Technical

Polycyclic Aromatic Hydrocarbons in Crude and Deodorized Vegetable Oils

B.K. Larsson*, **A.T. Eriksson** and **M. Cervenka**
Food Laboratory, National Food Administration, Box 622, S-751 26 Uppsala, Sweden

The efficiency of the refining process in removing polycyclic aromatic hydrocarbons (PAH) from crude vegetable oils was studied. Samples of the crude oils (coconut, soybean and rapeseed oils) and the corresponding refined, deodorized oil were taken on-line in three Swedish oil refineries and margarine manufacturing plants and analyzed for 20 different PAHs. Of the crude oils, coconut oil had by far the highest PAH levels. However, the PAH levels in the refined coconut oils were very low. This shows that the activated charcoal treatment used for removing PAHs from coconut oil achieves the desired effect. The crude soybean and rapeseed oils contained relatively low, but varying, amounts of PAH. At present these oils are not purified by activated charcoal. Nevertheless, the PAH levels in the refined oils were considerably lower than those in the corresponding crude oils. This probably is due to evaporation of PAH in the deodorization process, where steam is passed through the hot oil under high vacuum. However, deodorization has only a marginal effect on the high molecular PAHs, of which several are classified as carcinogens.

Polycyclic aromatic hydrocarbons (PAH) have attracted a wide interest in food analysis circles because of their mutagenic and carcinogenic properties. Smoked foods, in particular, have been subjected to a large number of surveys for PAH. A maximum limit for benzo[a]pyrene (BaP) in smoked meat products (1 μg/kg) has been established in food legislation of the Federal Republic of Germany, Austria and Poland.

The presence of PAH in vegetable oils has been reported by various investigators (1-12). Different routes of contamination have been suggested. These include:

- Uptake by the oil plants from contaminated soils (10,13).
- Atmospheric pollution of the oil plants (10,13).
- Direct drying of the oilseeds with combustion gases (5,10).
- Uptake from petroleum-based solvents used in the extraction of the oils from oilseeds (10,14).

Particularly high levels of PAH have been found in crude coconut oils (2-5), because the copra from which the oil is pressed is often dried directly with combustion gases. As a result of these findings, Biernoth and Rost (4) developed a special clean-up procedure to remove PAH from crude vegetable oils during the refining process. The clean-up is accomplished by filtration through activated charcoal and is carried out in connection with the bleaching of the oil. Many oil refineries, including those in Sweden, have adopted this clean-up procedure and use charcoal regularly in the refining of coconut oils.

A pilot study on PAH in edible oils and margarines was conducted in our laboratory in 1982-83 (15). High PAH levels, up to 11.7 μg/kg of BaP, were found in some retail samples of coconut oil. Soybean oils contained varying amounts, up to 3.4 μg BaP/kg. The average level of BaP in 13 samples of margarine was 0.6 μg/kg. Wide variations in PAH levels in different samples of the same retail oil or margarine product indicated that some batches of crude oils were more contaminated than others, or that the refining process in the oil industry removed PAH with varying efficiency.

Following discussions with representatives of the Swedish edible oil industry, it was decided to investi-

