International Council for the
Exploration of the Sea.

C.M. 1955
Shellfish Committee
No. 23

Factors affecting the pollution and self-purification of molluscan shellfish in a polluted estuary

4

P. C. Wood

Introduction

In this country little attention has been directed towards the seasonal variations of pollution which occur in oysters, although this aspect of shellfish pollution has been studied by workers in the North Atlantic States of America and Canada. Smith (1912) reported the coliform content of oysters taken from Narragansett Bay to be considerably lower in winter than in the summer, although there was no corresponding change in the number of coliforms in the seawater. Other workers reported similar findings, (Gorman 1912; Cumming 1916; Gage and Gorman 1925; Gibbard et alia, 1942) and now-a-days legislation in certain areas of the North Atlantic seaboard allows for the sale in winter of oysters from grounds which produce grossly polluted oysters in the warmer months (Needler 1951; Homans 1955).

Not many previous observations are available for England and Wales. In the 1930's during an investigation at the Conway Laboratory of the Ministry of Agriculture, Fisheries and Food, of methods for the cleansing of oysters under conditions of low water temperature, it was noted that oysters taken in winter from grounds which produced highly polluted mussels were virtually unpolluted. The significance of this was not appreciated at the time and oysters for experiment were later placed either in tanks of diluted sewage or directly beneath a sewage outfall. Such oysters could not fail to become highly polluted since the residual water within the mantle cavity alone would contain sufficient bacteria to give small samples of the tissue the appearance of gross contamination.

More recently, Cole (1954) in a limited series of observations, noted that oysters from a polluted ground of the river Crouch contained few bacteria of faecal origin during winter, whereas large numbers were present in the warmer spring and summer months. He pointed out that temperature mainly affected the rate of filtration of oysters, and did not consider that the observed variation of pollution was associated with seasonal changes in the amount of sewage entering the river, since the period of maximum population, namely August and September, did not co-incide with the period of maximum pollution of oysters. No observations were made of the incidence of Bact. coli in waters over the grounds during these experiments, although Vaccaro et alia (1950) showed that these organisms remain viable in seawater for a longer period in winter than summer.

It was thought desirable to obtain more data and to extend the observations to include the mussel (Mytilus edulis). A preliminary investigation into the seasonal variation of pollution was, therefore, started in 1954 and samples of native oysters (Ostrea edulis), mussels and water were collected simultaneously from a fixed sampling point on the Southward Laying, River Crouch, at weekly intervals.

The samples were examined bacteriologically for the presence of faecal Bact. coli using methods described later and it appeared that a rise in water temperature was associated with an increase in the counts of faecal Bact. coli per ml. of oyster tissue. Although observations with mussels were not made when water temperatures were extremely low, a similar, although less obvious correlation was evident. Water counts showed wide fluctuations which did not appear to be correlated with the levels of pollution attained by the shellfish. At this stage of the experiments, water and shellfish samples were collected at or near low water, but no attempt was made to collect them precisely at the time of observed low slack water. It was thought probable that the variations observed in water pollution were the result of tidal movements which altered the bacterial quality of the water passing over the grounds.

It is established that polluted molluscan shellfish cleanse themselves when placed in tanks of clean water but little is known on the processes of pollution and cleansing of those shellfish which occur under natural conditions in situations where the level of pollution of the water varies within the tidal cycle. These variations were, therefore, studied and this led to a comparison of these processes in oysters and mussels during winter and summer months.

Methods

The Southward Laying of the River Crouch selected for these experiments is situated within the closed area proscribed in the Public Health (Shellfish) Regulations (1934) and is approximately half a mile downstream of the main sewage outfall of Burnham (population approximately 4,000 with seasonal increases). This outfall discharges virtually raw sewage through a tidal valve for a period of approximately 2 hours either side of low water. Downstream of the oyster ground there are no polluted outfalls, and since the river is tidal, the oyster ground is subject to the flow of highly polluted water on the ebb, and of virtually clean seawater on the flood, a set of conditions which is by no means uncommon in estuaries where cultivation is practised.

