Influence of injection nitrogen fertilization on yield and seed composition of winter oilseed rape (*Brassica napus* L.)

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ABSTRACT

Influence of the nitrogen injection fertilization 'controlled uptake long term ammonium nutrition' (CULTAN) on winter rape yield and seed composition was studied in 2009 and 2010 at two sites with different soil-climatic conditions in the Czech Republic. Two conventional treatments consisted of fertilization using the system of divided doses applied on soil surface. Two CULTAN treatments used injection fertilization with the whole dose of nitrogen applied once in the vegetation period, in early spring at the BBCH 26 stage (6 side shoots detectable). The trial compared conventional and CULTAN treatments. The overall dose of nitrogen was 200 kg N/ha in each treatment. The seed yield in a two-year average was 4.83 t/ha at conventional and 4.80 t/ha at the CULTAN treatment. This difference was not statistically significant. The higher nitrogen content in seed was recorded mainly at CULTAN treatments at the Hněvčeves site in 2009. An inconclusively higher phosphorus content was recorded in winter rape seed fertilized with the CULTAN method at both sites in 2010. Content of K, Ca, Mg and S did not show statistically significant differences between the two treatments during both experimental years. In oiliness no differences between conventional and CULTAN methods were observed.

Keywords: ammonium nitrogen; winter rape; injection; macroelements; oiliness

CULTAN (controlled uptake long term ammonium nutrition) method is based on the injection application of fertilizer with high content of nitrogen in the ammonium form into soil near the plant roots, creating so called depots. Fertilizer is applied at one dose once in the vegetation period (Sommer 2003). The depots in soil are a stable source of nitrogen due to high concentration of nutrients which is toxic for activity of nitrification bacteria (Balík et al. 2008). Positively charged ammonium ion is bound to negatively charged clay particles and humus compounds (Kücke and Scherer 2006). Literature reports that the nitrate form of nitrogen is more suitable for plants than the ammonium ion. However, previous results of the CULTAN methods show the very opposite. These adverse findings may be explained by different distribution of ammonium ion in the space of the plant roots (Sommer 1991, Marschner 1995). At the CULTAN fertilization only a certain part of the root system participates at ammonium nitrogen uptake from the depot margin. The roots are able to uptake the ammonium nitrogen out of there only if the aboveground part of plant is sufficiently supplied with saccharides and plants may therefore involve the absorbed nitrogen to the metabolism of nitrogen compounds (Sommer 2005).

Based on the results of the trials carried out in Germany, injection point application in spring, at the beginning of vegetation recovery, is the best for winter rape. According to Spiess and Meier (2008), at CULTAN fertilization a higher content of proteins and glucosinolates was observed, while the oiliness was lower. Plant damage caused by the injection wheel followed by infestation of plant by diseases was not definitely confirmed (Felgentreu 2003).

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The aim of this paper was to assess the influence of the CULTAN nitrogen fertilization on yield and composition of winter rape seeds and to compare this method with the conventionally treated control sites.

MATERIAL AND METHODS

In small-plot trials the influence of the CULTAN method of fertilization on yield, content of macroelements, oiliness and crude fat content of the winter rape seeds at Hněvčeves and Humpolec sites was observed in 2009 and 2010. Soil characteristics of these sites were specified by Kozlovský et al. (2009). The content of sulphur in soil in spring at the Hněvčeves site was 3.5 mg/kg and at the Humpolec site it was 3.4 mg/kg in water infusion. In the trial seeds of winter oilseed rape (*Brassica napus* L.) cv. Artus were used, which is a semi-late hybrid of 00-type. The seeding rate was 4 kg/ha. In Hněvčeves, the precrop was winter wheat in both experimental years, in Humpolec it was winter barley in (2009) and winter wheat in 2010. The trial consisted of four treatments with four replications. Scheme of the trials is given in Table 1. Two treatments included fertilization onto soil surface (conventional 1 and conventional 2) and two treatments used injection fertilization (CULTAN 1 and CULTAN 2). Treatments conventional 2 and CULTAN 2 were fertilized with nitrogen fertilizer with sulphur amendment. CULTAN treatments were fertilized using the GFI 3A injector (Maschinen und Antriebstechnik GmbH, Güstrower, Germany) with a working scope of 3 m, application distance of 10 cm distance from plant rows and the application depth of 5 cm. Injection applicator has 12 application wheels with 12 fertilizing nozzles. Surface of a fertilized plot was 39 m^2 out of which 15 m^2 was harvested with a small-plot combine-harvester.

