

**The Ecological, Chemical and Histopathological Evaluation of  
an Oil Spill Site**

**By:**

**R.L. Dow; J.W. Hurst, Jr.; D.W. Mayo, C.G. Cogger, D.J.  
Donovan, R.A. Gambardella, L.C. Jiang, and J. Quan; and  
P.P. Yevich**

**Research Reference Document: 71/1**

The Ecological, Chemical and Histopathological Evaluation of an  
Oil Spill Site

R. L. Dow;<sup>1</sup> J. W. Hurst, Jr.;<sup>2</sup> D. W. Mayo, C. G. Cogger,  
D. J. Donovan, R. A. Gambardella, L. C. Jiang, and  
J. Quan;<sup>3</sup> and P. P. Yevich<sup>4</sup>

<sup>1</sup>Research Director, Maine Department of Marine Resources, Augusta,  
Maine 04330. (207) 289-2291

<sup>2</sup>Fisheries Research Laboratory Director, W. Boothbay Harbor, Maine  
04575. (207) 633-5572

<sup>3</sup>Chemistry Department, Bowdoin College, Brunswick, Maine 04011.  
(207) 725-8731, Ext. 601.

<sup>4</sup>Chief, Histopathology Unit, National Marine Water Quality Laboratory,  
EPA, P. O. Box 277, West Kingston, R. I. 02892. (401) 789-1427.

## EVALUATION OF AN OIL SPILL SITE

### ABSTRACT

I. An oil spill into Long Cove, Searsport, Maine, beginning on 16 March and lasting through at least 30 June 1971, resulted in immediate and continuing soft clam (Mya arenaria) mortalities which, based on before and after biological surveys, had by August 1974 exceeded 85% of the estimated 50 million market size clams occupying the area.

II. Sediment and animal samples analyzed by gas chromatographic procedures were found to contain significant quantities of petroleum hydrocarbons identified from adjacent shoreline tank farm samples to be #2 fuel oil mixed with JP5 jet fuel.

III. Histopathological examination during the four-year period has identified gonadal and other soft tissue cancers with an incidence ranging from 2 to 27% at the sites where samples were collected. Stations which showed the greatest incidence of tumors could be correlated with the areas of the highest hydrocarbon concentrations.

# THE ECOLOGICAL, CHEMICAL AND HISTOPATHOLOGICAL EVALUATION OF AN OIL SPILL SITE

## Part I. Ecological Analysis Section

Penobscot Bay, a major portion of the mid-coast embayment of the Gulf of Maine, has a long history of economically important shellfish production, especially in the upper bay communities of Searsport and Stockton Springs.

Even during the high sea temperature period of 1947-1963 when proliferating green crab (Carcinus maenas) and other predator populations had reduced soft clam (Mya arenaria) stocks more than 90%,<sup>1</sup> joint state-municipal management and predator restraint programs made the Searsport-Stockton Springs region one of the six most productive areas in the Gulf of Maine (Map 1).

Increased industrial development in the Penobscot River Valley and adjacent upper Bay area, greater human population density, and expansion of oil handling and storage facilities rapidly reduced the quality of estuarine and marine waters overlying clam flats despite state legal classifications to the contrary. Finally, on June 28, 1966, the remaining shellfish growing areas of Searsport and Stockton Springs were closed because of pollution.

Before the 1966 closure, employment in the production of clams in the two towns had been approximately one hundred, with nearly ten times that number engaged in recreational or subsistence fishing. Commercial production alone averaged more than 135 metric tons annually of edible meats.

Between August 1 and August 19, 1966, an inventory survey of the clam resource in the upper bay was carried out jointly by the Northeast

Shellfish Sanitation Research Center of the U. S. Public Health Service, the Federal Water Pollution Control Administration, and the State of Maine's Department of Sea and Shore Fisheries (now Department of Marine Resources), using standard survey methods developed by the Department in the late 1940's.<sup>2</sup>

Survey results indicated that the existing crop of market size clams in Searsport alone amounted to more than 140 metric tons of edible meats, with a community value ranging from \$400,000 to \$1.1 million, depending upon use. Of this population total, approximately 110 metric tons occupied Long Cove and the adjacent west shore of Sears Island.

Following the 1966 pollution closure and coincident optimum sea temperature conditions, the population of clams increased rapidly and by the early 1970's exceeded 150 metric tons.

Joint state-municipal efforts to salvage some of these clams for commercial use led to the construction and operation of a clam self-cleansing plant using flow-through sterilized sea water as a cleansing mechanism, resulting in the salvage on an experimental basis of approximately 25 metric tons of clam meats.

On March 16, 1971, an oil spill from the U. S. Air Force storage facility on Long Cove, Searsport, contaminated much of this cove and adjacent portions of Sears Island. Oil continued to drain on to the flats until at least late June 1971 and made it necessary for the entire area to be closed to cleansing plant use of the clams (Photo 1).

The spill volume was reported by the Coast Guard as "something less than moderate" and "possibly no more than a barrel and a half got into the salt water." Subsequent estimates by the Department placed the

volume of spill at a minimum four metric tons, since 80% of that amount was recovered by the company employed for clean-up purposes.

The acute toxicity of the oil, a mixture of #2 fuel and JP5, resulted in almost immediate and continuing die-off of clams. During initial die-off, many clams surfaced from their burrows and fell over on the flats. Apparently this behavior was induced by the irritant and smothering effects of the oil which drained out onto the growing areas from several culverts and drainage ditches. Damage to the shellfish adjacent to those oil sources was evident almost immediately after the spill (Photo 2).

Across upper Penobscot Bay, about 8 kilometers from Long Cove, a portion of the spilled oil stranded on the flats of Little River and by March 26 had killed between 5 and 10% of the clams in that area. By March 29 the mortality had increased to more than 4 metric tons of edible meats.

In both Long Cove and Little River small clams near the surface of the flats were the first to die. Later, as the oil penetrated the sediments, larger clams, more deeply burrowed into the sediments, were also killed. The rate of kill appeared to be related to the rate of oil penetration and the depth of the clams. Generally clams burrow about 2.5 times their long diameter.

In one small portion of the west shore of Long Cove, the hulk of a wooden barge inadvertently served to collect nearby sediments and to raise the elevation of the flats above adjacent areas. Here clams were not exposed to the oil, for no visible evidence of oil nor associated mortalities have been found.

Elsewhere in the cove oil continued to spread laterally as well as down the beach slope, and mortalities of clams and other organisms continued.

On March 22 when about 3% of the Long Cove clams had died, a series of systematic surveys was begun by biologists of the Department to determine the extent of the contaminated area and to monitor vertical, horizontal, and lateral oil penetration of the sediments, and to estimate the areal and temporal extension of clam mortalities caused by the oil. Cooperative institutions and agencies working with the Department in this study are: Woods Hole Oceanographic Institution, TRIGOM, Bowdoin College, and the National Marine Water Quality Laboratory of EPA. These studies are still in progress and have been augmented at times by the assistance of other federal personnel. One such survey in late March 1971 by the U. S. Environmental Protection Agency was summarized as follows:

"As a result of the spill, the entire Long Cove was closed to shellfishing and a sampling program initiated by the Maine Department of Sea and Shore Fisheries to assess the effects of the oil discharge on the shellfish resource.

"The long-term marine resource damage which this particular spill has caused is expected to be of a much greater magnitude than the present figures indicate. All of the clams sampled in Long Cove have been found in a distressed condition, and it appears that the entire shellfish population of this area will be killed."

Of the total pre-spill Long Cove and western Seares Island population, 13% had been killed by March 30 and 25% by July 31, 1971. Approximately 55% had died by August 1972 and 86% by August 1974.

Two populations currently occupy the growing area; clams less than 3 centimeters in long diameter, survivors of the 1971 through 1974 year classes are in the upper 7.5 cms of sediments, and those spawned prior

to 1971 having a long diameter normally in excess of 4.5 cms occupy sediment strata generally more than 11.0 cms beneath the surface of the intertidal flats.

During the August 1972 clam population inventory, oil was found in 23% of the 130 intertidal sample plots. At subtidal stations when bottom sediments were disturbed globules of oil rose to the water surface and formed slicks. In 1974 oil was observed in all sample plots taken near low water.

Soft clams have virtually doubled in value since the 1966 inventory of Long Cove. This increase means that the damage done by the 1971 oil spill represents an annual area loss ranging from \$.8 to \$2.5 million, depending upon use.<sup>3</sup> Only 22 of the original standing crop of 157 metric tons of shucked meats had survived to August 1974. Since it requires five to six years to produce a marketable crop of clams in this area, even without oil spills, the annual loss will continue for the foreseeable future. In other study areas where oil spills have occurred, the annual growth rate has declined up to 60%, depending upon the type and amount of oil spilled.<sup>4</sup>

Based on an August 7, 1974, resurvey of the Long Cove clam population, recent mortality has declined since 1973 to 32%, making a cumulative mortality since the 1971 spill of 86%, not including those clams which were spawned after the 1971 spill year. Representatives of all four year classes since then (1971, 1972, 1973, and 1974) were found occupying the upper sediment stratum. The current mortality rate of smaller clams is not known. It is also not possible to estimate what may happen to them as they grow and burrow more deeply into the sediments. If the entrapped



oil has not been sufficiently weathered, leached, or dissipated through transport mechanisms, many of these clams may be killed when they reach the oil-bearing sediments.

Based on continued observation of the persistence of oil spilled in other soft clam growing areas, residues may be present in significant amounts for many years.<sup>5</sup>

Although oil was observed leaching out of the sample plots dug along the low tide line, the amount of oil present in these sediments superficially does not appear to be as great as it was in preceding years. The odor of oil is noticeable at many sites along the intertidal section of Long Cove, particularly in those areas downstream from drainage culverts.

## Part II. Chemical Analysis Section

### Experimental:

The gas chromatographic analyses were all carried out on a Perkin-Elmer Model 990 instrument utilizing dual column operation. The carrier gas was Helium at a flow rate of approximately 30 cc/min. The columns were six feet by 1/8" stainless steel. The solid support was acid-washed chromasorb W, 80/100 mesh. The stationary phase was 3% Apiezon L. Injection block temperature was 240°C. Detectors were kept at 340°C. The temperature was programmed at 6%/min. with the initial temperature, room temperature, and the final temperature, 280°C.

Sample preparation was identical to Blumer<sup>6</sup> with the exception that sediments were initially air dried at room temperature and then directly pentane extracted. This procedure in our experience gives higher yields of hydrocarbons than the methanol partition step when dealing with light refined oils.

Discussion and Results:

The following discussion is an interpretation of gas chromatographic analytical data collected on sediment and culvert effluent samples taken in the Searsport Region during the period March 1971 to September 1972. The enlargement (figure 1) shows the main sampling sites within the prime area. Sample coding designates the coastal location (Roman numeral), station number and the month of collection (by a capital letter). There were four collections made during this part of the study: March, 1971 (A); July, 1972 (B); August, 1972 (C); and September, 1972 (D). The samples examined are listed in Tables 1 and 2 with a more detailed description of each site location.

The large majority of samples collected were sediments. These samples have been employed to develop a profile of the local spill area and the general hydrocarbon background content of the region. Initially, 13 collection sites were established on the northwest side of Long Cove (see figure 1). Estimates of concentrations in ppm dry weight are given in Table 3. The chromatographic data indicate that at that time stations 1-4 exhibited chronic oil pollution (see figure 2). The large background envelope evidenced in these chromatograms indicates that a sizeable fraction of the samples obtained from these sediments contain weathered hydrocarbon residues. It would appear likely that a considerable contribution to the material presently detected on this shore must have come from prior contamination. Low relative concentrations of recently deposited #2 oil (determined by the area of the linear chain peaks and the  $C_{17}$ -pristane ratio) were found at these sites, however, beginning at location 5 the proportion of fresh #2 oil rapidly increased and reached a maximum between sites 6 and 7 (see figure 3). Although the total

hydrocarbon content of the sediments in sites 5-8 actually decreases, contamination by #2 oil of recent origin reaches maximum levels at these locations. It should be noted that the prime source of contamination for the current spill is believed to be a culvert located near sites 6-7. A sample of the effluent collected directly from this culvert indicated contamination in excess of 1000 ppm and that the polluting material was a mixture of #2 oil containing significant quantities of JP5 jet fuel (see figure 4). North of the culvert, stations 8-11 (see figure 5) indicate the presence of material aged to a similar extent as at stations 1-5. Readily identifiable quantities of moderately aged #2 oil also appear in these areas. At stations 12 and 13 (figure 6) the relative level of recently deposited #2 oil again appears to rise. A second storm culvert is located in the area.

On the northeast side of Long Cove collecting sites 14-18 were also established during March 1971. Locations 14 and 16 show only trace contamination (see figure 6). The large majority of the material contributing to the hydrocarbon content of sites 14 and 16 is of natural origin. Areas 17, 18 and 19 show recent and significant contamination by #2 oil (see figure 7). This material is essentially identical to that found at the culvert near stations 6-7, the only significant differences can be ascribed to substances of natural origin. These compounds occur just before the C<sub>17</sub> and C<sub>21</sub> linear chain peaks on the chromatogram.

During the summer of 1972 locations on the southeast side of Long Cove (19-21) and subtidal sites in the center of the cove were established.

Near Mack Point samples from stations 28-31 were collected in the March 1971 period. These sites produced chromatographic data which indicate the presence of relatively low quantities of fresh #2 oil in the

sediments. They contained (with the exception of site 30) a mixture of aged and some recently deposited material. Site 30, on the other hand, appeared to possess little fresh material and seemed to be primarily heavily aged petroleum compounds.

The eighteen sediment samples collected in March 1971 indicated the following profile for Long Cove, Searsport, at that time: sampling sites 1, 2, 3, 4, 8, 9, 10, and 11 were dominated by rather well weathered petroleum residues; three localized regions of recently deposited #2 oil (sites 5, 6, 7; 12, 13; and 17, 18, 19) were identified; and sites 14, 15, 16 were shown to possess only trace contamination; the area around Mack Point (28-31) exhibited a well weathered hydrocarbon content with relatively light concentrations of new material.

A second series of sediments were collected in July 1972. As expected, samples were found to possess mostly well aged petroleum residues for they were acquired nearly one and one-half years following the major spill period. All samples were collected on the northwest side of Long Cove and all samples were subtidal. The single exception was station 7, which contained significant quantities of very fresh #2 oil (see figure 8). The apparent localization of this new material and its proximity to the source culvert imply that low level seepage of #2 oil was still continuing in the area of station #7 during July 1972.

A third collection of samples was carried out during August 1972. Intertidal samples were collected at stations 6, 8, 11, 13, and new sites 15, 20 and 21 in Long Cove. One of the sediment samples (site 11) from the northwest side of the cove again exhibited fresh contamination with #2 oil (see figure 8) as did site 15 on the northeast shore. These latter results are a further indication that a chronic spill situation still exists in Long Cove.

A small final collection from earlier established stations was carried out in September 1972 and consisted of three subtidal samples obtained from the Mack Point sites. The results indicated that measurable quantities of aged hydrocarbons are present in these sediments but that they have not, in this general area, reached very high levels.

A number of reference chromatograms were obtained to establish a relationship between freshly deposited material and the source of the pollution. Standard samples of #2 oil and JP5 jet fuel collected from the storage terminal were compared to the culvert sample in figure 4. The linear chain hydrocarbons peak at  $C_{14}$  in the #2 oil but at  $C_{11}$  in the jet fuel. The culvert sample was observed to possess double maximum at  $C_{11}$  and  $C_{13}$  indicating that the effluent is unquestionably a mixture of the two materials. The major constituent of the mixture appears to be #2 oil as the sample leaves the culvert.

