



# Effect of nitrogen and phosphorus availability on the emergence, growth and over-wintering of Rumex crispus and Rumex obtusifolius

# V. Křišťálová\*, M. Hejcman\*†, K. Červená\* and V. Pavlů\*†

\*Department of Ecology, Faculty of Environmental Sciences, Czech University of Life Sciences, Kamýcká, Prague, Suchdol, and †Crop Research Institute, Drnovká, Prague, Ruzyně, Czech Republic

#### **Abstract**

Rumex crispus and R. obtusifolius are known to be nitrophilous but it is not known whether either or both species require a high N supply at all developmental stages. Furthermore, it is not clear whether both species require a high P supply, attain flowering in the seeding year, or have similar levels of winter resistance. The effect of nutrient availability on the emergence, growth and over-wintering of both Rumex species was investigated in a pot experiment (ten N, P and K fertilizer treatments) in Prague, Czech Republic. In both species, emergence of seedlings was negatively affected by very high N, but positively affected by increased P availability in the soil. No effect of K supply on the emergence, or subsequent growth, was recorded. High flowering in the seeding season and high winter mortality of R. obtusifolius contrasted with no flowering and no mortality of R. crispus. Over-wintering was not markedly affected by N, P or K supply. Both Rumex species are sensitive to a very high N supply in early developmental stages but tend to require a high N supply from the fully developed rosette-stage onwards. In addition to high N requirements during stem growth, flowering and seed ripening, both species also require a high P supply. At least some Central European populations of R. crispus, in contrast to R. obtusifolius, do not flower in the seeding year. In Central Europe, the frequently reported high field winter mortality of R. obtusifolius can be caused by its low frost resistance.

Correspondence to: M. Heicman, Department of Ecology, Faculty of Environmental Sciences, Czech University of Life Sciences, Kamýcká 129, CZ 165 21 Prague 6, Suchdol, Czech Republic.

E-mail: hejcman@fzp.czu.cz

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#### Introduction

Rumex crispus L. (curled dock) and R. obtusifolius L. (broad-leaved dock) are perennial species native to temperate Europe (Cavers and Harper, 1964) and nowadays are cosmopolitan and are troublesome weeds worldwide, especially on arable land and in temperate grasslands (Jeangros and Nösberger, 1990; Hopkins and Johnson, 2002; Gebhardt et al., 2006; Zaller, 2006; van Evert et al., 2009; Stilmant et al., 2010). For farmers in temperate regions, the fear of infestation of pastures by *R. crispus* and *R. obtusifolius* is one of the major obstacles preventing the switch from conventional to organic farming (Zaller, 2004). Both Rumex species are detrimental for livestock production as they decrease forage quality and have adverse animal health effects, causing serious digestive problems if eaten in high quantities (Holm et al., 1977; Hejduk and Doležal, 2004).

Large numbers of viable and germinable seeds, which last in the soil for many years, are produced by both Rumex species; up to 60 000 seeds per plant per year have been reported (Cavers and Harper, 1964). In addition to generative reproduction, both Rumex species are able to regenerate from fragmented roots. High reproduction and regenerative capacity allows R. crispus and R. obtusifolius to be highly successful weeds (Honěk and Martinková, 2004; Šilc and Čarni, 2007), and their control without herbicides is extremely difficult (Pino et al., 1998; Hopkins and Johnson, 2002; Strnad et al.,

Although the ecology of both species has received much research attention (Hatcher et al., 1995, 1997a,b; Benvenuti et al., 2001; Honěk and Martinková, 2002), the timing of the first flowering and seed production after the establishment of seedlings still remains unclear. According to Cavers and Harper (1964),

flowering and seed set occasionally occur in the seedling year, but usually not until the second year after germination. According to Akeroyd and Briggs (1983), the flowering of R. crispus occurs from the second season after seedling establishment, and the same finding was also reported for R. obtusifolius (Hongo, 1989b). Unlike in Britain and the rest of Europe, plants of *R. crispus* in most North American populations are reported to require over-wintering before flowering can occur (Hume and Cavers, 1983; Bond et al., 2007).

