



NITRATE STATUS IN SOME PLANTS GROWN IN NORTH SINAI SOILS

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ABSTRACT

Examine the nitrate concentration of a few distinct plants from various places in North Sinai Governorate (Arish City). This study is concerned with three main topics: (i) the contribution of various well-known leafy plants to dietary nitrate consumption by people; (ii) the dietary, environmental, and physiological factors regulating nitrate accumulation in plants; and (iii) the adverse and beneficial effects of nitrate on human health. The Locations (Airport Road Location (A), Wadi EL-Arish1(W), and Wadi EL-Arish 2(WI)) include this district. Rosemary, Breckl and thyme, Mentha, Black Mulberry, Jute mallow, Alfalfa, Basil, Maize green chop, Kurrat, Pursley, Arugula, and Horse mint were among the 12 varieties of plants represented by the total of 19 plants that were randomly selected from farms in three distinct regions (Arish City). The results revealed that the plant has a significant impact on the nitrate concentration, with Arugula having the greatest nitrate content while Kurrat and Horse mint having the lowest. The plants has an impact on the nitrate content. The nitrate concentrations of the plants studied exhibit a considerable locational effect. The outcomes of the investigation showed that the amounts of nitrate content were appropriate. To elaborate these findings with more samples and diverse plants, however, more work is required.

INTRODUCTION

Along with nitrite, nitrate is one of the main groundwater contaminants in the world. It is an ion generated when nitrogen is oxidized (NO₂⁻). According to (Revilla *et al.*, 2020), vegetables are essential for both human and animal nutrition since they are a superb source of vitamins, minerals, and physiologically active chemicals. Leafy vegetables are frequently eaten either fresh or cooked. The consumption of vegetables is thought to be directly responsible for about 87% of the total nitrate concentration in a typical diet. The ionic form of nitrate is an additional source of nitrogen, a crucial plant nutrient. The maximum contamination limit (MCL) for nitrate as nitrate-nitrogen in public drinking water systems in the United

States (U.S.) is 10 mg/L. (NO₃⁻N). The World Health Organization's (WHO) recommended limit of 50 mg/L of NO₃ or 11.3 mg/L of NO₃⁻N (NO₃ mg/L multiplied by 0.2258) is nearly comparable to this concentration (Al-Bhar and Al-Saffawi, 2021). Other health effects, such as cancer and poor reproductive outcomes, were not considered. Nitrate is a precursor in the formation of N-nitrous compounds (NOC) *via* endogenous nitration, the majority of which are carcinogens and teratogens. The MCL was designed to guard against infant methemoglobinemia. As a result, exposure to NOC produced after ingesting nitrate from dietary and drinking water sources may result in cancer, birth abnormalities, or other negative health effects. Nitrate is present in a wide variety of foods, with some green leafy and root

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vegetables having the highest concentrations. **Ward *et al.* (2018)**, According to FAO/WHO, the acceptable daily intake (ADI) of nitrate is 3.7 mg/kg/day from all sources, including drinking water, vegetables, and food (**Kiani *et al.*, 2022**). One of the most significant challenges for public awareness nowadays is pollution. The danger of toxic chemicals getting into crops and water due to the overuse of pesticides and fertilizers in agriculture (**Afaf, 2006**). Nitrate buildup in plants is influenced by its metabolism and uptake. Nitrate reductase (NR) and other nitrogen metabolism enzymes in plant leaves convey the majority of the root-absorbed nitrates to the shoots where they are digested. Numerous internal cues (such as the expression of relevant genes and enzyme activity) as well as external environmental factors control the absorption, assimilation, and transport of nitrates in plants. Nitrate content in vegetables can be decreased by the manipulation of the growing environment, which will encourage the conversion of nitrate into organic nitrogen molecules in addition to better fertilization management tactics (**Zhonghua *et al.*, 2020**).