#### Conclusion:

The data indicate that a significant spill consisting of a mixture of #2 oil and JP5 occurred during the spring of 1971 in Long Cove, Searsport, Maine. The major impact of this spill was observed in two sampling areas (sites 5, 6, and 7; and sites 12 and 13) on the northwest shore of the Cove and in one sampling area (site 17, 18 and 19) on the northeast shore of the cove. Of these three areas the more northerly one on the northwest shore (sites 12 and 13) appears to have experienced less contamination than either of the other two locations.

Further studies indicated that contamination of the sampling sites with fresh material leached from saturated sediments located at higher elevations still continued up to 18 months following initial detection of the spill problem.

Part III. Histopathologic Findings

Soft shell clams, Mya arenaria, collected from the oil spill site at Long Cove, Searsport, Maine, have been submitted for histopathologic examination to the Histopathology Unit of the National Marine Water Quality Laboratory, Narragansett, R. I., since July, 1971. The first group was received for examination approximately five months after the oil spill occurred. Since then over 2000 soft shell clams have been looked at microscopically from this area. In addition, over 1000 clams from non-polluted areas of Maine were examined.

Histopathologic studies by Barry<sup>7</sup> of the National Marine Water Quality Laboratory, Narragansett, R. I., conducted in 1971, showed a 2% to 27% incidence of malignant gonadal tumors in the soft shell clams collected from stations around the culvert. In 1972 Mrs. Barry recorded a 2%-18% incidence of malignant gonadal tumors from the same stations. Yevich, in 1973, found that 2.5%-8% of the exposed clams had malignant gonadal tumors and in 1974, 9%-18% of the clams collected from the same stations contained the tumors (Figure 9, Table 4).

Malignant gonadal tumors originate from the germinal epithelium of the follicles of the gonadal tissue of both male and female clams. Tumors may eventually involve all of the follicles to such a degree that sexual differentiation cannot be determined (Fig. 12). The tumor cell mass in the follicles consists of mononucleated and multinucleated cells and cells with mitotic figures (Fig. 13, 14). If the tumor cells have not completely filled the follicle, cellular debris appropriate to the sex of the clam is seen (Fig. 15, 16).

In some of the soft shell clams examined the interfollicular connective tissue was invaded by the tumor cells (Fig. 17, 18). In other animals the gonadal tumor cells metastasized to the other organs of the body such as the walls of the pericardial cavity, the heart, kidneys, gills and genital pore (Fig. 19). One case was found in which the tumor cells had completely taken over the body of the clam and the normal histological architecture of the organs could not be seen. The development of malignant gonadal tumors could not be associated with any seasonal or cyclic changes.

Histopathologic studies were also carried out on soft shell clams collected from a #2 fuel oil and JP4 oil spill site at Brunswick, Maine. A 9% incidence of malignant tumors was observed in the animals examined. The tumors were possibly of mesenchymal tissue origin and were found in all areas of the body (Fig. 21, 22, 23).

At both oil spill sites the stations which showed the greatest incidence of tumors could be correlated with the areas of the highest hydrocarbon concentrations. No tumors were found in areas in which low amounts of hydrocarbons were detected or in any of the animals collected from the nonpolluted areas of Maine.

## EVALUATION OF AN OIL SPILL SITE

### References

- <sup>1</sup>Fluctuations in Gulf of Maine Sea Temperature and Specific Molluscan Abundance. R. L. Dow. J. Cons. int. Explor. Mer., 34(3), p. 532-534, 1972.
- <sup>2</sup>Shellfish Survey Methods. R. L. Dow. National Shellfisheries Assn. Convention, Atlantic City, N. J., August 12-14, 1952. Tech. Bull. 1, Dept. of Marine Resources, Augusta, Me.
- <sup>3</sup>A Multiplier for Computing the Value of Shellfish. Edward F. M. Wong, U. S. Dept. of Interior, Federal Water Pollution Control Admin., Needham, Mass., 1969.
- <sup>3</sup>Maine Landings, 1965 to 1973. U. S. Dept. of Commerce in cooperation with the Maine Department of Marine Resources.
- <sup>4</sup>Final Report on the Investigation of the Effects of the TAMANO Oil Spill on the Fauna of Casco Bay, Maine. Edward S. Gilfillan and Sherry Hanson. The Cape Ann Society for Marine Science, Inc., Gloucester, Mass. April 1974.
- <sup>4</sup>Supplement to the paper Long Term Weathering Characteristics of Iranian Crude Oil: The Wreck of the "Northern Gulf." D. W. Mayo et al. Dept. of Chemistry, Bowdoin College, Brunswick, Me.
- <sup>5</sup>Long Term Weathering Characteristics of Iranian Crude Oil: The Wreck of the "Northern Gulf." D. W. Mayo et al, Marine Pollution Monitoring (Petroleum) Symposium and Workshop, National Bureau of Standards, Gaithersburg, Md., May 13-17, 1974.
- <sup>6</sup>Correlation of Oils and Oil Products by Gas Chromatography. O. Zafiriou, M. Blumer and J. Myers, Woods Hole Oceanographic Institution Technical Report, WHOI-72-55, July 1972.
- <sup>7</sup>Histopathologic Studies. Patricia Barry. Histopathology Unit, National Marine Water Quality Laboratory, EPA, W. Kingston, R. I. (to be published.)



## EVALUATION OF AN OIL SPILL SITE

### List of Figures

- Fig. 1 - Location of oil analysis sampling sites
- Fig. 2-8 - Gas chromatographs
- Fig. 9 - Histopathological sample sites
- Fig. 10 - Gonadal tissue - normal male soft shell clam, Mya arenaria. Arrows point to follicles filled with sperm. H&E X360.
- Fig. 11 - Gonadal tissue - normal male soft shell clam, post spawning. Arrow A shows germinal epithelium beginning to develop again. Arrow B shows the atypical cell inclusions normally found in the male gonadal tissue. H&E X360.
- Fig. 12 - Gonadal tissue - soft shell clam. Follicles filled with tumor cells. H&E X360.
- Fig. 13 - Gonadal follicles - male soft shell clam. Arrow A points to normal spawned out follicle with atypical cell inclusions. Arrow B points to follicle filled with tumor cells. H&E X920.
- Fig. 14 - Gonadal tissue - post-spawning female soft shell clam. Follicle contains tumor cells. Arrows A point to multinucleated cells. Arrow B points to mononucleated cells. Arrow C cell containing mitotic figures. H&E X1480.
- Fig. 15 - Gonadal tissue - post-spawning female soft shell clam. Arrow A points to normal post-spawning follicles containing follicle cell inclusions. Arrow B points to follicles containing tumor cells. H&E X360.
- Fig. 16 - Gonadal tissue - post-spawning female soft shell clam. Arrow A points to normal post-spawning follicles containing follicle cell inclusions. Arrow B points to follicles containing tumor cells. H&E X920.
- Fig. 17 - Gonadal tissue - soft shell clam showing invasion of the tumor cells from the follicles to the connective tissue between the follicles. Arrow A points to follicle containing tumor cells. Arrow B shows connective tissue invaded by tumor cells. H&E X360.
- Fig. 18 - Gonadal tissue - soft shell clam showing invasion of the interfollicular connective tissue by the tumor cells. Arrow A points to follicles containing tumor cells. Arrow B points to connective tissue invaded by tumor cells. H&E X920.

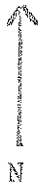
## EVALUATION OF AN OIL SPILL SITE

- Fig. 19 - Area of soft shell clam showing metastasis of tumor cells. Arrow A shows gonadal duct filled with tumor cells. Arrow B shows tumor cells invading the pericardial wall. Arrow C shows kidney invasion. Arrow D shows an area of involvement on the auricle wall. Arrow E shows the invasion of the branchial chamber by the tumor cells. H&E X360.
- Fig. 20 - Higher magnification of Fig. 19 showing metastasis of tumor cells from the gonadal tissue. Arrow B shows invasion of pericardial wall. Arrow C shows invasion of kidney tissue. Arrow D shows involvement of an area on the auricle wall. H&E X920.
- Fig. 21 - Soft shell clam collected from Brunswick, Maine, oil spill site. Tumor, possibly of mesenchymal origin, filling body mass. Arrow A points to tumor cells in the interfollicular connective tissue of the gonadal area. Arrow B points to the tumor cells in the submucosa of the intestine. Arrow C points to the tumor cells in the muscular area of the animal. H&E X360.
- Fig. 22 - Higher magnification of Fig. 21, possible mesenchymal tumor. H&W X920.
- Fig. 23 - Higher magnification of Fig. 21, possible mesenchymal tumor. Arrow D note-no invasion of gonadal follicles by the tumor cells. HEW X1480

EVALUATION OF AN OIL SPILL SITE

Figure 1

Location of oil analysis sampling sites.