Germination under natural conditions in Western Europe can occur throughout the growing season, but occurs mostly during spring and autumn because of a loss of dormancy at low temperatures (Roberts and Totterdell, 1981; van Assche and Vanlerberghe, 1989). Seeds of R. obtusifolius germinate more slowly than R. crispus, but early development of seedlings occurs more quickly in R. crispus (Cavers and Harper, 1964, 1979).

Both Rumex species are known to be nitrophilous (Novak and Slamka, 2003; Hejduk and Doležal, 2004), although there are doubts as to whether or not both Rumex species are nitrophilous in all developmental stages (Zaller, 2004). According to Klimeš (1996), low concentrations of nutrients have a stimulating effect on the germination of R. obtusifolius, whereas high concentrations slow down germination and decrease the proportion of germinating seeds. A low concentration of nitrates (0.01 mol L<sup>-1</sup>) was found to stimulate the germination of R. obtusifolius, whereas a high concentration (0·1 mol L<sup>-1</sup>) reduced its germination (Milberg, 1997). Although the requirement for high nitrogen (N) in mature plants of R. obtusifolius is generally known (Jeangros and Nösberger, 1990), little attention has been paid to the nutrient requirements of young plants or to the phosphorus (P) and potassium (K) nutrition of mature plants (Humphreys et al., 1999).

Similarly, the winter resistance of both species has received little research attention up to now. In a study performed in a cool-winter region of Central Europe, winter mortality of R. obtusifolius prevailed over summer mortality, and half of the plants died within the first 4 years of the field experiment and only 4% survived more than 8 years under a no-cutting management (Martinkova et al., 2009). In the same region, high mortality of mature R. obtusifolius plants on unmanaged semi-natural grassland was recorded (Pavlů et al., 2008a,b). High winter mortality of R. obtusifolius and R. crispus (approximately 50% mortality in the seeding year) was recorded in the cool winter season in Hokkaido, Japan (Hongo, 1989a). In the grassland experiment by Hongo (1989b) in the same region, 98% of transplanted seedlings of R. obtusifolius died in the third and fourth years, whereas the mortality of R. crispus was gradual and 17% of transplanted plants survived for 5 years. Differences in winter resistance may explain the differences in the world distribution of both these species, as R. crispus, in contrast to R. obtusifolius, can occur in northern and continental regions with severe winter frosts (Cavers and Harper, 1964). Although winter resistance may be a factor contributing to the different world distributions of the two species, differences in winter resistance between R. cripus and R. obtusifolius have not been investigated experimentally.

The aim of this study was to answer following questions: (i) is there any effect of high N, P and K availability on the emergence of R. crispus and R. obtusifolius? (ii) How is the performance (in terms of number of leaves, plant height and number of stems) of these two Rumex species affected by different N, P and K availabilities? (iii) How is winter survival different between the two species and how is it affected by different N. P and K availabilities?

#### Materials and methods

### **Experimental design**

In May 2008 and in May 2009, a factorial pot experiment was established in the open-air vegetation hall of the Crop Research Institute in Prague-Ruzyně (Czech Republic, 50°5'N; 14°18'E) with natural rain, temperature and light conditions. The investigated factors were species (two levels: Rumex crispus and R. obtusifolius) and nutrient availability (ten levels: Control, N1, N2, P1, P2, K, N<sub>1</sub>P<sub>1</sub>, N<sub>1</sub>P<sub>1</sub>K, N<sub>2</sub>P<sub>1</sub>K, N<sub>2</sub>P<sub>2</sub>K; see Table 1 for details) providing a total of 20 treatments. Each treatment was replicated five times (thus, 100 pots in total). The pots were fertilized twice per vegetation season, on 12 May and 20 July, using the following fertilizers dissolved in

Table I Fertilizer treatments and amount of nutrients applied in one dressing. Numbers given in brackets correspond to the total seasonal amount of applied nutrients (two dressings together).

