



Estimation of Nitrate Contents in Sixteen Varieties of Libyan Dates (*Phoenix dactylifera* L.) and Their Potential Health Risks

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ABSTRACT

Background: Dates play an essential role in the Libyan diet, particularly during Ramadan. As a result, there is considerable emphasis on ensuring that these dates are free from contaminants, especially nitrates, which are prevalent pollutants in food items. Therefore, this study aimed to evaluate the nitrate contents and the potential health risks associated with their consumption by adults and teenagers in various date varieties commonly consumed in Libya.

Methods: A total of sixteen date varieties, mainly gathered from southern Libya, were tested. The findings reveal that the average nitrate content in the date samples ranged between 498.66±135.35 mg/kg in the Tafsert variety and 1147.02±287.97 mg/kg in the Tagayat variety. To determine the potential health risks and safety of these dates, we assessed the Estimated Daily Intake (EDI), Acceptable Daily Intake (ADI) and Hazard Quotient (HQ).

Result: The results indicate that the EDI, ADI and HQ values remained below the thresholds established by the Joint FAO/WHO Expert Committee on Food Additives (JECFA). The EDI for nitrates was observed to range from 0.34 to 0.77 mg/kg body weight (bw)/day for adults and from 0.45 to 1.04 mg/kg bw/day for teenagers, while the acceptable limit is defined at 3.70 mg/kg bw/day. However, the findings also suggest that teenagers are more susceptible to nitrate levels in dates than adults. Therefore, the amount of dates consumed does not represent a risk to consumers. Additionally, the calculated HQ values for adults and teenagers varied from 0.21 to 0.48 and from 0.28 to 0.65, respectively. Since these HQ values are below 1, they indicate a low level of health risk for consumers.

Key words: Acceptable daily intake, Dates, Estimated daily intake, Hazard quotient, Health risk assessment, Nitrate.

INTRODUCTION

There are approximately 5,000 varieties of date palms worldwide, with 160 million palms cultivated in the Arab region, yielding about 6.6 million tons annually, which accounts for over 77% of global date production (Hammami *et al.*, 2024). Dates are nutritious fruits that are rich in essential vitamins, minerals, dietary fiber and antioxidants. They promote heart health, aid in digestion and help reduce oxidative stress and inflammation. Dates provide important nutrients, including carbohydrates, proteins, fats, vitamins A and C and minerals such as calcium, iron and magnesium (Shahdadi *et al.*, 2015). Most human nitrate exposure comes from plant-based foods, especially vegetables and certain fruits, accounting for 80% to 95% of daily intake (Pardo-Marín *et al.*, 2010; Quijano *et al.*, 2017; Qasemi *et al.*, 2024). Dates also contain antioxidants that may inhibit cancer-causing agents, influencing some countries to relax regulations on vegetable nitrate levels (Jannat *et al.*, 2022). Low levels of nitrate are generally considered safe, with the Joint Expert Committee on Food and Agriculture (JECFA) recommending an acceptable daily intake (ADI) of 0-3.7 mg per kilogram of body weight (Hmeljak and Cencič (2013)). The type of nitrogen significantly affects plant growth, with primary forms including ammonium (NH₄⁺), nitrate (NO₃⁻) and urea. Different plant species exhibit preferences for these sources, influenced by factors such as nitrogen levels and

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soil properties (Dong *et al.*, 2018; Al-Ethawi and Salem 2019). The overuse of chemical fertilizers leads to issues such as soil acidification and groundwater contamination

(Salem *et al.*, 2020; Fashaho *et al.*, 2020). Reports indicate alarming levels of heavy metals and nitrates in crops, often due to the irrigation with partially treated sewage water and excessive fertilizer application (Al-Busaidi *et al.*, 2015; Salehzadeh *et al.*, 2020; Jalali *et al.*, 2021), raising concerns about food safety (El-Nahhal, 2018; Abd-Elrahman *et al.*, 2022). High nitrate levels can be converted into harmful compounds, such as nitrosamines, while nitrite production can interfere with oxygen transport in the blood (Mensinga *et al.*, 2023; Bolaji *et al.*, 2025). Interestingly, nitrates and nitrites may lower blood pressure in healthy women (Ashworth *et al.*, 2015). Date palms, a key fruit crop in arid regions like Libya, contribute significantly to local economies and are cultivated in over 400 varieties (Racchi and Camussi, 2018). Despite extensive research on heavy metal and nitrate contamination in food, there is a limited number of studies on nitrates in palm dates. This study aims to examine the nitrate content in the studied dates and assess the potential health risks associated with their consumption.