AREA I

SEARSPORT  
STATION

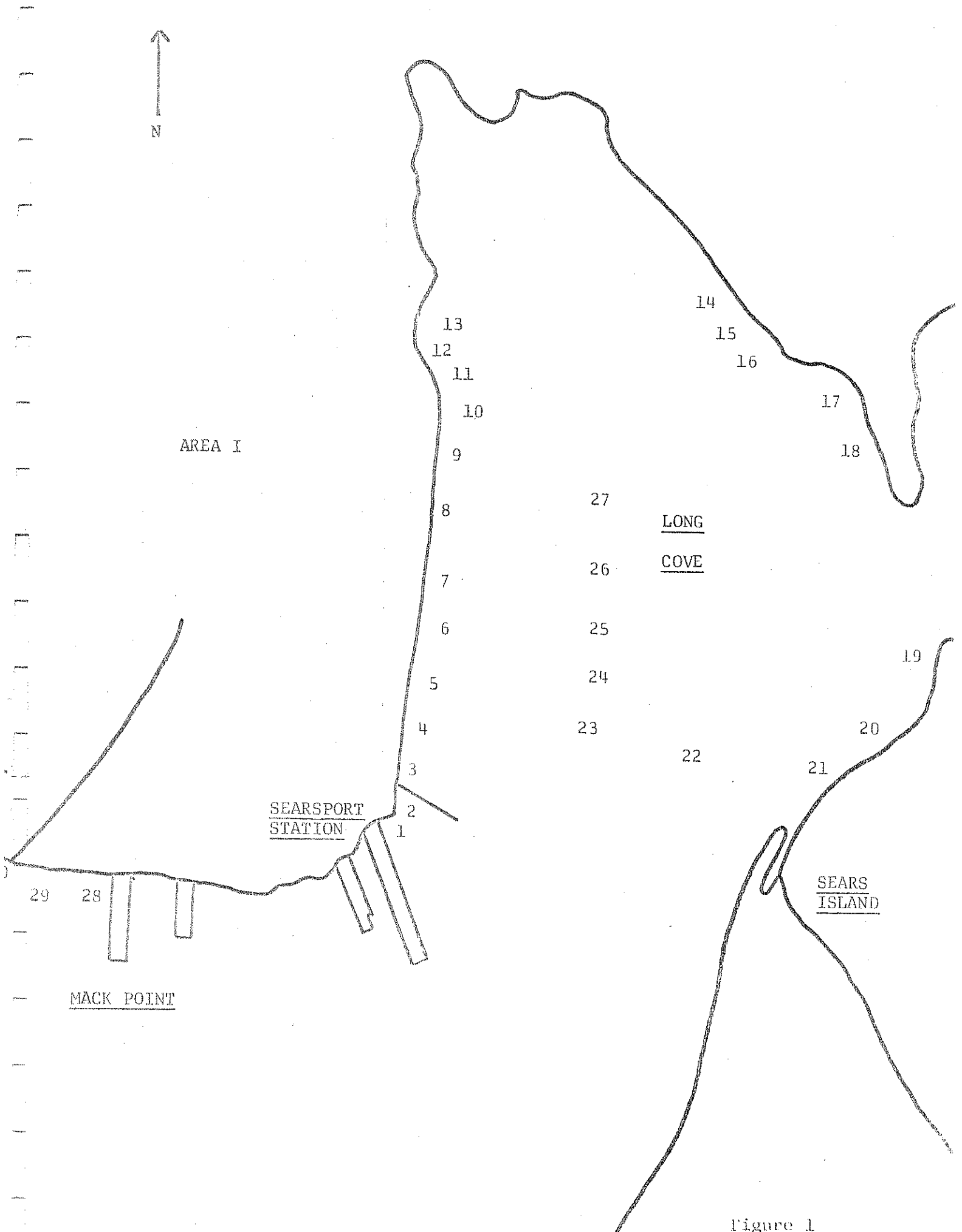
MACK POINT

LONG

COVE

SEARS  
ISLAND

Figure 1



EVALUATION OF AN OIL SPILL SITE

Figures 2 through 8

Gas chromatographs

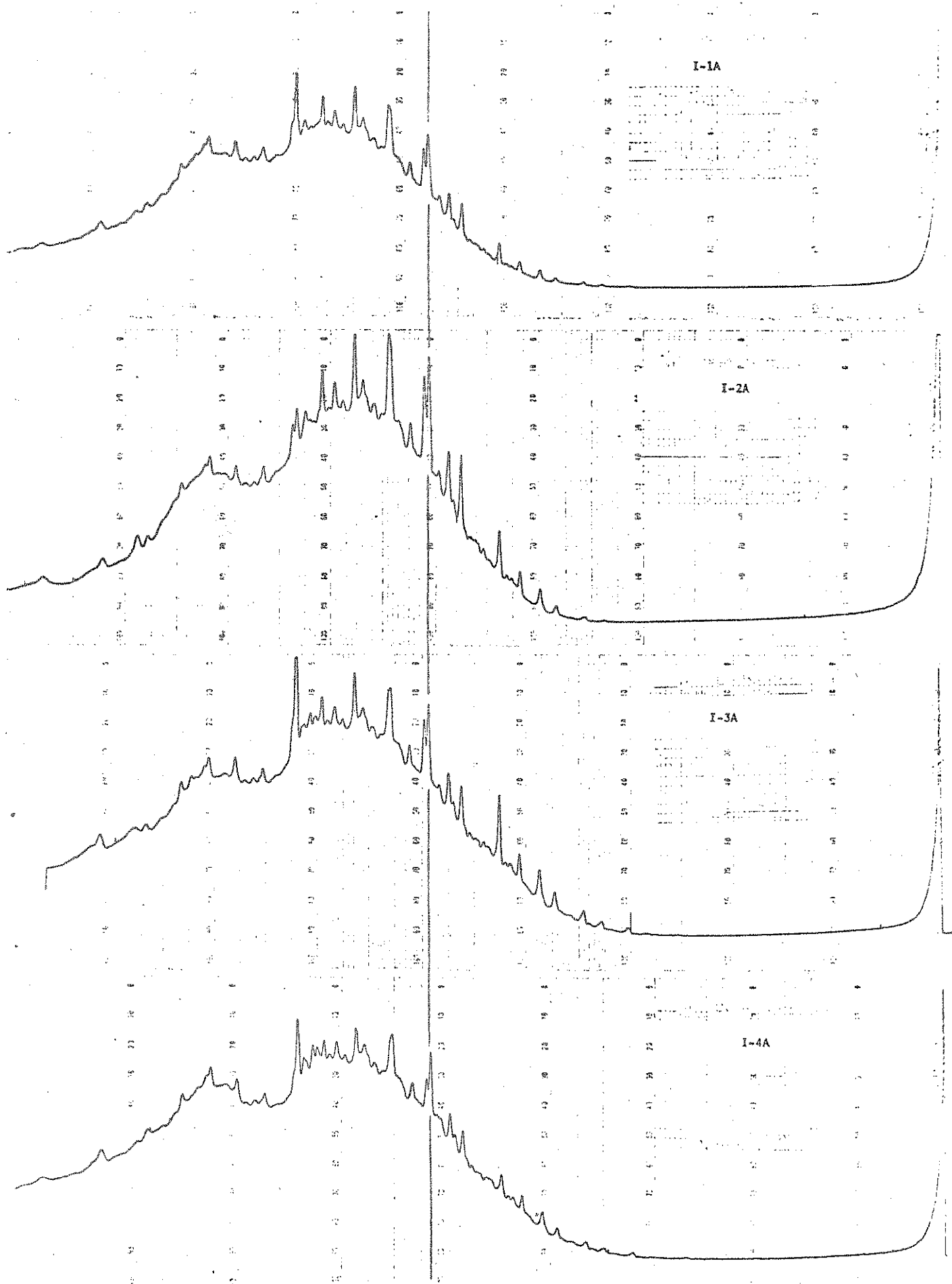


Figure 2

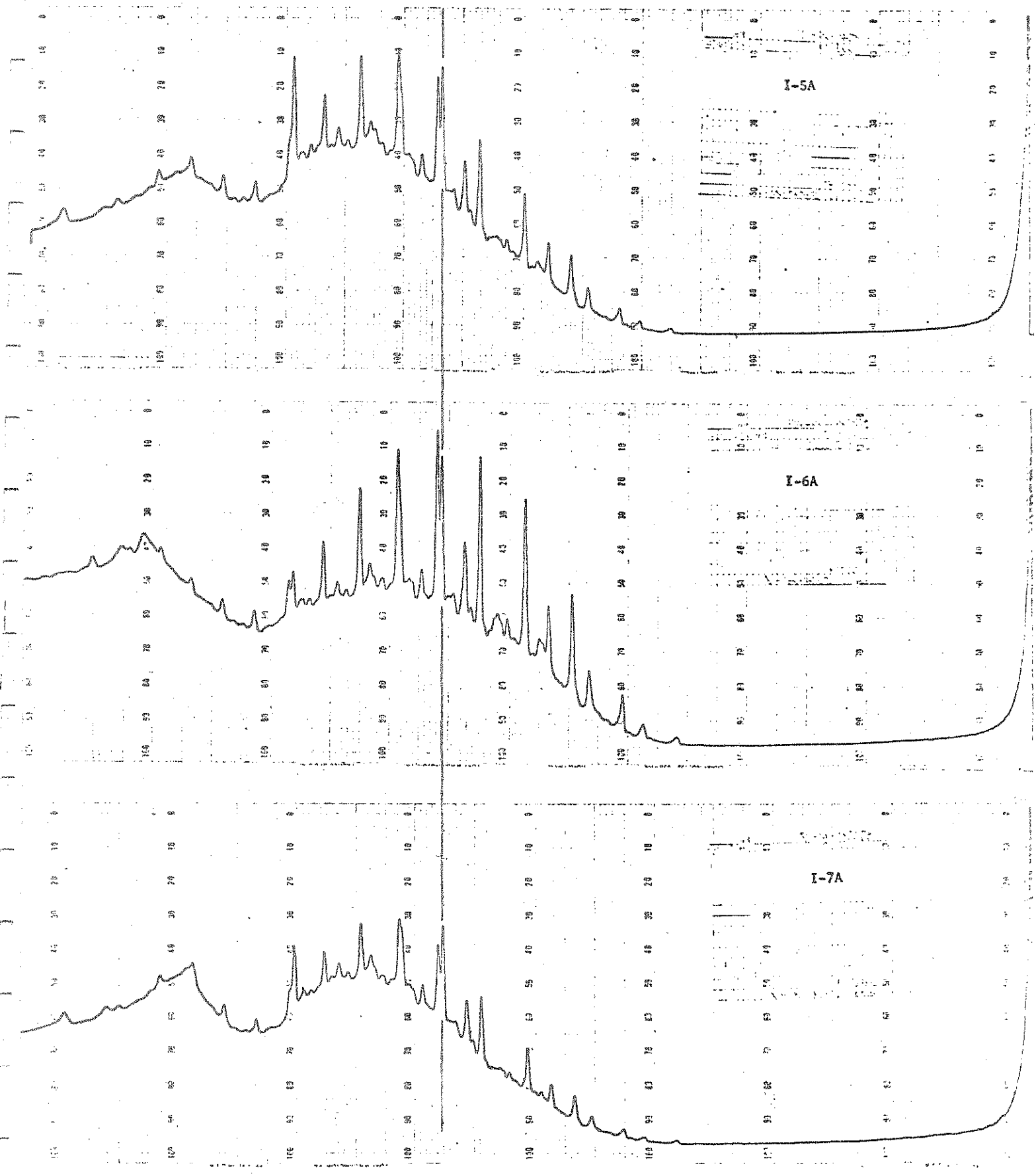


Figure 3

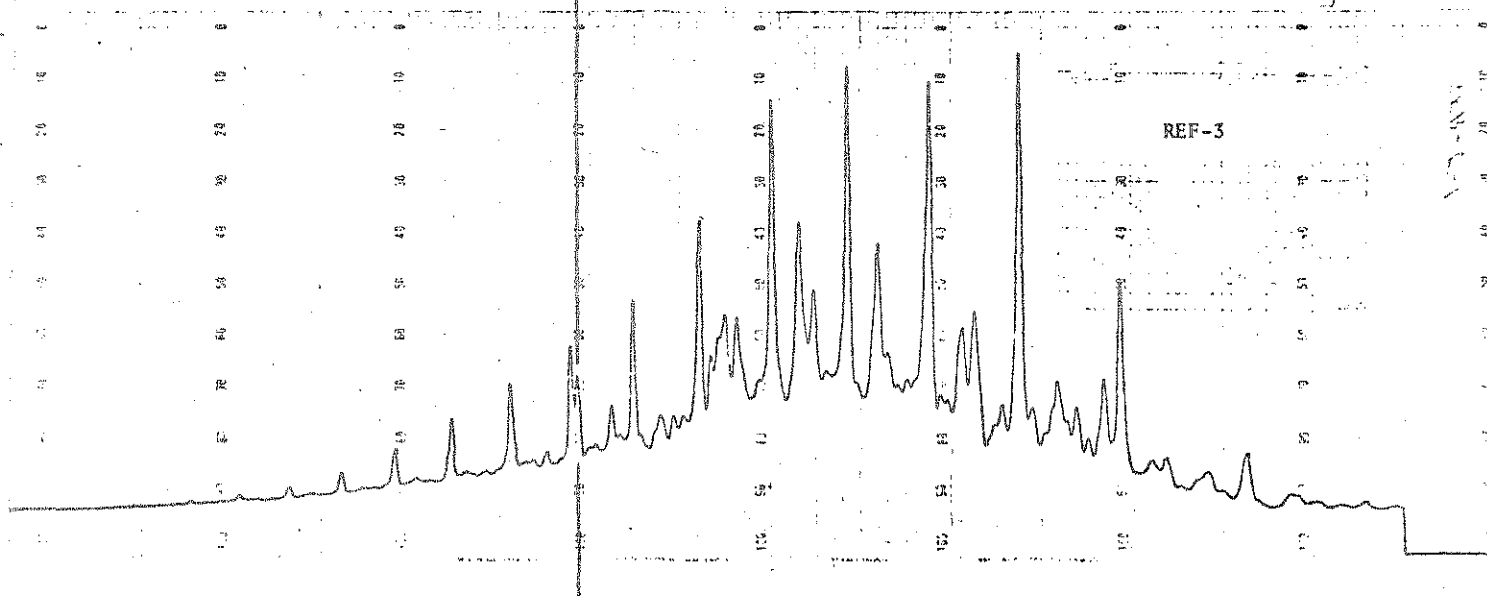
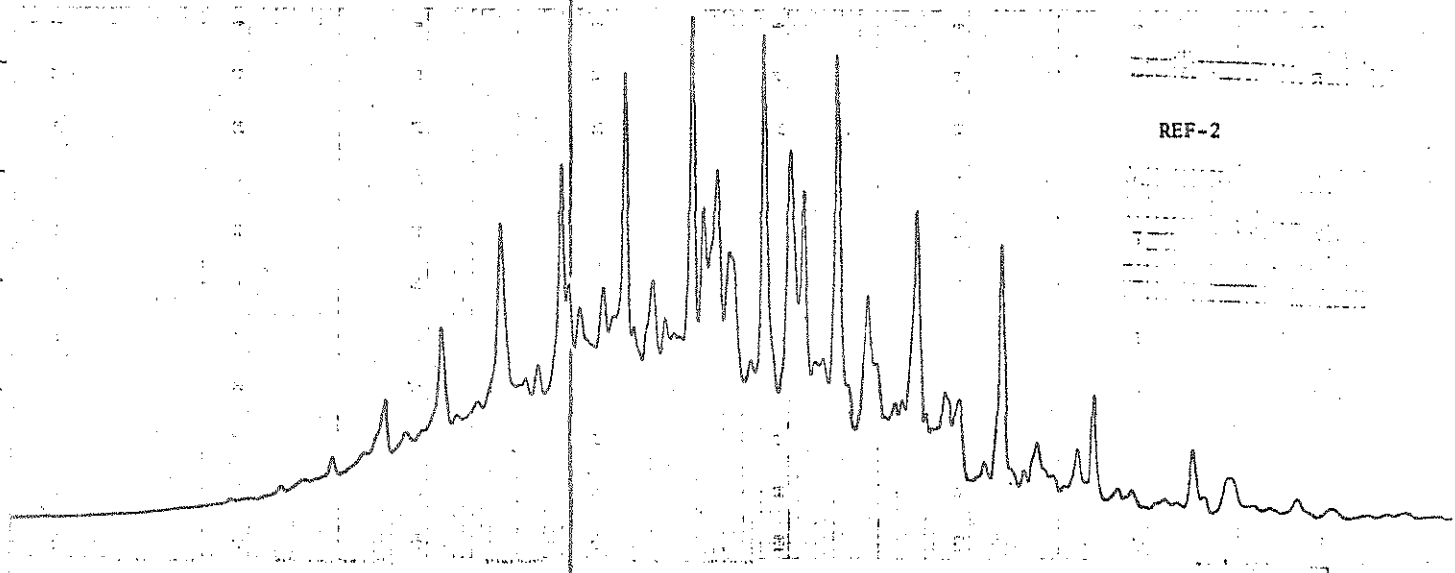
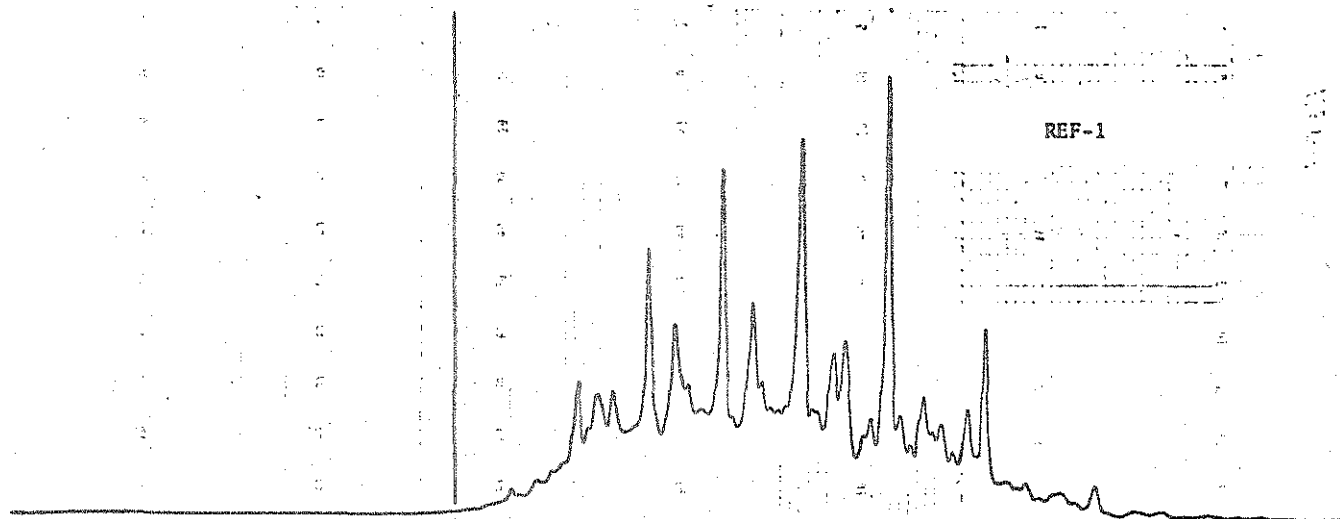


