Available online at www.sciencedirect.com

SCIENCE DIRECT®

Food Chemistry 89 (2005) 235–242

Food Chemistry

www.elsevier.com/locate/foodchem

Content of nitrates, nitrites, and oxalates in New Zealand spinach

Grażyna Jaworska *

Department of Raw Materials and Processing of Fruit and Vegetables, Agricultural University of Krakow, Krakow, Poland

Received 27 May 2002; received in revised form 19 February 2004; accepted 19 February 2004

Abstract

The levels of nitrates, nitrites, and oxalates were evaluated in usable parts of New Zealand spinach destined for consumption and technological processing. In a two-year investigation, plants with stems up to 15 cm in length, grown from sowing, were obtained in seven harvests and from seedlings in eight harvests. The New Zealand spinach contained in fresh matter, 449–2804 mg NO_3^-/kg, 0.09–0.77 mg NO_2^-/kg, and 506–981 mg/100 g of total oxalates; of this amount 69–98% constituted the water-soluble oxalates. In dry matter, the respective values were 9.4–47.4 g/kg, 0.9–12.1 mg/kg, and 8.8–15.4 g/100 g, respectively. The content of the discussed compounds depended on the usable parts of the plant, the year of investigation, and the time of harvest, while the method of growing did not affect the above amounts. More nitrates and nitrites were found in the stems with leaves than in leaves alone. Plants harvested in July showed the smallest content of nitrites and the greatest of nitrates, and in September the smallest contents of nitrates and oxalates, but the greatest of nitrites.

Keywords: New Zealand spinach; Nitrate; Nitrite; Oxalate; Harvest time; Part of plant; Method of growing

1. Introduction

With respect to its utilization, the New Zealand spinach is classed in the group of leafy vegetables, thus potentially disposed to accumulate excessive amounts of nitrates and oxalates (Grevsen & Kaack, 1996; Kabaskalis, Tsitouridou, & Niarchos, 1995; Siomos & Dogras, 1999; Tabekhia, 1980). Currently, this crop has negligible economic importance in Poland and throughout the world. However, as the investigations by Kmiecik and Jaworska (1999) show, it is worth propagating because of its high and uniform yields and value, recommending it for the food processing industry (Jaworska & Słupski, 2001).

Nitrates, nitrites, and oxalates compounds unwanted in vegetables because of their unfavourable effects on the human organism (Crawford & Glass, 1998; Eichholzer & Gutzwiller, 1998; Kabaskalis et al., 1995). Hence, numerous investigations concern factors conducive to the accumulation of these compounds in plants. Benoit and Ceustermans (1988), Biemond, Vos, and Struik (1996), Grevsen and Kaack (1996), Paschold (1985), and Takebe, Ishihara, Matsuno, Fujimoto, and Yoneyama (1995) stress the effects of plant species, cultivar, level and type of fertilization, light conditions, temperature, humidity and structure of the soil, and even the application of plant protection agents. The content of oxalates depends, among other factors, on species and cultivar, forms of fertilizers (chiefly nitric ones) growing period, and phases of plant growth (Kabaskalis et al., 1995; Makus & Hettiarachchy, 1999; Takebe & Yoneyama, 1997).

The aim of this work was to determine the effect of cultivation methods, the year of investigation, and the harvest time on the levels of nitrates, nitrites, and oxalates, in stems with leaves and in leaves alone, of the New Zealand spinach.

2. Materials and methods

The investigation concerned the New Zealand spinach reproduced by the Dutch breeding company

* Fax: +48-12-662-47-57.
E-mail address: rrgjawor@cyf-kr.edu.pl (G. Jaworska).

0308-8146/$ - see front matter
doi:10.1016/j.foodchem.2004.02.030

Topstar, and usable parts of that spinach, i.e., stems not exceeding 15 cm in length with leaves and the leaves alone being analysed. The plants were grown over two years, using two methods: from seedlings and from sowing directly in the soil, and successively harvesting with the increasing maturity. Depending on the method of growing, three or four harvests were conducted each year.

The plantation of the investigated vegetable lay on the western outskirts of Krakow, on brown soil developed from loess formations of the mechanical composition of silt loam, in the third year after farmyard manuring, with French beans as the fore crop. The fertilizations determined on the basis of soil fertility and the nutritional requirements of the vegetable, amounted to 80 kg P_2O_5 ha^{-1} and 150 kg K_2O ha^{-1}, 50% of these

Table 1
Harvest time of New Zealand spinach and percentage share of the weight of leaves alone (L) in weight of whole plant (leaves + stems – LS)

Items	Year of investigation	Grown from seedlings				Grown from sowing			
		1	2	3	4	1	2	3	4
Harvest time, dd.mm	I	15.06	5.07	2.08	4.09	29.06	25.07	21.08	26.09
	II	11.06	9.07	6.08	4.09	2.07	5.08	3.09	–
L/LS, %	I	75.2	71.7	68.3	73.8	79.1	68.2	71.1	77.6
	II	71.3	71.0	73.1	73.8	74.7	74.0	77.9	–

