Effect of Sulphur Fertilisation on Lycopene Content and Colour of Tomato Fruits

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Abstract: The effects of different sulphur (S) fertilisers (ammonium, sodium, potassium and calcium sulphates) in combination with nitrogen (N) on plant growth, yield and quality of fruits were investigated in two dwarf cultivars Proton and Šejk. Single N, applied as ammonium nitrate, stimulated growth of plants and significantly increased yield of fruits, but did not change content of lycopene as well as colour parameters (a^*, b^* and L^*) and decreased significantly S content in fruits. All S fertilisers significantly increased S and lycopene content in fruits (up to 39% in cv. Šejk and 92% in cv. Proton) and positively influenced colour of tomato puree, namely parameter a^*. The earlier cv. Šejk responded better to S supply than cv. Proton, which showed a negative yield effect esp. on variants where higher S doses were applied. Sodium sulphate undesirably significantly enhanced Na content of fruits in both cultivars.

Keywords: tomatoes; lycopene; colour; sulphur; nitrogen; fertilisation

INTRODUCTION

Tomatoes are important source of lycopene in human nutrition and their utilisation in food has been increasing word wide. Annual consumption of tomatoes in the Czech Republic increased in 1999–2006 years from 8.9 to 12.0 kg/person (Czech Statistical Institute 2007).

Lycopene is providing protection against various cancers and other chronic diseases, probably due to its antioxidative properties (Giovannucci 1999) and it is mainly responsible for the red colour of tomato fruits (Clinton 1998). Biosynthesis of lycopene and other carotenoids in plants comes from mevalonic acid and AcCoA containing S-amino acid cystein. Cystein biosynthesis in plants is the first step through that inorganic S is incorporated into organic compounds and correlates significantly with availability of S for crops. It is a major reaction in the direct coupling between S and N metabolism in plant (Brunhold 1993).

In the last decade of the last century S deficiency becomes widespread in European agriculture, namely as a consequence of reduction in sulphur dioxide emissions and also low S inputs with farm yard manure, the use of low S-containing fertilisers, high-yielding crop varieties and a declining use of S-containing fungicides (Scherer 2001). In the Czech Republic atmospheric sulphur deposition had been dramatically decreasing since 1990. In the most of arable areas sulphur dioxide supply is below 10 kg S/ha that is insufficient to meet the requirements for this nutrient by intensively growing rape and probably by other crops (Zelený & Zelená 2002).

The objective of this study was to investigate response of two cultivars of tomatoes designed for field growing to different kinds of S fertilisers in combination with N on plant growth, yield of fruits, lycopene content and other quality parameters of fruits.

MATERIAL AND METHODS

Material and treatments. Two cultivars of dwarf tomatoes (*Lycopersicon esculentum*) Šejk and Proton designed for field growing and bred for high production of lycopene were examined during the year 2007. Tomato seedlings were grown in pots filled with 250 g of soil in growth chamber under constant conditions of temperature 25/20°C (day/night), photoperiod 16 h light, 8 h dark, illumination 250 μE/s/m. At one-month age the plants were transplanted, transferred into the greenhouse and finally grown till the harvest in pots with 6 kg of soil. In the experiment there was used only fresh topsoil from the field from the region of Josefov (Loc: 48°50'31.73''N, 17°0'58.4''E), South Moravia, where both tomatoes cultivars were grown in the large scale. Total CNS contents of soil were 1.167%; 0.128% and 0.021%, respectively. The experiment design consisted of 8 variants (Table 1) with 4 replications.

In control (C var.), no fertilisers were used. All other variants were fertilised with the same dose of N, in total 1 g N/plant. Second variant (N var.) was treated only with N in the form of ammonium nitrate (NH_4NO_3). Sulphur in the other variants was applied in the form of ammonium, sodium, potassium and calcium sulphates in total dose 0.25 and 0.5 g S/pot (plant). Dose of N in combined variants was balanced using NH_4NO_3. Only distilled water was used for watering. Plants were 2 times preventively treated again *Phytophtora*. .The experiment was finished after 4 month of plants growing. Ripen fruits were used for all following analyses. Part of each fruit was separated for lycopene and colour analysis and stored in deep freezer. The rest of fruits was dried and used for nutrients analysis using ICP-AES TraceScan.

Tomato processing before analysis of lycopene. 100–200 g of frozen fruits were homogenised in the mixer under nitrogen atmosphere to avoid the oxidation. Purée was poured through the fine sieve with holes 1 × 1 mm and packed into PE/PA heat-sealed bags, frozen and stored at –18°C till analysis.

