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ANTS IN VESTERSKOVEN

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DANSK RESUMÉ

Myrer er ekstremt vigtige for skovens økosystem, da de bl.a. bidrager til et sundt jordmiljø og til frøspredning. Fire skovenge blev undersøgt for myrer i juli-august 2022 ved brug af pit fall traps samt manuel eftersøgning. I alt blev otte arter fundet (dog kan det være tvivl om artsbestemmelsen af to af disse, da det kan være meget svært at skelne de forskellige arter i gruppen Myrmica fra hinanden), og i alt 565 individer blev artsbestemt. De forskellige skovenge havde stor forskel i hvilke myrerarter, der var til stede samt antallet af individer.

Resultaterne fra myreundersøgelsen viser, at nogle af skovengene allerede understøtter en divers myrerfauna, hvilket indikerer et sundt jordmiljø. Andre skovenge vurderes til også at have et potentiale for dette, når de biodiversitetsfremmende tiltag bliver implementeret.

Særligt interessant er det, at myrerne er vigtige frøspredere af violslægten, som en række interessante sommerfuglearter såsom kejserkåbe og storplettet perlemorsommerfugl er afhængige af. At der er en divers myrefauna på nogle af engene, og at de to nævnte sommerfugle blev fundet i store bestande, understreger at skulle man være interesseret i at bringe f.eks. den meget sjældne rødlig perlemorsommerfugl tilbage til Vesterskoven, så er der allerede en stor violbestand til at understøtte denne art.

INTRODUCTION

Ants are important bioindicators of current soil quality. There exists no official protocol for ant species which inhabit Denmark as bioindicators for land management, making it hard to quantify soil health using ant biodiversity and abundance. Despite this, there is no doubt that having a diverse and large number of ants in a specific area indicates that the soil is healthy (Anderson, 2002). In Australia, the use of terrestrial invertebrates, namely ants, as an indicator of many taxa's colonisation has been pioneered. There, it was noticed, and proven after extensive studies, that the pattern of colonisation of ants was closely correlated to other animals' (Anderson, 2004). No causal relationship has been established and Australia's environment is very different to that of Denmark, but it is not farfetched to imagine a similar relationship considering the benefits ants provide to the soil and surrounding flora.

Healthy soil has an increased capacity to function as a vital living system, to maintain or enhance the quality of air and water environments, and, most importantly, to sustain plant and animal productivity (Alkorta, 2003). Ants present in the temperate region of Denmark promote soil health due to their many interactions with their environment. Many ants create biopores which facilitate water drainage, aeration, and root growth (Lobry de Bruyn, 1999). Certain species' mounds contribute to the resource base of surrounding biota (Dean et al. 1997) and the nutrients found within them are redistributed after their erosion resulting in ant modified soil containing higher concentration of organic carbon, phosphate, and nitrogen. Moreover, due to the soil movement involved in nest building soil particles, nutrients and organic matter are redistributed in the area. The extent of this is dependent on the amount of burrowing involved in specific species' nest building (Hole, 1980). Not all species contribute to each of these factors equally or at all, but a diverse array of species will ensure all these benefits for the soil and maintain its health.

Besides soil health, ants can contribute to their ecosystem by dispersing seeds for many plant species, especially angiosperms who have evolved a mutualistic relationship with specific ants (Prokop et al., 2022). This is called myrmecochory and provides food for ants while providing several documented benefits for the plants in its future growth. The seeds migrate to a microsite enriched with phosphorus and nitrogen (the ants refuse pile), they can get protection from herbivores from the nearby ant colony and will likely have travelled far enough so to not compete with its parent or siblings (Passos, 2002). Negative effects for the seeds can include increased competition due to high density of seeds on the ants refuse pile.

Another mutualistic relationship between plant and ant has developed. The plants, through extra-floral nectaries, provide a sugary meal for the carnivorous ants who in turn protect it from herbivorous insects. The extra-floral nectaries have also been shown to attract whole ants' nest to the plants vicinity which can be extremely beneficial due to the nutrient rich nature of the soil surrounding ants' nests (Marazzi, 2013). Lastly, ants are prey to many invertebrates and vertebrates alike, providing a source of food for secondary consumers, hence play an important role in the complex food webs of the ecosystems they inhabit.