Bacteriologically clean oysters and mussels were placed in a weighted, buoyed, cage at a point on the shore just below low water of spring tides. To eliminate any interference in the functioning rate which might result from the disturbance of the cage at hourly intervals during the collection of samples, a number of smaller cages, each holding one sample, was later substituted for the single cage.

Oysters were selected for evenness of size and were normally 3 - 4 years old. In later experiments some attempt was made to use only oysters showing indications of rapid growth (broad, thin bands of newly deposited shell) on the assumption that these were of similar filtering capacity, but no significant differences were noted as a result of this refinement. The mussels were 2 - 3 years old, and were selected for evenness of size and smoothness of shell.

Sufficient shellfish for each experiment was placed in the cages and left for at least 24 hours before the commencement of sampling, in order that they might assume the bacterial characteristics of the ground. Samples of 4 oysters and 4 mussels were then collected at approximately hourly intervals throughout the day, each sample being placed in a separate sterile tin. At the same time, water samples were collected from the bottom of the river near the sampling point.

All samples were examined bacteriologically within a few hours of collection using a modification (Reynolds and Wood 1955) of the roll-tube method of Clegg and Sherwood (1947). Shellfish tissue of each sample was pooled, extracts prepared and four tubes containing 5 ml. of modified MacConkey agar each inoculated with 1 ml. of extract. The bacterial content of water was determined using 2 ml. of water in each of 5 tubes holding 5 ml. of MacConkey agar. After rolling, tubes were incubated overnight at 44°C in an accurately controlled water-bath. The red colonies representing faecal <u>Bact. coli</u> were counted and results finally expressed as the number of faecal <u>coli</u> in 1 ml. of tissue or 10 ml. of water.

Results and discussion.

From topographical evidence, wide variations would be expected in the bacterial quality of water passing over the sampling point, and this was confirmed by these tests. During each experiment, there appeared to be a well-defined cycle of pollution characterised by high counts of faecal <u>Bact. coli</u> around low water, and low counts during the flood of fresh water from the sea and on the first of the ebb before the sewer commenced to discharge. (See Figs. I, II and III which are representative of a larger series of similar results). This pattern was evident in all except one of the many experiments done (bottom of Fig. I), but the levels of pollution in each cycle differed considerably. This was probably the result of the influence of a number of varying factors, but the volume of sewage discharged and the strength and direction of the wind appeared to be the most important.

In most of the summer experiments there was a close correlation between the bacterial content of the water and of shellfish, but as experiments continued through the winter, it appeared that the characteristic pattern of pollution of the oyster in winter differed from that observed in summer. In some experiments the expected changes in the level of pollution did not take place. It will, therefore, be convenient to divide the results into three groups.

(a) Pollution in Summer (Fig. I)

When water temperatures were between 10 and 16.5°C. there was a close association between the bacterial content of water and shellfish. Both oysters and mussels became grossly polluted in the period around low water, when water showed its maximum count, but they quickly cleansed themselves with the flood of bacteriologically clean water from the sea. In the first experiment, mussels cleansed themselves within 1 hour of maximum pollution, and oysters did so after six hours. At these temperatures, the oyster and the mussel are known to filter at a rapid rate, and it is understandable that bacteria were quickly removed from their alimentary tracts with the flood of clean seawater. It is noteworthy that these experiments were conducted during periods of calm, when winds were light; the significance of this will be discussed later.