Treatment			
abbreviation	$N (kg ha^{-1})$	P(kg ha <sup>-1</sup> )	K(kg ha <sup>-1</sup> )
Cont	_	_	_
$N_1$	150 (300)	_	-
$N_2$	300 (600)	_	_
$P_1$	_	40 (80)	_
$P_2$	_	80 (160)	_
K	_	_	100 (200)
$N_1P_1$	150 (300)	40 (80)	-
$N_1P_1K$	150 (300)	40 (80)	100 (200)
$N_2P_1K$	300 (600)	40 (80)	100 (200)
$N_2P_2K$	300 (600)	80 (160)	100 (200)

water: ammonium nitrate with lime (NH<sub>4</sub>NO<sub>3</sub> + Ca-CO<sub>3</sub>, 27.5% N, 10% Ca), superphosphate (Ca(H<sub>2</sub>- $PO_4$ )2 + CaSO<sub>4</sub>, containing 8.5% P, 20% Ca, 10% S) and potassium chloride (KCl, 50% K, 47% Cl). The position of individual pots was changed at weekly intervals to avoid any pseudoreplication and edge effects.

The pot volume was 30 L, and the pot surface area was 1963 cm<sup>2</sup> (pot diameter 50 cm). Clay soil with the following chemical properties was used: Ntotal = 919 mg  $kg^{-1}$ ,  $K_{MehlichIII}$  160 mg  $kg^{-1}$  (good K availability) (Madaras and Lipavský, 2009), P<sub>MehlichIII</sub> 16 mg kg<sup>-1</sup> (low P availability) (Kulhánek *et al.*, 2007),  $Mg_{MehlichIII}$  373 mg  $kg^{-1}$  (high Mg availability),  $Ca_{Me}$ hlichIII 10 501 mg kg<sup>-1</sup> (very high Ca availability) and CaCl<sub>2</sub> pH 7.96. Potassium-rich soil was used to avoid any risk of K limitation on growth, as the emphasis was placed on N and P nutrition in this study. To ensure that growth was not limited by K, treatments with K applications (K, N<sub>1</sub>P<sub>1</sub>K and N<sub>2</sub>P<sub>2</sub>K) were used.

At the study site, the long-term mean annual temperature was 8·2°C (ranging from 6·4 to 9·7°C), and the mean annual precipitation was 422 mm (ranging from 255 to 701 mm; Prague-Ruzyně meteorological station, 1955-2007). The mean temperature from June to September 2008 and 2009 (main vegetation season) was 17.6 and 17.9°C, respectively, and from December to March 2008/2009 and 2009/2010 (winter season), it was 2.4 and 0°C respectively. During the winter season 2008/2009, the lowest measured temperature (-13·2°C) was recorded on 3 January 2009, and during the winter season 2009/2010, the lowest temperature (-19.8 °C) was recorded on 27 January 2010.

# Seedling emergence

Seeds of R. obtusifolius subsp. obtusifolius (hereafter referred to as R. obtusifolius) and R. crispus were collected during autumn 2007 and 2008 from a region near Prague city in central Czech Republic. The collection sites were mainly roadside ditches or abandoned fields. Seed material was collected from a group of plants at three localities. Five plants were selected randomly at each site, taking care not to favour tall or small plants. The seeds were stored at room temperature, in paper bags in the dark. Germination was tested under laboratory conditions in a day/night light regime at 20°C. Germination was performed in four Petri dishes, each with fifty seeds for each species before seeding. Germination was 95% for both species. Twenty visually undamaged and fully ripened seeds of R. obtusifolius and R. crispus were sown into each pot with pre-fertilized soil on 19 May 2008 and the following year on 18 May 2009. The depth of sowing was 1-2 cm, and the pots were watered if necessary to maintain optimal moisture conditions for plant growth. The cumulative number of seedlings was recorded up to 26 June, in both 2008 and 2009, and no emergence of other seedlings was recorded after this date.