This study aims to get an idea of the current biodiversity of many taxonomic groups including Formicidae (ants) in Vesterskoven's meadows with the objective of later increasing it if it is thought to be needed. A baseline survey of ants will be undertaken in 4 meadows, labelled 1069, 1083 (1&2), 1088 and 1093, to establish the presence and abundance of ant species using pitfall traps and active searching. Ants that are obligate parasites are not expected to be found though they might be present as their lifestyle often confines them to their nest, making them difficult to capture.

Once the baseline is done, measures should be put in place to increase the ant biodiversity. One measure which has proven to increase small organisms' diversity within areas dominated by grass is the introduction of a few trees. It might be assumed that having additional trees in a meadow surrounded by forest is pointless, but these meadows have very different flora, fauna and often soil type to the forest. Introducing isolated trees creates microhabitats, within the meadows, which increases the heterogeneity of the ecosystems and allows ants which rely on trees in grass dominated landscapes to thrive. This is because the tree canopy can provide shelter and its trunk, its branches and dead wood can be inhabited by species. Importantly, the trees were shown to attract ants which had different modes of living hence contributing differentially to ecosystem health (Gaytan et al., 2021). In some meadows which were sampled trees were already present but increasing the diversity of trees within the meadows could also be a factor in ant biodiversity as they would provide different niches (Skarbek et al., 2019). More generally an increase in plant biodiversity would also increase ant biodiversity.

In many cases animal diversity increases plant diversity and vice versa but kick-starting this cycle can be challenging. Many approaches can be taken including introducing a body of water, dead wood, and a pile of rocks, providing new habitats for the fauna.

Proposed action for the improvement of the meadow's diversity includes a change in the management including how the grass is cut and cleared. Cutting the grass at regular times and clearing the grass soon after would reduce the nutrient content of the soil thereby opening up the top layer for more desirable non-generalist wildflowers. These play a vital role in the diversity of specialised pollinators. Another change would be to clear the topsoil from the meadows and use it to produce mounds which would have much greater sun exposure creating new niches for incoming plants and animals. The clearing of the topsoil would also expose the mineral soil, making it available to the non-generalist flowers.

METHODS

FIELD SURVEY

The study is conducted at five sites in four distinct areas of Vesterskoven (1069 “Lindeengen”, 1082 “Tsugasøen” (figure 1), 1088 “Salamandervandhullet” (figure 2), and 1093 “Rhododendronsøen”). Samples were collected during the months of July and August 2022.

Figure 1
One of the studied areas of the forest referred to as 1082.

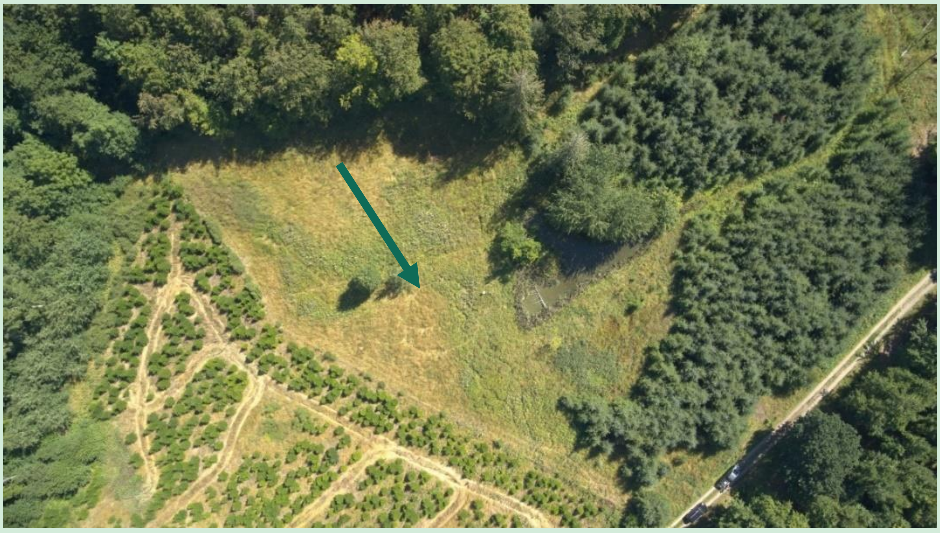


Figure 2
The site referred to as 1088 with an arrow showing tread marks where the pitfall traps were set up.

