Food Additives & Contaminants

ISSN: 0265-203X (Print) (Online) Journal homepage: https://www.tandfonline.com/loi/tfac19

Nitrate and nitrite in vegetables from north China: content and intake

Weike Zhong , Changmin Hu & Minjiang Wang

Food Additives and Contaminants, 2002, Vol. 19, No. 12, 1125–1129

Taylor & Francis
Taylor & Francis Group

Nitrate and nitrite in vegetables from north China: content and intake

Weike Zhong†*, Changmin Hu‡ and Minjiang Wang‡

† *Laboratory of Nuclear Analytical Techniques, Institute of High Energy Physics, Chinese Academy of Sciences, PO Box 918, Beijing 100039, P. R. China*

‡ *Research Center for Eco-Environmental Sciences, Chinese Academy of Sciences, PO Box 2871, Beijing 100085, P. R. China*

(Received 4 January 2002; revised 5 July 2002; accepted 12 July 2002)

The contents of nitrate and nitrite in potato, cabbage, Chinese cabbage, scallion (shallot), celery, cucumber, tomato, eggplant and wax gourd taken from the north China market from 1998 to 1999 were determined. These vegetables provide the major contribution to the nitrate intake from the diet. The highest content of nitrate was found in celery followed by Chinese cabbage, cabbage, scallion, wax gourd and eggplant. For all the products, a great variation in the content of nitrate was found. Generally, the nitrite content was low. The average intake of nitrate and nitrite from these vegetables was estimated as approximately 422.8 and 0.68 mg day^{-1}, respectively.

Keywords: nitrate, nitrite, vegetables, north China

Introduction

Nitrate is present in food naturally or it can be present as a result of the use of fertilizers on crops. The nitrate ion has a low level of acute toxicity, but it can be transformed into nitrite, which has much higher acute toxicity. It has been estimated that about 4–8% of the nitrate from the diet may be reduced to nitrite by the microflora in the oral cavity (Stephany and Schuller 1980, Gangolli *et al.* 1994, WHO 1996). Some studies showed that nitrate exposure is correlated with gastric cancer risk due to the endogenous formation of *N*-nitroso compounds (NOC) (Hotchkiss *et al.* 1992). The intake of nitrite is normally low compared with the dose that is acutely toxic, but nitrite in food is considered primarily to be a health problem because its presence both in food and in human body may lead to the formation of nitrosamines. Experiments in animals have shown that many nitrosamines are potent carcinogens and that they, finally, may also cause cancer in humans. Thus, there is increasing concern about the contamination of nitrate and nitrite in food, especially in vegetables (Petersen and Stoltze 1999, Vaessen and Schothorst 1999, Ysart *et al.* 1999), because vegetables are the largest source of dietary nitrate amounting for $> 90\%$ of nitrate exposure (Knight *et al.* 1987).

In China, monitoring programmes have been implemented to determine nitrate and nitrite levels in vegetables (Zhou *et al.* 2000). However, the results obtained might not be representative because of the limited areas covered and the lack of data on average consumption. In order to study the occurrence and source of nitrate and nitrite in vegetables in north China, a monitoring programme was established in 1998–99. This paper reports the results obtained from the monitoring of nitrate and nitrite in vegetables grown in north China and assesses the influence of fertilizer use on nitrate concentrations in these crops. The daily intake of nitrate and nitrite from these vegetables is then estimated.

Materials and methods

Vegetable sampling

The samples were obtained from the local wholesale markets in five average provinces in northern China. The nine main vegetables chosen for the study ac-

* To whom correspondence should be addressed. E-mail: zhongwk@mail.ihep.ac.cn

Food Additives and Contaminants ISSN 0265–203X print/ISSN 1464–5122 online
http://www.tandf.co.uk/journals
DOI: 10.1080/026520302100001480 6

count for about 80% of all the vegetables in the north China diet, including root vegetable (potato), leafy and stem vegetables (cabbage, Chinese cabbage, scallion and celery), and fruiting vegetables (cucumber, tomato, eggplant and wax gourd). In total, 200 samples were taken, and the sampling numbers were based on individual vegetable consumption in north China in 1997 to represent the best real market situation. The sampling period (February 1998–February 1999) was divided into four seasons (spring, summer, fall, winter). This distinction is logical because nitrate accumulation in vegetables is associated with many factors including plant species, light intensity and temperature. The vegetables were all produced locally and kept fresh. After they were purchased from local wholesale markets, the samples were bagged and kept at 4°C in a refrigerator before analysis.