MATERIALS AND METHODS

Study area and sample collection

The cultivation of date palms (*Phoenix dactylifera* L.) has grown significantly in Libya, with an estimated four million trees, mainly in the southern Saharan region (Racchi *et al.*, 2014). Ripe dates are harvested by the end of October. In November 2024, samples of sixteen local date varieties, Degla, Asapeer, Tafsert, Taleas, Aarrayag, Aglyen, Ammrayer, Saeyde, Adawy, Sallalow, Akdayer, Tagayat, Apil, Kadray, Majdool and Ajwa, were purchased from the market and brought to the laboratory for analysis.

Preparation of palm date samples

Date palm samples were prepared following the methods of Ajebe and Bahiru (2019) and Zhao and Wang (2017). One hundred grams of each variety were washed, pitted and cut into uniform pieces, then dried in an oven at 105°C for 24 hours. After cooling, the samples were ground and sieved through a 2 mm mesh. Finally, 4.0 grams of each sample were stored in clean plastic bottles for analysis.

Extraction and determination of nitrate in dates

Nitrate extraction was performed as per Lufei and Yong (2017). A 0.1 g date sample was suspended in 10 mL distilled water, heated to 80°C for 1 hour, cooled and filtered through Whatman filter paper No. 4. For nitrate determination, 0.1 mL of the extract was mixed with 0.4 mL salicylic acid solution in a 30 mL tube, allowed to sit for 20 minutes and then 9.5 mL of 2 N NaOH was added. The nitrate concentration was measured immediately using a Jenway 7205 spectrophotometer, following Equation (1).

$$Y = \frac{C \cdot V}{W} \quad \dots 1$$

Where,

Y = Nitrate content (mg/kg dwt).

C = Nitrate concentration from OD410 (abs * slope - intercept) using the calibration curve.

V = Total volume of extracted sample (ml).

W = Weight of sample (g).

Health risk assessment

A study evaluated the health risks associated with date consumption in adults and teenagers, focusing on nitrate levels in various date varieties. The average daily intake was estimated at 110g for adults and 70g for teenagers. The estimated daily intake (EDI), percentage of acceptable daily intake (%ADI) and hazard quotient (HQ) were calculated as per Ram *et al.* (2022).

Estimated daily intake (EDI)

People can be exposed to pollutants through ingestion, skin contact and inhalation; however, for nitrates, the primary exposure route is oral intake (Qasemi *et al.*, 2024). Excessive nitrate consumption from palm dates can pose health risks if it exceeds the Acceptable Daily Intake (ADI) of 3.7 mg per kilogram of body weight, as set by the European Food Safety Authority (EFSA, 2008). Adults consume about 110 g, while teenagers consume around 70 g of palm dates daily. High nitrate intake can lead to serious health problems or fatalities (Dezhangah *et al.*, 2022). We calculated the average daily nitrate intake based on body weight and bioavailability using the Estimated Daily Intake (EDI) equation, which reflects potential ingestion but does not consider metabolic excretion (USEPA, 2004).

$$EDI = \frac{EF \cdot ED \cdot DC \cdot MC}{BW \cdot AT} \quad \dots 2$$

Where:

EDI = Estimated daily intake (mg/kg bw/day).

EF = Exposure frequency (365 days/year).

ED = The exposure duration, which is 70 years for adults and 50 years for teenagers. For teens (7-17 years), the maximum exposure duration is 11 years, while adults over 70 can have a maximum of 30 years (EPA, 1989).

DC indicates daily date consumption: ~110 g/day for Libyan adults (70 kg) and ~70 g/day for teenagers (50 kg), potentially doubling during Ramadan.

MC = Mean nitrate concentration in samples (mg/kg dry weight). BW is the body weight of adult consumers (70 kg) and teenagers (50 kg) (Radfard *et al.*, 2018).

AT = Average Time: 25,550 days for adults and 18,250 days for children (Saha and Zaman, 2013).

Noncarcinogenic hazard quotient (NHQ)

Human health risk assessment, as defined by the U.S. Environmental Protection Agency (USEPA, 2004), estimates the likelihood of adverse health effects from chemical

exposure in contaminated environments. This study conducted a quantitative risk assessment of nitrate exposure from date consumption, utilizing data from the USEPA (2020). The reference dose (RfD) for nitrate is 1.60 mg/kg body weight per day (Gruszecka-Kosowska and Baran, 2017). The Hazard Quotient (HQ) compares individual exposure to the RfD: an HQ > 1 indicates a significant health risk, while HQ < 1 suggests safe exposure levels (Djahed *et al.*, 2018).