Once the sites were chosen six pitfall traps were placed, equally distanced, in a circle of 15m diameter (figure 7). Their position was chosen as to best represent the ecology of that specific area. The pitfall traps were made up of an outer cup which was placed in a hole dug for that purpose. Resting on it is a white funnel of 10cm diameter, placed flush with the ground, which leads into a smaller cup, the inner cup, which is placed within the outer cup (figure 3 and 4). The inner cup is filled with 99% ethanol with a few drops of detergent, its purpose: a killing and preservative agent. Above all this a protective plastic cover was placed, to avoid the build-up of rainwater within the inner cup, which was held up by stones or wood placed around the trap (figure 5). The outer cups were placed seven days before the experiment began to avoid digging-in effect which could affect the results (Jimenez-Carmona et al, 2019). Once the experiment started the traps were emptied thrice in 4 weeks with the ant specimens retained. About 12L of ethanol was required to complete the experiment.

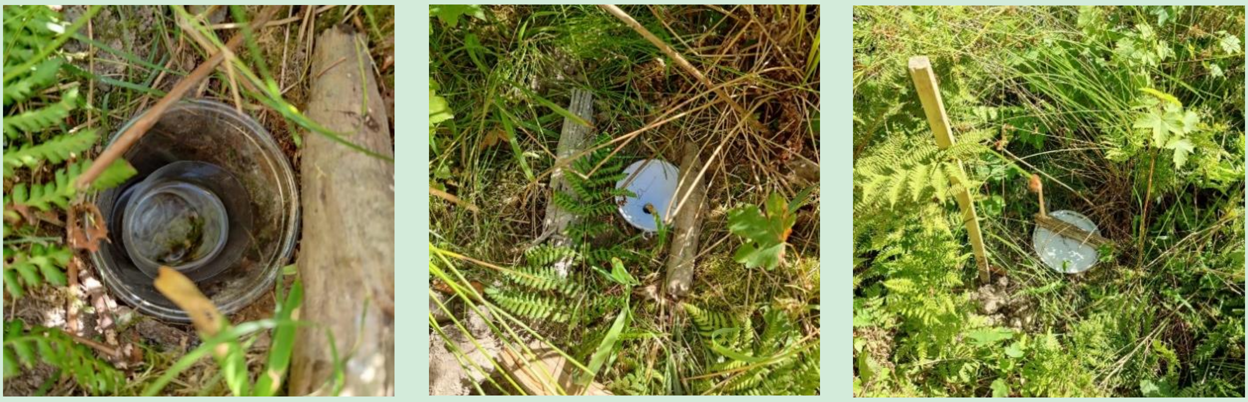


Figure 3, 4, and 5
The setup of the pitfall trap. Figure 3 shows the inside of the pitfall trap with the outer and inner cup. Six of these traps were placed in a circle of 15m in diameter in each site. Figure 4 shows the funnel flush with the ground and two sticks beside it to hold up the plate. Figure 5 shows a marker to show where the trap is placed as well as the plastic plate held down by a stick to stop rainwater entering the trap

The sites were additionally divided into 30 smaller circles of ~3m diameter equally spaced around the site where active searching took place. A rope of appropriate size was used to outline each area before searching. Within these areas all potential nest spots are investigated. Additionally, three random areas within each circle are inspected to try to identify any species which do not leave any above ground trace. In these small areas of about 15cm diameter the vegetation was removed if needed to inspect the surface of the soil. Each site is investigated for close to two hours with roughly 3-4 minutes devoted to each circle.