#### Plant performance

Three of the most developed plants were left in each pot after 26 June 2008 and 2009. The following data were collected at weekly intervals: (i) number of leaves per plant; (ii) number of leaves per main stem; (iii) length of the longest leaf; (iv) height of the plant; and (v) number of stems per plant.

# **Over-wintering**

The plants were not cut during the vegetation season or in the autumn. The pots were not protected by any covering or insulation from frost, and they were exposed to normal weather conditions during the winter. The number of plants that survived the winter was counted on the 1 April 2009 and 10 April 2010.

# Data analysis

All analysis was performed using STATISTICA 8.0 software (Statsoft, Tulsa, OK, USA). Emergence and winter survival data were evaluated by factorial Anova for species and treatment effects and their interactions. After obtaining significant results, multiple comparisons using Tukey's HSD test were applied to identify significant differences between treatments for each species. The number of stems per main stem, number of leaves per plant, length of longest leaf, number of stems or growing points and plant height data were evaluated using a mixed ANOVA model for species, treatment and time effects and time and treatment interactions.

As the results from both seasons, 2008/2009 and 2009/2010, were highly comparable, only the data from the first season have been included in the results section, and they are summarized in graphs.

#### Results

#### **Emergence**

When analysed by factorial ANOVA, it was found that emergence was significantly affected by species (F = 16.9, P < 0.001) and treatment (F = 19.4,P < 0.001), but not by their interaction (F = 1.2, P = 0.32). The mean emergence over all levels of nutrient availability was 39% and 49% for R. obtusifolius and R. crispus respectively. In both species, emergence was lowest in all treatments with N<sub>2</sub> applications  $(N_2, N_2P_1 K \text{ and } N_2P_2K)$ , and these treatments were

significantly different from the P<sub>1</sub> and P<sub>2</sub> treatments (Figure 1).

#### Plant performance

#### Number of leaves per main stem

The number of leaves per main stem was significantly affected by species (F = 1064, P < 0.001), treatment (F = 29, P < 0.001), time (F = 68, P < 0.001) and all of their interactions. In the case of R. obtusifolius, the greatest number of leaves (6.4) was recorded for the N<sub>2</sub>P<sub>1</sub>K treatment on 1 August. For the N<sub>1</sub>P<sub>1</sub>, N<sub>1</sub>P<sub>1</sub>K and N<sub>2</sub>P<sub>2</sub>K treatments, the greatest number of leaves (from 4.5 to 6) was recorded on 1 August, but for the Control, N<sub>1</sub>, N<sub>2</sub>, P<sub>2</sub> and K treatments, the greatest number of leaves (from 0.8 to 4.3) was recorded on either 21 or 28 August (Figure 2a). No stems of R. crispus developed in any of the fertilizer treatments during the seeding season.

#### Number of leaves per plant

The number of leaves per plant was significantly affected by species (F = 25.1, P < 0.001), treatment (F = 106.4, P < 0.001), time (F = 63.8, P < 0.001) and all of their interactions. In the first sampling dates (seedlings stage), the number of leaves in both species was greater in P<sub>1</sub> and P<sub>2</sub> than in other treatments (Figure 2c, d). On the other hand, the number of leaves was greatest in all treatments with N applications on the last sampling date. Generally, the greatest number of leaves was recorded in all treatments with N<sub>1</sub>P<sub>1</sub> or N<sub>1</sub>P<sub>1</sub>K applications in the period from 24 July to 21 August. In the case of R. obtusifolius, the absolutely greatest number of leaves (20 per plant) was recorded for the N<sub>2</sub>P<sub>1</sub>K treatment on 1 August. In the case of R. crispus, the absolutely greatest number of leaves (24 per plant) was recorded for the N<sub>1</sub>P<sub>1</sub> treatment on the 1 August. The number of leaves was significantly and positively affected by the number of growing points (Rumex obtusifolius: y = 2.0393 + 2.4171x;  $R^2 = 0.703$ , P < 0.001; Rumex crispus: y = 0.07 + 0.4x;  $R^2 = 0.95$ , P < 0.001) and by plant height (*Rumex obtusifolius*: y = 30.6 + 1.6x;  $R^2 = 0.15$ , P = 0.006; Rumex crispus: y = 2.38 + 0.22x;  $R^2 = 0.79$ , P < 0.001).

