Pak. j. sci. ind. res., vol. 36, nos. 2-3, Feb.-Mar. 1993

SCREENING OF VARIOUS LOCAL RAW FOOD COMMODITIES FOR AFLATOXIN CONTAMINATION. *Part -I*

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(Received December 31, 1990, revised April 1, 1993)

Fourteen food commodities (wheat, rice, maize, oat, sugar, pulse, gram, green gram, black gram, lentil pepper, coriander, turmeric, gur and cumin) procured from various areas of Punjab, NWFP, Sindh, Baluchistan, and Azad Kashmir were screened for aflatoxins B_1 and B_2 contamination. The results showed an incidence of about 43% aflatoxin contamination at moderate levels in six raw foods. The highest aflatoxin B_1 concentration (88.3 µg/kg) was found in maize from Multan, followed by a level of 82.1 µg/kg/ B_1 in green gram from Peshawar. Maize from D.I. Khan showed the lowest B_1 concentration (11.1µg/kg). Pepper from Sargodha contained 41.7 µg/kg aflatoxin B_2 . The result obtained are discussed in terms of seasonal and regional variations together with international standards laid down for the safe ingestion of aflatoxin contaminated raw foods.

Key words: Aflatoxins in foods, Aflatoxin analysis.

Introduction

During recent years incidents of food poisoning resulting from contamination of food grains with toxogenic fungi have been reported [1,2]. The presence of aflatoxins B_1 and B_2 at biologically significant levels in agricultural food commodities such as barley, bean, corn, cotton seed, rice and wheat, and in vegetable, spices, fruits and consumer products prepared from theses commodities is well known [3,4]. The correlation between aflatoxin contamination of the diet and occurrence of human liver cancer has convinced public health scientists to evolve measures to lessen human exposure to these toxins [5]. Prophage induction, filamentation and related effect caused by aflatoxin B_1 are established [6,7]. The B_1 toxicology for childeren is well known [8,9]. The aflatoxin induced DNA damage has also been reported [10]. Thus, there was a dire need to initiate a study on the evaluation of the aflatoxin content of local raw food commodities so that probable incidences of contamination could be assessed for evolving a future monitoring programe aimed at establishing a strict food quality control.

Materials and Methods

The raw food samples were collected from Azad Kashmir, and from the provinces of the Punjab, NWFP,Sindh, and Baluchistan during the production period of 1984 through 1986. The samples were procured from various districts of respective provinces where the relevant food was grown. Equal amount of each commodity (usually 1.0 kg) was mixed to produce a single representative sample at divisional level. The composed samples were cleaned to remove any foreign matter and gound in a Christy and Norris hammer mill into a coarse powder of uniform particle size (100 mesh). An accurately weighed portion of the sample (usually 10 g) was isolated for the detemination of the moisture and ash contents.The remainder of each sample was packed and sealed in thick polythene bags and deep frozen at -20° until analysed.

The mini-column method was used to determine the presence of aflatoxins in the samples. Reagent blanks were run to eliminate the chances of false fluorescence and to establish the purity of reagents and chemicals used. The details of the procedure are described elsewhere [11]. The samples showing positive test were quantified by the CB methods, using the chloroform extract for the measurement of aflatoxin by thin-layer chromatography using silica-gel G on coated glass plates, 0.5 mm thickness [12]. The spotting was done for the unknown samples superimposed with mixed standards of aflatoxin B_1 and B_2. The plates were developed in chloroform-methanol mixture and were radiated at 366 nm to identify the type of aflatoxin present, counterchecked by R_f values determined isothermally at 25.5° on triplicate subsamples.

Results and Discussion

The information on the total composite samples of food stuffs analysed is given in Table 1. The data on aflatoxin and moisture content of various contaminated food commodities are given in Table 2. The reported concentrations appear as averaged over triplicate runs, with precision between 1.5 - 2.0%. Of the 108 composed samples of cereals and grains, spices, condiments and sweetening agents analysed, 15 were found to be aflatoxin B_1 and/or B_2 contaminated. In the contaminated foods the incidence of aflatoxin $B_1$was about 64%, compared with 30% of that of B_1 B_2 combined. Wheat, rice, maize, green gram, lentil and pepper from various areas

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TABLE 1. DESCRIPTION OF FOOD COMMODITIES AND NUMBER OF COMPOSITE SAMPLES ANALYSED FOR AFLATOXIN CONTAMINATION.

