stages of early growth that extended from the time of larval affixture until the spat were firmly attached to the cultch, figure 11. These stages were identified by Roman numeral as I, II, III, IV and V, and determined as follows:

> Stage I - No new shell has been deposited, the larva having just settled. The hinges may be slightly parted but show no indication of expanding.
>
> Stage II - Some growth of the new shell has occurred. The spat has turned to its side, placing its left valve downward upon the cultch.
>
> Stage III - New shell growth is projecting outward, at right angles to the hinge, to approximately the width of the old shell. Parallel to the hinge, the new growth is nearly one-half way around the old shell.
>
> Stage IV - New growth has progressed outward to approximately two times the width of the old shell.
>
> Stage V - New growth has increased to approximately three times, or greater, the width of the old shell.

A few sample measurements of the new shell, taken at right angles to the hinge, ranged from 16.0 to 37.4 microns in stage II, 184.2 to 237.4 microns in stage III,

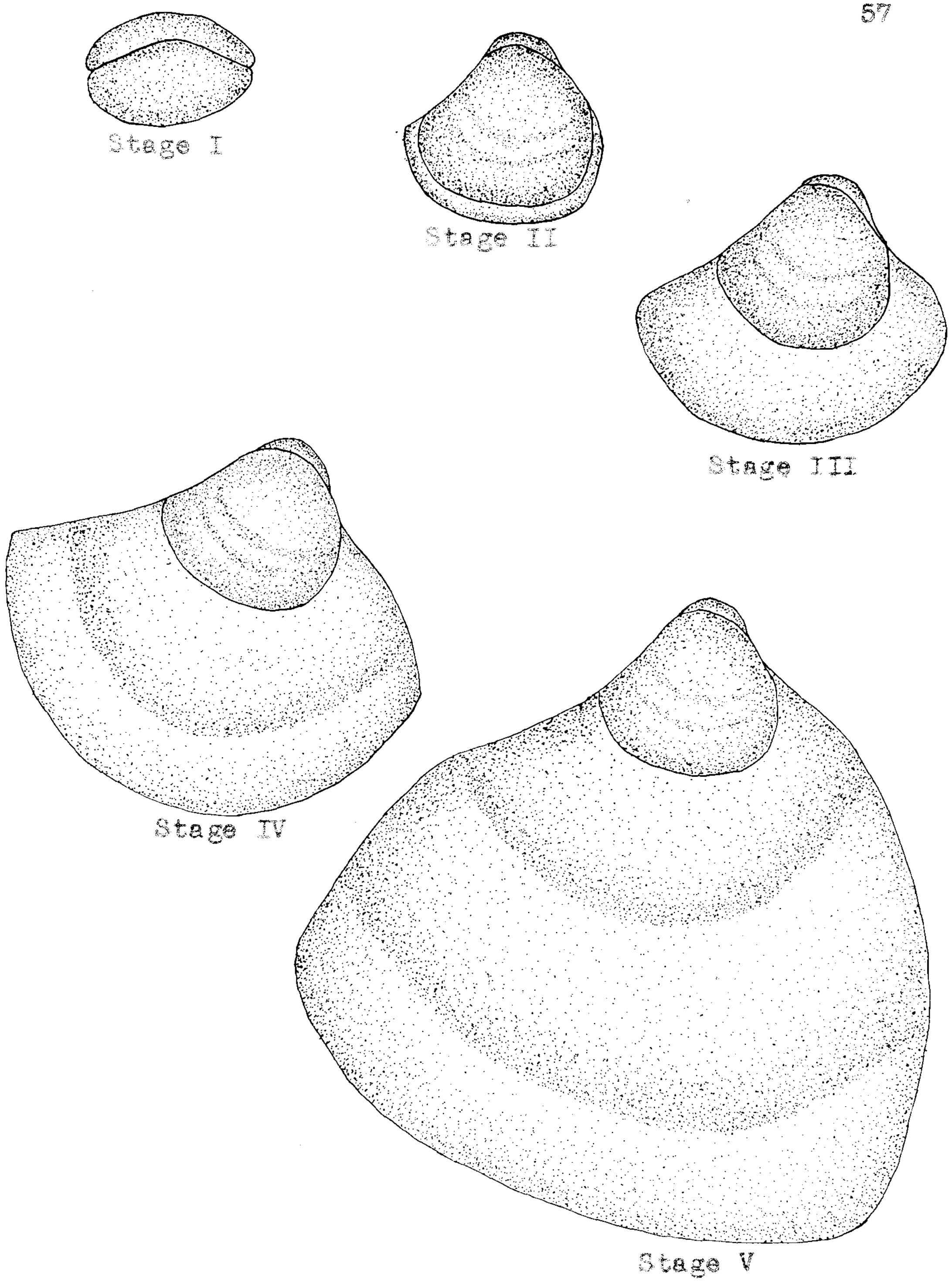


Figure 11. Stages of early spat growth.

211.6 to 445.5 microns in stage IV, and 466.7 to 643.6 or more microns in stage V. Stage I had no measurable new shell. The measurements tended to overlap as the stages were determined from the relationship of old to new shell. Also, the old shell showed considerable variation for the larvae settle at different sizes. In the samples measured, the width of the old shell varied from 195.6 microns to 256 microns. This measurement should not be confused with the length of the larvae, which is taken parallel to the hinge. Spatfalls in the rearing tanks usually began when the largest larvae reached about 250 microns in length.

Rates of Growth

Having established five distinct stages for young spat, observations were conducted upon the rates of growth within these stages. Shells were first placed in rearing tanks containing free-swimming larvae for a 24 hour period. The shells with attached spat were then transferred to another tank free of larvae. Identification of each individual spat upon the smooth inner surfaces of the shells was accomplished by encircling with a graphite mark and recording the position. Thereafter, for a seven to twelve day period, the shells were examined daily for growth and mortality of the spat. Care was taken to prevent losses from handling and dehydration. Spat indicating abnormally slow development and perishing during later examinations were discarded to avoid bias in the determination of growth rates. Table 12 shows the growth rate of young spat from microscopic inspection of individuals fastened to shell.

Table 12. Early growth stages of 362 oyster spat obtained from rearing tanks, July to September, 1954.

