Polynuclear aromatic hydrocarbons in South Indian diet

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Several commonly consumed food items of South India were screened for the presence of polynuclear aromatic hydrocarbons (PAHs). Samples were extracted with dichloromethane, cyclohexane and acetonitrile and PAHs were identified. Chrysene was detected in almost all the samples. Salted, sundried and oil-fried ribbon fish, and *kashayam* prepared from cumin seeds, mint leaves and cardamom, contained chrysene in high concentration. Benzo[*a*]pyrene was maximum in ribbon fish, while dibenzanthracene was quantified in high concentrations in the *kashayam* and oil-fried chillies.

POLYNUCLEAR aromatic hydrocarbons (PAHs) are a group of highly lipophilic chemicals that are present ubiquitously in the urban environment as pollutants in very small quantities. In general, PAHs are formed during combustion, pyrolysis and pyrosynthesis of organic matter[1]. Such thermal degradation products have been reported in trace concentrations in tobacco smoke, cigarette smoke, marijuana smoke, polluted air, water, industrial air, soil sediments, aquatic organisms, mineral oils and refined petroleum products[2-5]. PAHs have also been identified and quantified in several food items[6-9], which have been proved to be mutagenic and are related to the predominant form of alimentary tract cancers in Asian and Western countries[6]. Epidemiological studies have unequivocally established a relationship between the occurrence of PAHs and different types of cancer[1]. Since the incidence of stomach cancer is high in South India, we have screened several commonly consumed food items, such as deep fried *vathals*, salted and sundried fishes and pyrolysed portions of *uppuma* – a food product of rava, for mutagenicity, which proved positive[10]. We have detected the presence of some of the PAHs in these food items, quantified them and the results are presented here.

Five hundred mg of test sample (Table 1) was ground well in a pestle and mortar with 10 ml of water. This was extracted thrice in a separatory funnel, with 40 ml of dichloromethane. The extracts were pooled and 5 g of sodium sulphate was added to remove lipids and excess water. The clear supernatant was decanted and evaporated over a waterbath at 60°C to 5 ml. Forty ml of cyclohexane was added and again evaporated to a minimal volume. This was passed through a silica gel (activated at 110°C for 5 h) column of 1 × 5 cm size. The resulting solution was collected and 1 g of sodium sulphate was added. The supernatant was collected and evaporated at 60°C to dryness. The residue was dissolved in 0.5 ml of acetonitrile and used for HPLC analysis[11]. The extracted sample (10 ml) was injected into a Shimadzu LC 6A HPLC system (steel column, 150 × 4.6 mm, Zorbaks–ODS, column temp. 35°C; flow rate 1 ml/min, 254 nm, mobile phase: 100% acetonitrile) and PAHs were detected and quantified based on the chromatograms of the authentic samples/standards[10].

Chrysene was detected in almost all the samples (Table 1). Ribbon fish contained chrysene as high as 18.57 µg/g, while *kashayam III* had 12.61 µg/g. Pyrolysed portion of *uppuma* contained 2.65 µg/g of chrysene. In fried mustard, it was very low, 0.07 µg/g. Dibenzanthracene was also high in *kashayam III*. *Vathal* prepared from *Capsicum annum* had 27.48 µg/g of dibenzanthracene, whereas salted and sundried whitebait fish contained 12.34 µg/g. Benzo[*a*]pyrene was present in *kashayam III*, *vathal* of *Cyamopsis tetragonaloba* and in all the sundried fishes. Samples of ribbon fish revealed benzo[*a*]pyrene at a high concentration, 60.81 µg/g. All the three PAHs were detected and quantified in whitebait fish and *kashayam III* (Table 1). Oil used for frying (heated groundnut oil) did not reveal any peak which corresponded with that of authentic PAH standards.

Table 1. Polynuclear aromatic hydrocarbons (PAHs) in food substances.