Figure 4



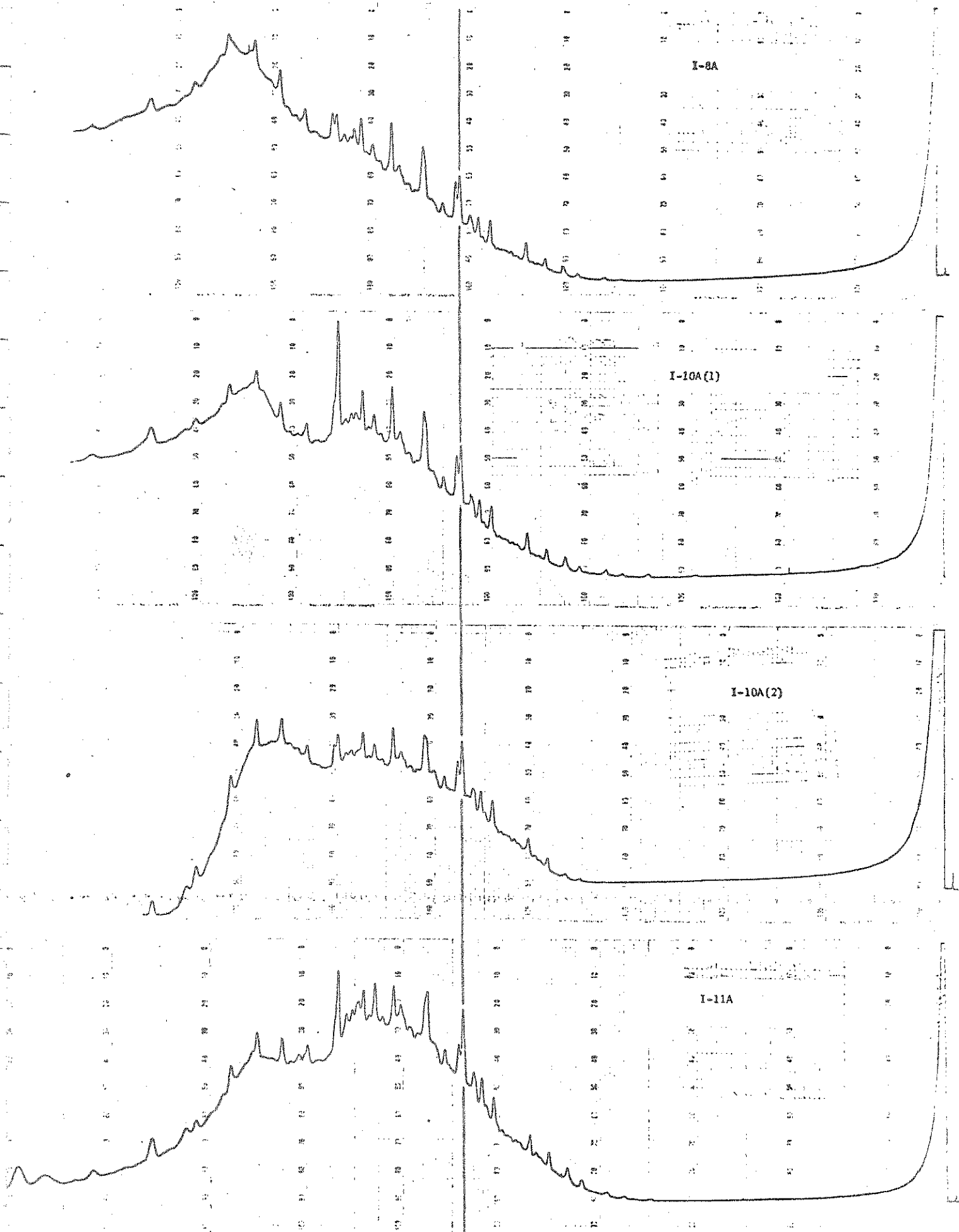


Figure 5

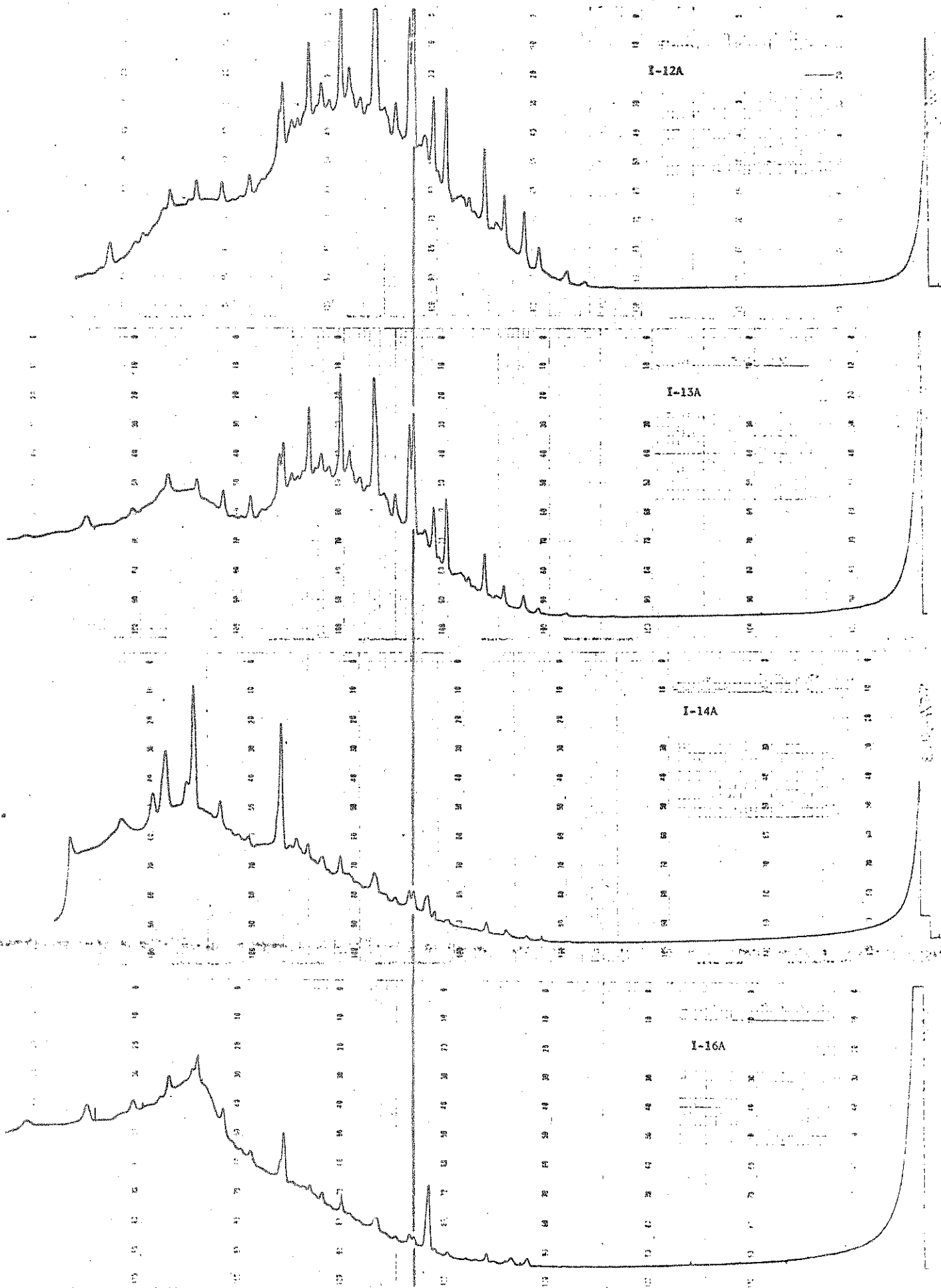


Figure 6

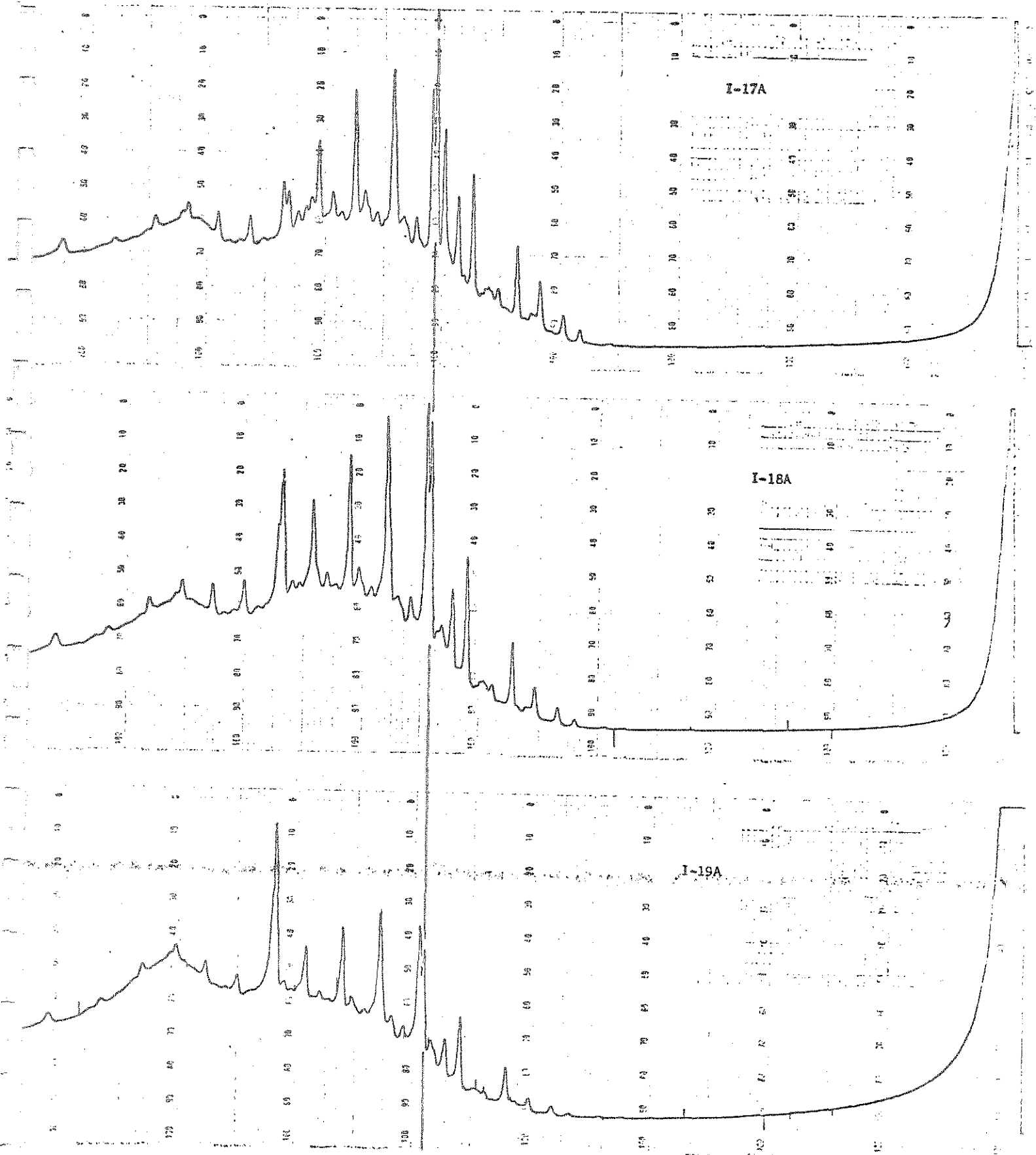


Figure 7

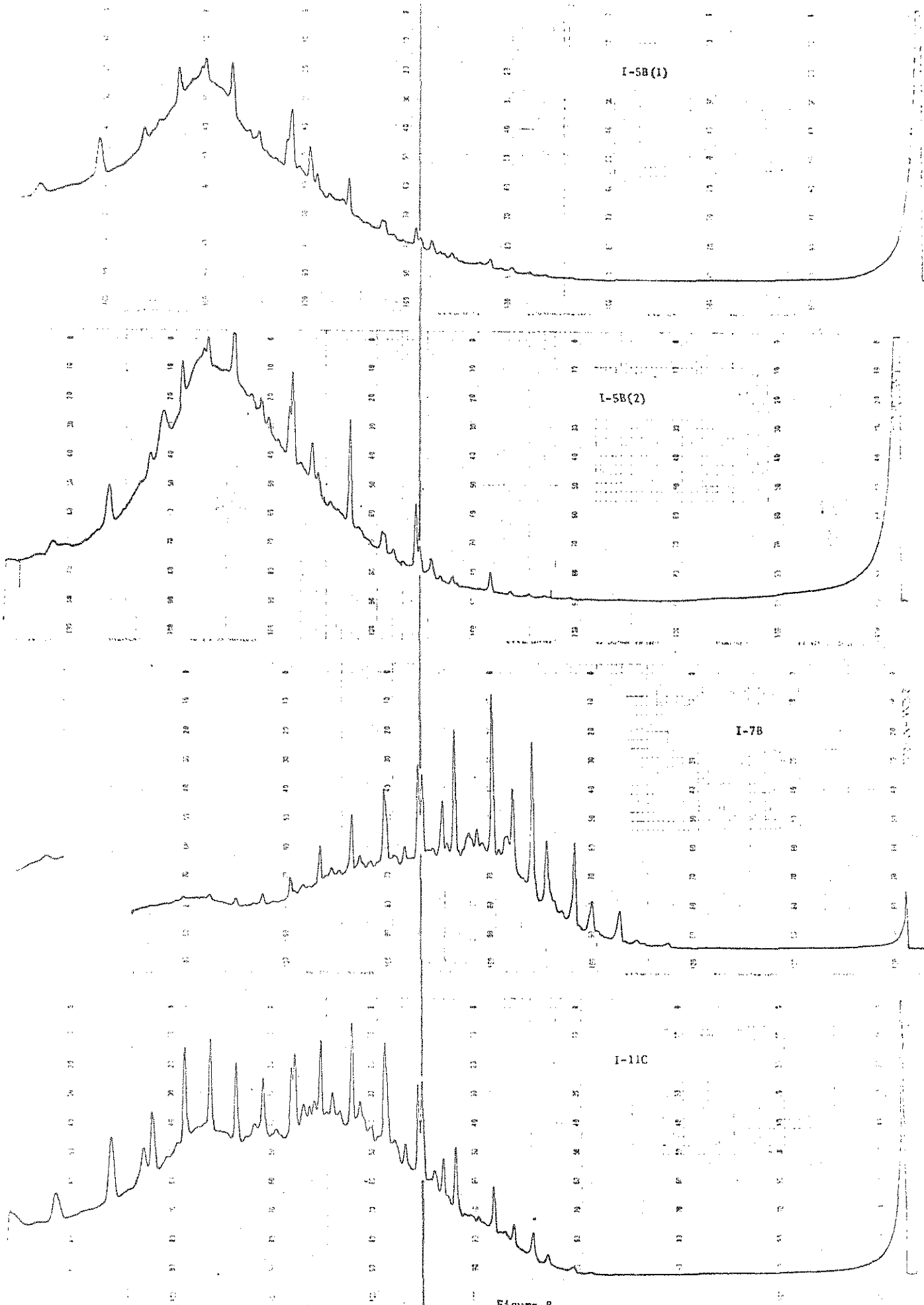
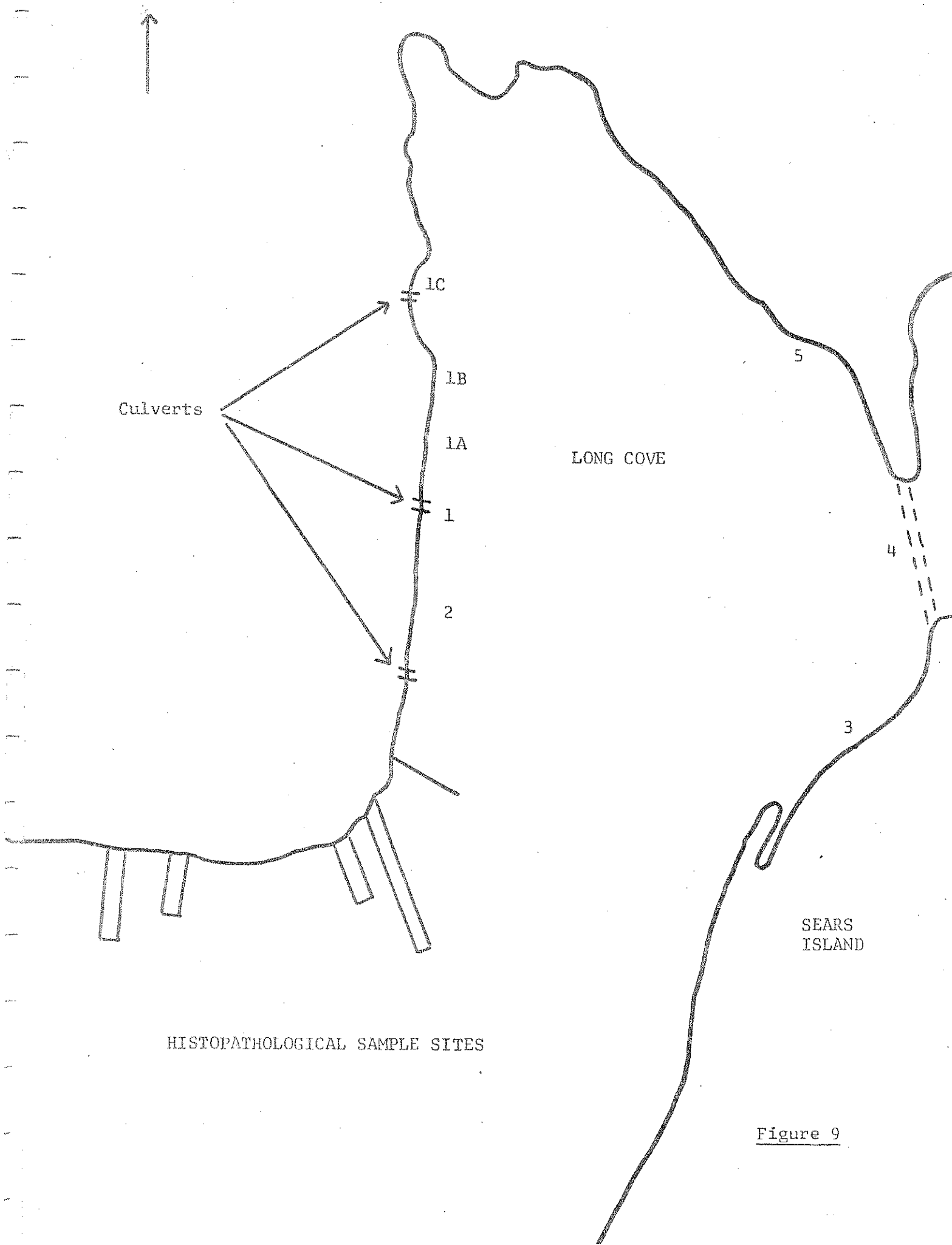


