

# **Enhancing Biodiversity on Industrial Land**

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## Abstract

The significant decline in flower-visiting invertebrate populations across Europe has been largely attributed to habitat loss and agricultural intensification. To facilitate and enhance population growth, reversal of biodiversity loss within the local landscapes should be addressed. This study identifies and assesses habitat features associated with flower-visiting invertebrates with the aim to develop management targets to enhance biodiversity. The abundance and diversity of flower-visiting invertebrates were compared between six habitat areas within the grounds of the Alcan Aluminium smelter. The majority of invertebrates were attracted to high flower density, rather than a particular habitat area. Underpinning the project, this study identified three priority habitats utilised by invertebrates; Grassland, Woodland, and Hedgerows.

Within grassland, the impact of mowing regimes on flower-visiting invertebrates and flowering plants was assessed. A reduction in mowing frequency resulted in a significant increase of both flower density and diversity within the sward. Comparisons to local flower-rich grasslands showed that following appropriate management the grassland at the Alcan smelter was comparable, if not better in some cases in terms of flower density, along with increasing stability of the plant and invertebrate network.

Steps to establish a diverse understorey within the woodland began with an assessment of the seedbank. Results closely mimicked the above ground flora, confirming a low abundance and diversity of flowering species. Germination trials followed to determine the suitability of current light levels to support woodland flowering plants; findings indicated the need for canopy management to enhance the woodland. Hedgerows are an important landscape feature for invertebrates, this study showed that species-rich hedgerows hosted a greater diversity of invertebrates than species poor hedgerows. Indicating the importance of hedgerow management, and the potential benefits of gap filling with native shrub species.

Finally, the presence of suitable nesting habitat is another important consideration when managing habitats for invertebrates. Here we assessed the use of three different substrates for suitability in building artificial nests (aimed primarily at bees and wasps). The uptake of these nests increased with exposure length, suggesting that artificial nests do indeed support breeding invertebrates, and could be successfully used as a tool for invertebrate conservation in the future.



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# Chapter 1 Introduction

Enhancing biodiversity is a goal of many conservationists and ecologists worldwide (Archibald et al. 2011, Kleijn et al. 2011, Turnhout et al. 2013), however, biodiversity loss as a result of human activity is undeniable and far outweighs any remedial work which is currently being undertaken (Myers et al. 2000). Over the past 20 years, a greater understanding of how biodiversity loss affects ecosystem functioning and services has been achieved (Cardinale et al. 2012), emphasising the importance of restoring and enhancing biodiversity; not just to retain and restore habitats worldwide, but also to support the expanding human population (Chopra et al. 2005). To slow or even reverse biodiversity loss actions are required on a large landscape scale. In 1992 the Convention for Biological Diversity (CBD) adopted the first global treaty to provide a legal framework to protect biodiversity, ensuring that contracted parties create and enforce national strategies and action plans to conserve, protect and enhance biological diversity. The success of national schemes, is dependent on providing connectivity on a local scale, and creating networks between fragments of high ecological value habitat and areas of low ecological value such as agricultural, industrial and urban habitats (McShane et al. 2011, Tscharntke et al. 2012), and providing a network for native flora and fauna to prevent extinction (Fahrig 1997).

## 1.1 Biodiversity

Putting an economic value on biodiversity is one method of understanding its global importance, however it is essential to first establish an understanding of the terms, threats, importance and implications of changes in biodiversity for ecosystems and mankind.

### 1.1.1 *What is biodiversity and why conserve it?*

The term biodiversity is frequently used, almost daily with regards to global environmental issues; although the word is often interpreted differently between different fields of study. The definition states that biodiversity is the degree of variation between living organisms, and is an indicator of environmental health (Gaston 2009), yet there are many expressions of biodiversity; including genetic, taxonomic, and ecosystemic (Noss 1990) and an understanding of all three are essential to maintain biological diversity

On a global scale, biodiversity provides mankind with many of the essential services that sustain life, therefore protecting and preserving species and habitats, improves the quality of life and standard of living of human populations. Providing protection to the diversity of plant species within watersheds can provide clean water to cities (Woodward and Wui 2001), and it has also been suggested that over 70% of developing nations worldwide depend directly on the biodiversity within their immediate surroundings for survival (Higman 2004). It has also been shown that areas with a higher biodiversity adapt more effectively to changes in environmental conditions (Cardinale et al. 2012). Critically, biodiversity is the basis of most ecosystem services including water cycling, water purification, natural flood defences, nutrient breakdown and pollination (Balvanera et al. 2006, Fischer et al. 2006, Cardinale et al. 2012), therefore biodiversity loss can lead to ecosystem breakdowns such as erosion, disease outbreaks, flooding and food shortages (Chopra et al. 2005), integrating the value of biodiversity into human life.

The Millennium Ecosystem Assessment (MA) was initiated in 2001, to assess the consequences of ecosystem change for human well-being, and provide scientific backing for action needed to enhance the conservation and sustainable use of ecosystems. The assessment provided a scientific appraisal of the condition and trends in the world's ecosystems, highlighting that 60% of a group of 24 ecosystem services are being degraded. As ecosystem changes are increasing, the likelihood of nonlinear changes in ecosystems changes such as disease emergence, abrupt alterations in water quality, the collapse of fisheries, and shifts in regional climate are increasingly likely. Furthermore, the MA highlighted that information within a local and national context is limited, particularly with regard to the economic value of non-marketed service.

Factors such as climate change, pollution, nutrient loading and CO<sub>2</sub> loading can cause an ecosystem to breakdown, facilitating the migration of species, and a reduction in the natural products such as food, timber, fuel, textiles, medicine and fresh water available (Chopra et al. 2005). The implementation of biodiversity conservation at an international level provides protection to locally important areas and aims to preserve, restore and reconnect them to prevent further decline (Chapin III et al. 2000, Balmford et al. 2005).



### **1.1.2 Threats to and causes of biodiversity loss**

Threats to biodiversity are many worldwide (Butchart et al. 2010) but habitat loss and fragmentation from urban and agricultural expansion are considered to be amongst the major causes of decline within Europe (Fahrig 2003, Di Giulio et al. 2009, Krauss et al. 2010, Breeze et al. 2012, Breeze et al. 2014). Further factors are considered on a global scale (Table 1.1). The underlying factor driving landscape changes is the exponential growth of the human population, currently exceeding 7 billion people (Chapin III et al. 2000, Cardinale et al. 2012). Although the human lifespan is constrained, the advances in medical science, ensures continued population growth, resulting in the need for urban expansion, agricultural intensification, and further exploitation of ecosystems for food and construction purposes (Pinstrup-Andersen and Pandya-Lorch 1998, Di Giulio et al. 2009).

Continual clearance of natural vegetation for agriculture, timber, housing and industry has resulted in significant habitat loss globally (Saunders et al. 1991, Lambin et al. 2003). Many specialised species and habitats have been lost through these processes, which has severely impacted habitat diversity (Boakes et al. 2010).

Increasing urbanisation has highlighted the importance of biodiversity conservation within towns and cities (Ingo 2011, Shwartz et al. 2014). Internationally landscapes have become dominated by man-made infrastructure which is continually increasing in size, encompassing remaining capsules of green space. As a result, city growth has impacted on biodiversity and species colonisation because of three main factors. Firstly; pollution, not just chemical, but noise and light have also had a significant impact on species diversity within urban areas (Rebele 1994, Connor et al. 2002). Secondly; habitat connectivity can be interrupted by infrastructure such as business parks, and housing developments (McIntyre 2000), and finally the introduction of invasive species into parks and gardens (McKinney 2006), can all have a negative effect on biodiversity value.

Although many species of flora and fauna are adapting and becoming integrated to urban landscapes (Matteson et al. 2008, Matteson and Langellotto 2010), the human population is expected to increase further, increasing the percentage of inhabitants within cities. This increase in urbanisation could further impact ecosystem services (Di Giulio et al. 2009), increasing the justification for a legal biodiversity framework, and the involvement of ecologists within town planning to ensure that the environment is

considered and mitigated against during construction to prevent further degradation (Bolund and Hunhammar 1999).

The other considerable cause of habitat fragmentation is agricultural expansion (McLaughlin and Mineau 1995). Agriculture is essential worldwide for the production of food to trade, and creates many employment opportunities within local communities (Rosegrant and Cline 2003). As a result, arable crops and pasture land are prominent landscape features worldwide. Historically, farming systems improved biodiversity within the UK enhancing botanical structural diversity for invertebrates and providing landscape connectivity by reducing forest dominated landscapes and converting them to arable fields, and traditional grazing pasture, with hedgerows and ditches as boundary markers (Pollard et al. 1974). During the 20th century however, farming was intensified, during both the first and second world wars arable farming was increased to alleviate the consequence of food import shortages (Robinson and Sutherland 2002, Firbank et al. 2008). Post war intensification continued, this combined with the introduction of the Common Agricultural Policy (CAP) during the 1960's, saw an increase in field size, the frequency of monocrops within the landscape, mechanisation and the use of inorganic fertilisers and pesticides to boost crop successes and reduce workforce and labour costs (Gliessman 2015). Consequently, natural habitats such as chalk and wet grassland, and traditional hay-meadows were no longer managed in a traditional way resulting in a rapid decline of wildflower and invertebrate diversity (Tscharntke et al. 2005, Henle et al. 2008, Potts et al. 2010).

Parcels of land unsuitable for access by machinery, or dense woodland remained, although these remaining parcels were fragmented, resulting in population isolation, as a result of reduced connectivity (Robinson and Sutherland 2002). (Franklin et al. 2002). Known as habitat fragmentation, at its extreme remaining parcels of habitat become too small to support viable populations, and too far apart for organisms to move between (Fahrig 2003). Populations of both *Bombus sylvarum*, and *B. distinguendus* for example, have been affected by habitat fragmentation, despite both species being mobile invertebrates, their specialist habitat requirements have resulted in a few individuals being found within a small habitat parcels with low genetic diversity.

### **1.1.3 Legislation and species protection**

Biodiversity loss could be considered inevitable, but the International Union for Conservation of Nature and Natural Resources (IUCN) continues to argue that whether it is through enacting laws and policies, implementing species recovery programmes,

establishing protected areas or restoring ecosystems, conserving nature does work (Vié et al. 2009). The reversal of decline as demonstrated by the reclassification of the black footed Ferret (*Mustela nigripes*) from extinct to endangered in 2008 (Jachowski and Lockhart 2009), and the population growth of the Iberian lynx (*Lynx pardinus*) following intervention in Spain (Palomares et al. 2011), show that counteracting biodiversity loss can be achieved. However, the amount of work, effort and money which is required to identify, monitor, and restore important habitats and individual species is extremely high, and conservation priorities need to be established, over an extended period of time for any such success. The integration of projects, research and providing information to local communities through outreach and education is vital for success.

### **1.1.3.1 Legislation within the UK**

As a response to the increasing concern regarding biodiversity loss, the government implemented methods of monitoring and controlling species loss and habitat degradation to prevent further decline. The prominent piece of legislation relating to nature conservation in the UK is the 'Wildlife and Countryside Act' of 1981 (as amended), supplemented by the Countryside and Rights of Way (CRoW) Act 2000 and the Natural Environment and Rural Communities Act 2006 (in England and Wales), and the Nature Conservation (Scotland) Act 2004 (in Scotland). This act consolidates and amends previous national legislation to protect wild animals, plants and habitats to implement and work in conjunction with the European Union (EU) 'Convention on the Conservation of European Wildlife and Natural Habitats' (Bern Convention) and 'Council Directive 79/409/EEC' on the conservation of wild birds (Birds Directive) in Great Britain. Statutory designations such as 'Sites of Special Scientific Interest' (SSSI), 'Special Protection Areas' (SPA), 'Special Areas of Conservation' (SAC), and 'Local Nature Reserves' (LNR) for the rare or declining habitats or species they support have been established through this legislation, however many habitats considered to be of lower scientific value as result of their size or ecological value, such as brownfield sites, grass verges and hedgerows could still be valuable ecological resources throughout the landscape, and could be vital for reaching biodiversity targets , but these areas not directly protected by legislation and under threat from development.

**Table 1.1:** Summary of additional factors which are considered causes of biodiversity loss worldwide

Factor	Problem	Examples	References
Invasive alien species	<ul style="list-style-type: none"> <li>• Introduced species of flora and fauna can have devastating effects on local species populations and entire ecosystems. A result of climate change, tourism, and global trade, the number of non-native species successfully inhabiting natural habitats, parks and gardens worldwide is increasing.</li> <li>• Alien species can also impact agriculture, forestry, and fisheries highlighting economic concerns in addition to the environment.</li> </ul>	Harlequin ladybird Japanese knotweed Himalayan balsam Goldenrod	(Gerber et al. 2008) (Majerus et al. 2006) (Usher et al. 1986, Perrins et al. 1993) (Moroń et al. 2009)
Pollution	<ul style="list-style-type: none"> <li>• Pollution has significantly reduced both terrestrial and aquatic species diversity. Any chemical in the wrong place or at the wrong concentration can be considered a pollutant.</li> <li>• Transport, industry, construction, extraction, power generation and agroforestry all contribute to air, land and water pollution, either directly affecting species, or causing a chemical imbalance in the environment.</li> </ul>	Heavy metals Carbon emissions Road run off	(Kandeler et al. 1996) (Lee and Power 2013) (Hsu et al. 2006), (Forman and Alexander 1998) (Siikamäki and Newbold 2012)
Climate change	<ul style="list-style-type: none"> <li>• Now widely accepted as a cause of global decline in species number, diversity, range and distribution.</li> <li>• Caused by emissions of greenhouse gases from fossil fuels, amongst other factors. Small effects recorded within the UK, however globally species ranges, population sizes, timing of biological effects (e.g. flowering), and sea level have been affected.</li> <li>• A temperature rise of 2-4 degrees in summer, milder winter, changes in rainfall and more temperature extremes has been recorded within the UK</li> </ul>	Climate becoming more unpredictable affecting species distribution  Increase in natural disasters e.g. flooding destroying vulnerable habitats	(Moss et al. 2010) (Memmott et al. 2007, Memmott et al. 2010) (Thomas et al. 2004a) (Sala et al. 2000) (Díaz et al. 2006) (Mooney et al. 2009)
Over exploitation	<ul style="list-style-type: none"> <li>• Over exploitation of species by humans causes massive destruction to natural ecosystems. Exploitation of biodiversity occurs for food, construction, industrial products, and the pet and botanical trade.</li> <li>• Over exploitation is not always direct it can be a result of pest control or by-catch of non-target organisms.</li> </ul>	Fisheries Forestry / Timber Traditional medicine Pet Trade	(Worm et al. 2009) (Hilborn et al. 1995) (Lange 2002) (Alves and Rosa 2007) (O'Brien et al. 2003)

To address the increasing threat to biodiversity from invasive alien species EU Regulation No 1143/2014 on the prevention and management of the introduction and spread of invasive alien species was introduced in October 2014. This regulation provides a crucial step to counteract threats posed by invasive species and increase the chance of achieving biodiversity targets set by the CBD and EU 2020 biodiversity targets. The regulation should provide EU countries with an effective system to prevent the introduction and colonisation of species that can cause adverse impacts on the environment, the economy, and human health (Sundseth 2014).

#### **1.1.3.1.1 Biodiversity Action Plans and Biodiversity 2020**

Over the last twenty years the importance of biodiversity has been highlighted. The rapid introduction of Biodiversity Action plans in 1994 set to reduce the impact of biodiversity loss to the United Kingdom, highlighted concern regarding recognition of valuable habitats and aimed at identifying the best management and methods to promote biodiversity. The UK Biodiversity Action Plan (UK BAP) was the UK Government's response to the Convention on Biological Diversity (CBD), which the UK signed up to in 1992 in Rio de Janeiro. The UK was the first country to produce a national BAP listing biological resources, and detailing plans for conservation of these resources.

These documents highlighted and supported the most threatened species and habitats by creating specific action plans to aid recovery, and slow degradation. National reports, produced at three to five year intervals, showed how the UK BAP was contributing to the UK's progress towards the reduction of biodiversity loss called for by the CBD.

Following the publication of the new 'UK Post-2010 Biodiversity Framework', the government's response to the new strategic plan of the CBD, the UK BAP partnership no longer operates, however, much of the information originally generated under the UK BAP still remains of use. Biodiversity 2020 is England's contribution to the framework, set up with the aim to;

"Halt overall biodiversity loss, support healthy well-functioning ecosystems and establish coherent ecological networks, with more and better places for nature for the benefit of wildlife and people."

Covering both terrestrial and marine environments throughout England, the strategy contains key outcomes, with specific actions aimed to protect and enhance

biodiversity. The outcomes encompass priority habitats and ecosystem services, marine habitats, species diversity, and people. Across England, 'Local Nature Partnerships' are working with local Government, conservation sector, and business communities to enhance the environment locally to achieve the outcomes of Biodiversity 2020.

#### **1.1.3.1.2 Local Biodiversity Action Plan (LBAP)**

Working at a local level is the optimum method of achieving the goals of Biodiversity 2020, with 119 local biodiversity action partnerships and nine regional biodiversity fora having already been established across England at the time of writing (November 2015). Aiming to classify and identify areas of local priority for biodiversity and conservation, these plans allows local business and communities to assess the value and management of their private land or community gardens, in addition to charities such as the wildlife trust working on a larger scale. LBAPs allow the translation of national targets into effective actions at a local and regional level, identify local species or habitats which are significant within the immediate surrounds and valuing the habitat on a national scale.

#### **1.1.3.1.3 Agri- Environmental Schemes (AES)**

Farmers and land managers are responsible for managing a large percentage of England's natural environment. Through the implementation of AES, land owners can continue to produce crops, but also support local wildlife through enhancements. AES are voluntary agreements implemented through CAP which pay farmers to farm in a more environmental friendly manner, and have the potential to revert farmland back to a time when it was a haven for wildlife (Natural England 2009). The schemes were introduced in 1987, with 'Environmentally Sensitive Areas', the first AES in the UK. As the scheme has progressed and evolved it has incorporated a number of management options to support wild pollinator and farm wildlife populations. The schemes support over 58,000 AES agreements to date, covering over 6 million hectares, which appear to be having a positive effect on habitat maintenance, reducing damage to local diversity (Natural England 2009) The schemes facilitates and funds the addition of flower rich field margins to enhance pollinator and parasitoid communities, winter bird seed mixes, tussocky grass margins, along with the reinstatement of diverse hedgerows, and assists in safeguarding areas of countryside and improving habitat connectivity for farmland species (Butler et al. 2007).

Following the CAP reform 2014, current AES schemes are focused towards supporting the National Pollinator Strategy 2014 a government collaboration to protect the future of pollinating invertebrates. Results of habitat creation are variable nationwide, nevertheless successes of AES have been observed (Potts et al. 2006, Carvell et al. 2007, Nisbet et al. 2010), particularly within species rich grassland restoration using green hay from existing sites (Vickery et al. 2004).

## **1.2 Flower-visiting Invertebrates**

Once overlooked by the general public, increased awareness of pollinator decline has engaged local communities and researchers to work together to establish the causes of decline within their local area. Using methods such as citizen science, where the general public are encouraged to record species visiting their garden, data can now be gathered on a national scale at relative low costs, improving our overall knowledge on population dynamics and changes. Despite the observed declines in invertebrate diversity, there are almost 30,000 species of invertebrate recorded within Great Britain, easily outnumbering the number of vertebrate and plant species recorded (Kirby 1992). However, in comparison invertebrates have received relatively little attention with regards to conservation issues and an underestimation of their decline is likely (Thomas et al. 2004b). Evidence of the severity of invertebrate declines, particularly within pollinating species were first highlighted by Biesmeijer et al. (2006), triggering further research into the causes and consequences of flower-visiting invertebrate declines (Goulson et al. 2008b, Potts et al. 2010, Breeze et al. 2011), and the implementation of the STEP (Status and Trends of European Pollinators) project initiated in 2010 to document further trends in the decline, interpret the traits and produce a Red list of European Pollinator species (Potts et al. 2011).

With over half the identified invertebrate species having some dependence on flowers, pollen and nectar within their annual lifecycle, these flower-visiting invertebrates have seen one of the largest population declines. Bees, butterflies and hoverflies, have all suffered declines which have correlated with agricultural intensification, overzealous pesticide use, and urban expansion (Goulson et al. 2005, Potts et al. 2010). The resulting impact this decline could have on the landscape and economy is staggering, in Britain alone it is estimated that the economic value of crop pollination by invertebrates is £510m per year, and would cost approximately £1.8bn annually to replace with alternative methods (Breeze et al. 2012).

### **1.2.1 Essential Functions of Flower-visiting Invertebrates – Pollination**

The greatest consequence from a reduction in flower-visiting invertebrates is the decrease in pollination services (Potts et al. 2011, Vanbergen et al. 2014). Defined as the transfer of pollen from the anthers on one plant to the stigma on the same or another plant, insects play a vital role. Bees in particular are important for pollination, as their larvae feed on pollen the adults have collected. Therefore, each adult bee maintains a high work rate, visiting many flowering plants. The process of pollination and the interactions between flowering plants and flower-visiting invertebrates are of great ecological and conservation importance. Their reliance on each other for reproduction and survival dictates that a reduction in flower-visiting invertebrates results in a significant reduction in crop success (Steffan-Dewenter and Schiele 2008). At the global level, 75% of primary crop species and 35% of crop production rely on some level of animal pollination (Klein et al. 2007) emphasising the severity of abundance declines (Moroñ et al. 2008), and the resulting effect to the economy from a reduction of exports such as cereals, fruits and vegetables.

Pollination also plays an important role in maintaining biodiversity at local and global scales (Bastolla et al. 2009). The number of species involved in the process is what makes the flower-visiting invertebrates so valuable for managed ecosystems, such as agriculture, natural plant populations and habitat longevity (Kremen et al. 2007). Aside from the ethical considerations of the importance of preserving pollinator diversity, from a purely economic standpoint, it is critical that our native wild bee populations are conserved (Stubbs et al. 1997).

### **1.2.2 Loss of flower-visiting invertebrate diversity**

All flower-visiting invertebrates are affected by multiple environmental pressures (Table 1.1), however, the greatest cause of decline has been attributed to habitat fragmentation and inappropriate pesticide use (Potts et al. 2010). Availability of suitable habitat is paramount for the survival of flower-visiting invertebrates (Westrich 1996), and, within the UK, agricultural expansion has dominated the landscape with monotypic crop fields, replacing valuable species rich grasslands, and nesting habitats (Tilman 1999, Walker et al. 2004). Although Oil seed rape (*Brassica napus*) provides an abundant pollen source for short tongued bees during spring (Knight et al. 2009), the relatively short flowering period is no benefit to late emerging long tongued bee species for example (Goulson et al. 2005, Williams et al. 2007). Furthermore, Oil seed



rape has also been argued by some as not being nutrient rich enough to support foraging adults in general (Memmott et al. 2010). In addition, the sheer density of flowers available at one time can also affect the pollination of wildflowers in the wider landscape (Holzschuh et al. 2011). Although the introduction of novel nectar rich wild flower margins are attractive to invertebrates (Pywell et al. 2005, Pywell et al. 2006, Pywell et al. 2011a), and can support species which forage in to late Autumn (Pywell et al. 2006, Carvell et al. 2007), the need for shelter, and breeding sites also needs to be addressed.

#### **1.2.2.1 Pesticide use**

The overuse and inappropriate use of pesticides within the agricultural environment is another major cause of pollinator decline (Goulson et al. 2005, Potts et al. 2010). A link has been identified between neonicotinoids and a reduction in brain function within foraging *Apis mellifera* and *Bombus sp* (Gill et al. 2012, Henry et al. 2012). Specifically, Henry et al. (2012) found that a sub-lethal exposure to thiamethoxam (a specific Neonicotinoid) significantly increased homing failure in foraging *A. mellifera*, and Gill and Raine (2014) recorded a significant difference in forage success between control and pesticide exposed *B. terrestris*, and a decrease in pollinator efficacy which combined resulted in an increase in hive mortality (Gill and Raine 2014). As a response to the increasing body of evidence the European Environment Agency and the European Commission supported a ban of the use of three types of neonicotinoid on crops attractive to pollinating insects (McGrath 2014). During July 2015 this ban was temporarily lifted under the caveat that some farmers should be able to use the insecticide under EU rules for “emergency” use, therefore allowing the use of banned chemicals to protect crops, however research into the extent of damage to invertebrate populations vis the use of systemic pesticides is ongoing (van Lexmond et al. 2015).