Analyses

Samples were washed with tap water and deionized water. The edible part of the samples was used for analyses. For each vegetable, a composite sample of many individuals (e.g. 10 Chinese cabbages, 50 tomatoes, etc.) was used. After washing, the vegetables were chopped into small sections and homogenized in a blender mill. The sample (10 g) was weighed in a beaker and 50 ml deionized water added. The beaker was placed in a water bath at 70°C for 30 min. The extract was filtered into a flask (100 ml) and diluted exactly to 100 ml with deionized water.

The nitrate content in the extract was detected by high-performance ion chromatography (HPIC) with UV detector (208 nm) on an Ionpac AG4A-SC pre-column and AS4A-SC column (Diones). The mobile phase was 2.8 mm $NaHCO_3$ and 2.2 mm Na_2CO_3. Blank samples were prepared to examine contamination during the treatment operation. When the samples were determined by HPIC, two injections of a nitrate standard were made for every five pairs of vegetable samples to calibrate any possible variation in the compound responses. Nitrite in the extract was reacted with sulphanilanide and *N*-(1-naphthyl)ethylene-diamine, and the absorbance of the violet azo compound was measured at 540 nm. The average recovery rates of nitrate and nitrite were 95 and 92%, respectively, and the limits of quantification attained by the present method were 1 and 0.004 mg kg^{-1} respectively. The results of the method were identical to those of the Chinese National Standards Analytical Method (GB/T15401-94, 1995), which was based on a conventional sulphanilamide and *N*-(1-naphthyl)ethylene-diamine chloride colorimetric determination of nitrite followed by cadmium column reduction to determine nitrate by difference.

Results and discussion

Nitrate

The contents of nitrate in nine vegetables in north China are shown in table 1. Nitrate accumulation was quite different both between different vegetables and within the same vegetables, ranging from 10^1 to 10^4 mg kg^{-1}. In general, leafy and stem vegetables have a high nitrate accumulation. Celery contained the highest nitrate concentration with an average of

Table 1. Content of nitrate in vegetables (mg kg^{-1} for fresh weight).

				No. of findings in the indicated range		
Specimen	No. of samples	Range	Average	< 1000	2000	> 2000
Potato	26	31.5–714	164	26		
Scallion	26	8.03–4240	704	19	5	2
Chinese cabbage	24	337–3600	2120	3	5	16
Cucumber	23	17.4–500	170	23		
Cabbage	23	26–2670	1530	9	4	10
Tomato	22	10.3–259	77.9	22		
Eggplant	20	67.3–1000	479	16	4	
Wax gourd	19	201–1390	635	14	4	1
Celery	17	446–10800	3600	2	2	13

3600 mg kg^{-1} and a maximum of 10 800 mg kg^{-1}. For Chinese cabbage, cabbage and scallion, the average contents of nitrate were about 2120, 1530 and 704 mg kg^{-1}, respectively. The root and fruit vegetables have relatively low nitrate contents. For tomato, the average content of nitrate is only 77.9 mg kg^{-1}. As the tolerance level for nitrate in vegetables has not been established in China, we simply classified our results in three ranges (nitrate content < 1000, 1000 < nitrate < 2000, and nitrate > 2000 mg kg^{-1} for fresh weight). Note that the nitrate concentrations in celery, Chinese cabbage and cabbage are very high.

As mentioned above, the nitrate accumulation in vegetables is affected by many factors, among which fertilizer application is the major one. Nitrate accumulation in vegetables often depends on the amounts and kinds of nutrients present in the soil and especially is closely related to the amounts of the fertilizers applied and the time of application. Some studies indicate that the proper application of nitrogenous, phosphate, potassium fertilizer, and green and farmyard manure could significantly reduce nitrate accumulation in vegetables. The optimal proportion for nitrogenous, phosphate and potassium fertilizer is 1.0:0.7:2.0 (Zhou *et al.* 2000). Our survey from 50 farms indicates that the actual proportion was 1.0:0.8:0.5. Thus, the heavy application of chemical nitrogenous fertilizer and the lack of potassium fertilizer could be the main cause of nitrate contamination in vegetables. Anther reason for the high level of nitrate in celery and Chinese cabbage may be the hydroponic cultivation method. Our experiment shows that the NO_3^- contents of celery were mostly dependent on the cultivation method and that celery cultivated by a hydroponic method contained 4500–9850 mg kg^{-1} whereas that cultivated by conventional methods contained 550–980 mg kg^{-1}.

