

Sulphur nutrition index in relation to nitrogen uptake and quality of winter wheat grain

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ABSTRACT

A sulphur nutrition index (SNI) is an analogue of the N nutrition index, which is a widely used simple indicator of plant N status. The aim of this study was to relate the SNI (ratio of S concentration in shoot biomass to critical S concentration - S_c) to N uptake, grain yield and breadmaking quality of winter wheat (*Triticum aestivum* L.) grain during 4-yr small-plot field experiments realized under non-limiting N conditions in three different locations in the Czech Republic. The model of S dilution curve (S_c) developed by Reussi et al. (2012) for spring wheat was used for expression of the SNI. According to the model, optimal S concentration in shoot biomass was calculated using nothing but shoot biomass weight. The constant value of $S_c = 0.55$ was determined for shoot biomass weight lower than 1.0 t ha^{-1} . A very strong correlation was recorded between the SNI and an N to S weight ratio (N:S) in shoot biomass. Both, optimal N:S weight ratio in shoot biomass and qualitative parameters of grain (particularly Zeleny sedimentation volume, grain protein content and wet gluten content) were recorded if the SNI exceeded values of 0.80 at the beginning of stem elongation, 0.70 at the late boot stage, and even 0.60 at the beginning of heading despite the fact that the Reussi et al.'s model of S dilution curve was originally applicable only until the end of stem elongation. Correlation between the SNI and relative grain yield was weak.

Key words: Critical concentration, dilution curve, gluten, protein content, sulphur status, *Triticum aestivum*, yield.

INTRODUCTION

Low content of bioavailable S forms in soils was recorded e.g. in Poland (Siebielec et al., 2017). Concurrently, negative S balance was mentioned by Lozek et al. (2011) in soils in Slovakia. Concurrently, S uptake positively influences both uptake and utilization of N (Salvagiotti et al., 2009). Precise application of S particularly in intensive wheat production systems is then necessary (Flaete et al., 2005). A concurrent management of N and S is important to reduce potential pollution of residual soil nitrate by increasing N recovery from the soil while sustaining high N use efficiency (Salvagiotti et al., 2009).

Critical values for leaf concentrations of total S, sulphate and glutathione change over time and are not suitable for diagnosing S deficiency early in the growth season. In contrast, an N:S ratio in shoot biomass is a more reliable indicator of plant S status (Blake-Kalff et al., 2000). Optimal values of the N:S ratio in shoot biomass were expressed for various growth stages of wheat by many authors: after 6 wk of growth (Qian and Schoenau, 2007), during tillering (Reussi et al., 2011), during stem elongation (Calvo et al., 2008), during ear emergence (Steinfurth et al., 2012), and particularly in grain (Zhao et al., 1999; Tea et al., 2007).

However, one of the problems using the N:S ratio is that a surplus of one of these elements may be interpreted as a deficiency of the other one (Scherer, 2001). Furthermore, determination of the N:S ratio requires two precise analytical measurements (Blake-Kalff et al., 2000). Hence, Reussi et al. (2012) expressed S dilution critical curve from the beginning of tillering to the end of stem elongation of spring wheat. This model takes account of decreasing plant S concentration during wheat growth because the optimal S concentration in shoot biomass is calculated using nothing but shoot biomass

weight (Reussi et al., 2012). The calculation of S dilution critical curve enables to express the S nutrition index (SNI) analogous to the N nutrition index (NNI), which is widely used for quantifying the N status (Sadras and Lemaire, 2014). Sulphur nutrition index is calculated as a ratio of measured S content in shoot biomass and the critical S concentration calculated from the model of S dilution critical curve. For $SNI > 1$ the crop S status can be considered as non-limiting, and for $SNI < 1$ the crop S status can be considered as limited by S supply. On the other hand, the model according to Reussi et al. (2012) of S dilution critical curve is relevant to weight of shoot biomass higher than 1.0 t DM ha^{-1} , which is a problem for S status determination at earlier growth stages.

The aim of this study was to answer the following questions: Can SNI be considered as an appropriate indicator of winter wheat S status under Central European field conditions? Does the value of $SNI = 1$ lead to the highest grain yield and the best quality of winter wheat grain? Is it possible to use the S dilution critical curve for earlier or later growth stages of winter wheat than the model according to Reussi et al. (2012) states?

