International Council for the
Exploration of the Sea

ICES C.M.1980/E:10
Marine Environmental
Quality Committee

STUDIES ON POLYCYCLIC AROMATIC HYDROCARBONS IN WESTERN ATLANTIC SHELLFISH

by

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Abstract

Levels of non-alkylated polycyclic aromatic hydrocarbons (PAH) were determined in lobsters, clams and mussels from several areas in New Brunswick and Nova Scotia. In general, there were no significant differences in PAH levels between animals from control areas and areas with known inputs. Analysis of individual animals or pooled samples showed that widely divergent levels of PAHs are present in animals from the same area. PAH studies by other nations are encouraged.

Résumè

Les concentrations d'hydrocarbures aromatiques polycycliques non alcoylés ont été étudiées chez des homards, des coques et des moules provenant de plusieurs régions distinctes et maintenus dans diverses conditions. Il a été en général impossible de trouver des témoins chez lesquels les niveaux étaient nettement inférieurs à ceux d'animaux provenant de régions à apports d'hydrocarbures connus. L'analyse d'animaux individuels démontre qu'il existe des concentrations de HAP très différentes parmi les sujets d'une même région. Nous recommandons aux autres pays d'entreprendre des études sur les HAP.

During the last couple of years the Marine Environmental Quality Committee has been receptive to studies on so-called "new" or previously unstudied marine pollutants. One group of pollutants which has not been studied as part of the ICES Coordinated Monitoring Studies are the polynuclear or polycyclic aromatic hydrocarbons (PAHs). These compounds (the smallest being three condensed rings such as anthracene) may or may not contain alkyl or other groups.

Many PAHs are known carcinogens, co-carcinogens or initiators. PAHs are, in general, organic fuel combustion products, and the quantity formed increases as combustion efficiency decreases. The ratio of alkylated PAHs to non-alkylated PAHs decreases as the combustion temperature increases (Blumer, 1977). On an industrial scale the destructive distillation of bituminous coal yields coal tar which is also a rich source of PAHs. Coal tar is redistilled to yield creosote and coal tar pitch, all three products which have wide useage in marine applications such as the preservation of wood structures and marine paints. Liquid petroleum hydrocarbons also contain PAHs. Bilge discharge, natural seepage and tanker disasters are potential sources of PAHs to the marine environment (Suess, 1976; Bravo, 1978).

PAHs are relatively water insoluble, stable, and are likely to be taken up by biota. However, many biota containing inducible microsomal enzymes (mixed function oxidases, aryl hydrocarbon hydroxylase) are able to metabolize PAH and excrete the metabolic products (National Academy of Sciences, 1972). Bivalves, such as mussels, and crustaceans, such as lobsters, lack these enzymes and elevated levels of PAH are likely to be found in such species (Payne, 1977).

Levels of PAHs of from 0.1 - 11 µg/kg wet weight have been reported in shellfish off the eastern coast of the United States (Pancirov, 1977; Brown, 1979), and studies by Hites et al. (1977) and Farrington et al. (1977) have demonstrated that an increase in non-alkylated PAHs has occurred in marine sediments from off the eastern coast of North America.

This paper reports some preliminary results of our investigation of non-alkylated PAH in clams, mussels and lobsters.

Materials and Methods

Shellfish homogenates were prepared and frozen in solvent washed glass jars. Following KOH saponification (Dunn, 1979), the residual lipids and PAHs were extracted with iso-octane then transferred to toluene for column chromatography on 30 g of 5% H_2O deactivated Florisil. PAHs were eluted with 200 ml of toluene, transferred to 5 ml of dimethyl sulfoxide (DMSO), and extracted into iso-octane from aqueous DMSO, washed with water and then finally prepared for HPLC in 100 µl DMSO. HPLC was carried out on a Vydac 201 TP reverse phase column (10 µ, 3.2 mm id, 25 cm L) using a linear gradient (70 to 94% acetonitrile/water [v/v] over 20 minutes).

Both internal and external standardization were used as required. Retention times with fluorescence and u.v. absorption detection were used for identification. Peak heights and areas were used for quantitation.

Results and Discussion

Levels of individual non-alkylated PAH compounds found in shellfish from a number of selected areas are shown in Table 1. Sample locations (areas 1-12) are shown in Figure 1. As part of our initial investigation into PAH compounds in shellfish, we initially determined PAH levels in lobster (Homarus americanus) from:

(1) areas selected as controls
(2) an estuary with known industrial input - the Miramichi Estuary New Brunswick, Canada
(3) an aquarium utilizing Halifax Harbour water - a metropolitan area with industrial port activities
(4) areas contaminated by the Kurdistan oil spill
(5) a large commercial lobster pound.

