

# **ORIGINAL ARTICLE**

# Investigation of total aflatoxin in nuts and dried fruits and consumption habits during pregnancy in Türkiye

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#### **ABSTRACT**

Background: During pregnancy, adequate energy, nutrient intake, and food safety are important. Aims: This study aimed to examine the presence of aflatoxin, which has toxigenic properties, in nuts and dried fruits consumed during pregnancy. Subjects and Methods: Forty-five pregnant women living in Istanbul and Balikesir cities provinces, who were selected using the convenience sampling method, were applied face-to-face interview method on their attitudes and behaviors regarding their preferences for consuming nuts and dried fruits and storage conditions between February and April 2022. For aflatoxin analysis, the samples were taken from the nuts and dried fruits consumed by the pregnant women. The total aflatoxin content in the samples was analyzed by the ELISA method with a quantitative aflatoxin highsensitivity test kit. Results: The most commonly consumed nuts by pregnant women during pregnancy; were walnuts and hazelnuts, respectively. It was determined that pregnant women buy nuts and dried fruits first from the nut shop and second from the market. Regarding storage preferences, it was seen that the participants mostly stored the nuts in the closed kitchen cabinet or refrigerator cabinet. For the storage material preferences, glass, porcelain, and packaging were preferred. For aflatoxin results, no detectable level of aflatoxin was found in 35 samples, and the presence of aflatoxin in 3 samples (1.43 ppb, 1.523 ppb, and 1.804 ppb, respectively) was detected. Conclusion: Nuts and dried fruit consumption preferences of pregnant women differed according to the products. No aflatoxin was found in the nuts and dried fruits at a level that could threaten the health of pregnant women.

Keywords: Aflatoxin, Nuts, Dried Fruits, Pregnancy, ELISA method

#### **ARTICLE INFORMATION**

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# **1** Introduction

Adequate and balanced nutrition is very important for sustaining life, physical growth and development, mental development, and maintaining health from infancy to old age. The role of nutrition in pregnancy is well known and it also has an impact on maternal and infant health after birth. Therefore, it is essential to have an adequate and balanced diet during pregnancy<sup>1</sup>. Moderate amounts of dairy products in a healthy diet; should include protein-containing foods such as lean meat, fish, eggs, and legumes, and limited amounts of

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high-fat and sugar-containing foods. Along with the changes that occur with pregnancy, there are important variations in vitamin and mineral levels. Since iron, calcium, magnesium, and zinc levels in the serum decrease during pregnancy, the need for these minerals increases. The need for thiamine, riboflavin, folate, vitamin A, vitamin C, and vitamin D increases, as does the need for energy and protein, which increases during pregnancy<sup>2</sup>. During pregnancy, vitamin E should be taken with food. Foods with the highest amount of vitamin E; vegetable oils, whole grains, nuts (such as hazelnuts, almonds, walnuts), and green leafy vegetables. It is

Total aflatoxin in nuts and dried fruits during pregnancy

important to include iron-rich foods such as red meat, poultry, legumes, dried fruits, molasses, whole grains, and enriched cereal products during pregnancy <sup>3</sup>. Adequate intake of energy and nutrients during pregnancy prevents chronic diseases in the baby that may develop in adulthood. With adequate and balanced nutrition, the risk of premature birth or limited intrauterine fetal growth, late birth, development of preeclampsia, and gestational diabetes can be reduced <sup>4</sup>.

Mold growth and mycotoxin formation in foods cause sensory (organoleptic) disorders such as color, odor, and taste as a result of chemical and enzymatic reactions. In addition, water and dry matter content decrease, and carbohydrates and fats deteriorate. As a result of spoilage, a decrease is observed in the nutritional and energy values of the food, and thus the products lose their commercial value <sup>5</sup>. Especially, aflatoxins cause various toxic effects on human and animal health <sup>6</sup>. Exposure of humans to aflatoxin occurs through the consumption of foods contaminated with aflatoxin 7. Exposure to aflatoxin; various factors such as age, diet, and hepatitis B infection affect the degree of toxicity of aflatoxins<sup>8</sup>. For the baby to start and continue a healthy life, attention should be paid to the foods consumed during pregnancy and the amount of aflatoxin in the foods, their consumption should be limited, or their consumption should be avoided <sup>3</sup>. Aflatoxin can be found mostly in dried fruits, corn and other grains, peanuts, spices, cocoa, unprocessed vegetable oils, figs, coffee beans, and coconut powder.

