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Nitrate and Nitrite Contents of Some Fresh and Processed Egyptian Vegetables

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ABSTRACT

The nitrate and nitrite contents of sixteen fresh vegetables, widely consumed in Egypt, were determined. The highest values of nitrate were observed in leafy vegetables, followed by root vegetables and then pulses. Of the leafy vegetables, spinach, roquette and chard contained the highest concentration of nitrate. Nitrite concentrations were detected at low levels in a few samples, while the others were free of nitrite.

Cooking had the effect of lowering the levels of nitrate in all types of fresh vegetables studied; no nitrite was formed during cooking.

Storage of frozen vegetables for six months decreased the levels of nitrate, while nitrite was formed at low levels after storage for three or four months.

INTRODUCTION

Nitrate and nitrite occur widely in human and animal foodstuffs, both as international additives and as undesirable contaminants. It has been recognized that high levels of these compounds are undesirable in certain foodstuffs such as baby foods. Comly (1945) observed methemoglobinemia in human infants given formulae prepared with well-water of high nitrate content. Von Oettingen (1958) stated that the ingestion of small doses of nitrate may cause cyanosis (methemoglobinemia) because of the reduction of nitrate to nitrite in the gastrointestinal tract. The toxicity of nitrites is due to their interaction with blood pigment to produce methemoglobinemia, and their presumptive toxicity is related to their possible reaction with amine or amides to form nitroso compounds (Wolff & Wasserman, 1972). Nitrate may be considered as the index of the amount of nitrite which may be formed. White (1975) showed that four-fifths of nitrate intake is from vegetables and less than one-sixth from cured meats, so it is important to maintain the nitrate content of vegetables at low levels. Very little information is available about the nitrate and nitrite contents of fresh vegetables in Egypt. The present work was undertaken to determine the nitrate and nitrite contents of some fresh vegetables commonly consumed in Egypt and to study the effect that cooking had on such contents. In addition, the effect of storage on nitrate and nitrite in frozen samples of vegetables was investigated.

MATERIALS AND METHODS

Materials

The sixteen samples of fresh vegetables studied included four categories—leafy vegetables (spinach, roquette, chard, parsley, jew's mallow, celery, lettuce and cabbage), pulses (okra, beans and peas) root vegetables (radish, carrots and potatoes) and others (squash and tomatoes). All samples were obtained from the local markets in Alexandria. Fresh samples were analysed on the day of purchase. Some samples of leafy vegetables, root vegetables and pulses were selected in order to study the effects of cooking and storage in the frozen state. Samples were prepared for cooking as for normal consumption and cooked in boiling water (1:2 v/v) for different times according to type. Frozen samples were prepared using a multiplate freezer, blanched in boiled water, packaged in polyethylene bags, frozen at −20 °C and stored for 6 months at −8 °C.

Methods

Reagents

(a) Alumina cream. Saturated solution of potassium aluminium sulphate.

(b) Ammonia buffer solution (pH 9·6–9·7): Twenty millilitres of concentrated HCl were added to 500 ml of water, then 50 ml of 0·880 ammonia were added and the solution was diluted to 1 litre.
(c) Diluted ammonia buffer: Ten millilitres of ammonia buffer were diluted to 100 ml with water.
(d) Orange I reagent: Fifty millilitres of glacial acetic acid and 360 ml of distilled water were warmed to 50 °C and poured into a dark glass reagent bottle (600 ml) containing 0·25 g of powdered sulphanilic acid and shaken until dissolved. Then 0·20 g of 1-naphthol were added and dissolved by shaking. The solution was cooled and 90 ml of 10% aqueous solution were added. The pH should be $4{\cdot}0 \pm 0{\cdot}05$.
(e) Reducing columns: Finely divided cadmium was placed in a 1·5 × 15 cm column as described by Follett & Ratcliff (1963).