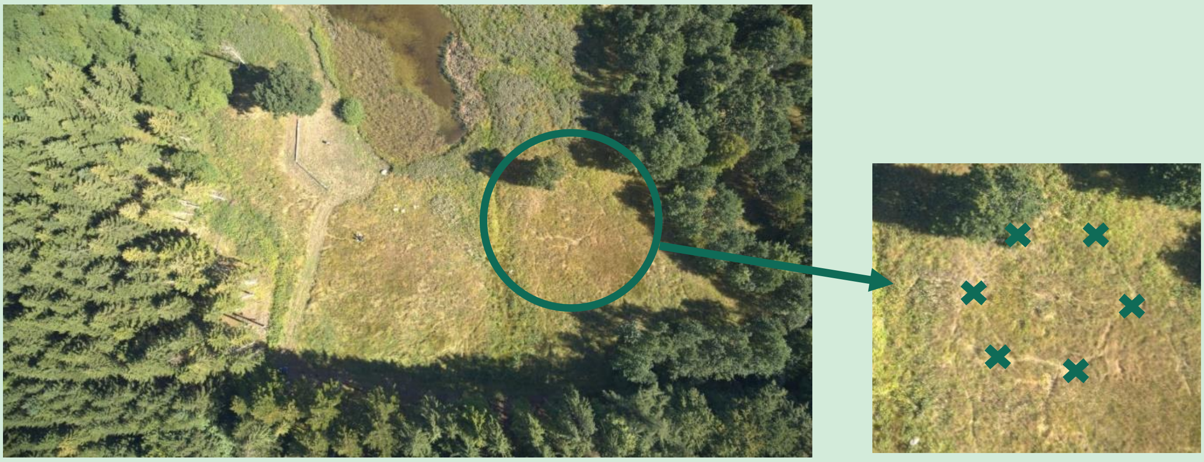


Figure 7
The pitfall trap distribution in 1082.2.

IDENTIFICATION

Once the specimens are collected, they are brought back to the laboratory where they are separated and identified using “Myrer i Danmark” (Secher et al., 2012) which was translated and simplified to end at ant genera. This simplified version used for identification is presented below. To get down to the species level “Nationalnyckeln Till Sveriges Flora Och Fauna” was used which included an English translation and pictures. A digital microscope (50X~ 1000X magnification) was use to use to examine the morphological details on the ants to aid identification.

The *Myrmica* ants were more speciose and tougher to identify, and it would not be surprising if there were more species than stated in the results. In the case of *M. rugulosa* and *M. salbuleti* these signify the species group or complex rather than the actual species due to the minor anatomical differences between species in their respective groups creating much uncertainty over the actual species. In the case of *M. rugulosa* the organisms are likely to be one of three species: *M. gallieni, M. hellenica* or *M. rugulosa.* In the case of *M. salbuleti* the ants are believed to be of either *M. specioides*, *M. lonae* or *M. salbuleti.*

1. A. Two petioles..2
   B. Gaster with clear construction and petiole....................Ponerinae, *hypoponera (punctalissima)*
   C. Petiole and gaster with no clearc
   onstriction...3

2. A. 2.5-4 mm, completely black to brown, large head and mesosoma, petiole with clear longitudinal stripes, smooth gaster (smaller than head), antennas with 10 segments..*tetramonium (caespitum)*
   B. 2.8-3.4 mm, reddish to yellow brown, whole body smooth and shiny, downward tooth like structure on post-petiole...*Formicoxenus (ntidulus)*
   C. 2.3-4 mm, slim, light and reddish brown often with a darker gaster, three outer segments larger than rest og antennae...*Leptothorax* or *Temnothorax*
   D. 4-6.5 mm, robust and amber with a darker gaster, three outer segments on antennae shorter than rest of antennae..*Myrmica*
   E. No teeth on mandibles..*Harpagoxenus*

3. A. 3-5 mm, small species (expect *Lasius sooty* 6-6.5 mm), yellowish, grey, or black, same color all over, small or indistinct oceilli...*Lasius*
   B. Small, 3.2-4.5 mm, two-colored with dark brown gaster and reddish brown mesosoma and head (pale)...*Lasius brunneus*
   C. Large species, 4.5-9 mm, can be black, grey, reddish, often multicolored, several species with dome shaped tufts of different material...*Formica*
   D. Large, very robust species, 6-14 mm, head large and black with black gaster (sometimes reddish)...*Camponotus*