Statistical analysis

All experiments were conducted in triplicate and SPSS version 26 was employed to calculate the minimum, maximum, mean and standard deviation of nitrate levels for the dates under study. The Pearson correlation coefficient (*r*) was utilized to assess the linear relationships among the various data varieties. The tested date varieties were categorized based on their nitrate content using Ward's hierarchical clustering analysis. A one-way analysis of variance (ANOVA) was performed to compare the mean nitrate concentrations across the different date varieties. The threshold for statistical significance was set at $p < 0.05$ (Dezhangah *et al.*, 2022).

Validation method

The validation method focused on data types, permissible limits and process performance characteristics, including precision, accuracy, calibration data and the linearity of the calibration curve (Dezhangah *et al.*, 2022). A linear calibration curve was established using nitrate standards from 10 to 120 mg/L, with three replicates per concentration. Deionized water served as a blank sample to determine the Limits of Quantitation (LOQ) and Limits of Detection (LOD) (Mehri *et al.*, 2019).

RESULTS AND DISCUSSION

Calibration curve and validation method

The calibration equation, which links the analyte to nitrate concentration, was used to determine the nitrate content

in palm dates. A calibration curve (Fig 1) was created by measuring seven concentrations of NO_3^- , ranging from 0.0 to 120 mg/L, with samples exceeding this range diluted for reassessment. A multiple linear regression model showed a strong correlation coefficient (R^2) of 0.9953. The limits of detection and quantification for NO_3^- were 6.670 mg/kg and 20.211 mg/kg, respectively. Comparable results were reported by Pardo-Marín *et al.* (2010) with a LOD of 7 mg/kg and a LOQ of 20 mg/kg and by Chung *et al.* (2011) with a LOD of 4 mg/kg and a LOQ of 20 mg/kg.

Nitrate content in palm dates

Table 1 shows the nitrate content of various palm date varieties from Brack City's local market. The mean nitrate levels (mean $\pm$ SD in mg/kg dwt) were as follows: Degla (735.38 $\pm$ 464.67), Asapeer (1012.31 $\pm$ 277.69), Tafsert (498.65 $\pm$ 135.35), Tales (711.27 $\pm$ 173.94), Aarrayag (637.48 $\pm$ 295.77), Aglyen (708.62 $\pm$ 218.36), Ammrayer (695.87 $\pm$ 69.78), Saeyde (734.52 $\pm$ 288.90), Adawy (500.26 $\pm$ 56.41), Sallalow (712.43 $\pm$ 198.29), Akdayer (676.27 $\pm$ 112.36), Tagayat (1085.65 $\pm$ 335.46), Apil (798.53 $\pm$ 378.75), Kadray (627.73 $\pm$ 6.12), Majdool (758.63 $\pm$ 198.25) and Ajwa (812.53 $\pm$ 38.9). Tafsert exhibited the lowest nitrate content, whereas Tagayat had the highest.

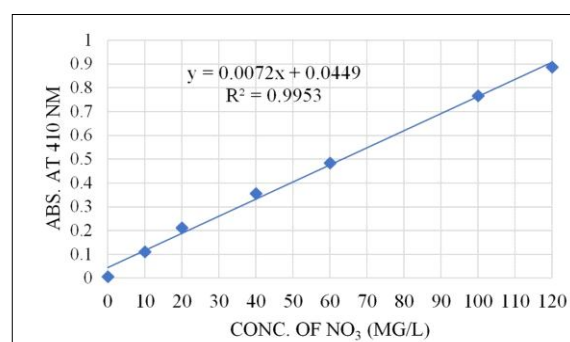


Fig 1: Nitrate standard curve.

Table 1: Descriptive analysis of Conc. of Nitrate in palm dates varieties (local name) (mg/kg. dwt).