Preparation of extracts
Fresh vegetables were prepared as for normal consumption, then sliced or cut into small pieces. Frozen samples were thawed and minced before analysis. Samples were extracted according to the method of Kamm *et al.* (1965). A 2-g sample was mixed with 5 ml of ammonia buffer solution, then 50 ml of alumina cream were added, followed by 50 ml of distilled water. The mixture was shaken for 10 min. on a mechanical shaker. Distilled water was added to the mixture until the total volume reached 200 ml and then it was filtered through Whatman No. 4 filter paper. Nitrite and nitrate were determined in the filtrate.

Nitrite determination
The method of Follett & Ratcliff (1963) was followed. Twenty millilitres of the extract were pipetted into a 100-ml volumetric flask, 5 ml of ammonia buffer solution were added and the flask was made up to volume with distilled water. Five millilitres of the solution were transferred to a test tube, 10 ml of Orange I reagent and 5 ml of distilled water were added. The optical density was measured at 474 nm using a Spekol spectro colorimeter against a blank prepared under the same conditions with the sample being omitted. A standard curve was prepared using sodium nitrite solution.

Nitrate determination
Nitrate was determined by reduction to nitrite using a cadmium column. Cadmium columns were prepared according to the method of Follett &

Ratcliff (1963). Prior to the determination the column was washed with 25 ml of dilute HCl (0·1N), followed by 50 ml of distilled water and, finally, with 25 ml of diluted ammonia buffer solution. Twenty millilitres of the extract were mixed with 5 ml of ammonia buffer solution and the mixture was poured onto the column, followed by distilled water. One hundred millilitres of the eluate were collected to determine nitrite as previously described.

RESULTS AND DISCUSSION

Nitrate and nitrite contents of some fresh vegetables

The results presented in Table 1 show that the highest values of nitrate were observed in leafy vegetables. Of the leafy vegetables, spinach contained the highest amount of nitrate (5830 ppm). In addition, roquette, chard, parsley and jew's mallow contained high levels of nitrate. Celery, lettuce and cabbage had the lowest levels of nitrate. The results are in agreement with those reported by Lee *et al.* (1971) who stated that the nitrate content of spinach was 2480 ppm. Pickston *et al.* (1980) found that the nitrate content of leafy vegetables lay between 15 and 2600 ppm.

The nitrate content of pulses varied from 30 to 600 ppm. Beans contained the highest level of nitrate. According to Walker (1975), the nitrate content of pulses was lower than that of leafy vegetables. The results are in agreement with those reported by Pickston *et al.* (1980) stating that the nitrate content of pulses ranged from 55 to 260 ppm.

The results indicate that, of the root vegetables, red and white radish had the highest nitrate contents. Walker (1975) reported that the nitrate content of radish was 5000 ppm and potato tubers usually contained less than 100 ppm of nitrate.

The results presented in Table 1 also show that nitrite was not detected in the fresh vegetables studied. In general, the leafy vegetables had higher nitrite contents than pulses and root vegetables. These results agree with those reported by other investigators who stated that the nitrite concentrations in fresh vegetables were usually very low, or less than 1 ppm (Siciliano *et al.*, 1975; Walker, 1975; Corsi *et al.*, 1981).

The results given in Table 2 show that cooking reduced the levels of nitrate in all vegetables. The loss upon cooking was more pronounced in leafy vegetables; it reached 79·4 % for spinach, 62 % for jew's mallow and

TABLE 1
Nitrate and Nitrite Contents of Some Fresh Vegetables
(ppm on dry weight basis)

Vegetables	*Per cent moisture*	*Nitrate*	*Nitrite*
Leafy vegetables			
Spinach	93·7	5 830	—
Roquette	91·8	4 780	—
Chard	88·8	2 180	20
Parsley	91·2	1 840	—
Jew's mallow	88·8	1 290	4
Celery	88·0	540	30
Lettuce	95·3	430	110
Cabbage	93·0	190	—
Pulses			
Beans	89·5	600	—
Okra (foreign)	85·6	150	—
Okra (local)	85·2	80	—
Peas	76·2	30	—
Root vegetables			
Radish (red)	91·6	3 400	10
Radish (white)	95·2	3 260	120
Carrots	92·8	430	—
Potatoes	83·8	160	—
Others			
Squash	91·7	1 380	—
Tomatoes	94·9	130	40

