



# Continuous and cumulative acidification and N deposition induce P limitation of the micro-arthropod soil fauna of mineral-poor dry heathlands

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## ABSTRACT

Phosphorus content of mineral-poor sandy soils is steadily decreasing due to leaching caused by continuous and cumulative acidification and N deposition. Sod-cutting as a traditional restoration measure for heathland vegetation appears to increase P limitation, as most of the P present is in the organic matter being removed by sod-cutting. Mineral weathering, the natural inorganic source of P, becomes limiting or has even ceased as a result of the depletion of minerals. Previous investigations indicate a P limitation of the macrofauna under these circumstances. If this also holds for the soil fauna, hampering of decomposition may occur. To test experimentally whether soil fauna is indeed limited by the amount of P in the system, we set up an experiment in sod-cut heathland in which we added P or Ca (as Dolokal), resulting in: P + Ca+, P + Ca-, P-Ca+ and P-Ca- (control) treatments and an extra reference block in the original grass encroached heathland vegetation. The Ca treatment was added because liming is used to recover from acidification effects, but as a side effect Ca may also bind P. Three growing seasons after the addition of P and Ca, we found a significant increase in herbivores and predators among the soil fauna, with herbivore numbers higher in the P+/Ca-plots than in the P+ /Ca + plots, indicating a lower availability of P in the presence of added Ca. Predators increased in all P+ plots. Fungivorous browsers responded negatively to the treatment after three growing seasons, both to P and to Ca addition. Phoretic species responded rapidly either to fewer numbers (when these are fungivorous browsers) or to greater numbers (when these are herbivorous browsers) to P addition. P addition induced also an allometric effect, via the medium-sized species increasing in greater numbers than both the larger and smaller species.

## 1. Introduction

Acidification and N deposition are considered major threats for the conservation of dry heathlands in Western Europe (Bobbink et al., 1998; Bobbink and Roelofs, 1995; De Graaf et al., 1997; De Graaf et al., 1998; Heil and Diemont, 1983). Critical loads of N deposition for dry heathland are exceeded for decades (Bobbink and Roelofs, 1995; Bobbink et al., 2010), resulting in a shift from a dwarf shrub dominated vegetation to a grass encroached vegetation in which herb species have also become much rarer (De Graaf et al., 2009; Kleijn et al., 2008; Roem et al., 2002). These shifts are explained by lifting the N limitation of particular plant species (Roem et al., 2002) and by differences in sensitivity to Al toxicity of others: N deposition causes severe acidification in the top soil, which releases Al at pH lower than 4.2 (Bobbink et al.,

1998; Houdijk et al., 1993). The focus in restoration management has been to combat these changes by removing as much N as possible by sod-cutting (Diemont, 1996) in order to restore the former N limited ecosystem.

Sod cutting has been proven highly efficient in restoring ericaceous dwarf shrub dominance in grass encroached vegetation (Diemont, 1996). However, plant species diversity, especially herb diversity, hardly recovers. Aerts (1990) and later Roem and Berendse (2000), Britton and Fisher (2007) and Von Oheimb et al. (2010) showed the importance of N:P stoichiometry for interspecific competition among plant species. In a recent study, Vogels et al. (2017) found an impoverished invertebrate fauna correlated with an increased N:P ratio in the dominant plant species. They found a response of increased plant N:P ratio in both herbivorous and detritivorous Diptera and

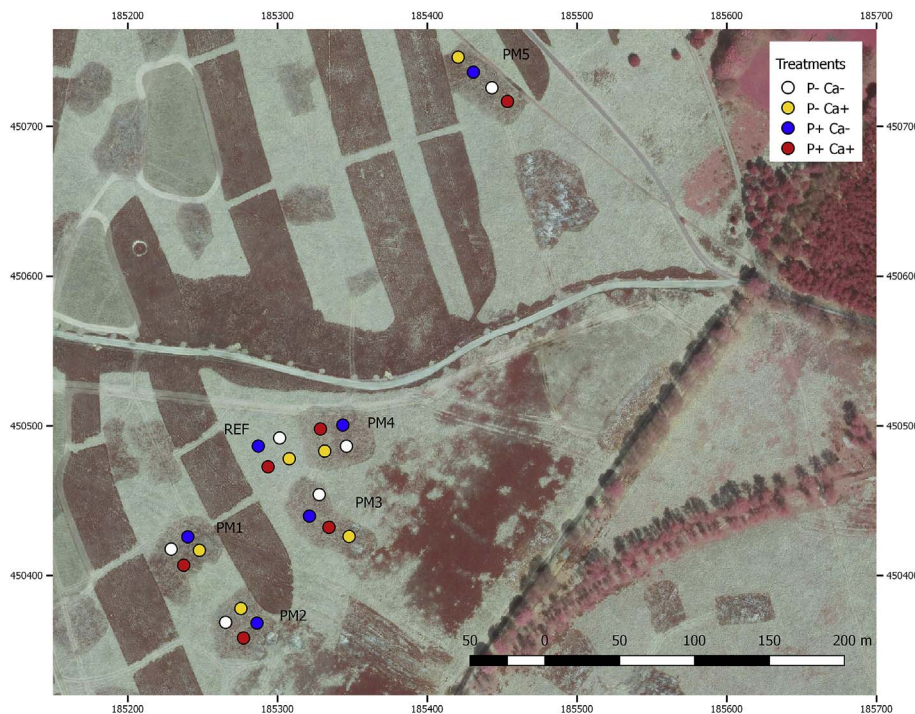
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**Fig. 1.** Aerial false-colour photo, taken in 2013, showing the lay-out of the experimental plots in De Hoge Veluwe National Park. Light blue surfaces are sod-cut. REF, reference (uncut) plots and replicates (PM) represent P +/Ca + red, P+/Ca-blue, P-/Ca + yellow and P-/Ca-grey plots of  $15 \times 15 \text{ m}^2$ . Distance between marginal numbers is 100 m.