Seed oiliness was determined spectroscopically on the minispec mq-one Seed Analyzer (Bruker, Ettlingen, Germany). Seed nitrogen content was determined by the Kjeldahl method using the Vapodest 50s (Gerhardt Gmbh & Co. KG., Königswinter, Germany). P and S contents were determined using the inductively-coupled plasma optical emission spectrometry ICP-OES (Varian, Victoria, Australia). Contents of K, C and Mg were determined by atomic absorption spectroscopy using the SpectrAA (Varian, Victoria, Australia). Statistical evaluation of the experiment was done in the Statistica 8.0 programme with the single-factorial ANOVA followed by the Tukey's test at the level of significance $P < 0.05$ (StatSoft, Tulsa, USA).

The period from August to December 2008 at the Humpolec site was characterized by the above-average air temperatures. Intensive precipitations were recorded in February and March 2009 at both sites. Precipitations in May 2010 were significantly above average at both sites.

RESULT AND DISCUSSION

In 2009 the average yield in the Czech Republic was the second highest during the winter rape cultivation in the country. It was confirmed by the results of our study, especially at the Hněvčeves site (Table 2). Thanks to favourable conditions the average seed yield at this site was 7.29 t/ha. At the Humpolec site it was significantly lower, on average by 3.37 t/ha. It is related to lower 1000 seed weight values and the number of plants per a unit of space compared to Hněvčeves. The Humpolec site should be more suitable for winter weed growing because it is located at the altitude of 525 m, average annual precipitations are 667 mm and the average yearly temperature is 6.5°C. Winter oilseed rape generally prefers the sites with the yearly average temperatures of 7–9°C and precipitations of 450–700 mm, altitude up to 650 m

Table 1. Fertilization scheme (kg N/ha)

Treatment	BBCH 25	BBCH 30	BBCH 26	BBCH 58	Total
Conventional 1	57 (CAN)	93 (CAN)	–	50 (CAN)	200
CULTAN 1	–	–	200 (UAN)	–	200
Conventional 2	57 (AS)	93 (CAN)	–	50 (CAN)	200
CULTAN 2	–	–	200 (UAS)	–	200

CAN – calcium ammonium nitrate (27% N); AS – ammonium sulphate (20% N, 23% S); UAN – urea ammonium nitrate (30%); UAS – urea ammonium sulphate (19% N, 5% S); BBCH – identification key of phenological growth stages

Table 2. Seed yield (t/ha, 12% moisture)

Treatment	Hněvčeves		Humpolec	
	2009	2010	2009	2010
Conventional 1	7.03[a]	4.61[a]	3.94[a]	4.69[a]
CULTAN 1	7.16[ab]	4.51[a]	3.80[a]	4.70[a]
Conventional 2	7.26[ab]	5.05[b]	3.93[a]	4.57[a]
CULTAN 2	7.71[b]	5.10[b]	4.03[a]	4.67[a]

Values within a column marked with the same letter are not statistically significant ($P < 0.05$)

Table 3. 1000 seed weight (g)

Treatment	Hněvčeves		Humpolec	
	2009	2010	2009	2010
Conventional 1	5.40[b]	5.41[a]	4.68[b]	5.53[ab]
CULTAN 1	5.76[a]	5.32[a]	4.53[a]	5.46[b]
Conventional 2	5.46[b]	5.05[b]	4.89[c]	5.60[a]
CULTAN 2	5.83[a]	5.10[b]	4.71[b]	5.62[a]

Values within a column marked with the same letter are not statistically significant ($P < 0.05$)

a.s.l. (Fábry 1992). The period from August to Decemeber 2008 was very warm at the Humpolec site, which supported more intensive plant growth and subsequent nitrogen deficiency in the following spring. Thus it is possible to consider more frequent nitrogen fertilization of crops in future. Another reason of nitrogen deficiency might have been washing off the fertilizer to lower layers of soil as a result of intensive precipitations in February and March 2009. In 2010, treatments with sulphur-amended fertilizers (conventional 2 and CULTAN 2) reached a higher seed yields, especially at the Hněvčeves site. Compared to application of fertilizer without sulphur a significant increase of seed yield was recorded, on average by 11.3%. Kozlovský et al. (2009) in their 2-year trials reported an increase of winter wheat grain yield after application of sulphur-amended nitrogen fertilizers irrespective of the method of application (conventional or CULTAN). 2010 season was specific as to the weather conditions. Precipitations in May 2010 were significantly above average at both sites. All treatments used in the trial were strongly infested with fungal diseases irrespective of the method of fertilization. These results correspond to the findings of Felgentreu (2003) that injection fertilization did not harm leaf surface and thus did not increase the risk of infestation of crop with fungal diseases. CULTAN treatments (1 and 2) significantly increased 1000 seed weight compared to conventional treatments (1 and 2) at the Hněvčeves site in 2009 (Table 3). These results correspond to the statements of Sommer (2005) that the CULTAN fertilization reduces nitrogen reutilisation, which delays ageing of the plant base and thus the time when assimilates are stored in seeds prolongs. Also Sedlář et al. (2011) observed higher 1000 seed weight in CULTAN-fertilized spring barley compared to conventional fertilization. At the Humpolec site in 2009 the situation was opposite. Both CULTAN treatments reached significantly lower 1000 seed weight than conventional treatments. These results are probably related to the findings of Christen and Sieling (1995) that 1000 seed weight is more influenced by year than fertilization treatment. In 2010 no significant differences in 1000 seed weight were observed between the fertilization methods.

