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Nitrogen Uptake by Winter Wheat (*Triticum aestivum* L.) Depending on Fertilizer Application

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The influence of the injection of nitrogen fertilizers with the CULTAN system (Controlled Uptake Long Term Ammonium Nutrition) on nitrogen uptake by winter wheat (*Triticum aestivum* L.) was observed at small-plot field experiments under conditions of the Czech Republic (central Europe) during 2007–2013. The CULTAN system consisting in the injection of all the nitrogen in one dose was compared with conventional broadcast surface fertilization which is carried out in three partial nitrogen doses. The total nitrogen dosage was 150 kg N.ha^{-1}. If the CULTAN fertilization was carried out at the beginning of tillering of winter wheat (BBCH 22) instead of at the end of tillering (BBCH 29), the CULTAN-treated winter wheat did not suffer from nitrogen deficiency at the BBCH 45 (boot stage) and BBCH 51 (beginning of heading) growth stages. Nitrogen utilization efficiency and biomass production efficiency were significantly higher with the CULTAN treatment compared to the conventional fertilization whereas nitrogen uptake efficiency tended toward lower values with the CULTAN treatment. Nitrogen use efficiency (NUE), post-heading nitrogen uptake, the contribution of nitrogen translocation to the total nitrogen in grain, the partial factor productivity of nitrogen as well as grain yield were not significantly influenced by the CULTAN system. Prolonged nitrogen uptake from the soil with the CULTAN treatment resulting in delayed plant senescence was not confirmed. Neither an application of sulphur-containing fertilizer nor the increased dosage of nitrogen (200 kg N.ha^{-1}) positively affected the studied parameters.

Keywords: depot, leaching and losses, nitrification, nitrate, remobilization

Introduction

The application of slow and controlled release fertilizers reduces environmental pollution in terms of hazardous gaseous emissions and water eutrophication (Azeem et al. 2014) and simultaneously enhances nitrogen use efficiency (Ladha et al. 2005) which is an important presupposition for the increased profit of production (Beatty et al. 2010; Gaju et al. 2011). However, the use of these fertilizers is substantially limited by their higher cost (Zebarth et al. 2009).

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The CULTAN (Controlled Uptake Long Term Ammonium Nutrition) system consists in the injection of the fertilizer with a significant ratio of nitrogen in ammonium form into the root space of plants. The place of fertilizer application in the soil is called the depot. The positive charge of ammonium and the high concentration of fertilizer in these depots result in the higher stability of fertilizer in the soil. Thus, with the CULTAN treatment, nitrogen can be applied in one dose using a common liquid fertilizer (Sommer and Scherer 2009; Kubesova et al. 2013). High stability of nitrogen in the depots and controlled ammonium uptake by plants lead to a prolonged vegetation period (Sommer and Scherer 2009) which is an important prerequisite for enhanced yield and better production quality (Pask et al. 2012).

Nitrogen uptake by CULTAN-treated spring barley is more gradual in comparison with the conventionally treated one (Sedlar et al. 2013) which corresponds with the results of Kozlovsky et al. (2010) and Peklova et al. (2012) who stated that CULTAN fertilization tends toward lower nitrogen content in winter wheat grain and winter rape seed respectively. A lower accumulation of nitrogen in plant biomass results in higher nitrogen utilization efficiency (Pask et al. 2012). Moreover, higher nitrogen rates decrease the efficiency of fertilization (Uzik et al. 2012) which can be maintained particularly by excellent management practices (Li et al. 2013).

The aim of this study is to investigate the long-term effect of fertilizer applied by the CULTAN system and the ability of nitrogen fertilizer injection to improve nitrogen use efficiency resulting in increased grain yield as well as the environmental and economic benefits.

Material and Methods

Small-plot field experiments with winter wheat (*Triticum aestivum* L.) cultivar Sulamit (sowing density of 450 seeds per m^2, size of a fertilized plot – 39 m^2, 15 m^2 harvested plot) were conducted during 2007–2013 at three sites with different soil-climatic conditions in the Czech Republic (central Europe): Hněvčeves (annual average precipitation 597 mm, annual average temperature 8.1 °C, clay loam, pH 5.9, available content of nutrients ($mg.kg^{-1}$): P 71, K 175, Mg 94, Ca 2092), Humpolec (annual average precipitation 667 mm, annual average temperature 6.5 °C, sandy loam, pH 4.9, available content of nutrients ($mg.kg^{-1}$): P 83, K 113, Mg 115, Ca 2255) and Ivanovice na Hané (annual average precipitation 548 mm, annual average temperature 9.2 °C, loam, pH 6.8, available content of nutrients ($mg.kg^{-1}$): P 177, K 573, Mg 215, Ca 3988). At the Ivanovice na Hané site the experiment was conducted only during 2007–2012.

