

Davidson's fixative (Shaw and Battle 1957) immediately upon collection. After 24-48 h, they were transferred to this same fixative, but without acetic acid, for long-term storage. Samples were shipped to the HSRL for histological processing and analysis. Small oysters which could not be readily dissected *in situ* were fixed whole, following careful prying open of their valves. Large oysters were shucked, and their shells were discarded. The shells of small oysters had decalcified by the time the oysters were processed for histology. Samples examined at Battelle were shipped approximately every 2 weeks, from mid-June through mid-November, by overnight mail from the Flower Co. They were then decalcified and fixed in Dietrich's fixative. A number of individuals were examined prior to fixation for evidence of fungal infection or other organisms that might have penetrated the shell.

Fixed tissues of large oysters were cut laterally from the hinge region, through the adductor muscle, to the posterior margin before being embedded in paraffin. Small individuals with decalcified shells were embedded intact with the ventral side down. This orientation allowed us to view the epitheliums under the hinge ligament, the myoepithelial attachment of the adductor muscle to the shell, the abnormal conchiolin layer when present, and the periostracum-secreting mantle edge, as well as various internal organs.

Embedded tissues were sectioned serially at 5-6 μ m, and stained with hematoxylin and eosin, or a Masson's Trichrome stain (Humason 1979) modified by the addition of Fast Green and Orange G. An initial sample of 55 individuals, collected during peak mortality and categorized as showing a) no sign of distress, b) early distress (some overgrowth of the left valve, some conchiolin deposition of inner valve, or both), or c) advanced distress (clear overgrowth of left valve, heavy abnormal conchiolin, weak muscle attachment), was examined microscopically for histopathological conditions. Pathological conditions recognized in the initial sample (see Results) were identified and rated (none, light, moderate, or heavy) in subsequent bi-weekly samples.

For transmission electron microscopy, mantle tissue and conchiolin were fixed in 2% glutaraldehyde and 1.570 paraformaldehyde in 0.1 M Millonig's phosphate buffer with 2.7% glucose at pH 7.3 for 2 hr followed by buffer rinses in 0.2 M Millonig's phosphate buffer with 5.4% glucose at pH 7.3, then post-fixed in 1% OsO₄ with 0.1 M Millonig's and 2.7% glucose at pH 7.3 for 1 h. After washing in distilled water, the tissue and conchiolin blocks were stained in 1% aqueous uranyl acetate for 1 h followed by dehydration in a graded series of ethanol solutions and transfer into propylene oxide. Infiltrations and embedding were made in Spurr's resin and thin sections were stained in Reynold's lead citrate and uranyl acetate. All procedures were accomplished at room temperature except 1) post-fixation which was done in an ice bath, and 2) resin polymerization which was performed at 58°C.

Sampling Program at the Fishers Island Study Site

A similar sampling protocol was implemented at a more oceanic site on the north shore of Fishers Island, NY: 2 cohorts of cultchless juvenile oysters were deployed in pearl nets (34 x 34 cm basal area), in vertical arrays of 4 nets, on June 12, 1991, in eastern West Harbor (depth = ca. 3 m at low water). Both cohorts were produced at the Aquacultural Research Corporation hatchery, Dennis, MA, using broodstock shipped from Island Pond, Fishers Island (G. Matthiessen, Ocean Pond Corp., pers. comm.). The small (1991) cohort (SC) was grown in trays at Ocean Pond Corp.'s nursery site in Island Pond, prior to transfer to West Har-

bor in mid-June. The large (1990) cohort (LC) overwintered in pearl nets in Island Pond until June 1991. Large cohort oysters were held in pearl nets with a 6 mm mesh, whereas SC oysters were held in 3 mm pearl nets (see Table 1 for initial sizes and stocking conditions). Low stocking-density groups were also maintained at this site (50 and 100 large and small oysters per pearl net respectively), and sampling was without replacement, as described previously. Live oysters were shipped to SUNY Stony Brook by overnight mail in coolers containing freeze-packs. Experimental oysters were thinned but not graded or culled, except at the time of initial deployment.

RESULTS

Sampling at the Oyster Bay Study Site

Mortality Patterns

Mortality estimates obtained *in situ* were generally in excellent agreement with those determined following dissection of oysters in the laboratory. Greatest discrepancies occurred on July 11, when field-determined mortalities were 4-9% and 9-12% for small and large oysters respectively, while laboratory-derived values were 10-15% and 18-21% respectively.

Despite variability between replicate stacks at any given sampling date, especially for small oysters, there was no consistent trend showing greater losses within one of the two replicates. Thus, these differences are attributed to sampling artifact, and data averaged for the two replicates.

Cumulative mortalities remained negligible until July 11, when they reached 12% and 20% for the small and large cohort, respectively, at a time when the water temperature reached 24°C (Fig. 1). Mortalities peaked at 62% and 54% on July 26 for small and large cohorts, respectively, and ceased thereafter. Grading/culling conducted on August 9 was not 100% effective in removing dead oysters from the LC. Therefore, an apparent increase in mortalities of large oysters between August 9 and August 23 is attributed to sampling error, rather than new mortalities. This was confirmed by determining the mean size of dead oysters over time (Fig. 1B), which remained constant, at 20.3-20.8 mm [the size of oysters in early July (Fig. 2)], between August 9 and September 20. Thus the dead oysters present in late August samples are clearly remnants from the July mortality outbreak.

Losses were consistently lower for oysters stocked at the lower density. They attained maxima of only 30-37% for small oysters and 28-38% for large oysters on July 26, but their timing coincided with that of oysters held at high density. Mortalities of the LC levelled off between 40 and 47% after August 23 (Fig. 1B). The outbreak of mortalities occurred during a period of elevated surface temperatures (range = 21.7 to 25.0°C), which were maintained between June and early September (Fig. 1). A comparison of Flower's temperature records from 1987 through 1991 (May 1 to July 26) indicates that late spring water temperatures were higher than usual in 1991. The mean for May was 17.2°C in 1991, compared to 14.2 and 15.3°C in 1990 and 1989, respectively. Cumulative day-degrees calculated for May were 8 to 16% higher in 1991 than in the 4 previous years. However, heavy oyster mortalities were also experienced in 1990, when the lowest May to July temperatures were recorded. Salinities ranged between 24 and 28 ppt, and only moderate to low levels (in late September and early October) of fouling and siltation of trays were observed throughout the study period.