#### Length of the longest leaf

The length of the longest leaf was significantly affected by species (F = 15.1, P < 0.001), treatment (F = 23, P < 0.001)P < 0.001), time (F = 82.9, P < 0.001) and all of their interactions. In the seedling stage, the length of the longest leaf was greatest for the P2, P1 and N1P1K treatments (Figure 3a, b) in both species. On the other hand, leaves of R. obtusifolius were longest for N2 and N1 treatments, but the leaves of R. crispus were longest for the N<sub>2</sub>P<sub>2</sub>K, N<sub>1</sub>P<sub>1</sub>, N<sub>2</sub> P<sub>1</sub>K and N<sub>1</sub>P<sub>1</sub>K treatments on the last sampling date. In the case of R. obtusifolius, the absolutely longest leaves (33 cm) were recorded on 1 August, but in the case of R. crispus, the absolutely longest leaves (34 cm) were recorded for treatment N<sub>2</sub>P<sub>2</sub>K on 4 September.

# Number of stems or growing points

The number of stems or growing points was significantly affected by species (F = 122, P < 0.001), treatment (F = 189.5, P < 0.001), time (F = 162.5, P <0.001) and all of their interactions. In the seedling stage, the number of growing points was highest for P<sub>2</sub> treatment in both species (Figure 4a, b). On the

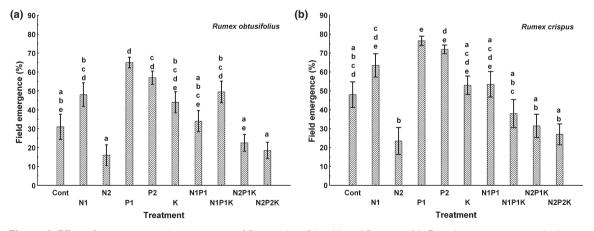


Figure 1 Effect of treatments on the emergence of Rumex obtusifolius (a) and R. crispus (b). Error bars represent standard errors of the means (SE). Treatment abbreviations are given in Table I. Using Tukey's post-hoc test, treatments with the same letter were not significantly different at the 0.05 probability value.

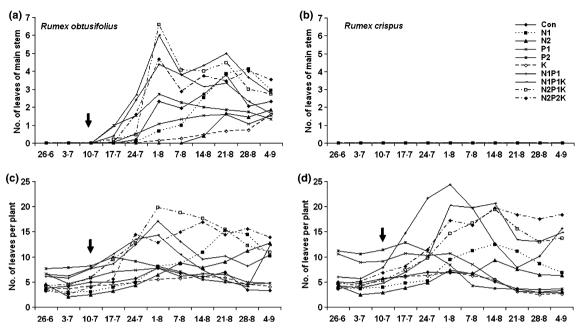


Figure 2 Effect of treatments on the number of leaves on the main stem of Rumex obtusifolius (a) and R. crispus (b) and the number of leaves on whole plants of R. obtusifolius (c) and R. crispus (d). Treatment abbreviations are given in Table I. Arrows indicate the beginning of the stem phase.