Stage	Day Examined						
	1	2	3	4	5	6	7
I	150	37	8	3	2	1	0
II	119	142	56	33	12	5	5
III	92	154	247	205	152	89	51
IV	1	29	51	120	190	239	249
V	0	0	0	1	6	28	57

Young spat usually began growth of the new shell immediately after settling upon the cultch. Spat known to have attached within the previous 24 hours were often found in stage II when first examined. Occasionally a larvae settled and reached spat stage III within 24 hours, this depending somewhat upon how early the larvae attached within the first 24 hours.

In general, from the data obtained, it can be stated that spat spent from one to two days in stage I, one to four days in stage II, one to seven days in stage III, and two to seven days in stage IV. Some "overlapping" undoubtedly occurred as an individual passing from stage I to stage III within 24 hours may have been in late stage I when first examined and early stage III when next inspected.

After seven days had passed, healthy growing spat were firmly attached and depositing new shell. Spat perishing during metamorphosis had no new growth at this time and in many instances had disappeared entirely from the cultch. Many spat first formed new shell slowly, increasing the rate of shell formation as they increased in size. Other spat developed uniformly through all five stages, while still others grew rapidly in the early stages and exhibited a decreased growth rate in later stages. Pale grey colorations were sometimes indicative of "sickly" spat which displayed slow growth and later perished. Normal feeding spat usually had a light brown coloration resulting from the color of their internal organs as seen through the semi-transparent shell. Many spat were noted with a dark grey tint. However, coloration could not definitely be used as an indication of either growth or condition.

Mortalities

Spat attached to shell utilized in growth studies were also employed to determine early mortalities. Each graphite encircled spat was checked daily for a seven day period and notations were made concerning crushed, missing or apparently dead individuals, table 13. Care was taken to avoid excessive mortality due to handling.

Certain considerations should be made concerning the relationship of spat to early mortalities. Frequently spat revealing no growth disappeared at subsequent examinations and were listed as missing. Probably some dead spat were accidentally crushed and mistakenly listed in the crushed column. Also, some spat listed as missing may have been crushed. The total mortality consisted of all individuals lost during the period of observations while the natural mortalities was the total mortality minus the crushed individuals. The natural mortality was indicative of the true loss as it included spat actually perishing during the seven day checking period.

Cultch shells possessing a convex ~~inner~~ outer surface were found to lo~~o~~se more spat from crushing than those with a concave inner surface. This was because of the protection provided by the concave surfaces from accidental scraping and bumping. Spat in stage I were often found missing in the first twenty-four hours of the examination period as they lacked sufficient time for firm attachment prior to handling.

In these five rearing experiments, the natural mortalities ranged from 8.9 to 43.0 per cent during the

Table 13. Early mortalities of spat during seven day examination periods in rearing tanks, July to September, 1954.

Allocations	Tank 2		Tank 4		Tank 5		Tank 5*		Tank 3	
	No. Spat	Per Cent	No. Spat	Per Cent	No. Spat	Per Cent	No. Spat	Per Cent	No. Spat	Per Cent
Total Spat Examined	121	--	127	--	124	--	100	--	101	--
Crushed	14	11.6	11	8.7	8	6.45	0	0.0	3	3.0
Missing	15	12.3	13	10.2	11	8.9	7	7.0	3	3.0
Apparently Dead	14	11.6	14	11.0	13	10.5	36	36.0	7	7.0
Total Mortality	43	35.5	38	30.0	32	25.8	43	43.0	12	11.9
Natural Mortality**	29	24.0	27	21.3	24	19.4	43	43.0	9	8.9

* Second spatfall received from tank 5.
** Total mortality minus crushed.

initial stages of spat growth. Of the 573 spat examined, 132 were listed as natural mortalities, which gave an average loss of 23.0 per cent seven days following setting. This loss was believed to be somewhat high due to undetermined inimical effects upon the spat from daily handling of the shells.

The second set obtained from tank 5 showed the highest loss, a comparison with glass slides indicating that these spat consisted largely of individuals in poor condition. In contrast, tank 3 contained fast growing healthy individuals that showed the least mortality. This indicated that the early mortalities were correlated with the general condition of the spat received from each rearing tank.

MORTALITY OF LATER SPAT STAGES

Once the young spat were firmly attached to the glass slides, following metamorphosis, they entered a period of rapid development that extended until late fall with the onset of lowering water temperatures. During this period of their life cycle, the spat were believed to be particularly influenced by the environmental conditions to which they were exposed. In hopes of increasing the numbers of surviving seed oysters, the spat were thus subjected to partially controlled conditions for various lengths of time prior to immersion in the waters of the bay. Three different types of conditions were provided by an inside holding tank, an outside water storage tank, and a floating live box in the bay. By enumerating at regular intervals, an estimate of the total mortalities incurring under each set of conditions was obtained. Also, the total accumulated mortalities for the entire observation period were determined and a comparison of mortalities upon the four surface combinations, upper smooth, lower ground, upper ground and lower smooth was made.

Environmental Conditions

When setting in rearing tanks terminated, the glass slides were removed from the plastic ladders and the spat enumerated. They were then placed in "cartons" of wood