(b) Pollution in Winter (Fig. II)

The second group of experiments, also done in calm weather, but when water temperatures were between 1.5 and 2.1°C, showed that the pattern of pollution of the oysters was different from that of the summer. Mussels continued to function at what appeared to be a normal rate, becoming polluted and then clean in a manner similar to that occurring during summer, whereas oysters remained virtually unpolluted, even when mussels and water became grossly polluted at low water. Mention has already been made of the works of Gorman (1912), Cumming (1916), Gage and Gorman (1925) and Gibbard et alia (1942) who observed similar conditions on the coasts of the North Atlantic. Parsons (Hunter and Harrison 1928) described seasonal variations in the level of pollution of oysters, and demonstrated that pollution became very low when the water temperature fell to 3.5-5.5°C. Experimentally, Galtsoff (1928) and Loosanoff (1950), were unable to detect water propulsion in the American oyster at temperatures below 5°C. The filtration rate in the mussel is not affected by the reduced temperature to the same degree (Gray 1928), and active filtration continues in the mussel in temperatures down to 0°C, (Dodgson 1928); this has been confirmed during many years mussel cleansing at Conway.

There is little information regarding the mechanism by which oysters in polluted waters remain but lightly polluted at low temperatures. Needler (1941) has suggested that the reduced filtration rate is accompanied by a slower rate of passage of food through the gut, during which time viable bacteria are subjected to the bactericidal substances of the digestive juices for a longer period. The unpublished work done by H. Marshall Webb at Conway indicated the presence of a thermolabile bactericidal substance in oyster tissue.

(c) Abnormal pollution (Fig. III)

Both the previous groups of experiments were made during periods when there was little wind. In the third series of experiments, done under more turbulent conditions at varying temperatures, the expected correlation between the levels of pollution of water and shellfish did not appear. Successive samples frequently showed wide deviations from one another, this being more evident for oysters than mussels. It was clear that some other factor was interfering with the "normal" pattern of pollution, and although this situation has not yet been extensively examined, observations of the weather showed that this irregularity occurred during periods when onshore winds caused heavy turbulence. The experimental cages used in these experiments were situated at the level of low water spring tides and for a large part of these experiments were in relatively shallow water where turbidity would, in these circumstances, be excessive. It is highly probable that the filtration rate was modified by the presence of suspended silt. The influence of turbidity has been examined experimentally by Kellogg (1915), Loosanoff and Engle (1947), and Loosanoff and Tommers (1948). The last-named workers demonstrated that the filtration rate of the American oyster (Crassostrea virginica) was reduced by as much as 80% in the presence of relatively small quantities of silt.

The possibility of other factors causing these discrepancies cannot be ignored. Salinity and oxygen tension probably varied but little in these experiments, as seawater flooded in and out of the estuary at a rapid rate and there was little fresh water entering the river. Other workers have examined shellfish to see if there is an association between the filtration rate and tidal movements. Loosanoff and Nomejko (1946) failed to detect a tidal or diurnal periodicity in the filtration rate of the American oyster, but Rao (1954) recorded pronounced tidal rhythms in Mytilus edulis and the Californian mussel (Mytilus californianus) which showed maximum filtration rates at the time of high water. It is possible that such a cycle was in operation during these experiments, but it is highly unlikely that it would lead to the discrepancies observed here.

Significance of these experiments in relation to public health

The experiments described above indicate the dynamic nature of the pollution and cleansing of shellfish from grounds which are subjected to intermittent pollution. A single sample can give but little information of the sanitary quality of a shellfish ground, and, as previous workers have indicated (Dodgson 1928; Knott 1951), a representative series of samples must be collected under varying conditions of wind and tide.

On oyster beds, investigation during summer will normally show the maximum level of pollution to which the shellfish are subjected, but will not provide a reliable guide to winter conditions.

Dodgson (1928) recommended that the mussel was a more reliable indicator of pollution than the oyster, and frequently used mussels to determine the level of pollution of an oyster bed. In view of the physiological differences between these two shellfish, this is to be deprecated, since under certain conditions pollution of the oyster will be light yet that of the mussel heavy.

Before the seasonal variation of pollution of oysters can be put to economic advantage, further work is required. Firstly, the effect of suspended silt upon natural purification must be more closely examined, and secondly the upper limit of water temperature at which oysters laid on polluted grounds fail to become significantly polluted must be determined. With these questions answered, it might be possible to utilize oysters in winter from certain grounds exposed to contamination; the fact that active filtration continues at normal winter temperatures precludes this possibility for the mussel.