herbivorous and carnivorous Carabidae. However, as detritivorous Diptera may feed on dead organic matter, fungal hyphae, or both, it remains unclear whether soil animals with different sizes, diets and life history strategies respond differently to sod-cutting and altered stoichiometry.

N:P ratios of terrestrial herbivores are significantly lower than those in plants (Elser et al., 2000), even in situations where these ratios have not become excessively high in plants. This indicates that P is more limiting for animals than N. An unforeseen effect of sod-cutting may therefore be that P limitation has become more severe for a large part of the fauna, as most of P was bound in the, by sod-cutting, removed organic matter in the humus form. Weathering, if still possible in the top soil by the presence of sufficient minerals, will take many years: Zehetner et al. (2008) found that about 50% of apatite P was taken up in organic matter in 500 years in Romania.

Barendregt and Siepel (2016) draw attention to the possible rigidity effects of nature conservation in keeping the same vegetation on the very same spot, as natural processes, such as acidification of the top soil continue and even are enforced by human influences. Mol et al. (2003) calculated that human-induced acidification in soils during the industrialized decades was equivalent in terms of leaching and weathering of minerals with the past 11,000 years. So, P containing minerals (e.g. apatite) in the top soil may be completely weathered and leached if P is not incorporated in organic matter. In heathlands, most P is bound in the organic matter in the A-horizon (Härdtle et al., 2009). However, most of the organic matter is removed by sod-cutting to combat the continuous influx of N, resulting in a possible severe P limitation. Under these conditions, only plant species can survive that are extremely efficient in P reallocation (indicated by a high N:P ratio in their tissues). As the fauna is even more susceptible to P limitation, effects must be seen there first. As also suggested in Vogels et al. (2017) an unforeseen negative side-effect of sod cutting may therefore be that P limitation has become more severe for a large part of the fauna.

Predominant fungi in heathlands are saprophytic, or form either ectomycorrhizal or ericoid mycorrhizal connections to plant roots. In the latter case they provide the plants with nutrients, particularly P via uptake from mineral soil (ectomycorrhiza) and/or from resistant organic matter (ericoid mycorrhiza) (Smith and Read, 2008). P contents

in fungi therefore are thought to be higher than in plants. So, it is hypothesized that fungivorous soil animals suffer less from P limitation than herbivorous animals do. Furthermore, larger animals need more P to complete their development, thus effects of P might be allometric. Mulder (2010) and Mulder and Elser (2009) show that larger animals especially suffer from P limitation as they found a clear allometric effect in occurrence of animal species under P limited conditions.

To test experimentally whether soil fauna is indeed limited by the amount of P in the system, we set up a full factorial experiment in sod-cut heathland. Liming in heathlands is often practised to restore the buffer capacity of the soil to acidification, however excess of Ca may inhibit P availability to plants (Haynes, 1982; Kooijman and Besse, 2002). Thus, in the experiment we added P or Ca (as Dolokal), addition noted as + and no addition as -, resulting in: P-Ca- (control), P + Ca-, P-Ca+ and P + Ca + treatments and an extra reference block in the original grass encroached vegetation (with *Molinia caerulea* dominance). Here 'control plots' refer to the sod-cut experiment without any addition and 'reference plots' refer to the original not sod-cut vegetation with or without additions. We focussed on the micro-arthropod soil fauna and formulate the following hypotheses: we hypothesize herbivores to respond positive to P addition, fungivores not to respond to P addition, but slightly to Ca addition and predators to respond positive to P addition, as a consequence of increased herbivore numbers. Concerning life-history tactics, we hypothesize a faster response in theytokous reproducing species than in sexual reproducing species, as the former have a faster reproduction, and a positive response of the better colonizers (phoretic species). Moreover, following the allometric rule of Mulder (2010), we hypothesize the most numerous positive response in populations of the larger species.

