Nitrate and glasshouse vegetables

JPNL ROORDA VAN EYSINGA

Institute for Soil Fertility, Haren (Gr.), The Netherlands, stationed at Glasshouse Crops Research and Experiment Station, Naaldwijk, The Netherlands

(Accepted: 13.06.1983)

Key words: nitrate in vegetables, nitrification inhibitors, nitrate and human health

Abstract. Leafy vegetables grown under glass in winter have a rather high nitrate content. Three possibilities of reducing this content are discussed: the use of nitrification inhibitors, growing on NFT and omission of nitrogen during a certain period before harvest, and the breeding of varieties with a low nitrate content.

In the last chapter the question whether high nitrate levels in vegetables are dangerous to human health is briefly discussed.

The quality of our food and its composition are being scrutinised more critically than ever before by the consumer. Many feel that the nitrate contents in a number of vegetables are too high. In Switzerland, an action level of 3500 mg NO₃ per kg fresh produce came into force for lettuce on 11 September 1981 (changed into a maximum permissible level of 4000 mg/kg in November 1982). Since 1 October 1982 the Dutch government laid down maximum permissible levels for nitrate in endive, spinach and outdoor lettuce (4000 mg NO₃ per kg fresh produce). Literature dealing with nitrate in vegetables was recently reviewed by Corré and Breimer [6].

The following aspects will be discussed in three headings:

- * the current situation in The Netherlands and in neighbouring countries.
- * the possibilities of reducing nitrate levels in glasshouse vegetables, and
- * the question why such reductions are necessary at all.

The current situation

Lettuce is the most important glasshouse salad vegetable in Western Europe. Nutrition experiments have shown that even if fertilizers are omitted, which would result in yield reductions of 10 to 20%, the nitrate contents in glasshouse lettuce will be reduced by an average of only 10% compared with a normally fertilised crop [14]. An example of more recent date is shown in Table 1. The trial was run in a commercial glasshouse on loamy sand after thorough leaching.

The nitrate levels in lettuce seem to depend mainly on the amount of light (radiation) received by the crop, which is well-illustrated with data from

Fertilizer Research 5: 149–156 (1984) © 1984 Martinus Nijhoff/Dr W. Junk Publishers, The Hague. Printed in The Netherlands.

Table 1. Yields and nitrate contents of lettuce (cv 'Pascal') harvested on 11 January 1982, with increasing amounts of nitrogen fertiliser

Ammonium nitrate limestone (26% N), kg/100 m ²	Mean head weight, g	Nitrate content, mg NO ₃ per kg fresh matter
0	182	3970
5	200	4410
10	211	4370

Statistical analysis of yield data: linear effect P < 0.01; nitrate content not tested

Switzerland in Figure 1. A similar relationship was found in The Netherlands between the harvest date and the nitrate contents of glasshouse lettuce, although in this case average values were about 1000 mg NO₃ per kg fresh produce higher. In other crops, for example in endive, the effects of nutrition were more pronounced in winter [3].

The provisional conclusion that may be drawn from this experience is that in the case of leafy vegetables, there is probably an interaction between the amount of light, or the time of year in which the crop is grown, and the nitrate content in the crop as affected by various nitrogen dressings. In addition, both the response and the interaction depend on the kind of crop.

High nitrate contents may be found in all leafy vegetables, as well as in some stem, tuber and root crops, but not in fruit vegetables such as tomatoes and cucumbers.

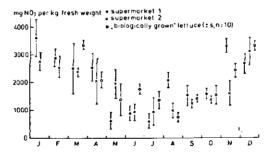


Figure 1. The seasonal pattern of nitrate contents of lettuce in shops in Switzerland according to Temperli et al. (1978/1979)

Methods of reducing the nitrate contents in crops

In the author's view, there are three possibilities: the use of a nitrification inhibitor, the use of solution culture techniques whereby nitrogen is omitted from the nutrient solution some time before the crop is harvested, and the breeding of varieties with a low nitrate content.

Nitrification inhibitors

There are many nitrification inhibitors. The author has gained some experience with nitrapyrin (N-serve) and dicyandiamide (Didin). Nitrapyrin is a product of Dow Chemical, available as N-serve 24. In experiments with lettuce, this product was mixed with ammonium sulphate usually applied in quantities of 10 kg per 100 m² and distributed over the soil surface. Shortly after, the fertiliser was incorporated into the soil by rotary cultivation.

N-serve 24 was tested in quantities ranging from 0 to 2 litres per 100 m². The results showed an average yield reduction of only 4%, whilst the nitrate contents were reduced, on average, by 25%. The emulsifiable product N-serve 24 E was also tested when mixed with 25% liquid ammionia in soil injection experiments.