31·5 % for cabbage. Phillips (1968) reported that cooking of fresh spinach decreased the nitrate content of the edible portions to approximately 20 % to 25 % of the initial value. Pickston *et al.* (1980) showed that the level of nitrate dropped in the cooked cabbage by an average of 18 %. According to Lutsoya & Rooma (1971) about 85 % of the nitrates and nitrites present in the vegetables passed into the cooking water.

The results indicate that cooking had a greater effect in lowering the nitrate content of beans than those of the other types of pulse. This loss reached 34 % while cooking of peas reduced nitrate content by only 16 %. In addition, cooking of root vegetables caused a loss in nitrate content as

TABLE 2
Effect of Cooking on Nitrate and Nitrite Contents of Some Fresh Vegetables (ppm on dry weight basis)

Vegetables	*Nitrate*	*Per cent loss*	*Nitrite*	*Per cent loss*
Leafy vegetables				
Spinach				
Raw	5 830	—	—	—
Cooked	1 200	79·4	—	—
Jew's mallow				
Raw	1 290	—	4·4	—
Cooked	490	62·0	1·6	40·9
Cabbage				
Raw	190	—	—	—
Cooked	130	31·5	—	—
Pulses				
Beans				
Raw	590	—	—	—
Cooked	390	34	—	—
Okra (foreign)				
Raw	150	—	—	—
Cooked	110	26	—	—
Okra (local)				
Raw	80	—	—	—
Cooked	70	13	—	—
Peas				
Raw	30	—	—	—
Cooked	25	16	—	—
Root vegetables				
Carrots				
Raw	430	—	—	—
Cooked	210	51·0	—	—
Potatoes				
Raw	160	—	—	—
Cooked	70	56	—	—