## 2. Material and methods

### 2.1. Site description and experimental design

Experimental plots were created in National Park De Hoge Veluwe, in the centre of the Netherlands ( $52^{\circ}.2'.30'' \text{ N}$ ;  $5^{\circ}.49'.50'' \text{ E}$ ). Plots are located on the gentle slope of a push moraine ridge with some cover sand. Soil type is a spodic dystrocept (Soil Survey Staff, 1999),

developed in moderately coarse sandy parent material containing about 13–18% loam in the deeper soil. The topsoil has been extensively leached, creating a more spodic topsoil, resembling the one of an orthod. The humusform is originally moderlike, but contains also larger proportions of amorphous organic compounds leaching into the B horizon as seen in orthods. Organic matter in the topsoil (0–10 cm) is  $6.8 \pm 0.3\%$ ,  $\text{pH}_{[\text{NaCl}]}$  is  $3.5 \pm 0.1$ . Vegetation in the reference plots (not sod-cut) is a dominance (facies) of purple moor grass (*Molinia caerulea*) in large tussocks. In the false-colour photograph in Fig. 1 the light-blueish surface has been sod cut. In the P-/Ca-plots in the sod-cut area recovering of vegetation is dominated by three species: *Molinia caerulea*, *Calluna vulgaris* and to a minor extent *Erica tetralix*. Vascular plant species diversity is low in the control plots ( $6.2 \pm 1.3$  [mean  $\pm$  sd] versus  $17.2 \pm 4.5$  in the P + Ca + plots). Vegetation cover of vascular plants is sparse in the control  $21.0\% \pm 7.1$  versus  $41.3 \pm 3.2$  in the P+/Ca + plots. Dominant moss species are the acid preferring species *Campylopus pyriformis* and *Dicranella heteromalla* in the P+/Ca-plots and the ruderal species *Funaria hygrometrica* and *Ceratodon purpureus* in the P-/Ca+ and P+/Ca + plots. Bryophyte species diversity ranges from  $2.7\% \pm 1.9$  [mean  $\pm$  sd] in the control to  $9.1 \pm 0.6$  in the P+/Ca + plots. Bryophyte cover is low in the control  $2.5 \pm 3.0$  versus  $55.1 \pm 3.2$  in the P+/Ca + plots.

Plots were  $15 \times 15 \text{ m}^2$  in size: five blocks with each four plots were randomly placed in the sod-cut area. Sod cutting was carried out in 2012 prior to the fertilizer additions. An experimental block design was used for P addition and Ca addition creating five replicate sets of P +/Ca+, P+/Ca-, P-/Ca+ and P-/Ca- (see Fig. 1). For reference to the original purple moor grass vegetation, an equally sized block of 4 plots was laid in there. P addition per plot was  $5 \text{ g m}^{-2}$  in the form of  $\text{NaH}_2\text{PO}_4 \cdot 2\text{H}_2\text{O}$ , wherein P is 19.85% by weight, so total fertilizer amount per plot was  $5 \times 15 \times 15 \times 1/0.1985 = 5.67 \text{ kg NaH}_2\text{PO}_4 \cdot 2\text{H}_2\text{O}$ . This addition created a 60% increase of total P in the soil ( $3.0 \pm 0.15 \text{ mmol P.l}_{[\text{soil}]}$  on 7 July 2012 and  $4.85 \pm 0.1 \text{ mmol P.l}_{[\text{soil}]}$  on 24 September 2013). P-fertilizer was spread out by hand on 24 April 2013 on the bare (sod-cut) soil, or on the vegetation in the reference plots. Ca was added as granular Dolokal (80% Ca and 20% Mg) five months prior to P fertilizer to reduce P adsorption to Ca-complexes, making P less available. Per Ca + plot 45 kg Dolokal has been used ( $200 \text{ g m}^{-2}$ ). Exchangeable Ca increased in the Ca + plots with 716.7% ( $600 \pm 50 \text{ mol l}_{[\text{soil}]}$  on 7 July 2012 and  $4300 \pm 450 \text{ mol l}_{[\text{soil}]}$  on 24 September 2013) and  $\text{pH}_{[\text{NaCl}]}$  increased from  $3.5 \pm 0.1$  to  $4.13 \pm 0.1$ .

## 2.2. Micro-arthropod sampling and identification

Soil samples were taken on 3 November 2015, three growing seasons after the application of P and Ca. At this time vegetation was in recovery phase, but not yet in full closure. On each replicate plot three soil cores of 5 cm depth and  $100 \text{ cm}^3$  content were sampled and extracted on a Tullgren funnel for 7 days. During that period temperature was increased from 35 to 45 °C. Ethanol 70% was used as conservation fluid and micro-arthropods obtained were put into lactic acid 40% for clarification and identification (Siepel and Van de Bund, 1988). Nomenclature and identification for the main groups is according to Weigmann (2006) for Oribatida, Karg (1993) for Gamasina, Karg (1989) for Uropodina and Bretfeld (1999), Potapow (2001), Dunger and Schlitt (2011) and Jordana (2012) for Collembola.