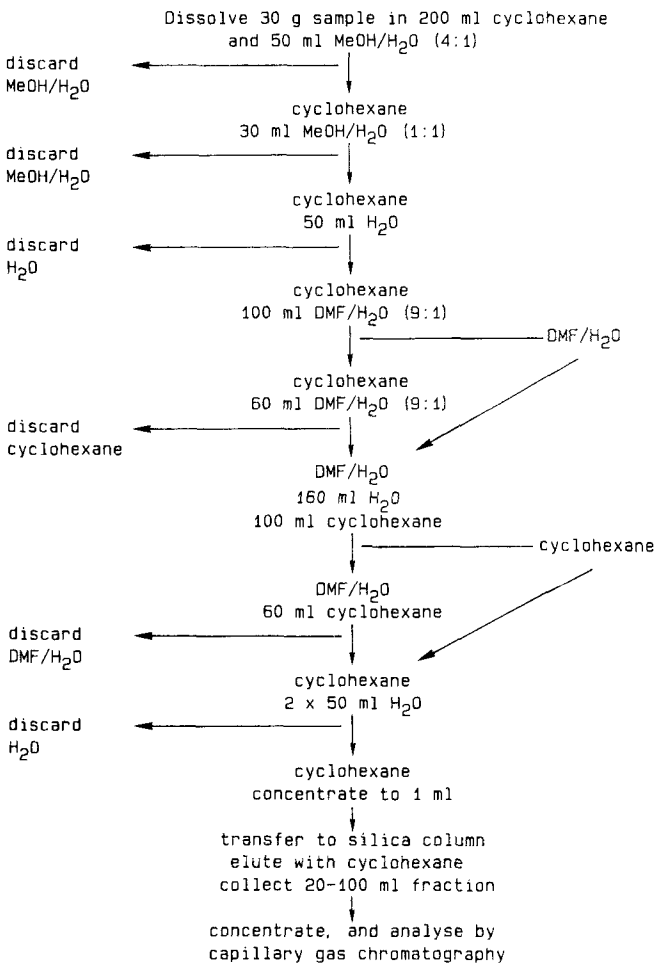

FIG. 1. Work-up scheme for analysis of PAH in oil samples.

*To whom correspondence should be addressed.

gate the efficiency of the refining process in removing PAH from crude oils. Therefore, samples of the crude oils and the corresponding refined, deodorized oils were taken from the processing line in three production plants.

MATERIALS AND METHODS

Samples. Samples of crude and deodorized oils were obtained from three different margarine factories in Sweden during the period December 1983–January 1984. Twelve samples of coconut oil, seven samples of soybean oil and four samples of rapeseed oil were taken. The samples were stored at 4 C before analysis.

Analyses. All samples were analyzed in duplicate. After addition of an internal standard (β,β'binaphthyl), the sample (30 g) was dissolved in cyclohexane and extracted as shown schematically in Figure 1. The method is essentially that described by Grimmer and Böhnke (16). When analyzing samples with very high levels of PAH (i.e. crude coconut oil), a 5-g sample was used and the amounts of solvents were reduced by half. The subsequent clean-up on silica has been described previously (17). The purified sample solution was analyzed by capillary gas chromatography. Each sample was injected twice on two different gas chromatographic systems: PYE Unicam GCD with falling needle injection and HP 5880 with on-column injection, each fitted with a flame ionization detector and a 25 m SE-54, i.d. 0.32 mm fused silica capillary column. Conditions were as follows: PYE Unicam: carrier gas, hydrogen 15 psi; injector temperature, 300 C; detector temperature, 330 C; oven temperature 150 C, programmed at 4 C/min to 300 C. HP 5880: carrier gas, helium 13 psi; injector temperature, 80 C; detector temperature, 310 C; oven temperature 80 C initially for 2 min, programmed 20 C/min to 170 C, 2 C/min from 170 C to 200 C, 5 C/min from 200 C to 300 C and held at 300 C for 30 min. The peaks were identified by comparing the retention times with those of standards. The identified compounds were quantified by comparing the integrated peak areas with that of the internal standard. A typical capillary gas chromatogram of a PAH extract from a sample of crude coconut oil is shown in Figure 2.

The peak identities in one sample of each type of oil were further confirmed by combined gas chromatography-mass spectrometry (GC-MS). The following conditions were used: GC column: fused silica, 25 m × 0.32 mm i.d. coated with OV1701; temp program: 90 C