Name of the date variety	No.	Min.	Max.	Mean	Std. D
Degla	27	333.5	1901.50	735.38	464.67
Asapeer	15	497.3	1348.50	1012.31	277.69
Tafsert	15	282.4	764.00	498.66	135.35
Taleas	24	375.5	1075.50	705.20	163.03
Aarrayag	13	278.9	1055.20	652.81	288.53
Aglyen	12	509.9	1095.80	723.63	201.18
Ammrayer	12	633.1	771.00	703.74	45.34
Saeyde	15	349.6	1129.40	730.07	283.02
Adawy	9	463.7	643.60	500.26	56.41
Sallalow	12	545.6	798.30	658.01	83.44
Akdayer	12	554.0	934.80	741.43	119.82
Tagayat	18	649.2	1450.00	1147.02	287.97
Apil	12	426.6	1176.30	799.29	311.45
Kadray	12	622.6	701.00	642.32	26.38
Majdool	12	556.1	952.30	789.78	125.51
Ajwa	12	768.2	841.00	811.72	22.22

The reduced nitrate levels in Tafsert are likely due to cultivation in an oasis without irrigation or fertilisers. Additionally, soil properties and cultivation practices significantly influence nitrate content (Alakhdhari *et al.*, 2025; Kaur *et al.*, 2025). The nitrate levels in our samples are moderate (500-1000 mg/kg), according to Hmeljak and Cenciè (2013). Although permissible limits for dates are not specified in the literature, we reference recommendations from Sayed and Rezvan (2014).

Health risk assessment

Estimated daily intakes of nitrate

Some individuals consume palm dates daily, while others do so occasionally; this study focuses on individuals who

consume them daily. The estimated daily nitrate intakes (EDI) for adults (70 kg) and teenagers (50 kg) are outlined in Table 2, showing higher nitrate levels in teenagers. Daily consumption was estimated at 110 g for adults and 70 g for teenagers, with nitrate exposure varying from 132.92 to 305.87 mg/day for adults and 95.63 to 219.97 mg/day for teenagers. The acceptable daily intake of nitrate is 0-3.7 mg/kg body weight/day (Luetic *et al.*, 2023), equating to 259 mg for adults and 185 mg for teenagers. Although Asapeer and Tagayat have high nitrate levels (1012.31 and 1147.0 mg/kg), they remain within JECFA's recommended limits.

Acceptable daily intake (ADI)

Nitrate toxicity can arise from the daily consumption of foods like dates. The Joint FAO/WHO Expert Committee on Food

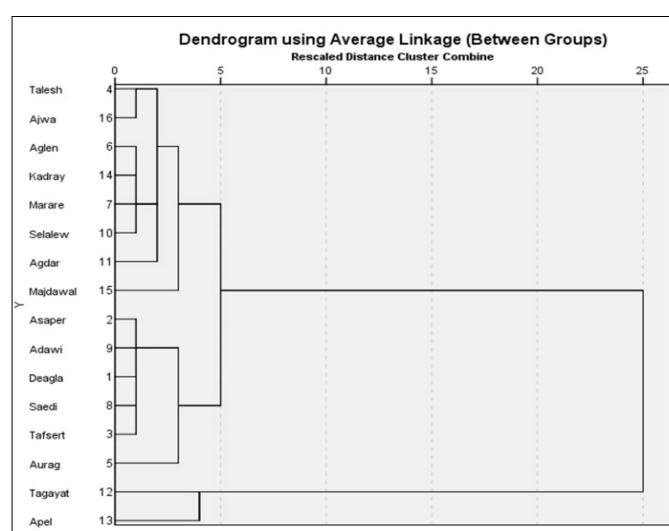


Fig 2: Dendrogram displaying hierarchical clustering for nitrate in date varieties.

Table 2: Estimated daily intake, mean nitrate intake and Hazards quotient of nitrate from consumption dates varieties.

Dates variety	NO ₃ (mg/kg)	DC (g/day)		EDI (mg/kg/bw/day)		ADI (mg/kg/bw/day)		Mean nitrate intake (mg/day/person)		HQ	
		AD	TN	AD	TN	AD	TN	AD	TN	AD	TN
Degla	735.38	110	70	0.50	0.67	0.14	0.18	1.05	1.47	0.31	0.42
Asapeer	1012.31	110	70	0.68	0.92	0.18	0.25	1.45	2.02	0.43	0.57
Tafsert	498.66	110	70	0.34	0.45	0.09	0.12	0.71	1.00	0.21	0.28
Taleas	705.2	110	70	0.47	0.64	0.13	0.17	1.01	1.41	0.30	0.40
Awrayag	652.81	110	70	0.44	0.59	0.12	0.16	0.93	1.31	0.27	0.37
Aglyen	723.63	110	70	0.49	0.66	0.13	0.18	1.03	1.45	0.30	0.41
Amrayer	703.74	110	70	0.47	0.64	0.13	0.17	1.01	1.41	0.30	0.40
Saeyde	730.07	110	70	0.49	0.66	0.13	0.18	1.04	1.46	0.31	0.41
Adawy	500.26	110	70	0.34	0.45	0.09	0.12	0.71	1.00	0.21	0.28
Sallalow	658.01	110	70	0.44	0.60	0.12	0.16	0.94	1.32	0.28	0.37
Akdayer	741.43	110	70	0.50	0.67	0.14	0.18	1.06	1.48	0.31	0.42
Tagayat	1147	110	70	0.77	1.04	0.21	0.28	1.64	2.29	0.48	0.65
Apil	799.29	110	70	0.54	0.72	0.15	0.19	1.14	1.60	0.34	0.45
Kadray	642.32	110	70	0.43	0.58	0.12	0.16	0.92	1.28	0.27	0.36
Majdool	789.78	110	70	0.53	0.72	0.14	0.19	1.13	1.58	0.33	0.45
Ajwa	811.72	110	70	0.55	0.74	0.15	0.2	1.16	1.62	0.34	0.46