In recent studies <sup>7, 9, 10</sup>, maternal aflatoxin exposure is known to cause some maternal complications such as maternal anemia, micronutrient deficiencies, increased proinflammatory cytokines, and increased oxidative stress during pregnancy <sup>7</sup>. It has been reported that aflatoxin levels, which are above reliable levels in the body, cause placental insufficiency, negatively affect fetal growth and development, and may cause neonatal jaundice and early childhood growth retardation <sup>7</sup>. It is known that aflatoxin is also present in human cord blood and passes to the developing fetus <sup>8</sup>.

Thus, this study aimed to examine the presence of aflatoxin in nuts and dried fruits consumed during pregnancy and the behaviors of pregnant women toward the preparation, storage, and consumption of nuts and dried fruits.

#### 2 Methods

### 2.1 Study area and design

In this cross-sectional descriptive study, the research population was pregnant women living in the Marmara region; The research sample consists of pregnant women living in two randomly selected provinces (Istanbul and Balikesir cities). A questionnaire was applied to 45 pregnant women selected using the convenience sampling method between February and April 2022, and samples were collected from the nuts and dried fruits consumed by the pregnant women. Inclusion criteria for the study; between the ages of 18 - 45, consuming nuts or dried fruits and not allergic to them, being pregnant, and volunteering. 38 pregnant women were selected by simple random sampling method among 45 pregnant women. The ethics committee approval was obtained from the Marmara University Faculty of Health Sciences Non-Invasive Clinical Studies Ethics Committee on 25/11/2021.

## 2.2 Data and sample collection

A structured questionnaire consisting of 40 questions about the consumption preferences of nuts and dried fruits and their attitudes and behaviors regarding storage conditions was applied to the pregnant women by face-to-face interview method. In the first part of the survey (17 questions), the sociodemographic characteristics of the participants, in the second part (21 questions), the eating habits, in the third part, consumption and purchasing options, and finally in the fourth part, the frequency of consumption of nuts and dried fruits, the consumption amount at one time, storage conditions and changes in consumption during pregnancy were questioned. For aflatoxin analysis, 50 grams of nuts and dried fruit were collected from all pregnant women. Sterile sample bags were used for the collection and storage of samples. Since aflatoxin has a structure affected by light and to avoid any change in the amount of aflatoxin, the samples were brought to the laboratory within two hours in a dark environment and cold chain, protected from light, and stored at -4 °C for two months.

### 2.3 Total aflatoxin analysis

To measure the total aflatoxin content of nuts and dried fruits, a quantitative aflatoxin high-sensitivity test kit (Veratox HS MAX, Neogen) was used with methanolic extraction. Briefly, the samples were ground and homogenized in an industrial water-cooling grinder and mixer. Ten milligrams of each ground sample were mixed in 50 ml methanol (70%) and vortexed for 3 min to homogenize. Then, each mixture was filtered by using a paper filter (Whatman No.1). The pH of the filtrates was measured to check availability (pH 6-8) for analysis. The conjugate of 100 µL was added to 100 µL of each standard (0-1-2-4-8 ppb for standard curve) and sample wells in a 96-well microplate. 100 µL mixture was transferred to the wells with antibody and the microplate was gently shacked back-forth and was incubated at room temperature for 10 min. After incubation, the wells were washed with ultra-distilled water three times and rinsed to dry. The conjugate of 100 µL was added to dried wells. It was incubated at room temperature for 10 min. 100 µL red stop solution was added to each well for colorization. The microplate was read

by a microplate reading spectrophotometer (Byonoy Absorbance 96, Gmbh, Germany) at 650 nm.

#### Table 1. Sociodemographic characteristics of pregnant

	x ± sd	Min-Max Value
Pregnancy week	23.03 ± 9.624	7 – 39
Age at marriage	24.61 ± 3.018	17 - 30
Number of pregnancies	1.63 ± 0.913	1 – 5
1 0	Number (n)	Percentage (%)
Age (year)		_
- 18 - 25	7	18.4
- 26 - 30	17	44.7
- 31 - 35	12	31.6
- 36 - 40	2	5.3
- 41 - 45	0	0.0
Breastfed Baby		
- Yes	1	2.6
- No	37	97.4
Educational Level		
- Illiterate	0	0.0
- Literate	0	0.0
- Primary school	1	2.6
- Middle school	2	5.3
- High school	7	18.4
- University	22	57.9
- Master's / Doctorate	6	15.8
Employment Status		
- Employed	26	68.4
- Unemployed	12	31.6
Occupation		
- Government employee	9	23.7
- Worker	5	13.2
- Self-employment	1	2.6
- Housewife	8	21.1
- Retired	0	0.0
- Other	15	39.5

### 2.4 Data analysis

Data were analyzed by SPSS statistic software (SPSS 15, IBM, USA). The normality and homogeneity of variance were controlled by Kolmogorov-Smirnov, Shapiro-Wilk, and Levene's Test. Data regarding descriptive statistics are shown in tables as numbers, percentages, mean and standard deviations. The total aflatoxin content of food samples was calculated by the standard curve generated with the linear regression between the dilutions of aflatoxin standard and the optic density values (OD650, nm) in Office Excel 2016 (Microsoft, USA).