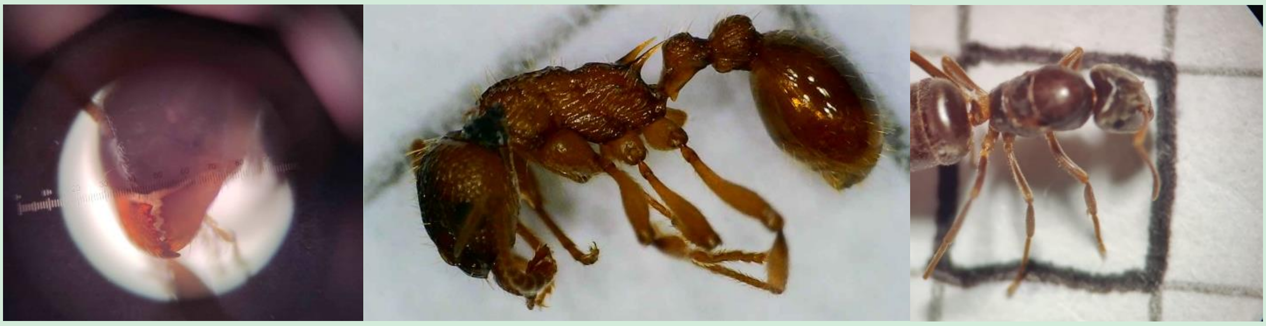


Figure 8, 9, and 10
Some morphological details used for identification. Figure 8 shows the mandibles of a nanitic *Lasius niger*. Figure 9 shows the full body of an individual of *Myrmica pugulosa* complex, and figure 10 shows a *Lasius umbratus* queen with a 5x5mm square for scale.

RESULTS

Table 1 – The distribution of the five species (or species groups) found during active search of the different plots.

Plot	Genera			Total
	Lasius	*Myrmica*	*Formica*	
1069	*niger* – 27	*rugulosa* – 1	/	28
1082.1	*niger* – 15	*rubra* – 1 *ruginodis* – 1 *rugulosa* – 1	/	18
1082.2	*niger* – 9	/	/	9
1088	*niger* – 17	*ruginodis* – 1 *rugulosa* – 6	*fusca* – 4	28
1093	*niger* – 1	*rubra* - 6	/	7

Table 2 – The breakdown of the ants found in the pitfall traps over the course of four weeks.

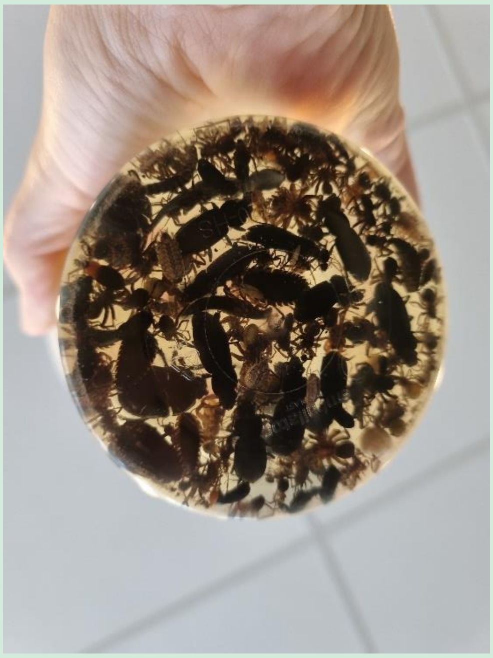
Plot	Species	Total per species	Collection			Total individuals
			1	2	3	
1069	*Lasius niger*	11	/	11	/	27
	Formica fusca	6	1	/	5	
	Myrmica ruginodis	5	2	3	/	
	Myrmica rugulosa	5	2	3	/	
1082.1	*Lasius niger*	18	2	13	3	30
	Formica fusca	10	6	4	/	
	Myrmica ruginodis	1	1	/	/	
	Myrmica rugulosa	1	1	/	/	
1082.2	*Lasius niger*	60	18	20	22	88
	Formica fusca	22	9	3	10	
1088	*Lasius niger*	182	63	70	49	367
	Lasius umbratus	1	/	/	1	
	Formica fusca	65	24	21	20	
	Myrmica rubra	2	2	/	/	
	Myrmica ruginodis	38	21	10	7	
	Myrmica rugulosa	44	14	20	14	
	Myrmica salbuleti	22	8	10	4	
	Myrmica scabrinodis	15	4	8	3	
1093	*Lasius niger*	2	1	/	1	53
	Formica fusca	1	/	1	/	
	Myrmica rubra	44	7	18	19	
	Myrmica rugulosa	4	/	4	/	
	Myrmica scabrinodis	2	/	/	2	

By-catch from the pitfall traps has not been sorted and identified at this stage, but clearly the samples include individuals from a number of different taxonomic groups (figure 11).

The by-catch samples will be stored with the aim of sorting and identifying them in the future.