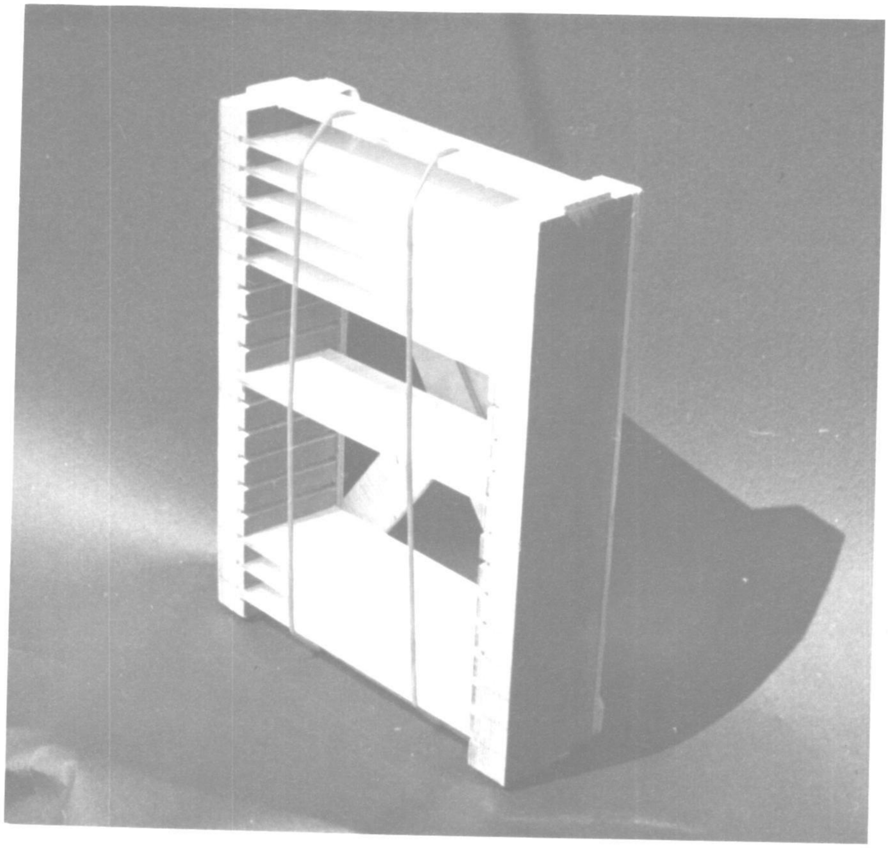


Figure 12. A wood carton used for retaining glass slides with attached spat during mortality periods in the inside holding tank, the outside storage tank, and the bay.

construction designed to retain the slides in a fixed position while the young spat were growing, figure 12. Two 18 inch sections of two by four lumber were used as sides of each carton. Seventeen or eleven grooves, according to the number of slides accommodated, were cut into each side. Strips of one quarter inch plywood were used to frame the sides, serving as a back for the grooves and as braces. Two pieces of rubber tubing were stretched from top to bottom of each carton to hold the inserted slides in position. Each carton was identified with a stencil as to the slide series it contained. To minimize the effects of toxic substances which might leach from the wood and harm the spat, the cartons were immersed in water several weeks prior to usage.

Rearing tank 4 was utilized for retaining the cartons with young spat inside the laboratory. This tank had previously served in obtaining spatfall. Sea water was filtered to remove organisms that might prove harmful to spat, as in the rearing experiments, and kept at temperatures between 19 and 22 degrees Centigrade. Salinity varied between 29 and 33 parts per thousand. Food media was introduced on alternate days and 75 gallons of water were changed every eight days. Aeration was provided by a two cylinder air pump. In general, the same water conditions prevalent in rearing the free-swimming larvae to the setting stage existed here.

In addition, a large wooden salt water storage tank was also utilized in retaining the spat covered slides for mortality periods. Here conditions differed from the inside holding tank, the sea water being pumped directly from the bay and remaining unfiltered. Since this tank was located outside of the laboratory the temperatures fluctuated throughout each twenty-four hour period, ranging from 12 to 18 degrees C. Bay water was pumped into the outside tank every three days to replenish the food material as the seed oysters did not receive food media. The salinity varied with the stage of tide when pumping occurred, ranging from 29.4 to 31.9 parts per thousand. A top on the tank prevented the direct rays of the sun from entering and warming the water to abnormally high temperatures. No means of aeration was provided, the large tank capacity and frequent water changes being considered sufficient to satisfy the oxygen requirements of the growing seed oysters.

Other groups of spat were placed directly into Yaquina Bay outside of the laboratory. To prevent excessive mortalities from siltation, the slide cartons were placed into a large floating live box, figure 13. Two 6 by 16 inch timbers served as floats and a large wire basket, 60 inches long, 28 inches wide and 18 inches high