S.No.	Food commodity	Composites analysed
1.	Cereals	
	Wheat (whole)	15
	Rice (raw)	12
	Maize (dry)	12
	Oat	2
2.	Pulses	
	Bengal Gram	10
	Green Gram	12
	Lentil	13
	Black Gram	10
3.	Condiments and spices	
	Pepper	6
	Turmeric	2
	Coriander	5
	Cumin	1
4.	Sweetners	
	Sugar	4
	Gur	4
		Total = 108

TABLE 2. DATA ON MOISTURE AND AFLATOXIN CONCENTRATIONS IN VARIOUS CONTAMINATED FOOD COMMODITIES.

S. No.	Food	Origin	Aflatoxin Type	Aflatoxin Concentration (µg/kg)	Moisture (%)
1.	Wheat(whole)	D.I.Khan	B_1 -	46.2	8.8
		Peshawar	B_1 -	48.2	9.8
2.	Rice (raw)	Lahore	B_1 -	52.7	10.8
3.	Maize (dry)	Multan	B_1 -	88.3	10.0
		Peshawar	B_1 -	66.6	9.9
		D.I.Khan	B_1 -	11.1	10.8
		Bahawalpur	B_1 -	27.8	11.3
		Sukhur	B_1 B_2	46.8	12.4
4.	Green Gram	Multan	B_1 B_2	51.2	18.4
		Peshawar	B_1 -	82.1	11.2
5.	Lentil	Sargodha	B_1 B_2	68.2	9.6
		Karachi	B_1 B_2	53.4	11.2
6.	Pepper (dry)	Lahore	B_1 -	29.5	15.8
		Sargodha	B_1 -	38.6	17.9
		Sargodha	- B_2	41.7	12.6

were found to be aflatoxin contaminated. Pepper from Sargodha was contaminated with aflatoxin B_2 only. In case of maize 4 samples out of 12 were found to be aflatoxin B_1 contaminated, giving an incidence of 33%. The levels of aflatoxins in various foods varied between 11.1 - 88.3 µg/kg, while the moisture content ranged between 2.5 - 12.6%. In red pepper the level of aflatoxin B_2 at 41.7 µg/kg was associated with a moisture content of 12.6%. On the whole, 6 food commodities out of 14 were found to be aflatoxin contaminated.

The aflatoxin contamination found in maize samples from various divisions of Multan, Bahawalpur, Sukkur, Peshawar and Dera Ismail Khan of the provinces of Punjab, Sindh and NWFP indicated a wide-spread contamination problem. The highest level of aflatoxin B_1 (88.3µg/kg) was found in maize procured from Multan (Punjab), while the lowest in the sample (11.1µg/kg) was encountered in the sample from D.I. Khan (NWFP). Comparable aflatoxin contaminations in maize were previously reported in an international survey[13]. However, the aflatoxin contamination in Pakistani maize has been reported at a higher level between 133-800 µg/kg by PCSIR Laboratories [14].

Moisture is considered to be the most important factor in the growth of *Aspergillus flavus* resulting in aflatoxin production. As the contaminated maize samples contained moisture content within limits laid down in food laws, it could be inferred that moisture was not responsible for the observed contamination. The samples might have been contaminated in the field before harvesting or, at a later stage, during storage under bad condition. It was most likely that the food commodity was invaded by the fungus and even though the generating fungal organisms had disappeared, the preformed toxin was still present to course the observed contamination. This could be a possible reason for the occurrence of aflatoxin in maize and other food samples.

The US and Canadian standards prescribe 30 and 20 µg/kg respectively as the upper acceptable limit of aflatoxin contamination, while Japan has prescribed a much higher limit of 1 mg/kg. The international agencies such as WHO, FAO and UNICEF set the tolerable aflatoxin limit at 30 µg/kg. The present study revealed that two samples of contaminated maize and one sample of pepper were within these allowed safe limits, while all other contaminated foods exceeded the prescribed safe limits.

In conclusion, the present data evidenced a potential aflatoxin contamination problem in local raw food commodities. A part from seasonal and regional variations responsible for the spread of aflatoxins in these foods, it is evident that good agricultural practices such as harvesting, fumigation, etc., are not being exercised by the cultivators, especially for the major wheat and rice crops and for the staple crops produced and consumed in large bulk in the country. Although only maize has emerged as a high risk crop, yet extensive surveillance programe of aflatoxin screening should be initiated for other foods as well. Regulatory programmes to control

aflatoxin contamination in various food species from different agricultural areas are, therefore, imperative.

Acknowledgements. We are grateful to FAO for technical and financial assistance, under the project NOR/078/Pak, that covered a major part of the present study. Collabortion by the Department of Chemistry, Quaid-i-Azam University is gratefully acknowledged.

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