Figure 8

EVALUATION OF AN OIL SPILL SITE

Figure 9

Histopathological Sample Sites



HISTOPATHOLOGICAL SAMPLE SITES

Figure 9

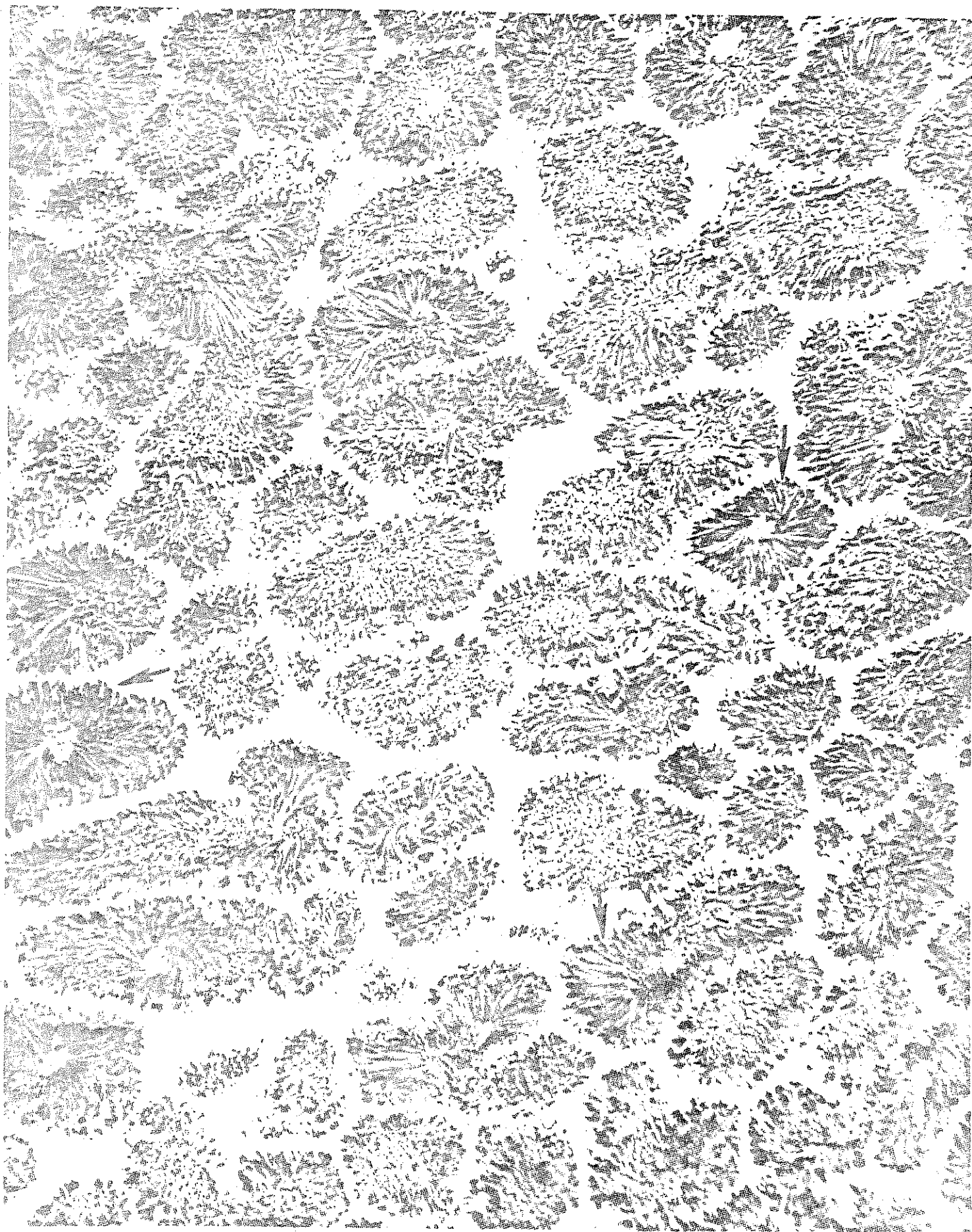
EVALUATION OF AN OIL SPILL SITE

Figure 10

Gonadal tissue - normal male soft shell clam, Mya  
arenaria. Arrows point to follicles filled with sperm.

H&E X360.

Fig. 10



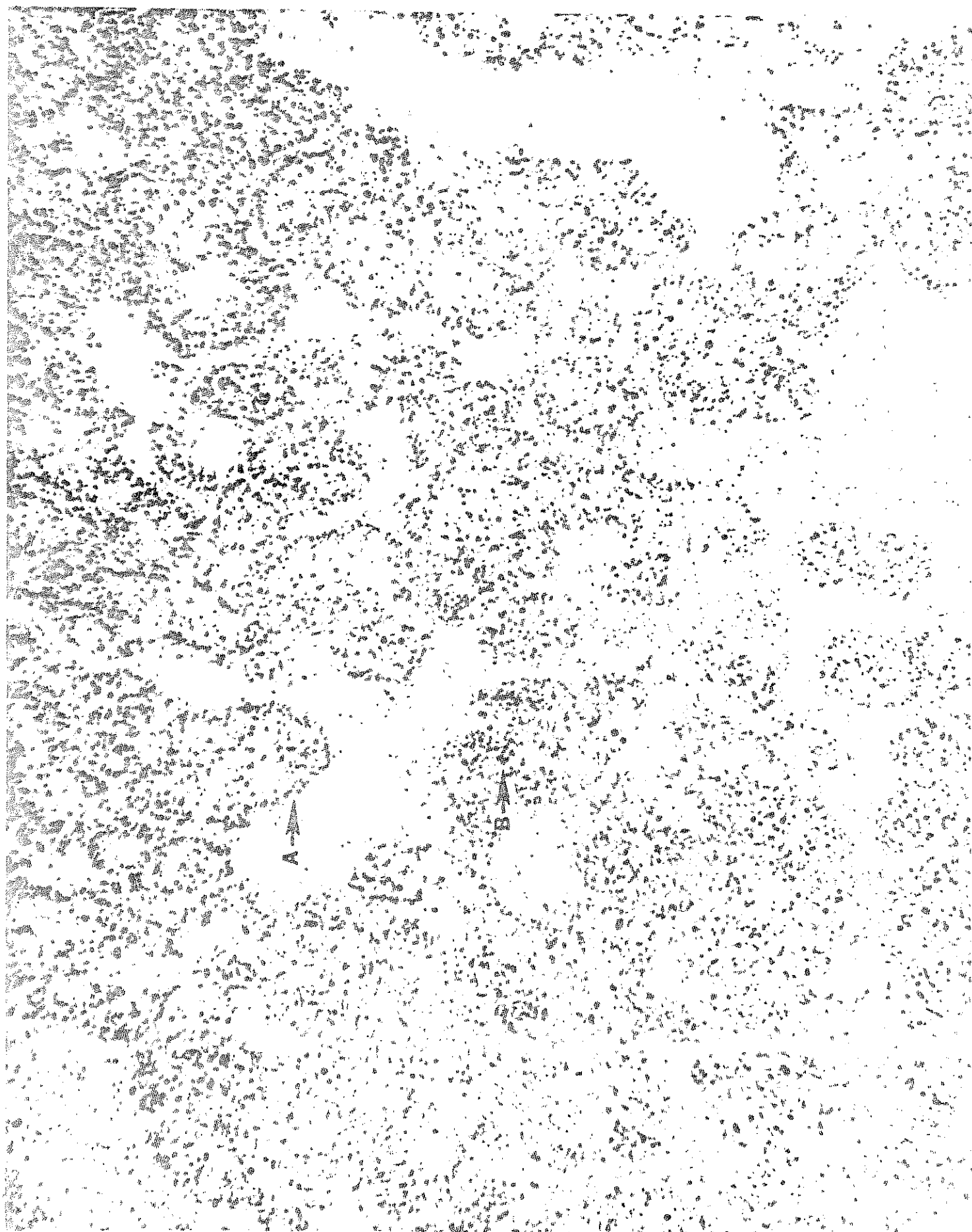


## EVALUATION OF AN OIL SPILL SITE

### Figure 11

Gonadal tissue - normal male soft shell clam, post spawning. Arrow A shows germinal epithelium beginning to develop again. Arrow B shows the atypical cell inclusions normally found in the male gonadal tissue. H&E X360.

Fig. 11

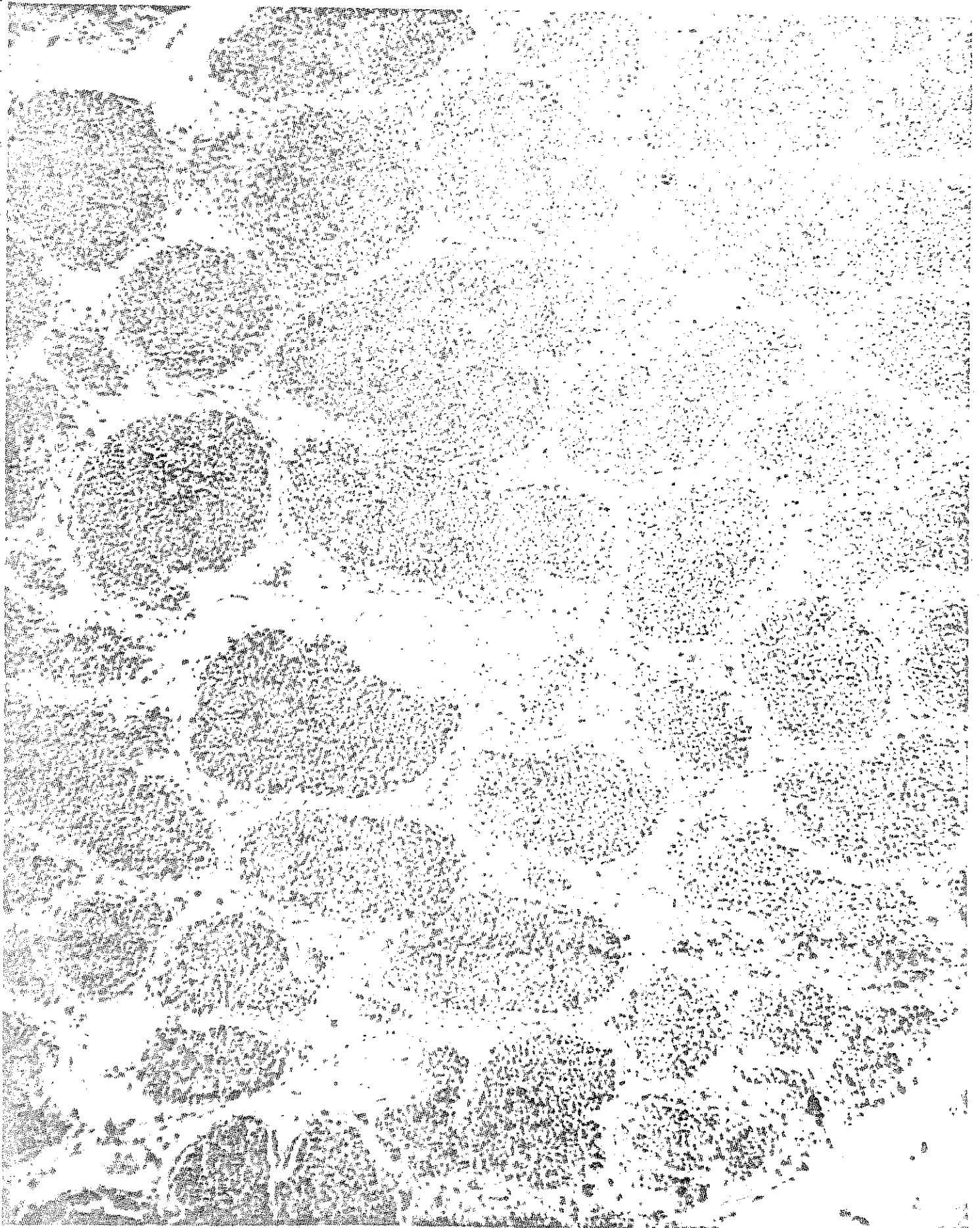


EVALUATION OF AN OIL SPILL SITE

Figure 12

Gonadal tissue - soft shell clam. Follicles filled with  
tumor cells. H&E X360.

Fig. 12



## EVALUATION OF AN OIL SPILL SITE

### Figure 13

Gonadal follicles - male soft shell clam. Arrow A points to normal spawned out follicle with atypical cell inclusions. Arrow B points to follicle filled with tumor cells. H&E X920.

Figure 13

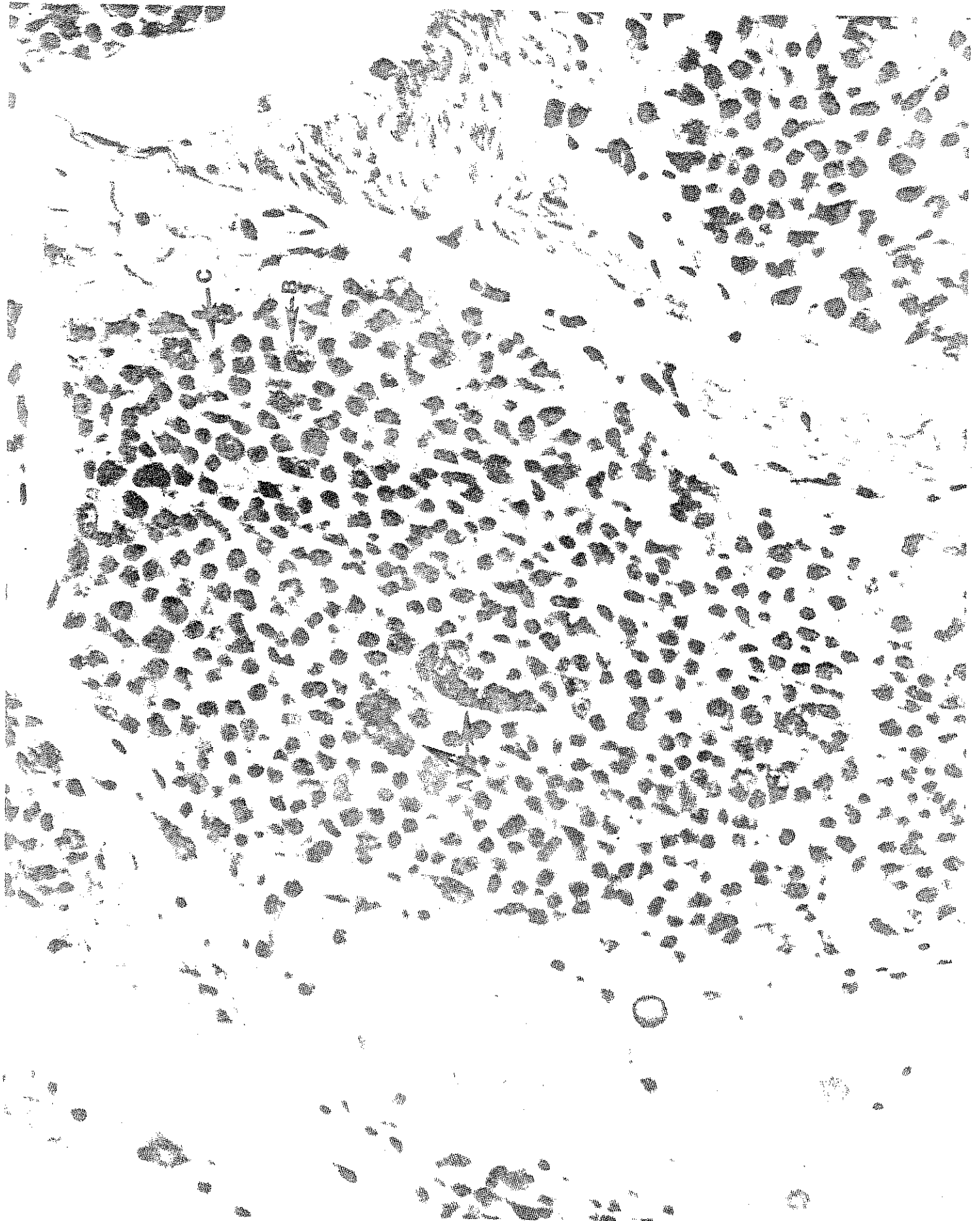


## EVALUATION OF AN OIL SPILL SITE

### Figure 14

Gonadal tissue - post-spawning female soft shell clam.  
Follicle contains tumor cells. Arrows A point to  
multinucleated cells. Arrow B points to mononucleated  
cells. Arrow C cell containing mitotic figures.  
H&E X1480.

