# INFLUENCE OF AMMONIUM NITRATE SULPHATE FERTILIZER ON IRON UPTAKE AND REMOVAL BY OILSEED RAPE PLANTS\*

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Influence of nitrogen and N-S fertilizers on the concentration of iron in the plants of winter oilseed rape was studied in precision field trials. A positive influence of N-S fertilizers on the contents of iron in individual parts of plants was determined. The highest Fe concentrations were found in the leaves and inflorescences, the lowest occurred in the stem. The concentrations were found in the interval of 13–201 mg Fe.kg<sup>-1</sup> of the dry matter depending on the growth period and the analysed part of the plant. No differences in the iron contents were determined in the seeds of individual treatments. The highest removal of Fe by the above-ground parts of the plants was recorded in the flowering period, reaching 906 g Fe.ha<sup>-1</sup> in the N-S variant.

iron; sulphur; winter oilseed rape; N-S fertilizers

### INTRODUCTION

The total iron contents in the soils fluctuate in the range of 0.2-5%. Iron occurs under aerobic conditions in the form of Fe(III)-oxides, in compounds with silicates and also in organo-mineral compounds. Goethite and hematite are important representatives of the oxide form. They occur in crystalline form and under aerobic conditions they are stable and iron from them is hardly utilized by the plants. Weakly crystalline oxides (eg ferrihydrite) and also organo-mineral compounds are a predominant source of iron that can be potentially mobilized. Due to the low solubility of all Fe(III) oxides, the iron soil solution concentration is, under aerobic conditions, exceptionally low (mostly less than  $0.2-0.3 \text{ mg.l}^{-1}$ ). At pH > 3.5 iron is fixed in soluble organic Fe complexes. Under extremely acidic conditions (pH < 3.5), usually found in forest soils and in the soils with a high sulphates content, the inorganic Fe(III) ions, [Fe<sup>3+</sup>, FeOH<sup>2+</sup>, and Fe(OH)<sub>2</sub><sup>+</sup>, FeSO<sub>4</sub> can also occur. Under anaerobic conditions there is a reduction of Fe(III) oxides to Fe<sup>2+</sup> and, subsequently, the increase of the Fe soil solution concentration takes place (Scheffer, Schachtschabel, 2002).

Plants usually take up iron in the Fe<sup>2+</sup> form. Grasses can also take up iron in the form of organic Fe(III) complexes (R ö m h e l d, M a r s c h n e r, 1986). In the grasses (strategy II), the phytosiderophores are produced and the whole Fe(III) phytosiderophore complex is transported via the plasma membrane. Contents of iron in the green parts of plants are present in the interval of 30–500 mg Fe.kg<sup>-1</sup> of dry matter. When the contents in the plants are below 50–80 mg Fe.kg<sup>-1</sup> of dry matter, deficiency symptoms may occur (S c h e f f e r, S c h a c h t s c h a b e l, 2002). Iron mobility is significantly influenced by the

processes occurring in the plants rhizosphere-microorganisms activities and the decomposition of organic matter as well as by root exudates. Fe(III)-chelates and, occasionally, Fe(II)-chelates, are the dominant form of soluble iron in the soil solution. In the xylem, iron is transported mainly in the form of Fe(III) complexes. Iron is characterised by a relatively easy change of the oxidation degree.

Depending on the ligand, the redox potential of the Fe (II/III) varies widely. On the basis of a very high affinity of iron to different ligands (organic acids, inorganic phosphates) the transport of iron over short or long distance in the forms of Fe<sup>3+</sup> or Fe<sup>2+</sup> is very unlikely.

Iron is a part of catalytic enzymes (catalase, peroxidase, nitrogenase) (Marschner, 2003). Most of iron in a plant cell is concentrated in the chloroplasts and mitochondria. A significant part of Fe is fixed in phosphoproteins called ferritins and serves as an iron storage for the development of plastids. Ferritin also plays a very important role in the cells defence mechanism against heavy metals (Prasad, Hagemeyer, 1999). Fe in the ferredoxine is fixed in protein by sulphur of the cystein molecules and thus it creates a highly effective redox system.

The critical deficiency level of iron content in the leaves is at 50–150 mg Fe.kg<sup>-1</sup> of dry matter (M a r s c h n e r, 2003). At the same time, the C4 group plants require a higher Fe concentration than the C3 plants. However, it is interesting (S m i t h et al., 1984) to note that the critical deficiency contents were almost the same for both groups (approximately 72 mg Fe.kg<sup>-1</sup> of dry matter in the C3 plants, and approximately 66 mg Fe.kg<sup>-1</sup> of dry matter in the C4 plants). The critical deficiency contents are much higher – approximately 200 mg Fe.kg<sup>-1</sup> of dry matter in the meristematic and fast growing tissues (e.g. root tips) (H ä u s s l i n g et al., 1985).