The conventional method of surface broadcast fertilization with three partial doses of calcium ammonium nitrate was compared with the injection of urea ammonium nitrate applied in one dose of fertilizer with the GFI 3A injection machine (Maschinen und Antriebstechnik GmbH Güstrow, Germany) to a depth of 5–10 cm into the soil. Both treatments were evaluated in two amounts of nitrogen fertilizers: 150 kg $N.ha^{-1}$ and 200 kg $N.ha^{-1}$. The effect of the nitrogen fertilizer containing sulphur was simultaneously examined: ammonium sulphate was applied instead of the first dose of the calcium ammonium nitrate in the conventional treatment and urea ammonium sulphate was applied instead of the urea ammonium nitrate in the CULTAN treatment. The total dosage of applied sulphur in the conventional and the CULTAN treatment was 47 kg S.ha^{-1} and 50 kg S.ha^{-1}, respectively (Table 1). The fertilizer types used in the experiment were specifically chosen to put our results into practice as effortlessly as possible.

Table 1. Fertilizer treatment, N amounts and timing of spring application of N-fertilizers

Treatment	Nitrogen dosage per ha (fertilizer)				Total nitrogen dosage per ha
	BBCH 22	BBCH 22 (29)*	BBCH 33	BBCH 52	
conv. 150	43 kg (CAN)		87 kg (CAN)	20 kg (CAN)	150 kg
CULTAN 150		150 kg (UAN)			150 kg
conv. 200	60 kg (CAN)		90 kg (CAN)	50 kg (CAN)	200 kg
CULTAN 200		200 kg (UAN)			200 kg
conv. + S	43 kg (AS)		87 kg (CAN)	20 kg (CAN)	150 kg
CULTAN + S		150 kg (UAS)			150 kg

Notes: * the CULTAN fertilization was carried out at the BBCH 29 growth stage in the years 2007–2008 (Experiment I) and at the BBCH 22 growth stage in the years 2009–2013 (Experiment II)
CAN – Calcium Ammonium Nitrate (27% N); UAN – Urea Ammonium Nitrate (30% N); AS – Ammonium Sulphate (20.5% N, 24% S); UAS – Urea Ammonium Sulphate (19% N, 6% S)

The experiment is divided into two parts depending on the growth stage of winter wheat in which the CULTAN fertilization was carried out: in experiment I, which was conducted in the years 2007 and 2008, the CULTAN fertilization was done at the end of tillering (BBCH 29) according to Sommer and Scherer (2009). In contrast, in experiment II, which was conducted in the years 2009–2013, the CULTAN fertilization was carried out at the beginning of tillering (BBCH 22) according to the recommendations of Kozlovsky et al. (2010) for the soil-climatic conditions of the Czech Republic.

Samples of the aboveground biomass in the growth stages of BBCH 45 (boot stage; to check the nitrogen content in aboveground biomass just after an intense nitrogen uptake during erect growth) and BBCH 51 (heading 1/4 complete, i.e. usual term of so-called qualitative fertilization used for increasing grain protein content) were taken from a 0.25 m^2 area. The total nitrogen concentration in the aboveground biomass was determined by the Kjeldahl method using the Vapodest 50s (Gerhardt, Königswinter, Germany). The mineral nitrogen content in the topsoil (0–30 cm) before the CULTAN fertilization was determined in the infusion of 0.01 mol.l^{-1} $CaCl_2$ (Houba et al. 1986) using a segmented flow analysis with colorimetric determination on the SKALAR SAN plus SYSTEM (Skalar Analytical, Breda, Netherlands).

Statistical analysis of the data was carried out using the Statistica version 12 (StatSoft, Tulsa, USA). A standard analysis of the variance (ANOVA) procedures with the Fisher LSD test was used to calculate significant differences between the individual treatments of nitrogen fertilization at a $P < 0.05$ level of significance. The results are presented across the three sites and all experimental years.