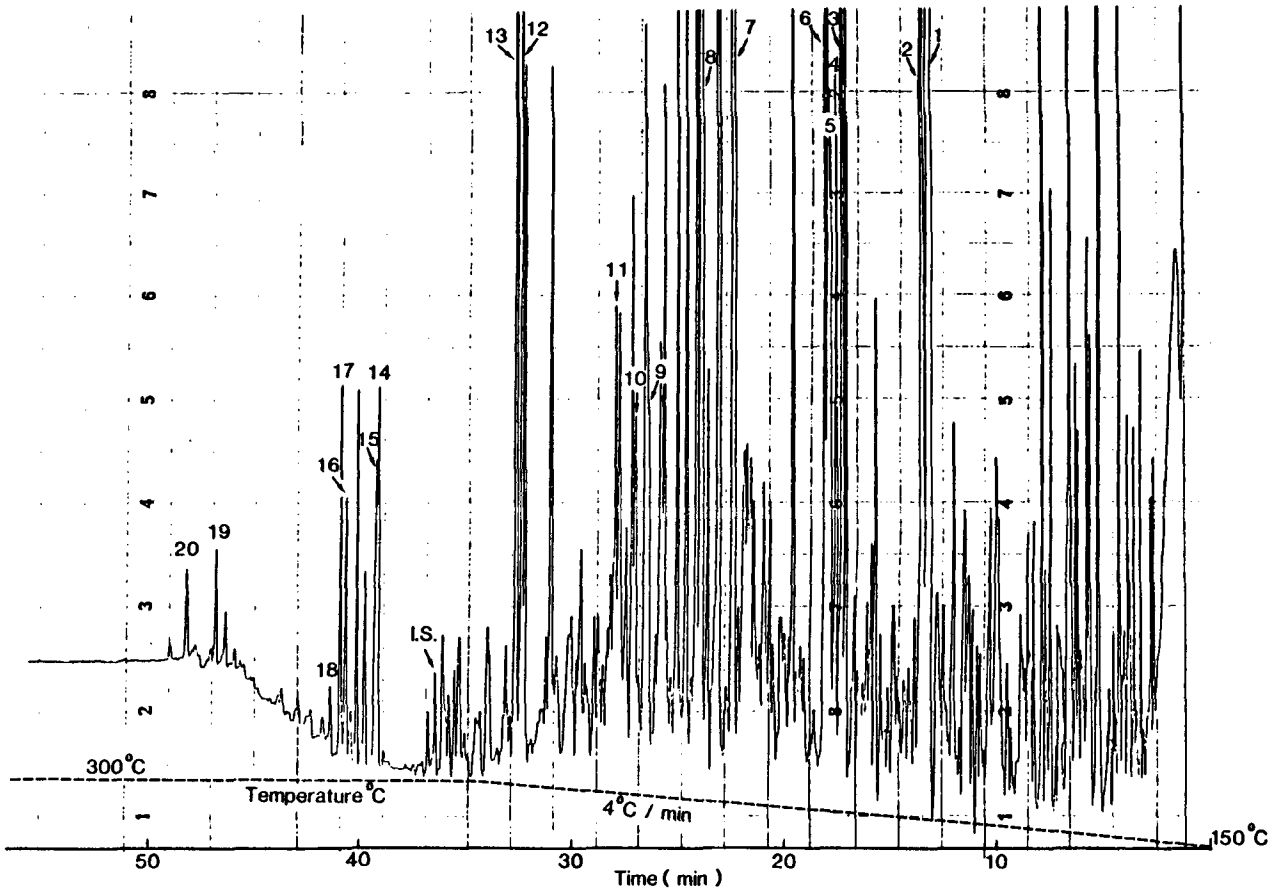

FIG. 2. Gas chromatogram of a PAH extract from a sample of crude coconut oil. 1, Phenanthrene; 2, anthracene; 3, 2-methylphenanthrene; 4, 2-methylanthracene; 5, 4,5-methylphenanthrene; 6, 1-methylphenanthrene; 7, fluoranthene; 8, pyrene; 9, benzo[a]fluorene; 10, benzo[b]fluorene; 11, 1-methylpyrene; 12, benz[a]anthracene; 13, chrysene and triphenylene; 14, benzo[b]-fluoranthene; 15, benzo[j]fluoranthene and benzo[k]fluoranthene; 16, benzo[e]pyrene; 17, benzo[a]pyrene; 18, perylene; 19, indeno-[1,2,3-cd]pyrene; 20, benzo[ghi]perylene, and (I.S.) β,β'-binaphthyl (internal standard).

TABLE 1

Recovery of PAHs in a Standard Mixture (NBS 1647) Added to Deodorized Oils[a]

	Deodorized coconut oil				Deodorized soybean oil				Deodorized rapeseed oil			
	Level I		Level II		Level I		Level II		Level I		Level II	
	Added µg/kg	Aver. rec. %	Added µg/kg	Aver. rec. %	Added µg/kg	Aver. rec. %	Added µg/kg	Aver. rec. %	Added µg/kg	Aver. rec. %	Added µg/kg	Aver. rec. %
Phenanthrene	1.2	58	3.2	82	1.1	65	3.4	72	1.2	68	3.3	61
Anthracene	0.8	47	2.2	59	0.7	82	2.4	57	0.8	58	2.3	41
Fluoranthene	2.4	89	6.3	100	2.3	98	6.6	93	2.3	96	6.4	90
Pyrene	2.4	87	6.2	100	2.2	96	6.5	94	2.2	94	6.3	92
Benzo[a]anthracene	1.2	94	3.2	106	1.1	100	3.4	106	1.2	129	3.3	111
Chrysene	1.1	101	3.0	116	1.1	122	3.2	119	1.1	103	3.0	125
Benzo[b]fluoranthene	1.2	94	3.2	116	1.1	164	3.4	136	1.2	132	3.3	115
Benzo[k]fluoranthene	1.2	99	3.9	93	1.1	147	4.1	96	1.2	143	4.0	96
Benzo[a]pyrene	1.3	86	3.8	109	1.2	159	4.0	109	1.2	120	3.9	117
Indeno[1,2,3-cd]-pyrene	0.9	144	3.8	95	0.8	132	4.0	100	0.9	143	3.9	108
Benzo[ghi]perylene	1.0	112	3.3	103	0.9	110	3.4	101	0.9	104	3.4	125