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### MATERIAL AND METHODS

A precision field trial was set up in Prague-Uhříněves at the experimental station of the Faculty of Agrobiology, Food and Natural Resources. The following treatments were investigated in the trials:

1) 100 kg N.ha<sup>-1</sup> (a single application of AN: nitrochalk, 27% N) – sidedress the first spring application,

2) 100 kg N.ha<sup>-1</sup> + 50 kg S.ha<sup>-1</sup> (single application of ANS: ammonium nitrate + ammonium sulphate, 26% N and 13% S) – sidedress the first spring application.

The size of the trial plot was 20 m<sup>2</sup>. Each treatment had four replicates. The station is situated at 295 metres above the sea level, with average annual temperature of 8.3 °C, and annual rainfall reaching 575 mm, of which 380 mm falls during the period between April and September. The level of the subterranean water table is at the depth of 1 metre and is of a permanent character. The favourable water regime is supported by developed illuvial horizons with a relatively good water retaining ability, which influences the stable capacity of moisture that can be utilized by plants. The soil is represented by Luvisol. The ground is a deep top soil (down to 0.32 cm) and the humus horizon to the depth of 0.70 cm. The sorption complex is saturated. Chemical analyses of the soils determined the following contents of available nutrients (Mehlich III): 220 mg.kg<sup>-1</sup> of potassium, 119 mg.kg<sup>-1</sup> of phosporus, and 123 mg.kg<sup>-1</sup> of magnesium. The total sulphur content was 850 mg.kg<sup>-1</sup>, the mineral S content was 4–7 mg.kg<sup>-1</sup> (before the application of fertilizers), and the total iron content was 9507 mg.kg<sup>-1</sup>. The value of pH/CaCl<sub>2</sub> was 6.2.

Winter oilseed rape (Bristol cultivar - two zero variety) was used as a trial crop. This is a typical representative of cultivars with strong branching and low height. It is characterized by very early flowering and it therefore better utilizes moisture during the spring period. The pods are able to assimilate during the period between flowering and harvest periods for up to 5-10 days longer, and this is very positively reflected in the yield formation. The Bristol cultivar has high oil content. Winter wheat was the preceding crop. Apart from the spring application of fertilizers, standard protection of the crop was carried out during the vegetation period. During the vegetation period plant samples were collected (3 plants per trial plot -i.e.12 plants per treatment). The plant was dissected into individual parts according to the growth period: root, upper leaves, lower leaves, stems, pods, branches, and flowers. Following their homogenisation, chemical analyses were carried out for individual replicates.

The plant material was decomposed by using the dry ashing procedure and the content of individual elements was determined by the optical emission spectrometry with induction fixed plasma (ICP – OES, Varian VistaPro, Australia).

# RESULTS AND DISCUSSION

The Uhříněves site is very fertile, as shown by the average yields achieved over a period of three years.

The seed yield on the unfertilized control treatment was 3.7 t.ha<sup>-1</sup>, the AN treatment showed by 49% higher, and the ANS treatment was 60% higher yields. During the entire spring growth period there was a distinct difference in the growth intensity between the unfertilized treatment and the treated treatments. The achieved results also confirmed the importance of sulphur fertilization in the cultivation of winter oilseed rape even on fertile sites. During all three experimental years there had always been a higher yield in the ANS treatment. After the application of the ANS fertilizer, the concentrations of particularly S, Mn, and Zn in plants were evidently increased (B a l í k et al., 2005b), while the B concentration did not change significantly over the period (B a l í k et al., 2005a). The lower molybdenum content was statistically significant (B a l í k et al., 2006).

