



**Battelle**

New England  
Marine Research  
Laboratory

Determination of Neoplasia in Soft-Shell  
Clams Mya arenaria Near the  
Seabrook Nuclear Plant

to

Yankee Atomic Electric Company

April 30, 1987

Study No. N-0954-9901

Report

Final Report

on

Determination of Neoplasia in Soft-Shell  
Clams Mya arenaria Near the  
Seabrook Nuclear Plant

to

Yankee Atomic Electric Company

April 10, 1987

Study No. N-0954-9901

by

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## EXECUTIVE SUMMARY

There have been recent indications of an increase in prevalence of neoplasia in soft-shell clams along the east coast of the United States. The disease has been implicated in increased mortalities in those populations. To determine whether the disease is present in soft-shell clam populations near the Seabrook Nuclear Power Station, and whether the incidence increases seasonally, representative samples of clams were collected in October 1986 and February 1987 from three clam flats routinely monitored as part of the Seabrook monitoring program. The clams were delivered to Battelle Ocean Sciences in Duxbury, Massachusetts, for histological examination. In October, 1 of 30 clams from Flat 1 and 2 of 28 clams from Flat 2 showed low levels of neoplastic cells. By February, the incidence of infection had risen to 2 of 30 clams from Flat 1 and 8 of 30 clams from Flat 2. Thirty clams from Flat 4 were examined during each period. No evidence of neoplastic cells was found in any clams from Flat 4.

The histological technique used to identify the neoplastic cells provides the most conservative estimate of the number of infected clams from a given population. It is likely that the estimate of 27 percent incidence of the disease in the Flat 2 populations is low, and that significant mortalities in that population might be expected this spring and summer. Disease-related mortalities are also likely to be elevated on Flat 1, but not to the extent of those on Flat 2.

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## INTRODUCTION

### Background

Reports of neoplasms in field-collected bivalve molluscs have increased in recent years (e.g., see Lauckner, 1983, and Couch and Harshbarger, 1985, for reviews). The incidence of a sarcomatous neoplasm in the soft-shell clam Mya arenaria has especially increased in certain areas of the United States east coast (e.g., Barry and Yevich, 1975; Farley, 1976, Yevich and Barczysz, 1976, 1977, Brown et al., 1977, 1979; Brown, 1980; Walker et al., 1981; Cooper et al., 1982; Reinisch et al., 1984), and is considered epidemic in some locations of Maine, Massachusetts, and Maryland (Farley et al., 1986). Evidence for a viral etiology for the sarcomatous neoplasm was provided by Appeidoorn and Oprandy (1980) and Oprandy et al. (1981), with possible enhancement of the disease by chemical pollutants (Reinisch et al., 1984).

Although the disease has been reported in most coastal states from Maryland north, no information was available from New Hampshire. In light of the possibility of neoplasms occurring in clam populations near the Seabrook Nuclear Power Station, with subsequent large-scale clam mortalities due to the disease, irrespective of plant operations, Yankee Atomic Electric Company contracted with Battelle Ocean Sciences to

determine whether neoplasms do indeed exist in Mya arenaria populations near the Seabrook Nuclear Power Station. Samples of clams were collected from three flats near the plant in October 1986 and processed histologically for evidence of the disease. It was found that the disease existed at two of the three flats, but at low levels. Because the clams were collected at a time of year (early autumn) when the disease was less likely to be prevalent (Reinisch et al., 1984), and because the technique used to diagnose the disease (histological observations of fixed tissues) would most probably yield the most conservative results, it was felt that a second sampling later in the winter would be useful to determine whether the incidence was higher than in October.

### Objective

The objective of the study was to determine whether neoplasms exist in any of the soft-shell clam populations near the Seabrook Nuclear Power Station, and whether there might be seasonal differences in prevalence of the disease. The existence of the disease was determined through histological examination of clams representative of the different populations.