#### **1.2.2.2 Increased urbanisation and habitat displacement**

Flower-visiting invertebrates have complex life cycles, reliant on a variety of habitat types; one of the primary reasons why increased urbanisation and agricultural intensification has had such a negative effect on local invertebrate populations. This loss of connectivity has caused a shift where industrial and brownfield sites which support low nutrient soils, and cycles of disturbance and abandonment have allowed the colonisation of a flower-rich botanical community, intermixed with areas of bare ground, spoil and water, providing a mosaic of optimal conditions to support foraging, basking, and nesting habitat for invertebrates away from their natural range. Although

increased urbanisation can displace insects from their natural environments, urban areas such as brownfield sites, domestic gardens and parks are increasingly important to support local invertebrate populations. Bumblebee nests have been reported to have a superior survival rate and higher density occurrence within gardens compared to both agriculture and woodland habitats (Osborne et al. 2008, Goulson et al. 2010) and, a greater species richness of bee species have been recorded in urban areas compared to nature reserves and farmland (Baldock et al. 2015). Although increased plant richness may be one of the guiding factors, modern gardens are moving away from traditional flower beds and vegetable crops, with the introduction of decking and paving, to minimise labour and save time in the garden. Therefore wildlife areas within gardens are important for invertebrates, and it is well documented that the presence of invertebrate species depends greatly on the foraging and nesting resources available (Westrich 1996). Although some urban gardens could be considered unattractive to insects, with the increased concern for wildlife throughout the UK, local authorities are encouraging wildlife areas within public parks and gardens, improving the availability of suitable habitat for invertebrates (Blackmore and Goulson 2014). Goulson et al. (2002) found that sub-urban habitats had a positive effect on bumblebee numbers, likely a result of the mosaic of habitats, and variety of forage and refuge available. Urban gardens, could never replace the importance of native wildflower meadows for invertebrates and a few flowering plants in your garden is not going to reverse the ongoing plant and pollinator decline, but availability of forage and nesting options within an ever expanding habitat type could be essential for invertebrate survival.

Urban habitats are completely different when compared to an invertebrate's natural habitat; characterised by high fragmentation and frequency of disturbance, in addition to processes such as construction, ground tilling, and foliage modifications are all factors which could inhibit invertebrate colonisation (Matteson et al. 2008). Furthermore, non-native flowering plants can dominate urban environments impacting on the availability of nest sites for locally important species (Frankie et al. 2005). The degree of mowing, pruning, weeding and clearing leaf litter can also reduce invertebrate visitation (Helden and Leather 2004, Gaston et al. 2005), therefore managing gardens to benefit invertebrates might involve allowing the accumulation of detritus structure in the form of leaf litter, which has been shown to greatly increase natural enemy abundance (Langellotto and Denno 2004).

### **1.2.2.3 Alien invertebrate species**

Non-native invertebrate species can colonise either as a result of an introduction via human interaction, or a natural progression as a result of global climate change (Vitousek et al. 1996). Alien species can forage on many native plants without effect, in some cases improving pollination success, however, they can be found to displace native pollinators in addition to transferring disease, and facilitating pollination of exotic weeds (Goulson 2010, Hulme et al. 2013). Bumblebee species such as *Bombus hypnorum*, first recorded in the UK in 2001 have colonised and spread rapidly across the UK from Europe (Goulson and Williams 2001). Although they are not native they have become residents in the UK, and do not appear to have an adverse effect on native bee populations. Conversely, the Harlequin Ladybird (*Harmonia axyridis*), frequently used as a biological control entered the U.K in 2004 colonising and spreading rapidly (Majerus et al. 2006). The population is currently outcompeting native ladybirds, causing devastating effects to native populations (Vilcinskis et al. 2013).

Furthermore, some invertebrate species are introduced to enhance crop pollination, with growers becoming increasingly reliant on placing commercially reared, and in some cases imported, bee hives of *Apis mellifera* and *Bombus sp* within glasshouses and polytunnels to boost pollinator numbers (Otterstatter and Thomson 2008, Fürst et al. 2014). However, despite their positive role in pollination, escaped commercial sub species such as *Bombus terrestris dalmatinus* could threaten endemic sub species, such as *Bombus terrestris audax*, by transferring pathogens and parasites (Fürst et al. 2014, Graystock et al. 2014), or through hybridisation and heightened competition (Ings et al. 2006).

### **1.2.2.4 Alien plant species**

Evidence of pollinating invertebrates utilising alien plant species as an additional forage source within their food networks has been recorded (Stout and Morales 2009). Alien plant species that form large dense populations could attract native invertebrates disrupting the pollination of native plants, as the reward in pollen and nectar is greater (Dietzsch et al. 2011). However, this may not have a large-scale effect, as species adapted for generalised foraging are more likely to feed on alien plants than specialised species (Lopezaraiza-Mikel et al. 2007, Kleijn and Raemakers 2008). Therefore, although alien plants can increase in local abundance outcompeting native plant

assemblages (Vila and Weiner 2004), Hulme et al. (2013) suggest that the potential negative effects of these plants are less than some studies have previously suggested.

Alien plant species can be contained within gardens, garden escapes colonising bare ground, or some crop species within agriculture. However, if a population does become large and the impact of the continued spread highlights concern, then conservation programs may need to remove the alien species to allow colonisation of native species. The removal of Himalayan Balsam (*Impatiens glandulifera*) from riverbanks however, can have a serious impact on invertebrates, small mammals and birds. Although the plant is non-native, it grows large and dense providing a good amount of shelter, and the flowers are heavily utilised by native bee populations. Therefore, the implications for native bees of any such removal scheme could be serious, and it has been suggested by many, that an alternative should be planted in its place if removed on a large scale (Fussell and Corbet 1992, Zavaleta et al. 2001, Moragues and Traveset 2005)

Gardens host a range of introduced flowering species for ornamental purposes. These plants are either imported or hybridised to emphasise their flower size, symmetry, corolla colour and flowering longevity, all of which can be responsible for attracting flower-visiting invertebrates (Miller et al. 2011). However, although some introduced flowers can be large and attractive to invertebrates, they are not necessarily producing the quality and quantity of nectar and pollen required as a food resource for bees (Comba et al. 1999). Therefore, although there is evidence for the benefits of alien species within gardens, these features may only benefit more generalised, broad feeders (Morales and Traveset 2009).

### **1.2.3 Importance of diversity within plants and invertebrates**

The decline in pollinator diversity can bring with it a significant decline in pollination of wild plants, 80% of wild plant species are reliant on invertebrate pollination for fruit and seed sets (Ashman 2004). Although flower-visiting invertebrates can have broad feeding strategies; adapting when a favoured plant is unavailable, there are some species of insects such as *B. sylvarum* and *B. distinguendus* that have specific habitat requirements. Even though bumblebees are extremely mobile, they can be restricted to feeding on just one or two species of plant. In both cases (*B. sylvarum* and *B. distinguendus*), this has resulted in low, volatile populations (Ellis et al. 2006, Charman et al. 2010). Furthermore, some invertebrate species have particularly complex life

cycles requiring different habitats for different life stages; what a larva requires can sometimes vary greatly from what the adult insect requires to thrive (Kirby 1992).

Invertebrates can identify a preferred plant species by subtle differences in flower morphology. Identifiers such as flower size, age, sex or symmetry, can allow an invertebrate to choose the most rewarding flowering plants. Some species such as bumblebees can determine the success of forage from ephemeral odours left on petals by previous visitors (Goulson 1999). When flowers are scarce, theory predicts that foragers will abandon specialisation in favour of generalisation (Goulson 1999). Despite invertebrates preferring constancy, within controlled experiments both honeybees, and hoverflies have been recorded to abandon constancy when presented with decreasing availability of their preferred flower *Brassica kaber* (Kunin 1993).

Different genera of invertebrates have adapted to pollinate different flowering plants, therefore when thinking of enhancing pollinator diversity it is important to consider the species you wish to encourage, the current local populations, the climate and natural fauna of your target area. Bumblebees in particular (except *B.terrestris*, the most polylectic) carefully select nectar sources, although overlaps between broad foraging species and their emergence times exist (Lye et al. 2009). Invertebrates are also heavily influenced by resources such as breeding sites, foraging habitat or overwintering sites on a landscape level, and consideration of the surrounding habitat, and the possibility of source-sink dynamics and neighbouring effects should be considered during works to enhance and monitor habitat suitability for invertebrate pollinators (Rusch et al 2010, Dunning et al 2009). The presence of any one resource within a habitat patch is complemented by the presence of another type of resource nearby in a different habitat patch, making it possible to enhance and support larger populations. Within the source-sink model an area of high quality habitat can sustain, and increase total population size particularly when a small amount of sink habitat is located within landscape, because excess invertebrates can disperse and reproduce within nearby sink patches. However, when sink habitat increases available source patches are not always discovered by dispersing individuals, affecting the population growth. These factors highlight the importance of habitat connectivity and working on a landscape level to establish management regimes for flower-visiting invertebrates (Dunning et al 2009).

### **1.3 Aims and Objectives**

As a response to the significant decline in flower-visiting invertebrates and the resulting impact on food security and sustaining natural habitats, research in to pollinating species, has increased over the past decade (Goulson et al. 2008b, Potts et al. 2011, Berg et al. 2013), investigating aspects of pollinator ecology, ecological enhancement and invertebrate behaviour, however the focus has largely been attributed to butterfly, honeybee and bumblebee species within agricultural environments. The effects of habitat management on flower-visiting invertebrates as a functioning group is relatively understudied, furthermore the assessment of habitats incorporated within an active industrial site, constrained by industry, intensive management and frequent disturbance and provides a fresh approach to enhancing connectivity and biodiversity on a landscape scale.

This thesis is novel in its approach, aiming to investigate and consider the benefits of habitat parcels within an area of industrial land. The effects of small management changes from the current landscape management plan will be monitoring by observing changes in botanical and flower-visiting invertebrate abundance over a three-year period. The data collected will provide essential information to guide the future management of the site to maintain a favourable ecological status, but also provide evidence based management suggestions to inform future landscaping and ecological management plans for new industrial units, and guidance material for planning urban infrastructure across the United Kingdom. Furthermore, results could be used to inform positive changes to methodology currently used to manage public open space by local authorities and private land owners further enhancing biodiversity on a wider scale.

Standard survey methods were used to collect data on flower density, and flower-visiting invertebrate abundance and diversity within the eight habitat parcels on site to establish a baseline dataset to allow further determination of the ecological value of the industrial site (Chapter 2). Specifically, predicting that i) Habitat type / site would have a significant effect on the abundance and diversity of flower-visiting invertebrates recorded, ii) Month surveyed will have a significant effect on the diversity and abundance of flower-visiting invertebrates observed, iii) Flower density will have a significant positive effect on flower-visiting invertebrate assemblages, and iv) Flowering plant diversity will have a significant positive effect on flower-visiting invertebrate assemblages.

The ecological value and current management of grassland is reviewed within Chapter 3, aiming to improve the botanical diversity of the grassland sward on site, by assessing the effect of cutting regimes and cutting removal to improve the abundance of flower-visiting invertebrates and increase flower density. We predict that, i) Removing grass trimmings from experimental plots will result in a higher plant species diversity and flower density compared to plots where grass trimmings have been left insitu, ii) Plots subjected to two cuts (2M, an early and a late cut) over the flowering period will support a significantly higher herb cover and great suppression of grasses compared to plots mown frequently (0M) and subjected to one cut (1M), iii) Flower density will significant effect the abundance and richness of flower visiting invertebrate on site, and iv) Plots subjected to one cut following flowering (1M) will support a significantly higher flower-visiting invertebrate diversity compared to plots mown twice a year (2M) and frequent cutting (0M).

Within Chapter 4, we aim to determine the success of grassland management at the Alcan smelter by undertaking assessments of flower density and invertebrate abundance on comparable habitat which have been managed by natural regeneration or under a seeding regime. This study aims to compares the response of flower-visiting invertebrates to determine the success of the management changes undertaken and use species interaction networks to determine flowering species of importance within the local area. Three hypotheses were investigated further i) There will be a significant relationship between the different plant assemblages on site on the invertebrate communities they attract, ii) Flower visiting invertebrate abundance and diversity will be significantly higher at nature reserve grasslands and sites managed with wildflower mix, and iii) Flower density and flowering plant diversity will be significantly higher at nature reserve grasslands and sites managed with wildflower mix, and iv) Ecological network analysis will highlight flowering plant species of importance to guide future management strategies.

Chapter 5 aims to determine a management strategy for the plantation woodland on site, by establishing an evidence based report, by assessing the underlying quality of the seedbank, to determine appropriate management techniques. A further investigation into canopy cover and germination success of a woodland understory mix, aims to find the ideal conditions to base a management scheme upon. Two hypotheses were formulated i) Seed diversity will be significantly higher in samples

collected from 10-15cm depth compared to the surface layer (0-5cm), and ii) light levels representative of the ride habitat will significantly increase germination rate and species diversity of a shaded woodland mix within a replicated trial.

Chapter 6 evaluates the value of two hedgerow types for flower-visiting invertebrates by assessing the invertebrate communities inhabiting, species rich and species poor hedgerows to consider their value as connective habitat for this species. Specifically, we predicted that, i) species rich hedgerows would support a significantly higher abundance of flower-visiting invertebrates than species poor hedgerows, ii) species rich hedgerows would support significantly higher species diversity of flower-visiting invertebrates compared to species poor hedgerows and iii) species rich hedgerows would host a significantly higher abundance of nectar feeding species and pollen collecting invertebrate guilds, than predator and parasitic guilds.

Finally, with suitable habitat loss being an influential factor in the continued decline of invertebrate populations, the effectiveness and uptake of artificial nests was investigated within Chapter 7. The trial aimed to determine the success of artificial traps to attract invertebrates. Specifically, predicting that i) the number of occupied stems within each nest will significantly increase with exposure time, ii) there will be no significant differences between the three substrate types utilised, and iii) there will be a significant difference between species and recorded stem diameter.



## **1.4 Introduction to the study site and its history**

### **1.4.1 Study Area -Alcan aluminium smelter, Lynemouth, Northumberland, UK**

Located on the Northumberland coastline (55.2016° N, 1.5396° W), the Alcan smelter covers 82.7 hectares supporting a number of large aluminium production buildings, and offices subject to a high level of noise pollution and heavy traffic. Established in the 1970's the smelter has been responsible for the production of aluminium and its distribution worldwide, however during 2014 the smelter ceased production, and is currently under decommission.

Although the intensity of production on site presents itself as unfavourable to local biodiversity, the smelter is surrounded by an area of buffer land supporting plantation woodland, grassland, and areas of scrub and hedgerows (Figure 1.1), providing approximately 21 hectares of land favourable for supporting a number of taxa including birds, mammals and invertebrates. The site is bordered by arable crop production and grazing pasture which dominates the wider landscape, however within 2km of the site boundary there are a number of designated sites for nature conservation; one RAMSAR site, four Sites of Special Scientific Interest (SSSI), and two Local Nature Reserves (LNR). The designations are largely attributed to the habitat suitability for breeding and overwintering waders and wildfowl, but other designations include unimproved neutral grassland and ancient woodland.

#### **1.4.1.1 Current habitat management**

At present, the site has been managed to achieve a pleasant working environment, with the planting of low maintenance shrub borders, scattered trees within amenity grassland and intensively managed grassland dominating the site. The buffer areas of trees and scrub have been left unmanaged for long periods of time reducing their ecological value, but provide refuge for a number of taxa. A survey of principle habitat types (Phase one Map, Figure 1.1) undertaken during 2011 indicated a number of areas where management could be implemented to improve biodiversity for invertebrates. Although the site is dominated by infrastructure, the location of the smelter lends itself to improvement, providing a pocket of habitat within an agriculturally dominated landscape and area valuable to wildlife.



**Figure 1.1:** Phase one habitat survey of Alcan smelter and the local landscape. The wider Ariel photography shows the close proximity of the site to the LNR (bottom Left) and SSSI / RAMSAR coastline (Top right). The site was dominated by A1.1.1 (Broad leaved semi natural Woodland), A1.1.2 (Broadleaved plantation woodland), B.5 (Marshy wet grassland), C3.1 (Tall ruderal herbs) and J1.1 (Arable fields)

## Chapter 2

### **Flower density more important than habitat type for increasing flower- visiting invertebrate diversity**

#### **2.1 Abstract**

Declines in flora and fauna are well documented and highlight the need to manage available habitats to benefit local biodiversity. Between May and September in 2011 the number, composition and diversity of flower-visiting invertebrates was assessed across eight sites, representing a range of habitats within an industrial site in the North East of England, UK. There was no significant difference in insect assemblages between the sites selected, but there was a significant difference between the months surveyed. Flower density was highlighted as the most important factor driving these changes between months and indicates that flower density is more important to a site for invertebrate diversity than the presence of specific habitats. Analysis of the invertebrate communities each month allowed comparison of dominant invertebrates to the flower density data, highlighting sites where management intervention could be initiated to benefit insect diversity, or alternatively specific management plans to encourage target species. Furthermore, this study highlights the importance of correct data interpretation to answer specific management objectives, and recommends analysing the invertebrate community interactions to determine the dominant species present prior to undertaking any management of the site in question.

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## 2.2 Introduction

As a result of human influences, habitats and ecosystems are continually fragmented by factors such as city expansion, and agricultural intensification (Sala et al. 2000, Krauss et al. 2010). Areas such as gardens, parks, brownfield sites and working industrial sites are becoming important 'islands' for wildlife between the ever increasing urbanised areas. However, these sites require management to conserve biodiversity, and to provide an optimum habitat network for species (Law and Dickman 1998). Sites left unmanaged over long periods of time can become dominated by rank grasses and pernicious weeds, reducing the biodiversity and value of the habitat (Wratten et al. 2012).

Previous studies have highlighted the impacts of environmental management schemes and the effects of habitat fragmentation on biodiversity within agricultural environments (Feehan et al. 2005, Whittingham 2006, Kenward et al. 2011). Additionally, the importance of gardens and parks for pollinating species within urbanised areas are becoming more apparent (Gaston et al. 2005, Goddard et al. 2009, Cameron et al. 2012). Nevertheless, the importance of industrial areas has not yet been considered. Despite industrial sites often being heavily utilised, a large proportion have the potential to create wildlife refuges within an urban and agriculturally dominant landscape. Industrial sites are found worldwide and frequently cover a large expanse of land, usually incorporating varied unmanaged habitats.

The aluminium smelter site at Lynemouth, UK, incorporates a range of these different habitats including scrub, grassland and wetland. Although it is widely accepted that increased habitat variety often results in higher species diversity, (Kunin 1997, Tschardtke et al. 2002, Benton et al. 2003) factors such as the botanical structure and flower density within the site can be more influential on invertebrate assemblages than distinct habitat types (Potts et al. 2003, Ghazoul 2006) . This highlights further questions as to what drives these trends within such habitats. For example, how variable do habitats need to be to improve invertebrate diversity? Does habitat connectivity affect habitat quality? And importantly, if a specific management strategy was implemented at sites such as Lynemouth, would it be possible to increase diversity of invertebrate species present?

The success of any programme to enhance biodiversity is dependent on how people manage land and invest in development. Within large companies, where employees have been encouraged to take an interest in biodiversity, site action plans have

become successful. However, an important step before management strategies can be employed is to determine the value of the site for biodiversity. By assessing different habitats and understanding which species are utilising these areas, informed decisions on future land management can be made.

This study aims to assess flower-visiting invertebrate abundance and diversity as a response variable to differing flower density and plant diversity within habitat parcels at the Alcan smelter. Flower-visiting invertebrates are a functional group of invertebrates involved in ecosystem processes such as pollination, and have been subjected to severe decline (Potts 2010). Invertebrates readily adapt to changes in habitat and are frequently used as bioindicator species as a sign of environmental health making them an ideal target to highlight different habitats.

Specifically, four hypotheses were investigated further i) Site/habitat will have a significant effect on the abundance and diversity of flower-visiting invertebrates recorded, ii) Month surveyed will have a significant effect on the diversity and abundance of flower-visiting invertebrates observed, iii) Flower density will have a significant positive effect on flower-visiting invertebrate assemblages, and iv) Flowering Plant diversity will have a significant positive effect on flower-visiting invertebrate assemblages

## **2.3 Study Design**

Eight individual sites representing different habitat types were identified before the start of the study and the diversity of flower-visiting invertebrates was assessed within these sites on a monthly basis for one survey season (May-September).

### **2.3.1 Study site**

The study was conducted on land surrounding the RioTinto Alcan Aluminium smelter in Lynemouth, UK (55.2016° N, 1.5396° W). Covering 82.7 hectares the site is typical of a working industrial site, with intensively managed grassland and shrub borders around offices, access roads, car parks and production units. However, 20.7 hectares (25%) of the site is predominately scrub, woodland and wetland forming a buffer zone. To the west of the smelter, hybrid poplar trees have been mixed with native European tree species such as *Sorbus aria*, *Acer pseudoplatanus* and *Quercus robur* to create a fast growing screen to the smelter. Since this planting, the densely populated woodlands have left a bleak understory, dominated by two species of plant; the *Rubus fruticosus* and *Urtica dioica*. Grassland which has been left unmanaged is becoming

dominated by *Cirsium sp.*, and rank grasses which are restricting the growth of other species.

Invertebrate flower visitors were sampled from eight sites around the smelter over the period of May – September 2011. The selected sites represented a range of habitats which include: Flower Rich grassland (FR), Mown Grassland (MG), New Hedge (NH), Old Hedge (OH), Plantation Woodland (PI), Pond (Po), Ridge and Furrow Grassland (RF) and Woodhorn Woodland (WW). All eight sites were contiguous and covered a similar area, therefore patch size is unlikely to be influential (Table 2.1, Appendix 1, Figure A-1.1).

## **2.3.2 Invertebrate Sampling**

### **2.3.2.1 Pan Traps**

Three pan traps (17cm diameter and 6cm depth) were placed at each site, 1m apart in a triangle formation. Blue, yellow and white UV reflective plastic bowls were used to account for colour preference by certain insects (Wilson et al. 2008). These colours were used for three principal reasons; they represent a range of wavelengths found in the visual spectrum, they are similar to flower colours, and have been proven to attract a variety of flower-visiting species (Moroń et al. 2008, Westphal et al. 2008). Traps were filled to the three quarter line with water, to which several drops of unscented dishwashing detergent (Ecover-Zero) were added to reduce the surface tension. Pan traps were set approximately 0.5 m above the ground at the height of the surrounding vegetation to allow the trap to be visible to flying insects. Wooden posts with brackets and wire were used to secure the pans in place during sampling.

Traps were exposed for a period of 30 hours (traps set at 10.00 and collected at 16.00 the following day) twice a month. On collection, the specimens were transferred into glass vials, labelled and preserved in 70% ethanol. All flower-visiting species considered to frequent flower heads as part of their lifecycle, and be directly or indirectly involved with pollination were identified to family, genus, or genus and species level (where possible) using a dichotomous key. From the order Diptera, frequent flower visitors are concentrated in three main families: Syrphidae, Bombyliidae, and Tachinidae (Larson et al. 2001). Families such as Empididae and Asilidae, known to frequent flowers for predatory reasons were also collected. All other Diptera families were not included within this study.

**Table 2.1:** Description of the eight sites used within the study, highlighting the name given, location, habitat type and a description of the dominant plant species present within each site.

Site	Location (British National Grid)	Habitat area (Hectares)	Description
Flower Rich (FR)	NZ29350 BNG89685	0.55	Well drained, poor quality soil. Flowering species such as <b><i>Lotus corniculatus</i></b> , <b><i>Trifolium pratense</i></b> , and <b><i>Dactylorhiza incarnata</i></b> present
Mown Grassland (MG)	NZ29674 BNG89205	0.30	Lawn areas regularly mown and dominated by <b><i>Taraxacum officinale</i></b> , <b><i>Bellis perennis</i></b> . Backing onto long unmanaged grassland surrounded by farmland
New Hedge (NH)	NZ29741 BNG89191	0.20	Running alongside Wheat field. Consisting of <b><i>Rosa canina</i></b> , <i>Crataegus</i> sp., <b><i>Prunus spinosa</i></b> (blackthorn) and <b><i>Rubus fruticosus</i></b>
Old Hedge (OH)	NZ28918 BNG89685	0.20	Single species hedge hawthorn, bordering <b><i>Brassica napus</i></b> field, with weed species such as <b><i>Matricaria discoidea</i></b> and <b><i>Leucanthemum vulgare</i></b> in the field no flowering plants in the field margin
Plantation Woodland (PI)	NZ29135 BNG89703	0.62	<b><i>Urtica dioica</i></b> as understory plants, 2m separated poplar trees with occasional <b><i>Acer pseudoplatanus</i></b> , <b><i>Quercus robur</i></b> and <b><i>Sorbus aria</i></b>
Pond (Po)	NZ29384 BNG89278	0.55	Pond surrounded by <b><i>Vicia sativa</i></b> , <b><i>Anthriscus cerefolium</i></b> , <b><i>Oenanthe crocata</i></b> , <b><i>Rubus fruticosus</i></b> and <b><i>Centaurea nigra</i></b> (common knapweed)
Ridge and Furrow Grassland (RF)	NZ29001 BNG89671	0.60	<b><i>Heracleum sphondylium</i></b> dominated grassland with tall dominant grasses such as <b><i>Elymus repens</i></b> and <b><i>Arrhenatherum elatius</i></b> surrounded by farmland
Woodhorn Woodland (WW)	NZ29079 BNG89616	0.55	Older woodland trees including <b><i>A. pseudoplatanus</i></b> and <b><i>Q. robur</i></b> , with an understory of <b><i>Hyacinthoides non-scripta</i></b> , <b><i>Galanthus</i> sp.</b> , and <b><i>Rubus fruticosus</i></b> .