After identification micro-arthropods were classified in life-history strategies according to Siepel (1994) and feeding guilds (Siepel and de Ruiter-Dijkman, 1993), as well as in size classes based on identification literature. Twelve life-history strategies are grouped into dispersal strategies (obligate phoretic, i.e. every generation is on the move (either as an adult – Strategy IV, or with special adaptation in the juvenile phase – Strategy III), or facultative (some generations can successively reproduce in the biotope – for instance a dead log – but eventually the species has to move to another site – Strategy II). Another group of

strategies are synchronization based: species having a real obligate diapause (Strategy V), combined with semelparity (Strategy VI), or anemochory (Strategy VII), or just have a form of quiescence (Strategy VIII). Finally strategies are discriminated based on their reproduction form: sexual reproduction (Strategy XI), for long living species with seasonal iteroparity (Strategy XII), apomictic thelytokous (asexual) reproduction (Strategy X), or automictic thelytokous reproduction (Strategy IX). Strategy I are animal parasites (Siepel, 1994). In terminology of feeding guilds for herbivores and fungivores, Siepel and de Ruiter-Dijkman (1993) is followed: defined on the activity of micro-arthropod gut enzymes (either produced by themselves or by micro-organisms in their gut): cellulase (plant feeding), trehalase (fungal contents feeding) and chitinase (also capable of digesting fungal cell walls) in combination with their preferred diet. Thus herbivorous grazers have cellulase activity, browsers not, fungivorous grazers have chitinase and trehalase activity, browsers only trehalase activity and herbofungivorous grazers have activity of all three enzymes, while opportunistic herbofungivores browse the fungi and graze the plant organic matter. Species feeding guilds not listed in Siepel and de Ruiter-Dijkman (1993) have been analysed later or are derived from literature (e.g. fungivorous and stylet-like chelicerae: fungivorous browsers). Predators can be grouped into specific nematode predators and general predators. Finally size classes used are: small ( $< 250 \mu$ ), medium ( $250\text{--}500 \mu$ ) and large ( $> 500 \mu$ ).

## 2.3. Statistical analysis

Differences in micro-arthropod species richness between the vegetated reference plots and the sod-cut plots were explored with linear mixed-effect models, with block as random factor. Similar analyses were done for the number of individuals per plot, but here we assumed a negative binomial error distribution (using the *glmer.nb* function in R) rather than a normal distribution. For the sod-cut plots we used *glmer.nb* to study the effect of P and Ca addition on the number of individuals. In these analyses species traits (either their feeding guild, size class or life history Strategy) were also used as an explanatory variable. As in all cases the effects of these species traits interacted significantly with the effects of P or Ca addition, we also performed separate analyses per level of that species trait. For the analysis of the effect of feeding guild we used a subset without omnivores, bacterivores and herbofungivorous grazers because these feeding guilds were represented by only 31, 8 and 20 individuals, respectively. The remaining 6 guilds had at least 119 individuals each, and 7229 individuals in total. Similarly, in the analysis of P+ and Ca + effects per life history Strategy, only strategies represented by at least 265 individuals were included.

## 3. Results

A total of 7251 individual micro-arthropods were caught and identified, distributed over 74 species. Highest species richness (35) was found in reference (uncut sod) plot P-/Ca+, while the lowest number of species (12) was found in the P-Ca-control plot in sod-cut block 2 and plot P-/Ca+ in block 5. Reference plots did not differ significantly for neither P nor Ca addition, therefore these were tested together against sod-cut plots. In reference plots species richness was higher than on the sod-cut plots ( $29.0 \pm 7.1$  vs.  $18.6 \pm 6.1$  [mean  $\pm$  sd];  $p = 0.015$ ). However, individual numbers showed the reverse, being much lower in the reference plots than in the sod-cut plots ( $99.8 \pm 33.5$  vs.  $342.5 \pm 269.2$ ;  $p < 0.001$ ) predominantly due to the fertilizer applications: P-/Ca-control plots had a lower number of individuals ( $159.0 \pm 68.9$ ), which did not differ significantly from the reference plots ( $p = 0.06$ ). Densities in the plots varied from  $9300 \text{ m}^{-2}$  (control P-/Ca-) to  $189,500 \text{ m}^{-2}$  (Block 4, P +/Ca-), so ranging from a situation comparable with the moss phase in drift sands to twice as high compared to average heathland densities (Siepel and Nijssen, 2010).

