Scientific Inquiry and Review (SIR)
Volume 8 Issue 1, 2024
ISSN (P): 2521-2427, ISSN (E): 2521-2435
Homepage: https://journals.umt.edu.pk/index.php/SIR

Article QR

Title:	**Assessment of Total Aflatoxin Content in Dry Fruits Samples Collected from Local Markets of Lahore, Pakistan**
Author (s):	Sadaf Sarfraz[1], Muhammad Ikram[1], Manzar Zahra[1], Shabbir Hussain[2], Abdul Barri[1]
Affiliation (s):	[1]Lahore Garrison University, Lahore, Pakistan [2]Khawaja Fareed University of Engineering and Information Technology, Rahim Yar Khan, Pakistan
DOI:	https://doi.org/10.32350/sir.81.06
History:	Received: September 3, 2023, Revised: November 14, 2023, Accepted: November 16, 2023, Published: March 15, 2024
Citation:	Sarfraz S, Ikram M, Zahra M, Hussain S, Barri A. Assessment of total aflatoxin content in dry fruits samples collected from local markets of Lahore, Pakistan. *Sci Inq Rev*. 2024;8(1):94–109. https://doi.org/10.32350/sir.81.06
Copyright:	© The Authors
Licensing:	This article is open access and is distributed under the terms of Creative Commons Attribution 4.0 International License
Conflict of Interest:	Author(s) declared no conflict of interest

A publication of
The School of Science
University of Management and Technology, Lahore, Pakistan

Assessment of Total Aflatoxin Content in Dry Fruits Samples Collected from Local Markets of Lahore, Pakistan

Sadaf Sarfraz[1*], Muhammad Ikram[1], Manzar Zahra[1], Shabbir Hussain[2], Abdul Barri[1]

[1]Department of Chemistry, Lahore Garrison University, Lahore, Pakistan
[2]Department of Chemistry, Khawaja Fareed University of Engineering and Information Technology, Rahim Yar Khan, Pakistan

ABSTRACT

This report presents an evaluation of the concentration of total aflatoxins in a range of dry fruits obtained from local markets in Lahore, Pakistan. It also proposes some risk mitigation strategies through detoxification. The analysis involved appropriate techniques to accurately quantify the total aflatoxin (AF) content in each sample. According to the findings, AFs were not found in any of the branded dry fruit samples. However, AFB1 contamination in open samples of almonds, peanuts, apricots, walnuts, raisins, figs, and coconut was found in concentrations exceeding the EU guidelines. Furthermore, dry fruit samples collected from branded companies showed no AF- contamination. These findings suggest potentially high health risks posed by using dry fruits from open markets. This fact further emphasizes the importance of detoxification methods for safer consumption.

Keywords: aflatoxins (AFs), contamination, dry fruits, ELISA, local markets, TLC

Highlights

- The research focuses on Lahore, Pakistan, making the findings directly applicable to the local community, serving as a foundation for targeted food safety measures.
- The report rigorously evaluates the presence and concentration of aflatoxins in various dry fruit types commonly available in Lahore's local markets, providing precise data on the extent of contamination.

*Corresponding Author: dr.sadafsarfraz@lgu.edu.pk

- The study investigates and proposes practical methods to detoxify these aflatoxin-contaminated dry fruits, offering actionable solutions to enhance food safety.

1.INTRODUCTION

Aflatoxins (AFs) are naturally occurring poisonous mutagens found in various food products. These poisonous mutagens are considered unsafe for both human beings and animals [1]. Aflatoxins are produced by certain species of molds named Aspergillus flavus and Aspergillus parasiticus. They are destructive for food and the expulsion of these mutagens from edibles is very important [1]. These microorganisms produce noxious secondary metabolites called mycotoxins via a series of synthetic and enzymatic reactions. The accumulation of AFs in human body may lead to cancer or may result in liver damage. Furthermore, their accumulation in circulatory system makes human beings and animals suffer from various hepatotoxic, mutagenic, carcinogenic, and teratogenic diseases [2].

Different groups of AFs, such as B1, B2, G1, and G2, have been identified. Among them, major genus including AFB1, AFB2, AFG1, and AFG2 are mostly found in animal feeds. AFB1 has been reported as the most well-known and poisonous AF adversely affecting human health, thus it is characterized as group 1 human carcinogenic compound [3]. Although different AF mutagens have been identified, yet they are closely associated with each other showing a slight difference in their chemical compositions.

According to one study, mycotoxins damage more than one-fourth of the world's protein yields [4]. At present, with a continuous increase in world’s population, there is a constant detrimental effect on natural food resources. Any damage to food products due to the growth of AF mutagens may generate a huge burden on various food protein supplies and may lead to food scarcity [5]. So, AF occurrence needs to be properly measured and steps may be taken to control its growth in order to avoid any food damage.