The content of macroelements in seed is given in Table 4. Higher content of nitrogen in seed was usually observed at treatments fertilized nitrogen fertilizer containing sulphur (conventional 2 and CULTAN 2) compared to the treatments without sulphur (conventional 1 and CULTAN 1). Schnug et al. (1993) reported that a lack of sulphur in soil is as a significant problem of winter oilseed rape fertilization, because the effect of other nutrients, primarily nitrogen, is thus reduced. The content of nitrogen in seed is predominantly influenced by site and also by the dose of nitrogen in fertilizer (Balík et al. 1997). The highest nitrogen content in seed at sulphur-amended injection application of fertilizer (CULTAN 2) was obtained at the Hněvčeves site in 2009 and 2010 and at the Humpolec site in 2009. Felgentreu (2003) reported a decrease in oiliness and an increase of the protein content in winter rape seeds after injection fertilizing with sulphur-amended nitrogen fertilization. He attributed this result to an increased activity of the asparagine synthase after ammonium fertilization. The increased content of amino acids in plants fertilized with ammonium nitrogen is a result of detoxification of ammonia using the synthase of amino acids with organic acids that are a source of carbon (Hayens and Goh 1978). In 2009 no differences were observed in the content of phosphorus in seed between the methods of fertilization at the Hněvčeves site. At the Humpolec site in 2009 significantly higher content of phosphorus in seed was recorded at CULTAN 1 treatment, compared to the conventional 1 treatment. This finding was not repeated following year. In 2010, CULTAN

Table 4. Content of macroelements in seed dry matter (%)

Site	Year	Treatment	Macroelements					
			N	P	K	Ca	Mg	S
Hněvčeves	2009	conventional 1	3.29^{a}	0.49^{a}	0.63^{a}	0.24^{a}	0.24^{a}	0.06^{a}
		CULTAN 1	3.49^{a}	0.51^{a}	0.67^{a}	0.25^{a}	0.25^{a}	0.06^{a}
		conventional 2	3.37^{a}	0.50^{a}	0.65^{a}	0.25^{a}	0.24^{a}	0.06^{a}
		CULTAN 2	3.64^{a}	0.48^{a}	0.65^{a}	0.24^{a}	0.24^{a}	0.05^{a}
	2010	conventional 1	3.38^{ab}	0.69^{a}	0.78^{a}	0.30^{a}	0.26^{a}	0.10^{a}
		CULTAN 1	3.24^{a}	0.73^{a}	0.80^{a}	0.33^{a}	0.26^{a}	0.10^{a}
		conventional 2	3.32^{ab}	0.71^{a}	0.81^{a}	0.33^{a}	0.26^{a}	0.11^{a}
		CULTAN 2	3.51^{b}	0.76^{a}	0.78^{a}	0.31^{a}	0.25^{a}	0.12^{a}
Humpolec	2009	conventional 1	3.15^{a}	0.46^{a}	0.71^{a}	0.25^{a}	0.27^{a}	0.07^{a}
		CULTAN 1	3.01^{a}	0.61^{b}	0.54^{a}	0.18^{a}	0.18^{a}	0.08^{a}
		conventional 2	3.44^{a}	0.51^{ab}	0.67^{a}	0.21^{a}	0.23^{a}	0.08^{a}
		CULTAN 2	3.48^{a}	0.52^{ab}	0.63^{a}	0.21^{a}	0.22^{a}	0.07^{a}
	2010	conventional 1	3.29^{ab}	0.70^{ab}	0.80^{a}	0.27^{a}	0.28^{ab}	0.10^{a}
		CULTAN 1	3.09^{a}	0.76^{a}	0.77^{a}	0.27^{a}	0.27^{a}	0.10^{a}
		conventional 2	3.39^{b}	0.68^{b}	0.78^{a}	0.29^{a}	0.28^{ab}	0.11^{a}
		CULTAN 2	3.32^{ab}	0.70^{ab}	0.79^{a}	0.28^{a}	0.28^{b}	0.10^{a}