Nitrite

The contents of nitrite in all the vegetables are shown in table 2. The nitrite contamination in vegetables was not as high as for nitrate. Although there were 13 samples exceeding the tolerance limit of China (GB15198-94, 1995), only four samples contained > 10 mg kg^{-1} nitrite with none > 50 mg kg^{-1}. Further, the contents of nitrite in 17 samples were below the detection limit. There were large variations in the amount of nitrite in the vegetables and no correlation was found between the contents of nitrate and nitrite.

Although only small amounts of nitrite are present in vegetables, nitrite can be formed via reduction of nitrate if they are stored incorrectly. This is especially a problem for leafy vegetables, because soil adhered to them may be difficult to remove completely.

Intake

Another aim of this paper was to estimate the average human potential exposure to nitrate and nitrite via vegetables. To accomplish this aim, the mean nitrate and nitrite concentrations in the vegetables were used in combination with the average daily consumption of the vegetables. The average consumption of different kind vegetables in north China was completed with our own calculations in cooperation with the Ministry of Agriculture (table 3). Potential human exposure to nitrates and nitrites through vegetation in the north China was estimated on average consumption, and the results are also shown in table 3.

The total intake calculated from nine vegetables was approximately 422.8 mg day^{-1} for nitrate and

Table 2. Content of nitrite in vegetables (mg kg^{-1} for fresh weight).

Vegetables	No. of samples	Range	Average	SD	Sample no. exceeding the tolerance limit
Potato	26	0.1–4.9	1.59	1.29	1
Scallion	26	n.d.–44.9	2.91	8.78	3
Chinese cabbage	24	n.d.–0.739	0.183	0.198	0
Cucumber	23	0.034–1.11	0.229	0.284	0
Cabbage	23	n.d.–4.77	0.472	0.973	1
Tomato	22	n.d.–2.08	0.233	0.419	0
Eggplant	20	0.055–48	4.63	10.7	3
Wax gourd	19	0.056–28.5	2.00	6.4	2
Celery	17	0.009–8.6	2.17	4.7	3

n.d., not detected

Table 3. Average daily intake of nitrate and nitrite from vegetables in north China.

Vegetable	Consumption ($g\,day^{-1}$)	Nitrate content ($mg\,kg^{-1}$)	Nitrite content ($mg\,kg^{-1}$)	Nitrate intake ($mg\,day^{-1}$)	Nitrite intake ($mg\,day^{-1}$)
Potato	85	164	1.59	13.9	0.135
Scallion	65	704	2.91	45.8	0.19
Chinese cabbage	54	2120	0.183	114.5	9.9×10^{-3}
Cucumber	53	170	0.229	9.1	1.2×10^{-2}
Cabbage	45	1530	0.472	68.9	2.1×10^{-2}
Tomato	44	77.9	0.233	3.4	1.0×10^{-2}
Eggplant	39	479	4.63	18.7	0.18
Wax gourd	31	635	2.00	19.7	6.2×10^{-2}
Celery	28	4600	2.17	128.8	6.1×10^{-2}
Total	444			422.8	0.68

0.68 $mg\,day^{-1}$ for nitrite. Celery and Chinese cabbage constitute the major contribution (about 57% of total nitrate) to the average nitrate intake of the north Chinese because of their higher nitrate content. It must be emphasized that nine vegetables in table 3 only accounted for 80% of the total consumption in the north Chinese diet. The other 20% include many different kinds of vegetables that are supposed to have the same distributions of nitrate and nitrite as in the other 80%. If all vegetables were included, the total nitrate and nitrite intakes from vegetables were about 528.5 and 0.85 $mg\,day^{-1}$.

It is interesting to compare the potential intake of nitrate and nitrite from vegetables in north China with those in other countries (table 4). From table 4, it can be concluded that the nitrate intake estimated in this study is substantially larger than the intake estimates reported from the UK, Denmark and Egypt. The most obvious explanation for this difference is the heavy consumption of vegetables in China.