Shallow injection (5 cm) produced more or less the same effects, but deep injection (20 cm) gave a yield reduction (Table 2).

Table 2. Yields and nitrate contents of lettuce (cv 'Pallas') harvested on 6 April 1981, following dressings of ammonium sulphate or liquid ammonia injections at 20 cm depth, with and without the addition of N-serve 24 E ($\frac{1}{2}$ litre 100 m²)

Fertiliser treatment, kg or 1/100 m ²	Mean head weight, g	Nitrate content, mg NO ₃ per kg fresh matter
9 kg ammonium sulphate		
(26.5% N)	184	3640
91 liquid ammonia (25% N) 91 liquid ammonia +	170	3290
N-serve 24 E	172	3030

Statistical analysis of yield data: ammonium sulphate versus liquid ammonia, with and without N-serve: P = 0.03; nitrate content not tested

Later experiments proved that deep injection with ammonia, with or without N-serve 24E, was unfavourable. Three injection depths were compared - 5, 10 and 20 cm - and the deeper injections resulted in significantly lower yields, even lower than the yields obtained from unfertilised control plots. Nitrapyrin appears to be a suitable nitrification inhibitor for crops under glass, but the product sold at the moment is dissolved in an organic solution, undesirable to be used in large quantities. It will be necessary to design a better formulation of the product to make its application technologically more attractive. The manufacturer has promised to set up a research programme for the purpose.

Dicyandiamide is marketed in West Europe by S.K.W. Trostberg, Munich, FRG, under the name of Didin. In experiments with lettuce, up to 1 kg of the product per 100 m², thoroughly mixed with ammonium sulphate, could be applied without great problems. Didin has more acute action than N-serve. The appearance of lettuce changes a little when the material becomes fully active. The crop becomes a little darker in colour and the leaves grow more

upright, giving the plants a somewhat tulip-shaped habit. Chlorosis of the leaf edges may occur when large quantities of ammonium become available. High rates of dicyandiamide also resulted in leaf scorch in lettuce. Usually, it is not possible to establish with any certainty whether the chlorosis and leaf scorch are the results of excessively high ammonium levels in the soil or whether they are caused by dicyandiamide. Other crops, in particular the *Brassicae* and also endive, are even more sensitive than lettuce. An application rate of 0.5 kg per 100 m² is already too high for these crops. Figure 2 is a black and white reproduction of a colour plate from Temme [16] which shows leaf damage to rye grown in soil dressed with dicyandiamide.



Figure 2. Leaf scorching in rye grown in soil dressed with dicyandiamide. (Taken from Temme, 1946.)

If a nitrification inhibitor is to be applied, the soil should be thoroughly leached first, as otherwise too much nitrate will remain available to the plants. In an experiment with lettuce, N-serve (0.51 per 100 m²) was applied with various quantities of calcium nitrate and ammonium sulphate (Table 3). As expected, N-serve performed better, the more of the nitrogen was applied in the form of ammonium.

,

Table 3. Yields and nitrate contents of lettuce (cv 'Orba') harvested on 9 April 1980, showing effects of different proportions of ammonium sulphate and calcium nitrate, applied mixed with N-serve (0.51 per 100 m²)

Ammonium sulphate (20% N), kg/100 m ²	Calcium nitrate (15.5% N), kg/100 m ²	Mean head weight, g	Nitrate content, mg NO, per kg fresh matter
10	0	314	2070
9	1.3	322	2060
8	2.6	328	2370
7	3.9	332	2430
6	5.2	339	2470
5	6.5	322	2960

No statistical analysis was carried out, the experiment had only two replications

Since the effect of leaching is greater on coarse — than on fine — textured soils, better results may be expected from the use of inhibitors on lighter soils. In author's experience, this was indeed the case.

Solution culture in which nitrogen is omitted some time before harvest

Growing lettuce on nutrient film (NFT) makes it possible to change the nitrogen supply instantly. The idea was to grow lettuce on NFT and to omit nitrogen from the nutrient solution when the crop was almost mature. Any excess nitrate in the crop would then be used up during the final stages of growth. The results obtained were disappointing.

Growth of the crop stopped almost immediately when demineralised water or nutrient solutions without nitrogen were introduced. The nitrate content of the crop decreased only very slowly. The explanation must be that nitrate is stored in the vacuoles and is very difficult to remove from there [11]. The nitrate in the vacuoles is often regarded as an osmoticum (Steingrover, pers. communication).