Values within a column marked with the same letter are not statistically significant ($P < 0.05$)

treatments, especially at the Hněvčeves site, did not reach higher values of phosphorus content in seeds compared to conventionally fertilized treatments. It corresponds to the conclusions of Sommer (2005) that CULTAN fertilization supports increased phosphorus storage into seeds. As soon as the roots are forced to uptake NH_4^+, it results in strong excretion of protons. The change of equilibrium in roots lowers the rhizosphere pH and activates phosphorus mobilization (Trenkel 1997). It is a long known fact that the toxicity of ammonium ion is stronger at the potassium deficiency (Wall 1939). No differences in the potassium content in seed were observed between the fertilization systems during the trial. The antagonistic effect of ammonium ion was not manifested and the toxic influence of ammonium nitrogen was not observed at the CULTAN treatments. According to Sattelmacher et al. (1993) potassium has a key role in assimilation mechanism of the ammonium ion. In the content of calcium in seed differences between conventional and CULTAN treatments were not observed. The findings of Hayens and Goh (1978) that plants growing in conditions of

Table 5. Oiliness (%, 8% moisture)

Treatment	Hněvčeves		Humpolec	
	2009	2010	2009	2010
Conventional 1	42.7^{b}	42.4^{a}	40.8^{ab}	42.7^{a}
CULTAN 1	42.2^{ab}	42.7^{a}	41.9^{b}	43.9^{b}
Conventional 2	42.5^{ab}	42.5^{a}	40.1^{a}	42.4^{a}
CULTAN 2	41.5^{a}	41.8^{a}	40.8^{ab}	42.5^{a}

Values within a column marked with the same letter are not statistically significant ($P < 0.05$)

Table 6. Crude fat yield (t/ha, 8% moisture)

Treatment	Hněvčeves		Humpolec	
	2009	2010	2009	2010
Conventional 1	2.85^{a}	1.86^{ab}	1.53^{a}	1.90^{a}
CULTAN 1	2.87^{a}	1.83^{a}	1.51^{a}	1.96^{a}
Conventional 2	2.93^{a}	2.04^{b}	1.50^{a}	1.84^{a}
CULTAN 2	3.04^{a}	2.03^{b}	1.56^{a}	1.88^{a}

Values within a column marked with the same letter are not statistically significant ($P < 0.05$)

the ammonium fertilization generally have a lower content of calcium, magnesium and potassium, and higher content of phosphorus and sulphur than plants fertilized only with nitrate, were not confirmed. In the content of sulphur in the rape seeds statistically significant differences between conventional treatments and CULTAN methods were not observed in a 2-year period. In 2009 sulphur content in seed dry matter lower by 0.05% and 0.03% was observed at the Hněvčeves and Humpolec sites, respectively, compared to 2010. This difference was probably caused by a dilution effect, as the high seed yield was obtained at all treatments, especially at the Hněvčeves site.

Seed oiliness is shown in Table 5. Filipek-Mazur et al. (2001) give the critical sulphur reserve of 10 mg/kg available in the soil in water infusion. Sulphur values at both sites are significantly lower than this limit. The highest oiliness was reached in 2010 at the Humpolec site at the CULTAN 1 treatment. Average oiliness of all treatments was higher in this year compared to 2009. In general, fertilization with nitrogen only, i.e. without sulphur, reduces oil production as the seed yield decreases (Joshi et al. 1998). Moreover, water deficit during the stage of flowering and silique formation may result in higher content of proteins and lower content of oil (Bouchereau et al. 1996). In Humpolec in 2010 a higher sum of precipitation in June and July was recorded compared to Hněvčeves in the same period. No significant differences in oiliness were observed between the two systems of fertilization in both experimental years. Nitrogen form generally did not influence the oil content in winter rape seed. Harris (1980) adds that slight differences were observed in rape seed oiliness in relation to the dose and term of nitrogen fertilization.

At the Hněvčeves site in 2009 high yield of crude fat was obtained, as the seed yield was record at all treatments in this year (Table 6). At the same site in 2010 the yield of crude fat was significantly higher at sulphur-amended treatments (conventional 2 and CULTAN 2) compared to treatments without sulphur (conventional 1 and CULTAN 1). The lowest yield of crude fat was observed at the Humpolec site in 2009. To some extent, it correlates with the obtained seed yields that were lower, probably as a result of higher rate of infestation with fungal diseases (*Sclerotinia sclerotiorum* and *Alternaria brassicae*). In 2010, both CULTAN treatments at the Humpolec site gave slightly higher yield of crude fat, compared to conventionally fertilized treatments.

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