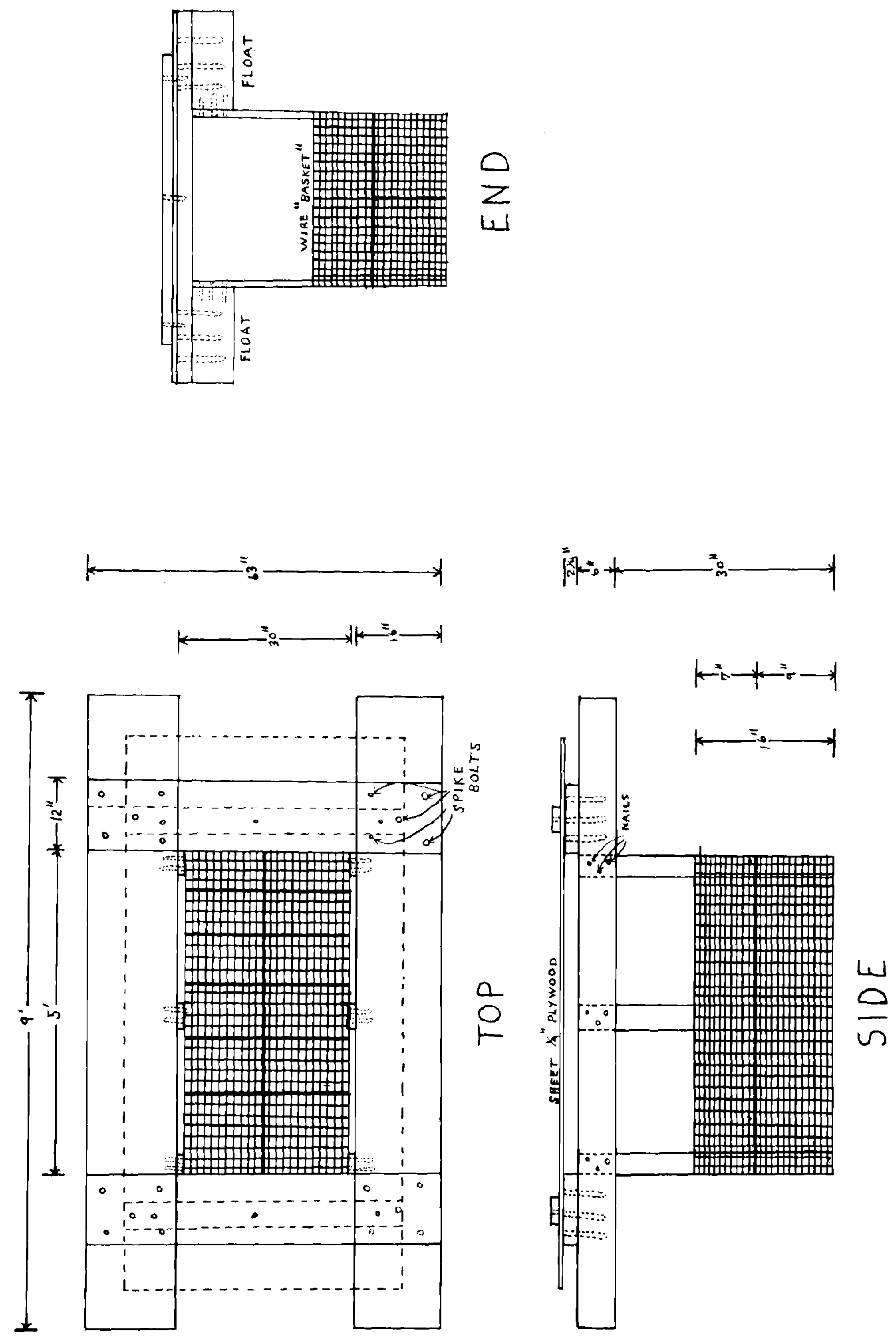


Figure 13. The floating live box placed in the bay, utilized for holding wooden cartons and slides for mortality periods.

was suspended underneath. To protect the glass slides from the sun's rays and restrict the growth of algae, a sheet of one quarter inch plywood was placed over the live box. Seasonal environmental conditions encountered included a temperature variation of 9.5 to 18 degrees C., and a salinity range of 26.8 to 34.0 parts per thousand. These two conditions changed with the ebb and flow of the tides and the amounts of fresh water discharged by the Yaquina River. Nutritive substances and food micro-organisms were obtained directly from the sea water in competition with other living organisms attaching to the glass slides.

Mortalities

Slides A

Slide series A were placed in the outside storage tank on July 20. Two weeks later, on August 3, the slides were removed, the spat enumerated and the mortalities noted, table 14. Most of the spat were accidentally destroyed at this time when the rubber tubing permitted the slides to slip from the carton, thus removing slides A from further observations.

After 14 days in the outside storage tank, the mortalities on the four main types of surfaces ranged from 36.6 to 21.7 per cent, being greatest on upper smooth and

Table 14. Spat mortalities, slide series A, through a two week period in the outside storage tank, July 20 to August 3, 1954.

Slide Surface	Spat Count July 20	Spat Survival August 3	Mortality (Per Cent)
Upper Smooth	134	85	36.6
Lower Ground	327	256	21.7
Upper Ground	295	218	26.1
Lower Smooth	28	20	28.6
Ground	622	474	23.8
Smooth	162	105	35.2
Upper	429	303	29.4
Lower	355	276	22.3
Totals	784	579	26.2

least on lower ground. Smooth surfaces with 35.2 per cent received a higher mortality than ground surfaces with 23.8 per cent. Upper surfaces with 29.4 per cent received higher losses than ground with 22.3 per cent. Total mortality for the two week period of growth in the outside storage tank was 26.2 per cent. Some silting occurred on the upper surfaces of all slides since the carton was suspended vertically.

Slides B

Slide series B were removed from rearing tank 4 on July 26 for the initial enumeration. The count was proven inaccurate by a second enumeration on August 9 so this second check, following two weeks in tank 4, was taken as the spatfall count. The slides were suspended in the large outside holding tank for two weeks until August 23 and then placed in the floating live box in the bay until the termination of observations, being removed only at intervals for the determination of losses. The accumulated mortalities determined from these experiments is presented in table 15 as percentages.

After 166 days, from August 9 to January 8, the mortalities on the four main types of surfaces ranged from 97.1 to 69.2 per cent, being greatest on upper smooth and least on lower ground. Smooth surfaces with 95.5 per cent received a higher mortality than ground surfaces with 80.9 per cent. Upper surfaces with 93.7 per cent received higher losses than lower surfaces with 74.1 per cent.

For this period the total mortality for all slides was 87.0 per cent, giving a survival of only 13.0 per cent. Mortalities were greatest the first two weeks in the outside holding tank where half of the initial set was found missing. This was believed due primarily to sediment present in bay water when pumped into this tank every three days, resulting in a moderate layer of silt being deposited upon the upper surfaces of all slides. In further holding experiments in the outside tank, the slide cartons were suspended horizontally instead of

Table 15. Spat mortalities, slide series B, through various experimental periods from inside tank into outside tank and then into bay, August 9, 1954 to January 8, 1955; percentages of accumulated mortality from first enumeration in parenthesis.

Slide Surface	Numbers of Surviving Spat					
	Inside tank*	Outside tank	Bay			
	Aug. 9	Aug. 23	Sept. 6	Sept. 20	Nov. 6	Jan. 8
U. S.	388	122 (68.6)	49 (87.4)	41 (89.4)	22 (94.3)	11 (97.1)
L. G.	299	222 (25.8)	170 (43.1)	141 (52.9)	123 (58.9)	92 (69.2)
U. G.	377	173 (54.1)	81 (78.5)	71 (81.2)	52 (86.2)	37 (90.2)
L. S.	99	57 (42.4)	38 (61.6)	28 (71.7)	20 (79.8)	11 (88.9)
Ground	676	395 (41.6)	251 (62.8)	212 (68.6)	175 (74.1)	129 (80.9)
Smooth	487	179 (63.3)	87 (82.1)	69 (85.8)	42 (91.4)	22 (95.5)
Upper	765	295 (61.4)	130 (83.0)	112 (85.4)	74 (90.3)	48 (93.7)
Lower	398	279 (29.9)	208 (47.7)	169 (57.5)	143 (64.0)	103 (74.1)
Totals	1,163	574 (50.6)	338 (70.8)	281 (75.8)	217 (81.3)	151 (87.0)

* Determined from second enumeration, after two weeks in the inside holding tank.

vertically so as to decrease mortalities from this source. Silting was less of a problem in the bay, for while the live box firmly retained the slide cartons in a vertical position, the action of the currents tended to free the glass of silt.

Within the bay, the mortalities sustained during each holding period gradually lessened as the spat began increasing in size. Undoubtedly, most of the weaker spat had been lost in the outside tank, but the increase of competitive organisms on the slides began to exert detrimental influences.

Abundant growths of marine organisms were first noted on September 20, one month after the slides were immersed in the bay. By November 6, barnacles, Balanus sp., were exceedingly numerous, crowding out the young seed oysters and growing upon their shells, while the growth of other fouling organisms had reached a peak.