[a]Consideration is taken to the original levels of PAH in the samples before addition.
[b]Aver. rec., average recovery, n=2.

TABLE 2

PAH Levels in Crude and Refined Deodorized Coconut Oil

	Refinery A				Refinery B				Refinery C			
	Sample 1		Sample 2		Sample 1		Sample 2		Sample 1		Sample 2	
PAH µg/kg	Crude[a]	Deod.	Crude	Deod.	Crude	Deod.	Crude	Deod.	Crude	Deod.	Crude	Deod.
Phenanthrene		1.0	1300	0.5	1100	0.9	970	2.8	1000	1.1	850	0.5
Anthracene		0.4	280	0.2	230	0.2	200	0.3	210	0.1	180	<0.1
2-Methylphenanthrene		0.5	240	<0.1	130	0.8	140	1.7	110	0.7	110	0.1
2-Methylanthracene		0.2	120	<0.1	94	0.3	60	0.6	73	0.1	54	<0.1
4,5-Methylphenanthrene		0.2	110	<0.1	75	0.7	59	1.5	66	in	in	<0.1
1-Methylphenanthrene		0.5	130	<0.1	130	0.9	120	2.5	120	0.5	100	0.3
Fluoranthene		4.5	590	0.3	490	8.3	520	18	460	4.4	490	3.5
Pyrene		5.6	500	0.4	400	9.9	440	20	380	6.2	390	4.5
Benzo[a]fluorene		in[b]	in	<0.1	65	in	in	in	95	in	in	0.3
Benzo[b]fluorene		0.5	40	<0.1	38	4.0	39	1.9	32	in	29	in
1-Methylpyrene		1.1	39	<0.1	43	3.1	38	3.6	34	in	26	1.3
Benzo[a]anthracene		1.4	86	0.1	74	0.9	76	1.3	62	0.7	72	0.5
Chrysene/triphenylene		3.2	130	0.4	110	2.7	120	4.1	100	2.6	110	1.9
Benzo[b]fluoranthene		1.5[c]	74[c]	0.3[c]	45[c]	0.4[c]	55[c]	0.7[c]	47[c]	in	39	0.5
Benzo[j,k]fluoranthenes		—	—	—	—	—	—	—	—	in	36	in
Benzo[e]pyrene		0.2	25	<0.1	22	0.4	20	0.4	22	0.3	29	in
Benzo[a]pyrene		0.3	33	<0.1	23	0.2	22	0.2	20	in	34	in
Perylene		<0.1	7.3	<0.1	7.2	<0.1	6.3	<0.1	5.6	in	7.1	in
Indeno[1,2,3-cd]-pyrene		0.4	14	<0.1	8.1	0.2	9.8	<0.1	8.7	0.4	19	0.5
Benzo[ghi]perylene		0.2	17	<0.1	8.7	<0.1	9.6	<0.1	7.1	in	16	0.4
Total PAH		22	3700	2	3100	34	2900	59	2900	17	2600	16

The identification is based on GC retention times.
[a]Not analyzed. Sample broken during transport.
[b]in, interference from co-eluting compounds; quantitation not possible.
[c]Benzo[b]fluoranthene and benzo[j,k]fluoranthenes.