### **3 Results**

Pregnant women (n = 38) who participated in the study were between the ages of 18 - 45 (mean  $29.0 \pm 4.39$  years) and between 7 - 39 (mean  $23.0 \pm 9.6$  weeks) gestational weeks of their pregnancy as given in Table 1. More than half of the pregnant women (57.9%) graduated from university. 23.6% of the pregnant women belonged to various occupational groups such as civil servants (23.6%), workers (13.1%), self-employed (2.6%), housewives (21%), and others (39.4%).

#### Table 2. Nutritional habits of pregnant

	xī± sd	Min-Max Value
Number of main meals consumed per day (n=38)	2.82 ± 0.393	2-3
Number of snacks consumed per day (n=32)	2.16 ± 0.767	1-4
	Number (n)	Percentage (%)
Meal Skipping	(/	(70)
- Yes	1	2.6
- No	18	47.4
- Sometimes	19	50.0
Most Frequently Skipped Meal		
- Morning	4	18.2
- Noon	17	77.3
- Evening	1	4.5
Reason for Skipping Meals		
- Nausea	3	7.9
- Lack of appetite	6	15.8
- I don't have a habit	4	10.5
- Lack of time	4	10.5
- Other	5	13.2
Snack Consumption		
- Yes	29	76.3
- No	5	13.2
- Sometimes	4	10.5
Most Consumed Nutrients in Snacks		
- Fruit	33	86.8
- Dried fruit	10	26.3
- Fruit juice	5	13.2
- Nuts	28	73.7
- Cake-biscuit	7	18.4
- Chocolate	6	15.8
- Milk	14	36.8
- Yogurt	12	31.6
- Ayran	4	10.5
- Bread/sandwich	4	10.5
Special Nutrition Program		
During Pregnancy - Yes	4	10.5
- res - No	4 34	10.5 89.5
Change in diet during pregnancy	54	07.5
- Quantity increased	19	50.0
- Amount decreased	6	15.8
- Hasn't changed	13	34.2
Food allergy		5 1.2
- Yes	4	10.5
- No	34	89.5
Nuts consumption during		
pregnancy - Yes	37	97.4
- No	37 1	2.6
Meals in which nuts are frequently consumed		

- Breakfast	2	5.3
- Mid-morning	11	28.9
- Noon	4	10.5
- Afternoon	14	36.8
- Evening	8	21.1
- Night	7	18.4
Dried fruit consumption during		
pregnancy		
- Yes	31	81.6
- No	7	18.4
Meals in which dried fruits are		
frequently consumed		
- Breakfast	1	2.6
- Mid-morning	7	18.4
- Noon	4	10.5
- Afternoon	13	34.2
- Evening	3	7.9
- Night	7	18.4

While 81.6% of the pregnant women who participated in the study consumed 3 main meals, 18.6% stated that they consumed 2 main meals. 47.4% stated that they never skip meals, 50% sometimes skip meals, and 2.6% always skip meals. It was observed that 18.2% of the pregnant women who skipped meals skipped their morning meal, 77.3% their lunch meal, and 4.5% their evening meal. Considering the reasons for skipping meals in pregnant women, it was determined that loss of appetite was the most common cause of 27.3%, and nausea was the least common cause of skipping meals with a frequency of 13.6% (Table 2).

76.3% of the pregnant women who participated in the study consumed snacks, 10.5% reported that they sometimes consumed snacks and 13.2% reported that they did not consume any snacks. In snacks, 86.8% of pregnant women were fruit, 26.3% dried fruit, 13.2% fruit juice, 73.7% nuts, 18.4% cake, 15.8% I prefer to consume chocolate, 36.8% milk, 31.6% yogurt, 10.5% ayran (beverage with yogurt), 10.5% bread/sandwich. 50% of the pregnant women stated that the amount of food intake increased during pregnancy, 34.2% stated that the amount did not change, and 15.8% stated that the amount of food intake decreased. 10.5% of pregnant women had food allergies. While 97.4% of pregnant women consumed nuts, 81.6% of them consumed dried fruit. Nuts (36.8%) and dried fruits (34.2%) were mostly consumed in the afternoon snack (Table 2).