Numerous crops produce nitrate that builds up. Livestock may be killed by plants with more than 1.5 percent dry weight of nitrate (as KNO_3). When livestock consume feed that contains between 0.5 and 1.5 percent nitrate, sub-lethal consequences may result. Animals using nitrate fertilizers, machine oil, and some naturally occurring well and pond waters are also susceptible to nitrate toxicity. The capacity of nitrate to accumulate in plants varies. The amount of nitrate in many of these plants is influenced by the kind of soil, the availability and form of nitrogen contained in the soil, numerous environmental conditions, and chemical or physical plant injury. For instance, nitrate-accumulating plants may develop an excessive nitrate buildup as a result of drought, frost, or 2,4-D treatment. While the seed is harmless, nitrate largely builds up in the vegetative tissue of plants (cite from

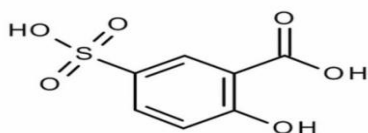
USDA). The goal of this study was to determine how some hazardous components changed in several green vegetables that are frequently eaten in Upper Egypt. The amount of nitrate in spinach was the largest, whereas coriander leaves had the lowest amount. Both the blanching and freezing processes caused a loss of nitrate. As reported by (**Sorour *et al.*, 2021**). The most frequently examined endpoints were thyroid disease, colorectal cancer, bladder, and breast cancer, each of which received three trials (four studies). Aside from methemoglobinemia, the best evidence for a link between drinking water nitrate consumption and harmful health outcomes from studies on colorectal cancer, thyroid disorders, and neural tube abnormalities. Nitrate levels in water were mostly below the corresponding regulatory limits in four of the five published studies on colorectal cancer, indicating an increased risk of colorectal cancer or colon cancer (**Ward *et al.*, 2018**). Vegetable plants' nitrate concentration fluctuated depending on a number of variables, such as how nitrogen fertiliser was managed, the amount of light, water stress, and soil pH. (**Abd-Elrahman *et al.*, 2022**). Future population expansion, increased nitrogen fertilizer use, and intensifying and concentrating animal agriculture will all likely result in higher nitrate concentrations in the world's water systems (**Ward *et al.*, 2018**). Groundwater, surface water from rivers, and treated wastewater are typically the three main sources used to irrigate agricultural crops. Additionally, a variety of synthetic and natural fertilizers are occasionally used to improve soil fertility, which in turn promotes the growth of agricultural goods, surface waters, and groundwater (**Kiani *et al.*, 2022**). Because of the aforementioned factors, the primary goal of this study was to evaluate the impact of various irrigation sources, including fresh water (third source), W (first source), and A (second source), on nitrate accumulation in soil, as well as the

inclusion of 12 different cultivated plant varieties. Furthermore, by consuming the aforementioned edible plants, the rate of nitrate factor transfer from soil to plants and the risk of nitrate-related human health issues were assessed. There are only a few global concurrent investigations on the effects of the three different types of irrigation sources on soil and plant pollution, nitrate risk assessments, and nitrate transfer factors that we are aware of. As a result, the finding of this study will be used to design future studies.

MATERIALS AND METHODS

Chemicals and Reagents

All chemicals used were of analytical grad. NaNO_3 - NaOH -



5-sulfosalicylic acid

Sample

Sampling of irrigation water sources

The following are the three different kinds of irrigation sources:

The first source of irrigation used to irrigate (WI) sections was well water (underground water).

Well water (underground water), which is used to irrigate (A) section, is the second irrigation source.

Fresh water from underground well is the third irrigation source used to water the (W) portion.

Sampling of the three sources for irrigation water were collected (W, WI, and A). Each sample underwent three iterations, yielding average samples for each type of irrigation water that were then tested for the desired pollutants.