Figure 14





EVALUATION OF AN OIL SPILL SITE

Figure 15

Gonadal tissue - post-spawning female soft shell clam.  
Arrow A points to normal post-spawning follicles  
containing follicle cell inclusions. Arrow B points  
to follicles containing tumor cells. H&E X360.

Figure 15

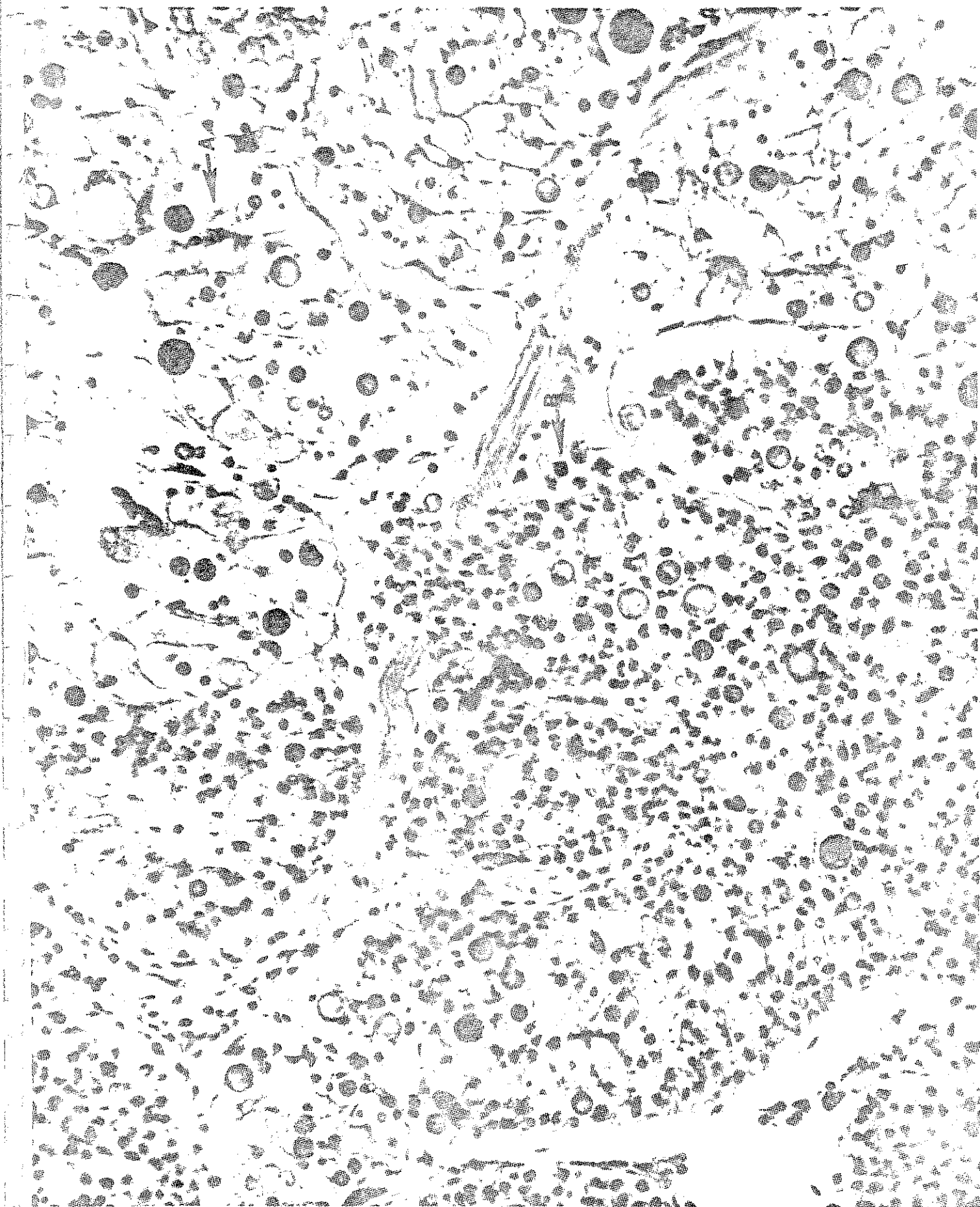


EVALUATION OF AN OIL SPILL SITE

Figure 16

Gonadal tissue - post-spawning female soft shell clam. Arrow A points to normal post-spawning follicles containing follicle cell inclusions. Arrow B points to follicles containing tumor cells. H&E X920.

Figure 16

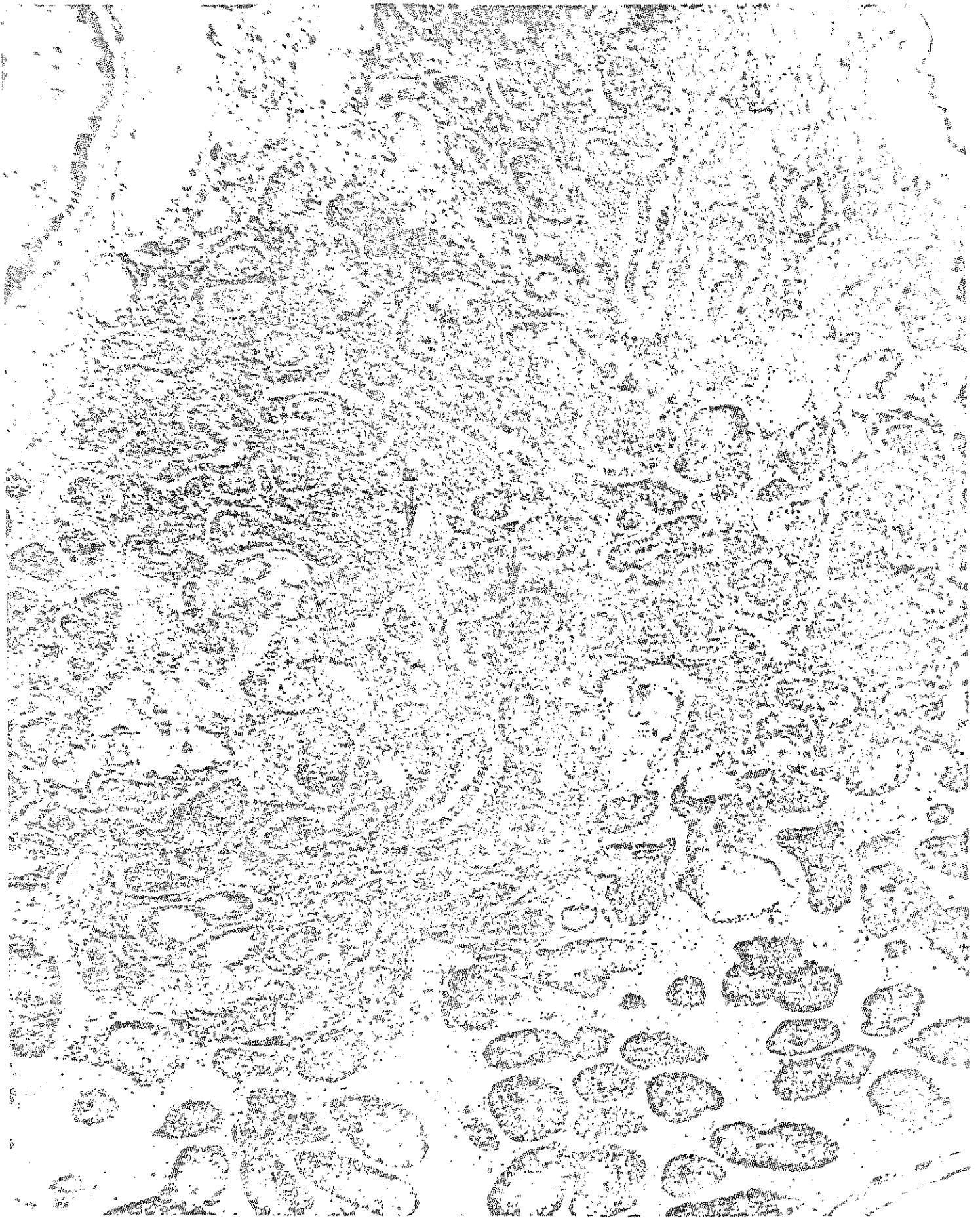


EVALUATION OF AN OIL SPILL SITE

Figure 17

Gonadal tissue - soft shell clam showing invasion of the tumor cells from the follicles to the connective tissue between the follicles. Arrow A points to follicle containing tumor cells. Arrow B shows connective tissue invaded by tumor cells. H&E X360.

Figure 17



EVALUATION OF AN OIL SPILL SITE

Figure 18

Gonadal tissue - soft shell clam showing invasion of the interfollicular connective tissue by the tumor cells. Arrow A points to follicles containing tumor cells. Arrow B points to connective tissue invaded by tumor cells. H&E X920.

## EVALUATION OF AN OIL SPILL SITE

### Figure 19

Area of soft shell clam showing metastasis of tumor cells. Arrow A shows gonadal duct filled with tumor cells. Arrow B shows tumor cells invading the pericardial wall. Arrow C shows kidney invasion. Arrow D shows an area of involvement on the auricle wall. Arrow E shows the invasion of the branchial chamber by the tumor cells.

H&E X360.



Figure 19

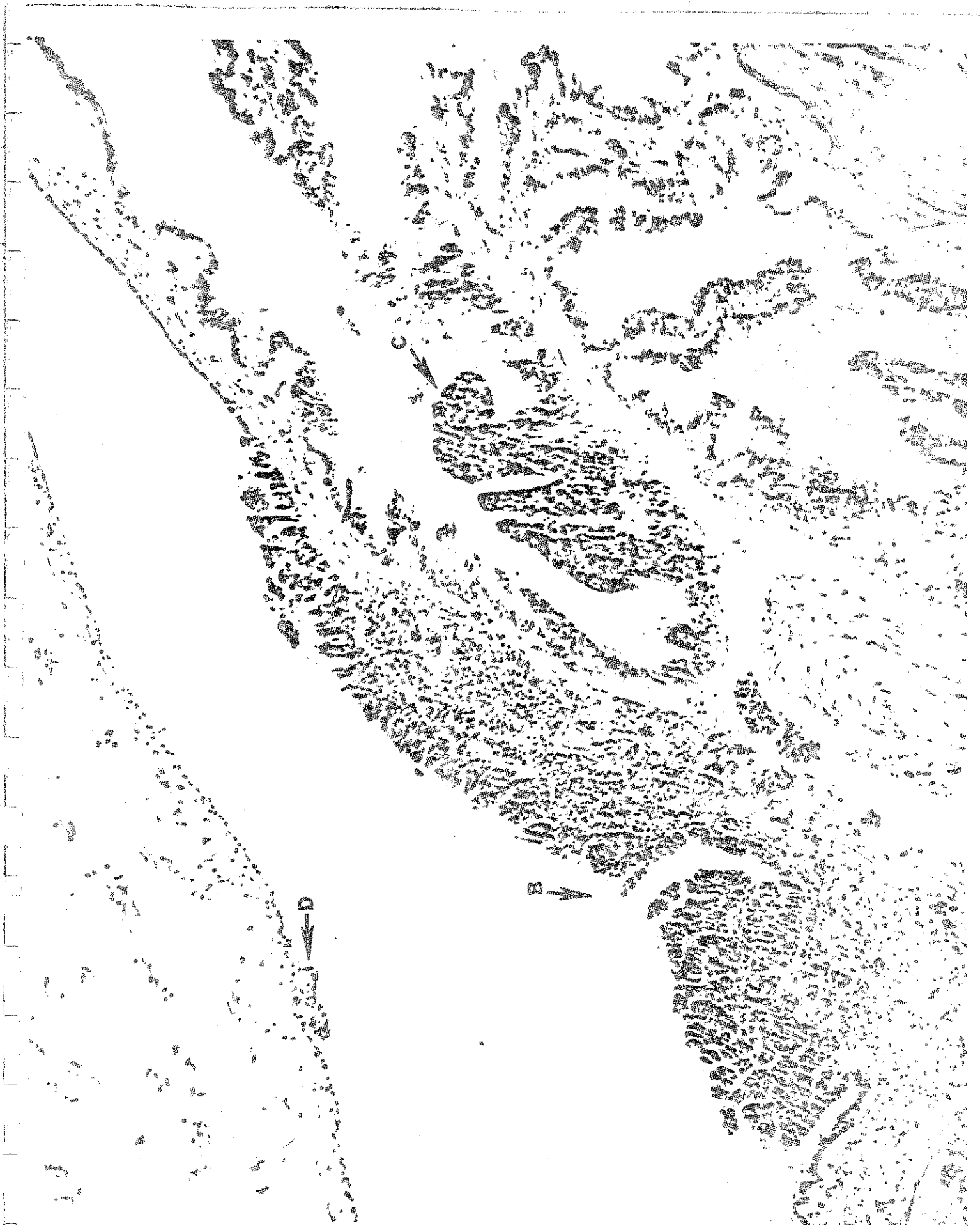


## EVALUATION OF AN OIL SPILL SITE

Figure 20

Higher magnification of Fig. 19 showing metastasis of tumor cells from the gonadal tissue. Arrow B shows invasion of pericardial wall. Arrow C shows invasion of kidney tissue. Arrow D shows involvement of an area on the auricle wall. H&E X920.

Figure 20



## EVALUATION OF AN OIL SPILL SITE

### Figure 21

Soft shell clam collected from Brunswick, Maine, oil spill site. Tumor, possibly of mesenchymal origin, filling body mass. Arrow A points to tumor cells in the interfollicular connective tissue of the gonadal area. Arrow B points to the tumor cells in the sub-mucosa of the intestine. Arrow C points to the tumor cells in the muscular area of the animal. H&E X360.