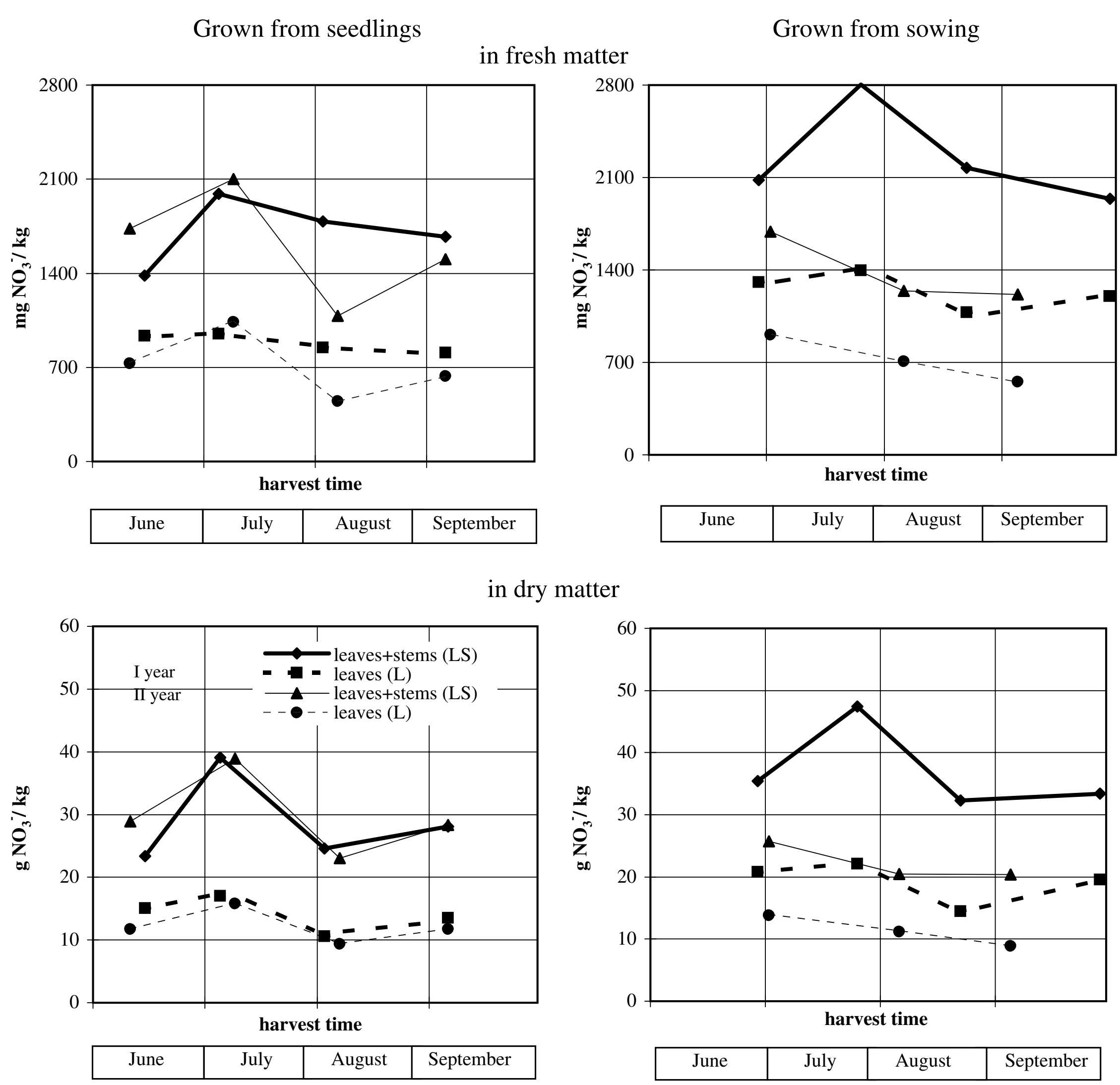

Fig. 1. Content of nitrates in New Zealand spinach.

Table 2
Mean contents of nitrates, nitrites, and oxalates in the New Zealand spinach depending on the usable part of the plant, the method of growing, the year of investigation, and time of harvest