Determination of lycopene. Frozen homogenates were thawed at refrigerator overnight at +5°C. 1–1.5 g homogenate was used for lycopene extraction following the procedure Bicanic *et al.* (2004). The mixture of hexane: acetone: ethanol (2:1:1) and 0.05% BHT was used for lycopene extraction. The top layer volume was read. 0.5–1.0 ml of hexane layer was dried by the gentle stream of nitrogen and redissolved in the mixture acetonitrile:methanol: tetrahydrofurane (30:55:15). The HPLC (Hewlett Packard 1100) under following conditions was used for lycopene determination:

Column: LichrospherR100 RP-18, 5 μm, 250 × 4 mm; pre-column: LichrospherR100 RP-18, 5 μm, 4 × 4 mm, temperature: 30°C, mobile phase: acetonitrile: methanol: tetrahydrofurane (40:55:5) with s 0.05% triethanolamine, flow rate: 1.5 ml per minute, sample loop 20 μl, spectrophotometeric detector λ = 475 nm. Standard of lycopene (Sigma) was used for peak identification and quantification.

Colour measurement. Thawed homogenate was filled into the white dish to create the layer with height of 20 mm. Minolta colour meter CR-300 (Japan) was used for prediction of the CIE L^*, a^*, b^* colour parameters. L^* is the brightness parameter, a* positive values represent red colour component and b^* positive values represent the yellow colour component. Each homogenate was measured at 10 places. Each place was measured by using the three flashes. Mean values and 95% confidentiality ranges were calculated.

Table 1. Variants of experiment

No.	Variant	Fertiliser	S	N
			(g/plant)	
1	C	control	0	0
2	N	NH_4NO_3	0	1.0
3	N+KS	K_2SO_4	0.25	1.0
4	N+NHS	$(NH_4)_2SO_4$	0.25	1.0
5	N+2KS	K_2SO_4	0.5	1.0
6	N+2NHS	$(NH_4)_2SO_4$	0.5	1.0
7	N+2NaS	Na_2SO_4	0.5	1.0
8	N+2CaS	$CaSO_4$	0.5	1.0

RESULTS AND DISCUSSION

The tomato cvs Šejk and Proton that were used in our experiment differed by growth rate, habitus, time of flowering and fruit ripening. Variety Proton had more vigorous growth, plants were higher, flowered later and fruits ripened later than cv. Šejk.

Yield and content of nutrients in tomatoes fruits

Fertilisation of plants with single N stimulated above all their longitudinal growth, growth of

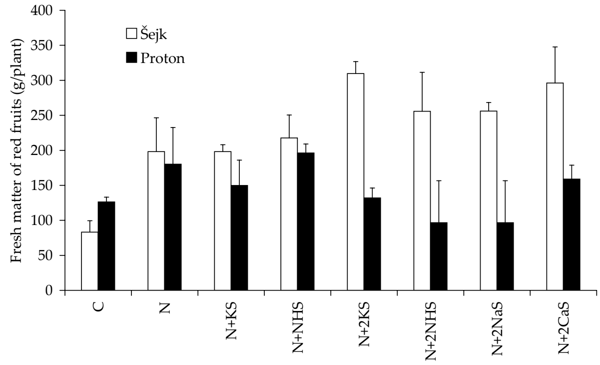


Figure 1. Effect of N in combination with S fertilisers on yield of red tomato fruits cv. Šejk and Proton. Data are expressed as average (n = 4) ± SD

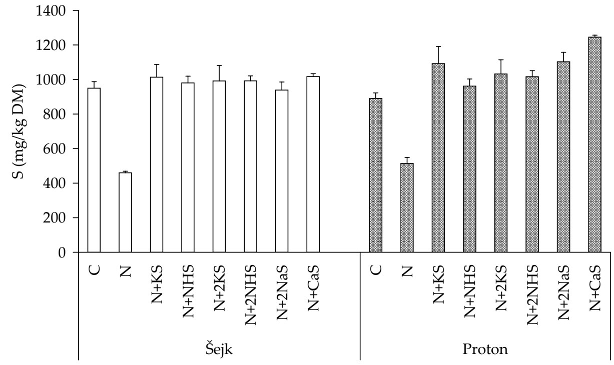


Figure 2. Effects of N in combinations with different kind of S fertilisers on S content in red tomato fruits cv. Šejk and Proton. Data are expressed as average (n = 4) ± SD

lateral branches in case cv. Proton and total matter production. It significantly increased yield of fruits of both cultivars (Figure 1), but significantly decreased S content in fruits to about 450 mg/kg of dry matter (Figure 2). The drop of S content in fruits may be partly explained by so called "dilution effect", considering that fruit matter in N variant was highly enhanced in comparison with control at the same S supply from the soil. Both cultivars responded similarly to S supplement by increase of S content in fruits however only in Šejk it increased also yield of fruits. Var. Proton showed even negative yield effect esp. on variants where higher S doses were applied, 0.5 g/plant. Sodium sulphate undesirably, very significantly enhanced Na content of fruits in both cultivars (Figure 3).