MATERIAL AND METHODS

Small-plot field experiments with winter wheat (*Triticum aestivum* L.) 'Sulamit' were realized during a 4-yr period (2007-2010) at three locations with different soil-climatic conditions in the Czech Republic (Central Europe): Hněvčeves (L1; $50^{\circ}18' \text{ N}$, $15^{\circ}42' \text{ E}$, 265 m a.s.l., Haplic Luvisol (FAO classification), average precipitations and temperature - 560 mm, 8.4° C , $\text{pH}(\text{CaCl}_2)$ 6.3, available nutrient contents (Mehlich 3): 89 mg P kg^{-1} , 292 mg K kg^{-1} , 158 mg Mg kg^{-1} , 2350 mg Ca kg^{-1}), Humpolec (L2; $49^{\circ}32' \text{ N}$, $15^{\circ}21' \text{ E}$, 525 m a.s.l., Gleic Cambisol, average precipitations and temperature - 678 mm, 6.7° C , $\text{pH}(\text{CaCl}_2)$ 6.1, available nutrient contents (Mehlich 3): 90 mg P kg^{-1} , 193 mg K kg^{-1} , 183 mg Mg kg^{-1} , 2250 mg Ca kg^{-1}) and Ivanovice na Hané (L3; $49^{\circ}18' \text{ N}$, $17^{\circ}5' \text{ E}$, 225 m a.s.l., Chernozem, average precipitations and temperature - 554 mm, 9.3° C , $\text{pH}(\text{CaCl}_2)$ 7.3, available nutrient contents (Mehlich 3): 137 mg P kg^{-1} , 390 mg K kg^{-1} , 245 mg Mg kg^{-1} , 4458 mg Ca kg^{-1}). Water soluble S varied among years, average contents were: 12.3 mg kg^{-1} for L1, 15.1 mg kg^{-1} for L2, and 8.2 mg kg^{-1} for L3.

The sowing density was 450 seeds m^{-2} , the size of each plot was 39 m^2 , of which 15 m^2 were harvested by a small-plot combine harvester; grain yield was determined by weighing grains from individual plots and the result was converted to 0% moisture. Samples of shoot biomass at the BBCH 30 growth stage (beginning of stem elongation), BBCH 45 growth stage (late boot stage) and BBCH 51 (beginning of heading) were taken from a 0.25 m^2 area. The N:S ratio was determined using the weight concentrations expressed in g kg^{-1} of N and S in shoot biomass according to Blake-Kalff et al. (2000).

Levels of 150 kg N ha^{-1} , 200 kg N ha^{-1} and 150 kg N ha^{-1} + 40 kg S ha^{-1} , respectively, were applied in mineral fertilizers using two techniques of fertilizers application: topdressing in three split N doses on the soil surface during spring vegetation and N fertilizers injection into soil at the beginning of spring vegetation, respectively. Each treatment had four replicates. Detailed characteristics of the experimental scheme are given by Sedlar et al. (2015).

Plant N status was assessed by critical N concentration (N_c) expressed by Justes et al. (1994). Nitrogen nutrition index (NNI) was calculated as a ratio of measured shoot total N concentration (%) and N_c (Sadras and Lemaire, 2014). To reduce the influence of N status on evaluation of S uptake, only plant samples with optimal range of N concentration in shoot biomass were included in our results, i.e. samples with $NNI = 0.90\text{-}1.10$ at the BBCH 30 and BBCH 45 growth stages and $NNI = 0.85\text{-}1.15$ at the BBCH 51 growth stage.

Total N concentration in shoot biomass was determined by the Kjeldahl method using the Vapodest 50s (C. Gerhardt, Königswinter, Germany). Grain protein content was expressed as $N \times 5.7$. Sulphur content in shoot biomass was determined using with optical emission spectroscopy with inductively coupled plasma (ICP-OES Agilent 720; Agilent Technologies, Santa Clara, California, USA). Zeleny sedimentation volume and a wet gluten content in DM of grain were determined using the NIR OmegaAnalyzer G (Bruins Instrument, Salem, Massachusetts, USA). Due to a significant effect of location on the previously mentioned variables, all variables were expressed as relative values related to the averages of individual locations (Table 1).

Critical S concentration (S_c) in shoot biomass was calculated according to Reussi et al. (2012):

$$S_c = 0.37 DM^{0.169} \quad [1]$$

where DM is shoot DM (t ha^{-1}). Based on our results, the constant value $S_c = 0.55$ was used for $DM < 1.0 \text{ t ha}^{-1}$ (see Results and Discussion).