According to the statement of pregnant women for the nut, and dried fruit consumption preference; 78.9% of them preferred to consume dried fruits, 50% of them preferred to consume dried fruit, and 44.7% of them preferred to consume both nuts and dried fruit. Nuts were often purchased from nut-dry fruit shops (63.3%), markets (56.7%), outdoor bazaars (10.0%), production sites (3.3%), and other places (3.3); dry fruits were purchased from nut-dry fruit shops (68.4%), market (36.8%), outdoor bazaar (5.3%), production site (5.3%) and other places (5.3%) (Table 3).

Pregnant women consumed frequently nuts in the form of crusty (20.0%), hulled (63.3%), or mixed (16.7%). Dried fruits were consumed in the form of crusty (5.3%), hulled (84.2%), or mixed (10.5%). Nuts consumed by pregnant women were examined in a process before consumption. Moreover, 26.3 of them did not process dried fruits before consumption, while washed (63.2%), rubbed/wiped (5.3), and processed differently (5.3%) (Table 3).

**Table 3.** Purchase and consumption preferences ofpregnancies nuts and dried fruits

	Nuts n (%)	Dried Fruits n (%)	Mixed n (%)
Consumption preference			
- Yes	30 (78.9)	19 (50.0)	17 (44.7)
- No	8 (21.1)	19 (50.0)	21 (55.3)
Place of purchase			
- Market	17 (56.7)	7 (36.8)	11 (64.7)
- Nuts Shop	19 (63.3)	13 (68.4)	11 (64.7)
- Market place	3 (10.0)	1 (5.3)	1 (5.9)
- Production place	1 (3.3)	1 (5.3)	0 (0.0)
- Other	1 (3.3)	1 (5.3)	0 (0.0)
Consumption type			
- Crusted	6 (20.0)	1 (5.3)	4 (23.5)
- Shelled	19 (63.3)	16 (84.2)	9 (52.9)
- Both of them	5 (16.7)	2 (10.5)	4 (23.5)
Process applied			
before consumption			
- Nothing	25 (83.3)	5 (26.3)	8 (47.1)
- Washing	1 (3.3)	12 (63.2)	7 (41.2)
- Rubbing/wiping	3 (10.0)	1 (5.3)	4 (23.5)
- Other	0 (0.0)	1 (5.3)	0 (0.0)

As pregnant women did commonly not consume moldy and rotten foods, a few of them consumed after throwing away the rotten part of foods (5.3%) or if tasted/smelled good (2.6%) (Table 4).

When the dried nut consumption frequency of the participants is examined; All of the pregnant women consumed walnuts at least 1-3 times a month. 36.8%, 28.9%, and 39.5% of the participants never consumed peanuts, almonds, pistachios, and cashews respectively. 7.9% of pregnant women stated to consume peanuts, pistachios, and cashews once a day. Overall, pregnant women in this study stated to regularly take care of the consumption of nuts such as walnuts (63.1%) and hazelnuts (34.2%) (Table 5). The highest amount of consumption was for sunflower seeds with an average of 42.1 g at one time. The lowest consumption amount was determined for cashew nuts (14 g) and peanuts (15.8 g).

	Number	Percentage
	(n)	(%)
Process applied to		
moldy/deteriorated foods		
- Throw it all away	36	94.7
<ul> <li>Stripping off the moldy part</li> </ul>	2	5.3
- Wash and consume	0	0.0
- Consume without throwing	0	0.0
- Other	0	0.0
Process applied to packaged		
products with expiry date		
- Do not consume after the expiry date	37	97.4
- Consume if 1-2 weeks have passed after the expiry date	0	0.0
- Consume if 4-7 days have passed after the expiry date	0	0.0
- Ignore it and consume it if it tastes and smells good	1	2.6

**Table 4.** Process applied by pregnant to moldy/deteriorated and expired products

Considering the purchasing preferences, the frequency of participants who preferred unpacked was 57.9%, while the frequency of preference for packaged was 42.1% for walnut. 66.7% of pregnant women prefer to buy packaged peanuts (Table 5). For the storage preferences of the nuts, at least 57.6% of the participants kept them in a cool and dry closed kitchen cabinet. None of the participants preferred to store in balconies, open areas in the house, or freezers. After the closed kitchen cabinet, the most preferred storage area has been the refrigerator. Generally, participants did not prefer to consume the nuts immediately (Table 5).