Lycopene content and colour of fruits

Influence of fertilisation on lycopene content is given in Figure 4. Single N significantly increased yield of fruits, but did not change content of lycopene as well as colour parameters (a^*, b^* and L^*) of fruit homogenate.

Supplement of S in all applied fertilisers manifested itself in both cultivares by significant increase of lycopene content in fruits and positively influenced colour of tomato puree, namely parameter of red colour component a^*. Increase of lycopene content after S supplement was higher in Proton but the differences between particular fertilisers were in Proton bigger than in Šejk (increase in average about 44.5% ± 28.9 and 32.15% ± 8.5, respectively). Ammonium sulphate had the

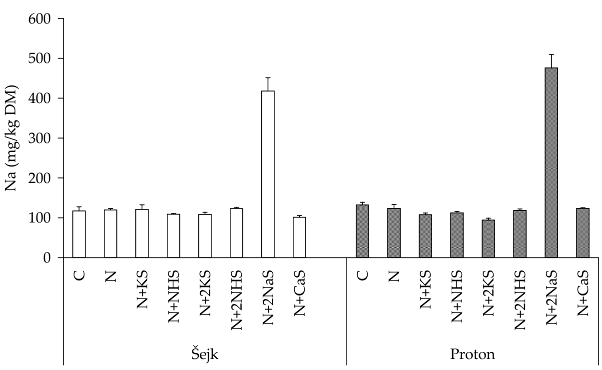


Figure 3. Effects of N in combinations with different kind of S fertilizers on Na content of red tomato fruits cv. Šejk and Proton. Data are expressed as average (n = 4) ± SD

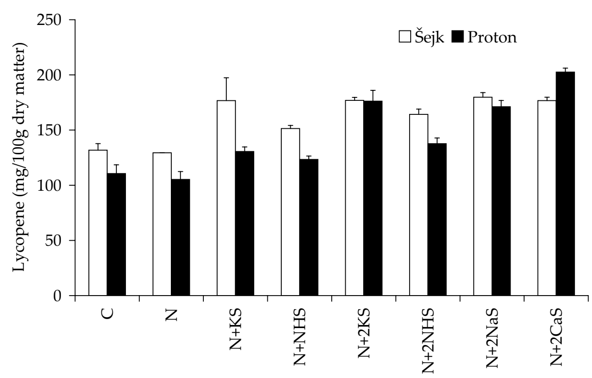
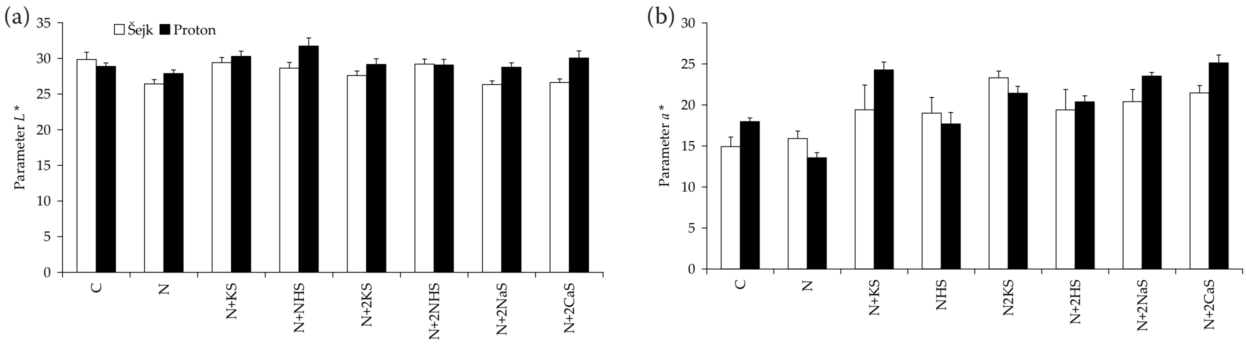


Figure 4. Effects of N in combinations with different kind of S fertilizers on lycopene content of red tomato fruits cv. Šejk and Proton. Data are expressed as average (n = 4) ± SD