The plants are adequately supplied with iron on an overwhelming majority of fields. The plants can suffer from deficiency only on carbonate soils. Plants sensitive to iron deficiency include the grape vine, different fruit varieties, citrus varieties, maize and soya (Mengel, 1984). Finck (1997) states in his paper that in relation to iron there is no need to be concerned about the nutrition of winter oilseed rape in Germany. As Bertrand and Hinsinger (2000) found, winter oilseed rape was able to mobilize Fe from less available forms in the soil better than peas and lupine. There is very little information in a literature about limiting concentrations of iron in the plants of oilseed rape. For example, there are no data in Ernährungstörungen bei Kulturpflanzen (Bergmann, 1993), an essential publication devoted to inorganic analyses of the plants. Similarly, in the Czech Republic, Neuberg et al. (1990) do not itemize any relevant criteria. Finck (1997) published values at the level of 50-150 mg Fe.kg<sup>-1</sup> in the dry matter of fully developed leaves in the budding period, while indicating the lower minimal limit of 50 mg as uncertain and, so far, unconfirmed. In our experiments, during this period and depending on the observed treatment, the iron content was 107-104 mg Fe.kg<sup>-1</sup> in the upper leaves and 529–665 mg Fe.kg<sup>-1</sup> in the dry matter of the lower leaves.

It is thus clear that the plants did not suffer from any deficiency of this microelement. The question of observing the changes in the iron content is interesting mainly in connection with the study of interactions of the risk elements with iron in different cell structures (Pavlíková et al., 2002; Cakmak, 2000; Misra, Ramani, 1991; Siedlecka, Krupa, 1999; Alcantara et al., 1994) and also when studying the effect of different fertilizing treatments on the uptake of nutrients (Gaur et al., 1971).

Fertilizing with sulphur and N-S fertilizers has become an inseparable part of the winter oilseed rape cultivation technology. However, the positive effect is being narrowed down only to the problem of sulphur but the complex approach to the effectiveness of the N-S fertilizers is missing. As it can be seen in Figs 1 and 2, distinctly higher concentrations in all parts of the plant were found in the ANS treatment. It must be emphasised that in this treatment there was a stable higher growth of the above-ground biomass and the influence of the diluting effect was greater

here that in the AN treatment. The results, shown in Figs 1 and 2, show clearly that the highest contents were found in leaves. Compared with zinc, copper and molybdenum, Fe concentrations were always higher in the leaves from the lower part of the stem. High contents were also determined in the inflorescence. The lowest concentrations were found in the stems or branches at all samplings.

Apparently, there are several causes of increased Fe content in plants. Increased uptake of the sulphate anion in the ANS treatment plants is associated with an increased uptake of cations (including iron). For example, during the flowering period the sulphur content in the AN treatment was 1.03% (upper leaves) and 0.67% (lower leaves), while for the ANS treatment the values reached up to 1.8% (upper leaves) and 0.97% (lower leaves).

The ANS treatment showed during the entire growth period an increased formation of the biomass. It is known that plants expel a significant part of their production in the form of root exudates. It can, therefore, be assumed that there was a greater quantity of exudates, particularly in the ANS treatment. As stated by Scheffer and Schachtschabel (2002), iron mobility is significantly influenced by the processes that take place in the plants' rhizosphere – i.e. by the soluble fulvo and humic acids, microorganisms activities and decomposition of organic matter, and also by the root exudates. Hinsin-

ger (1998, 2001a, b) and McLaughlin et al. (1998) state that the root respiration and exudation of organic acids contribute to the acidification of rhizosphere. According to the results of Nye (1986) and Hinsinger (2001b) the  $\rm CO_2$  respiration can significantly contribute to the acidification of rhizosphere only on calcareous soils, but not on the acidic ones as  $\rm H_2\rm CO_3$  dissociates significantly only in the ranges of neutral and alkaline pH (the pK value is 6.36). At the Uhříněves site the value was pH/CaCl<sub>2</sub> 6.2, so that a more significant influence of microorganisms and the root respiration on the change in the pH value cannot be considered.

It is well known from literature sources (Kirkby, 1968; Kirkby, Knight, 1977) that the form of nitrate nutrition has a considerable influence on the balance of cations and anions. Plants respond to the uptake of nitrate nitrogen by a relative increase in the pH value of the rhizosphere. However, in our experiments the different forms of nitrogen in the tested fertilizers did not, apparently, have any influence on the cations uptake. As determined by the soil analyses, no significant difference in the concentrations of NH<sub>4</sub> and NO<sub>3</sub> ions (extract of 0.01 M CaCl<sub>2</sub>) in the topsoil and subsoil was determined 35 days after the application of fertilizers. This is in good agreement with an intensive microbial activity of Luvizem at the Uhříněves site.