Besides using demineralised water to replace the nutrient solution, a solution of chlorides was tried in a number of experiments (see Table 4). The chloride solution gave a greater reduction in the nitrate content of the crop than demineralised water, but neither produced a particularly great satisfactory reduction. Alt and Struwe [2] obtained better results in terms of higher yield and lower nitrate content of lettuce. Their harvest (date not indicated) must have been under better light conditions. Under such conditions omitting nitrogen from the nutrient solution will give better results as growth rate is higher and leakage of nitrate from the vacuole is light-dependent (Blom-Zandstra, pers. communication).

Breeding

Lettuce cultivars have different nitrate contents. In experiments with modern cultivars at three experimental stations cultivars which were classed as having

Table 4. Yields and nitrate contents of lettuce (cv 'Salina') on NFT, harvested 9 and 19 days after change of nutrient solution. (Complete nutrient solution used in all plots until 5 November)

Treatments from 5 November onwards	Mean head weight, g	Nitrate content, mg NO ₃ per kg fresh matter
	harvested on 13 November	1981
Complete nutrient		
solution	172	4320
Demineralised water	171	3900
Chloride solution	174	3650
	harvested on 23 November	1981
Complete nutrient		
solution	196	4400
Demineralised water	156	2810
Chloride solution	168	2480

No statistical analysis was carried out

high nitrate levels in one experiment also had statistically high nitrate levels in the other two experiments.

However, the differences in nitrate contents of modern glasshouse cultivars were limited. To give some idea, they varied during winter conditions between 3500 and 4500 mg NO₃ per kg fresh produce. In the USA, Subramanya [15] published a report on the inheritance of nitrate-nitrogen accumulation in lettuce. This author mentions 'Valmaine' and 'Wonder van Voorburg' as cultivars with low nitrate contents. In our experiments, these cultivars were also classed in the low nitrate group.

At Wageningen, Eenink (pers. communication) is testing the lettuce cultivar gene bank for nitrate accumulation. The first results indicate that it must be possible to obtain lettuce varieties with low nitrate levels as a result of breeding. If the prognosis is correct, then it is likely that the nitrate reductions obtained via breeding could be many times greater than those achieved with nitrification inhibitors or by manipulation of the nutrient solutions. Reducing the nitrate contents by breeding is also the most secure method and economically the most attractive.

Some mention must be made of other vegetable crops. Breeding work on radish is in progress, be it not specifically for the purpose of reducing the nitrate contents. However, in The Netherlands and in other West European countries, there are vegetables which are grown under glass on a small scale and which have nitrate contents as high as 7000 mg NO₃ per kg fresh produce, such as purslane (Portulaca oleracea L.) and turnip tops (Brassica sp.; raapstelen in Dutch, Rübenstielchen in German). To the author's knowledge, crop improvement through breeding in these and perhaps many other vegetables has never been attempted. So the question arises whether it will be difficult to breed for low nitrate accumulation. Perhaps the study by Braaksma [4] could shed some light on these aspects.

Why is it necessary to reduce nitrate contents?

High nitrate contents in food for human consumption are considered undesirable for two reasons: a. the possibility of methaemoglobinaemia and b. the conversion of nitrate into nitrite in saliva which is thought to lead to the formation of carcinogenic nitrosamines in the intestinal tract. Leaving aside the risks to babies of high-nitrate foods, there are no cases recorded in the literature of methaemoglobinaemia having occurred in adults as a result of eating vegetables containing high nitrate levels and it probably has never happened. This leaves the question whether the consumption of vegetables with high nitrate contents increases the risk of cancer of the stomach or the colon. Research carried out in Colombia, [9] is often quoted in epidemiological discussions. Of three areas - A, B and C - investigated in the southern province of Narino, areas A and B were found to have an increased risk level of stomach cancer and more nitrate in the drinking water, compared with area C. However, the summary of this publication states: "The composite evidence from the comparisons indicated lettuce to be negatively associated with stornach cancer and raised the possibility of a positive association for corn". In the Dutch literature, it is sometimes suggested that there is an international norm for nitrate [12, 1]. However, in the reports of the WHO/FAO it is stated very clearly that the ADI (acceptable daily intake) standards apply only to food additives and not to the nitrate which is present naturally. There have been frequent discussions on this subject within the Commission (WHO 1962-1980) [18], but they have never led to a change in attitude. Finally, research is being conducted at the Roman Catholic University of Nijmegen on the use of ammonium nitrate for the treatment of kidney stones. The research worker concerned, Bruijns [5], gave patients oral doses of ammonium nitrate over prolonged periods of time, and tried to find nitrosamines in the saliva and the urine. The conclusion was: 'The hypothesis that the application of nitrate leads to clinically significant nitrosamine formation must be regarded as premature (Sen 1978, Lancet, Editorial 1977, Fraser 1980, Ruddell 1980)'.

The conclusion must be that a causal relationship between the consumption of vegetables with high nitrate contents and the incidence of intestinal cancer has never been demonstrated.