Mean for		Nitrates		Nitrites		Soluble oxalates		Total oxalates	
		mg/kg fm[a] ±SD	g/kg dm[b] ±SD	mg/kg fm[a] ±SD	mg/kg dm[b] ±SD	mg/100 g fm[a] ±SD	g/100 g dm[b] ±SD	mg/100 g fm[a] ±SD	g/100 g dm[b] ±SD
Part of plant	Leaves + stems	1760 ± 439.9	30.0 ± 7.60	0.49 ± 0.205	8.3 ± 3.24	533 ± 57.0	9.1 ± 0.80	651 ± 84.4	11.1 ± 1.36
	Leaves	900 ± 264.0	14.6 ± 4.16	0.34 ± 0.160	5.4 ± 2.47	666 ± 78.8	10.8 ± 0.75	777 ± 105.9	12.6 ± 1.37
	LSD $\alpha = 0.01$	173.8	2.94	0.088	1.38	33.0	0.37	45.9	0.65
Method of growing	Seedlings	1227 ± 502.7	21.4 ± 9.33	0.38 ± 0.190	6.4 ± 3.04	590 ± 100.8	10.1 ± 1.06	701 ± 122.0	12.1 ± 1.67
	Sowing	1447 ± 608.1	23.3 ± 10.40	0.46 ± 0.203	7.3 ± 3.36	610 ± 89.0	9.7 ± 1.24	729 ± 103.9	11.6 ± 1.39
	LSD $\alpha = 0.01$	ns	ns	ns	ns	ns	ns	ns	ns
Year of investigation	I	1519 ± 560.9	24.8 ± 10.15	0.52 ± 0.138	8.4 ± 2.26	619 ± 104.5	9.9 ± 1.34	764 ± 99.8	12.2 ± 1.06
	II	1113 ± 485.3	19.4 ± 8.71	0.30 ± 0.195	5.1 ± 3.28	577 ± 79.6	10.0 ± 0.92	657 ± 103.5	11.4 ± 1.89
	LSD $\alpha = 0.01$	252.7	4.56	0.080	1.33	ns	ns	48.7	0.72
Harvest time	June	1361 ± 468.8	22.6 ± 8.22	0.47 ± 0.144	7.7 ± 2.56	657 ± 81.2	10.8 ± 1.10	717 ± 68.5	11.8 ± 0.92
	July	1610 ± 635.9	27.8 ± 11.76	0.34 ± 0.173	5.8 ± 3.25	579 ± 84.5	9.9 ± 1.39	678 ± 100.7	11.6 ± 1.89
	August	1171 ± 539.5	18.3 ± 7.79	0.37 ± 0.269	5.4 ± 3.59	612 ± 117.3	9.6 ± 0.86	807 ± 116.4	12.8 ± 1.44
	September	1186 ± 477.8	20.5 ± 8.45	0.50 ± 0.132	8.5 ± 2.17	564 ± 69.4	9.7 ± 0.92	654 ± 95.1	11.2 ± 1.31
	LSD $\alpha = 0.01$	365.4	6.28	0.129	2.01	61.3	0.74	66.7	0.99

[a] Fresh matter.
[b] Dry matter.

doses being applied in the autumn of the preceding year and 50% in spring of the year of growth. Nitrogen fertilization, at a dose of 30 kg N ha^{-1} in the form of ammonium nitrate, was applied during the spring cultivation of the field. Moreover, ammonium nitrogen was applied as side dressing for the first time, at a dose of 20 kg N ha^{-1}, three weeks after spinach transplanting and, for seeded plants, at the same developmental phase, and then after each harvest at a dose of 30 kg.

The seedlings of New Zealand spinach were produced in plastic rings 8 cm in diameter. The rings, each containing six plants, were planted at a spacing of 50×50 cm about the 10th of May. At the same time, sowing directly in the soil was carried out, with a row spacing of 50 cm. After emergence, the plants in rows were thinned, leaving an 8 cm distance between them to obtain the same number of plants per 1 m of the row. Harvesting was carried out when the plants reached the height accepted in the method, cutting them at 4 cm above the soil during the first harvest and 2 cm higher in further harvests. The obtained crop was sorted yellow, wilted, and disease-infected plants being discarded. The dates of harvesting the New Zealand spinach and the share of leaves in the shoots with leaves are given in Table 1.

Chemical analyses were carried out directly after harvesting, in four replications, each in two parallel samples. The determinations concerned the level of dry matter (AOAC, 1984), nitrates, and nitrites using the ISO method (ISO/6635, 1984), and water-soluble (Wilson-III, Shaw, & Knight, 1982) and total oxalates (AOAC, 1984).

The obtained results were statistically verified using the Snedecor F test and the Student's *t* test, the least significant difference being calculated for the probability level $\alpha = 0.01$.

3. Results and discussion

Most literature data concerning the growth of New Zealand spinach take leaves into consideration as its usable part. In the presented paper, both leaves alone and stems with leaves were analysed. The investigations conducted by Jaworska and Słupski (2001) show that stems with leaves can be used as a valuable raw material for freezing unless they have become outgrown. The utilization of whole plants also eliminates the labour-consuming act of separating leaves from stems. However, the results obtained by Kmiecik, Gębczyński, and Korus (2001), Oguchi, Weerakkody, Tanaka, Nakazawa, and Ando (1996), Ottosson (1979) and Watanabe, Uchiyama, and Yoshida (1994) demonstrate that the leaf blades, petioles and stems differ in their chemical composition. A particularly great differentiation, resulting from the various intensities of metabolic processes in different parts of the plant, concerns nitrates. The content of these compounds is much greater in stems and petioles than in leaves. Contrary to nitrates, oxalates accumulate chiefly in leaves (Gębczyński, 1998; Kabaskalis et al., 1995; Tabekhia, 1980). The regularities observed by the above authors were confirmed in the present work with respect to the New Zealand spinach.