Calculations

Aboveground N content at maturity

$$\mathrm{AGNM} = (\mathrm{GN} \times \mathrm{GY}) + (\mathrm{SN} \times \mathrm{SY})\ (\mathrm{kg\ N.\,ha^{-1}})$$

where GN is grain N concentration (%), GY is grain yield (t.ha^{-1}), SN is straw N concentration (%) and SY is straw yield (t.ha^{-1}).

Nitrogen nutrition index at the boot stage (BBCH 45) and at the beginning of tillering (BBCH 51)

$$\mathrm{NNI} = \frac{\mathrm{N_t}}{\mathrm{N_{et}}}$$

where N_t is total nitrogen concentration measured and N_{et} is a critical nitrogen concentration corresponding to the amount of shoot dry matter produced (Justes et al. 1994):

$$\mathrm{N_{et}} = 5.35 \times \mathrm{DM}^{-0.442}$$

Nitrogen utilization efficiency

$$\mathrm{NUtE} = \frac{\mathrm{GY}}{\mathrm{AGNM}}\ (\mathrm{kg\ grain\ DM\ .\ kg^{-1}\ N\ uptake\ at\ maturity})$$

where GY is grain yield (t.ha^{-1}) and AGNM is aboveground N content at maturity (kg N.ha^{-1}) (Cerny et al. 2012).

Nitrogen uptake efficiency

$$\mathrm{NUpE} = \frac{\mathrm{AGNM}}{\mathrm{SAN} + \mathrm{FN}}\ (\mathrm{kg\ N\ uptake\ at\ harvest\ .\ kg^{-1}\ available\ N\ from\ soil\ and\ fertilizer})$$

where AGNM is aboveground N content at maturity (kg N.ha^{-1}), SAN is soil available N in spring (kg N.ha^{-1}) and FN is N applied in fertilizer, i.e. 150 kg N.ha^{-1} and 200 kg N.ha^{-1}, respectively (Gaju et al. 2011).

Nitrogen use efficiency

$$\mathrm{NUE} = \frac{\mathrm{GY}}{\mathrm{SAN} + \mathrm{FN}}\ (\mathrm{kg\ grain\ DM\ .\ kg^{-1}\ available\ N\ from\ soil\ and\ fertilizer})$$

where GY is grain yield (t.ha^{-1}), SAN is soil available N in spring (kg N.ha^{-1}) and FN is N applied in fertilizer, i.e. 150 kg N.ha^{-1} and 200 kg N.ha^{-1}, respectively (Gaju et al. 2011).

Contribution of nitrogen translocation to total nitrogen in grain

$$\mathrm{NTRg} = \frac{\mathrm{AGNH} - \mathrm{SN} \times \mathrm{SY}}{\mathrm{GY} \times \mathrm{GN}} \times 100\ (\%)$$

where AGNH is aboveground N content at the beginning of heading (BBCH 51) (kg $N.ha^{-1}$), SN is straw N concentration (%), SY is straw yield ($t.ha^{-1}$), GY is grain yield ($t.ha^{-1}$) and GN is grain N concentration (%) (Uzik and Zofajova 2012).

Post-heading (BBCH 51) nitrogen uptake

$$\mathrm{PHNU} = \mathrm{AGNM} - \mathrm{AGNH}\ (\mathrm{kg\ N.ha^{-1}})$$

where AGNM is aboveground N content at maturity (kg $N.ha^{-1}$) and AGNH is aboveground N at heading (BBCH 51) (kg $N.ha^{-1}$) (Sedlar et al. 2014).

Nitrogen remobilization efficiency

$$\mathrm{NRE} = \frac{\mathrm{AGNH} - (\mathrm{AGNM} - \mathrm{GY} \times \mathrm{GN})}{\mathrm{AGNH}}$$

where AGNH is aboveground N content at the beginning of heading (BBCH 51) (kg $N.ha^{-1}$), AGNM is aboveground N content at maturity (kg $N.ha^{-1}$), GY is grain yield ($t.ha^{-1}$) and GN is grain N concentration (%) (Arduini et al. 2006).

Nitrogen harvest index

$$\mathrm{NHI} = \frac{\mathrm{GY} \times \mathrm{GN}}{\mathrm{AGNM}}$$

where GY is grain yield ($t.ha^{-1}$), GN is grain N concentration (%) and AGNM is aboveground N content at maturity (kg $N.ha^{-1}$) (Montemurro et al. 2006).