Table 1. Average values of variables at individual experimental locations (L1-L3) across all experimental years.

Location	TGW	Zeleny sedimentation volume	GPC	WGC	Grain yield
	g	mL	—— % ——	—— % ——	t ha ⁻¹
L1	47.8c	45.7b	13.0a	26.5a	11.10a
L2	40.2a	52.2a	12.9a	24.9b	7.20b
L3	44.5b	52.8a	13.8b	28.8c	8.93c

Values within the column marked with the same letter are nonsignificantly different according Tukey test ($p \leq 0.05$).

TGW: Thousand grain weight; GPC: grain protein content; WGC: wet gluten content.

The SNI was calculated analogously to NNI expressed by Sadras and Lemaire (2014):

$$SNI = S_m/S_c \quad [2]$$

where S_m is total S content measured in shoot biomass (%) and S_c is a critical S concentration calculated from the model of S dilution critical curve (Reussi et al., 2012).

Linear regression and logarithmic regression were used for calculation of correlations among studied variables. A statistical analysis of data was carried out using Statistica 13 (TIBCO Software Inc., Palo Alto, California, USA). A standard ANOVA with the Tukey test was used to evaluate differences of variables among individual experimental locations.

RESULTS AND DISCUSSION

Critical S concentration for low shoot biomass

The calculation of critical S concentration for shoot DM weight lower than 1.0 t ha⁻¹ has not been defined by the model of S dilution critical curve (S_c) expressed by Reussi et al. (2012). However, in our experiments, 58% of all plant samples achieved lower weight of shoot DM than 1.0 t ha⁻¹ at the BBCH 30 growth stage indicating a need for definition of the critical S concentration for low shoot biomass. For the calculation of the NNI, Justes et al. (1994) and Yue et al. (2012) expressed a N_c for low shoot biomass by a constant value calculated as an average N concentration in shoot biomass of non-limiting N treatments.

Based on our results, the constant value of the critical S concentration for low shoot biomass was taken to be $S_c = 0.55$. The value of 0.55 is an average S concentration in shoot biomass when shoot DM did not exceed 1.0 t ha⁻¹ under non-limiting S conditions (data not shown). In that case, wheat plants had a N:S ratio in shoot biomass lower than 14.9:1 (median 7.32:1) at the BBCH 30 growth stage which was optimal for the highest grain yields of wheat according to the results of Blake-Kalff et al. (2000). To reduce the influence of plant N status on evaluation of S uptake, only plant samples with optimal N concentration in shoot biomass were included in our results, i.e. samples with values of NNI = 0.95-1.05 (0.99 median). On top of that, only plants with sufficient S content in grain i.e. grain N:S = 11-15:1 (median 12.6:1) (Tea et al., 2007) and simultaneously with sufficient S content in straw, i.e. straw N:S = 3-7 (4.79 median) (Steinfurth et al., 2012) were evaluated. The average S concentration was calculated out of 20 cases in total.

SNI at the BBCH 30 and 45 growth stages

Figure 1 describes a very strong correlation between the SNI at the BBCH 30 growth stage and the N:S ratio in shoot biomass at the BBCH 30 growth stage. After substituting the x-value in regression equation (Figure 1) for the upper limit of the optimal N:S ratio stated by Blake-Kalff et al. (2000), i.e. N:S = 14.9, SNI was equivalent to the value of SNI = 0.73 at the BBCH 30 growth stage.

The correlation between the SNI at the BBCH 45 growth stage and the N:S ratio in shoot biomass at the BBCH 45 growth stage was also very strong. After substituting the x-value in regression equation (Figure 2) for the N:S ratio determined by Calvo et al. (2008) at the BBCH 37 growth stage after the highest S supply, i.e. N:S = 11-13, SNI was equivalent to values of 0.84-0.72 at the BBCH 45 growth stage.

According to the findings of Hrivna et al. (2015) and Klikocka et al. (2016), S fertilization improves wheat grain yield, grain specific weight, protein content, gluten content and Zeleny sedimentation volume. Therefore, linear regressions

Figure 1. Correlation between the sulphur nutrition index at the BBCH 30 growth stage and the N:S ratio in shoot biomass at the BBCH 30 growth stage using the constant value of critical S concentration $[S_c] = 0.55$ for shoot DM < 1 t ha⁻¹ under non-limiting N conditions (N nutrition index = 0.90-1.10). The number of cases was 138.