Figure 21

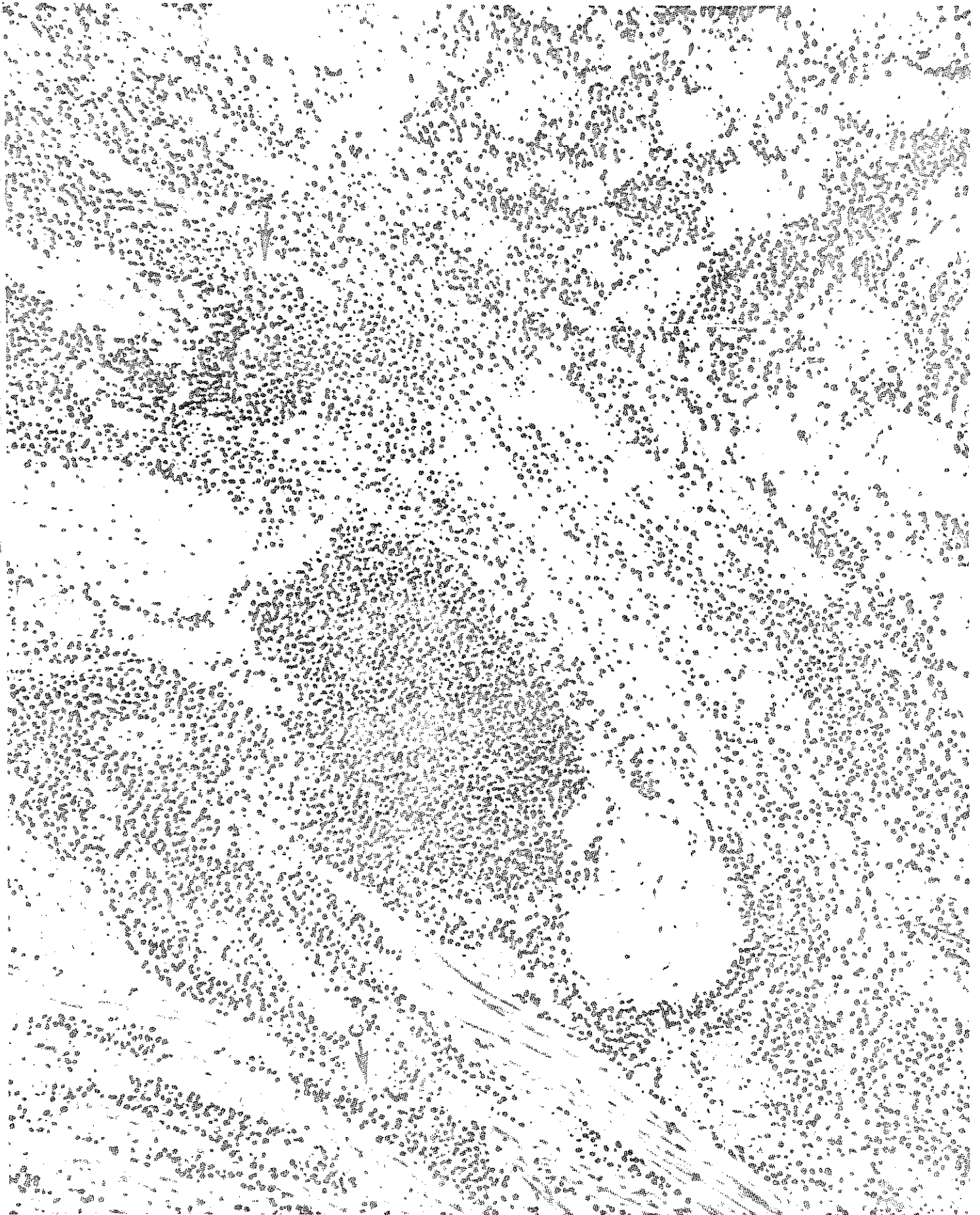


EVALUATION OF AN OIL SPILL SITE

Figure 22

Higher magnification of Fig. 21, possible mesenchymal tumor. H&E X920.

Figure 22



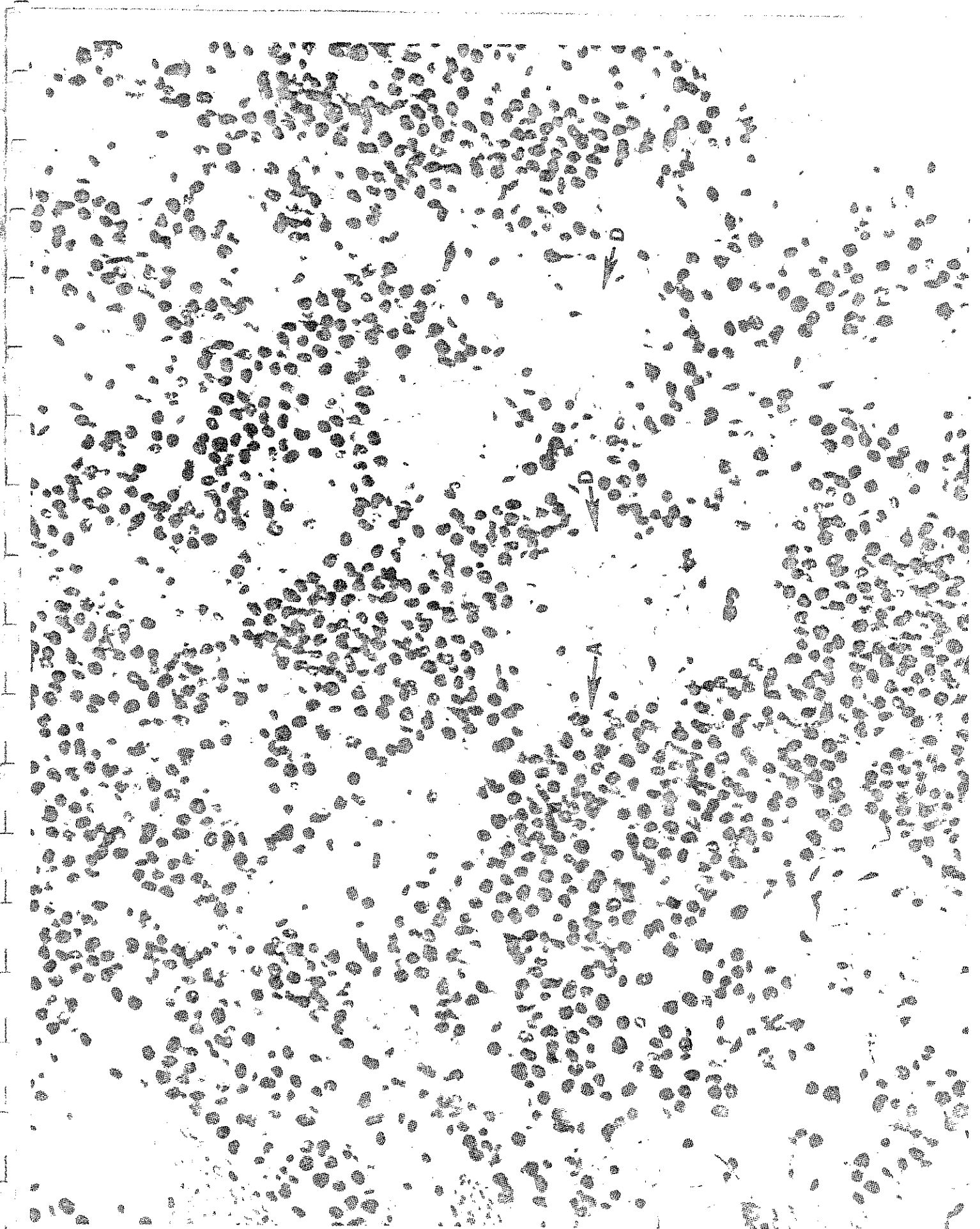
EVALUATION OF AN OIL SPILL SITE

Figure 23

Higher magnification of Fig. 21, possible mesenchymal tumor. Arrow D note-no invasion of gonadal follicles by the tumor cells. H&E X1080.



Figure 23



## EVALUATION OF AN OIL SPILL SITE

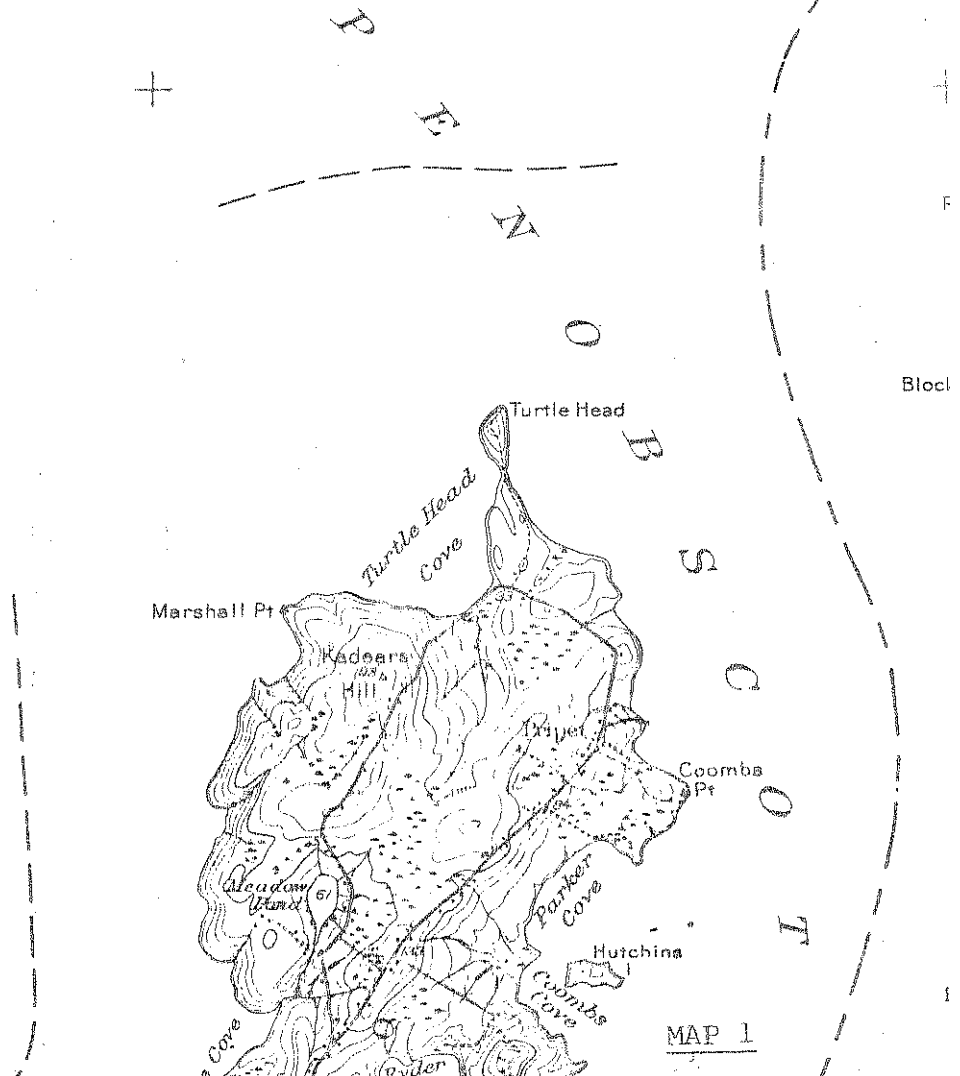
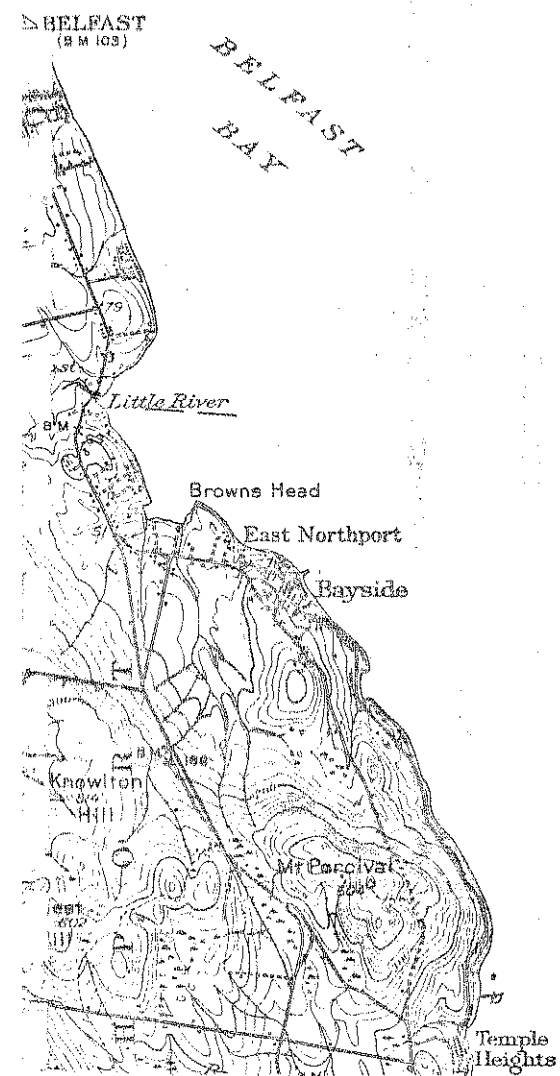
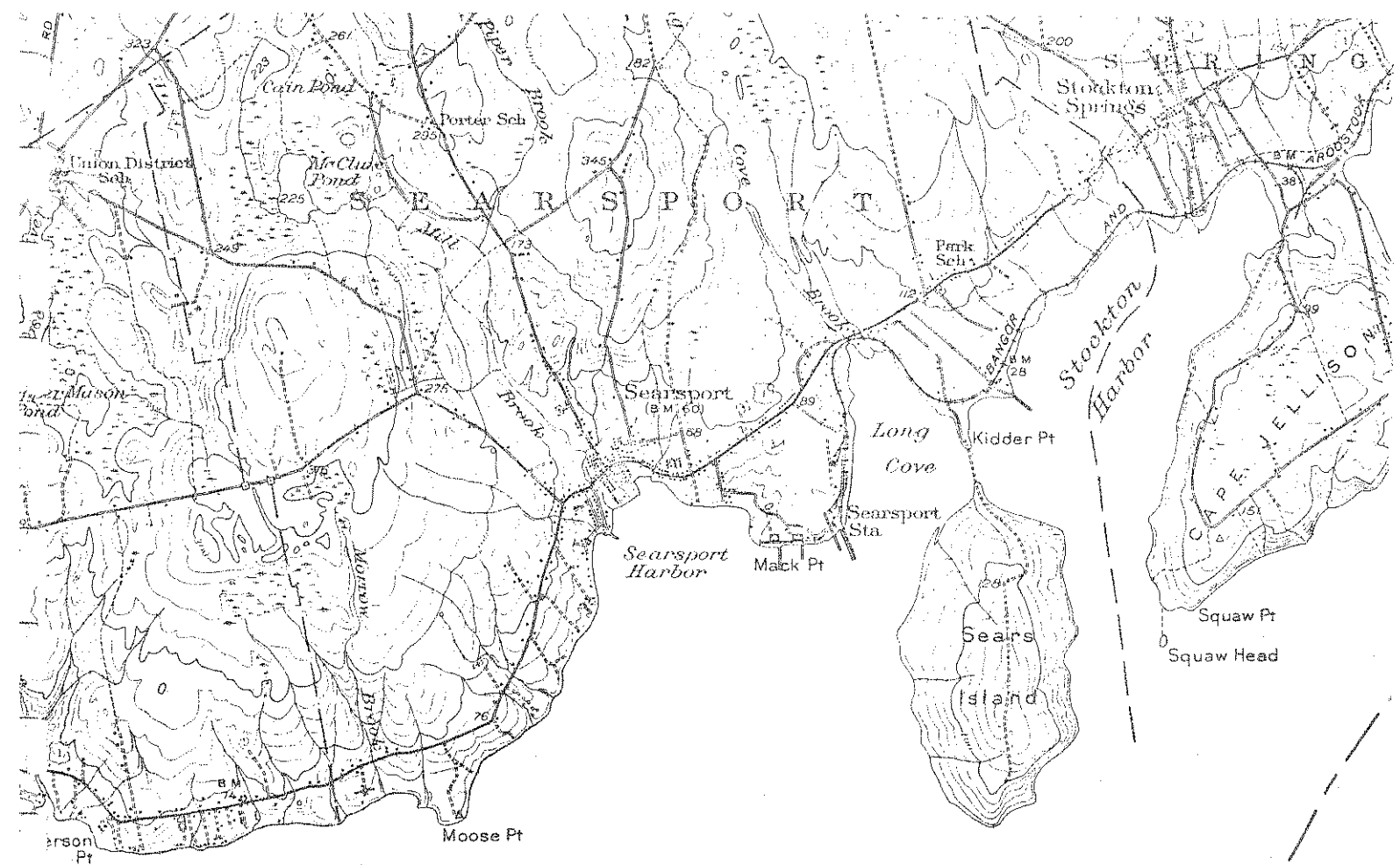
### List of Maps

Map 1 - A portion of Castine, Maine, quadrangle, scale 1:62500,  
showing the area of the spill in Long Cove and Little River.