TABLE 3

PAH Levels in Crude and Refined Deodorized Soybean Oil

	Refinery A						
	Sample 1		Sample 2		Refinery B		Refinery C
PAH, μg/kg	Crude	Deod.	Crude	Deod.	Crude	Deod.	Deod.
Phenanthrene	9.8	0.6	17	0.5	18	0.4	1.1
Anthracene	0.8	<0.1	1.4	<0.1	1.4	0.2	<0.1
2-Methylphenanthrene	2.5	0.2	4.7	<0.1	3.4	0.2	<0.1
2-Methylanthracene	<0.1	<0.1	0.5	<0.1	0.4	<0.1	<0.1
4,5-Methylphenanthrene	0.6	<0.1	1.6	<0.1	1.1	<0.1	<0.1
1-Methylphenanthrene	1.2	0.1	3.3	<0.1	3.0	<0.1	0.1
Fluoranthene	5.7	0.4	5.9	0.3	7.1	0.6	1.9
Pyrene	3.9	0.5	4.6	0.4	5.8	0.7	2.0
Benzo[a]fluorene	in[a]	<0.1	in	in	in	in	in
Benzo[b]fluorene	0.8	<0.1	0.2	0.3	in	0.2	in
1-Methylpyrene	in	<0.1	0.5	<0.1	0.5	0.2	0.3
Benzo[a]anthracene	1.0	0.4	1.2	0.3	1.4	0.7	1.2
Chrysene/triphenylene	1.7	0.7	2.7	1.0	3.0	1.7	2.5
Benzo[b]fluoranthene	2.4[b]	0.2	3.1[b]	1.4[b]	5.2[b]	2.0[b]	2.9[b]
Benzo[j,k]fluoranthenes	—	0.5	—	—	—	—	—
Benzo[e]pyrene	in	in	in	1.5	1.4	1.7	1.5
Benzo[a]pyrene	0.4	0.3	in	0.6	1.2	1.1	0.8
Perylene	<0.1	<0.1	in	<0.1	in	in	0.5
Indeno[1,2,3-cd]pyrene	0.9	0.7	0.7	0.6	2.1	2.1	0.8
Benzo[ghi]perylene	0.9	0.7	0.8	0.9	1.4	1.0	1.0
Total PAH	33	5	48	8	56	13	17

The identification is based on GC retention times.

[a]in, interference from co-eluting compounds; quantitation not possible.

[b]Benzo[b]fluoranthene and benzo[j,k]fluoranthenes.

for 1 min, 30 C/min to 180, 6 C/min to 270; carrier gas: helium; mass spectrometer: LKB 2091 GC-MS; electron impact 70 eV; ion source temp: 250 C. To improve sensitivity a narrow mass range was selected for scanning, m/z 160-280.

The accuracy of the method was checked by a series of recovery tests. Deodorized oils (30 g) were spiked with aliquots of a reference solution, NBS 1647 (18) with certified amounts of a number of PAHs. The spiking was done at two different levels, 0.7-2.5 μg and 2.2-6.7 μg of the different PAHs per kg oil.

The results of the recovery experiments are shown in Table 1. The recovery shows the same pattern in all oils, a relatively low recovery for the low molecular PAHs phenanthrene and anthracene, about 100% recovery for fluoranthene, pyrene and benz[a]anthracene, and a recovery sometimes well over 100% for the high molecular components chrysene to benzo[ghi]perylene. Direct gas chromatographic analysis of the spiking solution gave a peak area distribution in agreement with the certified levels. Thus, the results of the recovery experiments indicate a certain discrimination in the extraction in such a way that high molecular PAH are extracted more effectively than the internal standard, β,β′-binaphthyl, which, in turn, is more effectively extracted than the low molecular components. The large deviations from 100% recovery appeared mainly in the low level region (level 1) where possible errors in the determination of the original PAH levels of the unspiked oil have more significance on the calculated recoveries.

A slight discrimination in the extraction of PAH can be expected in view of the partition coefficients reported for PAHs in different solvent partition systems (19). It is possible that the presence of large amounts of fat in the extraction influences the distributions in such a way that a marked discrimination takes place. However, the obtained recoveries must be regarded as satisfactory when the low PAH levels and the complexity of the matrices in these types of samples are considered.

RESULTS AND DISCUSSION

The results of the PAH analysis of crude and deodorized oils are listed in Tables 2-4. The amounts of BaP and total PAH in crude coconut oil ranged from 20 to 34 μg/kg, and from 2600 to 3700 μg/kg, respectively, whereas the levels in the deodorized oils were <0.1-0.3 μg/kg and 2-59 μg/kg.

Crude soybean oil contained 0.4-1.2 μg BaP and 33-56 μg of total PAH per kg. The levels in the corresponding deodorized oils were 0.3-1.1 μg/kg and 5-17 μg/kg.

The levels of BaP and total PAH in crude rapeseed oil were 0.6-2.1 μg/kg and 43-61 μg/kg, respectively. The

deodorized rapeseed oils contained 0.3-1.3 μg BaP and 5-12 μg of total PAH per kg.