### 3.1 Presence of total aflatoxin

Total aflatoxin contents of the samples (n=38) collected from pregnant women were calculated with regards to the standard curve by linear regression analysis (Total aflatoxin, ppm = (OD650 – 0.163) / (-0,0087), R<sup>2</sup> =0.9648) (Figure 1). Except for three samples, aflatoxin was not found in 35 nuts and dry fruit samples with the minimum detectable level. In three samples, the aflatoxin was determined as 1.430 ppb (walnuts), 1.523 ppb (mixture of peanuts, walnuts, hazelnuts, raisins), and 1.804 ppb (mixture of almonds, peanuts, raisins, dates) (Figure 1).

### **4** Discussion

Jardi et al., (2019) reported that Spanish pregnant women consumed an average of 2.8 g of nuts per day according to the food consumption frequency questionnaire in Spain<sup>11</sup>. Also, Gignac et al., (2019) determined that a total of 2208 pregnant women consume approximately 6 g of nuts per day in four regions of Spain <sup>12</sup>. In Türkiye, 32.2% and 48.3% of 174 pregnant women increased their consumption of dried fruits and nuts respectively. Some (0.6-1.1%) of them gave up dried fruits and nuts consumption during pregnancy <sup>13</sup>. In the study with 120 Romanian pregnant women (18-44 age old), 93% of them declared consuming nuts while 58% of them consumed dried fruits. The consumption frequencies of nuts and dried fruits were mostly 1-3 times a month with 37% and 39% of pregnant women respectively 14. In a recent study with fifty Turkish healthy pregnant women and fifty with gestational diabetes, 39% and 9% of all participants daily consumed nuts and dried fruits. More than 50% of all women declared not consuming dried fruits during pregnancy. While comparing the participants, the daily intake of nuts was higher in pregnant women with gestational diabetes and the daily intake of dried fruits was higher in healthy pregnant women <sup>15</sup>. In this study, the pregnant women started consuming nuts and dried fruits at the rate of 97.4% and 81.6% respectively. Most of them declared an increase in the consumption amount of nuts and dried fruits during their pregnancy, specifically walnut (52.6%) and dried apricot (21.1%).

According to the legislation of the European Union and Turkish Food Codex, there are maximum limits (MLs) for mycotoxins in many foodstuffs including nuts and dried fruits. The MLs of total aflatoxin (sum of B1, B2, G1, and G2) are 4  $\mu$ g/kg in peanuts, oilseeds, and dried fruits and 10  $\mu$ g/kg in hazelnuts, almonds, and pistachios for direct human consumption according to European Commission Regulation (EC) No 1881/2006 (EC 2023) <sup>16</sup>. In the Turkish Food Codex, the MLs of total aflatoxin (sum of B1, B2, G1, and G2) are 10  $\mu$ g/kg in all nuts and dried fruits for direct human consumption <sup>17</sup>.

In 2004-2005, Garipoglu (2006) determined that the samples of peanut, almond, pistachios, and hazelnut contained the aflatoxins below the MLs following the codexes of Europe and Türkiye. But the total aflatoxin exceeded the MLs in hazelnut samples at the rate of 6.06% (8 ppb) and 15% (4 ppb) <sup>18</sup>.

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Table 5. Food Consumption Frequency and Storage Conditions