References

Clegg, L. F. L. and Sherwood, H. P. 1947. The bacteriological examination of molluscan shellfish. J. Hyg., Camb., 45, 504.

Cole, H. A. 1954. Purification of oysters in simple pits. Fish. Invest. Ser. II 18, 5.

Cumming, H. S. 1916. Investigation of the pollution and sanitary conditions of the Potomac Watershed. U.S. Pub. Hlth. Serv., Hyg. Lab. Bull. 104.

Gage, S. D. and Gorman, F. P. 1925. Self purification of oysters during liberation. Amer. J. Publ. Hlth., 15, 1057.

Galtsoff, P. S. 1928. Experimental study of the function of the oyster gills and its bearing on the problems of oyster culture and sanitary control of production, handling, and shipment. Bull. U.S. Bur. Fish., 44, 1.

Gibbard, J., Campbell, A. G., Needler, A. W. H., and Medcof, J.C. 1942. The effect of liberation on the content of coliform bacteria in oysters. Amer. J. Publ. Hlth., 32, 979

Gorman, F. P., 1912. Seasonal variation in the bacterial content of oysters. Amer. J. Publ. Hlth., 2, 24.

Gray, J. 1928. Ciliary movement, Cambridge University Press.

Homans, R. E. S., 1955. Personal communication.

Hunter, A. C. and Harrison, C. W. 1928. Bacteriology and chemistry of oysters, with special reference to regulatory control of production, handling and shipment. U.S. Dept. Agr. Tech.Bul. 64.

Kellogg, J. L. 1915. Ciliary mechanisms of Lamellibranchs, with descriptions of anatomy. J. Morph., 26, 625.

Knott, F. A. 1951. Memorandum on the Principles and standards employed by the Worshipful Company of Fishmongers in the bacteriological control of shellfish in the London markets. Fishmongers' Hall, London.

Loosanoff, V. L. 1950. The rate of water pumping and shell movements of oysters in relation to temperature. Anat. Rec., 108, 3.

Loosanoff, V. L. and Engle, J.B. 1947. Feeding of oysters in relation to density of micro-organisms. Science, 105, 2723.

Loosanoff, V. L. and Nomejko, C. A. 1946. Feeding of oysters in relation to tidal stages and to periods of light and darkness. Biol. Bull., 90, 244.

Loosanoff, V. L. and Tommers, F. S. 1948. The effect of suspended silt and other substances on the rate of feeding of oysters. Science, 107, 69.

Needler, A. W. H., 1941. Oyster farming in Eastern Canada. Bull. Fish. Res. Bd. Canada, No. 60.

Rao, K. P. 1954. Tidal rhythmicity of rate of water propulsion in Mytilus and its modifiability by transplantation. Biol. Bull., 106, 353.

Reynolds, N. and Wood, P. C. 1955. Improved techniques for the bacteriological examination of molluscan shellfish and sea-water. In the press.

Smith, G. H. 1912. Seasonal variation in the bacterial content of oysters. Science, 35, 235.

Vaccaro, R. F. et alia, 1950. Viability of Escherichia coli in sea water. Amer. J. Publ. Hlth., 40, 1257.