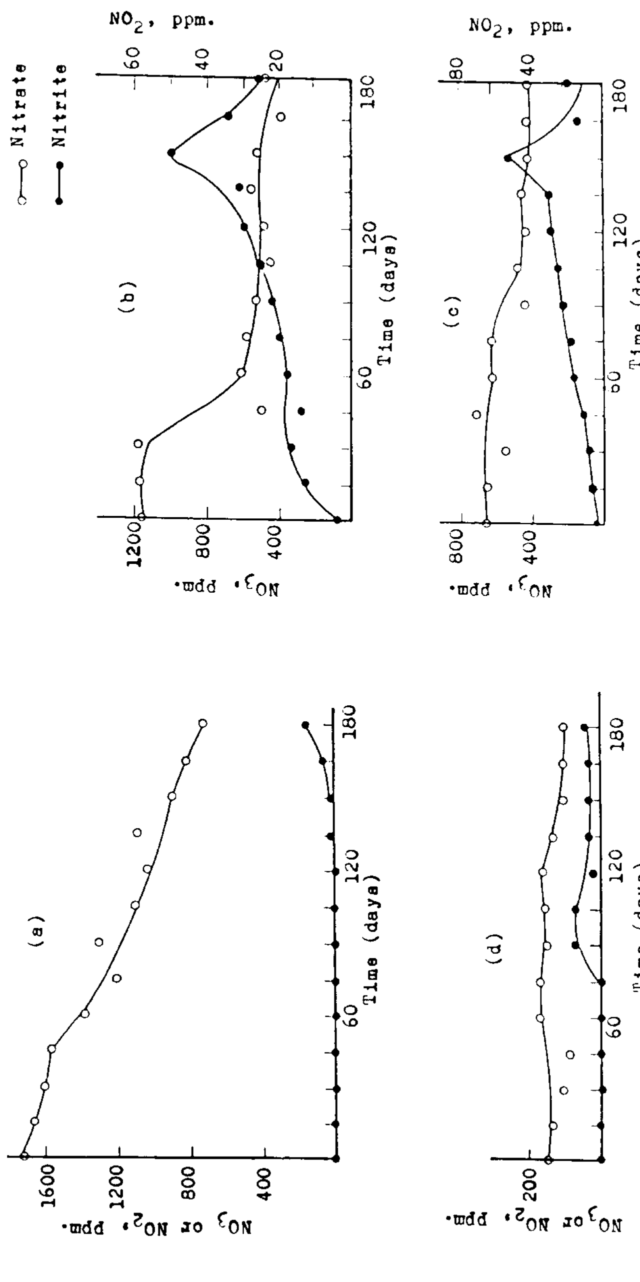

Fig. 1. Effect of storage on nitrate and nitrite contents of some frozen leafy vegetables. (a) Spinach. (b) Jew's mallow (leaf). (c) Jew's mallow (minced). (d) Cabbage.

indicated from the results in Table 2. Hata & Ogata (1971) showed that the nitrate and nitrite contents of potatoes were heat stable during cooking but losses occurred due to leaching from potato tissues into cooking water. Pickston *et al.* (1980) reported that the level of nitrate fell, after cooking, by an average of 24% for potatoes. The lost nitrate migrates into the cooking water and there is then a tendency to form nitrite.

Also, from the above results it can be seen that nitrite did not accumulate, after cooking, in all vegetable samples and, indeed, fell by 40·9% in jew's mallow.

Effect of storage on nitrate and nitrite contents of frozen vegetables

Samples of fresh vegetables were frozen to study the effect of storage on nitrate and nitrite contents. The samples were analysed at 15-day intervals

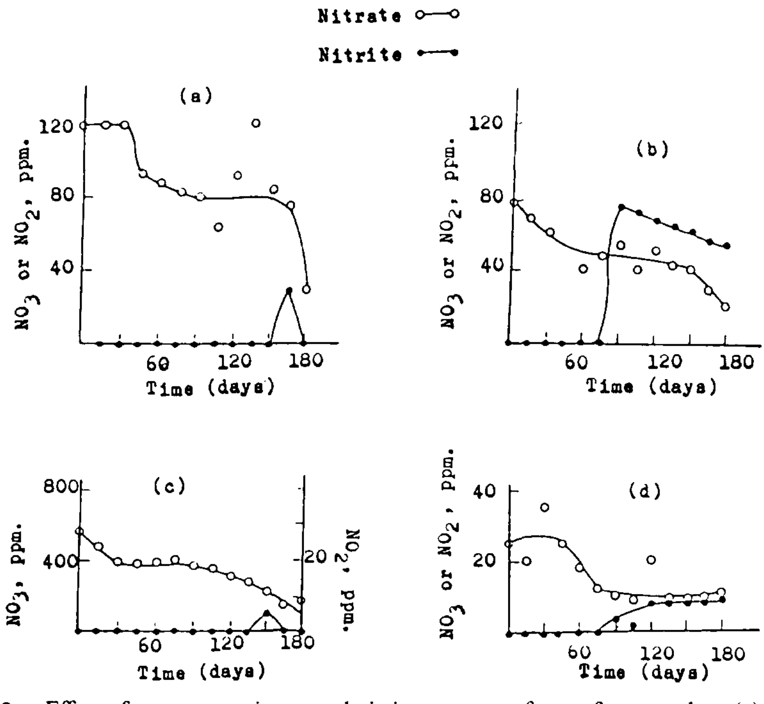

Fig. 2. Effect of storage on nitrate and nitrite contents of some frozen pulses. (a) Foreign okra. (b) Local okra. (c) Beans. (d) Peas.