Generally, in deprived states, filthy food supplies and inadequate safety measures make health products more prone to mycotoxin growth. For instance, hazelnut (Corylus avelanna L.), a widespread nut, is primarily cultured on the shoreline of Black Sea. Its hard shells have a good blockade against fungal contamination but AF development may occur due to storage and weather conditions. Thus, there is a possibility that reduced nutrition safety measures may lead to AF contamination [6].

Dry fruits are widely grown all around the world, especially in Pakistan. Pathogenic fungi can adhere to dry fruits and nuts during their cultivation, growth, ripening, overripening, handling, drying, storage, and transportation. The most common pathogenic fungi are Aspergillus, Fusarium, Penicillium, and Alterneria, which produce 78 toxigenic chemicals known as mycotoxins. Various countries have set criteria for the acceptable level of AFs in dry fruits and nuts due to their extreme toxicity. The European Commission, for example, has set maximum tolerable limits (MTL) of 4 and 10 g/kg for total aflatoxins in dry fruits and nuts, respectively[1]. The MTL for total AFs in dry fruits and nuts is 20 g/kg, according to the Food and Drug Administration (FDA) and the Food and Agriculture Organization of the United States of America.

The minimum toxicity level for mycotoxins in dry fruits and edible nuts has yet to be established in Pakistan. Based on the foregoing discussion, quick detection and quantification of AFs in dry fruits and nuts is vital to ensure safety, quality, and the execution of hazard analyses and critical control points (HACCP). Hence, the current study was designed to determine the level of AFs in Pakistani dried fruits and edible nuts. Furthermore, a comparison of the efficiency of various naturally occurring organic compounds in detoxifying AFs was also conducted.

2.MATERIALS AND METHOD

2.1 Sample Collection

All forty (40) samples of processed and unprocessed dry fruits were collected from various local markets in Lahore. The sample procedure was modified to conform to the approved AOAC approach. The collected samples were pulverized using the sample processor (Model ILP, FBRC/AL/05) to obtain a uniform blend. Afterwards, each sample was obtained into a final quantity of 100 g. Following this, 50g of each sample was isolated for AF testing, as per the standard method of AF determination. For experimental purposes, all samples were stored in opaque plastic bags until the analysis was performed.

2.2 Extraction and Analysis of AFs

AF standard of B1, B2, G1, and G2 in acetonitrile were purchased from Trilogy Analytical Laboratory (870 Vossbrink Dr, Washington, MO 63090,

[1] Commission of the European Communities

USA). Thin layer chromatographic (TLC) plates were imported from Merck (290 Concord Road Billerica Massachusetts USA). All standards were stored in freezer at -20°C till further use. Depending on the chemical content of the concerned food product, a variety of AF extraction methods have been reported in the literature. In the current study, AFs were analyzed in dry fruits using TLC plates imported from Merck (290 Concord Road Billerica Massachusetts USA), while chloroform (product of Sigma-Aldrich UK) was employed for the extraction process. For this purpose, 50g of each grinded sample was mixed with 25 g of diatomaceous earth in 25 ml of water and the final volume was increased to 150 ml using chloroform. The solution was then shaken vigorously for 30 minutes using a wrist arm shaker. The prepared sample solution was filtered using Whatman filter paper 1. Quantitative determination of AF was done using the method reported by [7]. ELISA (Enzyme linked immune sorbent assay) methodology reported by [8, 9] was used to analyze the samples [10–12].

2.3 Statistical Analysis

ANOVA was used to assess the differences in AFs concentrations in dry fruits and edible nuts, followed by a post hoc Tukey's honest significant difference (HSD) Test [13]. The *p*-value $P \leq 0.05$ was used to determine whether the mean values were substantially different. Using SPSS software, all experimental data was reported as the mean ± standard deviation (SD) in triplicate (IBM, PASW 117 Statistics19, USA). R^2 was calculated using regression analysis/correlation.

2.4 Detoxification Studies

To suppress pathogenic mycotoxin development in food, various researchers have reported a variety of physical, chemical, and biological approaches [7, 14–16]. According to a study conducted by Velazhahan [17] , the simplest strategy to reduce mycotoxin degradation is to use a brief procedure [17] at various levels, such as during processing and harvesting. Furthermore, natural compounds can be used to effectively reduce AF growth to safer levels by eliminating, degrading, and converting them into less hazardous AFs [17].

In this study, the methods used to detoxify AF included using garlic (Allium sativum) [17], Nigella sativa seed oil [18], citric acid [19], and sodium bicarbonate [20].