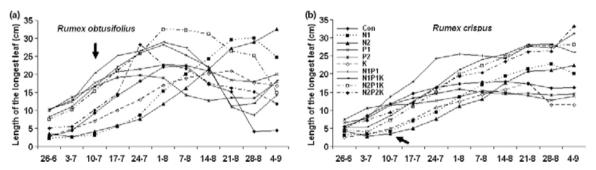


Figure 3 Effect of treatments on the length of leaves of Rumex obtusifolius (a) and R. crispus (b). Treatment abbreviations are given in Table 1. Arrows indicate the beginning of the stem phase.

other hand, the number of stems of R. obtusifolius was greatest for N<sub>2</sub>P<sub>1</sub>K and N<sub>1</sub>P<sub>1</sub>K treatments, and the number of growing points of R. crispus was greatest for the N<sub>2</sub>P<sub>2</sub>K and N<sub>1</sub>P<sub>1</sub>K treatments on the last sampling date. The greatest number of stems (4.9) and growing points (7.5) was recorded on the last sampling date, 4 September.

# Plant height

Plant height was significantly affected by species (F = 197.5, P < 0.001), treatment (F = 79, P < 0.001), time (F = 214, P < 0.001) and all of their interactions. In the seedling stage, the plant height of both species was not affected by treatment (Figure 4c, d). The growing points of both species started to become prolonged first in P<sub>1</sub> and P<sub>2</sub> treatments on 10 July, as the plant height was the highest of all treatments on this date. In both species, the highest plants were recorded for the  $N_1P_1$ ,  $N_2$   $P_1K$ ,  $N_1P_1K$  and  $N_2P_2K$ treatments on the final sampling date, 4 September. In the case of *R. obtusifolius*, the highest plants (66 cm) were recorded for N<sub>1</sub>P<sub>1</sub> treatment on 4 September, and in the case of *R. crispus*, the highest plants (6·7 cm) were recorded for treatment N<sub>2</sub>P<sub>2</sub>K on the same sampling date.

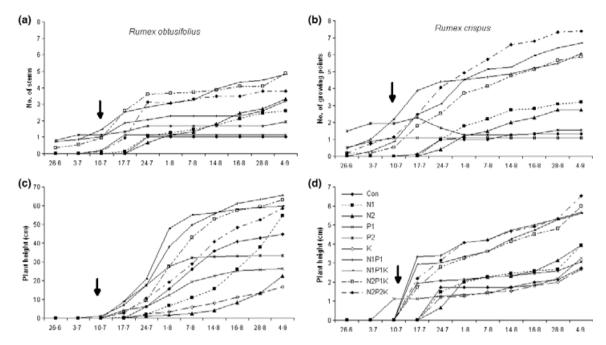


Figure 4 Effect of treatments on the number of stems of Rumex obtusifolius (a) on the number of growing points of R. crispus (b) and on the height of R. obtusifolius (c) and R. crispus (d). Treatment abbreviations are given in Table I. Arrows indicate the beginning of the stem phase.

### Over-wintering

Winter survival was significantly affected by species  $(F = 1 \ 009, \ P < 0.001), \ \text{treatment} \ (F = 5, \ P < 0.001)$ and species and treatment interaction. Winter survival over all levels of nutrient availability was 18 and 100% for R. obtusifolius and R. crispus respectively (Figure 5a, b). Survival of *R. obtusifolius* was zero in the K treatment and was more than 30% in the N<sub>1</sub>P<sub>1</sub> and N<sub>2</sub> P<sub>1</sub>K treatments. No mortality was recorded for R. crispus.

# Discussion

# **Emergence**

Emergence was negatively affected by very high N availability in both species, as the lowest rate of emergence was recorded in all the treatments with N2 applications. However, it should be noted that emergence was only evaluated after the first application of nutrients ( $N_2 = 300 \text{ kg N ha}^{-1}$ ). This was attributed to a negative effect of high NH<sub>4</sub><sup>+</sup> and/or NO<sub>3</sub><sup>-</sup> concentrations in the soil solution on seed germination. On the other hand, applications of lower N rates  $(N_1 = 150 \text{ kg N ha}^{-1})$  slightly stimulated germination. Negative effects of high NO<sub>3</sub><sup>-</sup> concentrations on the germination of R. obtusifolius, and stimulating effects of low concentrations, were also recorded by Milberg