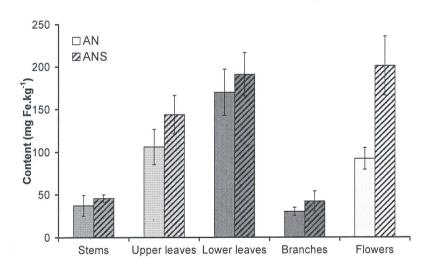


Fig. 1. Iron content in plants - flowering period

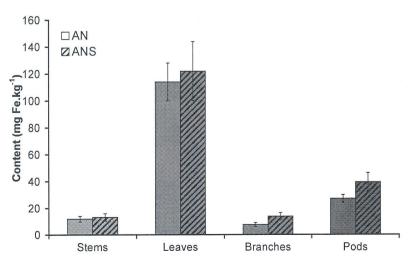
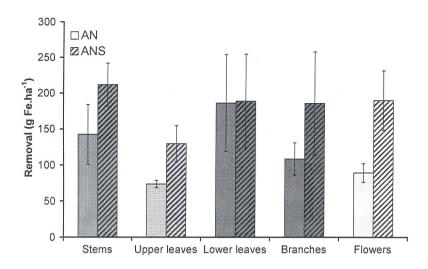
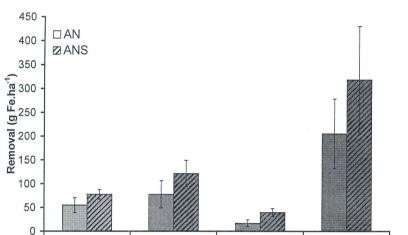


Fig. 2. Iron content in plants - green pod period





Branches

Fig. 4. Iron removal by plants - green pod period

Increased iron mobility in soil could also have been caused by the acidification effect of the ANS fertilizer. The results of the mineral sulphur contents in the topsoil provide direct, as well as indirect, evidence of this. The mineral S content in the topsoil during the elongation period of growth in the AN treatment was 3.9 mg.kg<sup>-1</sup>, and in the ANS treatment 9.2 mg.kg<sup>-1</sup>, in the flowering period of the AN treatment it was 6.3 mg.kg<sup>-1</sup>, and the ANS treatment 15.2 mg.kg<sup>-1</sup>, in the green pods period of the AN treatment it was 6.1 mg.kg<sup>-1</sup> and the ANS treatment 14.3 mg.kg<sup>-1</sup> (Sýkora, 2006).

Leaves

Stems

It is interesting that the differences in Fe contents of the vegetative organs did not transform into the differences in the seeds contents. The average concentration in the AN treatment was 31.2 mg Fe.kg<sup>-1</sup> and in the ANS treatment it was 30.1 mg Fe.kg<sup>-1</sup> of the seed dry matter.

Higher Fe concentrations in the plants of ANS treatment, together with the biomass growth, resulted in significant differences in the uptake of the element by the plant's biomass (Figs 3 and 4). During the flowering phase, the above-ground biomass of the AN treatment accumulated 600 g.ha<sup>-1</sup> and in the ANS treatment 906 g.ha<sup>-1</sup>. For the green pod period the values were determined as 355 g.ha<sup>-1</sup> (AN) and 557 g.ha<sup>-1</sup> (ANS). The displayed removals of iron by plants are lower than the frequently presented CETION data (in Fábry et al., 1992). They

show the total uptake as 683 g Fe.ha<sup>-1</sup> for the seed yield of 3.5 t.ha<sup>-1</sup>. Of this, the harvest transports away 332 g, while 351 g remains in the field in the postharvest crop residues. In our experiments the harvest removed approximately 20–40% of the element amount that had been taken up by the plant, depending on the treatment. The values of the Fe uptake dynamics show that the maximum uptake has been determined at the period of flowering, which also corresponds with the Cramer removal curves (in Vašák et al., 1997).

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Vliv hnojiva DASA na obsah a odběr železa rostlinami řepky.

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V přesných polních pokusech byl sledován vliv dusíkatých hnojiv a N-S hnojiv na koncentraci železa v rostlinách ozimé řepky. Byl stanoven pozitivní vliv N-S hnojiv na obsah železa v jednotlivých částech rostlin. Nejvyšší koncentrace železa byly zjištěny v listech a v květenství, nejnižší ve stonku. V závislosti na růstové fázi a analyzované části rostliny byly koncentrace v intervalu 13–201 mg Fe.kg<sup>-1</sup> sušiny. V semeni nebyly stanoveny rozdíly v obsahu železa u jednotlivých variant. Nejvyšší odběr železa nadzemní částí rostlin byl zaznamenán ve fázi květu a činil u varianty N-S 906 g Fe.ha<sup>-1</sup>.

železo; síra; ozimá řepka; N-S hnojiva

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