Nitrates are not neutral for human health, hence their content in fresh vegetables is statutorily regulated. In Poland, the regulation issued by the Minister of Health (Dziennik Ustaw Rzeczpospolitej Polskiej, 2001) permits the content of 2500 mg NO_3^-/kg in fresh spinach. In the present experiment, this level was exceeded only in one harvest of stems with leaves. The New Zealand spinach contained 449–2804 mg NO_3^- in 1 kg fresh matter and 9.4–47.4 in 1 kg dry matter (Fig. 1). The above values were lower than those obtained by Jaworska and Słupski (2001) in another experiment with New Zealand spinach. In comparison with spinach, the New Zealand spinach usually accumulates similar amounts of the discussed compounds (Grevsen & Kaack, 1996; Jaworska & Kmiecik, 1999; Takebe et al., 1995; Watanabe et al., 1994). However, in the original literature, some works concerning spinach show that this species can accumulate over 5000 mg NO_3^-/kg fresh weight (Michalik, 1996; Watanabe et al., 1994). In comparison with leaves alone, the stems with leaves of the New Zealand spinach contained 48–137% more nitrates in fresh matter and 55–147% more in dry matter. At all the harvest times, differences were statistically significant. It should be stressed that, among all the factors of the experiment, the differentiation between averages for the usable parts was most significant (Table 2). The method of growing did not affect the level of nitrates, while the year of the investigation and the time of plant harvesting did affect it. In the first year of the investigation, the content of these compounds was 35% higher than in the second year. It was also observed that the greatest content could be found in plants harvested in July and the smallest in August and September, the difference amounting to 37%. The investigations carried out by Breś et al. (1991), Kmiecik et al. (2001) and Siomos and Dogras (1999) on dill showed a smaller tendency to nitrate accumulation in summer months than in spring or autumn. Bąkowski, Michalik, and Horbowicz (1996) and Jaworska and Kmiecik (1999) found a distinctly higher level of these compounds in the autumn yield of spinach than in the spring harvest. No effect of harvest time on the level of nitrates was observed by Watanabe et al. (1994) in spinach or by Jaworska and Kmiecik (1999) in New Zealand spinach. Throughout the entire experiment, the mean content of nitrates was 1330 mg in 1 kg fresh matter and 22.3 g in 1 kg dry matter. The maximum deviations from the above averages were −66% to +111% and −58% to +113%.

The New Zealand spinach contained 0.09–0.77 mg of nitrites in 1 kg fresh matter and 0.9–12.1 mg NO_3^- in 1

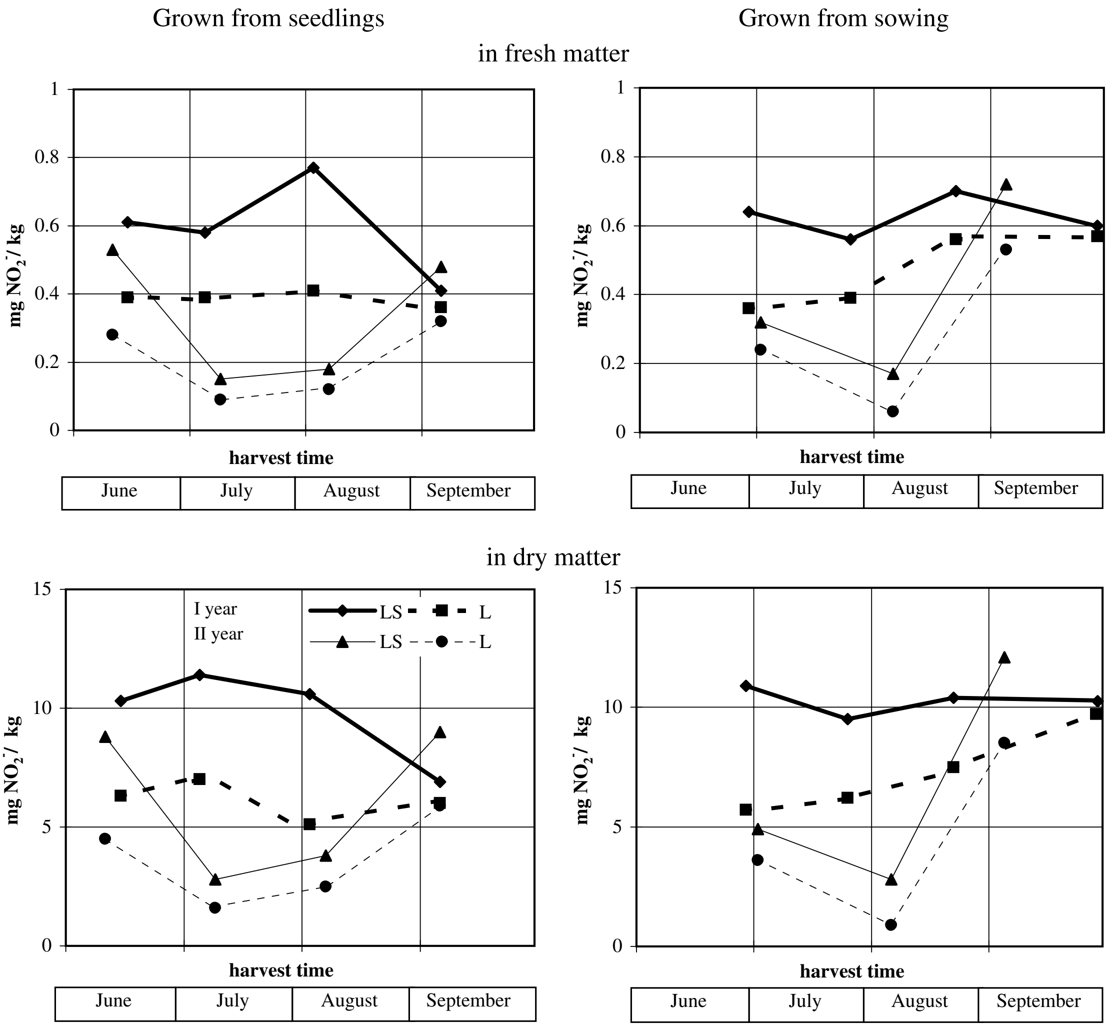

Fig. 2. Content of nitrites in New Zealand spinach.