(1997) and Klimeš (1996). Similarly, Mandák and Pyšek (2001) reported a negative effect of a high NO<sub>3</sub> concentration on the germination of Atriplex species. No germination, in either Rumex species, was recorded in an experiment with NH4NO3 fertilizer applications in contrast to normal germination in an experiment with Ca(H<sub>2</sub>PO<sub>4</sub>)<sub>2</sub> fertilizer applications in a laboratory germination performed by Křišťálová (2010). On the other hand, there was a clear positive effect of P1 and P2 treatments on the emergence of both species. This is consistent with the conclusion by Jenkins and Ali (1999) that P is an essential nutrient which is especially important in the promotion of early plant growth. Emergence was not affected by K applications because the clay soil was able to supply the seedlings with an adequate amount of K even without K application. The same results, i.e. a negative effect of a very high N supply and a positive effect of P supply on the emergence of both species, were also recorded in the second experimental year (data not shown here).

Although both species are generally believed to be nitrophilous (Milberg, 1997; Zaller, 2004), they suffer from the effects of very high N availability during seed germination and seedling emergence, in contrast to mature plants. The negative effect of a very high N application rate on the seedling emergence of both Rumex species seems to be a possible practical measure that might be used for decreasing Rumex seedling

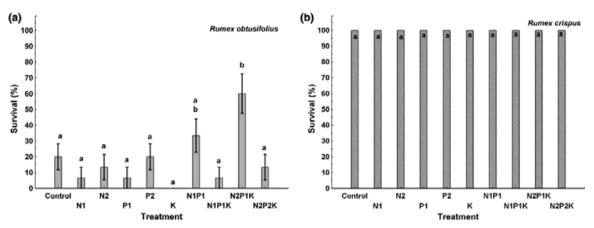


Figure 5 Effect of treatments on the over-wintering of Rumex obtusifolius (a) and R. crispus (b). Error bars represent standard errors of the means (SE). Treatment abbreviations are given in Table 1. Using Tukey's post-hoc test, treatments with the same letter were not significantly different at the 0.05 probability value.

emergence in locally disturbed patches after the removal of established Rumex plants in permanent grasslands, but this requires further investigation.

#### Plant performance

Three distinct life phases were recognized in the lifespan of R. obtusifolius: (i) a phase of germination and seedling emergence, (ii) a phase of rosettes and (iii) a phase of stem growth, flowering and seed ripening. Different effects of fertilizer treatments were recorded in each phase of plant growth. The germination, emergence and stabilization of seedlings took 3 weeks. Rosette growth was recorded from 19 May until 10 July, during which the supply of P appeared to play the main role. Seedlings of both species developed fastest in P<sub>1</sub> and P<sub>2</sub> in all treatments and had the greatest number of leaves during this phase. A further negative effect of a very high N supply on both species was recorded, as leaf growth during the rosette phase was depressed in all treatments with a N<sub>2</sub> supply (Figure 2). This clearly demonstrates a high P demand of Rumex plants in early development, but lower adaptation to a very high N supply.

The maturation, flowering and seeding period was recorded from 19 May until 4 September. Nitrogen, applied together with K and P, strongly increased plant performance (plant height, number of stems, number of leaves and length of the longest leaf) during the phase of stem growth, flowering and seed ripening. A delay of plant development was recorded for N1 and N2 treatments, probably due to P limitation in these treatments as the plants may have required time to increase their root systems to be sufficiently well developed to acquire enough P from the scarce amount in the soil. This indicates that a plant-available concentration of Mehlich III P of 16 mg kg<sup>-1</sup> was not sufficient for the optimal growth of both species. In addition to P limitation in the N treatments, high N availability prolonged the vegetation season and seed ripening, and this effect of high N supply is in accordance with the results of Vos and Biemond (1992).