3.RESULTS AND DISCUSSION

Table 1 summarizes the natural occurrence of AF contamination in various samples of dry fruits. The results indicated that AFs were not found in processed/packaged dry fruit samples; however, they were found in raw samples. According to the findings, 109 samples (77%) of the 140 unprocessed dry fruit samples were found to be contaminated with AFs at various concentrations. The contamination ranged from 1.04 to 15.12 µg/kg, with an average of 13.9 µg/kg, which is much higher than the EU's maximum tolerable limit (MTL = 4 and 10µg/kg, respectively) for dried fruits and nuts. The findings revealed that AFs B2, G1, and G2 were not found in any of the dry fruit samples, while AFB1 was found in all of them. However, these samples met the MTL (20 µg/kg) set by the United States and were suitable for human use.

It was discovered that the level of AF contamination in various types of dry fruits was highly variable. For example, raisins had the highest AF contamination, with nearly 50.0% of the samples tested positive for AFs over the MTL limit, with a mean level of 10.1µ g/kg, respectively. AF detection range for raisins was 5.89-14.68 µgk/g with 15 samples affected, whereas 11 samples were within the permissible limits and the remaining 04 were not. Walnut (7.7 µg/kg), coconut (7.8 µg/kg), and apricot (7.2 µg/kg) had the lowest AF mean levels, respectively. However, according to the findings (Table 2), the maximum quantity of AF was found in one walnut sample (15.12 µg/kg) and the amount violated FDA and WHO rules, as the acceptable limit is 10 µg/kg. Out of the 16 coconut samples tested for AF, 3 samples were beyond the limit and 13 were found within the acceptable limits. The maximum AF level in fig was 14g/kg, whereas the maximum AF level in almonds was 11.34 µg/kg. For almonds, 12 out of 16 contaminated samples were within permissible limits and 04 samples were above the European standard's permissible limits.

AF contamination level in several species of dry fruits and nuts from many countries including Pakistan have been documented by various studies. In this regard, [21] reported from Pakistan that AFB1 and total AFs were found in 132 (43%) samples of dry fruits and edible nuts, out of a total of 307 samples. Contamination ranged from 21.50 g/kg to 4.90 g/kg on average. According to Luttfullah [22], the contamination range of AFs in different varieties of dry fruits and nuts in Pakistan was 20% to 50%. In contrast, the findings of the current study revealed the content of AFs (more

than 70%) as substantially larger than the ones reported in prior research [22].

According to several studies, dried raisins do not provide a suitable surface or environment for the growth of Aspergillus flavus and production of AFs [23]. However, in the current investigation, the highest concentration of AFs was observed in dried raisins. This implies that climatic factors have a significant impact on the level of AF contamination in dried fruits and edible nuts during the growing season. Diverse farming and harvesting practices, soil type, microbial flora, and varied temperature and humidity all play a role in AF contamination in dried fruits and edible nuts across the country. Additionally, poor harvesting and management techniques, as well as mechanical damage during harvesting, minimal curing, low-quality materials, and insufficient storage and transit conditions, all aid the establishment of fungal diseases. As a result, high AF levels were observed in dry fruits. Figure 1 indicates a bar graph showing the percentage occurrence of AFs in various dry fruit samples analyzed in this study.

Table 1. Screening of Open Samples for AFs by TLC

AF	Samples	No. of Samples	No. of Contaminated samples	No. of Uncontaminated samples	Contamination %	Max μgkg^{-1}	Permissible Limit
B1	Almond	20	16	4	80%	14.12	10 μgkg^{-1}
	Peanuts	20	15	5	75%	13.93	10 μgkg^{-1}
	Apricots	20	15	5	75%	13.10	10 μgkg^{-1}
	Walnuts	20	17	3	85%	15.12	10 μgkg^{-1}
	Raisins	20	15	5	75%	14.68	10 μgkg^{-1}
	Figs	20	15	5	75%	14.61	10 μgkg^{-1}
	Coconut	20	16	4	80%	11.89	10 μgkg^{-1}

*AF- Aflatoxin

Table 2. Positive Open Samples eere Triplicate to Calculate Mean and SD for AFB1 Found in Contaminated Dry Fruits Samples

Contaminated Sample of Dry Fruits (Sample ID's)	Aflatoxin conc. Attempt 1 (µg/kg)	Aflatoxin conc. Attempt 2 (µg/kg)	Aflatoxin conc. Attempt 3 (µg/kg)	Average ± SD (µg/kg)
Alm 2	11.87	11.08	11.34	11.43 ± 0.40
Alm 3	9.56	9.12	9.31	9.33 ± 0.22
Alm 4	8.87	8.08	8.34	8.43 ± 0.40
Alm 5	13.86	14.12	14.01	13.99 ± 0.13
Alm 7	1.55	1.04	1.87	1.49 ± 0.42