The toxicology of nitrate and nitrite has been repeatedly evaluated by the World Health Organization (WHO), which retained the previously established acceptable daily intake (ADI) of nitrate at 3.7 mg $NO_3^-\,kg^{-1}$ body weight day^{-1}, but lowered that for nitrite from 0.13 to 0.06 mg $NO_2^-\,kg^{-1}$ body weight day^{-1} at the meeting of the Joint WHO/FAO Expert Committee in Food Additives (JECFA) in 1995 (WHO 1996). According to the ADIs of 3.7 mg nitrate and 0.06 mg nitrite kg^{-1} body weight, the maximal intakes of nitrate and nitrite are 222 and 3.6 mg, respectively, for an average Chinese person weighing 60 kg. Although, the average nitrite intake is substantially lower than the ADI, the average nitrate intake is nearly twice that of the established ADI. Although consumption of fruit and vegetables is thought to protect against cancer, a high nitrate intake must taken into account when considering the endogenous conversion of nitrate to the much more toxic nitrite in the oral cavity.

Table 4. Comparison of the intake of nitrate and nitrite in north China with other countries ($mg\,person^{-1}\,day^{-1}$).

Country	Vegetable consumption ($g\,day^{-1}$)	Nitrate intake ($mg\,day^{-1}$)	Nitrite intake ($mg\,day^{-1}$)
China	510	486	0.78
Denmark[a]	142	38.9	0.091
England[b]	322	108.5	2.2
Egypt[c]	159	296	–

[a] Petersen and Stoltze (1999), [b] Ysart *et al.* (1999), [c] Saleh *et al.* (1998).

Acknowledgements

The authors thank Professor Chai zhifang at the Laboratory of Nuclear Analytical Techniques, Institute of High Energy Physics, Chinese Academy of Sciences, for helping with the preparation of the manuscript.

References

Gangolli, S. D., Van den Brandt, P. A., Feron, V. J., Janzowski, C., Koeman, J. H., Speijers, G. J. A., Spielgelhaleder, B., Walker, R., and Wishnok, J. A., 1994, Assessment: nitrate, nitrite and *N*-nitroso compounds. *European Journal of Pharmacology, Environmental Toxicology and Pharmacology*, **Section 292,** pp. 1–38.

GB/T15401-94, 1995, *Standards of the People Republic of China* (Beijing: Chinese Standard Press).

GB15198-94, 1995, *Standards of the People Republic of China* (Beijing: Chinese Standard Press).

Hotchkiss, J. H., Helser, M. A., Maragos, C. M., and Weng, Y. M., 1992, Nitrate, nitrite, and *N*-nitroso compounds. *Food Safety Assessment*, edited by J. W. Finley, S. F. Robinson and D. J. Armstrong (Washington, DC: American Chemical Society), pp. 401–418.

Knight, T. M., Forman, D., Aldabbagh, S. A., and Doll, R., 1987, Estimation of dietary Intake of Nitrate and Nitrite in Great Britain. *Food Chemical Toxicology*, **25,** 277–285.

Petersen, A., and Stoltze, S., 1999, Nitrate and nitrite in vegetables on the Danish market: content and intake. *Food Additives and Contaminants*, **16,** 291–299.

Saleh, Z. A., Brumm, H., Paetzold, R., and Hussein, L., 1998, Nutrients and chemical residues in an Egyptian total mixed diet. *Food Chemistry*, **63,** 535–541.

Stephany, R. W., and Schuller, P. L., 1980, Daily dietary intakes of nitrate, nitrite and volatile *N*-nitrosamines in the Netherlands using the duplicate portion sampling technique. *Oncology*, **37,** 203–210.

Vaessen, H. A. M. G., and Schothorst, A. C., 1999, The oral nitrate and nitrite intake in the Netherlands: evaluation of the results obtained by PHLC analysis of duplicate 24-hour diet samples collected in 1994. *Food Additives and Contaminants*, **16,** 181–188.

WHO, 1996, *Toxicological Evaluation of Certain Food Additives and Contaminants*. WHO Food Additives Series No. 35 (Geneva: World Health Organization), pp. 269–360.

Ysart, G., Clifford, R., and Harrison, N., 1999, Monitoring for nitrate in UK-grown Lettuce and Spinach. *Food Additives and contaminants*, **16,** 301–306.

Zhou, Z. Y., Wang, M. J., and Wang, J. S., 2000, Nitrate and nitrite contamination in vegetables in China. *Food Review International*, **16,** 61–76.