Figure 11
By-catch from one of the pitfall traps including:

- Beetles (Coleoptera): Ground beetles (Carabidae), Carrion beetles (Silphidae), Rove beetles (Staphylinidae), Earth-boring dung beetles (Geotrupidae)
- Flies (Diptera): Brachycera, Mosquitoes (Culicomorpha)
- Myriapoda: Millipedes (Diplopoda), Centipedes (Chilopoda)
- Arachnids (Arachnida): Spiders (Araneae)
- Crustaceans (Crustacea): Woodlice (Oniscidea)
- Molluscs (Mollusca): Snails (Gastropoda)
- Reptiles (reptilia): Lizards (Lacertidae)



DISCUSSION

While my competence in ant identification is not developed enough to have 100% certainty in all species of Formicidae, I am certain of their genera. The ants of genera *Lasius* and *Formica* were more easily identified to species level whereas certain *Myrmica* specimens required a lot of time and even then, left uncertainty. This said, there is no doubt that there are significant differences in the ant composition of different fields which differ both in diversity and abundance of ants. For example, area 1088 shows many ants from at least 8 different species whereas area 1082.2 showed a small number of species and individuals. Various factors will play a part in these differences but whatever the reasons a meadow with high ant diversity is one which can support a richer ecosystem. This is expected to be reflected in the baseline surveys of other taxonomic groups.

Unfortunately, pitfall traps and active search, though good methods for catching certain species, are not effective for species which seldom forage above ground. This might have led to certain species and even genera going unnoticed throughout the study. One of these is *Lasius flavus,* a species of ant, common in such environments, but not found in this study. Commonly known as the yellow meadow ant, it has a subterranean mode of living and relies on mutualistic interactions with root aphids for nutrition (Depa, 2011). Their mounds can often be found in grasslands and are known to impact the soil composition adding to the heterogeneity of the environment hence are important for the habitat (Ehrle, 2019). Their presence is unknown in Vesterskoven though this is something worth investigating in the future.

Lasius niger was by far the most common ant found in every field except 1093, which is not surprising as it is one of the most common species of ant in Northern Europe and is often found in forests, their borders, and open meadows. Nesting habits for this species are varied having been found in dead wood, constructed earth mounds or under stones (Wilson, 1955). They feed mostly on honeydew from aphids which they tend to, guard and house over winter but also scavenge for dead insects and plant seeds. In their attempt to obtain nutrition from these seeds, they are inadvertently excellent seed dispersers. They retrieve seeds, bring them back to their nests where they remove the eliaosomes for consumption and discard the seed in their refuse pile by the entrance of their nest (Servigne, 2008) giving the plant seeds the aforementioned advantages for future growth. Plant species which are known to benefit from interactions with *Lasius niger* include *Chelidonium majus*, *Viola odorata*, *Galium*, and *Ranunculus.* Many species of the genus Viola, such as *V. reichenbachiana, V. odorata* and *V. hirta* rely on myrmecochory for seed dispersal. This interaction is particularly important as Viola plants are larval hosts for many species of Lepidoptera. Some of these butterfly and moth species, such as the high brown fritillary, are endangered and have declining populations (Searle, 2022) making it even more important to promote their diversity.

In the meadow 1093, the *Lasius niger* found were all nanitic. This would suggest that, while they are currently not very common in that area, one or several new colonies are establishing themselves as nanitic ants are often a sign of a colony's first brood.

