Quantitative Estimation of Aflatoxins, Orchratoxins and Citrinin in Dried Fruits and Nuts Samples from Indo-Gangetic Region of Bihar

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Abstract

The point of this investigation was directed to evaluate the quantitative assessment of aflatoxins, orchratoxins and citrinin in dried fruits and nuts samples from indo-gangetic region of Bihar. Aflatoxins, ochratoxin An and citrinin were normally identified mycotoxins from almond, pistachio, cashew nut and raisins samples while figs were polluted with just aflatoxins. A sum of 154 dried fruits and nut samples (32 every one of almond and pistachio and 30 samples of every cashew nut, raisins and figs) were gathered from indo-gangetic region of Bihar. The consequences of present examination propose that the almond, pistachio, cashew nut and raisins are powerless substrate for parasitic development and further mycotoxin creation. The amount of aflatoxin distinguished was adequately high to incite carcinogenesis.

Keyword: *Aflatoxins, Ochratoxin, Citrinin, Dried fruits, Nuts*

1. INTRODUCTION

Ganga is the biggest and holy of India and most populace lives in Gangetic plain region. 44900 square kilometer territory of Bihar goes under Indo-Gangetic plain where about 103.8 million populaces dwell. Mycotoxins are the toxic optional metabolites of parasites by and large created on wide scope of consumable substances under different conditions.

Aflatoxins are a class of toxic metabolites produced by specific organisms that are usually found in the climate. Aspergillus flavus and Aspergillus parasiticus are the predominant contagious species that produce aflatoxins under dry spell, warm and damp conditions. Ochratoxin A (OTA) is a significant mycotoxin. OTA is a nephrotoxic, hepatotoxic, embryotoxic, teratogenic, neurotoxic, immunotoxic, genotoxic and cancer-causing mycotoxin. Citrinin (CIT), frequently found in a similar food as OTA, is a ground-breaking nephrotoxin. In recurrent portion toxicity examines, the kidney was distinguished as the chief objective organ for CIT, and huge species contrasts in the powerlessness to CIT have been noticed.

2. LITERATURE REVIEW

Sofia Agriopoulou et. al. (2020) Mycotoxins are toxic substances that can contaminate numerous nourishments with cancer-causing, genotoxic, teratogenic, nephrotoxic, and hepatotoxic impacts. Avoidance, detoxification, and sterilization of mycotoxins can contribute in this reason in the pre-collect and post-gather stages. . In this manner, the reason for the audit is to expand on the ongoing advances with respect to the event of primary mycotoxins in numerous sorts of significant rural items, just as the strategies for inactivation and detoxification of nourishments from mycotoxins to lessen or completely dispose of them.

Kai Zhang and Kaushik Banerjee (2020) Aflatoxins are optional metabolites of different Aspergillus species, which are universal in the climate and can develop on an assortment of yields whereby collection is affected by atmosphere impacts. The scientific procedures, for example, gas chromatography (GC), fluid chromatography (LC), mass spectrometry (MS), fine electrophoresis (CE) and slender layer chromatography (TLC) are thought about as far as recognizable proof, quantitation and throughput. In conclusion, with the rise of new procedures, the audit finishes with possibilities of promising advances for aflatoxin examination soon.

Luigi Castaldo (2019) A complete methodology joining a quantitative strategy for 28 mycotoxins and a post-target screening for other 245 contagious and bacterial metabolites in dry pet food samples were created utilizing an acetonitrile-based extraction and a ultrahigh-execution fluid chromatography coupled to high-goal mass spectrometry (UHPLC-Q-Orbitrap HRMS) technique. All sure samples demonstrated co-event of mycotoxins with the concurrent presence of up to 16 analytes per sample. In the review screening, up to 54 parasitic metabolites were likely distinguished being cyclopiazonic corrosive, paspalitrem A, fusaric corrosive, and macrosporin, the most regularly recognized analytes.

Shahzad Zafar Iqbal (2018) A sum of 320 samples of consumable nuts (poppy seed, peanut, pistachio, cashew, almonds) and dry fruits gathered from significant urban communities of Punjab, and Khyber Pakhtunkhwa, Pakistan, were investigated for the presence of aflatoxins (AFs) and ochratoxin A (OTA). The outcomes have uncovered the co-occurrence of OTA and absolute AFs in 25% all out samples of dry fruits and nuts with a mean degree of OTA 3.58 ± 1.30 µg/kg and complete AFs mean degree of 4.13 ± 0.48 µg/kg, individually. The high recurrence particularly of AFs in dry fruits and palatable nuts is viewed as a medical problem for purchasers.