Of the crude oils investigated, the coconut oils had by far the highest levels of PAH. This suggests that these oils have been pressed from copra which has been dried directly by combustion gases. However, in the corresponding refined products, the PAH levels were very low. This shows that the clean-up procedure with activated charcoal is effective. The high PAH levels found earlier in individual samples of refined coconut oil probably were due to the fact that in some plants the charcoal was added manually and therefore could sometimes have been omitted inadvertently (hypothesis suggested by the industry). However, in the last two years the addition of charcoal in the purifying process has been carried out automatically and registered on a recorder chart.

The crude soybean oils contained relatively low, but varying, amounts of PAH. The origin of the contamination of these oils has not yet been established. Soybean oils are not treated with activated charcoal in the Swedish margarine industry. As can be seen in Table 3, the PAH levels in the refined oils are still considerably lower than those in the corresponding crude oils. This probably is due to evaporation of PAH in the deodorization process, where steam is passed through the hot oil under high vacuum (4). However, the deodorization process seems to have little effect on high molecular PAHs. BaP was found in the deodorized oils at levels up to 1 μg/kg.

The rapeseed oils also contained PAH. The rapeseed used is grown in Sweden and, according to the industry, the seeds are dried almost exclusively with indirect heating methods. Possibly, the presence of PAH is due to accumulation of environmental PAH during the growing period, or by contamination from the solvent (hexane) used in the extraction process. Rapeseed oil is not purified by activated charcoal, and the samples of deodorized oil still contained PAH, particularly in the high molecular fraction.

The levels of PAH in coconut, soybean and rapeseed oils found in this study are similar to those reported previously by other investigators. In a study on PAH in various crude oils, Grimmer and Hildebrandt (5) found BaP levels of 17.9-48.4 μg/kg for coconut oil, 1.3-4.0 μg/kg for rapeseed oil and 1.5-1.9 μg/kg for soybean oil. Howard (1) detected BaP in two of nine samples of retail soybean oil, at levels of 1.2-1.5 μg/kg. Lawrence and Weber (12) found 0.2 μg BaP/kg in a sample of commercial soybean oil. Much higher PAH levels in various commercial oils were reported by Kolarovic and Traitler (11), e.g. 28.45 and 2.14 μg BaP/kg, respectively, in samples of soybean and rapeseed oils.

This is, to our knowledge, the first systematic study concerning the reduction of PAH during the refining of edible oils in modern refineries. The use of high resolution capillary gas chromatography enables the determination of a large number of PAHs, which facilitates a detailed characterization of the PAH fraction in the crude and deodorized oil. The selective reduction of low molecular PAH during deodorization, observed in this study, was also reported by Biernoth and Rost (3,4), who used a thin layer chromatographic method (20) to determine 13 PAHs in vegetable oils sampled at various stages of the refinery process. From their fundamental studies (3,4) in the late sixties, they concluded that a combination of charcoal treatment, which they found particularly effective for high molecular PAH, and deodorization is an ideal method for producing PAH-free oils and fats.

TABLE 4

PAH Levels in Crude and Refined Deodorized Rapeseed Oil

	Refinery A			
	Sample 1		Sample 2	
PAH μg/kg	Crude	Deod.	Crude	Deod.
Phenanthrene	16	0.4	11	0.3
Anthracene	0.5	0.1	0.7	<0.1
2-Methylphenanthrene	6.3	<0.1	1.6	<0.1
2-Methylanthracene	0.2	<0.1	<0.1	<0.1
4,5-Methylphenanthrene	in[a]	<0.1	0.5	<0.1
1-Methylphenanthrene	4.8	<0.1	1.0	<0.1
Fluoranthene	9.4	0.3	5.5	0.3
Pyrene	10	0.3	4.1	0.4
Benzo[a]fluorene	in	<0.1	0.5	<0.1
Benzo[b]fluorene	1.6	<0.1	0.4	<0.1
1-Methylpyrene	2.8	<0.1	0.2	<0.1
Benzo[a]anthracene	1.0	0.2	1.7	0.6
Chrysene/triphenylene	5.6	0.7	3.0	1.3
Benzo[b]fluoranthene	1.1[b]	0.7[b]	3.8[b]	2.8[b]
Benzo[j,k]fluoranthenes	—	—	—	—
Benzo[e]pyrene	in	0.3	2.2	1.1
Benzo[a]pyrene	0.6	0.3	2.1	1.3
Perylene	in	<0.1	0.7	0.4
Indeno[1,2,3-cd]pyrene	0.9	0.3	1.7	1.4
Benzo[ghi]perylene	in	0.6	1.5	1.2
Total PAH	61	5	43	12

The identification is based on GC retention times.
[a]in, interference from co-eluting compounds; quantitation not possible.
[b]Benzo[b]fluoranthene and benzo[j,k]fluoranthenes.