Contaminated Sample of Dry Fruits (Sample ID's)	Aflatoxin conc. Attempt 1 (µg/kg)	Aflatoxin conc. Attempt 2 (µg/kg)	Aflatoxin conc. Attempt 3 (µg/kg)	Average ± SD (µg/kg)
Alm 8	7.71	6.98	7.56	7.42 ± 0.38
Alm 9	6.87	6.08	6.34	6.43 ± 0.40
Alm 10	3.86	4.12	4.01	3.99 ± 0.13
Alm 12	10.55	10.04	10.87	10.49 ± 0.42
Alm 13	5.71	4.98	5.56	5.42 ± 0.38
Alm 14	3.91	3.51	3.03	3.48 ± 0.44
Alm 16	9.94	9.45	9.25	9.54 ± 0.35
Alm 17	10.89	9.56	10.05	10.16 ± 0.67
Alm 18	7.46	7.22	7.64	7.44 ± 0.21
Alm 19	6.44	6.45	6.98	6.62 ± 0.30
Alm 20	9.26	9.68	9.43	9.45 ± 0.21
Pea 1	9.95	9.71	9.45	9.70 ± 0.25
Pea 2	7.03	7.89	7.77	7.56 ± 0.46
Pea 3	12.72	12.3	12.02	12.34 ± 0.35
Pea 5	5.73	6.01	5.60	5.78 ± 0.21
Pea 6	8.56	8.48	8.24	8.42 ± 0.16
Pea 7	9.93	9.89	10.05	9.95 ± 0.08
Pea 8	8.89	8.56	8.20	8.55 ± 0.34
Pea 9	13.93	13.56	13.01	13.51 ± 0.46
Pea 11	4.73	4.32	4.25	4.43 ± 0.25
Pea 14	9.59	9.6	9.35	9.51 ± 0.14
Pea 15	9.29	9.58	9.78	9.55 ± 0.24
Pea 16	8.63	8.34	8.78	8.58 ± 0.22
Pea 17	9.26	9.68	9.43	9.45 ± 0.21
Pea 19	6.85	6.87	6.45	6.72 ± 0.23
Pea 20	11.23	11.43	11.87	11.51 ± 0.32
Apri 1	6.09	5.97	6.32	6.12 ± 0.17
Apri 4	8.77	8.13	8.97	8.62 ± 0.43
Apri 5	12.20	12.03	11.90	12.04 ± 0.15
Apri 6	9.21	8.88	9.16	9.08 ± 0.17
Apri 7	4.53	4.35	4.05	4.31 ± 0.24
Apri 9	11.14	11.71	11.47	11.44 ± 0.28
Apri 10	8.23	7.96	8.31	8.16 ± 0.18
Apri 11	1.43	1.19	1.26	1.29 ± 0.12
Apri 12	12.78	13.10	12.99	12.95 ± 0.16
Apri 13	3.37	3.63	3.11	3.37 ± 0.26
Apri 14	3.26	2.89	3.45	3.20 ± 0.28
Apri 16	9.09	9.33	9.24	9.22 ± 0.12
Apri 18	7.41	7.28	7.22	7.30 ± 0.09
Apri 19	1.87	1.08	1.34	1.43 ± 0.40

Contaminated Sample of Dry Fruits (Sample ID's)	Aflatoxin conc. Attempt 1 (µg/kg)	Aflatoxin conc. Attempt 2 (µg/kg)	Aflatoxin conc. Attempt 3 (µg/kg)	Average ± SD (µg/kg)
Apri 20	9.56	9.12	9.31	9.33 ± 0.22
Waln 1	8.87	8.08	8.34	8.43 ± 0.40
Waln 2	12.86	13.12	13.01	12.99 ± 0.13
Waln 3	9.55	9.04	9.87	9.49 ± 0.42
Waln 4	7.71	6.98	7.56	7.42 ± 0.38
Waln 6	8.87	8.08	8.34	8.43 ± 0.40
Waln 7	14.86	15.12	14.91	14.96 ± 0.11
Waln 8	8.55	8.04	8.87	8.49 ± 0.42
Waln 10	12.71	11.98	12.56	12.42 ± 0.38
Waln 11	2.91	2.51	2.03	2.48 ± 0.44
Waln 12	4.94	4.45	4.25	4.54 ± 0.35
Waln 13	10.89	9.56	10.05	10.16 ± 0.67
Waln 14	7.46	7.22	7.64	7.44 ± 0.21
Waln 15	6.44	6.45	6.98	6.62 ± 0.30
Waln 17	9.95	9.71	9.45	9.70 ± 0.25
Waln 18	2.03	1.89	1.77	1.89 ± 0.10
Waln 19	2.72	2.30	2.02	2.34 ± 0.35
Waln 20	5.73	6.01	5.20	5.78 ± 0.21
Rais 2	6.25	5.89	5.95	6.03 ± 0.15
Rais 3	6.55	6.04	6.87	6.49 ± 0.42
Rais 4	7.71	7.98	7.56	7.75 ± 0.21
Rais 5	13.91	13.51	13.03	13.48 ± 0.44
Rais 6	8.94	8.45	8.25	8.54 ± 0.35
Rais 8	9.26	9.68	9.43	9.45 ± 0.21
Rais 9	12.85	12.87	12.45	12.72 ± 0.23
Rais 11	11.23	11.43	11.87	11.51 ± 0.32
Rais 12	10.09	9.97	10.32	10.12 ± 0.17
Rais 13	8.77	8.13	8.97	8.62 ± 0.43
Rais 15	14.26	14.68	14.43	14.45 ± 0.21
Rais 16	6.85	6.87	6.45	6.72 ± 0.23
Rais 17	11.23	11.43	11.87	11.51 ± 0.32
Rais 18	10.09	9.97	10.32	10.12 ± 0.17
Rais 19	13.77	13.13	13.97	13.62 ± 0.43
Fig 2	7.98	7.37	7.29	7.54 ± 0.30
Fig 3	12.25	12.56	12.05	12.28 ± 0.25
Fig 5	4.61	4.43	4.51	4.51 ± 0.07
Fig 6	11.46	11.22	11.64	11.44 ± 0.21
Fig 7	4.73	4.32	4.25	4.43 ± 0.25
Fig 9	9.59	9.6	9.35	9.51 ± 0.14
Fig 10	9.29	9.58	9.78	9.55 ± 0.24