Punam Jeswal (2017) The point of this examination was directed to evaluate the mycotoxigenic parasitic affiliation and cooccurrence of aflatoxins and citrinin tainting in raisins, pistachio nut, walnut and almonds from Indo-gangetic plain region of Bihar. The distinguished citrinin level was lower in focus than aflatoxins yet the sum was adequate to actuate nephrotoxic impacts. The consequences of this examination propose that raisins, pistachio, walnut and almonds are vulnerable substrate for aflatoxigenic just as citrinin creating parasites and further aflatoxins and citrinin creations.

3. MATERIAL AND METHODS

Sampling

A total of 154 dried fruits and nut samples (32 every one of almond and pistachio and 30 samples of every cashew nut, raisins and figs) were gathered from indo-gangetic region of Bihar.

Identification of fungi

All the samples of dried fruits and nuts were haphazardly plated on the newly arranged Potato dextrose agar (PDA) and Standard blotting surface paper and hatched at 28 ± 20 C for 7 days and analyzed every day. Contagious settlement check was recorded following 5 to 7 days. ID was completed by morphological attributes and followed and the ordered plans of Maren for variety Aspergillus, Pitt for Penicillium, Nelson for Fusarium and Funder for different genera.

Detection of mycotoxins

The subjective and quantitative identification for common event of mycotoxins in dried fruits and nuts samples were broke down by catalyst connected immunosorbent examine (ELISA). Aflatoxins were distinguished in the sample by utilizing Total Aflatoxin (AF) examine unit (TO-E0006) and Ochratoxin A test pack (To-E0001) was utilized for location of ochratoxin An and RIDASCREEN FAST citrinin Assay (R6302) for Citrinin discovery.

4. RESULT AND DISCUSSION

Percent incidence of toxigenic fungi

In our current examination, various of parasites were disengaged in which some of them are notable for their mycotoxin creation. A sum of 5 contagious genera have a place with 15 species were separated (Table 1). Growths wereidentified based on their way of life and morphological attributes, these were distinguished as Aspergillus parasiticus, A. niger, A. flavus, A. ochraceus , A. versicolor, A. fumigatus, A. terreus, Penicillium citrinum, P. islandicum, P. verrucosum, Fusarium oxysporum, F. moliniforme, Rhizopus nigricans R. Oryzae and Mucor hiemalis. Aspergillus was the most predominant genera followed by Penicillium and Fusarium (Fig.1). Alghalibi et. al. was likewise separated A. flavus, A. niger, A. terreus , A. ochraceus from raisins and figs samples of Yemen. Zohri revealed relationship of A. fumigatus, A. flavus and A. versicolor in figs and raisins samples and furthermore identified the degree of ochratoxin A was up to 120mg/kg in Fig. samples.

Table 1: Percent incidence of isolated fungi from dried fruits and nuts of Bihar

Name of Fungi	Dry fruit & nuts				
	Almond	Pistachio	Cashew nut	Raisin	Fig
Aspergillus parasiticus	5.4	5.1	3.4	4.5	1.2
Aspergillus niger	2.1	4.2	1.2	3.2	0
Aspergillus flavus	15.6	24.8	13.4	12.4	4.8
Aspergillus ochraceus	14.2	12.1	11.6	15.5	6 .5
Aspergillus versicolour	1.4	3.8	4.2	2.7	0
Aspergillus fumigatus	0	0	1.1	2.6	0
Aspergillus terreus	1.2	0	2.1	0	0
Penicillium citrinum	3.6	2.1	2.4	1.3	1.2
Penicillium islandicum	0	0	1.2	0	0
Penicillium verrucosum	12.4	11.6	13.4	13.8	0
Fusarium oxysporum	2.8	1.2	5.1	1.2	0
Fusarium moniliforme	3.1	0	1.2	0	1.8
Rhizopus nigricans	2.8	1.1	0	0	1.4
Rhizopus oryzae	0	2.5	1.8	0	0
Mucor hiemalis	2.1	1.1	3.1	2.1	1.2

(a) **(b)**

(c) **(d)**

Figure 1: (a) Vigorous contamination of *Aspergillus flavus* and *A.niger* on pistachio, (b) Raisin contaminated with *A. flavus* and *A. niger*, (c) Almond seed having toxigenic strain of *A. flavus* and *A. versicolor*, (d) Figs samples on petriplate having less contamination of toxigenic fungi.

Mycotoxin producing potentiality of isolated fungi

Aflatoxins, ochratoxin An and citrinin creating probability of Aspergillus flavus, A. parasiticus, A. ochraceus, Penicillium citrinium and P. verrucossum were introduced in Table 2. 44% of A. flavus was discovered to be toxigenic and delivered aflatoxins which range between 15.7µg/l – 22.8 µg/l wheras A. parasiticus likewise created aflatoxins yet the degree of possibility was not exactly A. flavus. A. ochaecus and P. verrucosum created ochratoxin A with probability upto 16.5 µg/l and 18.7µg/l resepectivly. In present discovering ochratoxin A was delivered by P. verrucosum and A. ocharecus both. A author done the atomic indicative of comparable organisms for ochratoxin A creation.