TIDAL CYCLE OF POLLUTION AND CLEANSING AT SUMMER TEMPERATURES

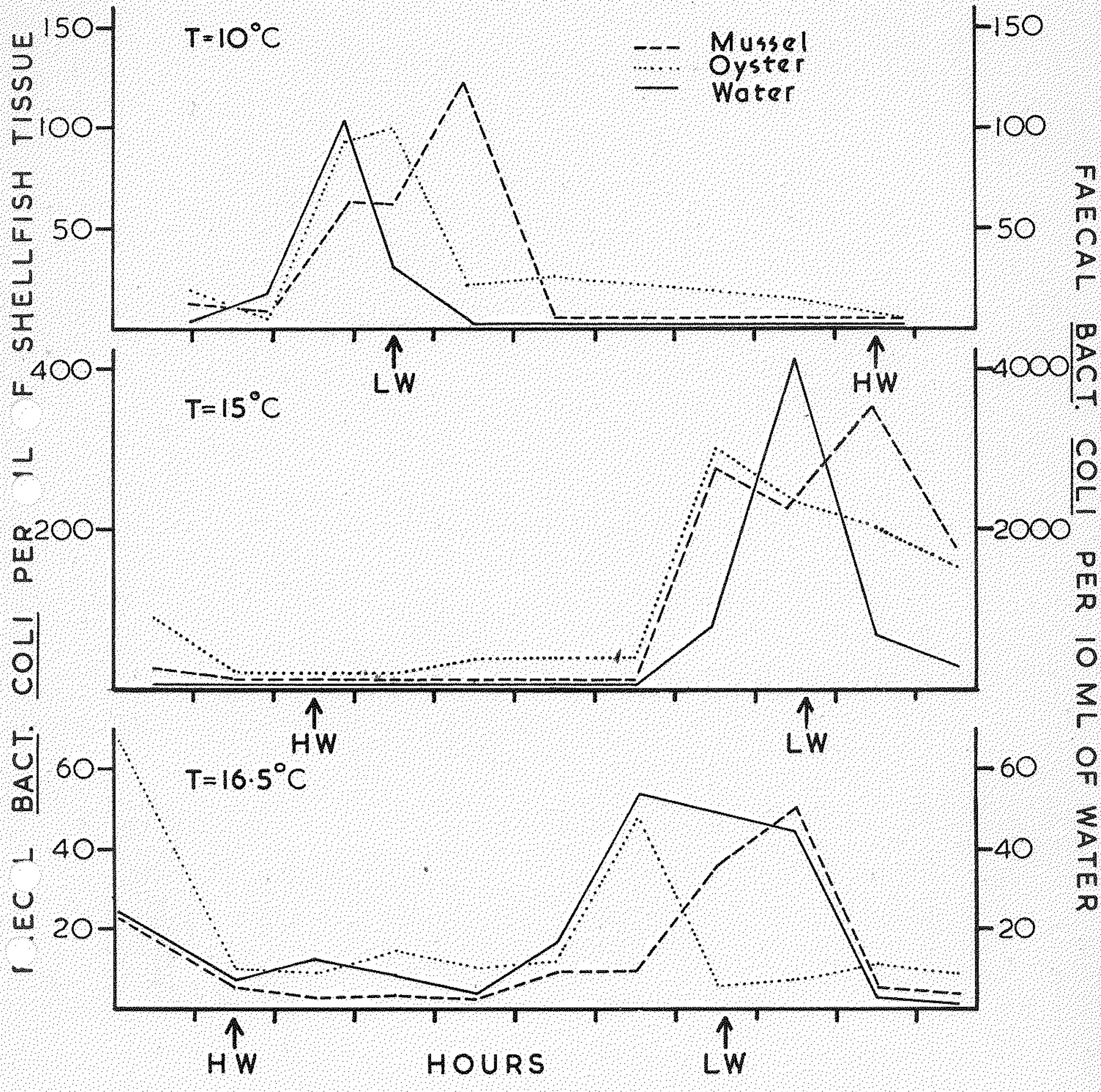

Fig. 1

TIDAL CYCLE OF POLLUTION AND CLEANSING AT LOW TEMPERATURES