### 2.3.2.2 Observation plots

Data was collected from observation plots at each site to complement the pan trap data as it allowed the monitoring of species less represented within pan trap samples, such as Lepidoptera and Apidae (Vrdoljak and Samways 2012). Initially net collecting

along a transect was proposed, however due to the access restrictions within industrial areas and the topography of the land this method was considered inappropriate. Each observation plot measuring 1m<sup>2</sup> was surveyed twice each month during May, June, July, August and September. Observations were made from a single point for a period of five minutes. Each insect seen to enter the observation plot was recorded, if the insect began to forage, the plant host was also recorded. Bumblebees and butterflies were identified to species, while hoverflies were identified to genus where possible. Due to the similarity between workers of *B. terrestris* and *B. lucorum* these species were treated as an aggregate species, as identification is unreliable in the field (Prys-Jones and Corbet 1991). Observations were only initiated between 10:00 and 17:00 h, when weather conformed to Butterfly Monitoring Scheme standards (Pollard and Yates 1993).

### **2.3.2.3 Botanical structure and flower density**

Estimates of flower density were collected for each site twice monthly. Ten randomly placed 625 cm<sup>2</sup> quadrats were used within the sample area (area surrounding the trio of pan traps). In each quadrat, the numbers of plants and the number of flower heads per plant species were recorded. The flower density was used as a surrogate measure for nectar availability as direct measurement of nectar parameters in the field is regarded as impractical (Potts et al. 2003).

### **2.3.3 Data analysis**

Data collected from the pan traps and the observation plots was combined to create one dataset. Minitab 16 was used to complete Correlation and ANOVA (Kruskal-Wallis) analysis on the total numbers of invertebrates recorded for each habitat. Shannon Weiner Diversity Index was used to determine a diversity value for each habitat. This value was derived from species richness and relative abundance of each species and quantifies how well species are represented within a community. Diversity was then compared against month, and flower density as individual factors. The R statistical and programming environment, version 3.0.2 (R Core Team 2013) was used to perform species accumulation curves to determine sampling completeness using the 'specaccum' function within the 'Vegan' package (Oksanen et al. 2013) (Appendix 1, Figure A1.1), and the Mantel's test function within the 'ade4' package (Dray and Dufour 2007). The Mantel's test was used to determine whether the observed differences in the dependent variable (species diversity), were associated with the predictor variable (i.e. site/geographical location), or whether differences were apparent because of



spatial variation, assuming that samples which are collected within a close geographical area will be similar and samples collected over a larger distance would be different.

As a result of the variation in taxonomic groups and functions of the invertebrates collected (Appendix 1, Table A1.1), analysis has been conducted with four guilds of flower-visiting invertebrate: (1) Nectar feeding only (Syrphidae, Vespidae, Lepidoptera, Tipulidae, Scioyzidae, and Tachinidae), (2) Parasitic invertebrates (Ichneumonidae, Tenthredinidae Chalcidoidea, and Chrysididae) (3) Pollen collecting (Apidae) and (4) Predatory invertebrates (Emphididae, Asilidae, and Panorpidae). Where trends were indicated, further analysis using the detailed dataset was undertaken.

Due to the multi-species nature of the data and the survey design utilised in the study, multivariate analysis was utilised (Clarke 1993, Clarke and Warwick 2001). Invertebrate assemblages within each habitat were compared using PRIMER 6.0, a non-parametric multivariate statistical package. Multi-dimensional scaling (MDS) plots based on Bray-Curtis similarity measures were used to compare invertebrate assemblages. Bray-Curtis was utilised because the technique is known to efficiently handle ecological data supporting high numbers of species absences (Zeros). MDS plots provided a visual representation of the community data showing relationships between samples ecological communities based on variation within the species composition. Further similarity percentage (SIMPER) analysis was also run on the data matrix, SIMPER decomposes Bray-Curtis similarities between all pairs of samples to identify those species that contribute most to the differences observed (Clarke and Warwick 2001).

## **2.4 Results**

Over the period May – September 2011, a total of 1138 individual invertebrates were sampled across the four guilds, within the eight sites (Appendix 1, Table A1.). A nested Analysis of similarity (ANOSIM) of sites within month showed there was no significant difference in the abundance of each flower-visiting guild between sites ( $R=0.054$ ,  $P=0.265$ ), but there was a significant difference between month sampled ( $R=0.387$ ,  $P<0.001$ ). The same trend was observed looking at assemblage for the site ( $R=-0.063$ ,  $P=0.268$ ) and month ( $R=0.342$ ,  $P<0.001$ ). Despite the lack of significant differences between flower-visiting invertebrates between the sites, there were strong patterns noticeable within the dataset. A mantel test showed that there was no spatial

correlation between distance and invertebrate diversity within this study ( $r=0.093$ ,  $P=0.65$ ).

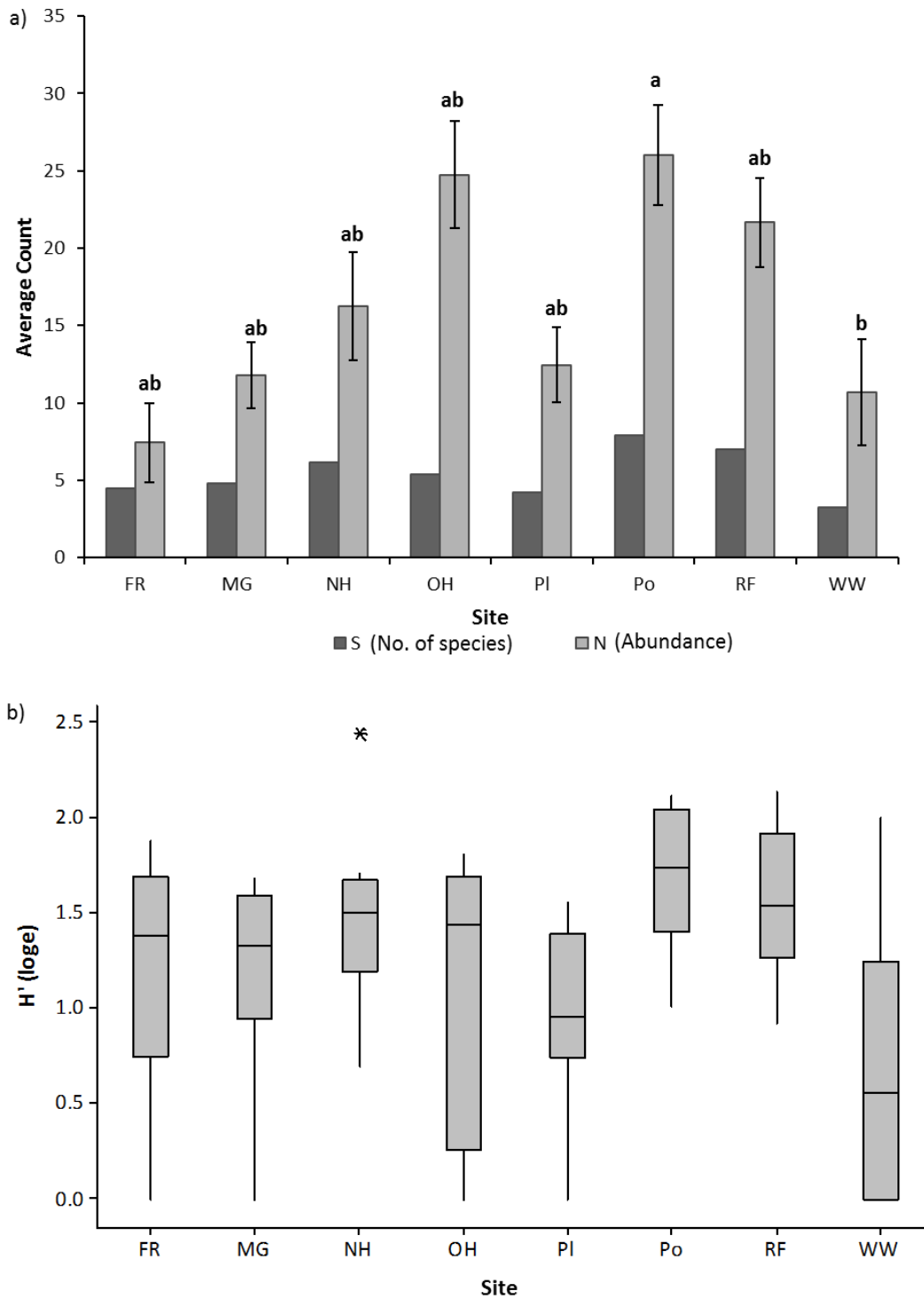
#### **2.4.1 Response of invertebrate assemblages to site**

The highest number of individuals were recorded within the Pond site ( $n = 234$ ) and the lowest within the Flower Rich Grassland ( $n = 67$ ). Few insect families recorded were site specific; most species were recorded across all sites; however, individuals from Satyridae, Panorpidae, Tipulidae and Coccinellidae were in isolated populations. Tenthredinidae occurred in all sites except the Plantation Woodland. The most abundant families recorded were the Ichneumonidae ( $n = 264$ ), and the Syrphidae ( $n = 375$ ). Within the family Syrphidae, 47% of records were from a single species *Episyrphus balteatus*, with 176 individuals recorded across the survey period. Individuals from both families were present across all sites.

Removing month as a factor, average count data showed that the Old Hedge, Pond and Ridge and Furrow Grassland, had more individuals compared to the other sites (Figure 2.1a), however the difference was non-significant (Kruskal-Wallis  $H=11.87$ ,  $P=0.105$ ). When using the number of species / families recorded for each site there was a significant difference between sites (ANOVA,  $F=2.43$ ,  $P < 0.05$ ). Tukey comparisons revealed that the differences between Pond and Woodhorn Wood were the cause of this variation ( $t=2.98$ ,  $P < 0.01$ ) (Figure 2.1a).

When looking at total diversity rather than abundance, a higher diversity of species was present in the sites; New Hedge, Pond and Ridge and Furrow (Figure 1.1b). This changes the result from the abundance data, whereby the Old Hedge was more important than the New Hedge. However, the data is highly variable between sample dates.

A significant difference in the diversity of flower-visiting invertebrates based on Shannon diversity scores was recorded across all sites (Kruskal-Wallis,  $H=17.75$ ,  $P < 0.05$ ) (Figure 2.1b). Repeated Mann Whitney tests revealed that the Pond had a significantly higher species diversity compared to Mown Grassland, Plantation and Woodhorn Wood. Woodhorn Wood had significantly lower species diversity than Mown Grassland, Pond and Ridge and Furrow (Table 2.2).



**Figure 2.1:** Response of invertebrates to site: (a) Mean Count for number of species (S) and abundance (N). Letters represent significant differences between sites following Tukey comparisons (b) Whiskered Box Plot showing the range of diversity scores. Box indicates median value, lower and upper quartiles. Whiskers indicate the range. Outliers indicated by an asterisk. (FR) Flower Rich Grassland, (MG) Mown Grassland, (NH) New Hedge, (OH) Old Hedge, (PI) Plantation Woodland, (Po) Pond, (RF) Ridge and Furrow Grassland and (WW) Woodhorn Woodland.

**Table 2.2:** Comparison of Mann Whitney Statistical test p-value results for Shannon diversity score between sites. Significant differences are highlighted in bold, and differences in significance threshold are highlighted by a different letter. (FR) Flower Rich Grassland, (MG) Mown Grassland, (NH) New Hedge, (OH) Old Hedge, (PI) Plantation Woodland, (Po) Pond, (RF) Ridge and Furrow Grassland and (WW) Woodhorn Woodland.

Site	FR	MG	NH	OH	PI	Po	RF	WW
FR		1a	0.665a	0.961a	0.269a	0.077a	0.289a	0.12a
MG			0.736a	0.923a	0.145a	<b>0.038b</b>	0.289a	0.0835a
NH				0.713a	0.075a	0.229a	0.665a	<b>0.0234b</b>
OH					0.267a	0.0829a	0.36a	0.2233a
PI						<b>0.0047c</b>	<b>0.0171b</b>	0.23a
Po							0.536a	<b>0.0046c</b>
RF								<b>0.0103b</b>
WW								

Bray Curtis Similarity analysis highlighted similarities between sites driven by the guilds (Figure 2.2a). Parasitic insects were found in high numbers in all sites; however, they did not occur in all replicate samples (Table 2.3 & 2.4; Figure 2.2a & 2.2b). There was a higher dominance of parasitic insects recorded within Plantation and Woodhorn Woodland indicating the importance of a woodland environment for this guild. The families Ichneumonidae and Tenthredinidae behave in a similar manner with records across all sites, however in contrast to Ichneumonidae, Tenthredinidae were underrepresented within the woodland sites (Table 2.3).

The pollen collecting, nectar feeding and predatory insect guilds were also present throughout all habitats. The pollen collecting guild saw a marked reduction in numbers within the Plantation and Old Hedgerow sites, whereas the other guilds were in higher abundance (Table 2.3). This difference appeared to be influenced by two *Bombus* species (*B. lapidarius* and *B. lucuorum/terrestris*) which were found in similar abundances throughout all sites, however larger numbers were recorded in the Flower Rich site for *B. lapidarius* and the New Hedge for *B. lucuorum/terrestris*. There was no record of *B. lucuorum/terrestris* within the Old Hedge (Table 2.5).

The nectar feeding guild was also influenced heavily by species within the family Pieridae including *Anthocharis cardamines*, *Pieris napi* and *P. brassicae* which were observed across six of the eight sites, yet were more dominant within the Pond site (Table 2.4).

**Table 2.3:** Heatmap-table summarising the relative abundance (%) of insect guild within each site, based on SIMPER analysis of count data recorded during the survey period. (FR) Flower Rich Grassland, (MG) Mown Grassland, (NH) New Hedge, (OH) Old Hedge, (PI) Plantation Woodland, (Po) Pond, (RF) Ridge and Furrow Grassland and (WW) Woodhorn Woodland.

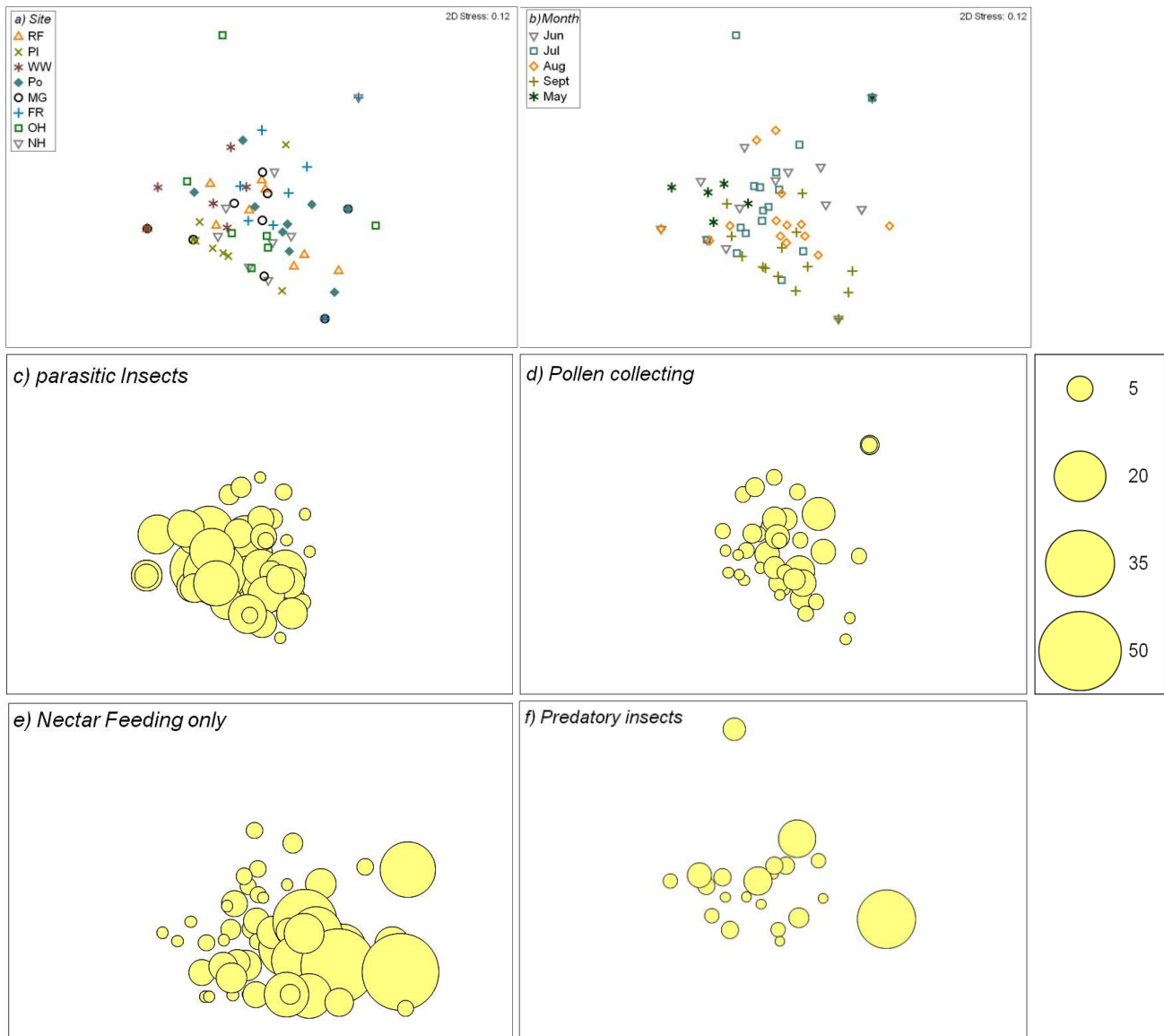
Insect Guild	FR	MG	NH	OH	PI	Po	RF	WW
Nectar Feeding only	3.86	5.27	5.57	5.26	4.53	6.15	5.23	3.2
Parasitic Insects	3.85	5.73	5.41	4.86	7.67	4.35	6.03	7.25
Pollen collecting	6.28	2.98	3.31	0.9	0.35	4.16	3.2	1.93
Predatory insects	0.41	0.77	1.06	3.93	1.21	1.2	1.05	1.7

Key %	0.00-1.00	1.01-2.00	2.01-3.00	3.01-4.00	4.01-5.00	5.01-6.00	6.01-7.00	7.01-8.00
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**Table 2.4:** Heatmap-table summarising the relative abundance (%) of dominant taxon within each site, based on SIMPER analysis of count data recorded during the survey period.

	FR	MG	NH	OH	PI	Po	RF	WW
<i>Icheumonidae</i>	2.27	4.4	3.12	3.98	6.54	2.71	3.87	7.18
<i>Tenthredinidae</i>	0.89	1.25	1.41	0.73	0	2.29	3.42	0.31
<i>Episyrphus sp</i>	0.3	1.31	1.99	3.08	1.28	2.96	2.38	1.68
<i>Chalcidoidea</i>	1.02	0.9	1.44	1.55	0.84	0.83	1.43	2.36
<i>Syrphus sp</i>	0.71	0.41	1.31	1.99	1.5	1.38	0.91	0.45
<i>Platycheirus sp</i>	0.99	0	1.01	1.6	1.1	0.41	0.92	0
<i>Bombus lapidarius</i>	4.77	1.44	0.8	0.46	0.24	1.3	1.61	0.38
<i>Bombus lucuorum / terrestris</i>	1.12	0.45	2.47	0	0.24	1.99	1.59	1.25
<i>Asilidae</i>	0.34	0.75	0.98	3.59	1.03	0.65	0.63	1.93
<i>Emphididae</i>	0	0	0.25	0.29	0.53	0.78	0.67	0
<i>Eupeodes sp</i>	0	1.03	0.87	1.69	0	0.51	0.93	0
<i>Melanostoma sp</i>	0.28	0	0	0.19	0.8	0.17	0.16	0.31
<i>Noctuidae</i>	1.24	0	0.42	0	0.37	0.33	0.6	0
<i>Coccinellidae</i>	0.74	0	0	0	0	0	0.75	0
<i>Hesperiidae</i>	0.32	0	0	0	0	0.53	0.69	0
<i>Megachilidae</i>	0.74	1.49	1.18	0.29	0	0.21	0.51	0
<i>Apis mellifera</i>	0.6	0	0	0	0	0	0.31	0.31
<i>Bombus pascuorum</i>	0.32	0.24	0	0.25	0	1.53	0	0.9
<i>Vespidae</i>	0.3	0.73	0.45	0	0.24	0.72	0	0.45
<i>Pieridae</i>	0.24	0.64	1.03	0.36	0.24	2.19	0	0
<i>Helophilus sp</i>	0.43	0.71	0.66	0	0	1.27	0.37	0
<i>Epistrophe sp</i>	0	0	0	0	0	0.57	0	0
<i>Eristalis sp</i>	1.33	0.79	0.29	0	0	0.44	0.16	0
<i>Sphaerophoria sp</i>	0	0	0.34	0	0	0	0.33	0
<i>Satyridae</i>	0	0	0.47	0	0	0	0	0
<i>Panorpidae</i>	0	0	0.42	0.34	0	0	0	0

Key %	0	0.01-1.00	1.01-2.00	2.01-3.00	3.01-4.00	4.01-5.00	5.01-6.00	6.01-7.00	7.01-8.00
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**Figure 2.2:** (a) Multi-Dimensional Scaling (MDS) plot of flower-visiting guilds at each of the eight sites based on Bray-Curtis similarity. (b) MDS plot of flower-visiting guilds at each of the five sampling months based on Bray-Curtis similarity. (c-f) Bubble plot overlaid on the MDS sample points indicating patterns driven by the abundance of each guild. These highlight the role of each guild in shaping the community, bubble size relative to the number of individuals at that point.

#### 2.4.2 Response of invertebrate assemblage to month surveyed

The numbers within each flower-visiting guild were significantly different between months (ANOSIM,  $R=0.387$ ,  $P < 0.001$ ), with the dominance of each guild affected (Figure 2.2). Furthermore, invertebrate assemblages were also significantly different between months (ANOSIM,  $R=0.342$ ,  $P=0.001$ ), with pairwise comparisons indicating that all months were significantly different to each other with regard to invertebrate assemblage except for the months of May and June (Table 2.5).

**Table 2.5:** Summary of p-value results for Pairwise comparisons of invertebrate assemblages between Months. Significant differences are highlighted in **bold**, and differences in significance threshold are highlighted by a different letter.

Month	May	June	July	Aug	Sept
May		0.086a	<b>0.014b</b>	<b>0.015b</b>	<b>0.004c</b>
June			<b>0.002c</b>	<b>0.003c</b>	<b>0.001c</b>
July				<b>0.001c</b>	<b>0.001c</b>
Aug					<b>0.006c</b>
Sept					

The Parasitic insect guild dominated throughout the entire survey season (Table 2.6 & 2.7: Figure 2.2c). Higher numbers of this guild were recorded in May, but reduced through the survey season with a marked reduction in September. Ichneumonidae and Tenthredinidae appear to be the most influential families for this trend (Table 2.7). The nectar feeding guild also showed significant seasonal changes in abundance; however, the trend was directly opposite to that of the parasitic guild, with records increasing over the survey season with a peak in September (Table 2.6). This trend was clearly influenced by late feeding Syrphidae present during this month (Table 2.7).

**Table 2.6:** Heatmap-table summarising the relative abundance (%) of dominant guilds for each month, based on SIMPER analysis of count data recorded during the survey period.

Insect Guild	May	June	July	August	September
Parasitic Insects	7.07	5.24	5.95	6.31	4.4
Pollen collecting	2.72	3.98	3.28	3.23	1.47
Nectar Feeding only	2.24	3.88	4.41	4.7	7.76
Predatory insects	2.98	1.34	1.98	0.83	0.5

Key%	0.00-1.00	1.01-2.00	2.01-3.00	3.01-4.00	4.01-5.00	5.01-6.00	6.01-7.00	7.01-8.00
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**Table 2.7:** Heatmap-table summarising the relative abundance (%) of dominant taxon for each month, based on SIMPER analysis of count data recorded during the survey period.