Biomass production efficiency

$$\mathrm{BPE} = \frac{\mathrm{GY} + \mathrm{SY}}{\mathrm{AGNM}}\ (\text{kg aboveground DM . kg}^{-1}\text{ N uptake at maturity})$$

where GY is grain yield ($t.ha^{-1}$), SY is straw yield ($t.ha^{-1}$) and AGNM is aboveground N content at maturity (kg $N.ha^{-1}$) (Pask et al. 2012).

Partial factor productivity of nitrogen

$$\mathrm{PFP_N} = \frac{\mathrm{GY}}{\mathrm{FN}}\ (\text{kg grain DM . kg}^{-1}\text{ N applied in fertilizer})$$

where GY is grain yield ($t.ha^{-1}$) and FN is N applied in fertilizer (Li et al. 2013).

Results

As shown in Figure 1, the total nitrogen concentration in the aboveground biomass of winter wheat at the boot stage (BBCH 45) did not significantly differ from the optimal value calculated according to Justes et al. (1994) with the exception of the CULTAN treatment when 150 kg N.ha^{-1} at the end of tillering (BBCH 29) was applied. In this case, the nitrogen concentration in the aboveground biomass was significantly lower than the optimum.

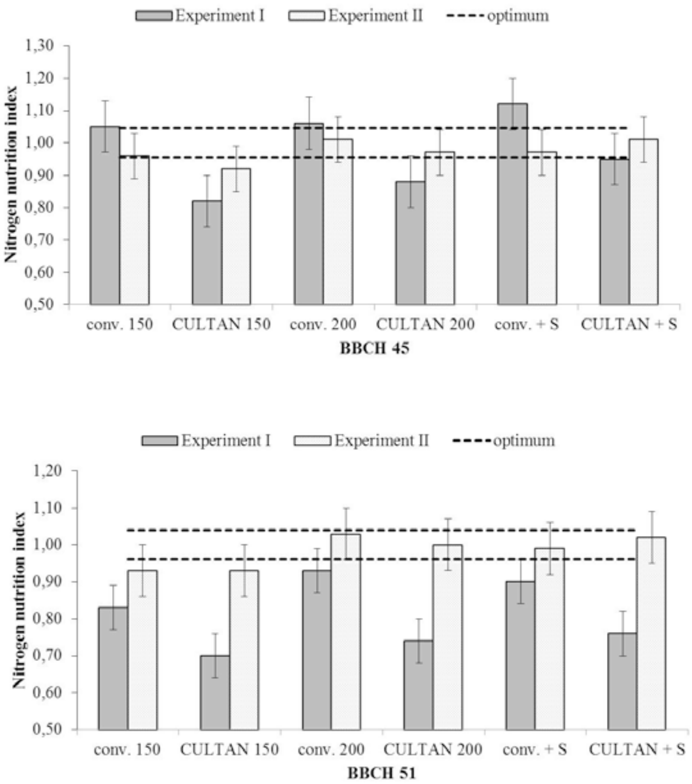

Figure 1. Nitrogen nutrition index at the BBCH 45 and BBCH 51 growth stages. Intervals indicate standard error at $P < 0.05$. In experiment I the CULTAN fertilization was done at the end of tillering of winter wheat (BBCH 29), in experiment II the CULTAN fertilization was carried out at the beginning of tillering (BBCH 22)

If the CULTAN fertilization was carried out at the end of tillering (BBCH 29), the concentrations of nitrogen in aboveground biomass at the beginning of heading (BBCH 51) were significantly lower in CULTAN-treated plants compared to the optimal value (Fig. 1). However, the earlier application of CULTAN fertilization, i.e. at the beginning of the tillering (BBCH 22) of winter wheat, led to no significant differences in the nitrogen nutrition index (NNI) between the optimal value and all other treatments.

Nitrogen utilization efficiency (NUtE) was significantly higher in the CULTAN treatments compared to the conventional ones with the exception of the CULTAN treatment when sulphur-containing fertilizer at the beginning of tillering (BBCH 22) was applied

(Table 2). By contrast, CULTAN-treated winter wheat tended to lower nitrogen uptake efficiency (NUpE) compared to the conventionally treated one, particularly when the CULTAN-fertilization was carried out at the end of tillering.

Nitrogen use efficiency (NUE), the contribution of nitrogen translocation to the total nitrogen in the grain (NTRg), the partial factor productivity of nitrogen (PFP) as well as post-heading nitrogen uptake (PHNU) did not differ in the CULTAN and the conventional treatments (Tables 2, 3 and 4).