Sampling of plants and soils

Three samples of each kind of plant that grew with a certain irrigation source were taken for this purpose from each type of plant. All plants were collected at reproductive stage. All farms fertilized by organic manure (cow manure) $15 \text{ m}^3/\text{fad}$. Consequently, 57 plant samples were gathered and sent to the laboratory as a result of 3 irrigation sources and 12 different types of plants. In this study part, an average of 19 plant samples were assessed for nitrate after each of the collected samples was examined. Rosemary, Breckl, Thyme, Mentha, Black Mulberry, Jute mallow, Alfalfa, Basil, Maize green chop, Kurrat, Pursley, Arugula, and Horse mint were among the 12 plant species that were gathered for this investigation, according to Table 1. Each sample came from one of three basic regions (A, W and WI). October 2021 saw the collection of samples. Soil samples were taken from the three sources, a table for the study sites, as shown in Table 1.

Nitrate measurement in various samples

Nitrate levels in the plant samples were determined by measuring nitrate in several samples. In order to estimate the amount of nitrate in each sample, an extraction step was also carried out using the necessary reagents and chemical solutions (per the instructions). The samples' absorbance was then calculated using a spectrophotometer. The amount of nitrate in plant sample was then determined after drawing the absorption-concentration diagram. It should be emphasized that the level of nitrate was determined using samples of plants that were collected. A spectrophotometer was used to measure the nitrate concentration in water, groundwater (well water), and fresh water in accordance with industry standards.

Table 1. Identification of the investigated plants that were collected from several sites in Arish, North Sinai Governorate, Egypt

No.	Name	Common name	Scientific name	Family	Origin	Growth habit
A ₁	Rosemary	Rosemary	<i>Rosmarinus officinalis</i>	Lamiaceae,	Mediterranean region	Evergreen shrub
A ₂	Breckl and Thyme	Creeping Thyme	<i>Thymus serpyllum</i>	Lamiaceae,	Europe, North Africa	Evergreen shrub
A ₃	Mentha	Mint	<i>Mentha spicata</i>	Lamiaceae,	Greek	Herbaceous plant
A ₄	Black Mulberry	Blackberry	<i>Morus nigra</i>	Moraceae	Ukraine, Iraq	Tree
A ₅	Jute mallow	Jew's mallow	<i>Corchorus olitorius</i>	Malvaceae	Tropical Africa	Herbaceous plant
A ₆	Alfalfa	Lucerne	<i>Medicago sativa</i>	Fabaceae, Leguminosae	South-central Asia, Iran	Perennial
A ₇	Basil	Great basil	<i>Ocimum basilicum</i>	Lamiaceae,	Central Africa, Southeast Asia	Annual, Perennial herb
W ₁	Maize green chop	Maize green forage	<i>Zea mays</i>	Poaceae, Graminaceae	Southern Mexico	Warm-season Annual herb
W ₂	Kurrat	Egyptian leek, salad leek	<i>Allium ampeloprasum</i>	Amaryllidaceae, Alliaceae	Southern Europe to Western Asia	Herbaceous biennial
W ₃	Jute mallow	Jew's mallow	<i>Corchorus olitorius</i>	Malvaceae	Tropical Africa	Herbaceous annual
W ₄	Maize green chop	Maize green forage	<i>Zea mays</i>	Poaceae, Graminaceae	Southern Mexico	Annual herb
W ₅	Pursley	Common purslane,	<i>Portulaca oleracea</i>	Portulacaceae	North Africa and Southern Europe	Annual (Tropical Perennial)
W ₆	Arugula	Garden rocket	<i>Eruca vesicaria</i>	Brassicaceae	Mediterranean region	Annual plant
W ₇	Jute mallow	Jew's mallow,	<i>Corchorus olitorius</i>	Malvaceae	Tropical Africa	herbaceous annual
WI ₈	Alfalfa	Lucerne	<i>Medicago sativa</i>	Fabaceae	South-central Asia	Perennial forage
WI ₉	Maize green chop	Maize green forage	<i>Zea mays</i>	Poaceae Graminaceae	Southern Mexico	Annual herb
WI ₁₀	Kurrat	Egyptian leek, Salad leek	<i>Allium ampeloprasum</i>	Amaryllidaceae, Alliaceae	Southern Europe to Western Asia	Herbaceous biennial
WI ₁₁	Horse mint	Filly mint	<i>Mentha longifolia</i>	Lamiaceae	Europe	Perennial Herb
WI ₁₂	Arugula	Garden rocket	<i>Eruca vesicaria</i>	Brassicaceae	Mediterranean region	Annual plant

A = Airport Road Location; 33° 48' 46" N, 31° 08' 06" E, W = Wadi EL-Arish1; 31° 07' 14" N, 33° 48' 44" E, WI = Wadi EL-Arish2, 31° 07' 13" N, 33° 49' 04" E,;

Preparation of reagents and standard solutions

Sulfosalicylic acid

100 ml of distilled water were used to dissolve 0.5 g of 5-sulfosalicylic acid.