DC = Daily consumption; EDI = Estimated daily intake; ADI = Acceptable Daily Intake; AD = Adult; TN = Teenager; HQ = Hazard quotient.

Additives (JECFA) has set an acceptable daily intake (ADI) for nitrates at 3.70 mg/kg body weight per day (EFSA, 2008). The %ADI from date consumption for adults and teenagers ranges from 9.08 - 12.21 and 20.88 - 28.08 for Tafsert and Tagayat, respectively. Mean nitrate intake for adults is 0.71 to 1.64 mg/kg/d and for teenagers, it is 1.0 to 2.29 mg/kg/d. Both are within the acceptable limit. Higher ADI values have been observed in some fruits and vegetables in Iran, Bangladesh and Egypt due to high EDI (Mehri *et al.*, 2019; Sebaei and Refai, 2019; Uddin *et al.*, 2021; Zendeabad *et al.*, 2022).

Noncarcinogenic risk

Several factors influence health risks associated with nitrates, including consumption frequency, inherent risk and individual body weight (Mehri *et al.*, 2019; Ali *et al.*, 2021). Our findings indicate that the Hazard Quotient (HQ) values for all date samples were below 1, suggesting no noncarcinogenic risks associated with date consumption. Specifically, HQ values for nitrates ranged from 0.21 in

Tafsert to 0.46 in Tagayat for adults consuming 110 g/day and from 0.28 to 0.65 for teenagers consuming about 70 g/day. The risk from nitrates may vary based on date type, environmental factors, farming practices and storage conditions (Ghasemidehkordi *et al.*, 2018). Notably, teenagers show higher HQ values than adults, indicating they are more vulnerable to nitrate concentrations in dates. Our results align with findings from Mehri *et al.* (2019), Ali *et al.* (2021) and Qasemi *et al.* (2024).

Statistical analysis

The Pearson correlation coefficient demonstrated a strong association between specific date varieties and their nitrate concentrations, highlighting significant relationships among several of these varieties. These findings will enhance the management of polluted dates, ultimately helping to mitigate the risks they pose to consumers. (Khan *et al.*, 2021). Table 3 depicts Pearson's correlation matrix for nitrate accumulation in the sixteen varieties of dates (Shown in bold). The results show a strong positive

Table 3: Pearson's correlation matrix for nitrate concentrations in dates varieties.

Deagla	Asaper	Tafsert	Talesh	Aurag	Aglen	Saedi	Adawi	Selalew	Agdar	Tagayat	Apel	Kadray	Majdawal	Ajwa	
Deagla	1														
Asaper	.457	1													
Tafsert	-.267	.217	1												
Talesh	-.330	-.324	-.501	1											
Aurag	.798**	.582*	.581*	-.895**	1										
Aglen	.566	.624	.445	-.656	.382	1									
Saedi	.512	.175	.793**	-.944**	.872**	.282	1								
Adawi	-.510	-.573	-.007	.187	-.525	-.387	-.050	1							
Selalew	-.112	1.000**	.765	-.855	-.588	.278	.809	.859	1						
Agdar	.955	-.397	.281	.819	-.503	.767	-.864	-.815	-.403	1					
Tagayat	-.948**	-.947**	-.909*	.921**	-.903*	-.406	-.970**	.556	.118	.862	1				
Apel	-.946**	-.923**	-.899*	.885*	-.891*	-.372	-.962**	.619	.893	.052	.994**	1			
Kadray	.953	-.401	.277	.821	-.500	.765	-.866	-.817	-.406	1.000**	.860	.048	1		
Majdawal	-.852	.610	-.037	-.935	.276	-.587	.961	.932	.615	-.969	-.711	.195	-.970	1	
Ajwa	.311	-.978	-.619	.943	.412	-.077	-.911	-.945	-.979	.580	.086	-.784	.583	-.762	1

*Correlation is significant at the 0.05 level (2-tailed).