In addition, blue mussels (Mytilus edulis) and clams (Mya arenaria) from the Kurdistan oiled area were analyzed as were samples of creosote used to treate marine piles and oil from the Kurdistan spill. Individual PAH levels in lobster vary widely from location to location. Within the coastal areas sampled it was not possible to find lobsters free of PAHs. This is not to imply that lobsters essentially free of PAH do not exist in coastal regions but that apparently more widespread contamination by PAHs exists in the region than had been previously suspected. Oil from the tanker "Kurdistan" proved difficult to analyze due to the presence of a large broad peak underlying the individual PAH peaks (Fig. 2). This meant that quantitative measurements should only be taken as a first approximation.

The presence of a large complex PAH profile in petroleum is not unexpected since it should be remembered that although petroleum hydrocarbons are known to contain PAHs, the bulk of the PAHs contained in crude oil are alkylated (Blumer, 1977). Processing of crude oil, especially with catalytic cracking, can increase the amount of non-alkylated PAHs (Trosset et al., 1978) so it is not unexpected to find that bunker "C" is enriched in PAH. We do not have the facilities required for separation and characterization of such a complex mixture of PAHs. It was hoped that a non-alkylated PAH pattern could be obtained from the bunker "C" from the Kurdistan to utilize it as a confirmatory technique in analyzing oiled shellfish. Consideration of lobster tail muscle and digestive gland PAH profiles suggest that, unlike material such as PCB, PAHs undergo substantial metabolism in that very dissimilar PAH patterns are present in each tissue. Bivalves such as clams and mussels from oiled areas have appreciable quantities of non-alkylated PAHs in their tissue and generally show the same qualitative and semi-quantitative patterns. This pattern is, however, somewhat closer to that present in the creosote sample than that of Kurdistan oil (Figs. 2 & 3).

Lobsters were taken from four sampling stations across the Miramichi estuary (Fig. 1, Nos. 9-12). Samples of tail meat and digestive gland from each sampling station were prepared by pooling equal weights of these tissues from 25 lobsters. Large variations in PAH concentrations were obtained as is common with other contaminants and a large number of samples would have to be pooled in order to obtain results within reasonable confidence limits.

The variability from animal to animal can be seen in the highly different levels of PAH found in digestive glands of two lobsters which had been held in our aquarium for over one year (Table 1).

The sample of impounded lobster had substantially higher PAH levels than levels from other samples. This is expected since Dunn and Fee (1979) have shown lobsters pick up PAHs over the duration of their impoundment.

Finally the animals which were obtained from areas not obviously oiled did not have the large broad underlying peak in their chromatograms. This suggests that other sources are responsible for the PAH contamination observed in these animals.

Coastal contamination by PAHs is undoubtedly a result of multiple sources and types of input. Creosote is given only as an example of one source of direct input into the marine environment. Other sources such as internal combustion, space heating, discharge of used petroleum products, asphalts and pitches all have pathways of handling leading to the oceans.

We will continue investigation into the presence of PAHs in shellfish, their occurrence, nature, uptake and depuration and would encourage activity by other nations in this area.

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Table 1

PAH Concentrations (µg/kg) in Selected Marine Samples

Specimen Location	Creosote* (Marine Application)	Bunker* "C"**	Control Lobster Area 2		Control Lobster Area 1	Oiled Lobster Area 4		Aquarium Lobster Halifax Lab		Pound Lobster Commercial		Lobster Areas 9-12	Oiled Area 7	Oiled Area 8
Tissue		Kurdistan	Tail Muscle	Digestive Gland	Digestive Gland	Tail Muscle	Digestive Gland	Digestive Gland	Digestive Gland	Tail Muscle	Digestive Gland	Digestive Gland	Blue Mussel Meat	Clam Meat
Number of Individuals	-	-	17	12	>1	14	14	1	1	10	10	4 X 25	10	10
Phenanthrene	292	43	n.d.	n.d.	-	n.d.	1588	n.d.	n.d.	28	11	618 + 120 ($\bar{19}$.9%)	462	733
Fluoranthene	24	2	2	n.d.	64	1	318	11	507	43	373	36.8 + 5.2 ($\bar{14}$.1%)	62	18
Triphenylene	trace	60	25	n.d.	-	n.d.	-	-	-	-	-	trace	-	-
Pyrene	109	1	5	n.d.	491	n.d.	488	16	1595	44	1571	63.5 + 34.5 ($\bar{54}$%)	131	85
Chrysene	13	3	7	140	-	n.d.	445	20	1119	81	375	29.6 + 9.0 ($\bar{30}$%)	205	209
Benzo(a)-anthracene	75	trace	37	655	146	23	684	14	592	11	1215	71 + 42 ($\bar{59}$%)	203	172
Benzo(e)pyrene	11	5	3	n.d.	n.d.	8	57	6	325	29	626	76.3 + 46.3 ($\bar{61}$%)	10	17
Benzo(b)-fluoranthene	3.2	0.5	2	17	12	1	24	3	305	7	114	10.8 + 4.4 ($\bar{41}$%)	25	23
Benzo(k)-fluoranthene	1.0	trace	0.2	2	2.3	0.3	7.6	4	1.3	1.3	22	1.5 + 0.7 ($\bar{47}$%)	1.3	2.3
Benzo(a)pyrene	1.5	0.3	1.5	18	10	0.2	24	6	582	1.7	40	4 + 3.4 ($\bar{85}$%)	21	12
Benzo(ghi)-perelene	trace	trace	n.d.	n.d.	n.d.	n.d.	n.d.	10	1395	5	129	trace	-	-