By January 8, most of the competing organisms that profusely covered the slides in the fall had perished, leaving but a few skeletal remains. However, the barnacles were still excessive, readily revealing their inimical effects upon the young oysters. In addition to setting upon the oysters' valves and covering the slides, the combined outward and upward growth of the barnacles forced spat from the glass. While many of these were still alive, they were no longer attached and therefore recorded as losses.

On slides B, it was found that the initial mortalities per surface showed a wider variation than the final mortalities, as the continuous action of detrimental factors tended toward equalization of losses on all surfaces. This is illustrated by a range of 42.8 per cent, from 68.6 to 25.8, on August 23 and a range of 27.9 per cent, from 97.1 to 69.2, on January 8.

Slides C

Slide series C were placed directly into the bay from the rearing tank on July 27. They were removed at intervals for the determination of mortalities until November 6, when the studies were terminated. On August 24, slide C-9 was found to be broken and thus eliminated. Accumulative mortalities calculated from four two-week periods and one seven-week period in the bay are presented in table 16 as percentages.

After 102 days, from July 27 to November 6, mortalities for the four main types of surfaces ranged from 93.5 to 82.3 per cent, being greatest on upper smooth and least on lower ground. Smooth surfaces with 92.5 per cent received a slightly higher mortality than ground with 88.0 per cent. Upper surfaces with 90.5 per cent

Table 16. Spat mortalities, slide series C, through various experimental periods in the bay, July 27 to November 6, 1954; percentage of accumulated mortality from first enumeration in parenthesis.

Slide Surface	Set July 27	Numbers of Surviving Spat from Bay				
		Aug. 10	Aug. 24*	Sept. 7	Sept. 21	Nov. 6
U. S.	1,077	492 (54.5)	380 (64.7)	264 (75.5)	198 (81.6)	80 (93.5)
L. G.	520	407 (21.7)	334 (35.8)	198 (61.9)	130 (75.0)	90 (82.3)
U. G.	2,674	1,835 (31.4)	1,314 (45.1)	800 (66.6)	630 (73.7)	260 (89.2)
L. S.	138	69 (50.0)	57 (50.6)	43 (62.9)	38 (67.2)	19 (83.6)
Ground	3,194	2,242 (29.8)	1,648 (43.5)	998 (65.8)	760 (73.9)	350 (88.0)
Smooth	1,215	561 (53.8)	437 (63.4)	307 (74.3)	236 (80.2)	89 (92.5)
Upper	3,751	2,327 (37.9)	1,694 (51.2)	1,064 (69.4)	828 (76.3)	330 (90.5)
Lower	658	476 (27.7)	391 (38.5)	241 (62.1)	168 (73.6)	109 (82.9)
Totals	4,409	2,803 (36.4)	2,085 (49.2)	1,305 (68.2)	996 (75.8)	439 (89.3)

* Slide C-9, with 172 spat on upper ground and 1 spat on lower smooth, was broken so the following spatfall were used in determining per cent from 8-24-54 on.

Upper ground	2,397	Ground	2,917	Upper	3,474	Total	4,110
Lower smooth	116	Smooth	1,193	Lower	636		

mortality lost more spat than lower with 32.9 per cent. When first submerged in the bay, slides C received a high mortality of spat on smooth surfaces, further mortalities tending toward equalization of these losses on all surface combinations.

For this period, the mortality for all slides totaled 89.3 per cent, giving a survival of only 10.7 per cent. The first check on August 10, after 14 days in the bay, showed the highest numbers of perishing spat with 36.4 per cent. The majority of spat in stage I when first enumerated were believed to have been missing two weeks later from the conditions encountered in the bay, as the remaining seed oysters indicated exceptionally good growth.

Marine organisms first appeared in profusion on September 7, after the carton had been immersed six weeks in the bay, figure 14. Barnacles at this time were small but their continued increase in size were later detrimental to numerous seed oysters.

Slides D

On August 27, slide series D were placed directly into the bay from the rearing tank for two two-week experimental periods. The attached spat showed little indication of growth when first examined on September 14.

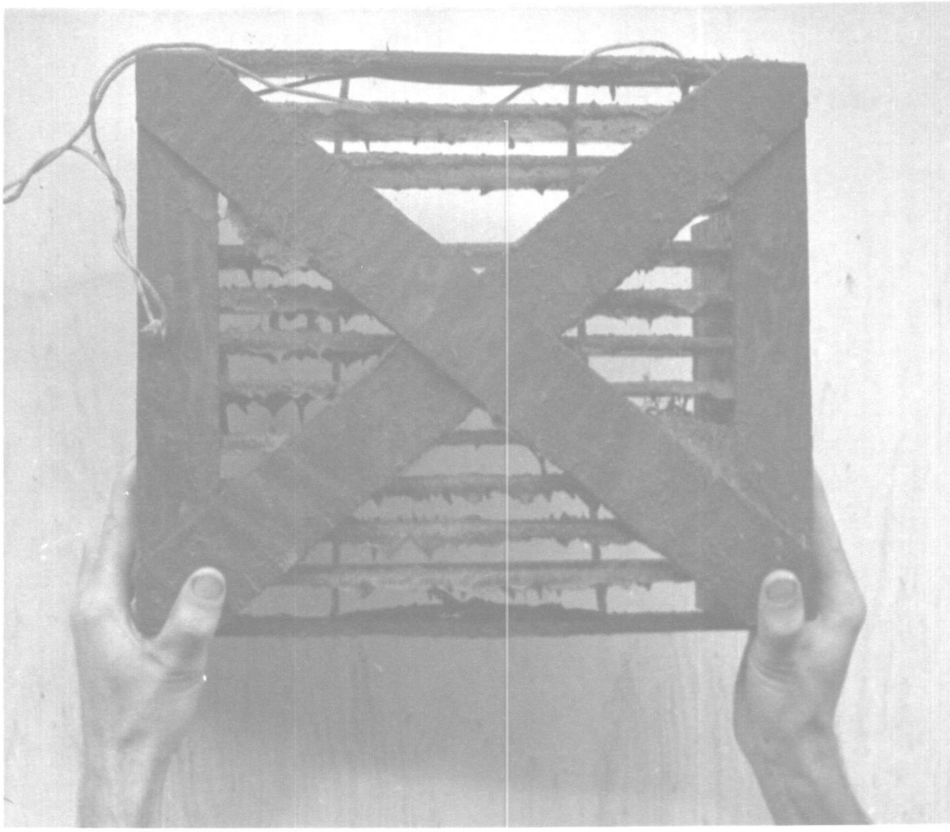


Figure 14. Growth of marine organisms upon carton and enclosed slide series after six weeks immersion in the bay, September 7, 1954.