kg dry matter (Fig. 2). Similar levels of these compounds in the New Zealand spinach, spinach, and parsley were reported by Jaworska and Kmiecik (1999), Jaworska and Słupski (2001) and Lisiewska and Kmiecik (1997). There are reports, however, that the level of nitrites in leafy vegetables can exceed 5 mg $NaNO_2$/kg fresh matter (Rostkowski, Borowska, Omieljaniuk, & Otłog, 1994). At all the harvest times, stems with leaves contained more nitrites than leaves alone, the differences ranging from 5% to 183% in fresh matter and from 7% to 196% in dry matter. The growing method did not affect the level of the discussed compounds, their amount depending on the year of the investigation and the harvest times. Moreover, the data in Table 2 suggest that the greatest effect on the level of nitrites is the year of plant growth. The mean content in fresh matter was 73% greater in the first year of growth than in the second. In dry matter, a 67% increase was noted. It should be stressed that the first year was hot and dry and the second decidedly cool and wet, as shown by the sums of temperatures and precipitation from May to September, amounting to 3284 °C and 333 mm and 2344 °C and 533 mm, respectively, for the two years. The mean content of nitrites was greater in June and September harvests and smaller in July and August. In all the treatments of the experiment, the mean content of nitrites was 0.42 mg/kg fresh matter and 6.9 mg/kg dry matter. The maximum deviation from the above averages ranged from −79% to +83% in fresh matter and from −87% to +75% in dry matter.

The content of water-soluble oxalates ranged from 442 to 827 mg in 100 g fresh matter and from 7.5 to 12.2 g in 100 g dry matter (Fig. 3). These values constitute 69–98% of total oxalate content. In earlier studies on this plant, Jaworska and Kmiecik (1999) also found a considerable share of the soluble form in the total content of oxalates, i.e., 78–81%. Watanabe et al. (1994) determined this share as 80–87% in spinach. According to other authors (Takebe et al., 1995; Yamanaka, Kuno, Shiomi, & Kikuchi, 1983), the ratio of soluble oxalates