Table 2: Mycotoxins producing potentiality of fungi isolated from dried fruit and nuts

Fungi examined	Positive/N.I.Aj	% toxicity	Mycotoxin detected	Potential range (µg/l)
Aspergillus flavus	11/25	44.0	Aflatoxins	15.7 – 22.8
Aspergillus ocharecus	9/25	36.0	Ochratoxin A	6.5 – 16.5
Aspergillus parasiticus	5/18	27.7	Aflatoxins	5.4 – 9.2
Penicillium citrinum	7/18	38.8	Citrinin	3.5 – 9.3
Penicillium verrucosum	3/10	30.0	Ochratoxin A	8.4 – 18.7

[i] **Number of isolates analyzed**

Natural occurrence of mycotoxins in dried fruits and nuts

In present examination, aflatoxins, ochratoxin A and citrinin were distinguished in the samples of dried fruits and nuts gathered from various region of Bihar. The consequences of normal rate of afltoxins, ochratoxins A and citrinin in 5 unique sorts of dried fruits and nuts has been appeared in Table 3. 68.7% of pistachio samples were tainted with aflatoxins followed by 59.3% of almond and 52.5% raisin samples and figs samples had just 12.5% defilement. Most elevated measure of aflatoxins was recorded in pistachio samples (442.8 ng/g) where as in almond and raisins samples, it was 245.8 ng/g and 184.1 ng/g separately. The most minimal measure of aflatoxin was available in figs samples (Fig. 2). Masood et. al. [23] have additionally announced aflatoxins defilement in dried fruits of Pakistan. The sum identified was goes from 3.28 µg/kg to 7.89 µg/kg. They noticed least measure of aflatoxins in dried figs (3.28 µg/kg) and most elevated in Pistachios without shell (7.89 µg/kg).

Table 3: Natural occurrence of amount of mycotoxins contamination in dried fruits and nuts

Samples	Numbe r of sample analyze d	% Contaminati on	Amount (ng/g) Mean ± S.E		
			Aflatoxi ns	Ochratoxi n A	Citrini n
Almond	32	59.3	245.8 ± 34.2	154.2 ± 18.8	184.1 ± 28.5
Pistachi o	32	68.7	442.8 ± 35.1	193.8 ± 35.7	158.0 ± 45.7
Cashew nut	30	43.3	214.5 ± 37.2	179.5 ± 28.5	98.8 ± 47.3
Raisin	30	52.5	184.1 ± 20.2	215.1 ± 30.2	174.1 ± 28.5

Fig.	30	12.5	35.4 ± 24.8	ND	ND

In this investigation, Ochratoxin A and citrinin was likewise distinguished from a wide range of dried fruits and nuts with the exception of figs. The most elevated measure of Ochratoxin A was recognized in raisin samples (215.1 ng/g) and least in almonds (154.2 ng/g). Citrinin pollution was most extreme in almonds (184.1 ng/g) and least in cashew nut (98.8 ng/g). Every one of the 5 sorts of dried fruits and nuts were debased with aflatoxins though ochratoxin An and citrinin was not distinguished from Fig. samples.

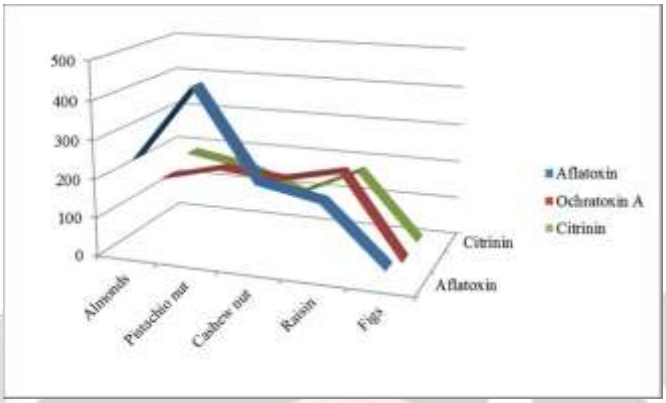

Figure 2: Showing pistachio had maximum aflatoxin contamination

5. CONCLUSION

Based on the current investigation, it could be reasoned that the dried fruits and nuts are rich substrate for contagious development and further mycotoxin creations. Each of the 5 kinds of dried fruits and nuts are debased from gangetic plain with aflatoxins and the identification level was amazingly higher than the admissible furthest reaches of EU. Ochratoxin An and citrinin was likewise distinguished from pistachio, raisins, almond and cashew nut samples and the recognized sum was adequately high to actuated toxicity from Fig. samples were impervious to ochratoxin An and citrinin creation and none of the samples of figs were discovered sullied with octratoxin An and citrinin. It is critical to mind in preparing, taking care of and transportation to lessen the defilement of these dangerous mycotoxins in dried fruits and nuts.

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