Since the greatest spatfall occurred on the upper surfaces of these slides, it was believed that many larvae settled downward in a weakened condition resulting in abnormal attachments. Due to the lack of apparent growth upon fixation, future mortalities were expected to be great. The determined accumulating mortalities are given in table 17 as percentages.

After one month in the bay, mortalities had taken an excessive 98.3 per cent of the original spatfall, 95.0 per cent having perished during the first two weeks. Because of the high mortality no definite conclusion concerning the value of surface types for spat survival could be

Table 17. Spat mortalities, slide series D, through two-week experimental periods in the bay, August 27 to September 28, 1954; percentage of accumulated mortality from first enumeration in parenthesis.

Slide Surface	Set Aug.27	Numbers of Surviving spat from bay Sept. 14		Sept. 28	
Upper Smooth	270	5	(98.1)	1	(99.7)
Lower Ground	124	13	(89.5)	4	(96.8)
Upper Ground	290	8	(97.2)	0	(100.0)
Lower Smooth	134	15	(88.8)	9	(93.3)
Ground	414	21	(95.9)	4	(99.0)
Smooth	404	20	(95.0)	10	(97.5)
Upper	506	13	(97.4)	1	(99.8)
Lower	258	28	(89.1)	13	(95.0)
Totals	818	41	(95.0)	14	(98.3)

reached. It is possible that had slides D been held in the inside tank for two weeks, providing the spat with opportunity to establish themselves by growth, the survival would have been greater. However, it is unlikely that such an action would have proved beneficial because of the apparent abnormal condition of the spat.

Slides E

Slide series E were placed in the outside holding tank on September 7. During the examination on September 21, twelve slides were broken. Thus, mortality data was calculated only from the five remaining slides for two two-week periods in the outside tank and two longer periods in the bay, table 18.

After 123 days, from September 7 to January 8, mortalities on the four main types of surfaces ranged from 97.7 to 87.1 per cent, being greatest on upper smooth and least on lower ground. Smooth surfaces with 94.1 per cent received a higher mortality than ground surfaces with 86.8 per cent. Upper surfaces with 93.9 per cent received higher losses than lower surfaces with 71.7 per cent.

For this period the total mortality for all slides was 88.1 per cent, giving a survival of only 11.9 per cent. Again the initial mortalities per surface showed a wider variation than the final mortalities and the

Table 18. Spat mortalities, slides E-1, 5, 11, 16 and 17* through various experimental periods in the outside tank and bay, September 7, 1954 to January 8, 1955; percentages of accumulated mortality from first enumeration in parenthesis.

Slide Surface	Set Sept. 7	Numbers of Surviving Spat: Outside tank: Sept. 21		Outside tank: Oct. 5		Bay: Nov. 6		Bay: Jan. 8	
U. S.	45	6	(86.6)	1	(97.7)	1	(97.7)	1	(97.7)
L. G.	76	48	(36.8)	36	(52.6)	29	(61.8)	25	(67.1)
U. G.	234	65	(72.2)	29	(87.6)	20	(91.5)	16	(93.2)
L. S.	23	7	(69.6)	4	(82.6)	4	(82.6)	3	(87.0)
Ground	310	113	(63.5)	65	(79.0)	49	(84.2)	41	(86.8)
Smooth	68	13	(80.9)	5	(92.6)	5	(92.6)	4	(94.1)
Upper	279	71	(74.6)	30	(89.2)	21	(92.5)	17	(93.9)
Lower	99	55	(44.4)	40	(59.4)	33	(66.6)	28	(71.7)
Totals	378	126	(66.6)	70	(81.5)	54	(85.7)	45	(88.1)

* The remainder of slides E had been broken so the data concerns only these surviving slides.

greatest number of spat perished during the first four weeks when in early stages of development.

The growth of fouling organisms, with the exception of barnacles, proved less a problem as the slides were not installed into the floating live box until fall. At this time the bay water had gradually become cooler, reducing the attachment and growth of these creatures. The lowering of the water's temperature, while slowing the growth rate, did not reduce the numbers of attaching barnacles.

Slides F

Slide series F received the most abundant set obtained in all larvae rearing attempts on August 6. The majority of spat indicated traces of growth at the initial examination and were believed to be exceptionally healthy individuals. These slides were first installed in the inside holding tank on August 6 for two two-week periods, being placed into the bay on September 3. By November 6, the numerous spat had increased in size so that crowding conditions existed upon the slides. This intercompetition plus the growth of bay organisms made enumeration difficult on lower ground surfaces where the seed oysters were exceptionally abundant. Some of these surfaces were not counted at this time because of possible resulting inaccuracies. When the slides were finally removed from the bay on January 8, most glass plates were missing from the carton as a result of wave action from winter storms. The accumulated mortalities, determined from periods in the inside tank and bay, are presented in table 19 in percentages.

After 92 days, from August 6 to November 6, mortalities on the four main types of surfaces ranged from 92.4 to 67.3 per cent, being greatest on upper smooth and least on lower ground. Smooth surfaces with 84.9 per cent received a higher mortality than ground surfaces with 78.9 per cent. Upper surfaces with 90.0 per cent had a greater mortality than lower surfaces with 67.8 per cent. Again the range of early mortalities on all surfaces was quite wide, extending from 12.8 to 80.7 per cent. Each succeeding enumeration resulted in a narrowing of this range.

Once these slides were in the bay the presence of fouling marine organisms was noticeable. Due to the heavy spatfall, more space was required by the rapidly growing oysters than was available. This resulted in inter-competition among the spat as well as competition with attaching barnacles. It was likely that a lighter set upon lower ground surfaces would have proven advantageous to the survival of spat. By November 6, the barnacles were quite large, resulting in considerable loss of seed

Table 19. Spat mortalities, slide series F, through various experimental periods in the inside holding tank and bay, August 6 to November 6, 1954; percentage of accumulated mortality from first enumeration in parenthesis.