	Walnut I	Walnut Hazelnut Peanut	Peanut	Almond	Almond Pistachio	Cashew I nut	Cashew Pumpkin Roasted nut seeds chickpea			Raisins	Dried apricots	Dried fig N	Dried Mulberry	Dried plum	Bluebe-	Cranbe- rıy	Date
	n (%)	u (%)	u (%)	n (%)	n (%)	n (%)	(%) u	(%) u	n (%)	0%) u	n (%)	(%) u	u (%)	n (%)	0%) u	n (%)	(%) u
Consumption Frequency																	
- Once a day	20 (52.6) 12 (31.6)		3 (7.9)	6 (15.8)	3 (7.9)	3 (7.9)	0 (0.0)	1 (2.6)	3 (7.9)	4 (10.5)	8 (21.1)	7 (18.4)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	5 (13.2)
- Several times a day	4 (10.5)	1 (2.6)	1 (2.6)	3 (7.9)	0 (0.0)	0 (0.0)	1 (2.6)	2 (5.3)	2 (5.3)	6 (15.8)	3 (7.9)	1 (2.6)	1 (2.6)	0 (0.0)	0 (0.0)	0 (0.0) 0	4 (10.5)
- 1 time per week	3 (7.9)	4 (10.5) 4 (10.5)		4 (10.5)	7 (18.4)	2 (5.3)	2 (5.3)	3 (7.9)	4 (10.5)	6 (15.8)	8 (21.1)	5 (13.2)	2 (5.3)	2 (5.3)	0 (0.0)	0 (0.0)	1 (2.6)
- 2-3 times a week	4 (10.5)	6 (15.8)	4 (10.5)	4 (10.5)	5 (13.2)	3 (7.9)	1 (2.6)	3 (7.9)	3 (7.9)	2 (5.3)	2 (5.3)	3 (7.9)	1 (2.6)	1 (2.6)	0 (0.0)	0 (0.0)	4 (10.5)
- 3-4 times a week	3 (7.9)	2 (5.3)	1 (2.6)	2 (5.3)	0 (0.0)	2 (5.3)	1 (2.6)	1 (2.6)	0 (0.0)	1 (2.6)	0 (0.0)	0 (0.0)	2 (5.3)	2 (5.3)	0 (0.0)	0 (0.0)	3 (7.9)
- 1-3 times a month	4 (10.5) 6 (15.8) 4 (10.5)	6 (15.8)		5 (13.2)	7 (18.4)	8 (21.1) 4 (10.5)	4 (10.5)	3 (7.9)	5 (13.2)	6 (15.8)	6 (15.8)	3 (7.9)	1 (2.6)	1 (2.6)	0 (0.0)	0 (0.0)	2 (5.3)
- Less than 1 per month	0 (0.0)	2 (5.3)	7 (18.4)	3 (7.9)	1 (2.6)	5 (13.2)	12 (31.6)	5 (13.2)	5 (13.2)	2 (5.3)	1 (2.6)	4 (10.5)	5 (13.2)	4 (10.5)	6 (15.8)	3 (7.9)	3 (7.9)
- Never	0 (0.0)	5 (13.2) 14 (36.8)		11 (28.9)	15 (39.5)	15 (39.5)	17 (44.7)2	11 (28.9) 15 (39.5) 15 (39.5) 17 (44.7) 20 (52.6) 16 (42.1) 11 (28.9) 10 (26.3) 15 (39.5) 26 (68.4) 28 (73.7) 32 (84.2) 35 (92.1) 16 (42.1) 16 (	16 (42.1)	(1 (28.9)	10 (26.3)	15 (39.5)2	26 (68.4)	28 (73.7)	32 (84.2)3	5 (92.1)	6 (42.1)
Amount of Consumption in One Time (g)																	
- $\bar{x} \pm sd$	22.7+8.9 23.1±9.5 15.8±8.9	3.1±9.5	15.8±8.9	18.4±8.0	18.8±5.9	$(8.4\pm8.0 \ 18.8\pm5.9 \ 14.0+8.0 \ 32.8\pm27.0 \ 0 \ 0 \ 0 \ 0 \ 0 \ 0 \ 0 \ 0 \ 0 \$	32.8±27.2 0	$\begin{array}{c} 23.3\pm12, \left  42.1\pm29, \\ 7 \right  \\ 4 \\ 8 \\ 8 \\ 8 \\ 8 \\ 8 \\ 8 \\ 8 \\ 8 \\ 8$	42.1±29.	20.9±9.7	24.1±16. 4	23.1±9.3	13.1±9.4	17.8±13. 8	$\begin{array}{c c} 12.7 \pm 11. \\ 5 \\ 4 \\ 4 \end{array} \left  \begin{array}{c} 18.3 \pm 14. \\ 22.6 \pm 8.7 \\ 4 \end{array} \right $	$\frac{8.3\pm14}{4}$	22.6±8.7
- Minimum-Maximum	10-50	10-50	3-35	7-35	7-35	3-35	8-100	5-50	10-100	10-50	10-100	10-50	5-35	10-50	3-35	10-35	5-40
Way of Purchasing																	
- Packed	16(42.1) 19(57.6) 16(66.7)	9 (57.6)		17 (63.0)	13 (56.5)	15 (65.2)	11 (52.4)	17 (63.0) 13 (56.5) 15 (65.2) 11 (52.4) 10 (55.6) 16 (72.7) 13 (48.1) 11 (39.3) 8 (34.8)	16 (72.7)	13 (48.1)	11 (39.3)		8 (66.7) 7 (70.0)		6 (100)	3 (100) 12 (54.5)	2 (54.5)