Table 2. N utilization efficiency (NUtE; kg grain DM . kg^{-1} aboveground N content at maturity), N uptake efficiency (NUpE; kg N . kg^{-1} N available) and N use efficiency (NUE; kg grain DM . kg^{-1} N available)

Experiment	Parameter	conv. 150	CULTAN 150	conv. 200	CULTAN 200	conv. + S	CULTAN + S
I	NUtE	38.4^{a}	47.7^{b}	36.9^{a}	46.8^{b}	38.6^{a}	43.8^{c}
	NUpE	1.12^{a}	0.84^{bc}	0.94^{b}	0.69^{c}	1.14^{a}	0.96^{b}
	NUE	42.7^{a}	39.2^{ab}	34.4^{bc}	31.6^{c}	43.2^{a}	41.1^{a}
II	NUtE	38.5^{ab}	42.9^{d}	36.6^{a}	41.3^{cd}	38.4^{ab}	39.4^{bc}
	NUpE	1.06^{c}	0.94^{b}	0.88^{ab}	0.79^{a}	1.07^{c}	1.05^{c}
	NUE	40.9^{a}	39.5^{a}	32.3^{b}	32.3^{b}	40.8^{a}	41.1^{a}

Notes: Values within the row marked either with the same letter or without exponents are not statistically different at $P < 0.05$

Table 3. Contribution of N translocation to total N in grain (NTRg; %), post-heading N uptake (PHNU; kg N.ha^{-1}), N remobilization efficiency (NRE)

Experiment	Parameter	conv. 150	CULTAN 150	conv. 200	CULTAN 200	conv. + S	CULTAN + S
I	NTRg	65.3	71.7	70.4	72.9	67.8	66.5
	PHNU	66.1	42.8	59.0	39.6	57.9	57.8
	NRE	0.75	0.78	0.77	0.78	0.76	0.77
II	NTRg	80.7	95.3	91.7	98.4	86.3	97.6
	PHNU	36.0^{a}	6.2^{ab}	18.2^{ab}	2.7b	27.2^{ab}	8.8^{ab}
	NRE	0.76^{a}	0.81^{b}	0.76^{a}	0.81^{b}	0.77^{ab}	0.79^{ab}

Notes: Values within the row marked either with the same letter or without exponents are not statistically different at $P < 0.05$

Nitrogen remobilization efficiency (NRE) was not significantly influenced by the treatment when CULTAN treatment was carried out at the end of tillering (BBCH 29) (Table 3). The earlier term of CULTAN application (BBCH 22) led to significantly higher NRE in the CULTAN treatment compared to the conventional one when 150 kg N.ha^{-1} and 200 kg N.ha^{-1} was applied.

The nitrogen harvest index (NHI) was significantly higher in the CULTAN treatment compared to the conventional one only when 200 kg N.ha^{-1} was applied at the beginning of tillering (Table 4).

Table 4. N harvest index (NHI), biomass production efficiency (BPE; kg aboveground DM . kg^{-1} N uptake at maturity), partial factor productivity of N (PFP; kg grain DM . kg^{-1} N applied in fertilizer)

Experiment	Parameter	conv. 150	CULTAN 150	conv. 200	CULTAN 200	conv. + S	CULTAN + S
I	NHI	0.85	0.86	0.85	0.84	0.84	0.85
	BPE	70.4^{a}	88.6^{b}	67.6^{a}	87.8^{b}	72.2^{a}	83.0^{b}
	PFP	57.3^{ac}	52.6^{a}	43.4^{b}	39.8^{b}	58.2^{c}	54.9^{ac}
	grain yield	9.99^{ab}	9.18^{a}	10.10^{ab}	9.26^{ab}	10.15^{b}	9.57^{ab}
II	NHI	0.83^{ab}	0.85^{b}	0.82^{a}	0.85^{b}	0.83^{ab}	0.84^{ab}
	BPE	69.6^{ab}	77.3^{d}	66.4^{a}	74.1^{cd}	69.2^{ab}	70.7^{bc}
	PFP	50.8^{a}	49.0^{a}	38.3^{b}	38.2^{b}	50.8^{a}	51.0^{a}
	grain yield	8.85	8.54	8.90	8.88	8.86	8.90

Notes: Values within the row marked either with the same letter or without exponents are not statistically different at $P < 0.05$

Biomass production efficiency (BPE) (Table 4) achieved significantly higher values in the CULTAN treatments compared to the conventional ones excluding the application of 200 kg N.ha^{-1} when the CULTAN fertilization was carried out at the beginning of tillering.