	May	June	July	Aug	Sept
<i>Icheumonidae</i>	6.23	4.05	4.64	4.07	2.87
<i>Bombus lapidarius</i>	2.07	1.45	1.44	1.71	1.2
<i>Asilidae</i>	2.21	1.01	1.89	0.68	0.46
<i>Bombus lucorum / terrestris</i>	0.48	2.26	1.33	0.96	0.34
<i>Tenthredinidae</i>	1.77	1.2	1.08	2.07	0.71
<i>Emphididae</i>	1.51	0.36	0.19	0.14	0.13
<i>Chalcidoidea</i>	0.48	0.99	1.19	1.51	1.53
<i>Pieridae</i>	0.93	0.49	1.16	0.35	0.3
<i>Megachilidae</i>	0.4	0.85	0.92	0.62	0
<i>Nymphalidae</i>	0.91	0.32	0.16	0	0
<i>Coccinellidae</i>	0	0.96	0	0	0
<i>Noctuidae</i>	0	0.83	0.4	0.15	0.43
<i>Melanostoma sp</i>	0.57	0	0.12	0.27	0.44
<i>Helophilus sp</i>	0	0.53	0	0.16	1.37
<i>Panorpidae</i>	0	0.41	0	0	0
<i>Vespidae</i>	0	0.32	0.46	0.38	0.47
<i>Chrysididae</i>	0	0.32	0.19	0	0
<i>Hesperiidae</i>	0	0.18	0.69	0	0
<i>Bombus pascuorum</i>	0	0.25	0.73	0.74	0
<i>Syrphus sp</i>	0	0.32	0.88	1.19	2.31
<i>Episyrphus sp</i>	0	0.32	1.74	2.73	3.08
<i>Apis mellifera</i>	0	0.35	0.34	0	0
<i>Eupeodes sp</i>	0	0.17	0.11	1.3	1.1
<i>Platycherius sp</i>	0	0.24	0.11	0.82	2.17
<i>Eristalis sp</i>	0	0.17	0	0.48	1.22

Key %	0	0.01-1.00	1.01-2.00	2.01-3.00	3.01-4.00	4.01-5.00	5.01-6.00	6.01-7.00
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### 2.4.3 Response of Invertebrate assemblages to Flower density

Mean flower density across the site ranged between 0 and 17.6 per m<sup>2</sup> over the season. Only one site had no flowering plants recorded and this was in the plantation (Figure 3a). Flowering plants were available across all other sites during July and August, yet by September half of the eight sites (NH, PI, RF and WW) had no flowering plants available to insects (Figure 2.3b). The Flower Rich site was the only one to have flowering plants available all season (Figure 2.3b), although surprisingly this site had the lowest recorded invertebrate visitation. In contrast, the pond had the largest invertebrate assemblages, but no plants available within the month of May.



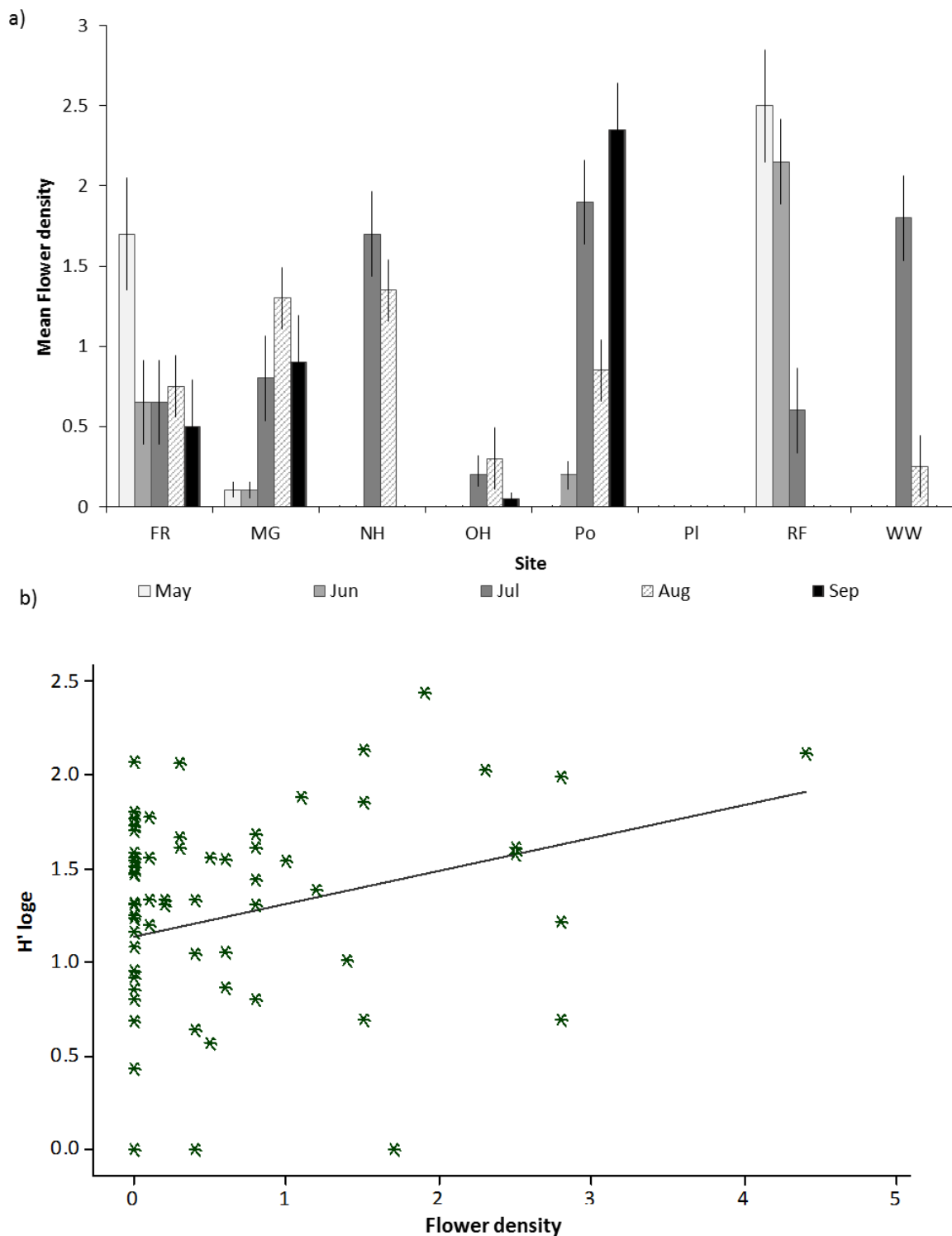
Following an ANOSIM on flower-visiting guilds, an influence of insect density was observed but not significant ( $R=0.14$ ,  $P=0.06$ ). However, when the whole data set was analysed flower density was highlighted as a significant factor with regard to invertebrate assemblage (ANOSIM,  $R=0.233$ ,  $P < 0.01$ ). Although flower density was not significantly different between months (Kruskal-Wallis,  $H = 8.68$ ,  $P = 0.070$ ), invertebrate assemblages were significantly affected by flower density when nested within month (ANOSIM,  $R=0.178$ ,  $P < 0.01$ ). Analysis of the entire data set also highlighted a significant correlation ( $\rho = 0.245$ ,  $P < 0.01$ ) between Shannon Wiener Diversity of invertebrates and flower density (Figure 2.3c).

#### 2.4.4 Response of invertebrate assemblages to Flowering plant diversity

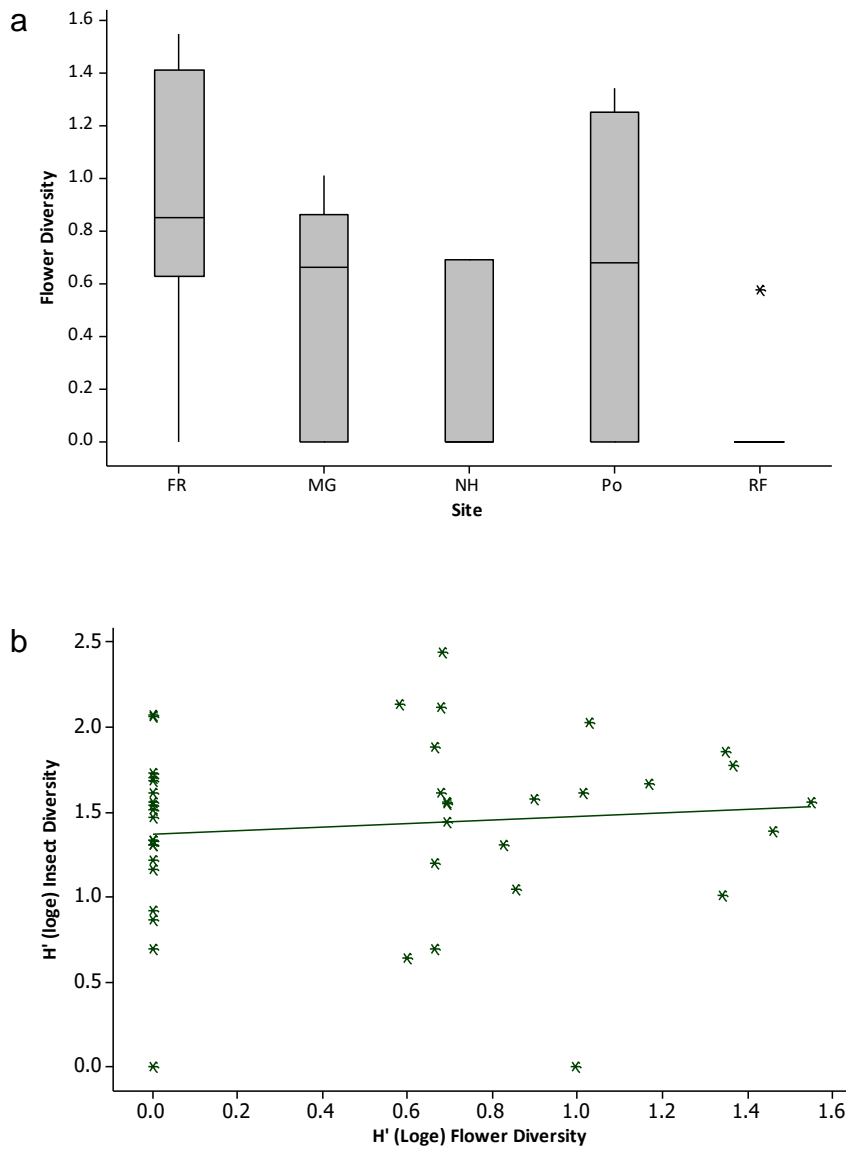
The Shannon Wiener diversity of flowering plants observed within each site was calculated. Three of the eight sites (OH, WW, and PI) had diversity scores of zero and therefore have been omitted from further analysis. The Flower Rich site had the highest recorded diversity (Figure 4), and a significant difference was observed between sites (Kruskal wallis  $H = 13.37$ ,  $P < 0.01$ ). Repeated Mann-Whitney tests highlighted which sites saw the greatest differences (Table 2.8). The Flower Rich and Ridge and Furrow Grasslands were the greatest drivers for this result. Flowering plant diversity was not identified as a significant factor for flower-visiting guild assemblage (ANOSIM,  $R=0.082$ ,  $P=0.824$ ) and there was no correlation of flower diversity with insect diversity ( $\rho = 0.162$ ,  $P = 0.298$ ).

**Table 2.8:** Comparison of Mann Whitney Statistical test p-value results for Shannon diversity score (flower diversity) between sites. Significant differences are highlighted in **bold**, and differences in significance threshold are highlighted by a different letter.

Site	FR	MG	NH	Po	RF
FR		0.1971a	<b>0.0387b</b>	0.3722a	<b>0.001d</b>
MG			0.2212a	0.585a	<b>0.012c</b>
NH				0.2736a	0.1741a
Po					<b>0.0354b</b>
RF					



**Figure 2.3:** Floral resource across the sites: (a) Mean Flower Density for each site subcategorised by month (b) Scatterplot with line of best fit ( $r^2$ ) showing the correlation between flower density and Shannon Weiner Diversity Score for insects. Flower rich (FR), Mown grassland (MG), New Hedge (NH), Old hedge (OH), Plantation (PI), Pond (Po), Ridge and Furrow (RF), and Woodhorn Woodland (WW)



**Figure 2.4:** (a) Whiskered Box Plot showing the range of diversity scores. Box indicates median value, lower and upper quartiles. Whiskers indicate the range. Outliers indicated by an asterisk. (FR) Flower Rich Grassland, (MG) Mown Grassland, (NH) New Hedge, (Po) Pond, and (RF) Ridge and Furrow Grassland (b) Scatterplot with line of best fit ( $r^2$ ) showing the correlation between Shannon Weiner Diversity Score for invertebrates and flowers.

## 2.5 Discussion

Results highlighted differences between the habitat parcels recorded across the Alcan site, and highlighted that the Pond and surrounding habitat supported a more diverse assemblage of invertebrates in terms of Shannon-Weiner diversity than other plots. Kunin (1997) highlighted that many habitat types should be incorporated within a reserve area to capture the species variation caused by habitat discontinuities, suggesting that more habitat types are generally better than one to enhance biodiversity. This study however, shows that flower-visiting invertebrates recorded at the Lynemouth Smelter site were influenced predominantly by the flower density recorded within the habitat parcels, rather than the distinct sites/habitats themselves, a finding which is widely supported (Potts et al. 2003, Ghazoul 2006, Molina-Montenegro et al. 2008). Although, site/habitat type did not significantly affect invertebrate assemblage within this study, flower density was a significant factor, suggesting that varied botanical structure and the presence of a large number of certain flowering plants are more important than the number of different types of habitats specifically.

Conversely, within this study flowering plant diversity did not have a significant impact on the invertebrate assemblage which was not expected as flower diversity is indeed an important factor in determining flower visitor presence (Potts et al. 2003, Frund et al. 2010). This result could be influenced by the low numbers of plant species recorded within the short-term trial and could identify the need for further analysis. The type of flowerhead available to insects will determine which species feed, hunt and breed within a particular habitat (Comba et al. 1999, Stang et al. 2006). Ideally, a variety of host plants for larvae and immature insects are required (Jermy et al. 1968, Lawton 1983, Haddad et al. 2001), in junction with factors such as flower longevity and nectar resource which have both previously been shown to have an effect on the invertebrate assemblages recorded within different habitats (Potts et al. 2009, Albrecht et al. 2012).

As expected, month had a significant effect on invertebrate assemblages, again complementing results seen in previous studies (McCall and Primack 1992, Brown and Schmitt 2001). Certain taxa were present at different times of the year, likely due to variation in emergence and breeding periods as a result of an insect's dependence on factors such as weather, as well as the availability of host and food plants for different invertebrate groups. Only one of the eight sites had plants flowering for the entire survey period (the Flower Rich site), however due to regular mowing of this area the

flower density was low, resulting in this area having the lowest recorded invertebrate visitation. Nevertheless, invertebrates were recorded across the whole season suggesting that this was not the only factor affecting the site's attractiveness. For example the pan traps could have been less attractive to the foraging invertebrates than the flowering plants present, resulting in the catch being proportional to the flower density as suggested by Cane et al. (2000). Additionally, despite a relatively high diversity of plant species within the Flower Rich site, it was dominated by one particular plant species, *Lotus corniculatus*. *L. corniculatus* was predominantly foraged by one particular species of pollinating invertebrate, *Bombus lapidarius*, a species known to have a preference for yellow flowers and the appropriate mouth parts to access the nectar from this plant (Comba et al. 1999). This dominance of the site by one plant species could in part explain the low invertebrate visitation rates recorded in this particular study. *L. corniculatus* may not have been an appropriate flower resource for other invertebrate species. Therefore, improving diversity of flowering plants within certain pre-existing habitats will undoubtedly have a significant effect on the pollinating species present within the sites. Alternatively, there may have been other factors such as noise pollution and forage distance to consider which were not assessed during this study.

The number and diversity of invertebrates could be influenced by improvements to the assessed sites, not only diversifying the flowering species available for forage, but also increasing diversity with regard to flower head shape. Ensuring a variety of flower heads such as umbels or composite heads will increase the number of flower-visiting invertebrates (Gilbert 1981). For example, low abundance of the hoverfly *Episyrphus balteatus* was recorded within the Flower Rich habitat, where hoverflies would normally be expected (Stubbs and Faulks 2002). The dominant flower, *L. corniculatus*, is not preferred by hoverfly species as their mouthparts do not allow access to the nectar. In contrast larger numbers of hoverflies were observed within the Ridge and Furrow Grassland, Old Hedge, and Pond sites as a result of the flowering plant species present: umbelled flowers such as *Oenanthe crocata* and *H. sphondylium* and simple flowering species such as *Crataegus monogyna*. A reduction in the mowing frequency at the Flower Rich site may diversify the flowering species present, allowing species which are less hardy to germinate from the seedbank (Gaujour et al. 2011). Furthermore, a study by López-Mariño et al. (2000) also highlights that due to the high proportion of perennial grasses often present within semi-improved habitats, only half

the species stored within a grassland's natural seedbank are present above the soil surface. This suggests that the sward can be diversified by managing the grass species present, proving a beneficial management strategy to enhance invertebrate diversity for this site.

The Pond site had the largest invertebrate assemblages, despite the limited floral resource available within the month of May. Some invertebrate species are locating nest sites around this time, particularly bumblebees which start searching around April and peak in May (Kells and Goulson 2003). Ensuring flowering plants are available throughout April to September is therefore an important management strategy, particularly for invertebrate diversity. The availability of nest and forage sites are essential to pollinating invertebrates and improvement in this area would likely have a positive effect on the overall insect diversity of this site (Westrich 1996, Winfree 2010). Conversely, there are few species of plant which flower early, therefore improvement to surrounding hedgerows, or planting of species such as *Salix cinera* and *Malus sp.* complemented with *Laminum album* and *Glechoma hederacea* could be more beneficial to the communities around the Lynemouth smelter (Pywell et al. 2011b).

Interestingly, this study highlights the importance of the method in which data sets are analysed, which could result in different interpretation of the data and therefore have a serious impact on the proposed management of a particular site. Firstly, the data was organised into guilds of insects to minimise taxonomic variation between data samples. The trends reported in this study were mirrored in both the guild analysis and complete taxon analysis showing that many of the results reported hold fast when analysing the data in different ways. However, some trends were weaker when looking at guilds, rather than when analysis was completed on the whole data set, which means significant findings could be overlooked if only one type of analysis was utilised. Furthermore, the data in this study showed that two sites, the Plantation and Woodhorn Woodland had lower mean invertebrate abundance, species richness and Shannon Weiner diversity score than all the other surveyed sites, suggesting these two sites may not be as important for the flower-visiting invertebrate communities (although these differences were not all significant). However, when comparing invertebrate assemblages these habitats were highlighted as important for parasitic insects such as Ichneumonidae. Following the initial analysis comparing mean averages of invertebrate abundance per site, the importance of these habitats could have been overlooked and the site managed inappropriately for the dominant inhabitants. Until

the community analysis was performed the dominance of Ichneumonidae and influence on the community may not have been appreciated, highlighting that community analysis is invaluable within biodiversity assessment. Secondly, with regard to Shannon Weiner diversity, a higher diversity of species was found to be present in three particular sites; the New Hedge, the Pond and the Ridge and Furrow grassland. In contrast when looking at the abundance data, the Old Hedge is a more important site for invertebrate assemblages than the New Hedge highlighted in the diversity data set.

The methods of analysis used in this study highlights the importance of data interpretation before management action plans are devised. The two variations in interpretation of the results above were both made with the same original data set, however one utilised abundance data whilst the other used total diversity. The differences are likely caused as the diversity index takes into account both species richness, and the relative abundance of each species to quantify how well species are represented within a community. Many management plans and advice provided to site managers contain information collected in a similar manner to this study to achieve specific objectives. Whether these objectives are to maximise species diversity or simply abundance, we recommend that the community interactions are assessed before management plans are drawn up to avoid the potential loss of valuable habitats and species through inappropriate management.

## **2.6 Conclusion**

In conclusion, this study highlights the importance of data interpretation to determine management objectives, and recommends analysing the community structure and identifying the dominant species prior to undertaking any land management. Although no significant difference was found between flower-visiting invertebrate diversity at sites when month was taken into account, flower density was highlighted as a factor driving the insect diversity showing that increasing the number of flowering plants rather than increasing the amount of specific habitats is a more cost effective management tool for industrial sites. Sites such as Flower Rich Grassland, which would be expected to attract the highest diversity of flower-visiting invertebrates, did not in the case of this study. This is likely a result of the high dominance of one particular species, *L. corniculatus*, which may exclude certain invertebrate assemblages. Importantly, this study highlights that with relatively low cost, industrial sites such as the Lynemouth smelter could be improved with regard to invertebrate

diversity by specific seed planting or a refinement of the mowing practices to allow diversification of flora within and between the sites, improving the overall ecological value of these sites.



## Chapter 3

### **Rapid effects of reduced grassland management on flower and invertebrate abundance**

#### **3.1 Abstract**

Following significant declines in flower-visiting invertebrates a key conservation objective is to maintain or improve grassland habitats to boost connectivity and the frequency of optimal foraging habitat within the wider landscape. An industrial site located on the Northumberland coastline was the focus of the study, where grassland on site was heavily constrained by mowing management. This study reduced the mowing frequency on site by implementing six mowing regimes to establish the best management type for the grassland sward present. Results show that a reduction in mowing frequency significantly improved the flower density recorded within treated plots and the resulting invertebrate diversity was significantly increased compared to the control (heavily constrained) mown plots. Although the results suggest that success of this scale could be site and seed bank dependent, a reduction in mowing can provide a cost effective method of enhancing biodiversity on land considered to be of low ecological value.

### 3.2 Introduction

Pollination is an essential function within ecosystems as well as being of vital importance for food production. In Britain it is estimated that the economic value of the pollination of crops by invertebrates is £510m per year, and it would cost approximately £1.8bn per year to replace (Breeze et al. 2012). However, pollinating invertebrates such as bees and butterflies have suffered marked reductions in numbers over the past decade (Biesmeijer et al. 2006, Gallai et al. 2009, Potts et al. 2010), this decline has been largely attributed to loss of habitat, agricultural intensification, disease, and pesticide use (Goulson et al. 2008b, Potts et al. 2010, Potts et al. 2011). The effect of loss of suitable habitat has been substantial and a key driver behind this has been the fragmentation of suitable habitat (Steffan-Dewenter and Tscharntke 1999). Therefore, a central conservation objective in mitigating declines in flower-visiting invertebrates is maintaining and improving grassland habitats.

Across the UK there are many areas of grassland which could be considered unfavourable to flower-visiting invertebrates. These range from ungrazed pasture left to grow tall or being encroached upon by scrub and trees, to overly managed grazing land, and managed amenity grassland within parks, gardens and industrial sites. These types of grassland are considered to have low ecological value, due to the limited species diversity, homogeneous sward length and, in the case of amenity grassland, intensive management (Lindemann-Matthies and Marty 2013). Without widespread intervention to these managed sites, populations and numbers of pollinating species could continue to decline (Tscharntke et al. 2005).

The regeneration of grassland habitats to enhance invertebrate diversity is not a new concept. Numerous studies have focused on arable landscapes (Thomas and Marshall 1999, Carvell 2002, Pywell et al. 2005, Carvell et al. 2007, Potts et al. 2009, Pywell et al. 2011b, Woodcock et al. 2014), and research into roadside verges (Hopwood 2008, Noordijk et al. 2009) and urban parks and gardens is becoming increasingly popular (Comba et al. 1999, Gaston et al. 2005, Konvicka et al. 2007, Goddard et al. 2010, Matteson and Langellotto 2011, Lindemann-Matthies and Marty 2013, Blackmore and Goulson 2014). However, to our knowledge there is no record of work within active industrial sites to improve amenity grassland and scrubland to benefit pollinating and flower-visiting invertebrate communities.

Although industrial areas are dominated by large buildings and offices, habitat landscaping is an important factor within site design; not only for adhering to planning

requirements, but to provide a visually pleasant and healthy working environment. Planting shrub borders similar to those found in parks and gardens is often utilised, combined with areas of intensively mown amenity grassland, a cost effective method of maintaining green space. However, although these green spaces are pleasing to the eye, they can lack many resources for native fauna.

Within the UK, a greater understanding of the value and importance of native, semi-natural, green spaces is increasing, and, as a result, habitats within parks and gardens are becoming more extensively managed. Many industrial grasslands and shrub borders remain intensively managed, suffering from over pruning and frequent mowing which can significantly impact the attractiveness to flower-visiting invertebrates and reduce forage opportunities (Lindemann-Matthies and Marty 2013). Depending on the soil type and sward quality of grasslands, simple management changes could substantially enhance flowering plant communities, at the same time reducing the need for expensive high maintenance shrub and flower borders and providing increased resources for pollinators.

A reduction in mowing allows growth of less hardy flowering communities which in turn reduces the grass cover, resulting in an increase in floral diversity (Kruess and Tschardt 2002, Gaujour et al. 2011). Success has been recorded on roadside verges (Noordijk et al. 2009) and intensively managed agricultural grassland (Potts et al. 2009) following a reduction in mowing frequency, in addition to an increase in invertebrate visitors. Humbert et al. (2010) also concluded that the frequent use of lawnmowers had devastating effects on invertebrate populations, causing mass mortality in the larval stages of Lepidoptera, and a significant negative effect on Carabidae and Orthoptera numbers, indicating distinct benefit to biodiversity.