Slide Surface	Set Aug. 6	Numbers of Surviving Spat: Inside tank, Aug. 20		Inside tank, Sept. 3		Bay, Sept. 17		Bay, Nov. 6*	
U. S.	2,401	464	(80.7)	340	(85.8)	261	(89.1)	183	(92.4)
L. G.	13,802	12,036	(12.8)	9,199	(33.3)	7,040	(58.9)	458	(67.3)
U. G.	3,338	1,715	(48.7)	1,324	(60.3)	1,117	(66.5)	254	(87.1)
L. S.	1,078	745	(30.9)	565	(47.6)	461	(57.2)	341	(68.3)
Ground	17,140	13,749	(19.8)	10,523	(38.6)	8,157	(52.4)	712	(78.9)
Smooth	3,479	1,209	(65.2)	905	(74.0)	722	(79.2)	524	(84.9)
Upper	5,739	2,177	(62.1)	1,664	(71.0)	1,378	(76.0)	437	(90.0)
Lower	14,880	12,781	(14.1)	9,764	(34.4)	7,501	(49.6)	799	(67.8)
Totals	20,619	14,958	(27.5)	11,428	(44.6)	8,874	(56.9)	1,283	(81.6)

* Several lower ground surfaces were impossible to enumerate at this time due to the crowded conditions of the spat and overlying barnacles. Only slides possible to enumerate went into these figures. Spatfalls used in determining percentages:

Lower Ground	1,401	Ground	3,377	Lower	2,479
Upper Ground	1,976	Upper	4,377	Total	6,856

oysters.

Discussion

The value of holding spat under controlled conditions prior to placement in the bay was quite questionable and might have depended upon the character of the set as received from the rearing tanks, table 20. Ordinarily the greatest mortalities were found to have occurred two weeks following the termination of spatfall, when the young spat were in their initial stages of growth.

Slide series A, B, and E received mortalities of 26.2, 50.6 and 66.6 per cent after two weeks in the outside

Table 20. Percentages of accumulating spat mortalities on all slide series after two and four week periods from rearing tanks; series F held in inside tank, series A and E in outside tank, series C in bay, series B in both outside tank and bay.

Slide Series	Inside tank		Outside tank		Bay	
	2 weeks	4 weeks	2 weeks	4 weeks	2 weeks	4 weeks
A	----	----	26.2*	----	----	----
B	----	----	50.6	----	70.8	----
C	----	----	----	----	36.4	49.2
E	----	----	66.6	91.5	----	----
F	27.5	44.6	----	----	----	----

* Slides accidentally broken at end of first two weeks from rearing tank.

holding tank. Many of the spat may have perished due to the settlement of silt from freshly pumped bay water. However, slides E were suspended with their surfaces vertical, eliminating the problem of siltation, yet a high loss occurred.

Slides F were held in tank 4 for two two-week periods and received low mortalities of 27.5 and 44.6 per cent. While the survival was comparatively high, these spat also showed every indication of excellent growth immediately upon setting. Direct placement into the bay might have resulted in a comparable survival. This is supported by slides C which were immediately immersed in the bay following spatfall. A loss of 36.4 per cent occurred the first two weeks, increasing to 49.2 per cent in four weeks, which was slightly greater than on slides F.

Therefore, it appeared that, if the set consisted of normal individuals, direct submergence in the bay was unlikely to result in excessive mortalities and a developmental period in either the inside or outside holding tanks may have been unnecessary. Slides in the floating live box were exposed to the continuous action of bay currents, which prevented accumulations of silt and provided constant access to nutritive substances and food organisms.

The value of various glass surfaces for spat survival was found to be more definitely defined, table 21. For all slide series containing apparently normal spat, upper smooth surfaces had received the greatest mortality at the conclusion of the examinations, followed by upper ground and lower smooth. Lower ground surfaces received the least mortality, indicating that this surface combination is best for the survival of seed oysters. Also, smooth surfaces sustained higher losses of spat than ground and upper surfaces received greater mortalities than lower. In all cases, the per cent of mortalities the first two weeks following spatfall varied widely with the four combinations of glass surfaces; the percentages of accumulated mortalities on each surface type at the end of the experiments (12 to 24 weeks) showed a smaller variation.

Over an average period of 98 days and under various environmental conditions, the per cent of mortality for all slide series was determined as 74.4.

Known factors which, when present, resulted in the foremost losses of young oysters were silt accumulation and barnacle competition. In the outside tank, silt settlement resulted in high mortalities upon upper horizontal surfaces. In the floating live box, silting was less a problem because of the action of bay currents. Had the glass slides been placed on the bottom of the bay the

Table 21. Percentages of mortalities upon various glass surfaces, slide series A, B, C, E and F, at the termination of observations; the average mortality for each type of surface and the average total mortality being determined for the first 98 days from the rearing tank.

Slide Series	Days from Rearing tank	SLIDE SURFACE								
		U.S.	L.G.	U.G.	L.S.	Ground	Smooth	Upper	Lower	Total
A	14	36.6	21.7	26.1	28.6	23.8	35.2	29.4	22.3	26.2
B	168	97.1	69.2	90.2	88.9	80.9	95.5	93.7	71.4	87.0
C	102	93.5	82.3	89.2	83.6	88.0	92.5	90.5	82.9	89.3
E	124	97.7	67.1	93.2	37.0	86.8	94.1	93.9	71.7	88.1
F	82	92.4	67.3	87.1	68.3	78.9	84.9	90.0	67.8	81.6
Averages	98	93.5	61.3	77.2	71.3	71.7	80.4	79.5	63.2	74.4

detrimental effects of mud could not have been escaped.

Marine organisms grew profusely upon the slides and retaining cartons during the summer, making their appearance two to four weeks after the slides were first immersed in the bay. Most of these organisms, with the exception of barnacles, Balanus sp., had perished by mid-winter. In addition to growing upon the seed oysters and glass slide, the combined outward and upward growth of the rapidly increasing barnacles forced spat from the glass. An estimated 10 per cent of the seed oysters were lost during the winter from this action alone.