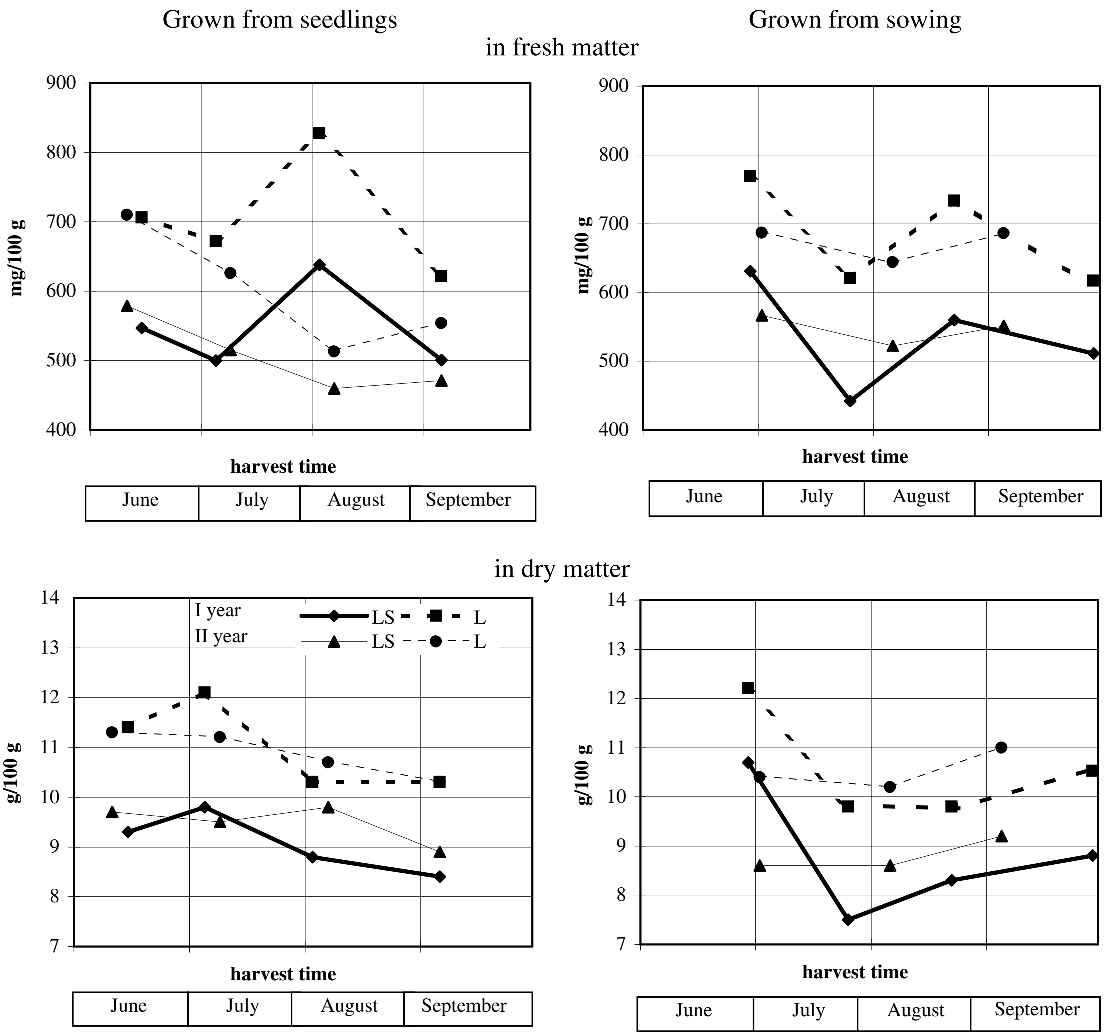

Fig. 3. Content of soluble oxalates in New Zealand spinach.

to total oxalates can be smaller, amounting to 54–68%. Of the factors analysed in this work, only the usable part of the plant and the harvest time affected the level of water-soluble oxalates, the effect of the former factor being more distinct (Table 2). The stems with leaves contained 10–29% smaller amounts of these compounds in fresh matter and 9–24% smaller in dry matter. The highest average content of soluble oxalates was found in plants harvested in June, the smallest content in fresh matter being noted in the September harvest and in dry matter in the remaining harvests, i.e., in July, August, and September. With the average content amounting to 599 mg soluble oxalates in 100 g fresh matter and 9.9 g in 100 g dry matter, for all the combinations of the experiment, the greatest deviations from the above values were −26% to +38% and −24% to +23%, respectively.

The data given in the literature, concerning the limit values of total oxalates in the New Zealand spinach, range from 276 to 894 mg/100 g fresh matter (Jaworska & Kmiecik, 1999; Yamanaka et al., 1983). In spinach, the respective values can range from 605 to 1760 mg/100 g fresh matter (Grevsen & Kaack, 1996; Yamanaka et al., 1983). In the present investigation, the obtained values for total oxalates were 506–981 mg/100 g fresh matter and 8.9–15.4 g/100 g dry matter (Fig. 4). The level of the above compounds depended on the usable part of the plant, the year of investigation, and the harvest time, being unaffected by the method of growing of the New Zealand spinach (Table 2). For the usable parts of plants, the differentiation between averages was 19% in fresh matter and 14% in dry matter in favour of the leaves, and for averages for the year of the investigation 16% and 7%, respectively, with an indication of the first year of growth. The greatest difference in the content of total oxalates was recorded between the harvest times. The highest content of the above compounds was found in August when, in fresh matter, the quantity of oxalates was 23% greater than in the September harvest, characterized by their lowest level. Watanabe et al. (1994) also demonstrated a higher ac-