Sodium nitrate standard solution

(1) Stock solution 0.1 M in 100 ml distilled water

Sodium hydroxide:

60 g was dissolved in 500ml distilled water

Nitrate extraction from soil and plants

A household blender was used to combine a 0.5-gram sample of the processed crop with 40ml of distilled water. Whatman No. 2 filter paper was used to filter the mixture (Afaf, 2006).

Nitrate determination from water

Add 20 ml of 5-sulfosalicylic acid and 10 ml of NaOH to 30 ml of water samples.

Quantitative determination of nitrate

A portion of the nitrate-containing solution was poured into a volumetric flask with a 100 ml capacity. Next, 10 ml of 5-sulfosalicylic acid and 5 ml of NaOH were added. To allow for colour development, the volume was filled with water and left for 15 minutes. At 410 nm, the absorbance was calculated in comparison to a blank solution. The calibration curve was created as follows, and it was used to determine the nitrate concentration: To create standard solutions of 0.01, 0.02, 0.03, 0.05, 0.001, 0.002, 0.003, 0.004 and 0.005M, 1, 2, 3, and 4 ml of the nitrate standard solution were placed into 100 ml volumetric flasks. NaNO_3 exhibits table 1. Following the addition of 10 ml of the 5-sulfosalicylic acid reagent, the volumes in each flask were finished to the markings. After 15 minutes, the absorbance was measured at 410 nm. By graphing the absorbance vs.

concentration, the calibration curve was created.

Standard curve

Among many other techniques, the visible spectrophotometer was picked and selected. Its low pollution consequences, simplicity, speed, and adaptability to show the kinetic change in nitrate concentration are the reasons for this. Figure 1 depicts a typical calibration curve for nitrate (NO_3^-) analysis.

Transfer factor evaluation

The transfer coefficient (TF) of each vegetable was calculated using Equation (1), and the actual amount of nitrate taken up by each vegetable. The transfer coefficient is the ratio of a contaminant's concentration in the plant to its concentration in the soil (Jolly *et al.*, 2013). It should be emphasized that the transfer coefficient should be calculated using a soil sample from each type of produce.

$$TF = \frac{CP}{CS} \quad (1)$$

Where TF is the transfer factor; CP is the concentration of contaminants in the plant (mg/kg); CS is a concentration of contaminants in soil (mg/ kg) (Kiani *et al.*, 2022).

RESULTS AND DISCUSSION

Chemical Reaction of Sulfosalicylic Acid with NO_3

Sulfosalicylic acid, which is produced when sulfuric acid and sodium salicylate react, reacts with nitrates to produce a mixture of ortho- and para-sodium nitrosalicylate in anhydrous media. The Nitrosalicylate anion is released in a basic environment, and its stable brownish-yellow color permits a colorimetric titration at 410 nm.