Competition for space by the seed oysters was noticed upon the lower ground surfaces of slides F, which received an exceptionally heavy set of 31.9 spat per square inch. The young spat tended to crowd against each other, prohibiting normal outward growth and contributing to the overall mortality. In future laboratory rearings, the introduction of sufficient quantities of suitable cultch to the tanks would possibly result in a lighter set per square inch. Thus a higher percentage of spat might eventually reach the full adult size.

GROWTH OF LATER SPAT STAGES

Young seed oysters from laboratory rearings usually were quite large and firmly attached to the glass slides two months following the termination of the spatfall. An attempt was made at this time to determine the growth rates of certain sets when subjected to various environmental conditions. While the data collected cannot be accepted as true average rates of growth due to inadequacies of the sampling method, an indication of the rate of shell formation was obtained.

Measurements were taken from slide series B, C, E and F during latter examinations for mortality. All individuals were selected at random from various glass slides and measured with calipers. The measurements were taken in the direction of an oyster's greatest growth, at right angles to the hinge. To avoid bias due to personal selection of individuals, once a glass surface was chosen each attached spat was measured. Factors considered in the selection of samples included the loss of certain individuals to mortality. These losses were likely to have been greater on slower growing spat, increasing the average daily shell formation of the larger surviving oysters. Measurements were not taken from slides containing a high abundance of seed oyster, the crowded conditions plus the presence of barnacles handicapping accuracy. Growth data

Table 22. Growth of oyster spat obtained from laboratory rearings, July to September, 1954.

	Date*	No. Spat Measured	Days from Rearing Tank	Average Size (mm)	Average Growth Per Day (mm)
Slides B	9-20-54	88	56	4.44	.079
	11-6-54	44	103	9.0	.086
	1-8-55	35	166	10.48	.063
Slides C	9-21-54	108	56	3.93	.070
	11-6-54	80	102	7.2	.070
Slides E	11-6-54	51	60	3.72	.062
	1-8-55	42	123	4.2	.039
Slides F	9-17-54	97	42	2.69	.0875
	11-6-54	53	92	6.23	.068

* All spat were measured following at least two weeks in bay.

of spat obtained from glass slides is presented in table 22.

Before the first measurements were obtained on September 20, slides B were installed in the inside tank for two weeks, two weeks in the outside tank and four weeks in the bay. The average growth rate per day for the spat measured at this time was .079 mm. By November 6, this had increased to .086 mm., dropping to .063 mm., by January 8. This average daily decrease was believed due to the lowering of bay temperatures during the winter months.

Slides C were placed directly into the bay from the rearing tank. The sample measured on September 21 indicated an average daily growth rate of .070 mm. for 56 days. By November 6, 46 days later, this daily average growth rate remained at .070 mm.

Two weeks in the outside tank and 46 days in the bay preceded the first measuring of spat on slides E. An average daily growth rate of .062 mm. was determined on November 6. Nine weeks later on January 8 this average had dropped to .039, which was believed caused by lower winter temperatures in the bay.

Slides F were held in the outside tank four weeks before being placed in the bay. On September 17, a 42-day growth period indicated an average daily growth of .0875 mm. for the spat measured. On November 6 this had decreased slightly to .068.

The data obtained indicates little determinable variance in daily growth of spat held under controlled conditions. Once in the bay, growth remained uniform until the onset of falling winter temperatures, which curtailed feeding activities of the seed oysters. By November 6, abundant populations of barnacles had settled upon all slides which may have decreased the rate of growth. Observations on lower ground surfaces of slides F, with 31.9 spat per square inch, revealed that spat crowded together grew less rapidly than spat that were scattered because of competition for space.

SUMMARY

1. Studies concerning the setting of free-swimming native oyster larvae in rearing tanks, the mortalities and the growth of young artificially reared oysters under controlled conditions were conducted at the Yaquina Bay Fisheries laboratory from June 1954 to January 1955.
2. Preliminary experiments showed that a wide variation existed in the numbers of spat attaching to both old and new shells, that graphite pencil marks appeared to have no inimical effects upon young oysters, and that a thirty-minute air exposure period was a fairly safe time limit for removal of spat from rearing tanks.
3. Spatfalls were obtained upon seven series of glass slides during the summer's rearing attempts, with the average numbers of spat attaching to each series ranging from .24 to 12.6 spat per square inch. The numbers of spat attaching were independent of the number of free-swimming larvae placed in the tank.
4. The greatest numbers of spat were usually found upon lower ground surfaces of glass slides, the smallest numbers upon lower smooth. A definite preference for ground surfaces was indicated by free-swimming larvae.

5. Intensities of spatfall upon upper and lower surfaces may have depended on the physical condition of each group of larvae, individuals of poor health tending to settle downward under the influence of gravity.
6. The spatfalls obtained indicated a greater set in the top three quarters of the rearing tanks than in the lower quarter, presumably because of insufficient current and perhaps inadequate aeration in the lower area.
7. Observations of young spat revealed that normal individuals began growth of the new shell immediately after setting and were firmly attached to the glass after a seven day period. These spat showed an average mortality of 23.0 per cent seven days after setting while retained in the rearing tanks.
8. Holding periods in an inside tank, an outside tank, or in a floating live box indicated that, if a set consisted of normal rapid-growing individuals, direct submergence into the bay was unlikely to result in excessive mortalities of spat.
9. Lower ground surfaces of glass slides received the least mortalities of spat, while upper smooth surfaces received the greatest mortalities, at the conclusion of the examinations. Also, smooth surfaces had greater mortalities than ground and upper surfaces

greater mortalities than lower.

10. The greatest mortalities on all slide series were found to occur within two weeks after the termination of setting, when the young spat were in initial stages of growth.

11. The per cent of mortalities the first two weeks following spatfall varied widely with the four combinations of glass surfaces; the percentages of accumulated mortalities on each surface type at the end of the experiments (12 to 24 weeks) showed a smaller variation.

12. Over an average period of 98 days and under various environmental conditions, 74.4 was the average per cent of spat mortality determined for all slide series.

13. Known factors which resulted in the greatest losses of seed oysters were deposits of silt and growths of barnacles, Balanus sp.

14. Measurements of spat showed that growth was quite uniform on all slides under all conditions, averaging approximately .062 to .087 mm. a day, until the lowering of bay temperatures occurred in the fall.

15. Spat crowded together (in densities of 31.9 spat per square inch) were observed to show less growth than spat that were scattered due to direct competition for space.

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