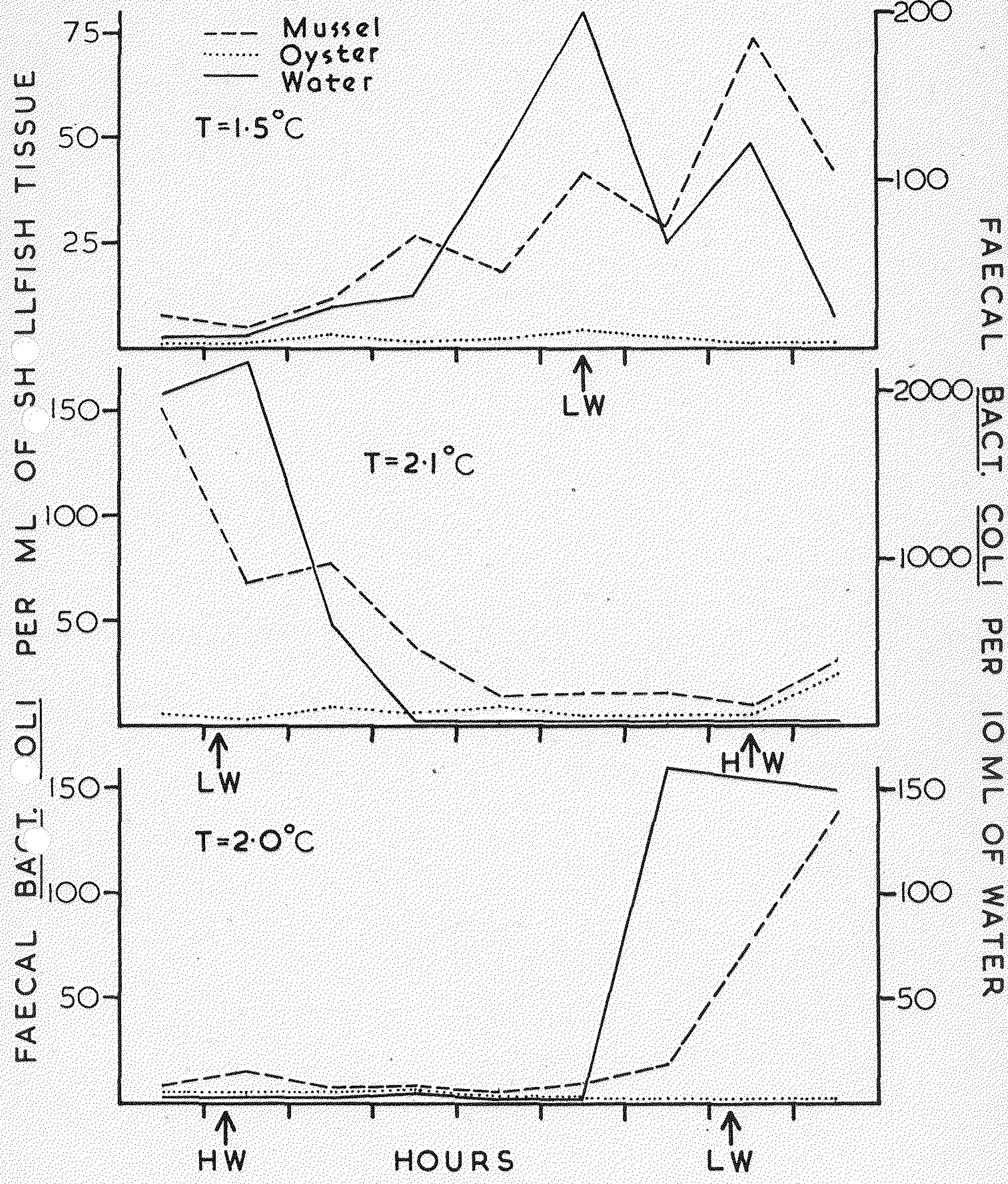

Fig. 11

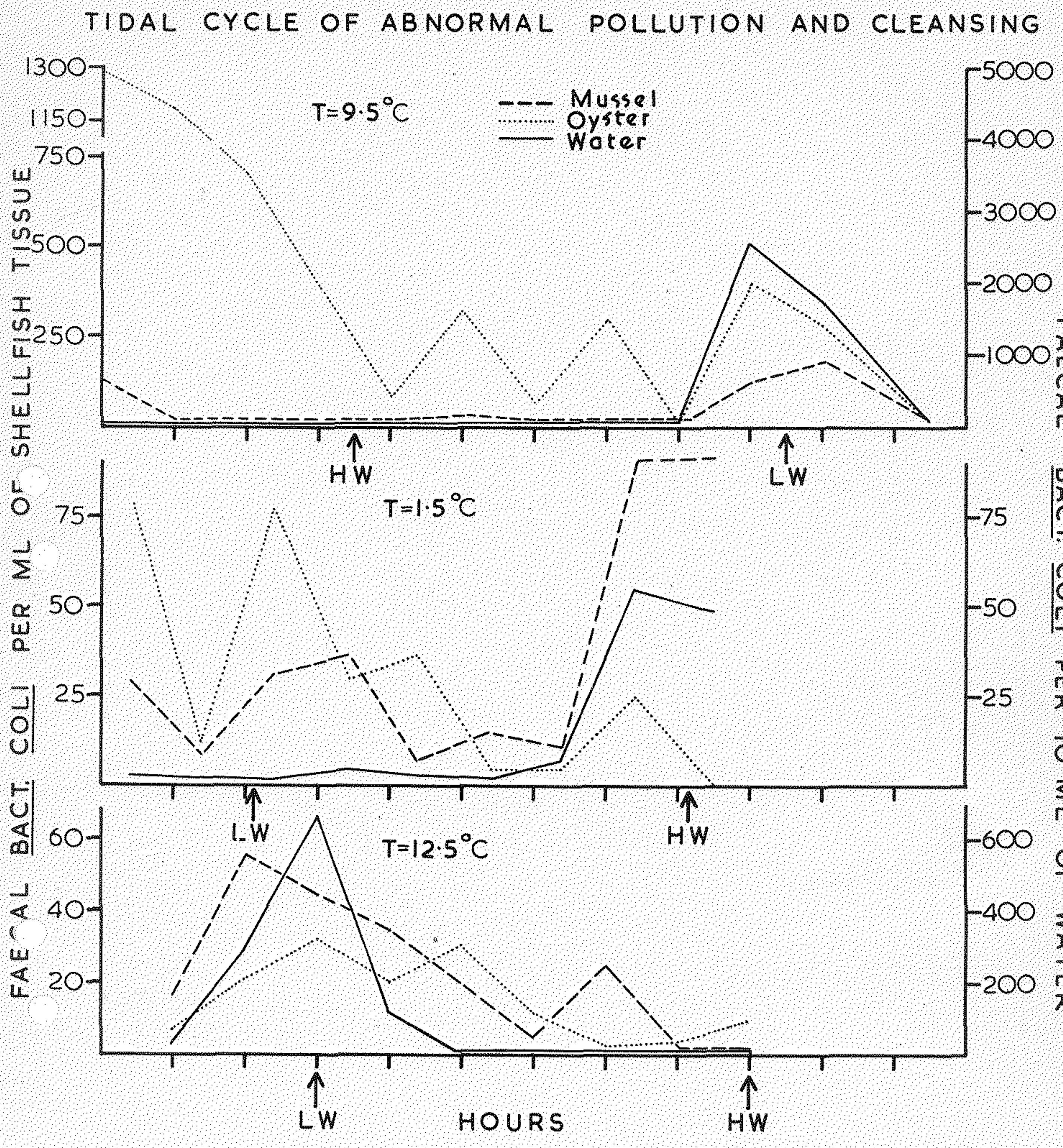
TIDAL CYCLE OF ABNORMAL POLLUTION AND CLEANSING
T=9·5°C
Mussel
Oyster
Water
1300
1150
750
500
250
5000
4000
3000
2000
1000
HW
LW
T=1·5°C
75
50
25
LW
HW
T=12·5°C
60
40
20
600
400
200
LW
HOURS
HW
FAECAL BACT. COLI PER ML OF SHELLFISH TISSUE
FAECAL BACT. COLI PER 10 ML OF WATER

Fig. III