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- Unpacked	22 (57.9)	22 (57.9)14 (42.4) 8 (33.3)	8 (33.3)	10 (37.0)	0 (37.0) 10 (43.5)	8 (34.8)	10 (47.6)	8 (44.4)	6 (27.3)	14 (51.9)	17 (60.7)		4 (33.3)	3 (30.0)	0 (0.0)	0 (0.0)	10 (45.5)
Storage place																	
- I consume right away	1 (2.6)	2 (6.1)	1 (4.2)	0 (0.0)	1 (4.3)	2 (8.7)	0 (0.0)	1 (5.6)	1 (4.5)	1 (3.7)	1 (3.6)	1 (4.3)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
- Refrigerator	11 (28.9)	11 (28.9) 9 (27.3) 4 (16.7)		6 (22.2)	3 (13.0)	3 (13.0)	3 (14.3)	4 (22.2)	4 (18.2)	8 (29.6)	11 (39.3)	9 (39.1)	4 (33.3)	3 (30.0)	3 (50.0)	0 (0.0)	11 (50.0)
- Deep freezer	0 (0.0)	0 (0.0)	1 (4.2)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
- Closed in the kitchen cabinet	22 (57.9)	22 (57.9) 19 (57.6) 16 (66.7) 1	16 (66.7)	18 (66.7)	8 (66.7)16 (69.6)15 (65.2)16 (76.2)11 (61.1)15 (68.2)16 (59.3)14 (50.0)11 (47.8) 6 (50.0)	15 (65.2)1	l 6 (76.2)	11 (61.1)	15 (68.2)	16 (59.3)	14 (50.0)	11 (47.8)		5 (50.0)	2 (33.3)	2 (66.7)	9 (40.9)
- On the open balcony	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
- On the covered balcony	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
- In the pantry	4 (10.5)	3 (9.1)	2 (8.3)	3 (11.1)	3 (13.0)	3 (13.0)	2 (9.5)	2 (11.1)	2 (9.1)	2 (7.4)	2 (7.1)	2 (8.7)	2 (16.7)	2 (20.0)	1 (16.7)	1 (33.3)	2 (9.1)
- At home outside the cupboard	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
Stored Material																	
- Own packaging	11 (28.9)	11 (28.9) 10 (30.3) 9 (37.5)	9 (37.5)	10 (37.0)	8 (34.8)	7 (30.4)	9 (42.9)	7 (38.9)	8 (36.4)	7 (25.9)	9 (32.1)	6 (26.1)	3 (25.0)	2 (20.0)	1 (16.7)	0 (0.0)	6 (27.3)
- Plastic container	4 (10.5)	3 (9.1)	3 (12.5)	2 (7.4)	2 (8.7)	2 (8.7)	1 (4.8)	1 (5.6)	2 (9.1)	1 (3.7)	2 (7.1)	3 (13.0)	1 (8.3)	1 (10.0)	2 (33.3)	1 (33.3)	1 (4.5)
- Plastic bag	2 (5.3)	2 (6.1)	1 (4.2)	2 (7.4)	2 (8.7)	2 (8.7)	4 (19.0)	3 (16.7)	5 (22.7)	6 (22.2)	4 (14.3)	5 (21.7)	2 (16.7)	2 (20.0)	1 (16.7)	1 (33.3)	3 (13.6)
- Cloth bag	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	1 (4.3)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
- Sack	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)