* = expressed in g/kg

** = quantification of PAHs difficult because of large underlying broad peak

\- = not measured due to peak uncertainty

n.d.= none detected (either uncertain in peak identification or too small to measure <<5 x baseline)

Figure 1

1 Halifax Harbour
2 3 Port Hood
4 5 6 Chedabucto Bay
7 Dominion Beach (Lingan)
8 Scatarie I (south side)
9 10 11 12 Miramichi estuary

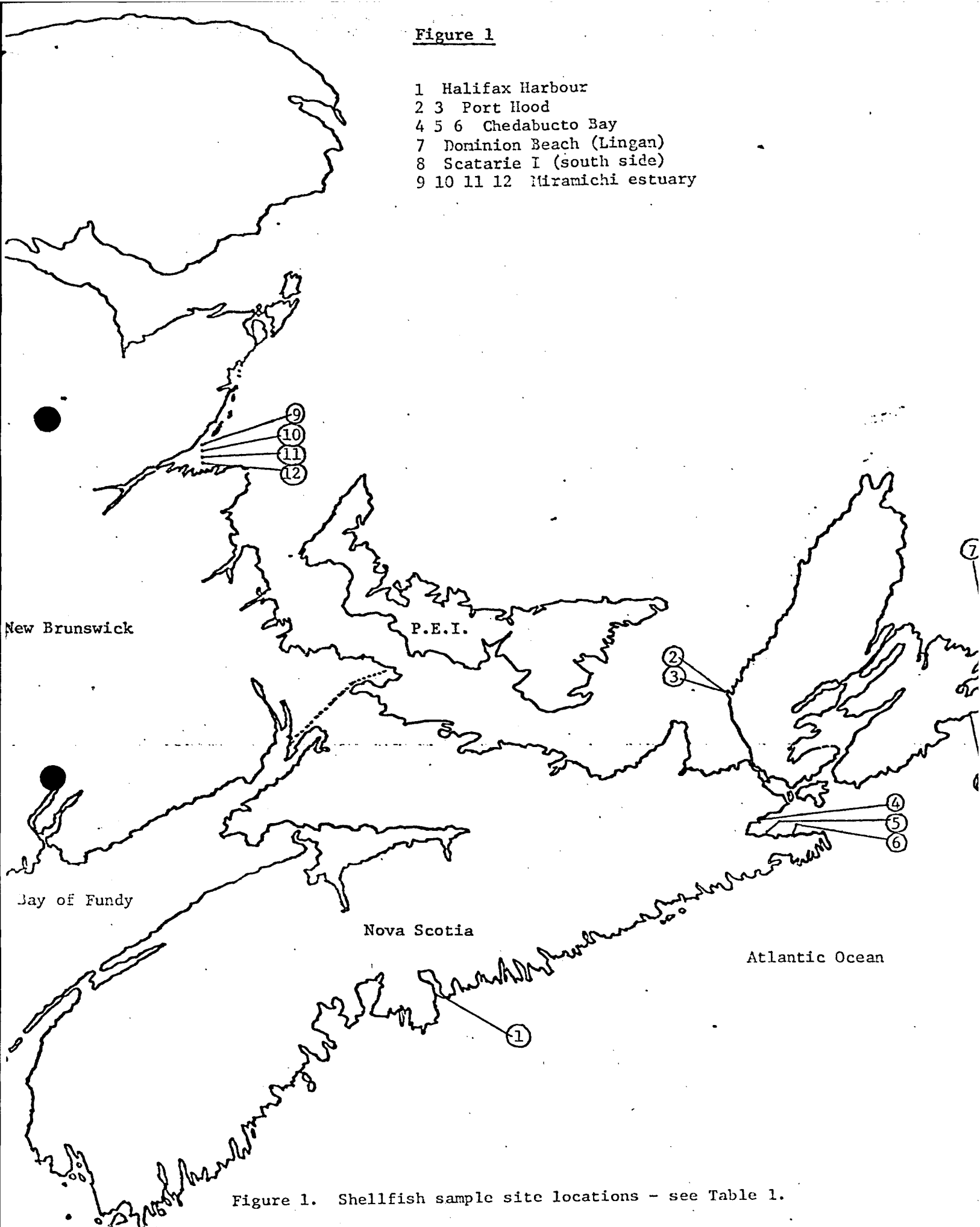

Figure 1. Shellfish sample site locations - see Table 1.

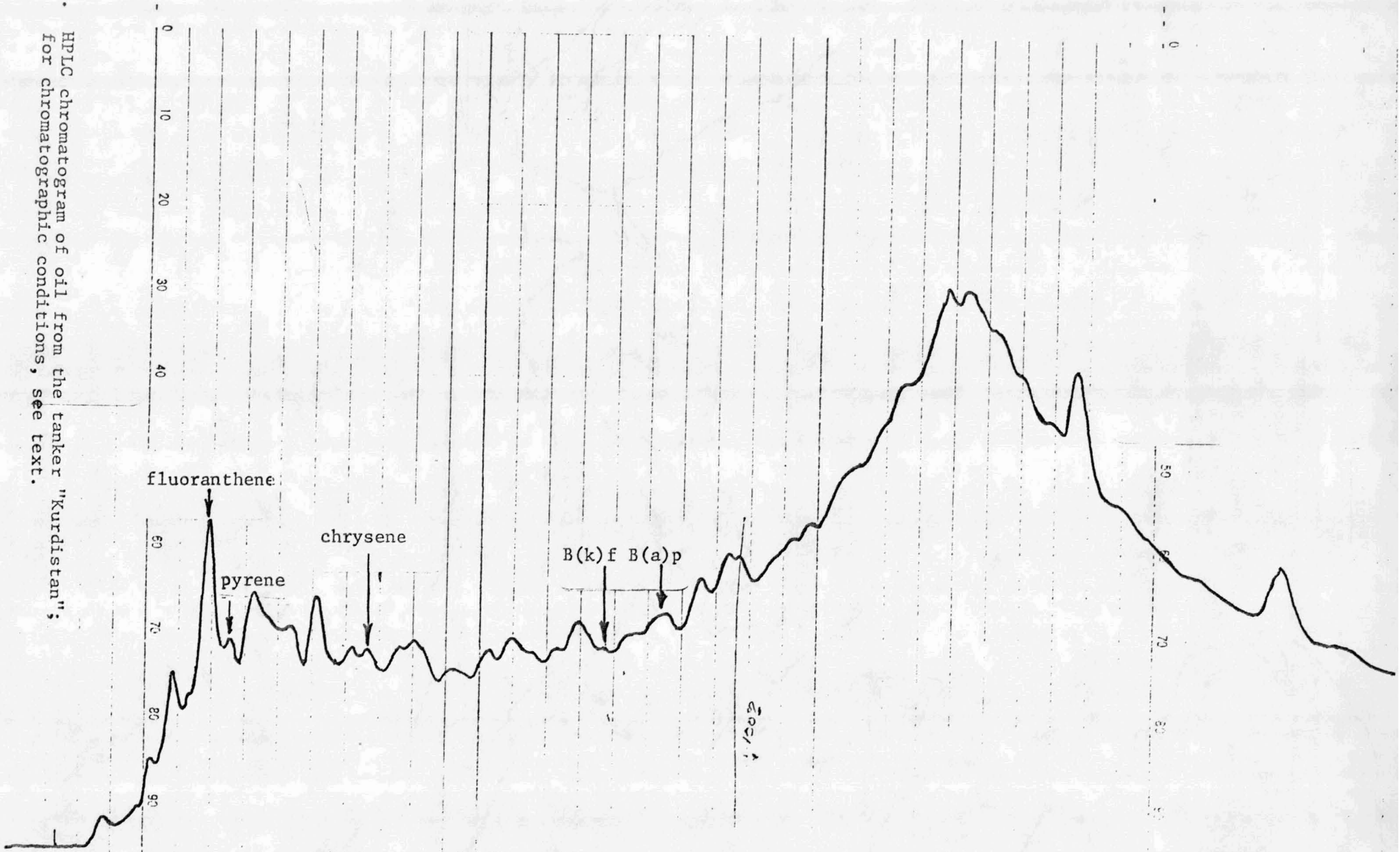

Figure 2. HPLC chromatogram of oil from the tanker "Kurdistan"; for chromatographic conditions, see text.