Neither the increased dosage of nitrogen (200 kg N.ha^{-1}) nor the addition of sulphur to the nitrogen fertilizers brought benefits to winter wheat production.

Discussion

If the CULTAN fertilization was carried out at the beginning of tillering (BBCH 22) of winter wheat instead of at the end of tillering (BBCH 29), CULTAN-treated winter wheat did not suffer from nitrogen deficiency at the BBCH 45 (boot stage) and the BBCH 51 (beginning of heading) growth stages resulting in no significant differences in grain protein content between the CULTAN-treated winter wheat and the conventionally treated one recorded by Kozlovsky et al. (2010).

Significantly higher values of nitrogen utilization efficiency (NUtE) recorded with the CULTAN treatment compared to the conventional one can be explained according to Gaju et al. (2011) by higher biomass production efficiency with the CULTAN treatment (Table 4) which is according to Pask et al. (2012) characterized as aboveground dry matter at harvest per unit of aboveground nitrogen content at harvest. Even though higher NUtE values could be explained according to Montemurro et al. (2006) as a result of increased post-anthesis nitrogen uptake from the soil, the higher NUtE recorded in the CULTAN treatment could be explained rather by tendency to lower grain protein content in the CULTAN treatment compared to the conventional one which is in accordance with the findings of Albrizio et al. (2010) and Sedlar et al. (2013).

Higher nitrogen use efficiency (NUE) is an important prerequisite for increased profitability, either through greater yields or reduced nitrogen losses (Gaju et al. 2011). To improve the nitrogen use efficiency it is necessary to improve both nitrogen utilization efficiency (NUtE) and nitrogen uptake efficiency (NUpE) (Beatty et al. 2010). The CULTAN treatment, however, led solely to significantly increased nitrogen utilization efficiency which is insufficient to improve nitrogen use efficiency.

No significant differences in the partial factor productivity of nitrogen (kg grain . kg^{-1} N applied) were found between the conventional and the CULTAN treatment, which according to Li et al. (2013) means the same nitrogen use efficiency in both application types of nitrogen fertilizers.

Significantly higher nitrogen remobilization efficiency (NRE) was recorded with the CULTAN treatment compared to the conventional treatment in the applications of 150 kg $N.ha^{-1}$ and 200 kg $N.ha^{-1}$ when the CULTAN fertilization was carried out at the beginning of tillering (BBCH 22). This phenomenon indicates, according to Bahrani et al. (2011), a lower bioavailability of soil nitrogen in the latter growth stages when the CULTAN system is used. However, significant differences in both the contribution of nitrogen translocation to the total nitrogen in the grain (NTRg) and the post-heading nitrogen uptake from the soil (PHNU) between the CULTAN treatments and the conventional ones were not recorded. Therefore, according to the findings of Uzik and Zofajova (2012), a significant influence of the CULTAN system on the duration of winter wheat vegetation cannot be assumed.

Sinebo et al. (2004) recorded a positive correlation between barley grain yield and the nitrogen harvest index (NHI). The nitrogen harvest index was however significantly higher only with the CULTAN treatment compared to the conventional one in the application of 200 kg $N.ha^{-1}$ when the CULTAN fertilization was applied at the beginning of tillering (BBCH 22). This phenomenon can be explained by no significant differences in grain yields among the treatments (Table 4). Thus, a prolonged nitrogen uptake from soil by CULTAN-treated wheat resulting in delayed plant senescence stated by Sommer and Scherer (2009) was not confirmed in our experiment.

Increased dosage of nitrogen (200 kg $N.ha^{-1}$) brought no benefits to winter wheat production indicating 150 kg $N.ha^{-1}$ as the optimal nitrogen dosage under the conditions of the experimental sites.

Sulphur-containing fertilizers did not positively influence the studied parameters. In accordance with the findings of Kulhanek et al. (2011), sufficient sulphur content for winter wheat production at the experimental sites in the Czech Republic can be concluded.

Using the CULTAN system it is possible to supply winter wheat with nitrogen just in one application of nitrogen fertilizer achieving the same grain yields in comparison with the conventional nitrogen split application. In case of the correct timing of CULTAN fertilization, a higher risk of both nitrogen losses from soil and insufficient nitrogen uptake by CULTAN-treated winter wheat compared to the conventionally treated one is avoided.

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