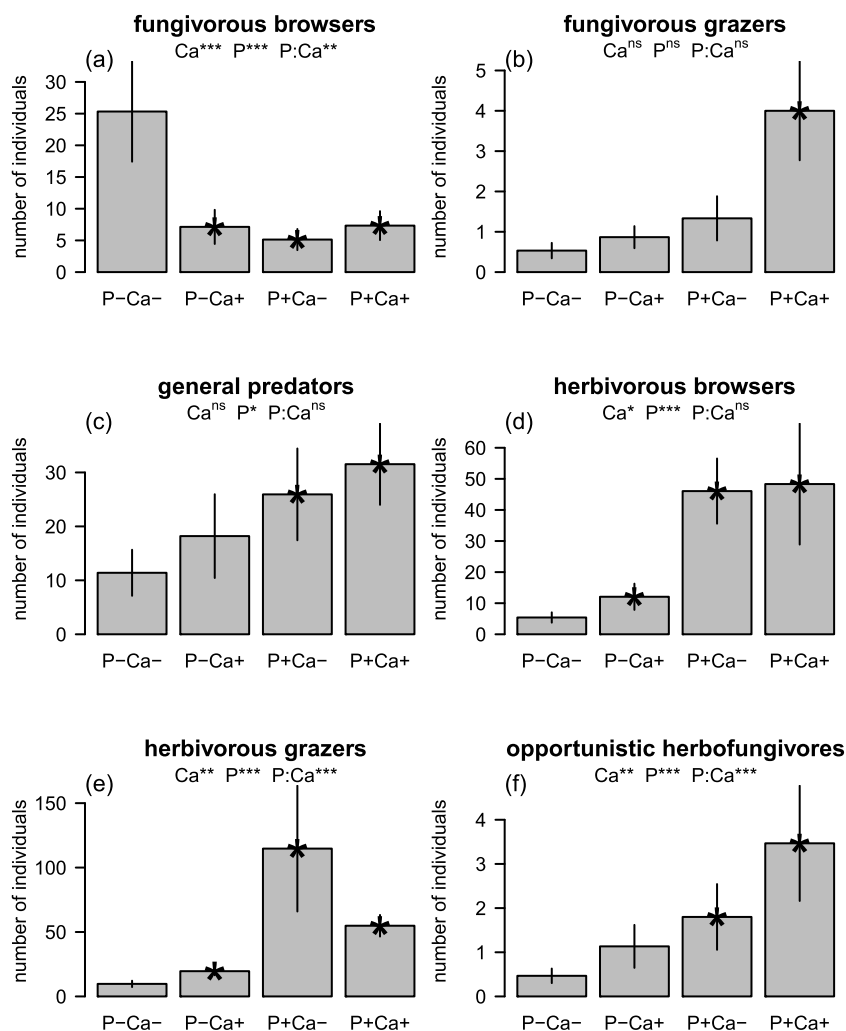
**Table 1**  
Systematic group, name, feeding guild and reference for the ten dominant species in the experiment.

Systematic group	Name	Feeding guild	Reference
Sarcoptiformes; Endeostigmata	<i>Alycus roseus</i> C.L. Koch, 1842 <i>Nanorchestes arboriger</i> (Berlese, 1904)	nematode predator herbivorous grazer	Walter (1988) Schuster and Schuster (1977)
Mesostigmata: Gamasina	<i>Asca bicornis</i> (Canestrini and Fanzago, 1887) <i>Rhodacarus coronatus</i> Berlese, 1921	general predator general predator	Hurlbutt (1963) Sardar and Murphy (1987)
Prostigmata: Heterostigmatina	<i>Scutacarus quadrangularis</i> (Paoli, 1911) <i>Hemitarsonemus</i> sp.	fungivorous browser herbivorous browser	Karafiati (1959) Schaarschmidt (1959)
Prostigmata: Eupodides	<i>Microtydeus subterraneus</i> (Wood, 1965) <i>Eupodes</i> sp.	herbivorous browser herbivorous grazer	Siepel and Nijssen (2010) Booth et al. (1985)
Collembola	<i>Isotoma viridis</i> Bourlet, 1839 <i>Sphaeridia pumilis</i> (Krausbauer, 1898)	herbivorous grazer herbivorous grazer	Poole (1959) Blanquaert et al. (1982)