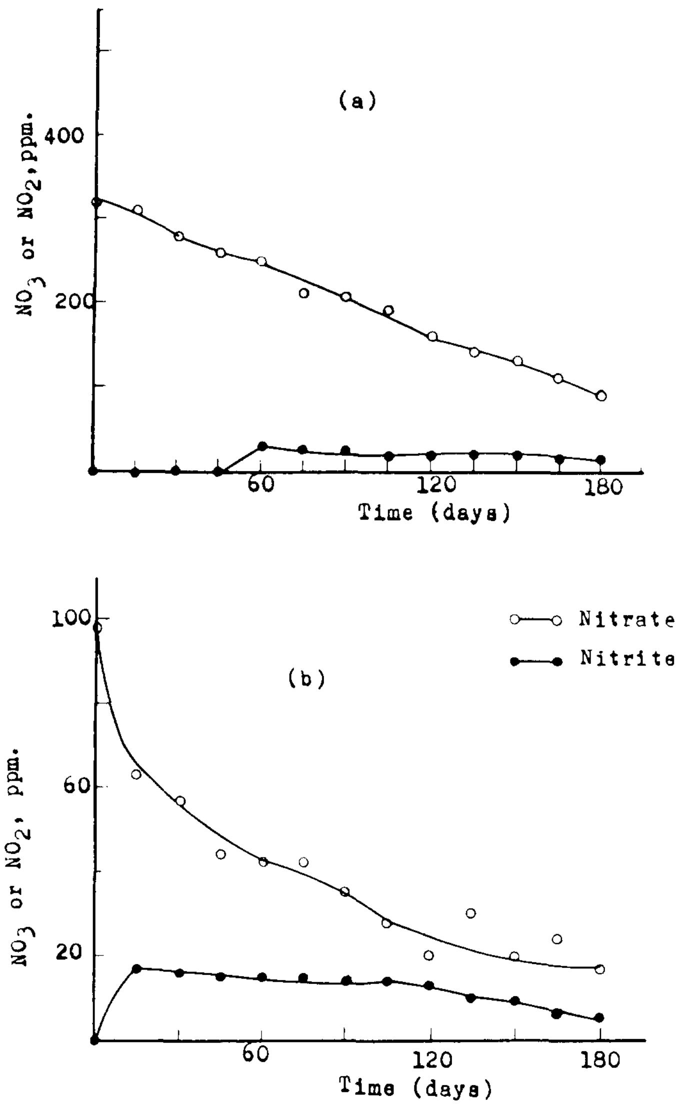

Fig. 3. Effect of storage on nitrate and nitrite contents of some frozen root vegetables. (a) Carrots. (b) Potatoes.

during the period of storage. The results in Fig. 1 also show that storage of frozen spinach for up to 6 months caused a reduction in nitrate content. No nitrite was found in frozen spinach during the first 4 months of storage but it was formed after storage for 6 months. Phillips (1968) reported that there was no significant increase in the nitrite content of frozen spinach with period of storage up to 5 months. Walker (1975) showed that frozen spinach did not accumulate large amounts of nitrite (less than 4 ppm) but, on thawing, nitrite concentration rose rapidly. During storage of frozen jew's mallow nitrate content decreased; on the other hand, nitrite content increased due to the reduction of nitrate to nitrite. The results show the same trend when frozen cabbages were stored—the level of nitrate decreased and no nitrite was detected during the first period of storage, then it accumulated to a high level after 3 months' storage.

The results illustrated in Fig. 2 show that the same trend was found with regard to the nitrate content of frozen pulses (beans, okra and peas); when stored for 6 months they showed a moderate decrease. No significant conversion of nitrate to nitrite took place within the first months of storage of frozen pulses, then it was formed at a low level after 3 months in all samples of pulses. In Okra (local) it accumulated at a high level.

As shown in Fig. 3, during storage, frozen root vegetables (carrots and potatoes) also show a decrease in nitrate content. Nitrite did not accumulate during the first period of storage but appeared after 2 months in frozen carrots and after 15 days in frozen potatoes.

In conclusion, cooking of fresh vegetables in boiling water had the effect of lowering their nitrate and nitrite contents due to leaching into the cooking water. Frozen vegetables stored for more than 3 or 4 months showed an increase in nitrite concentration, whilst, in the case of potatoes, it increased after only 15 days.

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