## MATERIALS AND METHODS

Samples of soft-shell clams Mya arenaria were collected from three clam flats, Flat 1, Flat 2 and Flat 4, in the vicinity of the Seabrook Nuclear Power Station by personnel of Yankee Atomic's contractor. Clams were collected during the regular sampling program in October 1986 and again in February 1987. In each case, the clams were placed whole in Dietrich's fixative (Preece, 1965) and delivered to Battelle for histological examination for neoplasia.

Upon arrival at the laboratory, the clams were removed from their shells, rinsed briefly in running tap water, and transferred to 80 percent ethanol. Each clam was cross-sectioned into 3 to 5-mm cross sections. One to four sections were selected as representative of the animal and processed for sectioning and staining.

The tissues were processed in an automatic tissue processor for routine paraffin sectioning. The paraffin-impregnated tissues were embedded in paraffin, sectioned at a thickness of 5 to 7  $\mu$ m, and stained with Feulgen picromethyl blue (Farley, 1969). The stained slides were examined by light microscopy for evidence of neoplastic

cells. The slides were examined under high power (400x) for up to 10 minutes per slide. If no neoplastic cells were seen after 10 minutes, the tissue was rated as negative.

## RESULTS

The results of histological observations carried out on clams from the October 1986 and February 1987 samplings from three clam flats near the Seabrook Nuclear Power Station are summarized in Table 1.

Neoplastic cells were found in clams from Flats 1 and 2 during both the October and February samplings, with an increase in prevalence of the disease between October and February in the clams from each site. In February, 2 of the 30 clams from Flat 1 contained neoplastic cells in an intermediate developmental stage as compared to 1 of 30 clams in an early stage in October, an increase in prevalence from 3 percent in October to 6 percent in February. In October, 2 of 28 clams showed evidence of the disease, a prevalence of 7 percent. Both clams were in an early stage of infection. In February, 8 of 30 clams contained neoplastic cells, a prevalence of 27 percent. In addition, 2 other clams from Flat 2 contained cells that were possibly early neoplastic cells. These clams were rated negative. Five of the 8 infected clams were in an early stage of the disease, 2 were in an intermediate condition, and 1 was in an advanced stage of the disease.

## DISCUSSION

Sarcomatous neoplasms of the type discussed in this study have been reported in east coast populations from both polluted and non-polluted areas (Barry and Yevich, 1975; Farley, 1976; Yevich and Barszcz, 1977; Brown et al., 1977, 1979; Brown, 1980; Cooper et al., 1982; Reinisch et al., 1984; Farley et al., 1986). The apparent initial cause of the disease is a virus similar to the B-type retroviruses (Oprandy et al., 1981), although the effects may be enhanced by environmental factors, including domestic and industrial pollutants (Reinisch et al., 1984).

Laboratory and field studies indicate that infected clams die from the disease. Farley et al. (1986) reported that individual diseased clams monitored in laboratory aquaria for a 5-month period progressed from early to advanced stages with 100 percent mortality. In addition, the same authors reported declines in prevalence on natural clam flats from high prevalences to zero back to high prevalences, again indicating 100 percent mortality among the infected clams and subsequent infection of the new year class.

On the basis of the existence of the disease in populations near the Seabrook Nuclear Power Station, it can be predicted that there will be some neoplasia-related mortalities, irrespective of power plant operations, in those populations. The highest prevalence of the disease was found on Flat 2. The techniques used in this study to ascertain the existence of neoplasia are very conservative. That is, they will confirm the existence of neoplasias but the prevalence is likely to be considerably higher than the technique reveals. It would seem, therefore, that significant disease-related mortalities of clams on at least Flat 2 are likely by late spring and early summer. Flat 1, where the prevalence of the neoplasias was only 6 percent in our study, may also have noticeable mortalities.



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TABLE 1. INCIDENCE OF NEOPLASIA IN SOFT-SHELL CLAM POPULATIONS NEAR THE SEABROOK NUCLEAR POWER STATION

Site	Number of Clams Examined		Number Infected		Prevalence of Infection (%)	
	Oct.	Feb.	Oct.	Feb.	Oct.	Feb.
Flat 1	30	30	1	2	3	6
Flat 2	28	30	2	8	7	27
Flat 4	33	30	0	0	0	0