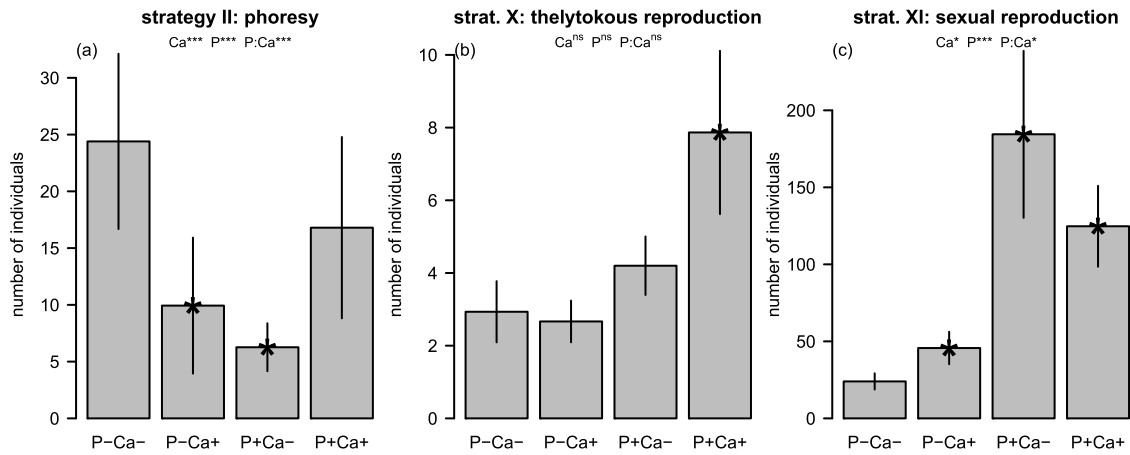
Dominant species are listed with their feeding guilds and references in Table 1. These ten dominant species form together 87.5% of the total number of individuals. In the supplementary material a PCA is presented on the feeding guilds of the micro-arthropods (displayed are all plots [numbers], feeding guilds [arrows] and treatments [circles]), as well as the complete dataset of species and numbers per plot.

The effect of P addition interacted with the effect of feeding guild on

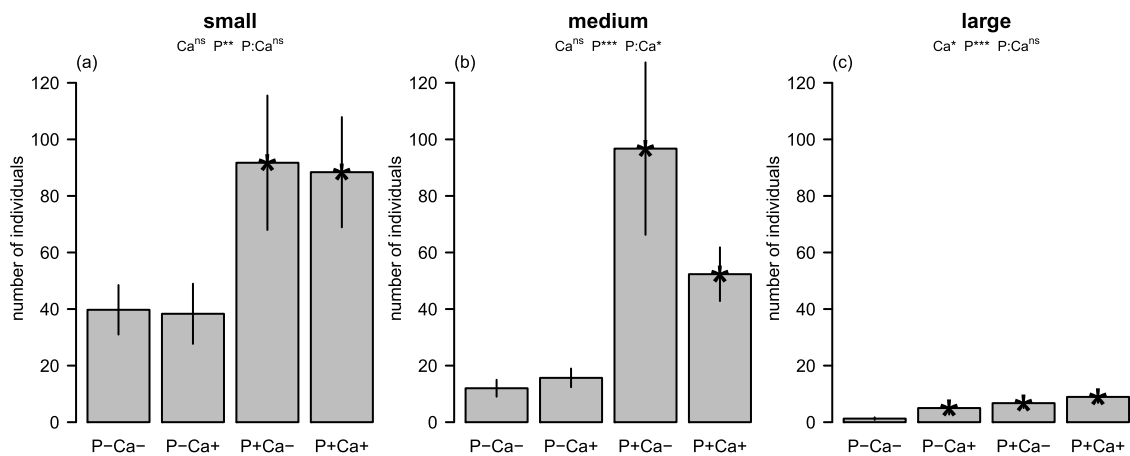
the number of individuals. We therefore performed separate analyses for each of the 6 most common feeding guilds (Fig. 2). In Figs. 2–4, data of the replicate plots have been pooled (without reference plots) and mean and s.d. are presented as well as significance levels of treatments and interaction. The guilds general predators (Fig. 2c), herbivorous browsers (Fig. 2d) and opportunistic herbofungivores (Fig. 2f) showed a significant positive effect of P addition, while the effect of Ca addition



**Fig. 2.** For each of the 6 most common feeding guilds, the mean number of individuals is given per plot, separately for the 4 treatments (bars are standard errors). The text above the plots give the p-values of P, Ca and the P:Ca-interaction effects. Stars in the plots indicate levels that are significantly different from the P-Ca- (left) bar. Please note the different scales on the y-axes.



**Fig. 3.** For each of the 3 most common life history strategies, the mean number of individuals is given per plot, separately for the 4 treatments (bars are standard errors). The text above the plots give the p-values of P, Ca and the P:Ca-interaction effects. Stars in the plots indicate levels that are significantly different from the P-Ca- (left) bar. Please note the different scales on the y-axes.