Contaminated Sample of Dry Fruits (Sample ID's)	Aflatoxin conc. Attempt 1 (µg/kg)	Aflatoxin conc. Attempt 2 (µg/kg)	Aflatoxin conc. Attempt 3 (µg/kg)	Average ± SD (µg/kg)
Fig 12	14.59	14.61	14.35	14.51 ± 0.14
Fig 13	10.21	9.83	10.37	10.13 ± 0.27
Fig 14	9.26	9.68	9.43	9.45 ± 0.21
Fig 15	6.85	6.87	6.45	6.72 ± 0.23
Fig 16	1.23	1.43	1.87	1.51 ± 0.32
Fig 18	7.09	6.97	7.32	7.12 ± 0.17
Fig 19	8.77	8.13	8.97	8.62 ± 0.43
Fig 20	3.44	3.45	3.98	3.62 ± 0.30
Coco 1	5.95	5.05	5.45	5.48 ± 0.45
Coco 2	10.03	9.89	9.77	9.89 ± 0.13
Coco 4	4.73	4.32	4.25	4.43 ± 0.25
Coco 5	9.59	9.6	9.35	9.51 ± 0.14
Coco 6	9.29	9.58	9.78	9.55 ± 0.24
Coco 8	1.59	1.61	1.35	1.51 ± 0.14
Coco 9	10.21	9.83	10.37	10.13 ± 0.27
Coco 10	9.26	9.68	9.43	9.45 ± 0.21
Coco 11	6.85	6.87	6.45	6.72 ± 0.23
Coco 12	11.33	11.41	11.89	11.54 ± 0.24
Coco 14	10.09	9.97	10.32	10.12 ± 0.17
Coco 16	8.77	8.13	8.97	8.62 ± 0.43
Coco 17	2.19	2.73	2.36	2.42 ± 0.22
Coco 18	6.73	6.01	6.6	6.44 ± 0.38
Coco 19	3.56	3.48	3.24	3.42 ± 0.16
Coco 20	7.93	7.56	7.01	7.50 ± 0.46