Coconut oil, soybean oil and rapeseed oil are important components of Swedish margarines. The problem with the occasional high PAH levels in refined coconut oils has, hopefully, been solved. The measures taken recently by the industry to control the addition of charcoal seem to be satisfactory. During 1984, 11 samples of Swedish margarines were analyzed and the results compared to those in the pilot study carried out in 1982-83. The average levels of BaP and total PAH were 0.4 μg/kg and 10 μg/kg, respectively, in 1984, as compared to 0.6 μg/kg of BaP and 20 μg/kg of total PAH in 1982-83. This decrease probably is due to the fact that the activated charcoal treatment has been automated and thus no refined oils with higher levels are used in the manufacturing of margarines.

The levels of PAH in crude soybean and rapeseed oil seem to vary considerably from batch to batch. As these oils are not treated with activated charcoal, some batches of refined oils do contain relatively high levels of high molecular PAH. Most of the PAHs which are classified as carcinogens are found in the high molecular fraction. According to the industry, the

introduction of charcoal treatment for soybean and rapeseed oils would be expensive because the volumes are so large. It is desirable that the industry can trace heavily contaminated batches of crude oils, so that these batches can be treated separately. However, a suitable method for the simple and rapid tracing of contaminated oils is not available at present. It is important that such a method be developed and also that the sources of PAH in soybean and rapeseed oils be further explored.

ACKNOWLEDGMENTS

This work was partly supported financially by Margarinbolaget AB and AB Karlshamns Oljefabriker. H. Grahn, Margarinbolaget, and G. Johansson, Karlshamns Oljefabriker, supplied the samples and took part in valuable discussions.

REFERENCES

1. Howard, J.W., E.W. Turicchi, R.H. White and T. Fazio, *J. Assoc. Off. Anal. Chem. 49*:1236 (1966).
2. Grimmer, G., and A. Hildebrand, *Chem. and Ind. 1967*:2000 (1967).
3. Biernoth, G., and H.E. Rost, *Ibid. 1967*:2002 (1967).
4. Biernoth, G., and H.E. Rost, *Arch. Hyg. 152*:238 (1968).
5. Grimmer, G., and A. Hildebrandt, *Ibid. 152*:255 (1968).
6. Fabian, B., *Ibid. 152*:231 (1968).
7. Franzke, C., and W. Fritz, *Fette, Seifen, Anstrichm. 71*:23 (1969).
8. Siegfried, R., *Naturwissenschaft 62*:576 (1975).
9. Swallow, W., *N. Z. J. Sci. 19*:407 (1976).
10. Heddeghem, A., A. Hyghebaert and H. De Moor, Z. *Lebensm. Unters. Forsch. 171*:9 (1980).
11. Kolarovic, L., and Traitler, H., *J. Chromatogr. 237*:263 (1982).
12. Lawrence, J.F., and D.F. Weber, *J. Agric. Food Chem. 32*:794 (1984).
13. Tilgner, D.J., *Food Manuf. 45*:47 (1970).
14. Dunn, B.P., *Carcinogens and Mutagens in the Environment 1*:175 (1982).
15. Larsson, B.K., National Food Administration, internal report (1983).
16. Grimmer, G., and H. Böhnke, *J. Assoc. Off. Anal. Chem. 58*:725 (1975).
17. Larsson, B.K., *Z. Lebensm. Unters. Forsch. 174*:101 (1982).
18. National Bureau of Standards, U.S. Department of Commerce.
19. Jacob, J., and G. Grimmer, *Environmental Carcinogens. Selected Methods of Analysis, Vol. 3*, edited by M. Castegnaro, P. Bogowski, H. Kunte, and E.A. Walker, IARC Scient. Publ. no. 29, International Agency for Research on Cancer, Lyon, 1979, pp. 85-86.
20. Biernoth, G., *J. Chromatogr. 36*:325 (1968).

[Received March 31, 1986]