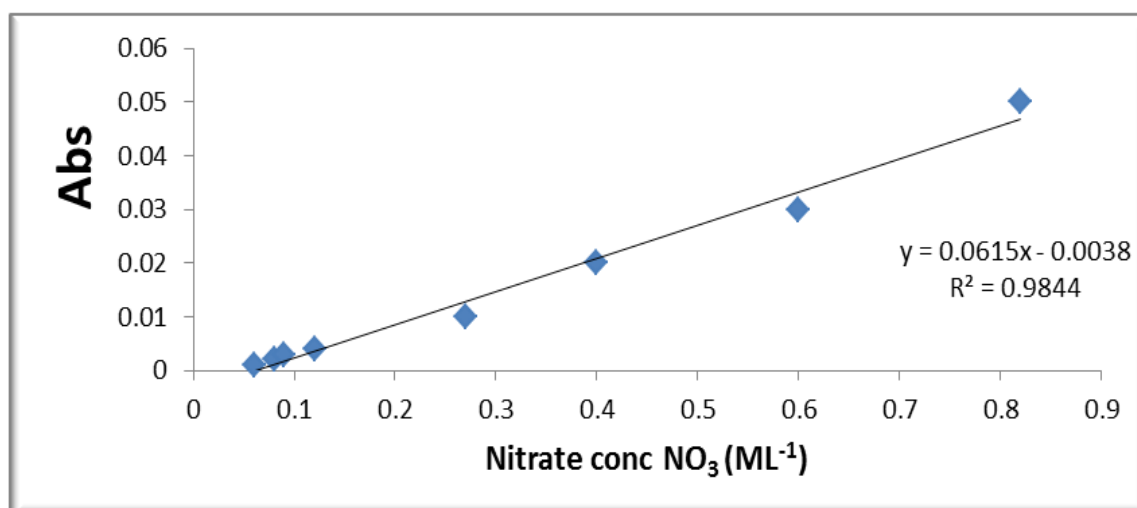
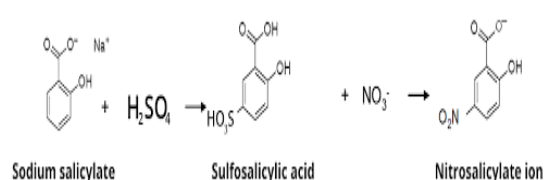


Fig. 1. A typical calibration curve for nitrate analysis (NO₃⁻) by spectrometric method



Concentrations NO₃ in plants

Twelve plants were tested for nitrate concentration in three different locations of El-Arish district, as indicated in Tables 2, 3, and 4 for nitrate. According to the region, they were split into three groups as follows:

Airport Road Location (A)

Average nitrate contents in 7 plants were determined in location A for nitrate, A1.... A7 are shown in Table 2. Legumes plants can fix atmosphere nitrogen.

Wadi EL-Arish (W)

Nitrate contents in 7 plants were determined in location W for nitrate W1.... W7 (Table 3).

Wadi EL-Arish (WI)

Nitrate contents in 5 plants were determined in location WI for nitrate, W8 W12 as shown Table 4. Legumes plants can fix atmosphere nitrogen.

Concentration NO₃ in sources of water

Nitrate content, pH and EC in water determined in the three sources of El-Arish district as shown in Table 5 and Fig. 2.

According to the findings, the concentration of nitrate in the water samples ranged from highest to lowest as AWWI (Fig. 2), taking into account that, Legumes plants can fix atmosphere nitrogen. According to **Zhonghua *et al.* (2020)**, irrigation regimes have an impact on crops cultivated in controlled environments' nitrate uptake, dispersion, and accumulation in addition to yield. For instance, root hypoxia brought on by insufficient water in the rhizosphere and excess nitrate accumulation due to shortfall and over watering contributes to excessive nitrate accumulation in wild rocket grown in greenhouses, respectively. (**Zhonghua *et al.*, 2020**) Others, on the other hand, discovered that by reducing the water level of the nutrition solution in hydroponic systems, proper rhizosphere drought stress reduced the nitrate concentration by 18% without affecting the lettuce crop's final output.