EVALUATION OF AN OIL SPILL SITE

Map 1

A portion of Castine, Maine, quadrangle, scale 1:62500, showing the area of the spill in Long Cove and Little River.



## EVALUATION OF AN OIL SPILL SITE

### List of Photographs

Photo 1 - Oil slick from March 1971 spill into Long Cove, Searsport.

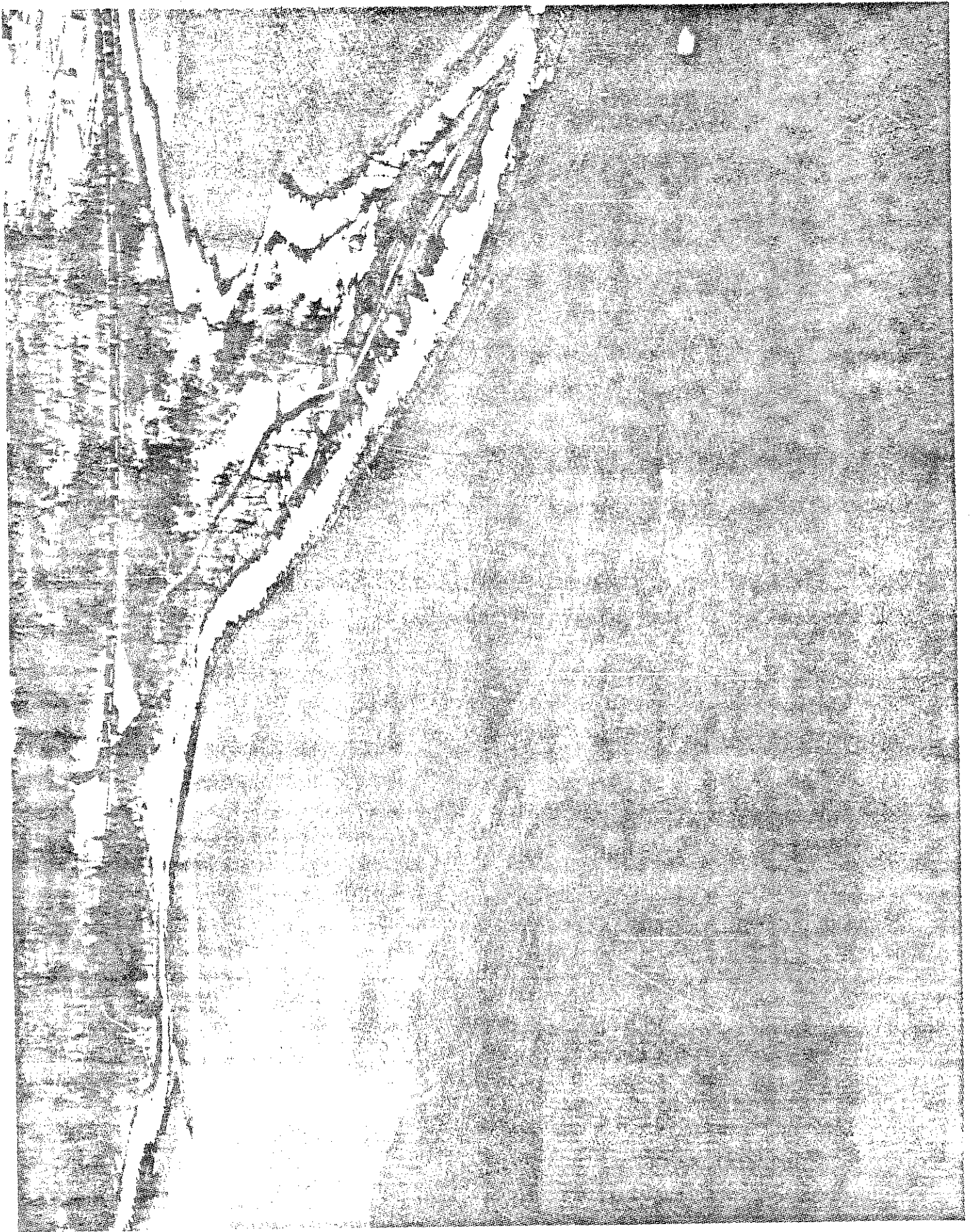
Photo 2 - During initial die-off, many clams surfaced from their burrows and fell over on the flats.

EVALUATION OF AN OIL SPILL SITE

Photo 1

Oil slick from March 1971 spill into Long Cove,  
Searsport.

Photo 1

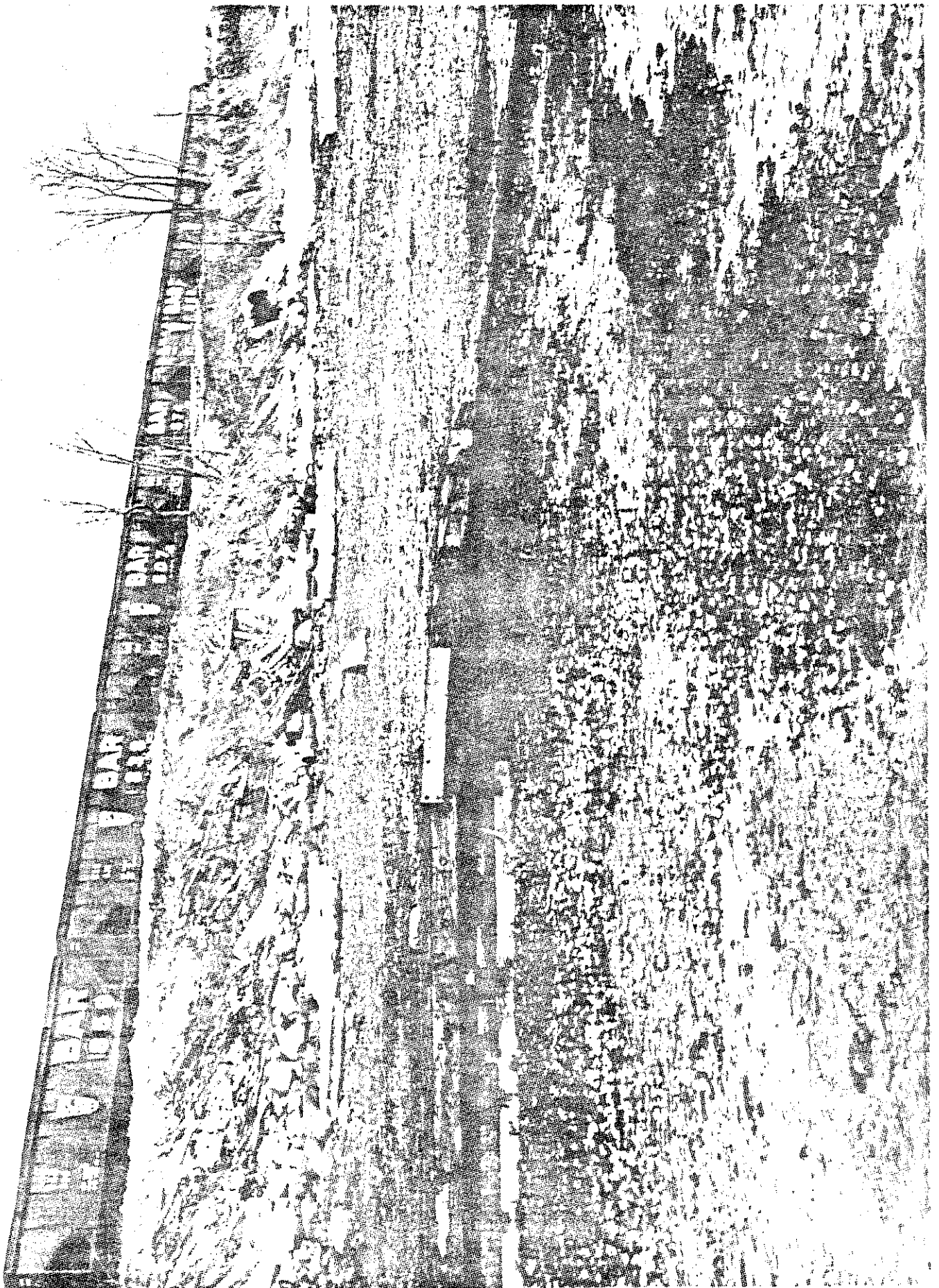


## EVALUATION OF AN OIL SPILL SITE

### Photo 2

During initial die-off, many clams surfaced from their burrows and fell over on the flats.





## EVALUATION OF AN OIL SPILL SITE

### List of Tables

Table 1 - Sediment Samples

Table 2 - Reference Samples

Table 3 - Hydrocarbon Concentrations (Sediments)

Table 4 - Long Cove (Searsport) Clam Samples - Incidence of Oil-Related Tumors

## EVALUATION OF AN OIL SPILL SITE

Table 1 - Sediment Samples

<u>Code Name</u>	<u>Detailed Location Description</u>	<u>Collector</u>
I-1A	#8 400 yds south of culvert, near concrete building	(R&S)
I-2A	#7 300 yds south of culvert	(R&S)
I-3A	#6 250 yds south of culvert	(R&S)
I-4A	#5 200 yds south of culvert	(R&S)
I-5A	#4 140 yds south of culvert	(R&S)
I-5B(1)	area 1 bottle A subtidal	
I-5B(2)	area 1 " B "	
I-6A	#3 70 yds south of culvert	(R&S)
I-6B	area 2 subtidal	
I-6C	area 2-4 (100 ft below MHT)	
I-7A	#2 culvert halfway between H&L tides, 100 yds from #1	(R&S)
I-7B	area 3 subtidal	
I-8A	#1 NW side	(R&S)
I-8B	area 4 subtidal	
I-8C	area 4-3 (180 ft below MHT)	
I-9B	area 5 subtidal	
I-10A(1)	#1 NW side, along RR tracks	(Sheldon)
I-10A(2)	#1 W side - back of RR station	(Sheldon)
I-11A	#2 back of RR station, going N	(Sheldon)
I-11B	area 6 subtidal	
I-11C	area 6-6 (30 ft below MHT)	
I-12A	#3 back of RR station, going N	(Sheldon)
I-13A	#4 100 ft from #3 towards water	(Sheldon)
I-13B	area 7 subtidal	
I-13C	area 7-6 (160 ft below MHT)	

# EVALUATION OF AN OIL SPILL SITE

Table 1 continued

<u>Code Name</u>	<u>Detailed Location Description</u>	<u>Collector</u>
I-14A	#5 E side	(Sheldon)
I-15C	area 8-7 (180 ft below MHT)	
I-16C	#7 E side	(Sheldon)
I-17A	#3 NE side	(Farrin)
I-18A	#5 SE side	(Farrin)
I-19A	#8 east end bar	(Farrin)
I-20C	area 9-8 (115 ft below MHT)	
I-21C	area 10-3 (410 ft below MHT)	
I-22C	#10 subtidal	(Ricker)
I-23C	#1 "	(Ricker)
I-24C	#2 "	(Ricker)
I-25C	#3 "	(Ricker)
I-26C	#4 "	(Ricker)
I-27C	#6 "	(Ricker)
I-28A	#10 back of Purina	(R&S)
I-28C	#12-4 (80 ft below MHT)	
I-28D	#12 subtidal	
I-29A	#9 NW of Purina - 300 yds	(R&S)
I-29C	#11-1 (117 ft below MHT)	
I-29D	#11 subtidal	
I-30A	#10 next cove near stream	(R&S)
I-30C	#13-2 (150 ft below MHT)	
I-30D	#13 subtidal	
I-41A	next cove south of Purina	(R&S)

( ) Indicates the collection of subsamples  
MHT = Mean High Tide Mark

# EVALUATION OF AN OIL SPILL SITE

Table 2 - Reference Samples

<u>Code Name</u>	<u>Detailed Description</u>
REF-1	reference sample of JP5 jet fuel
REF-2	reference sample of #2 oil
REF-3	oil taken coming out of culvert by F. Ricker used as reference sample

Table 3 - Hydrocarbon Concentrations (Sediments)

<u>Code Name</u>	<u>HC (ppm)</u>	<u>Code Name</u>	<u>HC (ppm)</u>	<u>Code Name</u>	<u>HC (ppm)</u>	<u>Code Name</u>	<u>HC (ppm)</u>	<u>Code Name</u>	<u>HC (ppm)</u>
I-1A	137								
I-2A	(93)								
I-3A	254								
I-4A	212								
I-5A	143	I-5B(1)	256						
		I-5B(2)	53						
I-6A	88	I-6B	125	I-6C	150				
I-7A	74	I-7B	210						
I-8A	52	I-8B	82	I-8C	43				
		I-9B	230						
I-10A(1)	53								
I-10A(2)	88								
I-11A	39	I-11B	705	I-11C	43				
I-12A	58								
I-13A	49	I-13B	330	I-13C	93				
I-14A	59			I-15C	40				
I-16A	7								
I-17A	158								
I-18A	(82)								
I-19A	32								
				I-20C	54				
				I-21C	46				
				I-22C	200				
				I-23C	160				
				I-24C	140				
				I-25C	64				
				I-26C	52				
				I-27C	152				
I-28A	81			I-28C	18	I-28D	41		
I-29A	47			I-29C	16	I-29D	14		
I-30A	46			I-30C	168	I-29D	35		
I-31A	54								

Table 4

LONG COVE (SEARSPORT) CLAM SAMPLES  
INCIDENCE OF OIL-RELATED TUMORS

Sta. #	Number of Tumors										Total Tumors	Total Clams	% with Tumors
	July '71	Aug. '71	Jan. '72	Aug. '72	June '73	July '73	Oct. '73	Dec. '73	Aug. '74	Sept. '74			
1	4	12	1		3	4	5	1		13	4 12 1 3 4 5 1 13	15 200 50 50 51 59 40 115	27 6 2 6 8 8 3 11
1-A		1	4						2		1 4 2 4 8 10 9	46 50 22 47 47 50 51	2 8 9 8 17 20 18
1-B		4									1 3 1 5 6 1	14 100 49 28 28 47	7 3 2 18 21 2
1-C		10									1 3 1 5 6 1	14 100 49 28 28 47	7 3 2 18 21 2
2	1	3	1								1 3 1 5 6 1	14 100 49 28 28 47	7 3 2 18 21 2
4					6						1 3 1 5 6 1	14 100 49 28 28 47	7 3 2 18 21 2
5		1									1 3 1 5 6 1	14 100 49 28 28 47	7 3 2 18 21 2
Total	5	31	23	6	3	4	5	1	7	13	98	1159	8.5

EVALUATION OF AN OIL SPILL SITE