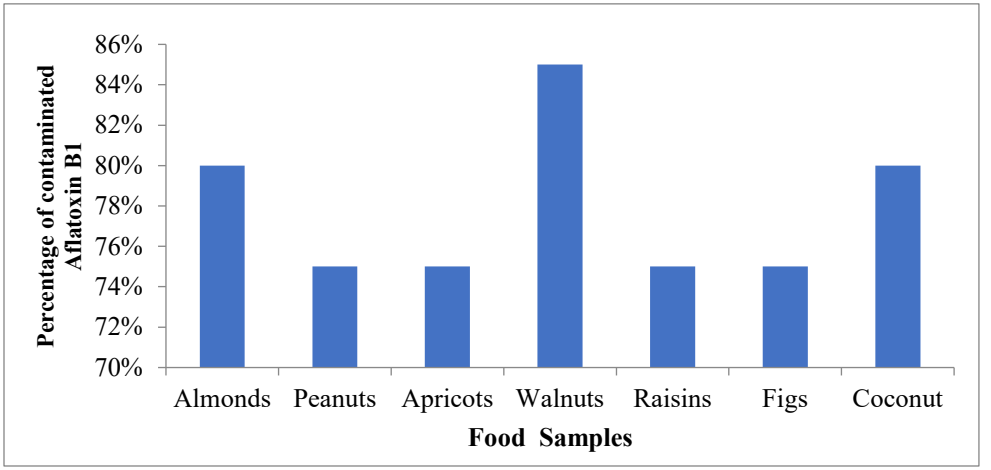

Figure 1. Occurrence of AFs in Dry Fruit Samples

3.1 Chemical Detoxification of AFs

Several studies have proposed to recover contaminated goods physically or chemically by lowering AFs to an acceptable level. Biological detoxification mechanisms, such as fermentation, profoundly change the characteristics of edibles and hence are not recommended. Chemical detoxification procedures are appealing because of their high efficiency and low cost. The main goal of chemical treatment is to activate AF molecules by oxidation, hydrolysis, or addition reactions, causing AFs to disintegrate. Keeping in view the efficacy of chemical compounds, citric acid and sodium bicarbonate were used to detoxify AFs, while black seed oil and garlic were found to be useful in reducing fungus in the current study. Based on the findings, black seed oil was the most effective decontaminating agent employed in this study. Figure 2 shows the results of the comparative examination of the decontaminating efficacy of the chemical agents used. Although these chemical approaches can efficiently detoxify AFs, however, their applicability is limited due to the safety of the degraded compounds and the removal of leftover chemicals after treatment with detoxifying compounds. In regions where AF contamination is common, successful detoxification methods can support local agriculture and trade by enabling safe production and export of dry fruits.

Table 3. Detoxification in Almonds with Natural Compounds

Sr. No	Sample	Aflatoxin conc. before Treatment (ppb)	Treatment with Natural Compounds	Aflatoxin conc. after Treatment (ppb)	Aflatoxin Reduction (%)
1	Almond	14.12	*Allium sativum* (Garlic)	1.32	90.65%
2	Almond		Black Seed Oil	0.0	100%
3	Almond		Citric Acid	3.41	75.84%
4	Almond		Sodium Bicarbonate	2.70	80.87%

Table 4. Detoxification in Peanut with Natural Compounds

Sr. No	Sample	Aflatoxin conc. before Treatment (ppb)	Treatment with Natural Compounds	Aflatoxin conc. after Treatment (ppb)	Aflatoxin Reduction (%)
1	Peanut	13.93	*Allium sativum* (Garlic)	1.50	89.23%
2	Peanut		Black Seed Oil	0.0	100%
3	Peanut		Citric Acid	2.95	78.82%
4	Peanut		Sodium Bicarbonate	2.78	80.04%

Table 5. Detoxification in Apricots with Natural Compounds

Sr. No	Sample	Aflatoxin conc. before Treatment (ppb)	Treatment with Natural Compounds	Aflatoxin conc. after Treatment (ppb)	Aflatoxin Reduction (%)
1	Apricot	13.10	*Allium sativum* (Garlic)	1.15	91.22%
2	Apricot		Black Seed Oil	0.0	100%
3	Apricot		Citric Acid	3.11	76.25%
4	Apricot		Sodium Bicarbonate	2.89	77.93%

Table 6. Detoxification in Walnut with Natural Compounds

Sr. No	Sample	Aflatoxin conc. before Treatment (ppb)	Treatment with Natural Compounds	Aflatoxin conc. after Treatment (ppb)	Aflatoxin Reduction (%)
1	Walnut	15.12	*Allium sativum* (Garlic)	1.43	90.54%
2	Walnut		Black Seed Oil	0.0	100%
3	Walnut		Citric Acid	3.50	76.85%
4	Walnut		Sodium Bicarbonate	2.67	82.34%

Table 7. Detoxification in Raisin with Natural Compounds

Sr. No	Sample	Aflatoxin conc. before Treatment (ppb)	Treatment with Natural Compounds	Aflatoxin conc. after Treatment (ppb)	Aflatoxin Reduction (%)
1	Raisin	14.68	*Allium sativum* (Garlic)	1.77	87.94%
2	Raisin		Black Seed Oil	0.0	100%
3	Raisin		Citric Acid	2.87	80.44%
4	Raisin		Sodium Bicarbonate	2.65	81.94%

Table 8. Detoxification in Fig with Natural Compounds

Sr. No	Sample	Aflatoxin conc. before Treatment (ppb)	Treatment with Natural Compounds	Aflatoxin conc. after Treatment (ppb)	Aflatoxin Reduction (%)
1	Fig	14.61	*Allium sativum* (Garlic)	0.85	94.18%
2	Fig		Black Seed Oil	0.0	100%
3	Fig		Citric Acid	3.44	76.45%
4	Fig		Sodium Bicarbonate	2.71	81.45%

Table 9. Detoxification in Coconut with Natural Compounds

Sr. No	Sample	Aflatoxin conc. before Treatment (ppb)	Treatment with Natural Compounds	Aflatoxin conc. after Treatment (ppb)	Aflatoxin Reduction (%)
1	Coconut	11.89	*Allium sativum* (Garlic)	0.62	94.78%
2	Coconut		Black Seed Oil	0.0	100%
3	Coconut		Citric Acid	2.53	78.72%
4	Coconut		Sodium Bicarbonate	2.12	82.16%