B(k)f: Benzo(k)fluoranthene
B(a)p: Benzo(a)pyrene

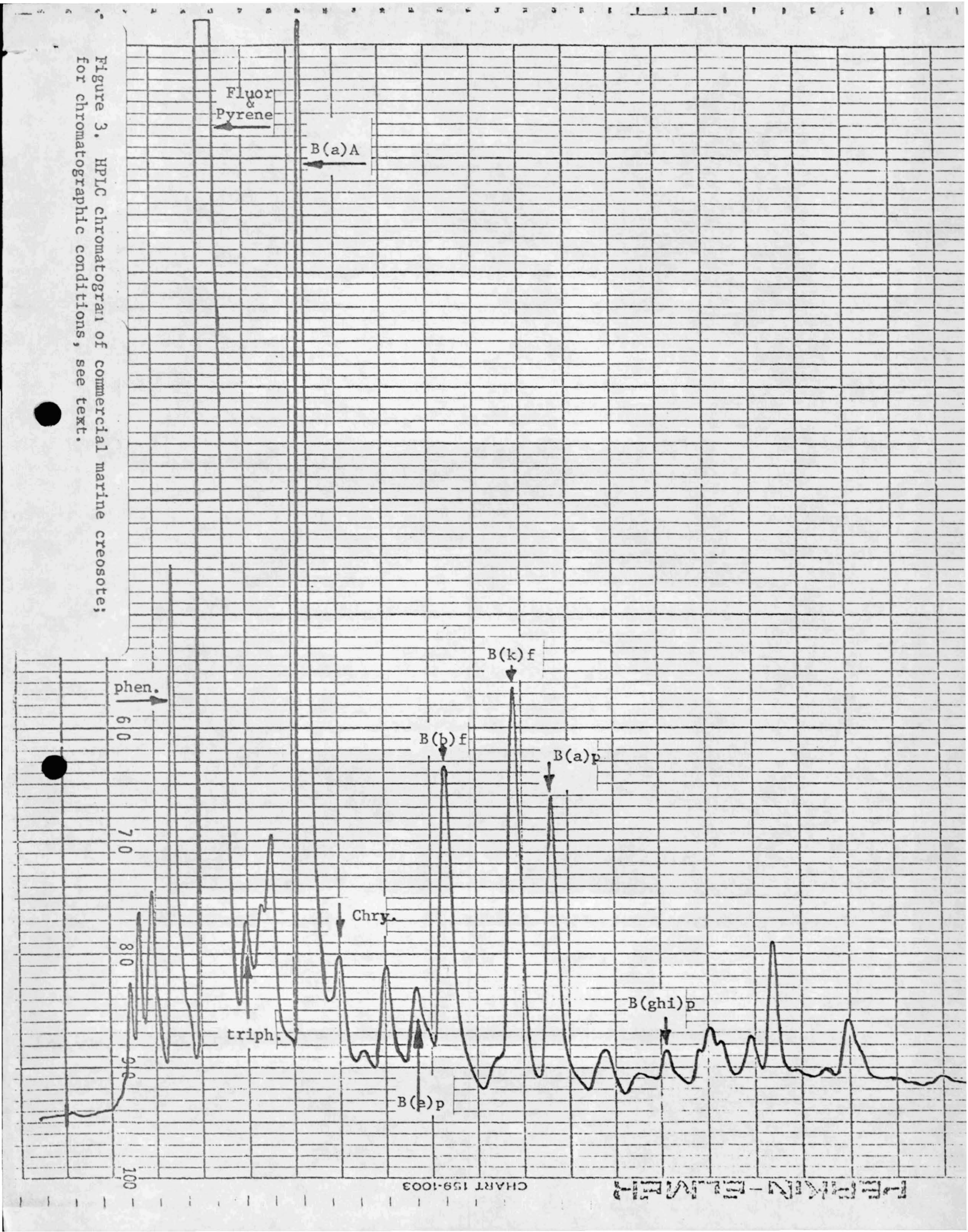

Figure 3. HPLC chromatogram of commercial marine creosote; for chromatographic conditions, see text.