Although R. obtusifolius was recognized by Humphreys et al. (1999) as being highly K demanding, no response to K application was recorded in our study as no obvious differences in plant performance were recorded between  $N_1P_1$  and  $N_1P_1K$  treatments. This indicates the ability of the clay soil that was used to supply an adequate amount of K for the growth of both species, even without K application. Evidently, a plantavailable (Mehlich III) K concentration of 160 mg kg<sup>-1</sup> was sufficient for the growth of both species. The results of our pot experiment are consistent with the conclusions by Kayser and Isselstein (2005) that K limitation in grassland is less likely on clay soils.

Stem production and the flowering of R. obtusifolius were recorded in the year of seeding, but there was no stem elongation or flowering of R. crispus in the year of seeding. The same result was recorded with genetically similar plants in the pot experiments on sandy soil as well as under field conditions on several soil types at different altitudes and in different years (Křišťálová, 2010). It seems that R. crispus requires a cold period and a sufficient amount of carbohydrates to induce stem growth and flowering. According to Cavers and Harper (1964), flowering and seed set can occasionally occur in the year of seeding, but not usually until the second year after germination. According to Hume and Cavers (1983) and Bond et al. (2007), plants of R. crispus in most North American populations require over-wintering before flowering can occur, unlike in the populations in Britain and the rest of Europe. The results of this study indicate that, at least in some populations of R. crispus in the Czech Republic, no flowering can occur in the year of seeding under all levels of nutrient availability. On the other hand, R. obtusifolius can flower in the year of seeding.

# **Over-wintering**

According to Cavers and Harper (1964), both R. crispus and R. obtusifolius have a tendency to be monocarpic and to die after producing seeds. Monocarpic behaviour was supported by almost no over-wintering of R. obtusifolius after flowering in this study, but high over-wintering of the same genotype of R. obtusifolius was recorded in our field experiments in the same and other winter seasons (Křišťálová, 2010). Furthermore, there was no difference in over-wintering between sterile and fertile plants. It seems that the main reason for low over-wintering in the pot experiments was the low resistance of mature plants of R. obtusifolius to frost, as pots were exposed to winter conditions without any protection. On the other hand, R. crispus possessed substantially higher cold resistance as no winter mortality was recorded. Similar results were recorded after the next winter season (2009-2010) which had heavy and long-term frosts: 100% mortality of R. obtusifolius in contrast to no mortality of R. crispus in the pot experiments on clay and sandy soils (Křišťálová, 2010). The low frost resistance of R. obtusifolius probably explains why a high winter mortality of adult R. obtusifolius plants was recorded in field experiments by Hongo (1989a,b), Martinkova et al. (2009) and Pavlů et al. (2008a). Furthermore, the high mortality of R. obtusifolius after severe winters was recorded by many farmers in the Czech Republic. The substantially higher frost resistance in R. crispus than in R. obtusifolius revealed in this study may explain why R. crispus is common in regions with a continental climate and heavy winter frosts, whereas R. obtusifolius is more common in oceanic regions with less severe winters. In addition, R. crispus, in contrast to R. obtusifolius, is able to grow up to latitudes of 69°N and 65°N in Europe and North America respectively (Cavers and Harper, 1964). The over-wintering of R. obtusifolius was only marginally affected by N, P and K availability, and the overwintering of R. crispus was not affected.

#### **Conclusions**

Seedling emergence of both R. crispus and R. obtusifolius was negatively affected by very high N availability but positively affected by increased P availability in the soil. Similarly, the seedlings were negatively affected by a very high N supply and developed fastest and had the greatest number of leaves in P treatments during the rosette phase. Both of these Rumex species are therefore sensitive to a very high N supply in early developmental stages, and they tend to tolerate a very high N supply from the phase of fully developed rosettes. In addition to high N requirements during the phase of stem growth, flowering and seed ripening, both species also require a high P supply. Similarly to most North American populations, at least some Central European populations of R. crispus do not flower in the seeding year, in contrast to R. obtusifolius. The high winter mortality of R. obtusifolius contrasted markedly with the absence of winter mortality in R. crispus, indicating that R. crispus possesses higher frost resistance. Over-wintering was only marginally affected by N, P and K availability.

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