SHORT REPORT

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DETERMINATION OF AFLATOXIN B_1 IN GROUNDNUT EXTRACTS AND ITS MUTAGENICITY

PRASONG KOONANUWATCHAIDET

Department of Biochemistry, Faculty of Medicine, Khon Kaen University, Khon Kaen, Thailand

LOUISE Y.Y. FONG

Department of Biochemistry, Faculty of Medicine, University of Hong Kong, Hong Kong

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Summary

Partially purified aflatoxin B_1(AFB_1) from ten batches of groundnut extracts in Hong Kong was quantitatively determined. Its mutagenesis was also assayed by S. typhimurium microsomal system. There is a good correlation between the presence of AFB_1 and its mutagenic activity in those extracts.

Aflatoxins (AF) constitute a group of toxins mainly produced by *Aspergillus flavus* which contaminate foods and feeds[1,2]. These compounds have been shown to possess potent hepatocarcinogenic activities in several animal species[3,4]. Four types of aflatoxins have been found, namely AFB_1, AFB_2, AFG_1 and AFG_2. Among these, AFB_1 is the most powerful toxin, fellowed by AFG_1, AFB_2 and AFG_2 in decreasing potency. Recently, a few rapid *in vitro* assay systems for screening environmental carcinogens as mutagens have been developed. The test of Ames *et al.*[5,6], using *Salmonella typhimurium* and mammalian microsomal enzyme has been widely introduced for detection of those mutagens, including AFB_1. This paper reports a low but definite presence of AFB_1 in commercial groundnuts in Hong Kong.

Ten samples of groundnuts were purchased from various markets in Hong Kong. One hundred grams of each batch was used for solvent extraction, and the extract was then passed through silica gel column[7]. Definite estimation was obtained

by thin-layer chromatographic and spectrophotometric techniques[8]. One gram of each sample was separately extracted with two millilitres of dimethyl-sulfoxide(DMSO) for mutagenesis assay[5,6]. Two bacterial tester strains, TA 98 and TA 100 were employed throughout experiments.

TABLE 1 THE QUANTITY OF AFB_1 CONTAMINATING GROUNDNUTS AND MAXIMAL MUTAGENICITY.

Sample	Quantity of AFB_1 (ppm)	Maximal revertant colonies/plate of DMSO extract (+ S–9 MIX)	
		TA 100	TA 98
1	0.875	955	1,643
2	0.615	427	974
3	0.724	579	1,144
4	0.638	682	995
5	0.797	585	908
6	0.559	535	759
7	0.729	690	936
8	1.234	1,088	2,187
9	1.344	1,591	2,064
10	1.550	1,935	2,534

The presence for AFB_1 in our extracts is quite low, ranging between 0.56 and 1.55 ppm. (Table 1). The determination is subject to some minor errors, eg. destruction of this toxin during extraction, elution, evaporation, and also quenching effect of spectrophotometric measurement due to some impurities. The general correlation observed, however, appears to be valid. Increasing mutagenicity is shown for both frameshift mutation (TA 98) and base-pair substitution (TA 100), with increase in the quantity of AFB_1. Results on mutagenic action of Sample Number 5 is illustrated in Fig. 1.

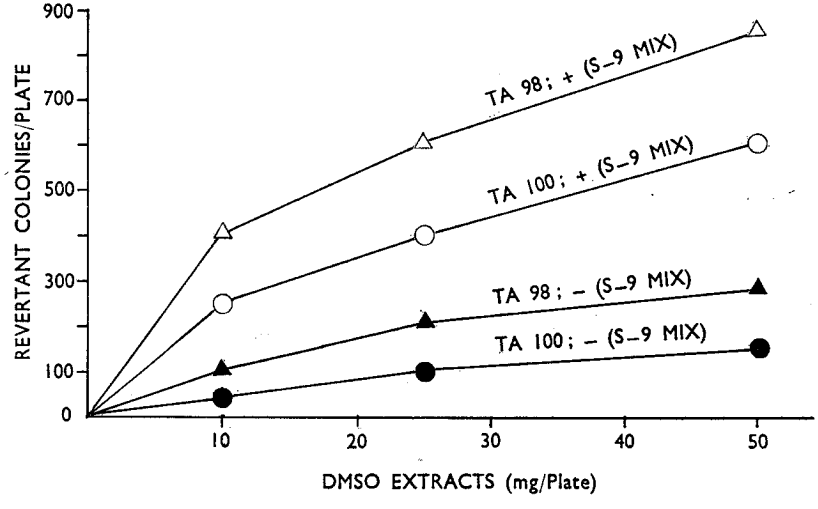

Fig. 1 The mutagenic dose-response curves of DMSO extract of Sample Number 5. The quantity of DMSO extracts refers to the original weight of the groundnuts.

The mutagenicity was enhanced in the case of TA 98 with rat liver microsomal activation system (S–9 MIX). It was evident that the extracts tended to cause mutation and microsomal enzymes were also needed (Fig. 1). Our results supported the previous reports of Ames *et al.*[6,9] and Gurtoo *et al.*[10] respectively. Further studies are needed, however, to identify clearly that the mutagenicity observed is due to AFB_1 alone and not also to other impurities present in the extracts.

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บทคัดย่อ

ได้มีการสะกัดเอาสารพิษเอฟล่าท็อกซิน B_1 จากถั่วลิสงในฮ่องกง 10 ตัวอย่าง แล้วนำไปหาปริมาณและวัดคุณสมบัติในการกลายพันธุ์โดยใช้บักเตรี *S. typhimurium* ร่วมกับเอ็นซัยม์ของไมโครโซม พบว่ามีความสัมพันธ์ดีพอสมควรระหว่างปริมาณและคุณสมบัติดังกล่าวของสารพิษนี้