**Correlation is significant at the 0.01 level (2-tailed).

Table 4: ANOVA Variance of nitrate against sites, varieties and Hazards quotient.

		Sum of squares	df	Mean square	F	Sig.
Site	Between groups	4516.936	113	39.973	1.595	.051
	Within groups	952.583	38	25.068		
	Total	5469.520	151			
HQ adult	Between groups	.572	113	.005	2.003	.008
	Within groups	.096	38	.003		
	Total	.668	151			
HQ children	Between groups	1.022	113	.009	2.015	.008
	Within groups	.171	38	.004		
	Total	1.193	151			
Varieties	Between groups	2156.603	113	19.085	1.563	.058
	Within groups	463.917	38	12.208		
	Total	2620.520	151			

correlation between Deagla and Aurag ($r = 0.798$), Asaper and Selalew ($r = 1.000$), Tafsert and Saedi ($r = 0.793$), Talesh and Tagayat ($r = 0.921$), Aurag and Saedi ($r = 0.872$), Agder and Kadray ($r = 1.000$) and Tagayat and Apel ($r = 0.994$). A positive correlation was observed between Asaper and Aurag ($r = 0.852$), Tafsert and Aurag ($r = 0.581$) and Talesh and Apel ($r = 0.885$). Whereas, strong negative correlations were observed between Deagla and Tagayat ($r = -0.948$) and between Asaper and Tagayat ($r = -0.923$), respectively and between Deagla and Apel ($r = -0.946$) and between Asaper and Apel ($r = -0.947$), respectively. Talesh, Aurag and Saedi ($r = -0.895$, $r = -0.944$) respectively. Saedi, Tagayat and Apel ($r = -0.970$, $r = -0.962$) respectively. Negative correlation observed only between Tafsert, Tagayat and Apel ($r = -0.909$, $r = -0.899$, respectively). Aurag, Tagayat and Apel ($r = -0.903$, $r = -0.891$). Fig 2 illustrates the hierarchical clustering of nitrate accumulation in dates, revealing three distinct clusters. The first cluster includes Tagayat and Apel. The second cluster comprises Aurag, Tafsert, Saedi, Deegla, Adwai and Asaper. The third cluster consists of Majdawal, Agdar, Salalew, Amrear, Kadray, Aglen, Ajwa and Talesh (Bahadoran *et al.*, 2016). Lower nitrate levels in dates may stem from reduced fertilizer use. Variability in nitrate levels can arise from factors such as soil type, fertilizer application, agricultural practices and groundwater contamination (Mehri *et al.*, 2019). The results of a one-way ANOVA between nitrate, sites, varieties and hazard quotients for children and adults are displayed in Table 4. The results show a significant difference in nitrate concentration and hazard quotient between children and adults, with a difference of 0.008 at $P < 0.05$. At the same time, there is no significant difference in nitrate concentration across sites or varieties.

CONCLUSION

This survey was conducted to evaluate the concentrations of nitrate in selected palm dates and assess the estimated daily intake (EDI) and health risks using the Hazard Quotient (HQ) approach. Data indicated that the mean $\pm$ SD nitrate level ranged from 498.66 ± 135.35 mg/kg in Tafsert to 1147.02 ± 287.97 mg/kg in Tagayat. Based on the results, the nitrate concentration in all samples is lower than the permissible limit suggested. The total EDIs of nitrate through the consumption of dates were lower than the ADI level. HQ values of nitrate by the consumption of dates were < 1 , indicating that health risks associated with nitrate exposure were not significant. Therefore, the intake of nitrate through date consumption could be considered safe for consumers. Therefore, it is essential to sustainably conserve and manage date palm resources to ensure future agricultural and economic stability.

Author's contributions

Mansour Awiadat Salem, Saeda Maatoq Ali Mohamed, Yasser Fathi Nassar . designed the study; Aisha Abdulgader Alshanoky, Masouda Mohamed Alshatory, Samera Musa

Alwaleed. analyzed the data; Mansour Awiadat Salem, Yasser Fathi Nassar wrote the manuscript.

Conflict of interest

The authors declare no conflicts of interest.

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