Food item	PAHs	Concentration (µg/g)
Vathal of:		
Capsicum annum	Chrysene	1.995
	DBA	27.480
Cyamopsis tetragonaloba	Chrysene	0.965
	DBA	9.540
Solanum torvum	BP	0.060
Fried mustard	Chrysene	0.070
Salted and sundried fishes:		
Whitebait fish		
(Stolephores bataviensis)	Chrysene	4.280
	DBA	12.340
	BP	5.730
Ribbon fish		
(Trichurus leptures)	Chrysene	18.570
	BP	60.810
Seer fish		
(Scomberomorus commersonii)	Chrysene	3.570
	BP	10.290
Pyrolysed portion of *uppuma*	Chrysene	2.650
Kashayams (from pyrolysed):		
Cumin seeds + aniseeds	Chrysene	0.520
Cumin seeds + aniseeds + ginger	Chrysene	2.011
Cumin seeds + mint leaves + cardamon	Chrysene	12.610
	DBA	31.150
	BP	6.400

BP, Benzo[*a*]pyrene; DBA, 1,2,5,6 Dibenzanthracene

All the test compounds are widely consumed in South India. Dimethyl sulphoxide extracts of the vathals displayed appreciable mutagenic activity[10] and this can be attributed to the presence of PAHs, suggesting that they are formed during deep frying. Kosage *et al.*[12] identified a series of heterocyclic amines and aromatic hydrocarbons from pyrolysates and ordinary cooked foods. Heterocyclic amines are also isolated from broiled sundried sardines[13, 14]. Our results reveal that sundried and salted fishes contain the potent carcinogens, chrysene, benzo[*a*]pyrene and dibenzanthracene in appreciable concentrations. Incidentally, Ikeda *et al.*[15] reported that people who frequently consumed charred fish items exhibited high incidence of gastric cancer. Benzo[*a*]pyrene and chrysene induce papillomas and carcinomas while dibenzanthracene induces sister chromatid exchanges and chromosomal aberration in experimental animals[1]. Heterocyclic amines are carcinogenic in various organs and most of them are carcigenic in two or more organs[9]. There is often a high degree of correlation between *in vitro* mutagenicity and *in vivo* carcinogenicity[16, 17]. Most of the food items we have screened, are mutagenic as revealed through Ames assay[10] and the probable causative agents, PAHs are detected in this study. We suggest, therefore, that PAHs, present in alarming quantities, in the commonly and widely consumed food items, may play a major role in the high incidence of gastric cancers in South India.

1. IARC monographs, 1983, part 1, vol. 32, International Agency for Research on Cancer, Lyon, France.
2. IARC monographs, 1984, part II, vol. 33, International Agency for Research on Cancer, Lyon, France.
3. IARC monographs, 1984, part III, vol. 34, International Agency for Research on Cancer, Lyon, France.
4. IARC monographs, 1985, part IV, Vol. 35, International Agency for Research on Cancer, Lyon, France.
5. Yamashita, M. *et al.*, *Jpn. J. Cancer Res.*, 1986, 77, 419.
6. Sugimura, T., *Environ. Health Perspect.*, 1986a, **67**, 5.
7. Sugimura, T., *Science*, 1986b, **233**, 312.
8. Yano, M. *et al.*, *Mutat. Res.*, 1988, **201**, 181.
9. Zhang, X. M. *et al.*, *Mutat. Res.*, 1988, **201**, 181.
10. Balachandran, B., Sivaswamy, S. N. and Sivaramakrishnan, V. M., 1989, 10th Annual Session AEB, Loyola College, Madras.
11. Federal Register, Environmental Protection Agency, USA, 1984, **49**, 112.
12. Kosage, T. *et al.*, *Chem. Pharm. Bull.*, 1978, **26**, 611.
13. Kasai, H. *et al.*, *Proc. Jpn. Acad.*, 1980a, **56B**, 278.
14. Kasai, H. *et al.*, *Chem. Lett.*, 1980b, 1391.
15. Ikeda, M. *et al.*, *Jpn. J. Cancer Res.*, 1983, **74**, 640.
16. Huberman, F., *Mutat. Res.*, 1976, **39**, 285.
17. Lijinsky, W., *Environ. Mol. Mutagen.*, 1989, **14**, 78.

ACKNOWLEDGEMENTS. We thank the Director & Scientific Director and the Advisor, Cancer Institute for their keen interest and constant encouragement. Financial assistance from the Ministry of Environment and Forestry, New Delhi, is gratefully acknowledged.

12 January 1990