**Fig. 4.** For each of the 3 size classes, the mean number of individuals is given per plot, separately for the 4 treatments (bars are standard errors). The text above the plots give the p-values of P, Ca and the P:Ca-interaction effects. Stars in the plots indicate levels that are significantly different from the P-Ca- (left) bar.

was smaller (but also positive). P and Ca addition effects were additive in these guilds and did not interact significantly, except for opportunistic herbofungivores. In herbivorous grazers (Fig. 2e) there was a significant interaction: numbers were increased most when P was added without Ca. The fungivorous browsers (Fig. 2a) were the only group to show an overall negative effect of P and Ca addition, independent of which combination of P and Ca was applied. For fungivorous grazers (Fig. 2b) numbers were too small to conclude an effect: only 2.3% of the individuals in the sod-cut plots versus 21.9% in the uncut reference plots. The latter was only slightly lower than the average in heathland vegetation (25%; Siepel and Nijssen, 2010).

Composition of life-history strategies among reference and experimental plots, however, was rather different: the fraction of Strategy X (thelytokous species) in the sod-cut plots was 7.4%, while in the uncut reference plots it was 40.6% (overall average in heathlands is 25%; Siepel and Nijssen, 2010). P and Ca addition had differential effects on micro-arthropods with respect to their life history strategies (Fig. 3). For phoretic Strategy (II) (Fig. 3a), either P or Ca addition resulted in a reduction in the number of animals, but when both were added the decrease was smaller. Sample sizes were small for the thelytokous reproduction Strategy (X) (Fig. 3b), but when both P and Ca were added,

numbers seem to increase, but not significantly. For the most abundant life history Strategy, sexual reproduction (XI) (Fig. 3c), Ca and especially P had significant positive effects, although their effects were not additive: the P + Ca + treatment resulted in an increase that was intermediate to the separate Ca + and P + effects; the Ca addition hampered the P effect significantly.

The smallest size class contained the most individuals, followed by the medium size class, while the large size class was almost absent. Addition of P significantly increased the number of individuals in all three size classes (by a factor 2.3 in the small size class (Fig. 4a), by a factor of 8.1 in the medium class (Fig. 4b) and by a factor 5.3 in the large size class (Fig. 4c)). The large size class, however, was very small in numbers. Ca addition, however, only significantly increased (by a factor of 4) the number of large-sized micro-arthropods. Only in the medium-size class did P + and Ca + significantly interact negatively and hampered the Ca addition the P effect.

#### 4. Discussion

Micro-arthropod species richness was highest in the reference plots (which were characterized by a vegetation dominated by purple moor

grass), and comparable to that of a dwarf-shrub dominated heathland vegetation (Siepel and Nijssen, 2010). Densities in the reference plots were, however, about four times lower than in a heathland vegetation, indicating that decomposition was rather hampered in the purple moor grass tussocks. As shown previously, sod-cutting does affect the species richness of soil fauna, as species of larger sizes are not able to enter the deeper soil layers (Siepel, 1996) and will be removed with the sods such as the moss mite species *Nothrus silvestris* Nicolet, 1855, *Spatiodamaeus verticillipes* (Nicolet, 1855), *Ceratoppia bipilis* (Hermann, 1804), *Trichoribatidites incisellus* (Kramer, 1897) and *Pergalumna nervosa* (Berlese, 1914) and consequently cannot react to additions in the experimental plots. Unlike the larger predatory mites, having a phoretic behaviour, these moss mites have a very limited dispersal capacity (Siepel, 1994). The sod-cutting thus contributed very much to the founder effect, one of the major reasons to analyse the data grouping individual species to feeding guilds and life-history tactics (Verberk et al., 2013). The vast majority of the species is very small, ranging from 150 to 250  $\mu\text{m}$ . Given the low density of micro-arthropods in the reference plots, even lower than in the sod-cut control P-/Ca-plots, we may also conclude that neither Ca, nor P addition had any effect when spread out on the current vegetation, all seems to be absorbed by the purple moor grass. So, sod-cutting prior to P addition is a prerequisite, if the additions have to become available in the soil system.

Numbers in sod-cut P-/Ca-plots did not significantly differ from reference plots, the tendency ( $p = 0.06$ ) towards higher numbers is mainly explained by an increase in phoretic fungivorous browsers (see equally sized bars P-/Ca-in Figs. 2a and 3a), taking advantage of a faster decomposition of remaining organic matter at the sod-cut plots due to higher temperatures (Siepel and van Wieren, 1990; Lang et al., 2014). Fungivorous browsers (unlike fungivorous grazers) can show this increase because of their faster dispersal (facultative phoretic, Strategy II; Siepel, 1994). These species are phoretic with flies and beetles that locate new spots of blooming hyphae. Fungivorous grazers did not show a clear effect. Summarizing, for fungivores we did not see an effect of P nor Ca addition, as hypothesized, but we see an increase of fungivorous browsers in the sod-cut P-/D-plots, where temperature increase was greatest due to the lowest vegetation cover. Herbivores, both opportunistic herbivores (in small numbers) and herbivorous browsers, showed positive, and additive, effects of P and Ca addition. On the contrary, herbivorous grazers (largest feeding guild in numbers here) did show an interaction effect: numbers increased most with only P addition and less in combination with Ca, which may reflect a minor availability of P in an Ca enriched environment (Haynes, 1982; Kooijman and Besse, 2002). General predators (e.g. *Asca bicornis* (G. Canestrini and Fanzago, 1875)) showed a positive reaction on both P and Ca addition, effects were cumulative, no interaction was found. Their response was directly related to the increase of micro-arthropod numbers in general (most notably in P-treatments) and most of these predators are capable of responding quickly as result of by their phoretic behaviour, or otherwise higher degree of mobility.