However, grassland does need to be carefully managed in order to prevent domination by unfavourable plant species, and maintain flower density and flowering plant species diversity (Pywell et al. 2005). Therefore, there is a requirement for a balance to be maintained in the mowing regime that is applied, being infrequent enough to allow the plant to grow and seed, but frequent enough to maintain sward diversity. This study utilised different cutting regimes applied to grassland areas within an industrial site in Lynemouth, UK, to establish their effects on flower density and invertebrate abundance and diversity. Four hypotheses were identified i) Removing grass trimmings from experimental plots will result in a higher plant species diversity and flower density compared to plots where grass trimmings have been left insitu, ii) Plots subjected to

two cuts (2M, an early and a late cut) over the flowering period will support a significantly higher herb cover and greater suppression of grasses compared to plots mown frequently (0M) and subjected to one cut (1M), iii) Flower density will significantly effect the abundance and richness of flower visiting invertebrate on site, and iv) Plots subjected to one cut following flowering (1M) will support a significantly higher flower-visiting invertebrate diversity compared to plots mown twice a year (2M) and frequent cutting (0M).

### **3.3 Study Design**

#### **3.3.1 Study site**

The study was conducted on the Aluminium smelter site of Rio Tinto Alcan, Lynemouth, UK (55.2016° N, 1.5396° W) located on the Northumberland coastline. Covering 82.7 hectares the site is typical of a working industrial site, with intensively managed grassland and shrub borders around offices, access roads, car parks, and production units. However, 20.7 hectares (25%) of the site is predominately scrub, woodland, and wetland forming a buffer zone. The smelter is located within an agriculturally dominated area, however within the buffer zone contains different habitat zones. To the west of the smelter, hybrid poplar trees have been mixed with native European tree species to provide a privacy screen around the smelter. Some areas of grassland have been left unmanaged, now dominated by thistles (*Cirsium sp.*) and rank grasses restricting the growth of other species.

The trial area was well drained with a shallow covering of nutrient poor soil. The area is dominated by intensively managed mown grassland, but hosted flowering species within the sward such as *Trifolium spp.*, *Lotus corniculatus*, and *Prunella vulgaris*, which were all frequented by pollinating species. Historically wetland habitat was associated with the site, however refinement to drainage has reduced moisture, but many sedges remain within the trial grassland area. Located centrally within the smelter grounds, an active railway line dissects the area into two distinct sections. The treated plots are surrounded by tarmac roads, with shrubs and trees located within the amenity grassland. A two metre buffer was in place around the edges of the railway track for safety reasons, and ease of mowing.

### 3.3.2 Experimental design

Fixed experimental plots measuring 3m x 10m were established in March 2012, and monitored during the mowing season (April to October) over a period of two years (2012 and 2013). Thirty individual plots were identified by coloured canes, and a one metre buffer zone was present around each plot. Plots were arranged in two rectangle blocks (1 x 18 and 1 x12), one each side of the railway line (Appendix 2, Figure A2.1). Each plot was assigned one of six treatments (Table 1; a 2-way orthogonal design, cutting regime (3 levels) X removal regime (2 levels)), with each treatment being replicated five times. The three mowing regimes (Table 3.1) reflect a range from normal practice (one cut per month), to minimal management (one cut per year). Additionally, grass cuttings were either removed or left on the plots, reflecting potential management options in practice.

**Table 3.1:** Summary of the management regimes applied to experimental plots at the Lynemouth smelter UK during the trial

Treatment	Number of cuts	Month of cut	Length of cut	Cuttings removed?
0M	One cut per month	April – Sept	3 inches	NO
0M+	One cut per month	April – Sept	3 inches	YES
1M	One Cut	Mid July	3 inches	NO
1M+	One Cut	Mid July	3 inches	YES
2M	Two Cuts	June and September	3 inches	NO
2M+	Two Cuts	June and September	3 inches	YES

Flower-visiting invertebrate surveys were undertaken between May and September and only on days when conditions were favourable for insect flight, coinciding with peak insect activity and as recommended by the butterfly recording scheme guidelines (Pollard and Yates 1993). Plant species and density was recorded from April when the first flowering plants came into flower. Surveys were conducted every 2-3 weeks, the

exact date would be decided upon weather conditions and having a sufficient length of time after mowing had been undertaken.

The three mowing regimes reflect a range from normal practice (one cut per month to minimal management (one cut per year). Additionally, grass cuttings were either removed or left on the plots, reflecting potential management options in practice. Treatments were designated to plots following a randomised block design; each treatment was replicated five times and replicates were spread across both sides of the railway track in order to eliminate bias as a result of differences in natural floral emergence.

### **3.3.3 Mowing**

Mowing was conducted during dry weather conditions following the experiment design (Table 3.1). A John Deere ride-on mower was used to cut the grass and treatments requiring the removal of cuttings were raked over and the cuttings removed by hand. Cutting removal was undertaken 72 hours following the mowing in order to facilitate seed dispersal into the sward.

### **3.3.4 Invertebrate sampling**

Counts of flower-visiting invertebrates (species observed to frequent flower heads as part of their lifecycle, and be directly or indirectly involved with pollination) were made along a transect bisecting the width of each 3 x10m plot. The transect was walked at a steady speed (~1–2 km/h), and each individual flower visiting invertebrate observed crossing the transect or landing to forage within the boundary of the plot was recorded and identification to family, genus or species (Appendix 2, Table A2.1) as an indication of insect activity. Insects found to be foraging within the plots were only recorded once, and the individuals were monitored to avoid including the same individual twice. During data collection assessment order of the plots was randomised to limit the effects of disturbance from human activity between plots positioned adjacent to one another, a limitation to Insect activity was only recorded when weather conditions were suitable for these species i.e. dry and without strong winds. This constraint led to the recording of pollinating insects not starting until May/June, even though the trial was initiated in April. Once the data set was complete the total abundance, richness and diversity of flower-visiting invertebrates were calculated.

### **3.3.5 Flower Density**

Estimates of flower density within each plot were recorded on the same days as the invertebrate sampling using 10 randomly placed 625cm<sup>2</sup> (25 X 25cm) non-overlapping quadrats within the experimental plots. Flower density was estimated by counting the number of flower heads per plant species recorded from plants rooted within the quadrat area. The flower density represented an estimate of the level of nectar resources available to foraging insects. Once the data set was complete, the data was adjusted to the common scale of 30m<sup>2</sup> for comparison against other factors recorded within the trial.

### **3.3.6 Grassland composition (The proportion of flowering plants and grass)**

To monitor the effects of the different cutting regimes on dicotyledonous species, each experimental plot was divided into three fixed subplot replicates. Subplots were randomly located and non-overlapping, measuring 1m x 1m and marked by inconspicuous permanent markers. Within each sub-plot the percentage cover of dicotyledonous species was recorded. The first assessment was completed on 19/04/2012, prior to the first cuttings there were no statistical differences between % herb cover (Kruskal Wallis  $\chi^2 = 3.5928$ , d.f. = 5,  $P = 0.6094$ ). The plots were reassessed in October 2012 and October 2013.

### **3.3.7 Statistical Analysis**

All statistical analyses were completed within the R statistical and programming environment, version 3.0.2 (R Core Team 2013). The percentage cover of flowering plants data (grassland composition) was arcsine transformed prior to analysis to improve normality, and was compared across different mowing regimes using a two-way ANOVA (mowing regime X cutting removal protocol). The Shannon diversity index, and species accumulation curves were calculated using the 'Vegan' package (Oksanen et al. 2013). Shannon diversity was used to compare invertebrate diversity between treatments types. As a result of the data not conforming to a normal distribution Kruskal-Wallis, followed by pairwise Wilcoxon tests (P-adjusted bonferonni) was utilised. The species accumulation curve was utilised to determine sample completeness for each treatment (Appendix 2, Figure A.2)

A preliminary linear regression with flower density as the dependent variable and regime, year and month as independent variables, including all potential interactions, showed strong heterogeneity of variance in the residuals. Therefore, linear regression with a generalised least-squares (GLS) extension (Pinheiro and Bates 2000) was

implemented. This adds a variance-covariate structure to the linear model, thus explicitly incorporating the heterogeneity of variance. We followed the procedure outlined in Zuur et al (2009). The most appropriate variance-covariate structure was determined using a full model estimated using restricted maximum likelihood (REML). Suitability was determined using the Akaike information criterion (AIC) in conjunction with visual inspection of fitted residuals. Following this, the minimum adequate model (MAM) was obtained by conducting a manual backwards stepwise selection using maximum likelihood methods. The significance of the highest order interaction terms were assessed at each stage, with nested terms not being tested, following Underwood (1998). Once the MAM was obtained, residual diagnostics were used to determine if model assumptions had been met. Assumptions were met and the significance of each independent term within the MAM was assessed using likelihood ratio tests between the MAM and a model based on the MAM but with the particular independent term completely removed. All analyses were performed using the 'nlme' package (Pinheiro et al. 2013) within the 'R' statistical programming environment (R Core Team 2013).

The invertebrate abundance and richness data contained more zeros than expected from a Poisson or a negative binomial distribution. Therefore, to model these variables we used zero inflated negative binomial models, with flower density, month and year as potential explanatory variables. The minimum adequate model was obtained using manual stepwise selection as with the GLS models, with AIC scores used as a second supporting method of selection. These analyses were conducted using the 'pscl' and 'lmtree' packages (Jackman et al. 2007, Hothorn et al. 2014b).

### **3.4 Results**

Over the two-year trial 952 flower-visiting invertebrates were sampled within 7 orders, and 20 plant species were observed (Appendix 2, Table A2.1). The number of invertebrates recorded increased between the two years 2012 (409) and 2013 (543), as did the average flower density: 2012 (99) and 2013 (123).

#### **3.4.1 Effect of experimental factors on flower density**

The minimum adequate model for flower density included two significant two-way interaction terms; Regime X Month ( $L$ -ratio = 211.90, d.f. = 10,  $P < 0.0001$ ), and



Regime X Year ( $L$ -ratio = 17.62, d.f. = 2,  $P < 0.001$ ). Thus, the fourth potential explanatory variable, cutting removal, was not part of any significant terms.

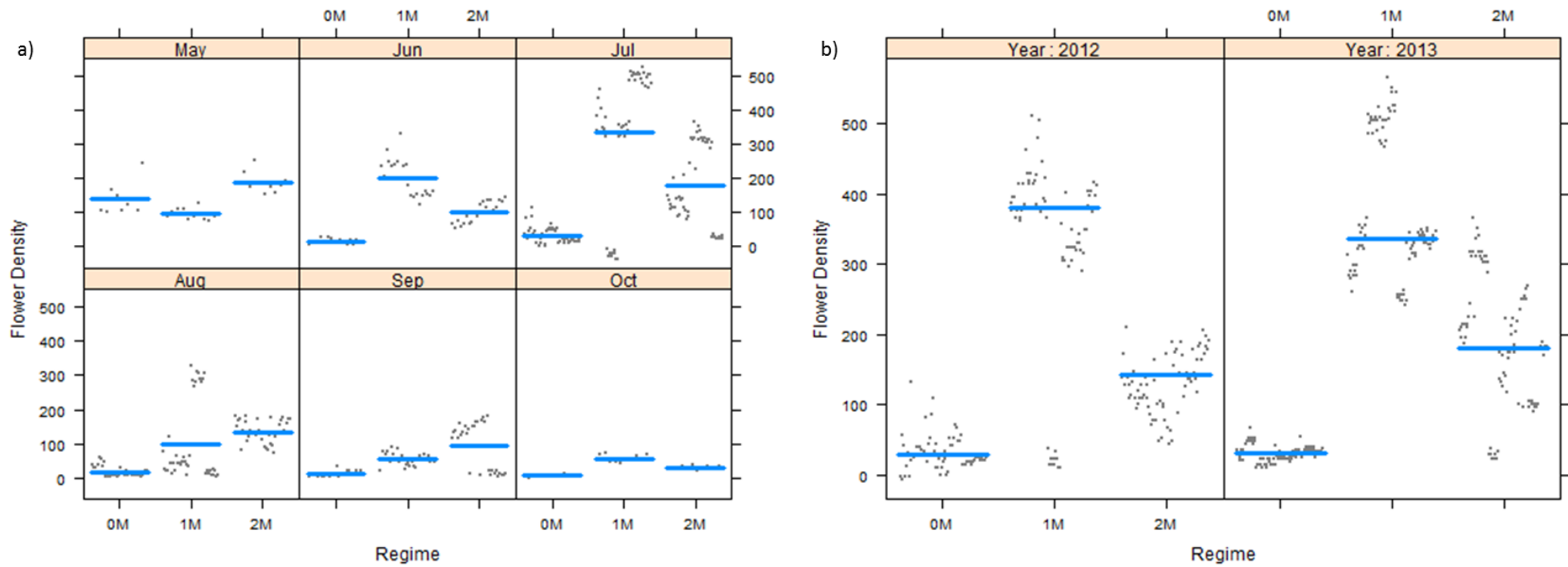
The  $L$ -ratios obtained from excluding individual terms completely from the minimum adequate model can be used to assess the order of importance of the individual variables in the model. Mowing regime and month appear to be of equal importance, and of much greater importance than year (Table 3.2). The variation in flower density between months is clearly observed (Figure 3.1a). A strong peak in flower density is indicated within 1M treatments during June and July, with 2M treatments showing higher flower densities during May, August and September. Figure 3.1b shows the predicted values for flower density, the values for 1M plots appear to decrease and 2M plots increase between years 2012 and 2013, however this difference is not significant within the data collected (Kruskall Wallis,  $\chi^2 = 0.274$ , d.f. = 5,  $P = 0.601$ ).

**Table 3.2:** Estimates of the overall impacts of each of the independent variables in the minimum adequate model, on flower density at the Lynemouth site (using likelihood ratio test between minimum adequate model and reduced model [all terms involving the relevant term removed, including interactions]):

Variable	d.f.	$L$ -Ratio	P Value
Regime	14	379.70	< 0.0001
Year	3	17.97	< 0.001
Month	15	386.48	< 0.0001

### **3.4.2 The effect of grass trimmings removal from experimental plots on flower density and grassland composition**

The removal or not, of grass trimmings was not a significant variable with regards to increasing flower density ( $L$ -Ratio = 0.037, d.f. = 15,  $P = 0.847$ ). However, following a two way ANOVA on grassland composition (Oct 2013), both mowing regime (ANOVA,  $F = 98.886$ , d.f. = 2,  $P < 0.001$ ) and cutting removal (ANOVA,  $F = 4.516$ , d.f. = 1,  $P < 0.05$ ) significantly increased the percent flower cover recorded. The interaction between mowing regime and cutting removal was not significant (ANOVA,  $F = 1.802$ , d.f. = 2,  $P = 0.171$ ), highlighting that the effect of cutting removal was similar across all regimes.



**Figure 3.1:** Visualisation of the two significant two-way interaction terms of the linear regression model with a GLS extension, with flower density as the dependent variable; (a) Regime X Month interaction, and (b) Regime X Year interaction. The blue lines represent model predictions and the points are partial residuals.

### **3.4.3 Effects of the mowing regimes on grassland composition**

By the end of October 2013 there were clear differences between the three mowing regimes, with the two-cut treatments (2M and 2M+) resulting in a significant reduction in the total area dominated by grass (Figure 3.2). All treatments had a similar percent herb cover (ANOVA,  $F = 0.778$ , d.f. = 5,  $P = 0.569$ ), and after just one season (Oct-2012) a significant difference was observed between the six mowing regimes (ANOVA,  $F = 11.94$ , d.f. = 5,  $P < 0.001$ ).

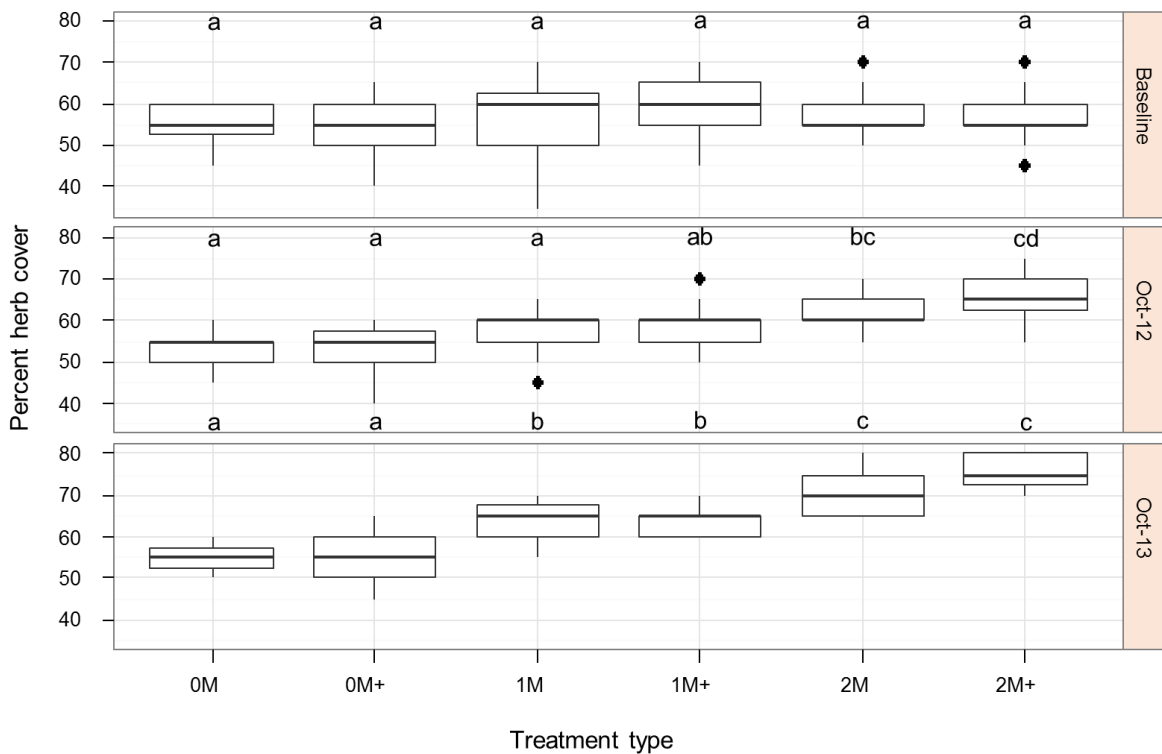
Tukey HSD comparisons revealed plots maintained with treatments 2M and 2M+ had significantly higher percentage flowering species cover compared to the other treatments. After the second season (Oct-2013), the differences in grassland composition between the treatments had increased (ANOVA,  $F = 41.18$ , d.f. = 5,  $P < 0.001$ ) (Figure 3.2), with 2M and 2M+ the most successful at suppressing grass dominance and supporting more flowering species.

### **3.4.4 Response of invertebrate abundance and richness to flower density**

Following spearman rank correlation tests on the data collected, a significant positive correlation between flower density and flower-visiting invertebrate abundance was recorded for each year (2012,  $\rho = 0.141$ ,  $P < 0.05$ , and 2013  $\rho = 0.680$ ,  $P < 0.001$ ).

Between the two years there was a temporal difference in the timings of the presence of invertebrate assemblage. In 2012 flower-visiting invertebrate activity occurred during May but was absent in October. In contrast, in 2013 we saw invertebrate activity in October but not in May. This is likely to be due to a combination of the weather conditions observed within the earlier seasons and the improved flowering density following the different cut regimes in 2013. Therefore, analysis has been conducted on data collected between June and September, where all months had invertebrates recorded in both years.

Zero inflated negative binomial models were used to determine the influence flower density, month and year had on invertebrate richness and abundance. For invertebrate richness the 3-way interaction term was significant in both the main model component ( $\chi^2 = 39.27$ , d.f. = 4,  $P < 0.001$ ) and the inflation component ( $\chi^2 = 20.11$ , d.f. = 4,  $P < 0.001$ ). For invertebrate abundance this 3-way interaction term was also significant in

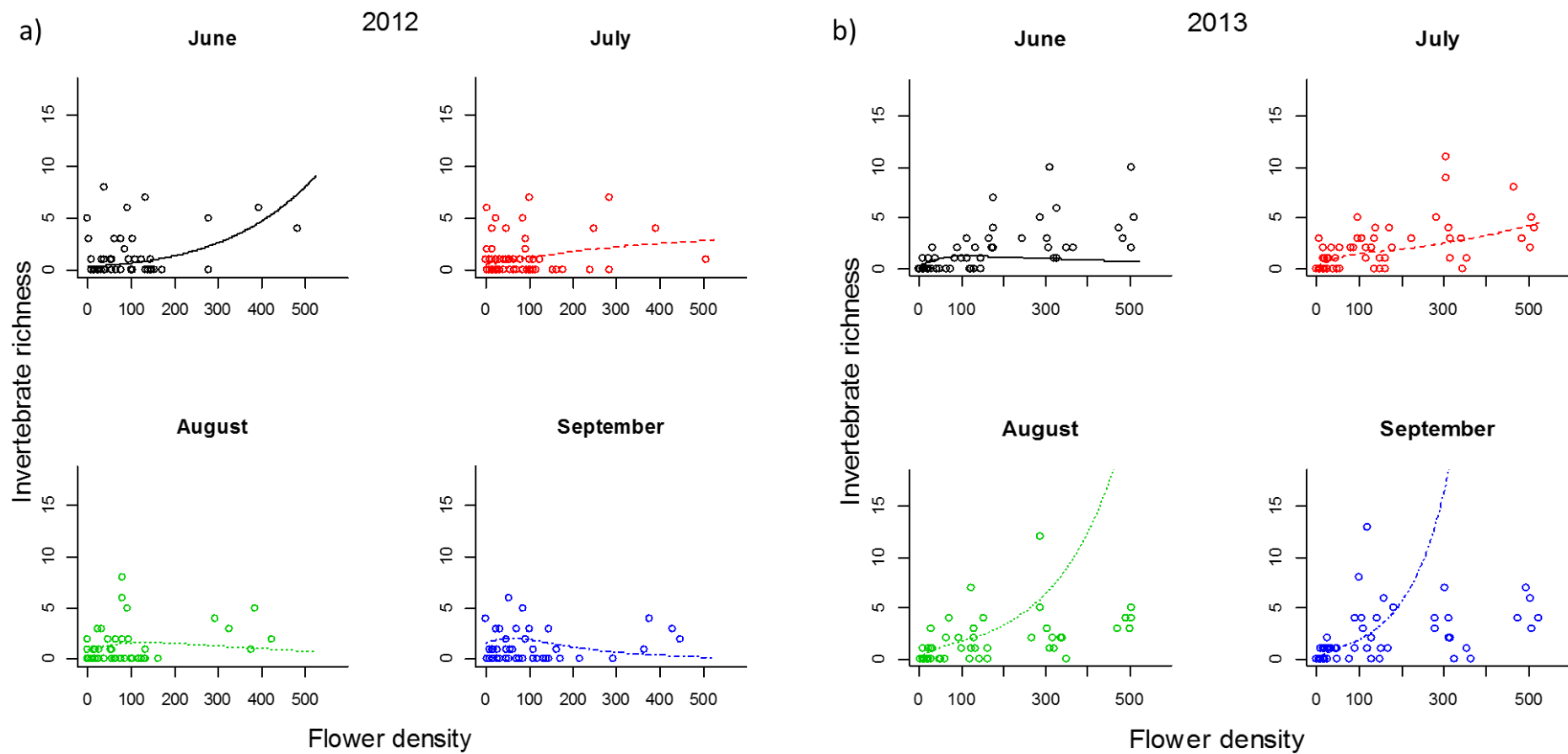


**Figure 3.2:** Whiskered box plot showing percentage herb cover at the three sampling times. The box plot is constructed from the median, and upper and lower quartiles. Whiskers show the maximum and minimum data points recorded, excluding any outliers. Outliers in the data are identified by black dots. Letters represent differences following Tukey HSD Comparison tests, after one way ANOVA on each year.