For recent publications in the field of toxicology Magee [10] and Ellen and Schuler [8] can be mentioned. As indicated by the title Magee discusses the aspects of nitrogen in various forms. Ellen and Schuler mentioned nitrate only. The last sentences of their concluding remarks are: "Scientific data applicable to man available at present provide no basis for alarm, but questions remaining unanswered justify a cautious approach. This means that extreme high nitrate levels in vegetables must be avoided, and that in general human exposure to nitrate and nitrite should be kept at an acceptable technological and economical minimum."

It is perhaps appropriate to end this short dissertation by commenting on an article published in a magazine for alternative life styles (Roest [13]). Roest recommends readers not to eat glasshouse produce, but only vegetables in season. Stinging nettles (*Urtica* spp.) and sorrel (*Rumex* spp.) are also recommended. Stinging nettles, collected from a coastal wood in December, were found to contain 6150 mg NO₃ per kg fresh matter. Stinging nettles have also been analysed in Switzerland where they were found to contain 2900 mg NO₃ per kg fresh matter in May [7].

References

- Aldershoff WG (1982) Nitraat in groenten en onze gezondheid. Bedrijfsontwikkeling 13, 273-280
- Alt D and Strüwe S (1982) Decline of the nitrate content in lettuce (Lactuca sativa var. capitata L.) by means of monitoring the nitrogen content of the nutrient solution in hydroponic systems. In: Plant Nutrition, Proc. 9th Internat. Plant Nutrition Colloquium Warwick, Aug. 22-27, CAB, Vol. 1, 17-21
- Bommeljé S (1981) Nitraatgehalten in groentegewassen. Resultaten inventarisatie 1979/1980. Consulentschap Bodemaangelegenheden Tuinbouw, Wageningen, 9 pp
- Braaksma F (1982) Genetic control of nitrite reduction in Arabidopsis thaliana. Diss. Rijksuniversiteit Groningen, 109 pp
- Bruijns E (1982) De behandeling met ammoniumnitraat bij nierstenen. Diss. Kath. Universiteit, Nijmegen, 220 pp
- Corré WJ and Breimer T (1979) Nitrate and nitrite in vegetables. Pudoc, Wageningen, 85 pp
- Eidgenössische Forschungsanstalt für Obst-, Wein- und Gartenbau, Wädenswil (1981) Nitrate sind kein Grund den Gemüsekonsum einzuschranken! Gartenb. (l'Horticulture Suisse) 102, 1471
- Ellen G and Schuller PL (1982) Nitrate, origin of continuous anxiety. Zur Gelegenheit des Abschlusskolloquiums des DFG. Bad Godesberg, 7-8 Oktober 1982 (in press)
- Haenszel W, Correa P, Cuello C, Guzman N, Burbano LC, Lores H and Munoz J (1976) Gastric cancer in Colombia. II Case-control epidemiologic study of precursor lesions. J. Natl. Cancer Inst. 57 (1976) 1021-1026
- Magee PN (1982) Nitrogen as a potential health hazard. Phil. Trans. R. Soc. Lond. B 296, 543-550
- Martinoia H, Heck U and Wiemken A (1981) Vacuoles as storage compartments for nitrate in barley leaves. Nature 289, 292-293
- Mol HJ (1979) Preventie van schadelijke stoffen in tuinbouwprodukten. Bedrijfsontwikkeling 10, 948-854
- 13. Roest B (1981) Hoe gevaarlijk is nitraat? Kleine Aarde 10, 38, 41-43
- 14. Roorda van Eysinga JPNL (1966) Bemesting van kropslaonder glas met bloedmeel en kalkammonsalpeter. Versl. Landbouwk. Onderz. 681, 18 pp
- 15. Subramanya R (1977) Inheritance of nitrate-nitrogen accumulation in lettuce (Lactuca sativa L.) Diss. Michigan State University, 84 pp
- Temme J (1946) Over de afbraak van Ca-cyaanamide in den grond. Diss. Landbouwhogeschool, Wageningen, 99 pp
- Temperli A, Kunsch U and Schärer H (1978/79) In: Eindgenössische Forschungsanstalt für Obst-, Wein- und Gartenbau, Wädenswil. Tätigkeitsbericht, 576
- World Health Organization: Evaluation of certain food additives. Report of the Joint FAO/WHO Expert Committee on Food Additives. 6th Report, WHO Techn. Rep. Series 228 (1962); 8th Report, WHO Techn. Rep. Series 309, 1964 (1965); 18th Report, WHO Techn. Rep. Series 557 (and FAO Nutrition Meetings Report Series 54) (1974); 23rd Report, WHO Techn. Rep. Series 648 (1980)

,