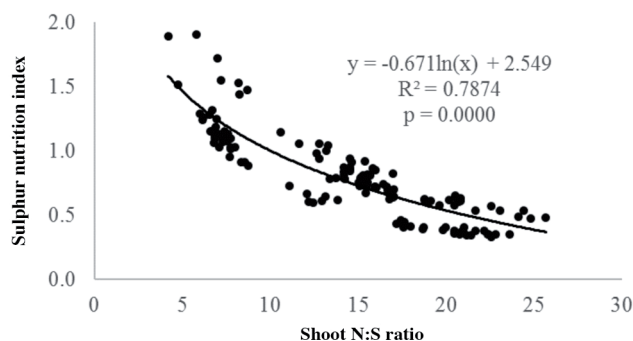
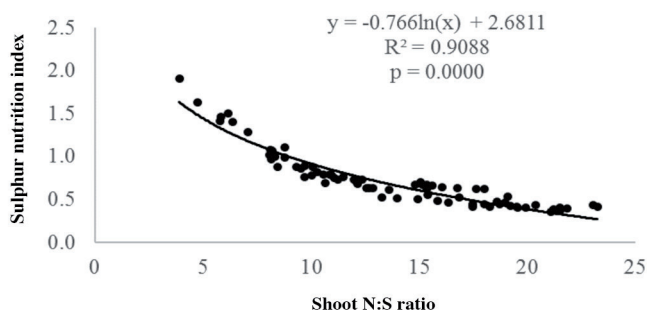


Figure 2. Correlation between the sulphur nutrition index at the BBCH 45 growth stage and the N:S ratio in shoot biomass at the BBCH 45 growth stage under non-limiting N conditions (N nutrition index = 0.90-1.10). The number of cases was 74.



describing correlations between the SNIs (y-values) at the BBCH 30 and BBCH 45 growth stages and both quantity and quality of winter wheat grain (x-values) are given in Table 2. The strongest correlation was recorded between the SNI at the BBCH 30 growth stage and relative Zeleny sedimentation volume in which case the correlation was moderate. The other variables correlated with the SNI at the BBCH 30 growth stage only weakly. To express the optimal values of the SNI, the x-value in regression equation was substituted for a value of 1 which is an average of studied relative variables. In that case, SNI achieved a value of 0.85 for average Zeleny sedimentation index. In case of SNI > 0.88, all studied relative parameters reached above-average values. With respect to grain quality of winter wheat, the SNI should be higher than the results suggest from correlation between the SNI at the BBCH 30 growth stage and the N:S ratio in shoot biomass at the BBCH 30 growth stage. On the other hand, only very weak correlations were recorded between the SNI at the BBCH 45 growth stage and studied relative variables (Table 2).

Table 2. Relations between relative both quantitative and qualitative parameters of winter wheat grain (x-values) and the sulphur nutrition index (SNI) (y-value) at the BBCH 30 (using the constant value of critical S concentration $[S_c] = 0.55$ for shoot DM < 1 t ha⁻¹) and BBCH 45 growth stage, respectively.

Relative parameter (x)	SNI 30 (y)			SNI 45 (y)		
	Linear regression equation	R ²	SNI-value if x = 1	Linear regression equation	R ²	SNI-value if x = 1
TGW	$y = 1.1338x - 0.2489$	0.111***	0.88	$y = 0.0228x + 0.6622$	0.000	0.69
Zeleny SV	$y = 0.6919x + 0.1563$	0.273***	0.85	$y = -0.0912x + 0.7599$	0.003	0.67
GPC	$y = 0.5941x - 0.0252$	0.051*	0.87	$y = -0.8242x + 1.5042$	0.032	0.68
WGC	$y = 0.766x + 0.1033$	0.095***	0.87	$y = -0.0644x + 0.7355$	0.001	0.67
Grain yield	$y = 0.5018x + 0.3726$	0.015	0.87	$y = -0.06255x + 1.2887$	0.032	0.66

*, **, ***Significant at the 0.05, 0.01, and 0.001 probability levels, respectively.

TGW: Thousand grain weight; Zeleny SV: Zeleny sedimentation volume; GPC: grain protein content; WGC: wet gluten content.