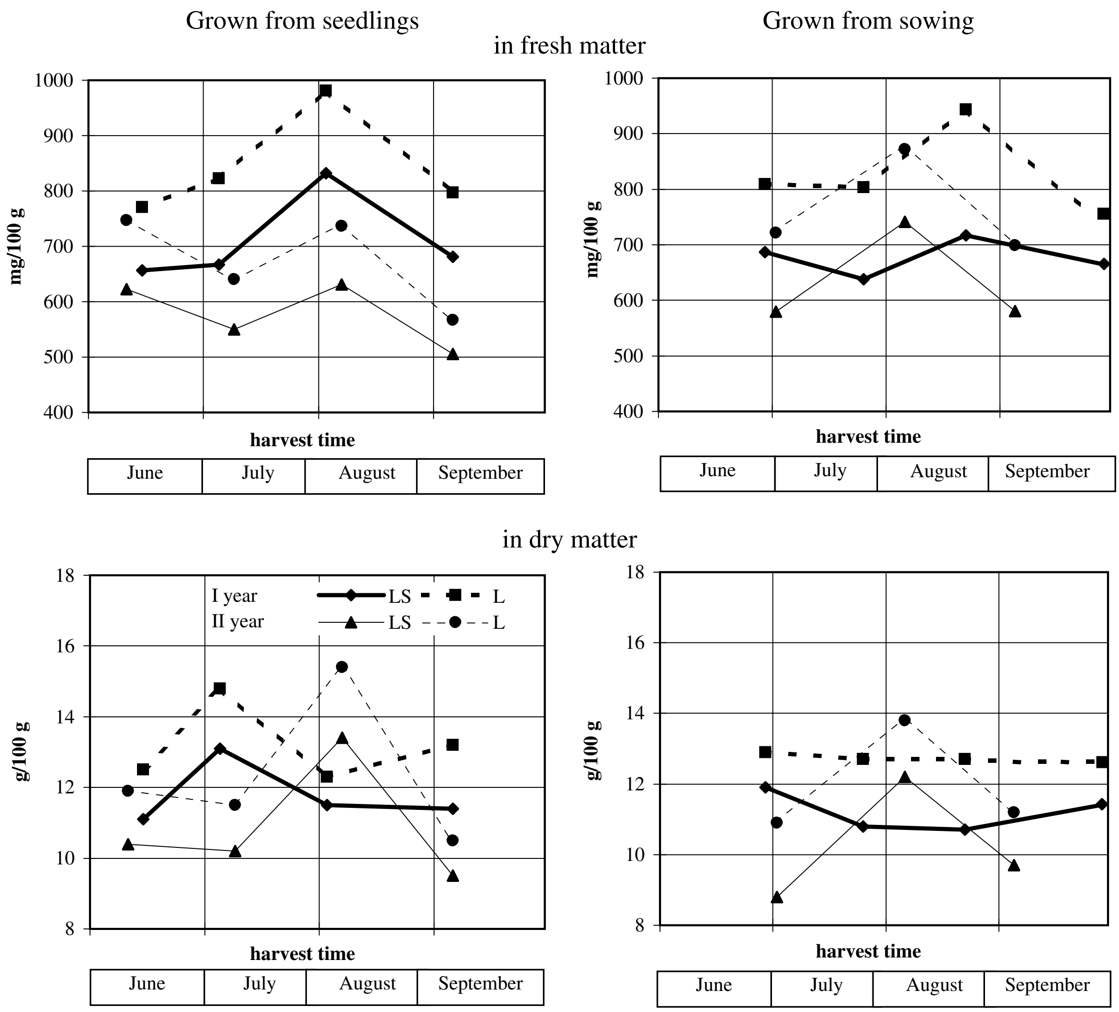

Fig. 4. Content of total oxalates in New Zealand spinach.

cumulation of oxalates in spinach from the summer period of growth than from the autumn harvest. The greatest deviations from an average for the entire experiment ranged from −29% to +37% in fresh matter and from −25% to +31% in dry matter.

4. Conclusion

In the New Zealand spinach, the contents of nitrates, nitrites, and oxalates depended on the usable part of the plant, the year of investigation, and the harvest time. The method of growing did not affect the level of these compounds. In stems with leaves, a 96% increase in the content of nitrates, 44% in nitrites, 20% in water-soluble oxalates, and 16% in total oxalates was recorded. In the first hot and dry year of the investigation, New Zealand spinach accumulated more of the investigated compounds than in the second cool and wet year. The harvest period had a considerably greater effect on the level of nitrates and nitrites than on the content of oxalates. In the entire period of the investigation, the deviations from averages exceeded 100% for nitrates and nitrites, amounting to 40% in the case of oxalates.

References

AOAC (1984). *Official methods of analysis of the Association of Official Analytical Chemists*. (14th ed. (pp. 611–613)). USA: Arlington.

Bąkowski, J., Michalik, H., & Horbowicz, M. (1996). Wpływ opakowania i warunków składowania na niektóre cechy jakościowe szpinaku. *Biuletyn Warzywniczy, 45*, 91–103.

Benoit, F., & Ceustermans, N. (1988). Phytotechnie et nitrates en culture maraîchère. *Revue de L'Agriculture, 41*(1), 23–43.

Biemond, H., Vos, J., & Struik, P. C. (1996). Effects of nitrogen on accumulation and partitioning of dry matter and nitrogen of vegetables. 3. Spinach. *Netherlands Journal of Agricultural Science, 44*, 227–239.

Breś, W., Komosa, A., Golcz, A., Kozik, E., Roszyk, J., & Tyksiński, W. (1991). Zawartość azotanów i azotynów w warzywach z targowisk w Poznaniu. *Poznańskie Towarzystwo Przyjaciół Nauk, Wydział Nauk Rolniczych i Leśnych, Prace Komitetu Nauk Rolniczych i Leśnych, 71*, 3–9.

Crawford, N. M., & Glass, A. D. M. (1998). Molecular and physiological aspects of nitrate uptake in plants. *Trends in Plant Science, 3*(10), 389–395.

Dziennik Ustaw Rzeczpospolitej Polskiej (2001). *Rozporządzenie Ministra Zdrowia z dn 27.12.2000 r. w sprawie wykazu dopuszczalnych ilości substancji dodatkowych i innych substancji obcych dodawanych do środków spożywczych lub używek, a także zanieczyszczeń, które mogą znajdować się w środkach spożywczych lub w używkach.*, nr 9, poz. 72, 5 luty, Warszawa.

Eichholzer, M., & Gutzwiller, F. (1998). Dietary nitrates, nitrites, and *N*-nitroso compounds and cancer risk: A review of the epidemiologic evidence. *Nutrition Reviews, 56*(4), 95–105.

Gębczyński, P. (1998). Wpływ wybranych czynników technologicznych na jakość mrożonych ogonków liściowych buraka liściowego. *Zeszyty Naukowe Akademii Rolniczej w Krakowie, s. Technologia Żywności, 342*, 41–48.