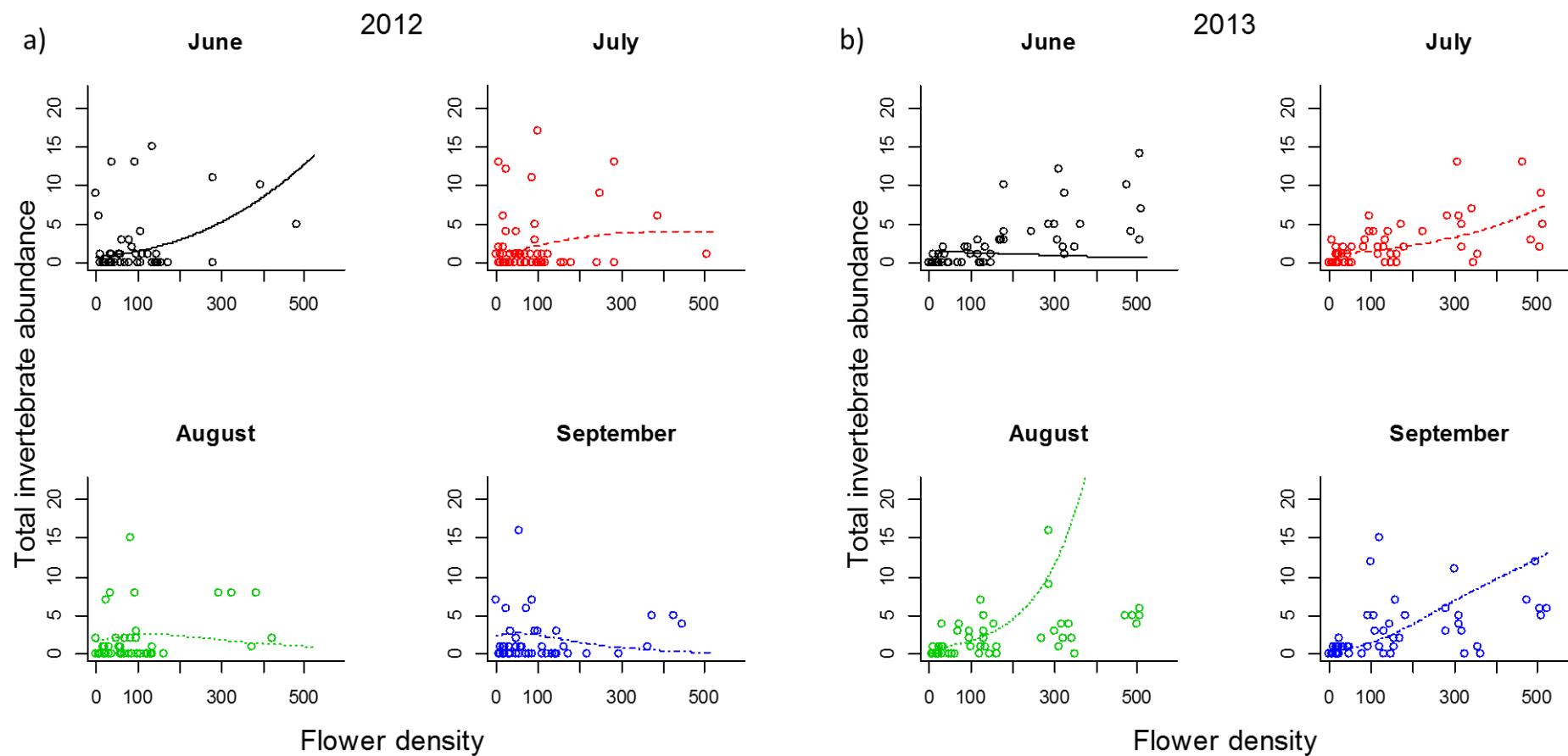
Treatments: (0M) One cut per month, (0M+) One cut per month, with cutting removal, (1M) One Cut, summer, (1M+) One Cut, summer with cutting removal, (2M) Two Cuts, one spring, one autumn, (2M+) Two Cuts one spring, one autumn, with cutting removal.

both the main model component ( $\chi^2 = 38.13$ , d.f. = 4,  $P < 0.001$ ) and the inflation component ( $\chi^2 = 16.65$ , d.f. = 4,  $P < 0.01$ )

Visualisations of the statistical models of the invertebrate richness and abundance are shown in Figure 3.3 and Figure 3.4, respectively. There was a much stronger positive relationship for the months July, August and September in 2013 compared to 2012. However, there was a flatter and lower trend in June 2013 compared with June 2012. It is important however to remember that the data spread for the flower densities is restricted within each month, with only July showing a range reasonably close to the full range. As a result predictions from these models can only be utilised within the independent variable ranges found in this study.



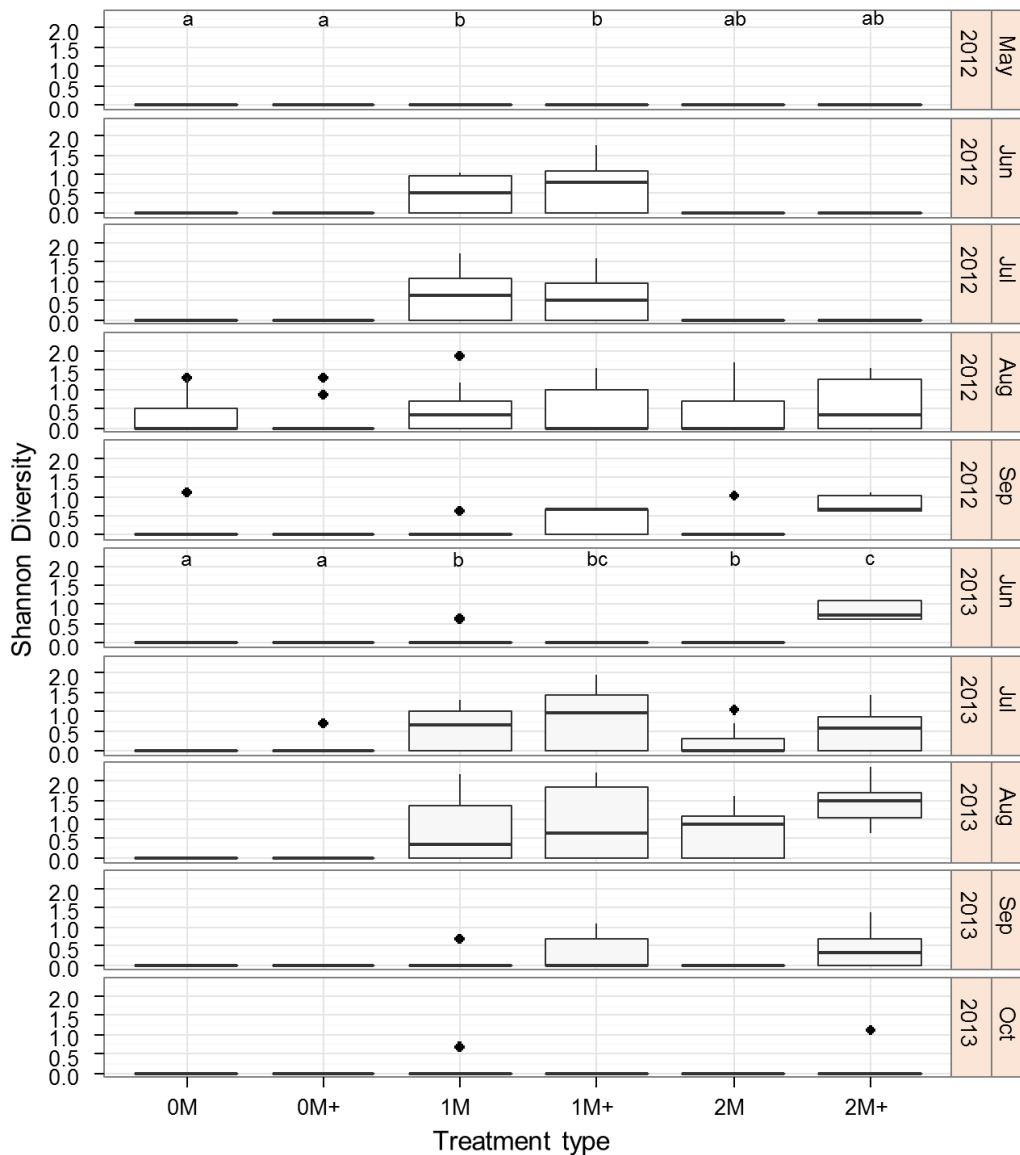
**Figure 3.3:** Visualisation of the significant three-way interaction (Flower density X Month X Year) of the zero inflated negative binomial model used to determine the relationship between invertebrate richness and flower density between a) 2012, and b) 2013. Circles represent data points, and the lines show the model predictions.



**Figure 3.4:** Visualisation of the significant three-way interaction (Flower density X Month X Year) of the zero inflated negative binomial model used to determine the relationship between total invertebrate abundance and flower density between a) 2012, and b) 2013. Circles represent data points, and the lines show the model predictions.

### **3.4.5 Effects of mowing regimes on invertebrate diversity**

The introduced mowing regimes had a positive significant effect (Kruskal-Wallis  $\chi^2 = 94.285$ , d.f. = 5,  $P < 0.001$ ) on invertebrate diversity. Each year was assessed separately, followed by Pairwise Wilcoxon tests (P-adjusted bonferonni) to determine significance between treatments (Figure 3.5). Invertebrate diversity also increased significantly between 2012 and 2013 (Wilcoxon,  $W = 10691.5$ ,  $P < 0.05$ ) within the treated plots (1M, 1M+, 2M, 2M+). During 2013 treatments with cuttings removed showed a higher diversity of invertebrates, compared to plots with cuttings left after mowing, highlighting the potential benefits of removing cuttings over a longer time frame.



**Figure 3.5:** Whiskered box plots showing the difference in Shannon Diversity Index between treatments. The box plot is constructed from the median, and upper and lower quartiles. Whiskers show the maximum and minimum data points recorded, excluding any outliers. Outliers in the data are identified by black dots. Letters represent differences following Pairwise Wilcoxon tests (P-adjusted bonferonni), following Kruskal- Wallis on each year (2012,  $\chi^2= 27.98$ ,  $P < 0.001$ , and 2013,  $\chi^2= 80.11$ ,  $P < 0.001$ ). Treatments: (0M) One cut per month, (0M+) One cut per month, with cutting removal, (1M) One cut, summer, (1M+) One cut, summer with cutting removal, (2M) Two cuts, one spring, one autumn, (2M+) Two cuts one spring, one autumn, with cutting removal.



### **3.5 Discussion**

The grassland sward responded rapidly to changes implemented through this trial, accepting the prediction that plots subjected to the 2M regime would support a significantly higher percentage herb cover compared to other treatments, plots subjected to 1M regimes would support a significantly higher flower-visiting invertebrate diversity and that a significant correlation between invertebrate abundance and flower density would be revealed. However, the removal of grass trimmings from experimental plots was not identified as a significant factor.

The increase in abundance and diversity of invertebrates recorded within treated plots would be expected as a result of an increase of forage and botanical structure. It is well documented that pollinator diversity can be readily enhanced by the introduction of novel flower rich habitat (Carvell 2002, Carvell et al. 2004, Pywell et al. 2005, Pywell et al. 2011b, Blackmore and Goulson 2014, Woodcock et al. 2014). However, here we have demonstrated that the same effect can be achieved through amendments to management practises, without requiring relatively costly seed mixtures. Furthermore, we highlight the important role management plays in increasing and maintaining flower density within grassland, and show how quickly these changes can become apparent, once the change in management is initiated. Reducing mowing frequency enhanced flower diversity, which in turn enhanced invertebrate abundance and richness (Tscharrntke et al. 1998, Goddard et al. 2009). There was a much stronger positive relationship between invertebrate abundance and flower density for the months July, August and September in 2013 compared to 2012, emphasizing the shift of flower density to the higher end of the range.

This was a relatively short-term study of two years, and although we saw a positive effect of management, over a longer period of time the natural regeneration of plants may start to decline, requiring seeding to remain valuable to invertebrates (Carvell et al. 2004). With this caveat, as a first step to enhancing floral and invertebrate abundance and diversity within grassland constrained through restrictive management, this method proved very successful here. The reduction in mowing frequency caused flower density to increase rapidly for the duration of the trial. After the initial assessments flowering species were recorded within treated plots that were not observed within the control plots (0M). The two cut method (2M) allowed a second flowering period into September with a higher flower density than the one cut method (1M), providing forage for invertebrates flying later in the season. This finding supports

the work of Noordijk (2009) where, within roadside verge maintenance, a two cut method was the most effective to achieve greater biodiversity of flower-visiting invertebrates. However, within our study the early cut could reduce insect visitation during June and July, potentially hindering the reproductive success of both plants and insects. Buri (2013) saw a similar effect during his study on orthopterans, highlighting the importance of site specific management timings, flexibility to allow for seasonal variations, and guidelines for staff. The one cut after flowering method (1M) did record the highest number of flower-visiting invertebrates during peak insect activity, suggesting it is the most effective method for increasing invertebrate diversity (Potts et al. 2009). The method is commonly used as it mimics traditional hay meadow cutting/grazing. Care should be taken in recognising that these findings are over the relatively short-term period of two years. Future work will need to be done in determining which of the methods (1M or 2M) is better at maintaining the grassland composition, and flower and invertebrate density over longer time periods.

Month and year both significantly influenced the flower density throughout the trial, emphasising that the emergence rate and species presence varied over the short-term (month) and over the longer term (year). As the result of the reduction in mowing frequency, plants were able to flower and set seed for the next year, reducing grass cover and allowing germination of the natural seed bank. Month was particularly influential in determining flower density. Invertebrate abundance was significantly influenced by flower density, and the monthly variation of flower density could be detrimental to forage availability over the season. Introducing additional flowering species via a seed mix could extend the flowering period and help to reduce the monthly variation in flower density. However, it must be remembered that many invertebrate species require variation in forage plants throughout a season to suit life cycles, and this must be accounted for in any plan to maintain floral diversity over the season.

Cutting removal had a significant effect on percentage herb cover during October 2013, however, cutting removal was not found to be a significant predictor of flower density. Previous studies (Schaffers et al. 1998, Schaffers 2002, Endels et al. 2007, Noordijk et al. 2009), found that cutting removal significantly improved grassland quality, indicating that the removal of hay cuttings are paramount to establishing greater plant diversity, particularly within nutrient rich soils. The removal of hay cuttings not only prevents the build-up of nutrients in the soil which can result in ruderal grass

domination (Pywell et al., 2011), but also opens up the sward for seedling dispersal and seed recruitment (Endels et al. 2007). Therefore, it is likely that the lack of a significant effect of cutting removal in this study is likely to be due to the relatively short runtime of the trial. and under a prolonged period, leaving grass cuttings will heavily nitrify the soil, reducing the diversity of the sward (Schaffers et al. 1998).

### **3.6 Conclusions**

The diversity and number of flowering plant species significantly increased as a result of changes in the mowing regime employed at the Alcan smelter, highlighting the importance and potential of relatively small and inexpensive management changes in promoting local biodiversity. Decreasing mowing frequency, increased flower density within treated plots having a positive effect on the abundance and diversity of invertebrates visiting the site, showing that a more targeted and complementary change in management of grassland associated with industrial sites, and the wider environment can significantly benefit biodiversity, at least in the short term. Long term benefits require constant monitoring of changes onsite, and further work here could establish whether the long term composition changes within the 2M will provide a greater flower density than 1M plots subjected to single cut in the future. It is essential that habitat improvements to benefit flower-visiting invertebrates over longer time scales are considered in conjunction with the shorter term management strategies addressed here, and reviewed to ensure success. Similarly, the benefits of increasing forage resources at the smaller scale must be considered within the context of the larger landscape scale for the full benefits to be properly assessed (Heard et al. 2007).

#### **3.6.1 Future management recommendations**

This study demonstrated that simple changes in the management of grasslands within an industrial site could significantly increase floral and invertebrate abundance and diversity. However, there is substantial potential to introduce further improvements. Firstly, the introduction of mid to late summer flowering species such as *Centaurea nigra* and *Malva moschata*, could improve the longevity of forage resources, in particular during the reproductive stages of bumblebees (Westphal et al. 2009). Towards the end of the season the new queens need to gather substantial resources in preparation for hibernation. Ensuring forage at this time is essential for preventing species decline (Mommott et al. 2010).

Secondly, this study has shown that both cut methods had their own advantages. Buri (2013) highlights the importance of uncut patches within grassland as refuge for invertebrates, in addition to the importance of delaying the first cut as long as possible, which supported by our findings. It is also well documented that unmanaged grassland is more suitable for Carabidae, Orthoptera and Lepidoptera larvae. However, in the long term, management by grazing or mowing is essential for the success of flowering grassland. Therefore, a regime which incorporates a mixture of cut timings and cut lengths on a rotation system with the cuttings removed would be the ideal method to enhance grassland biodiversity.

## Chapter 4

### **Determining the success of grassland management intervention by assessing flower-visiting invertebrate abundance, and species interaction networks between sites.**

#### **4.1 Abstract**

Semi natural grassland is valuable for flower-visiting invertebrates, providing a native foraging resource within agriculturally and urban dominated landscapes. Across the UK, schemes have been introduced to enhance wildflower abundance either by management regimes (Alcan Smelter, Lynemouth, UK) or through seeding regimes (Growing wild scheme, Northumberland, UK). Flower-visiting invertebrate abundance and flower density were recorded at the Alcan smelter, two growing wild sites, and two local wildlife sites designated for their semi-natural grassland, to establish the success and value of intervention at the Alcan smelter to benefit flower-visiting invertebrate populations. Results showed that the abundance and diversity of flowering plants and invertebrates found within the grassland at the Alcan smelter were significantly higher than both the introduced seeded regimes and local wildlife sites. Comparisons of the plant and flower-visiting invertebrate networks showed that the grassland at the Alcan smelter supported a habitat which was more stable, hosting a greater species diversity of both plants and flower-visiting invertebrates compared to the seeding sites in particular. The study highlights the success of management intervention within the grassland at the Alcan smelter, but also draws attention to the importance of continued management to maintain these improvements.

## 4.2 Introduction

Increased urbanisation and intensified agricultural practices have resulted in a reduction of natural flower rich grasslands throughout Europe (Blackstock et al. 1999, Biesmeijer et al. 2006). Exacerbated by habitat fragmentation, poor habitat connectivity between remaining habitat parcels has resulted in invertebrate population declines (Biesmeijer et al. 2006). Managed *Apis mellifera* colonies have shown declines since 1985, with a mean loss of 54% in England (Potts et al. 2011), and declines have also been evidenced in approximately 2,000 wild bee species (Goulson et al. 2008a, Potts et al. 2011). As a response to the decline in suitable habitat, many European countries have attempted to reverse invertebrate decline by enhancing and reintroducing flower and nectar rich grasslands across agricultural and urban environments (Noordijk et al. 2009, Haaland et al. 2011). Implemented through agri-environment and council schemes, it is proposed that a greater network of suitable habitat could be provided for invertebrates, improving the connectivity of semi-natural grasslands which in turn will encourage and support species rehabilitation (Wratten et al. 2012).

The planting of wildflower and nectar rich field margins is a popular concept, and is increasingly becoming common practice within agricultural systems, particularly because of government funding through agri-environment schemes (Carvell et al. 2004, Pywell et al. 2005, Woodcock et al. 2014). These margins increase botanical structure and diversity within predominately monotypic agricultural landscapes, providing additional forage and refuge for invertebrate fauna. Although these planting schemes generally achieve a positive response in invertebrate abundance (Thomas and Marshall 1999, Denys and Tschardtke 2002), it is argued that the majority of invertebrate records are already common frequenters of agricultural landscapes, resulting in no overall improvement in biodiversity (Haaland et al. 2011). Nevertheless, these schemes were not necessarily designed to encourage rarer species in to agricultural habitats (Potts et al. 2006), only to increase pollinator abundance, for which success is undeniable. The increase in flower abundance and subsequent increase in flower visitation rates is essential to maintain crop production and local wildflower populations within Europe (Aizen et al. 2009, Potts et al. 2010). Furthermore, any rarer species from a variety of taxa utilising these novel flower rich habitats (Woodcock et al. 2005, Pywell et al. 2006, Zurbrugg and Frank 2006, Carvell et al. 2007) indicate a positive effect in the face of an increasingly negative outlook for flower-visiting invertebrates.

Responding to the success of planting schemes associated with agricultural lands, local authorities within the UK have introduced wildflower seeding schemes within urban areas with similar success (Blackmore and Goulson 2014). Enhanced amenity grassland within housing estates, parkland and roadside verges is becoming a common sight within the UK, increasing the coverage of native meadow plant species, providing a mosaic of suitable forage and refuge for flower-visiting invertebrates within intensely urbanised areas, helping decrease the effects of habitat fragmentation. However, as with the agricultural schemes, continued management of these seeded areas is essential to ensure flowering longevity, encourage a variety of invertebrate species, suppress dominant grass species, and ensure the planted herb species can germinate and thrive (Carreck and Williams 2002, Marshall and Moonen 2002, Pywell et al. 2011b). Despite a reduction in mechanical mowing allowing successful diversification of a species poor sward in an economic manner (Chapter 3) it is inevitable that some reseeded areas may be required to sustain flower density for invertebrate foragers, Carvell et al. (2004) highlighted that within agricultural field margins the density of flowers from natural regeneration was not enough to sustain bumble bee populations. It is therefore, very likely that a similar effect will be echoed in the urban environment with sowing of particular plant seeds being required in future to maintain flower density and diversity.

Although semi-natural grassland cover has declined within the British landscape (Critchley et al. 2004), fragments have been preserved because of their species diversity or plant communities. These sites are valuable to local insect biodiversity, firstly as they have hosted invertebrate communities over many years providing a true representation of local species diversity, and secondly because they have plant species known to succeed and support flower-visiting invertebrates in the geographical region. Information from these sites can be a valuable tool to researchers and management bodies giving a baseline of the insect pollinators in that area for any given habitat and/or a target for management plans to aspire to. These islands of natural habitat have remained diverse despite relatively little human interference or management, providing refuge for invertebrates during times of habitat loss, highlighting the benefits of semi-natural grasslands to pollinators and other invertebrates within agricultural landscapes (Öckinger and Smith 2007).

During 2012, simple cost-effective management changes were introduced within the grounds of the Alcan aluminium smelter, Lynemouth, UK. At this site the mowing

frequency was reduced, resulting in a significant increase in flower density, in addition to a positive response in both abundance and richness of flower-visiting invertebrates (Chapter 3). This study aims to compare the response of flower-visiting invertebrates to management at the Alcan smelter, to sites managed by manual seeding (undertaken by local authorities in the area), and two naturally diverse wild flower grasslands in the region to determine the success of the management changes undertaken and use species interaction networks to determine flowering species of importance within the local area. Three hypotheses were investigated further i) There will be a significant relationship between the different plant assemblages on site on the invertebrate communities they attract, ii) Flower visiting invertebrate abundance and diversity will be significantly higher at nature reserve grasslands and sites managed with wildflower mix, iii) Flower density and flowering plant diversity will be significantly higher at nature reserve grasslands and sites managed with wildflower mix, and iv) Ecological network analysis will highlight flowering plant species of importance to guide future management strategies.

### **4.3 Study Design**

The grassland community within the Alcan smelter was compared to four other sites (Table 4.1) within approximately 15 km of the smelter (Appendix 3, A3.1). Two of the sites were classed as nature reserves because of their natural plant communities (Potland Burn, and Brenkley Colliery), and two had been artificially enhanced following a local council scheme to improve species diversity (Hartford Bridge and Fallowfield) within amenity grassland (Table 4.1). The flower density, invertebrate abundance, and foraging choice of flower-visiting invertebrates were assessed for each site over the peak foraging period from May to September.



**Table 4.1:** Brief description of each of the five sites used within the comparison study

Site Name	Location	Description	Current Management
<b>Brenkley Colliery</b>	NZ220744	Naturally colonised scrubland. Flower rich grassland community on poor soil	Unmanaged
<b>Potland Burn</b>	NZ256888	Naturally colonised Meadow known for its flower rich grassland and vetch species.	Grazing – October onwards
<b>Alcan</b>	NZ292897	Naturally colonised flower rich grassland, poor soil	Mixed Mowing regimes
<b>Fallowfield</b>	NZ269861	Planted wildflower meadow	Mown June and October
<b>Hartford Bridge</b>	NZ241801	Planted wildflower meadow	Unmanaged

#### **4.3.1 Invertebrate sampling**

Data was collected from three, 10 metre transects which crossed each site. Each transect was surveyed twice a month over the survey period of May – September 2013. Transects were walked at a steady speed (~1–2 km/h) and observations were recorded in a zone two metres either side of the transect line. Each time a flower-visiting invertebrate (any species known to forage on flowers or use flowers as part of their life cycle, and could be considered important for pollination) entered the sample zone it was recorded, if the insect visited a plant, that species was also recorded. All invertebrates were identified to species where possible. Syrphids were identified at least to genus and to species where possible. Due to the similarity between workers of *B. terrestris* and *B. lucorum* these species were treated as an aggregate species (Prys-Jones and Corbet 1991). Observations were only made between 10:00 and 17:00 h, when weather conformed to Butterfly Monitoring Scheme standard (Pollard and Yates 1993).

#### **4.3.2 Flower density**

Estimates of flower density were collected at the same time as the invertebrate sampling, twice monthly. Five randomly placed 625cm<sup>2</sup> quadrats were used within three permanently fixed 5x5 m quadrats. In each 625cm<sup>2</sup> quadrat, the numbers of

plants and the number of flower heads per plant species were recorded. For each site the mean flower density was calculated.