SNI at the beginning of heading

According to the results of Reussi et al. (2012), the model of S dilution curve was originally applicable only to the end of stem elongation of wheat. However, a very strong correlation was found between the SNI at the BBCH 51 growth stage and an N:S ratio in shoot biomass at the BBCH 51 growth stage (Figure 3). After substituting the x-value in regression equation (Figure 3) for the N:S ratio in shoot biomass stated by Steinfurth et al. (2012) as the optimum for the ear emergence growth stage, i.e. N:S = 13-14, SNI was equivalent to values of 0.64-0.59 at the BBCH 51 growth stage.

Moderate correlation was recorded between the SNI at the BBCH 51 growth stage and a relative Zeleny sedimentation volume and relative wet gluten content (Table 3). After substituting the x-value in regression equation (Table 3) for an average value of variable, i.e. $x = 1$, the SNI corresponded to values of SNI = 0.61 and SNI = 0.60, respectively. These values comply with the optimal SNI values, i.e. SNI = 0.64-0.59, derived from the relation between the SNI and the N:S ratio in shoot biomass for the BBCH 51 growth stage (Figure 3).

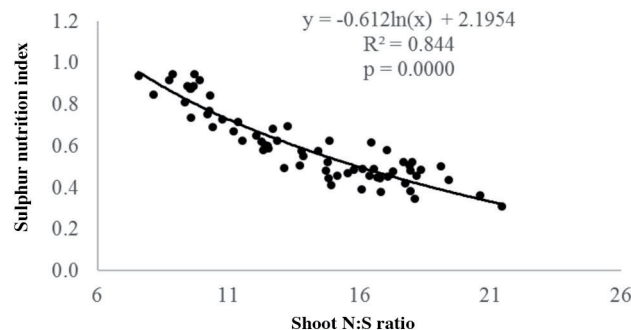
Table 3. Relations between relative both quantitative and qualitative parameters of winter wheat grain (x-values) and the sulphur nutrition index (SNI) (y-value) determined at the BBCH 51 growth stage.

Relative parameter (x)	Linear regression equation	R ²	SNI-value if x = 1
TGW	$y = 1.0357x - 0.4337$	0.029	0.60
Zeleny SV	$y = 0.6511x - 0.0380$	0.196***	0.61
GPC	$y = 0.9152x - 0.3171$	0.062*	0.60
WGC	$y = 1.0097x - 0.4094$	0.158***	0.60
Grain yield	$y = 0.6295x - 0.0401$	0.070*	0.59

*, **, ***Significant at the 0.05, 0.01, and 0.001 probability levels, respectively.

TGW: Thousand grain weight; Zeleny SV: Zeleny sedimentation volume; GPC: grain protein content; WGC: wet gluten content.

Figure 3. Correlation between the sulphur nutrition index at the BBCH 51 growth stage and the N:S ratio in shoot biomass at the BBCH 51 growth stage under non-limiting N conditions (N nutrition index = 0.85-1.15). The number of cases was 66.



The SNI at the BBCH 51 can be utilizable with respect to late S fertilization because Steinfurth et al. (2012) state that S fertilization at ear emergence growth stage prevents S deficiency in late stages of wheat growth. Zorb et al. (2009) state that the late S fertilization was found to improve the composition of gluten proteins and baking quality of winter wheat grain.

The fact that a nutrition index equal to 1 may not be always optimal for yield and quality of harvested products was also shown in case of NNI of various crops (Debaeke et al., 2012; Hu et al., 2014; Sedlar et al., 2017).

The SNI only weakly correlated with relative grain yield of winter wheat. Klikocka et al. (2016) also recorded higher effect of S fertilization on grain quality of wheat rather than on grain yield. According to the results of Kulhanek et al. (2014), nonsignificant effect of S fertilization on grain yield of winter wheat was also found under field conditions of the Czech Republic.

CONCLUSIONS

Calculation of the S nutrition index (SNI) using the model of Reussi et al. (2012) of S dilution curve was proved to be a reliable indicator of winter wheat S nutrition status even at the beginning of heading (BBCH 51). Calculation of the SNI was feasible also at the beginning of spring vegetation when shoot dry biomass weight was lower than 1.0 t ha⁻¹. In that case, the constant value of the critical S concentration in shoot biomass 0.55 was determined. Both optimal N:S weight ratio in shoot biomass and qualitative parameters of grain (particularly Zeleny sedimentation volume, grain protein content and wet gluten content) were recorded if the SNI exceeded values of 0.80 at the beginning of stem elongation (BBCH 31), 0.70 at the late boot stage (BBCH 45) and 0.60 at the beginning of heading (BBCH 51). Correlation between the SNI and relative grain yield was weak.

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