Differences in life-history strategies, as shown in Fig. 3 are to a certain degree related to differences between feeding guilds: the larger number of phoretic individuals (Strategy II) on P-/Ca-is entirely composed of fungivorous browsers (especially *Scutacarus quadrangularis* (Paoli, 1911)), while the larger number at P+/Ca+ is largely composed of herbivorous browsers (*Hemitarsonemus* sp.). The interaction effect seen at Strategy XI (sexual reproduction) is in majority accounted for by herbivorous grazers. Life-history Strategy X (thelytokous reproduction) is of minor importance here: the fraction in the sod-cut plots was very low. Thelytoky as a strategy (as in e.g. *Oppeilla nova* (Oudemans, 1902)) requires a predictable environment (Siepel, 1994; Gulvik et al., 2007). It is obvious that predictability of moisture and temperature regimes in a thick litter layer of purple moor grass tussocks is higher than in average heathland vegetation and indeed much higher than in recently sod-cut plots. Observed numbers were greatest in the P +/Ca + plots, covered with the most dense vegetation compared to other sod-cut plots. Apparently these treatments resulted in a quicker

recovery towards higher levels of environmental predictability.

Following the allometric rule of Mulder (2010), we hypothesized positive reactions of P addition in populations of the largest species. We did, indeed, find a significant increase of all size classes (Fig. 4) in response to P addition. Medium and large size classes responded however more than the smallest size class (a factor 8.1 and 5.3 versus 2.3 respectively). In the medium size class a negative interaction effect between P and Ca was seen, which is accounted mostly for by predators (e.g. *Rhodacarus coronatus* Berlese, 1921) and a somewhat by some herbivorous grazers. The largest size class showed both a positive effect of P as well of Ca addition (without interaction) and can be accounted for by predators (e.g. *Asca bicornis*). As a result of the sod-cutting before the addition experiments were executed, most of the larger species (moss mites) were removed with the sods and could therefore not react to any addition. In the small size class we saw an increase in numbers of the herbivorous grazers (largest feeding guild) *Nanorchestes arboriger* (Berlese, 1904) and *Eupodes* sp. As we take into account that medium and small size class species are more multivoltine and the large size class contains in majority univoltine species in still low numbers, we may conclude that our results follow our hypotheses in general given the restricted experimental period of three years.

The dominant plant species (*Molinia caerulea*, *Calluna vulgaris* and *Erica tetralix*) are all known to be highly efficient in their phosphate handling: *M. caerulea* is well adapted to P limitation (Kirkham, 2001) possibly because of its deep rooting strategy, enabling this plant species to acquire P that is stored in B and mineral C-horizons as well, while *C. vulgaris* and *E. tetralix* use ericoid mycorrhiza in acquiring P (Smith and Read, 2008). In addition, all three species are able to sustain very high N:P ratios (Von Oheimb et al., 2010), probably leading to the observed effects in heathland herbivorous macrofauna by Vogels et al. (2017), when herbivore diet becomes more and more restricted to these dominant species. Adding phosphorus, as we showed here, does indeed increase the number of smaller herbivores in the soil fauna, confirming the results found in the correlative study of Vogels et al. (2017). These smaller micro-arthropods are thought to react first and our results show that they do so within a period of three growing seasons after application. Effects in fungivores were not observed in the timeframe of the study; probably the added P was still in the live organic matter (efficiently recycled in the perennial plants). Moreover, fungivores feed on the main supplier of phosphate to the vascular plants and hence will be the last to suffer from shortages in P. Nevertheless, the observed P limitation of the soil fauna was probably caused by continuous depletion of phosphate in our heathland systems, and maybe in forests on the same soil types as well, creates the dominance of a litter increasingly difficult to decompose. At the same time it is known that litter quality and P soil content is the best predictor of decomposition rate (Aponte et al., 2012). For restoration purposes, we need to calculate the loss of P due to increased weathering and leaching as a result of increased N deposition and acidification, in order to restore the nutrient balance in soil before decomposition and productivity in the system comes to a halt. In doing so shock effects must be prevented, thus applications must be in repeated small doses or in a slow-release system, having a timing of years.

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## Appendix A. Supplementary data

Supplementary data related to this article can be found at <http://dx.doi.org/10.1016/j.soilbio.2018.01.025>.

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