Table 2. NO₃⁻(mg kg⁻¹) content in 7 plants in location A

Plant	NO ₃ (mg kg ⁻¹)	TF
A ₁	991.5	104
A ₂	533.9	56
A ₃	991.5	104
A ₄	NIL	NIL
A ₅	NIL	NIL
A ₆	NIL	NIL
A ₇	762.7	80

Table 3. NO₃⁻(mg kg⁻¹) content in 7 plants in location W

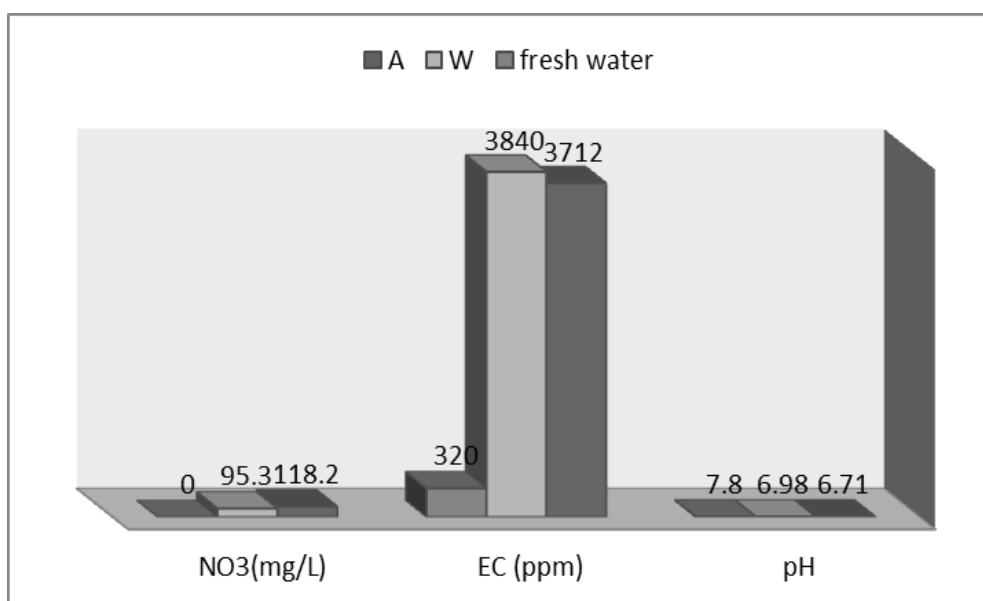
Plant	NO ₃ (mg kg ⁻¹)	TF
W ₁	NIL	NIL
W ₂	NIL	NIL
W ₃	NIL	NIL
W ₄	NIL	NIL
W ₅	NIL	NIL
W ₆	NIL	NIL
W ₇	NIL	NIL

Table 4. NO₃⁻(mg kg⁻¹) content in 5 plants in location WI

Plant	NO ₃ (mg kg ⁻¹)	TF
WI ₈	NIL	NIL
WI ₉	839.0	68.8
WI ₁₀	152.5	12.5
WI ₁₁	76.3	6.3
WI ₁₂	1525.5	125.0

Table 5. NO₃⁻(mg l⁻¹) content in water at the three sources of El-Arish district

Water	pH	EC(mg l ⁻¹)	NO ₃ (mg l ⁻¹)
W	6.71	3712	118.22
A	6.98	3840	95.34
WI	7.8	320	NIL

**Fig. 2.** NO₃⁻(mg l⁻¹) content in water at the three sources of El-Arish district

These contradictory findings may be the result of various growth strategies and nitrogen management practices, which have an impact on the uptake and metabolism of nitrates in various vegetable species.

Saline water and treated wastewater have been used for irrigation in many nations due to the lack of freshwater resources, particularly in dry and semi-arid regions. Reusing wastewater and saline water can help agricultural production use less water, but it's important to consider their negative effects on crop quality and development. (Conversa *et al.*, 2021) This might be explained by the competition of the same anion channel between nitrates

and chloride (Cl⁻) at the location of the xylem parenchyma cell membranes.