- Paper bag	0 (0.0)	0 (0.0) 0 (0.0) 0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	3 (0.0)     0 (0.0)     0 (0.0)     0 (0.0)     0 (0.0)     0 (0.0)     0 (0.0)     0 (0.0)     0 (0.0)     0 (0.0)     0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
- Glass, porcelain	21 (55.3)	18 (54.5)	21 (55.3) 18 (54.5) 11 (45.8) 13 (48.1) 11 (47.8) 1 (47.8) 7 (33.3) 7 (38.9) 7 (38.9) 7 (31.8) 13 (48.1) 13 (46.4) 9 (39.1) 6 (50.0) 5 (50.0) 2 (33.3) 1 (33.3) 12 (54.5)	13 (48.1)	11 (47.8)	11 (47.8)	7 (33.3)	7 (38.9)	7 (31.8)	13 (48.1)	13 (46.4)	9 (39.1)	6 (50.0)	5 (50.0)	2 (33.3)	1 (33.3)	12 (54.5)
- Tin box	0 (0.0)	0 (0.0) 0 (0.0) 0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0) 0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0) 0 (0.0) 0 (0.0) 0 (0.0) 0 (0.0) 0 (0.0) 0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0.0) 0 (0.0) 0		0 (0.0)
- Clay pot	0 (0.0)	0 (0.0) 0 (0.0) 0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0) 0 (0.0)	0 (0.0)	0.0) 0	0 (0.0)	0 (0.0)	0 (0.0) 0 (0.0) 0 (0.0) 0 (0.0) 0 (0.0) 0 (0.0) 0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	(0.0) 0 (0.0) 0		0 (0.0)
- Wrapped in aluminum foil		0 (0.0) 0 (0.0) 0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0) 0 (0.0)	0 (0.0)	0.0) 0	0 (0.0)	0 (0.0)	0 (0.0) 0 (0.0) 0 (0.0) 0 (0.0) 0 (0.0) 0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0) 0 (0.0)		0 (0.0)
- Wrapped in ding film 0 (0.0) 0 (0.0) 0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	(0.0) 0 (0.0) 0 (0.0)	0 (0.0)	0 (0.0) 0 (0.0) 0 (0.0) 0 (0.0) 0 (0.0) 0 (0.0) 0 (0.0) 0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0) 0 (0.0)	0 (0.0)	0 (0.0)
Consumption Status in Pregnancy																	

20 (52.6) 16 (48.5) 10 (41.7) 13 (48.1) 11 (47.8) 9 (39.1) 6 (28.6) 8 (44.4) 9 (40.9) 13 (48.1) 17 (60.7) 16 (69.6) 5 (41.7) 4 (40.0) 2 (33.3) 1 (33.3) 15 (68.2)

2 (9.1)

1 (33.3)

0 (0.0)

1(10.0)

1 (8.3)

2 (8.7)

2 (7.1)

3(11.1)

1 (4.5)

1 (5.6)

1 (4.8)

1 (4.3)

1 (4.3)

2 (7.4)

5 (15.2) 1 (4.2)

5 (13.2)

Decreased

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Increased

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Hasn't changed

1

1 (22.7)

1 (33.3)

4 (66.7)

5 (50.0)

6 (50.0)

5 (21.7)

(3.(34.2)|12.(36.4)|13.(54.2)|12.(44.4)|10.(43.5)|13.(56.5)|14.(66.7)|9.(50.0)|12.(54.5)|11.(40.7)|9.(32.1)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40.7)|11.(40

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Kanık (2018) chromatographically analyzed a total of 80 almond samples from retail markets in Türkiye and measured the aflatoxins (0.118 - 0.508 ppb) in 15% of almond samples <sup>19</sup>. Uğur (2021) suggested that there are aflatoxins in most (95-100%) peanut, hazelnut, dried grape, and dried fig samples from several cities of Türkiye. The total aflatoxin measurement was higher in processed (drying, shelling, and roasting) nuts than in raw nuts samples and also higher in raisin samples than in dried figs. However, the aflatoxin contents in all 140 samples (except one) were below the MLs (4, 8, and 10 µg/kg) of aflatoxins for direct human consumption. Therefore, a significant difference was not found between packaged and unpackaged samples. So, It was suggested that there was no health risk for adult consumers according to the estimated tolerable daily intake (0.001-0.022 ng/kg v.a./day) value and margin of exposure <sup>20</sup>. In this study, the total aflatoxin content of nuts and dried fruit consumed by the pregnant did not exceed the legal limits.

The study on 1675 mothers and infants from pregnancy through 2 years of age presents the presence of aflatoxins (especially AFB1 and AFM1) in their blood and breast milk and the significant correlation between the aflatoxin exposure and child body growth parameters <sup>21</sup>.

The present study has some limitations, including the pregnant women's self-report data. The generalizability of our findings is limited because this is a small sample size. However, we also investigated the amount of aflatoxin in nuts and dried fruits consumed as well as the consumption habits of these pregnant women comprehensively using objective measures.

In conclusion, despite the low aflatoxin presence, and below the legal limit, increasing daily nuts and dried fruit intake might raise aflatoxin exposure during pregnancy and breastfeeding.

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Authors' Contribution: Ş.A, S.K, and B.İ.O.K conceived and designed the study, Ş.A, T.C., B.Ç., and M.N.G. undertook the literature research. All authors participated in the experiment and data acquisition. Ş.A, S.K, and B.İ.O.K performed the data analysis. Ş.A, T.C., B.Ç., and M.N.G carried out the statistical analysis, prepared, reviewed, and all authors drafted the manuscript. All authors approved the final version before submission. All authors have read and agreed to the published version of the manuscript.

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