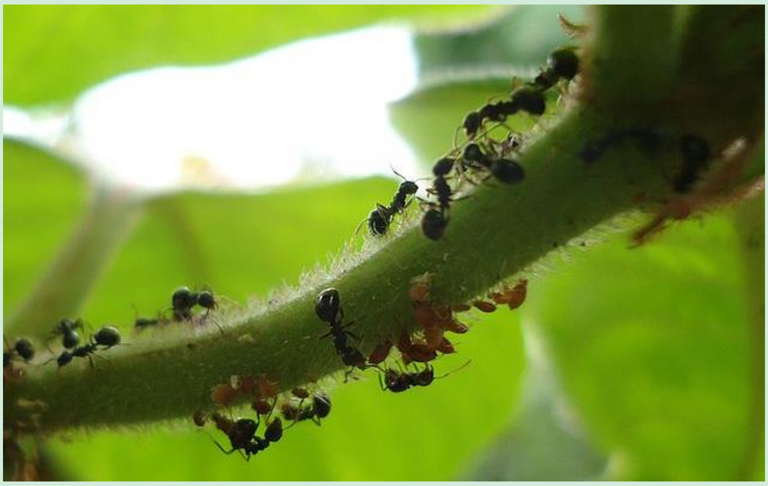


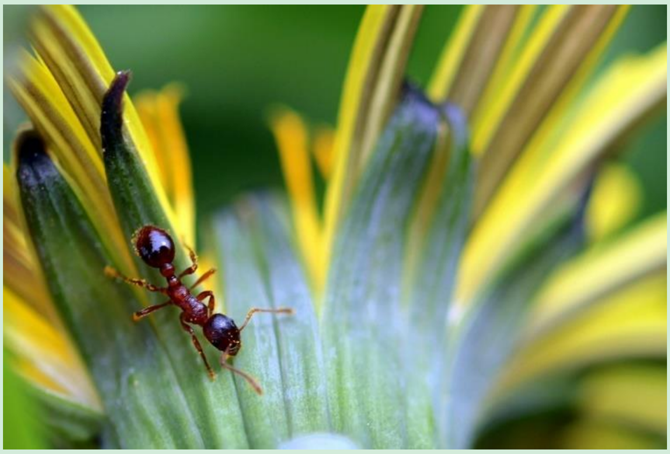
Figure 12
Formica fusca (tending aphids in the photo) was found in pitfall traps in all study sites as well as during active search.

They are not the only species which are involved in myrmecochory which is a common practice for many ants. *Formica fusca* was another species found in the meadows which disperses seeds. It is a species that often inhabits woodlands and mid-succession stage old fields where it usually builds its nest in dead wood. They are described by Zhu (2018) as “the major seed dispersers of the forest understory” and are known to interact in this mutualistic fashion with many species. They also predate on small insects, feed on extra floral nectaries and tend to aphids and consume their honeydew. As previously described, ants attracted by sugary rewards predate on small insects which benefits the plants, now protected from herbivores.

Myrmica is a genus which seldom builds mounds (Elmes, 1982) but, like so many other Formicidae found, are seed dispersers. *Myrmica rubra* and *ruginodis* appear in several literary papers on the subject (Prokop, 2022) (Bologna, 2015). It was the most speciose group in the forest meadows, but its individuals were not evenly distributed and rarely found in large numbers. Their low numbers may be due to the presence of the aggressive *Lasius niger* which would explain why there is a high number of *Myrmica* in the only meadow with few *Lasius niger* (1093).

Myrmica species are some of the most parasitised in the Formicidae family, which is saying a lot seeing as there are over 10,000 species which parasitise ants. *Maculinea* are a group of obligate parasitic butterflies which specialise in different species of *Myrmica*. Their life cycle requires a stage within an ant colony, this can be to eat larvae or to be fed by the ants directly. The European species of *Maculinea* are threatened and several are on the European Red List of Butterflies (Tartally, 2011). Due to their complex life cycle, Maculinea distribution is patchy, but an increase in *Myrmica* density and diversity in Vesterskoven would certainly increase their, and so many other *Myrmica* parasites’ chances of getting a foothold in the forest.

Figure 13
The common red and (*Myrmica rubra*) was found in multiple pit fall traps, especially in site 1093.



Finally, one individual queen of the species *Lasius umbratus* was found. It is a parasite of several *Lasius* species, in this case its likely host is *Lasius niger*. Queens, having found their host, rarely venture above ground so the individual found is believed to be searching for a host colony. *Lasius umbratus* queens cannot start their own colony without workers so it approaches small colonies, which attack it but struggle to kill it, so to acquire the colony's scent. Once it has acquired the scent, it enters the nest and kills the current queen. It will then slowly take over the colony by laying eggs of its own species using resources from the parasitised *Lasius niger*. This new colony has a different lifestyle of tending to aphids underground. It is rarely observed above ground and the literature on this species is limited.

CONCLUSION

The different meadows showed very different compositions of ants with some having few or no ants which specialise in the environment we collected them in. Present in all meadows was the generalist *Lasius niger* and the often forest dwelling *Formica fusca*. The 1093 meadow was the only one dominated by a grassland specialist: *Myrmica rubra*; and 1088 was the only meadow with a healthy array of many ants. There is evidence that these areas can support a diverse community of ants and hopefully new measures will, especially in the less speciose meadows, increase the abundance and diversity of ants which will help support the entire ecosystem.

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