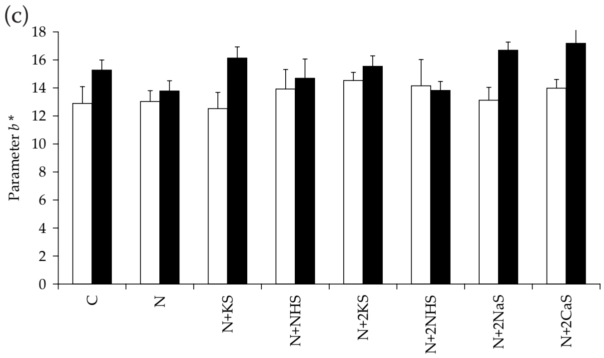


Figure 5. Effects of N in combinations with different kind of S fertilisers on colour parameters of red tomato fruit homogenates cv. Šejk and Proton (a) parameter L*, (b) parameter a*, (c) parameter b*. Data are expressed as average (n = 10) ± 95% confidentiality interval

smallest effect, namely in Proton. Further nutrients present in fertilisers may have additional effect. A lot of studies on lycopene content in tomatoes were done on variety effect, growing season, cultivation conditions, ripeness of fruits (Thompson *et al*. 2000), but we did not find any report on the effect of plant nutrition on lycopene biosynthesis, namely on effect of sulphur. Tomato is not ranged between plants highly demanding this nutrient. There may be cultivar differences in effect of S on plant growth and yield, like in case of Proton and Šejk. However S in both cultivars increased lycopene content in fruits, that is probably due to the importance of S in its biosynthesis.

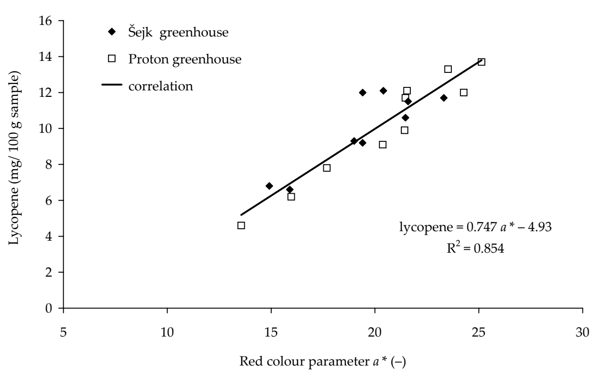


Figure 6. Correlation between lycopene content and red colour component parameter a* for Šejk and Proton cultivars

The higher content of lycopene influenced significantly the colour of homogenate from tomato fruits. Supplement of S in all combined variants significantly increased also the colour parameter a* of homogenate from fruit (Figure 5b).

We have tested the existence of the mathematical correlation between lycopene content and red colour component a* of homogenate of cv. Šejk and Proton (Figure 6).

It is apparent that very good correlation exists with correlation coefficient 0.924.

CONCLUSION

Sulphur applied in the form of different fertilisers (ammonium, sodium, potassium and calcium sulphates) in all cases significantly increased content of lycopene in fruits and red colour of tomato homogenate in both cultivars. Although it had very similar effect on content of lycopene in tomato fruits in both cultivars, growth of plants and yield of fruits were influenced differently in dependency on cultivars. Higher dose of S positively influenced yield of fruit in cv. Šejk, but decreased yield of fruits in cv. Proton.

Fertilisation with single N without S stimulated growth of plants and increased yield of fruits, but it did not change lycopene content in fruits and decreased content of S in fruits in both cultivars.

Natrium sulphate undesirably significantly increased Na content in fruits. Very good correlation exists between lycopene content and red colour component *a**.

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References

Bicanic D., Swarts J., Luterotti S., Pietraperzia G., Dóka O., De Rooij H. (2004): Direct quantification of lycopene in products derived from thermally processed tomatoes: Optothermal Windows as a selective, sensitive and accurate analytical method without the need for preparatory steps. Analytical Chemistry, **76**: 5203–5207.

Brunhold C. (1993):Regulatory interactions between sulfate and nitrate assimilation. In: De Kok L.J., Stulen I., Rennenberg H., Brunold C., Rauser W.E. (eds): Sulphur Nutrition and Assimilation in Higher Plants. SPB Academic Publishing, The Hague: 61–75.

Clinton S.K. (1998): Lycopene: chemistry, biology and implication for human health and disease. Nutritional Review, **56**: 35–51.

Giovannucci E. (1999): Tomatoes, tomato-based products, lycopene, and cancer. Review of the epidemiological literature. Journal of National Cancer Institute, **91**: 317–331.

Scherer H.W. (2001): Sulphur in crop production. European Journal of Agronomy, **14**: 81–111.

Thompson K.A., Marsnall M.R., Sims C.A., Wei C.I., Sargent S.A., Scott J.W. (2000): Cultivar, maturity, and heat treatment on lycopene content in tomatoes. Journal of Food Science, **65**: 791–795.

Zelený F., Zelená E. (2002): Changes in sulphur supply and need for oilseed rape in the Czech Republic. Phyton, **42**: 265–270.

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