Grevsen, K., & Kaack, K. (1996). Quality attributes and morphological characteristics of spinach (*Spinacia oleracea* L.) cultivars for industrial processing. *Journal of Vegetable Crop Production, 2*(2), 15–29.

ISO/6635 (1984). Fruits, vegetables and derived products – Determination of nitrite and nitrate content – Molecular absorption spectrofotometric method. *International Organization for Standarization*, Geneve 20, Switzerland.

Jaworska, G., & Kmiecik, W. (1999). Content of selected mineral compounds, nitrates III and V, and oxalates in spinach (*Spinacia oleracea* L.) and New Zealand spinach (*Tetragonia expansa* Murr.) from spring and autumn growing seasons. *Electronic Journal of Polish Agricultural Universities, Food Science and Technology, 2*(2), Available from Internet: http://www.ejpau.media.pl/series/volume2/issue2/food/art.-03.html.

Jaworska, G., & Słupski, J. (2001). Badanie przydatności szpinaku nowozelandzkiego do mrożenia. *Żywność, 2*(27), 92–102.

Kabaskalis, V., Tsitouridou, R., & Niarchos, M. (1995). Study of oxalic acid content in vegetables and its implication on health. *Fresenius Environmental Bulletin, 4*, 445–448.

Kmiecik, W., Gębczyński, P., & Korus, A. (2001). Wpływ odmiany, rodzaju części użytkowej i terminu uprawy na zawartość azotanów V, III i szczawianów w koprze (*Anethum graveolens* L.). *Bromatologia I Chemia Toksykologiczna, 34*(3), 213–220.

Kmiecik, W., & Jaworska, G. (1999). Effect of growing methods of New Zealand spinach on its yield and pattern of harvests. *Folia Horticulturae, 11*(1), 75–85.

Lisiewska, Z., & Kmiecik, W. (1997). Effect of freezing and storage on quality factors in hamburg and leafy parsley. *Food Chemistry, 60*(4), 633–637.

Makus, D. J., & Hettiarachchy, N. S. (1999). Effect of nitrogen source and rate on vegetable amaranth leaf blade mineral nutrients, pigments and oxalates. *Subtropical Plant Science, 51*, 10–15.

Michalik, H. (1996). Zawartość azotanów w warzywach w zależności od gatunku i odmiany. *Hodowla Roślin i Nasiennictwo, 3*, 15.

Oguchi, Y., Weerakkody, W. A. P., Tanaka, A., Nakazawa, S., & Ando, T. (1996). Varietal differences of quality-related compounds in leaves and petioles of spinach grown at two locations. *Bulletin of the Hiroshima Prefectural Agriculture Research Center, 64*(7), 1–9.

Ottosson, L. (1979). Changes in ascoric acid in vegetables during the day and after harvest. *Acta Horticulturae, 93*, 435–442.

Paschold, P. J. (1985). Einfluss agrotechnischer Massnahmen auf den Nitratgehalt von Spinat. *Gartenbau, 32*(10), 297–299.

Rostkowski, J., Borowska, M., Omieljaniuk, N., & Otłog, K. (1994). Występowanie azotanów i azotynów we wczesnych warzywach i ziemniakach dostępnych w handlu Białegostoku w 1992 roku. *Roczniki Państwowego Zakładu Higieny, 45*(1/2), 81–87.

Siomos, A. S., & Dogras, C. C. (1999). Nitrates in vegetables produced in Greece. *Journal of Vegetable Crop Production, 5*(2), 3–13.

Tabekhia, M. M. (1980). Total and free oxalates, calcium, magnesium and iron contents of some fresh vegetables. *Deutsche Lebensmittel-Rundschau, 76*, 280–282.

Takebe, M., & Yoneyama, T. (1997). Effect of ammonium–nitrogen supply on oxalic acid content in spinach grown in hydroponics and fields. In T. Ando et al. (Eds.), *Plant nutrition – for sustainable food production and environment* (pp. 957–958). Dordrecht: Kluwer Academic Publishers.

Takebe, M., Ishihara, T., Matsuno, K., Fujimoto, J., & Yoneyama, T. (1995). Effect of nitrogen application on the contents of sugars, ascorbic acid, nitrate and oxalic acid in spinach (*Spinacia oleracea* L.) and komatsuna (*Brassica campestris* L.). *Japanese Journal of Soil Science and Plant Nutrition, 66*(3), 238–246.

Watanabe, Y., Uchiyama, F., & Yoshida, K. (1994). Compositional changes in spinach (*Spinacia oleracea* L.) grown in the summer and in the fall. *Journal Society for Horticultural Science, 62*(4), 889–895.

Wilson-III, Ch. W., Shaw, P. E., & Knight, R. J., Jr. (1982). Analysis of oxalic acid in carambola (*Averrhoa carambola* L.) and spinach by high-performance liguid chromatography. *Journal of Agricultural and Food Chemistry, 30*, 1106–1108.

Yamanaka, H., Kuno, M., Shiomi, K., & Kikuchi, T. (1983). Determination of oxalate in foods by enzymatic analysis. *Journal of the Food Hygienic Society of Japan, 24*(5), 454–458.