### **4.3.3 Data Analysis**

All statistical analyses were conducted within the R programme (R Core Team 2013), utilising the 'vegan' package (Oksanen et al. 2013) for ecological datasets. The 'specaccum' function was used to perform species accumulation curves to determine sampling completeness (Appendix 3, Figure A3.2) Following Hellinger transformation (Legendre and Gallagher 2001) to counteract the large number of zeroes within the data set, Principal Component Analysis (PCA) was completed on both the plant and invertebrate data sets for each assessment date to determine the effect of flower density on flower-visiting invertebrate abundance. The matrix for PCA1 and PCA2 were constructed and compared using Procrustes rotation (Gower 1975, Oksanen et al. 2013), which quantifies the multivariate similarity between plant and invertebrate matrices by rotating and rescaling the ordinations so as to achieve maximum similarity. To determine the significance of the factors flower density and site, variance partitioning was completed on the data within the 'vegan' package (Oksanen et al. 2013). The significance of the fractions produced during this method were determined following redundancy analysis (RDA), allowing us to systematically partial out the effects of each variable to determine the individual or joint effects on the response variable, in this case total invertebrate abundance.

#### **4.3.3.1 Assessing abundance and diversity data**

The response of invertebrate abundance and plant abundance to the factors site and date surveyed were tested using a Generalised Linear Model (GLM) with a Poisson error structure. Pairwise post-hoc comparisons were made using the glht function within the 'multcomp' package (Hothorn et al. 2014a).

Shannon-Weiner species diversity was calculated for each site using the 'vegan' package. Between sites comparisons were made using the non-parametric Kruskal-Wallis test, followed by Pairwise Wilcoxon comparisons, with the P-value Bonferonni adjusted (Adbi and Valentin 2007) to correct for multiple testing.

#### **4.3.3.2 Descriptors of ecological network structure**

Each site and its resulting ecological network was assessed following construction of ecological network matrices using the 'bipartite' package (Dormann et al. 2014). This package allows visualisation of simple network structures and displays the strength of each relationship between higher and lower level species, allowing an insight into the complex working of the site rather than just comparing the abundance of plants and invertebrates recorded.

The package can also calculate a number of indices which are used in network theory link analysis can be used to evaluate relationships (connections) between species at each site. Three common indices are connectance, linkage density and interaction evenness (Tylianakis et al. 2007). Connectance on the scale of 0 to 1 is the proportion of possible links that are realised amongst the potential links; allowing connectance to predict key dynamic properties of ecological networks and providing the standardised number of combinations (Dormann et al. 2014). Linkage density refers to the realised number of possible links within the matrix and is calculated as the number of higher trophic level species multiplied by the number of lower trophic level species. Finally, interaction evenness or Shannon's evenness ranges from 0 to 1, when the score reaches 1 the number of interactions between invertebrate and plants species is uniformly distributed, showing a homogeneous distribution of interactions within the network (Tylianakis et al. 2007, Morris et al. 2014).

The generality of the higher trophic level species and the vulnerability of the lower trophic level species were also calculated, The values represent the mean effective number of consumers per plant and vice versa. Lower values indicate higher levels of specialisation by invertebrates within the ecological networks (Morris et al. 2014).

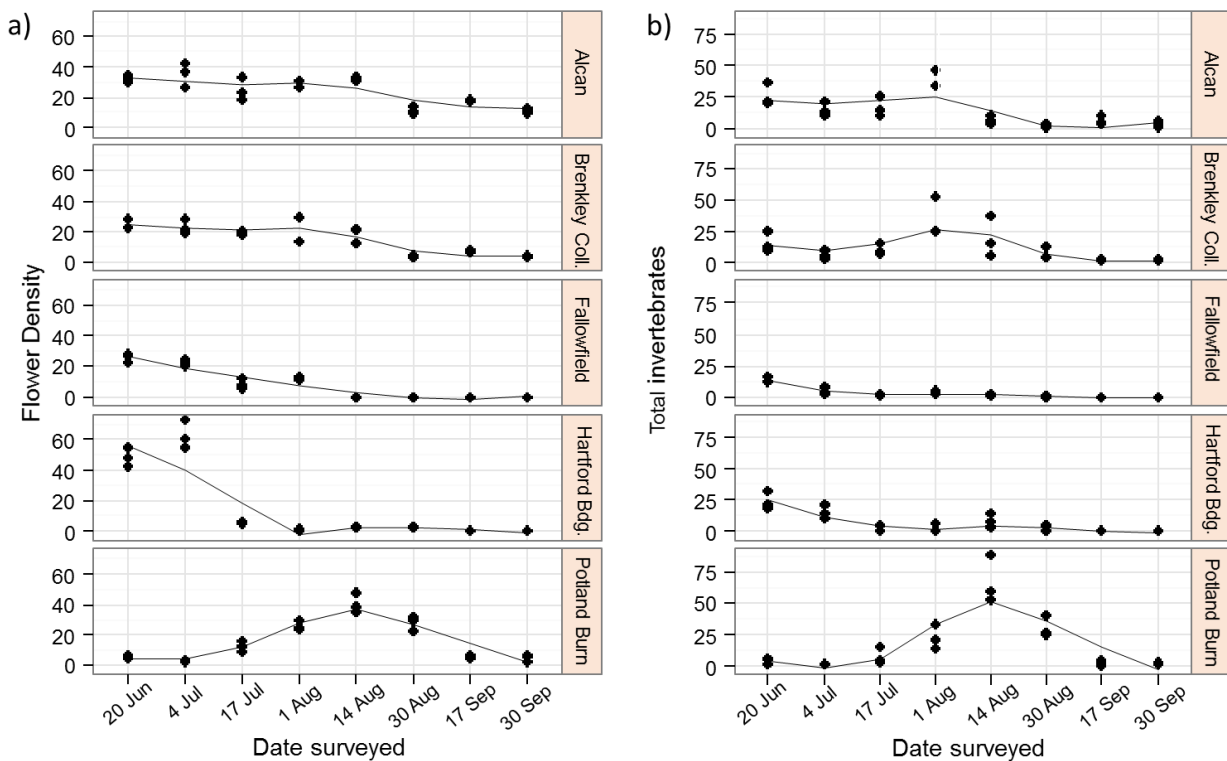
The precision of each of the descriptive indices used was calculated using the 'vaznull' function within the 'bipartite' package. 'Vaznull' is a constrained null model which was used to generate 1000 permutations of the index result from the pollination network, by randomising the recorded results. This allowed us to plot the data with an estimate of the index accuracy and provide 95% confidence intervals (Dormann et al. 2014).

As the variance partitioning analysis was completed on PCA results, we were unable to directly identify the key flowering species utilised by invertebrates throughout the trial. Therefore, the species strength scores calculated via the ecological network indices were used. The species strength represents a sum of the level of dependencies

of other species on the target, quantifying the importance of a species within a network (Dormann et al. 2014). These species strength scores however, do not take into account the length of availability through the season; they are calculated from the total species scores recorded at the end of the study.

#### 4.4 Results

Flower density and invertebrate abundance decreased across the survey period (Figure 4.1). The planted meadows at Hartford Bridge and Fallowfield both had high flower density in June, however showed a sharp decline with no regeneration for the remaining survey period. Interestingly, Potland Burn showed a contrasting trend to the other four sites, with a slow increase in flower density, peaking in August then gradually declining (Figure 4.1a). The total invertebrate abundance followed the same trend as the flower density, however the three naturally colonised sites had a peak of activity between July and the end of August (Figure 4.1b).



**Figure 4.1:** Scatterplot with a loess smooth curve (calculated within the ggplot2 package within R), showing the change in a) the flower density and b) flower-visiting invertebrate abundance recorded, for each of the five site

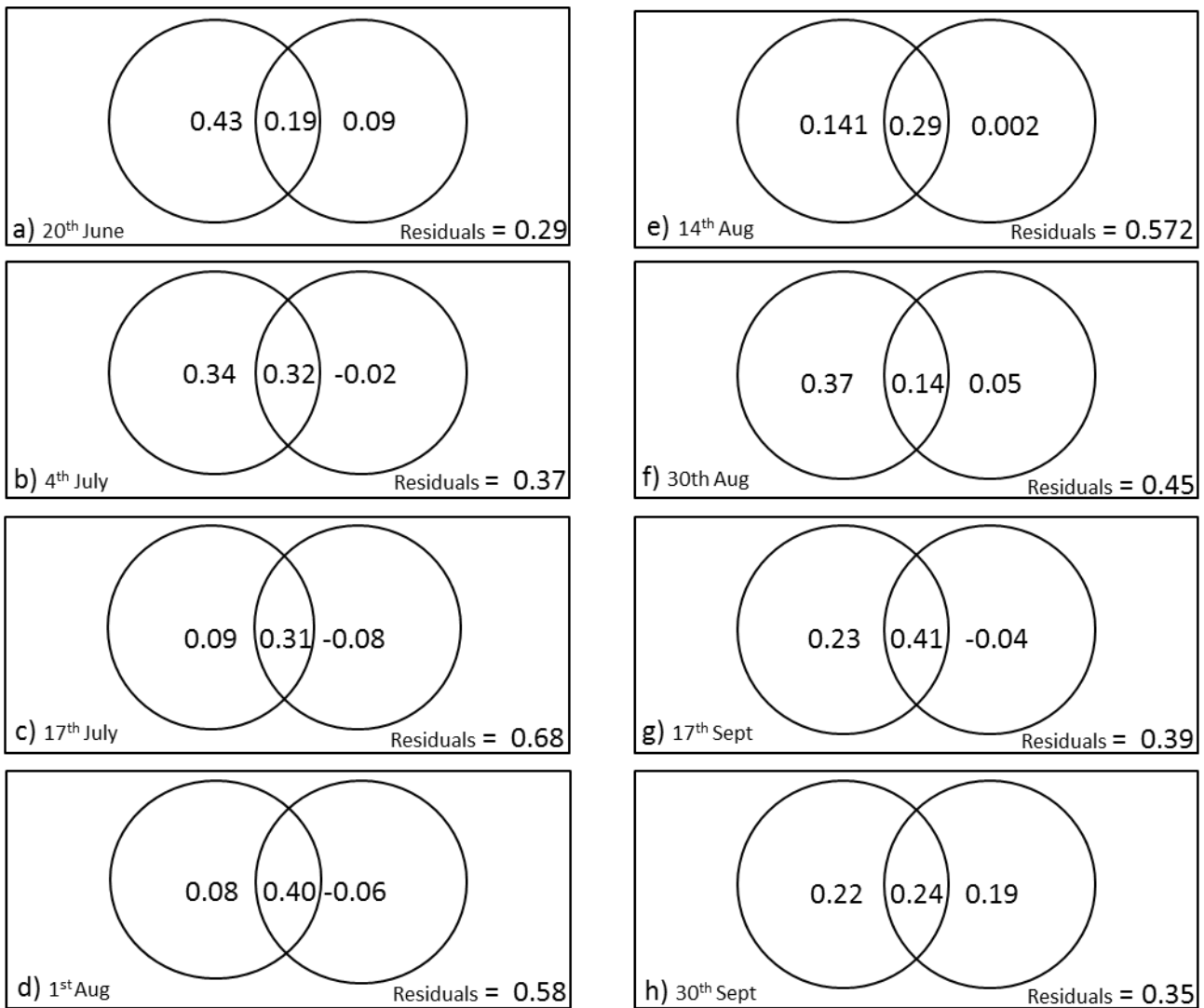
#### **4.4.1 Determine the relationship between the different plant assemblages on the invertebrate communities they attract**

Each assessment date was tested to determine the effect of plant communities on invertebrate abundance. Procrustes analysis on all survey dates showed a significant correlation between plant and invertebrate community composition (Table 4.2).

**Table 4.2:** Results from Procrustes analysis showing the correlation between the invertebrate and plant community ordinations measured via the m12 squared values (interpretation analogous to that of an  $R^2$  value), correlations were significant on all sample days. The data was Hellinger transformed (Legendre and Gallagher 2001) prior to analysis as a result of the zero-inflated content of ecological count data.

<b>Date assessed</b>	<b>Correlation in a symmetric Procrustes rotation</b>	<b>Procrustes Sum of Squares (m12 squared)</b>	<b>P value</b>
<b>20th June</b>	0.73	0.48	<b>0.001</b>
<b>4th July</b>	0.78	0.39	<b>0.001</b>
<b>17th July</b>	0.66	0.56	<b>0.004</b>
<b>1st Aug</b>	0.62	0.62	<b>0.004</b>
<b>14th Aug</b>	0.59	0.66	<b>0.009</b>
<b>30th Aug</b>	0.55	0.70	<b>0.031</b>
<b>17th Sept</b>	0.63	0.50	<b>0.004</b>
<b>30th Sept</b>	0.80	0.36	<b>0.001</b>

To determine the role of site and flower density on invertebrate abundance, variance partitioning was completed, allowing identification of the percentage of variation relating to each predicting factor. Figure 4.2 summarises the results showing that the joint effect of site and flower density, or the site alone were the most significant factors influencing invertebrate abundance. The residuals ranged from 0.29 – 0.68, highlighting that other unspecified factors were influencing the results.



**Figure 4.2:** Variance partitioning showing the percentage of variation relating to two explanatory variables. Left circle is site, right circle is flower density and their joint effect is in the middle. The calculated residuals are presented in the bottom right hand corner.

To determine the significance of the proportions presented within Figure 2, Redundancy analysis (RDA) was completed. Both factors, site and flower density were included within the model and showed that site was the most significant influence on changes in invertebrate abundance between sample dates, however, flower density was also recorded as a significant factor at the beginning (20 June) and end (30 September) of the investigation (Table 4.3).

**Table 4.3:** Summary of F and P values for the effects of flower density and site on pollinator community composition via RDA analysis. Significant results are highlighted in **bold**.

Date assessed	F value Plants	P value Plants	F value Sites	P value sites
20th June	2.5967	<b>0.048</b>	5.3583	<b>0.001</b>
4th July	0.6692	0.789	3.7737	<b>0.001</b>
17th July	0.4266	0.948	1.4104	0.121
1st Aug	0.4928	0.892	1.4193	0.140
14th Aug	1.0206	0.427	1.7367	<b>0.049</b>
30th Aug	1.5230	0.130	3.4462	<b>0.002</b>
17th Sept	0.5321	0.699	2.7462	<b>0.018</b>
30th Sept	3.7663	<b>0.036</b>	2.9313	<b>0.039</b>

#### 4.4.2 Success of management at Alcan compared to other management techniques and sites of interest.

##### 4.4.2.1 Total invertebrate abundance and diversity

To determine the success of the management at each site, the total invertebrate abundance and Shannon diversity were compared (Figure 4.3). Likelihood ratio tests (LRT) were undertaken to determine the significance of effects of individual parameters on invertebrate abundance. Both site and date surveyed significantly influenced invertebrate abundance (GLM, LRT = 324.27, d.f. = 4,  $P < 0.001$ , and GLM, LRT = 598.08, d.f. = 7,  $P < 0.001$ , respectively). Tukeys HSD comparisons revealed that the two seeded sites (Fallowfield, and Hartford Bridge) had a significantly lower invertebrate activity compared to Alcan and the two naturally managed sites (Brenkley Colliery and Potland Burn) (Figure 3a), and highlighted the variation in invertebrate abundance between survey dates (Table 4). The Shannon diversity of invertebrates recorded was also significant between sites (Kruskal-Wallis,  $\chi^2 = 19.65$ , d.f. = 4,  $P < 0.001$ ). Pairwise Wilcoxon tests showed that Fallowfield (one of the seeded sites) had the lowest diversity, compared to all other sites (Figure 4.3b).

The parameters site and survey date also had a significant effect on flower density (GLM, LRT = 187.15, d.f. = 4,  $P < 0.001$ , and GLM, LRT = 600.27, d.f. = 7,  $P < 0.001$ , respectively). Tukeys HSD comparisons showed that Alcan had a significantly higher total flower density compared to all other sites (Figure 4.4a) and flower density changed significantly between sampling dates (Table 4.5). Plant species diversity was also significantly different between sites (Kruskal-Wallis,  $\chi^2 = 17.36$ , d.f. = 4,  $P < 0.01$ ). Pairwise Wilcoxon tests showed Alcan to have the highest overall diversity of plant species and Fallowfield the lowest (Figure 4.4b).



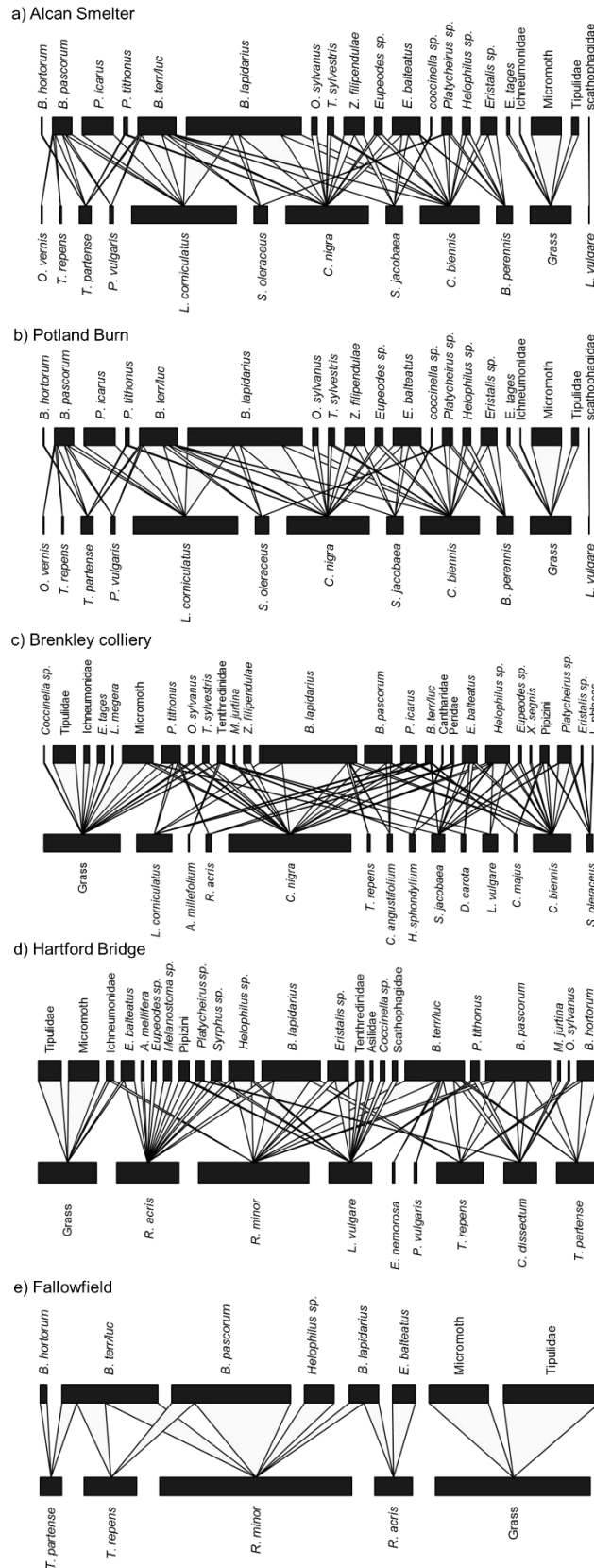




#### **4.4.2.2 Ecological networks**

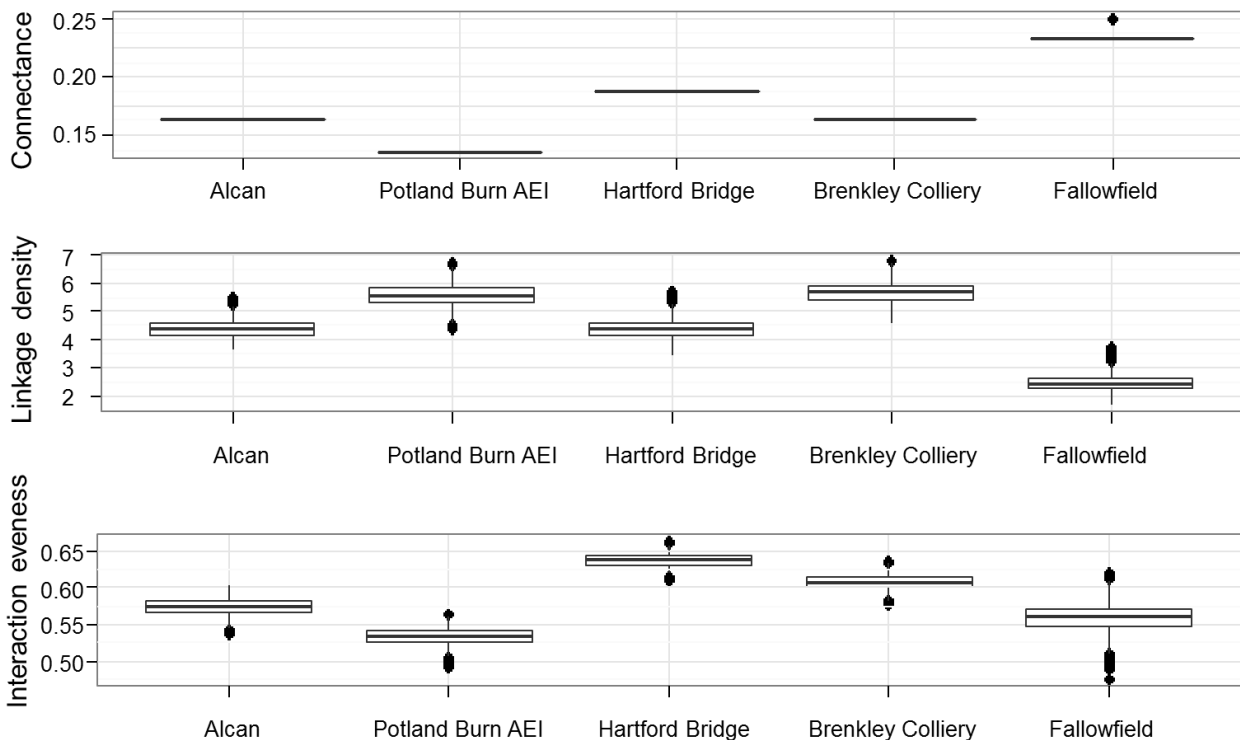
Bipartite interaction matrices were produced for each site (Figure 4.5a-e), allowing visualisation of interactions between trophic levels, the preferences of different invertebrates, and the habitat structure on each site. Most strikingly there is a distinct reduction in the number of higher and lower trophic level species present at Fallowfield (Figure 4.5e), and compared to the other sites.

Potland burn (Figure 4.5b) shows that a number of invertebrate species foraged on *Centaurea nigra*, whereas Alcan, Brenkley colliery, and Hartford Bridge (Figure 4.5a-d) show a broader range of forage species.



**Figure 4.5:** The ecological networks at the five field sites a) Alcan Smelter, b) Potland Burn, c) Brenkley Colliery, d) Hartford Bridge, and e) Fallowfield. The black blocks represent the participants, and the downward arrows indicate links between higher and lower level species. The arrow size is proportional to interaction strength (Appendix 3. Table 3.1 contains a full species list).

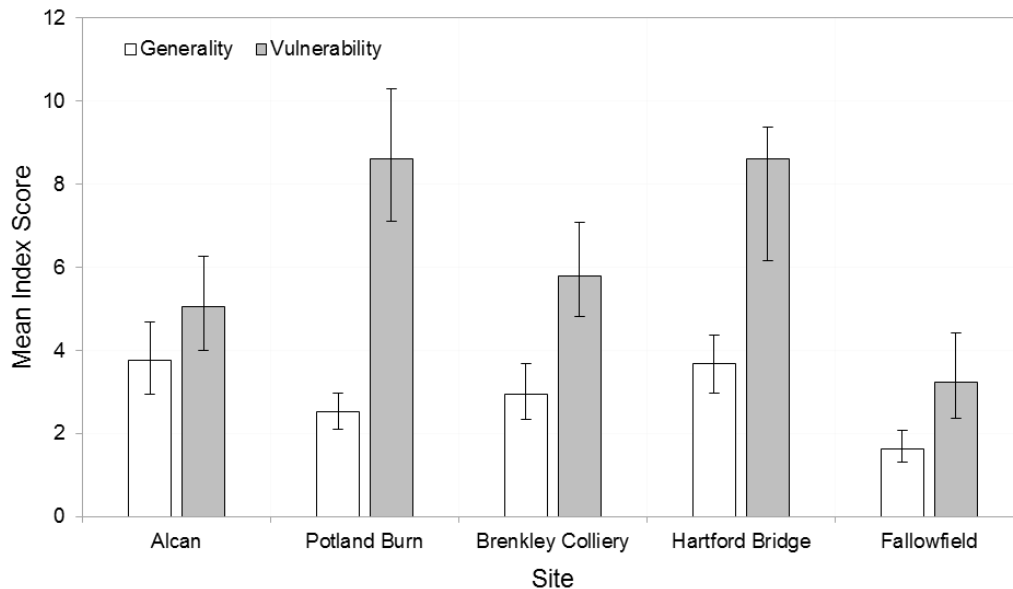
A series of indices were calculated to describe the ecological networks, at the network and species level. Three indices were used, connectance, linkage density and interaction evenness (Figure 4.6).