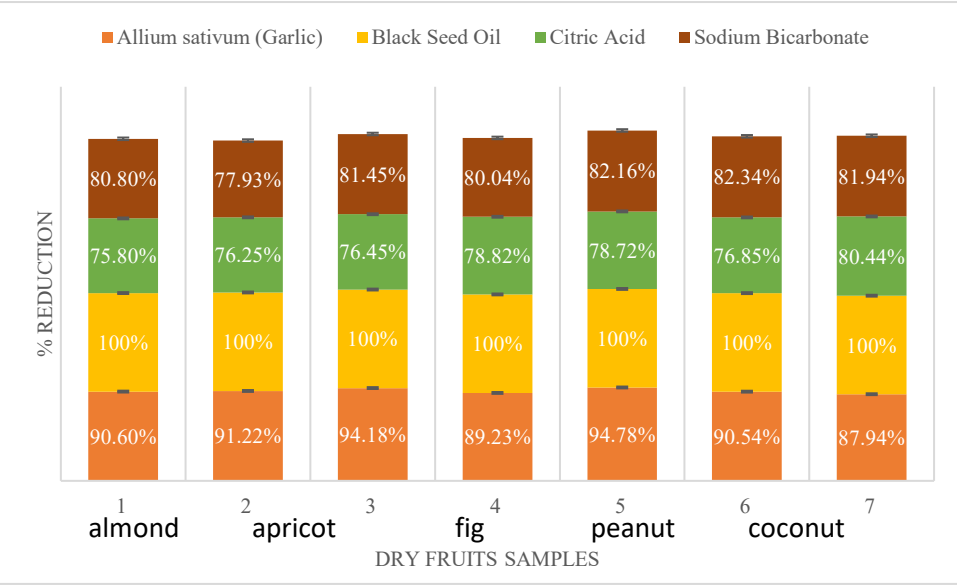

Figure 2. Comparative Analysis of the Ability of Chemical Compounds to Detoxify Collected Dry Fruit Samples

3.2. Conclusion

Regular consumption of AF-contaminated food might result in serious health hazards, potentially leading to liver cancer and other health issues due to their carcinogenic properties for the end user. Dried fruits, which play a major role in the daily diet, are more likely to contain AFs. AFs were not discovered in processed samples, according to the findings. Although, they were found in 109 (39%) of the 280 samples of unprocessed dry fruits.

AFs in agricultural commodities can be minimized by avoiding fungal growth at the farm level. Food and feed handlers should be made aware of

improper practices that lead to AF- contamination, such as the Department of Public Health and the Ministry of Agriculture. It is essential to employ measures, such as good farming practices, appropriate drying, handling, packaging, and adequate storage and transportation to increase the export of dry fruits and edible nuts from Pakistan.

Furthermore, diverse control procedures, such as dry heating, roasting, traditional microwave baking, gamma radiation, UV exposure, hydrogen peroxide treatment, and storage in various climates show differing degrees of AF count destruction and could be used as food safety measures. It is recommended that consumers purchase dried fruits from reputable retailers and have them processed. Furthermore, the materials should be maintained in a cool, dry environment and any filthy, unsealed, or damaged packing should be discarded.

ACKNOWLEDGEMENT

The authors would like to acknowledge the technical support and lab facilities received from Food and Biotechnology Research Centre, PCSIR, Laboratories, Lahore.

CONFLICT OF INTEREST

The authors of the manuscript have no financial or non-financial conflict of interest in the subject matter or materials discussed in this manuscript.

DATA AVAILABILITY STATEMENT

The data associated with this study will be provided by the corresponding author upon request.

REFERENCES

1. Huwig A, Freimund S, Käppeli O, Dutler H. Mycotoxin detoxication of animal feed by different adsorbents. *Toxicol Lett*. 2001;122(2):179–88. https://doi.org/10.1016/s0378-4274(01)00360-5
2. Qazi JI, Fayyaz Z. Aflatoxin contaminated foods and health risk perspective for Pakistani population. *Mycopath*. 2006;4(2):27–34.
3. Iqbal A, Khalil IA, Shah H. Aflatoxin contents of stored and artificially inoculated cereals and nuts. *Food Chem*. 2006;98(4):699–703. http://dx.doi.org/10.1016/j.foodchem.2005.06.034
4. Kabak B, Dobson AD, Var Il. Strategies to prevent mycotoxin contamination of food and animal feed: a review. *Crit Rev Food Sci*