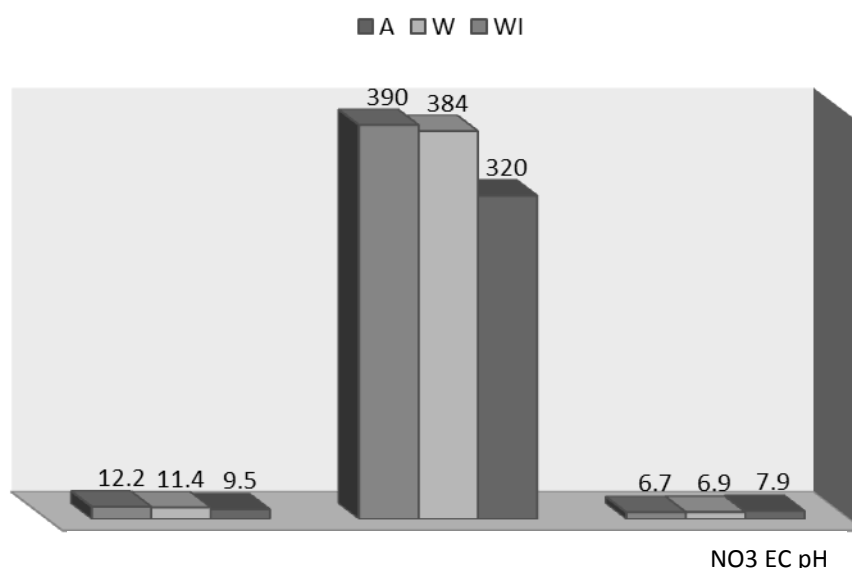
Concentration NO₃⁻ in soils

Nitrate content, pH and EC in soils determined in El-Arish district as shown in Table 6 and Fig. 3.

Irrigation schedules and water quality should not be disregarded in vegetable cultivation under controlled circumstances, according to Zhonghua *et al.* (2020). Producing low-nitrate veggies and saving water at the same time in controlled situations may be accomplished with the use of an irrigation regime that is based on water conditions.

Table 6. NO₃⁻(mg Kg⁻¹) content in soils at the three sources of El-Arish district

Cites soil	pH	EC(mg Kg ⁻¹)	NO ₃ (mg Kg ⁻¹)
A	7.9	320	9.5
W	6.9	384	11.4
WI	6.7	390	12.2

**Fig. 3.** NO₃⁻ (mg Kg⁻¹) content in soils at the three sources of El-Arish district

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المخلص العربي

حالة النترات في بعض النباتات المزروعة في تربة شمال سيناء

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دراسة محتوى النترات في عدد من النباتات المختلفة من مواقع مختلفة بمحافظة شمال سيناء (مدينة العريش). تركز هذه الدراسة على (1) مساهمة بعض النباتات الورقية الشهيرة في تناول النترات الغذائية من قبل الإنسان، (2) العوامل الغذائية والبيئية والفسولوجية التي تؤثر على تراكم النترات في النباتات و (3) الآثار الضارة والمفيدة للنترات على صحة الإنسان. هذا هو جزء من المواقع (A مطار العريش، 31° 08' 06" E, 33° 48' 46" N, وادي العريش 31° 07' 14" W و 33° 48' 44" E, WI وادي العريش 31° 07' 13" N, 33° 49' 04" E). تم جمع مجموعه 19 نباتاً من 12 نوعاً مختلفاً من النباتات (إكليل الجبل، زعتر بريكلاند، النعناع، التوت الأسود، الجوت الملوخية، البرسيم، الريحان، قطع الذرة الخضراء، الكوروات، البورسلي، الجرجير والنعناع) 57 عينة تم جمعها عشوائياً من مزارع مختارة في ثلاث مناطق مختلفة (مدينة العريش). أظهرت النتائج أن نوع النبات له تأثيراً كبيراً على محتوى النترات، حيث وجد أعلى محتوى نترات في الجرجير، بينما كان أقل محتوى نترات في الكوروات والنعناع. يتأثر محتوى النترات بنوع النباتات. أظهرت المواقع المختلفة تأثيراً معنوياً على محتوى النترات في النباتات المختبرة في هذه الدراسة. أشارت النتائج إلى مستويات مقبولة من محتوى النترات. ومع ذلك، هناك حاجة إلى مزيد من العمل لتوضيح هذه النتائج بمزيد من العينات والنباتات المختلفة.

الكلمات الاسترشادية: النترات في الماء، الأعلاف، النباتات الورقية، تقدير النترات، علاقة التربة بتراكم النترات.

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