Nutr. 2006;46(8):593–619. https://doi.org/10.1080/10408390500436185

5. Juan C, Zinedine A, Molto J, Idrissi L, Manes J. Aflatoxins levels in dried fruits and nuts from Rabat-Salé area, Morocco. *Food Control*. 2008;19(9):849–53. https://doi.org/10.1016/j.foodcont.2007.08.010
6. Bhat R, Ramakrishna Y, Beedu S, Munshi K. Outbreak of trichothecene mycotoxicosis associated with consumption of mould-damaged wheat products in Kashmir Valley, India. *Lancet*. 1989;333(8628):35–37. https://doi.org/10.1016/S0140-6736(89)91684-X.
7. Gratz S, Wu Q, El-Nezami H, Juvonen R, Mykkänen H, Turner P. Lactobacillus rhamnosus strain GG reduces aflatoxin B1 transport, metabolism, and toxicity in Caco-2 cells. *Appl Environ Microb*. 2007;73(12):3958–3964. https://doi.org/10.1128/AEM.02944-06
8. Eslami M, Mashak Z, Heshmati A, Shokrzadeh M, Nejad ASM. Determination of aflatoxin B1 levels in Iranian rice by ELISA method. *Toxin Rev*. 2015;34(3):125–28. https://doi.org/10.3109/15569543.2015.1074925
9. Nejad ASM, Ghannad MS, Kamkar A. Determination of aflatoxin B1 levels in Iranian and Indian spices by ELISA method. *Toxin Rev*. 2014;33(4):151–54. https://doi.org/10.3109/15569543.2014.942319
10. Romer TR, Campbell AD. Collaborative study of a screening method for the detection of aflatoxins in mixed feeds, other agricultural products, and foods. *J Assoc Off Anal Chem*. 1976;59(1):110–117. https://doi.org/10.1093/jaoac/59.1.110
11. Romer TR. Screening method for the detection of aflatoxins in mixed feeds and other agricultural commodities with subsequent confirmation and quantitative measurement of aflatoxins in positive samples. *J Assoc Off Anal Chem*. 1975;58(3):500–506. https://doi.org/10.1093/jaoac/58.3.500
12. Nazir A, Asif M, Kalim I, Ahmad N, Iqbal M. Quantification of aflatoxins in dry fruits using ELISA with reference to standards for human consumption. *Chem Intl*. 2021;7(4):224–229.
13. Asghar MA, Ahmed A, Zahir E, Asghar MA, Iqbal J, Walker G. Incidence of aflatoxins contamination in dry fruits and edible nuts collected from Pakistan. *Food Contl*. 2017;78:169–175.

14. Phillips TD, Lemke SL, Grant PG. Characterization of clay-based enterosorbents for the prevention of aflatoxicosis. *Adv Exp Med Biol*. 2002:157–171. https://doi.org/10.1007/978-1-4615-0629-4_16

15. Tian J, Zeng X, Zeng H, Feng Z, Miao X, Peng X. Investigations on the antifungal effect of nerol against Aspergillus flavus causing food spoilage. *Sci World J*. 2013;2013:e230795. https://doi.org/10.1155/2013/230795

16. El-Aziz A, Abeer R, Mahmoud MA, Al-Othman MR, Al-Gahtani MF. Use of selected essential oils to control aflatoxin contaminated stored cashew and detection of aflatoxin biosynthesis gene. *Sci World J*. 2015;2015:e958192. https://doi.org/10.1155/2015/958192

17. Velazhahan R, Vijayanandraj S, Vijayasamundeeswari A, et al. Detoxification of aflatoxins by seed extracts of the medicinal plant, Trachyspermum ammi (L.) Sprague ex Turrill–structural analysis and biological toxicity of degradation product of aflatoxin G1. *Food Cnt*. 2010;21(5):719–725. https://doi.org/10.1016/j.foodcont.2009.10.014

18. Karlovsky P, Suman M, Berthiller F, et al. Impact of food processing and detoxification treatments on mycotoxin contamination. *Mycotoxin Res*. 2016;32(4):179–205. https://doi.org/10.1007/s12550-016-0257-7

19. Rastegar H, Shoeibi S, Yazdanpanah H, et al. Removal of aflatoxin B1 by roasting with lemon juice and/or citric acid in contaminated pistachio nuts. *Food Cont*. 2017;71:279–284. https://doi.org/10.1016/j.foodcont.2016.06.045

20. Samarajeewa U, Sen A, Cohen M, Wei C. Detoxification of aflatoxins in foods and feeds by physical and chemical methods. *J Food Prot*. 1990;53(6):489–501. https://doi.org/10.4315/0362-028X-53.6.489

21. Masood M, Iqbal SZ, Asi MR, Malik N. Natural occurrence of aflatoxins in dry fruits and edible nuts. *Food Contl*. 2015;55:62–65. https://doi.org/10.1016/j.foodcont.2015.02.041

22. Luttfullah G, Hussain A. Studies on contamination level of aflatoxins in some dried fruits and nuts of Pakistan. *Food Contl*. 2011;22(3-4):426–429. https://doi.org/10.1016/j.foodcont.2010.09.015

23. Onyeaka H, Ghosh S, Obileke K, et al. Preventing chemical contaminants in food: Challenges and prospects for safe and sustainable food production. *Food Contl*. 2024;155:e110040. https://doi.org/10.1016/j.foodcont.2023.110040