**Figure 4.6:** Boxplots showing the three ecological network descriptors, connectance , linkage density, and interaction evenness, for each of the four sites following network visualisation. The box plot is constructed from the median, and upper and lower quartiles. Whiskers show the maximum and minimum data points recorded, excluding any outliers. Outliers in the data are identified by black dots.

Connectance was less than 0.30 for all sites, likely a result of forbidden links and a low number of interactions within the flower-visiting invertebrates recorded. There were no significant differences between sites and this factor (Kruskal-Wallis  $\chi^2 = 11.45$ ,  $P = 0.32$ ), however, Fallowfield had the highest connectance, whereas Potland Burn had the lowest (Figure 4.6). There was no significant difference between Linkage density between sites (Kruskal-Wallis,  $\chi^2 = 8.24$ ,  $P = 0.61$ ) with Hartford Bridge and Potland Burn scoring higher than Alcan, but Fallowfield considerably lower (Figure 4.6). The sites were comparable for interaction evenness with scores between 0.45 and 0.7, Hartford Bridge was at the top of the range (Figure 4.6) but again these differences were not significant (Kruskal-Wallis,  $\chi^2 = 12.41$ ,  $P = 0.26$ ).

Out of the five sites Fallowfield and Alcan had the closest generality and vulnerability scores (Figure 4.7), indicating a stable plant and invertebrate population. Index scores at Hartford Bridge and Potland Burn however, were considerably further apart suggesting higher specialism by invertebrate foragers on site



**Figure 4.7:** Bargraph showing the generality of the higher trophic level species (Flower-visiting invertebrates) compared to the vulnerability of the lower trophic level species (Flowering species) of the five surveyed sites. The indices were calculated using the bipartite package within R. Errorbars show 95% confidence intervals following vaznull model with 1000 permutations.

**4.4.2.3 Determine the most valuable plant species in this area for flower-visiting invertebrates to provide future management guidance to local authorities and businesses aiming to improve biodiversity.**

Invertebrate foragers at Alcan and Fallowfield did not have a single dominant food plant species. The other three sites, Potland Burn, Brenkley Colliery, and Hartford Bridge, all showed strong preference to individual species (Table 4.6). Species strength scores calculated within the ‘specieslevel’ function of the ‘Biparite’ package show *Centaurea nigra* was a species of importance at Potland Burn and Brenkley Colliery, and at Hartford Bridge *Ranunculus acris* and *Leucanthemum vulgare* were also of importance (Table 4.6a).

**Table 4.6:** a) Heat map displaying the species strength scores of the lower trophic level (Plants), and b) Heat map displaying the species strength scores of the higher trophic level (Invertebrates). Species with the highest scores at each site were selected and have been displayed below showing the most important species per site. This list is not exhaustive, additional plants were recorded across the sites, however only the species scoring greater than 1.0 were included.

Key	0	0.1-1	1.1-2	2.1-3	3.1-4	4.1-5	>5
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a)	Alcan	Potland Burn	Brenkley Colliery	Hartford Bridge	Fallowfield
<i>Bellis perennis</i>	1.29	0.00	0.00	0.00	0.00
<i>Crepis biennis</i>	3.10	3.37	4.11	0.00	0.00
<i>Centaurea nigra</i>	4.11	8.31	6.23	0.00	0.00
<i>Lotus corniculatus</i>	2.26	0.01	1.24	0.00	0.00
<i>Trifolium pratense</i>	1.78	1.11	0.00	1.15	1.15
<i>Succisa pratensis</i>	0.00	2.85	0.00	0.00	0.00
<i>Ranunculus arcis</i>	0.00	2.08	0.00	6.13	1.50
<i>Rhinanthus minor</i>	0.00	0.00	0.40	3.36	2.85
<i>H. sphondylium</i>	0.00	0.00	1.39	0.00	0.00
<i>Senecio jacobaea</i>	1.70	0.00	1.65	0.00	0.00
<i>Sonchus oleraceus</i>	0.42	0.00	1.81	0.00	0.00
<i>Cirsium dissectum</i>	0.00	1.00	0.00	3.33	0.00
<i>L. vulgare</i>	1.00	0.25	0.75	5.48	0.00
<i>D. carota</i>	0.00	1.00	0.42	0.00	0.00

b)	Alcan	Potland Burn	Brenkley Colliery	Hartford Bridge	Fallowfield
<i>B. lapidarius</i>	1.75	2.35	2.54	0.75	0.47
<i>B. pascorum</i>	3.20	2.17	1.00	1.46	0.93
<i>B. terr/luc</i>	1.42	2.04	0.94	2.81	1.50
<i>Empididae</i>	1.00	1.00	0.00	0.00	0.00
<i>Ichneumonidae</i>	0.02	1.00	0.65	0.09	0.00
<i>Syrphus sp.</i>	1.13	1.13	0.00	0.20	0.00
<i>E. balteatus</i>	0.74	0.23	1.71	0.21	0.60
<i>Helophilus sp.</i>	0.34	0.59	1.36	0.30	0.15
<i>Tenthredinidae</i>	0.00	0.10	1.85	0.14	0.00
<i>Scathophagidae</i>	1.00	0.00	0.00	0.12	0.00

## 4.5 Discussion

This study assessed and compared the success of different grassland management techniques at five different sites within Northumberland, UK. The study was novel in its approach as it assessed the response of flower density and invertebrate abundance to different grassland management techniques, allowing us to consider the successes and problems faced by each method. The seeded and natural regeneration sites were compared and contrasted to the Alcan smelter, highlighting management concerns, and identifying flowering species which could further enhance the Alcan smelter or grassland of similar composition in the local area.

At the seeded sites, Hartford Bridge in particular, flower density was high at the beginning of the sampling period, however low flower density was recorded from June until the end of the flowering season. Peak activity of flower-visiting invertebrates occurs between May and September (Pollard and Yates 1993), therefore low flower density could have an adverse effect on invertebrate populations. The high abundance of *Rhinanthus minor* early in the season, and absence of *C. nigra* in late summer explains this trend compared to the other sites, however, *C. nigra* and other flowering species had previously been recorded on the site. No management had been employed to control the dominance of *R. minor*, therefore as beneficial as this species can be for suppressing grass species to allow growth of dicotyledon species (Westbury and Davies 2005), the sheer density on this site could have had a negative effect on seedling germination. Gibson et al. (1992) recorded a similar effect of *R. minor* dominance, emphasising the importance of the correct balance of flowering species within created seedmixes, possibly to the extent of seedmix creation for each individual habitat and soiltype. Despite being planted with the same flowering seedmix at the same time of year, the diversity and density of species recorded were much higher at Hartford bridge compared to Fallowfield. The sward recorded at Fallowfield was dominated by amenity grassland species, which could be preventing seedling germination. Continued management to control grass dominance could easily improve the flowering species diversity (López-Mariño et al. 2000, Noordijk et al. 2009). In fact, Fallowfield could benefit from a higher density of *R. minor* (Davies et al. 1997), although success of grass suppression can be dependent on the species *R. minor* is exposed to (Westbury et al. 2006).

Grassland sites require management as a response to flowering stage, weather conditions, and the specific aim of the management on site; therefore when grassland

is cut or grazed too early or too late, the flower density and invertebrate abundance can be affected for the remainder of that season. Hartford Bridge was scheduled for cutting after the first flowering period. If the site had been monitored, the mowing schedule could have been amended to reduce the amount of *R. minor* seeds setting and falling back to the soil. Mowing was omitted at Hartford Bridge during the trial period, allowing the site to become dominated by tall grasses and Apiaceae stems from previous years. *Cirsium* sp. were the only recorded floral resource for flower-visiting invertebrates between July and September, yet if the grassland had been mown once the first flowering period had finished it is likely that flower density would have improved later in the season, diversifying the sward in September (Chapter 2, Noordijk et al. 2009, Scriven et al. 2013). Although the long term grassland species composition may not be affected, management by mowing will continue to improve the sward structure and prevent grass dominance and dicotyledon suppression (Chapter 2).

In contrast, although Fallowfield was mechanically mown the cut was undertaken before the first flowering plants had set seed. The mowing had been scheduled for a particular date and time, and despite it not being the optimal time for the grassland to be cut, mowing was still undertaken. As the seeds of the sward had not fallen, a reduction of flower density is likely the following year affecting the success of the management scheme at this location. Cutting or grazing of grassland is essential, as evidenced by the problems at Hartford Bridge, but timing of the cut(s) is also important as described at Fallowfield; Therefore the key to success is ensuring long term management goals are enforced, and conducted at the appropriate time to be beneficial for flowering communities.

The two semi-natural grasslands were managed in different ways, and it is important to consider the evidence from both. Potland Burn, which was dominated by *S. pratensis* and *C. nigra* was grazed over winter with the cattle removed during summer, and Brenkley colliery was left unmanaged. Cattle grazing at Potland burn provides a good cut back of grasses to allow regeneration of many flowering species to occur in the subsequent year, and furthermore reduces the need for mechanical mowing, which has been shown to have devastating effects on ground dwelling invertebrates (Humbert et al. 2010, Buri et al. 2013). Furthermore the cattle trample the site allowing gap recolonisation, and assist with seed dispersal, providing vital functions to enhance grassland diversity (Vickery et al. 2001, Rook and Tallowin 2003). Potland Burn had



low flower density at the start of the trial, improving throughout the season, both *C.nigra* and *S.pratensis* are late flowerers explaining this difference, however habitat to encourage early foragers and support nest seeking queen bees, could prove beneficial. In contrast, Brenkley Colliery supported a large number of plant species, which hosted a diverse selection of invertebrates across the survey period, despite the site being unmanaged. As with Alcan, all flowering plants at Brenkley Colliery had been allowed to germinate from natural colonisations, however a single cut management regime could be introduced to maintain flower density and diversity in the future.

Evaluation of the ecological networks were undertaken to represent the biotic interactions at each of the sample sites. In which Higher and lower trophic species are connected by pairwise interactions (links), allowing comparison and description of real ecosystems and also to investigate the effects of network properties on network models. Ecological networks provide characterisation of the prominent patterns within species matrices, but cannot provide a complete inventory of all possible interactions, merely a subset of the total number of higher level species interacting with a subset of lower level species (Dormann et al. 2015). The connectance (proportion of possible interactions which occur) within the ecological networks was low for all sites (less than 3), which is considered usual for plant pollinator networks, where communities appear to show a higher degree of specialism, and the maximum number of interactions is impossible as a result of forbidden links (Bosch et al. 2009, Dormann et al 2015). Forbidden links are unobservable in nature, and cannot be prevented by increased sampling, simply they are restrictions imposed by the species life history, particularly relevant when describing invertebrates where difference life stages occupy different habitat niches for survival. Reasons for forbidden links can be from unrecorded interactions, and from low probability of interspecific encounter for potential pairwise interactions. To truly evaluate the completeness of ecological interactions, a vast understanding of natural history is required to discount forbidden links irrespectively of sampling effort (Dormann et al. 2015)

In terms of habitat stability, Fallowfield and Alcan had the smallest gap between the generality and vulnerability scores, and therefore were identified as the most stable networks out of the five sites (Morris et al. 2014). Both Fallowfield and Alcan provided flowering plants for generalist invertebrates across the season, despite the density and

diversity of flowering plants being lower at Fallowfield. When considering a site such as Potland Burn or Hartford bridge, the number of plant species foraged on by invertebrates was time limited, putting a considerably higher vulnerability value on the lower levels (plants), emphasising the importance of studying a habitat over time to determine the habitat quality and flowering longevity for flower-visiting invertebrates.

To facilitate and maintain habitat suitability for flower-visiting invertebrates, it is important to encourage flowering density as well as diversity (Chapter 2, Scriven et al. 2013). The density of flowers attracts the invertebrates, and the diversity of plant species provides forage for different taxa. The indicator species of importance revealed with the ecological networks for the five study sites supported the theory that invertebrate diversity can be influenced by the diversity of flowering plants available for forage (Comba et al. 1999, Stang et al. 2006). The species strength scores indicated that *C. nigra*, *R. acris*, and *R. minor* were all dominant flowering species, and attracted a higher diversity of invertebrates. However, their flowering availability during the season was limited to short periods, showing the need of additional plant diversity onsite. Potland Burn, had a high density of *C. nigra* available for forage by bumblebees over a two week period, however did not have a diverse flower assemblage, or sufficient flowering period, at a high enough density to support other invertebrates. A similar problem was recorded at Hartford Bridge with the density of *R. minor*, however, although Hartford Bridge did have a diverse community, the flowering longevity was not enough to provide a stable habitat. Nevertheless, the strong species scores associated with these species emphasises the attraction power certain flowering species can have on flower-visiting invertebrates; identifying additional species which could be introduced within the Alcan site to improve flower diversity and density throughout the summer period.

It is acknowledged that within this trial, the ecological networks were utilised to visualise the key drivers in the results observed and to establish the strength of species within the networks regarding interactions between adult invertebrates and flowering plants. Full comprehension of the complexities of network ecology were not required for this study, however the inclusion of an understanding of specific invertebrate life histories and increasing sampling to observe zoo-centric (e.g pollen on bees) and phyto centric (Species which visit plants) interactions would improve the discovery rate of interactions and provide a more accurate insight to the species interactions, providing a more robust and comprehensive tool for ecological management planning.

## **4.6 Conclusions**

Compared to sites in the local area, the Alcan smelter has a diverse network of lower and higher trophic level species. The flowering period is currently a good length to allow forage for flower-visiting invertebrate, and hosts a diverse range of flowering plants. The site is statistically comparable to local nature reserve grasslands and outperforms sites seeded with a wildflower mix in relation to total abundance and diversity of plant and invertebrates recorded. This study underpins the significance of the reduction of mechanical mowing on site, and the resulting positive effect on biological diversity, providing evidence based results to guide future ecological management plans for use by local authorities to manage public open space and to planners to guide future landscape and management plans for development.

### **4.6.1 Future management recommendations**

From the analysis conducted, the Alcan site could benefit from the addition of some of the species highlighted in Table 4.6; *R. arcis*, *S. pratensis* and additional *L. vulgare*. These three species were heavily utilised by flower-visiting invertebrates, would add structural diversity, and lengthen the foraging period on site. Short flowering periods are common problem recorded within grassland sites, as bumblebees in particular require early and late flowering forage for queens (Memmott et al. 2010).

## Chapter 5

### Methods to value and enhance understorey vegetation within plantation woodland

#### 5.1 Abstract

Woodland trees can limit the light radiating to the understorey ground flora, reducing the species diversity and flower density available to foraging invertebrates. To increase overall invertebrate diversity at a site, improving the floral diversity of these habitats could offer a cost effective way of managing invertebrate populations. Clearing areas of trees to allow more light to pass through, should in theory increase the understorey and thus increase the number and diversity of flower-visiting invertebrates. However, this does depend on the availability of a seedbank in the first place. In an assessment of the seedbank in a plantation woodland at the Alcan Smelter in Northumberland, the seedbank composition at three soil depths was found to be similar, and the majority of species that germinated were similar to those already growing at the site. Therefore, at this site, the simple management strategy of allowing regeneration of the seedbank by removing the top layer of soil (to remove dominant weeds and current above ground flora), and increasing light levels would not improve the floral diversity of the woodland. As an alternative the effect of different light levels on seed mix germination success was undertaken within replicated seedtrays. At the current light levels recorded at the site, there was significantly less germination from the mixture in comparison to light levels found at the edge and ride areas of the woodland.

## 5.2 Introduction

Woodland was once a dominant habitat type throughout the UK, however a large proportion has been cleared since Neolithic times allowing development of agricultural practices and villages, as well as providing resources for building materials and fuel (Woodbridge et al. 2012). Remaining woodland still covers approximately 8.4% of the UK (Smith and Gilbert 2001), and this value is increasing as a result of farmland reversions through agri-environment schemes (Wilson and Hart 2001), and restoration of native woodlands (Harmer et al. 2010). Woodland habitats can have a high ecological value (Fry and Sarlöv-Herlin 1997, Hopkins and Kirby 2007), supporting biologically diverse communities including fungi, lichen, wildflowers, invertebrates and large mammals. It is the combination of microhabitats in the form of canopy cover, scrub, herb layer, fungi and deadwood which allows woodland to support such diverse assemblages (Hopkins and Kirby 2007).

Ancient woodlands, classed as any continually managed woodland predating 1600AD, host the highest biological diversity of all woodlands (Spencer and Kirby 1992, Alexander et al. 1998). Characterised by ancient and veteran trees, diverse ground flora, scrub layer and fallen deadwood, many ancient and semi-natural woodlands have been protected as Sites of Special Scientific Interest and Biodiversity Action Plan habitats (Hopkins and Kirby 2007). In contrast, plantation woodlands, often a result of woodland planting projects for land regeneration and wildlife, or solely for the production and harvest of timber are also dotted across the landscape. These woodlands, are made up of evenly spaced and aged trees, which often results in little to no understorey vegetation, and often the tree species are non-native, which provides limited connectivity within the current landscape (Godefroid et al. 2006, Anderson 1996) devaluing this particular type of woodland habitat.

The woodland at the Alcan smelter, Lynemouth UK was planted during the 1980's to provide a screen to the smelter site. This site has become dominated by the non-native fast growing *Populus nigra 'Italica'*, of which the long-term suitability within woodland is questionable; *P. nigra 'italica'* have a shorter lifespan than native slower growing trees, can be easily uprooted in windy conditions, and are more susceptible to disease (Wood 1994). Although other tree species such as *Acer pseudoplatanus*, *Sorbus aria*, and *Betula pendula* were also planted, they are poorly established, possibly because of competition with the *P. nigra 'italica'* stands, resulting in a dense tall canopy which prevents light reaching the understorey. Combined with a dense leaf litter and dense

cover of *Urtica dioica*, *Ranunculus repens* and *Rubus fruticosus*, the establishment of a diverse scrub layer and understorey sward has been prevented, which limits the value of the woodland for biodiversity. Although this site was identified as being an important habitat for parasitic invertebrate species (Chapter 2, Scriven et al. 2013), diversification of the understorey flora and improving structural diversity could substantially improve the overall biodiversity (Fuentes-Montemayor et al. 2012, Graham et al. 2014, Irwin et al. 2014).

Clear felling, tree removal over an extended period, litter clearance, the planting of native trees and scrub, and wildflower seed planting are all possibilities for restoring biodiversity in a woodland understorey (Harmer et al. 1997, Harmer et al. 2010, Stanturf et al. 2014, Atkinson et al. 2015). However, before undertaking any such management or mitigation strategy understanding the scope of what is contained within the current seedbank may significantly reduce the need for expensive and time demanding management schemes. Such information can reveal historical plant species composition on any given site (McDonald et al. 1996), and in some cases allow natural regeneration once the right conditions are met. Such assessments have been used in agricultural settings to determine weed population dynamics within crops (Mayor and Dessaint 1998, Menalled et al. 2001), and by botanists wanting to understand the effects of soil disturbance on flower abundance. Furthermore they can successfully guide habitat restoration by identifying once dominant species which have been suppressed by previously unfavourable conditions (Bossuyt and Hermy 2003, Erenler et al. 2010, Gaujour et al. 2011, Kalamees et al. 2012). In natural seedbanks, any number of dormant seeds may be present, the viability of which can vary from days to decades, and species not currently found in above ground flowering plant surveys have routinely been shown to be present (McDonald et al. 1996, Cohen et al. 2004). Erenler et al. (2010) studied understorey species richness within woodland, and discovered that within Plantation on Ancient woodland sites diverse seedbanks of ancient woodland plant indicators species were present, despite the significantly reduced above ground flora diversity.

In this study we aim to develop an evidence based evaluation of habitat quality to inform a management strategy for plantation woodland located at the Alcan smelter site, Northumberland, UK. Specifically, two hypotheses are to be investigated further i) Seed diversity will be significantly higher in samples collected from 10-15cm depth compared to the surface layer (0-5cm), and ii) light levels representative of the ride

habitat will significantly increase germination rate and species diversity of a shaded woodland mix within a replicated trial.

### **5.3 Study design**

#### **5.3.1 Study Site**

Within the grounds of the Rio Tinto Alcan Aluminium smelter, Lynemouth, UK, a woodland plantation was established during the 1980's. The woodland, located along the South West side of the smelter provides a screen from the main road, and any surrounding residential properties. As a result of the close planting proximity, a dense canopy has developed providing less than 5% light penetrating to the understorey. There is no tree understorey or shrub layer within the woodland, and the ground flora is dominated by pernicious weeds such as *Urtica dioica*, and *Rubus fruticosus* providing a limited resource for invertebrate species. Within the grassland ride however, flowering grassland species are present indicating that the seedbank may contained dicotyledonous species currently constrained by unfavourable conditions.

#### **5.3.2 Seedbank Study**

A trial to establish the seedbank viability within the woodland was conducted during spring 2012. Soil samples were collected during the last week of March (to allow for natural vernalisation of seeds within the soil prior to collection), from sample pits positioned at 50 metre intervals along three 150 metre transects. Transect locations were identified using OS mapping and were positioned across the centre of the woodland running approximately from east to west. The use of transects allowed us to identify sampling points from mapping software prior to collection to prevent sampling bias, and allowed consideration of differences in seed density and microclimate throughout the woodland (Van der Valk et al. 1992).

Each sampling pit, positioned at 0m, 50m, 100m, and 150 metres along each transect, measured 1 m<sup>2</sup>. From each sampling pit, three soil samples of 750ml were taken using a hand trowel extracting the soil from the edges of the pit at three depths, 0-5 cm, 5-10 cm, and 10-15 cm (Warr et al. 1994) A total of 36 soil samples were collected from the woodland understorey, providing four replicates for each of the three sample

depths, and taken to the Newcastle University Field Station, Northumberland, UK for further analysis.

To prepare the soil for the trial, each sample was passed through a 1-cm sieve to remove stones and grass roots. The soil sample was then spread on a layer of sand and John Innes no. 2 compost in a ratio for 1:1 (approximately 3cm deep), within standard seed trays (37 cm x 23 cm x 5 cm) and lightly mixed. Trays of sand and compost were intermixed with the experiment to provide controls and determine effects from windborne seeds.

Samples were watered regularly to maintain moisture in the soil, and the trays were protected from slug damage. Once germinated, seedlings were identified (Rose and O'Reilly 2006, Poland and Clement 2009), counted and removed. Any seedlings which were difficult to identify were potted on and allowed to grow to aid identification. Trays were monitored for 18 months, until all seedlings had emerged. Grass and rush species were not counted as part of the trial because we wanted to establish the seed bank quality of flowering plants for the benefit of flower-visiting invertebrates.

### **5.3.3 Seedling emergence trial**

To establish the efficacy of a seed mix application to the understorey as a management technique to enhance biodiversity, a shading and emergence trial was undertaken to determine the rate of clearance required to establish ground cover. Standard seed trays (37 cm x 23 cm x 5 cm) filled with 1:1 measures of John Innes no 2 compost, and top-soil collected from the field site were used to grow a "woodland and shaded areas" seed mix supplied by Wildflower Lawns and Meadows Ltd, East Sussex (34 native wildflower species (30% wildflower to grass seeds). During September 2013, the mix was applied evenly to seedtrays at a rate of 3 g per m<sup>2</sup>, and then each tray was subjected to one of the three shading regimes, Central Canopy (T1), Ride (T2) and Edge (T3) (Table 5.1), using shading mesh supplied by a horticultural supplier. Metal stakes were used to support mesh over each individual tray at a height of 30-40 cm. The amount of mesh per tray was calculated using a light meter to replicate the percent shading recorded under the woodland canopy (Table 5.1).

Each treatment was replicated 5 times for statistical robustness. The 15 trays (treatments n=3, replicates n=5) were positioned within a randomised block formation, located and maintained at Cockle Park farm, Morpeth, UK. All trays were treated with Metaldehyde to protect from slug damage, and watered regularly to maintain soil



moisture. If grass species became too dominant, the sward was cut back using scissors, avoiding any emerged seedlings present

Once germinated, the abundance and species of seedlings was recorded once a month until August 2014, creating 11 datapoints per treatment (Data collection was postponed within February due to adverse weather conditions). On each visit, emerged seedlings were identified and removed from the tray, If identification was not possible, the seedlings were potted on to allow development to stage where a confident identification could be achieved.

#### **5.3.4 Statistical analysis**

All data analysis was completed within the R program (R Core Team 2013). The abundance and Shannon Weiner Diversity index of seedlings were calculated during both the seed bank trial and germination trial. The samples collected for the seedbank trial, and the different germination regimes were then compared using either an Analysis of Variance (ANOVA), or Kruskal-Wallis depending on whether the data conformed to normality and heterogeneity of variance. Post hoc tests were undertaken to identify differences between regimes when a significant effect had been observed. Differences between treatments were highlighted by Pairwise Wilcoxon tests (P-adjusted, Bonferonni) when Kruskal-Wallis had been undertaken, and Tukey's Higher Significant Difference (HSD) tests on ANOVA results.

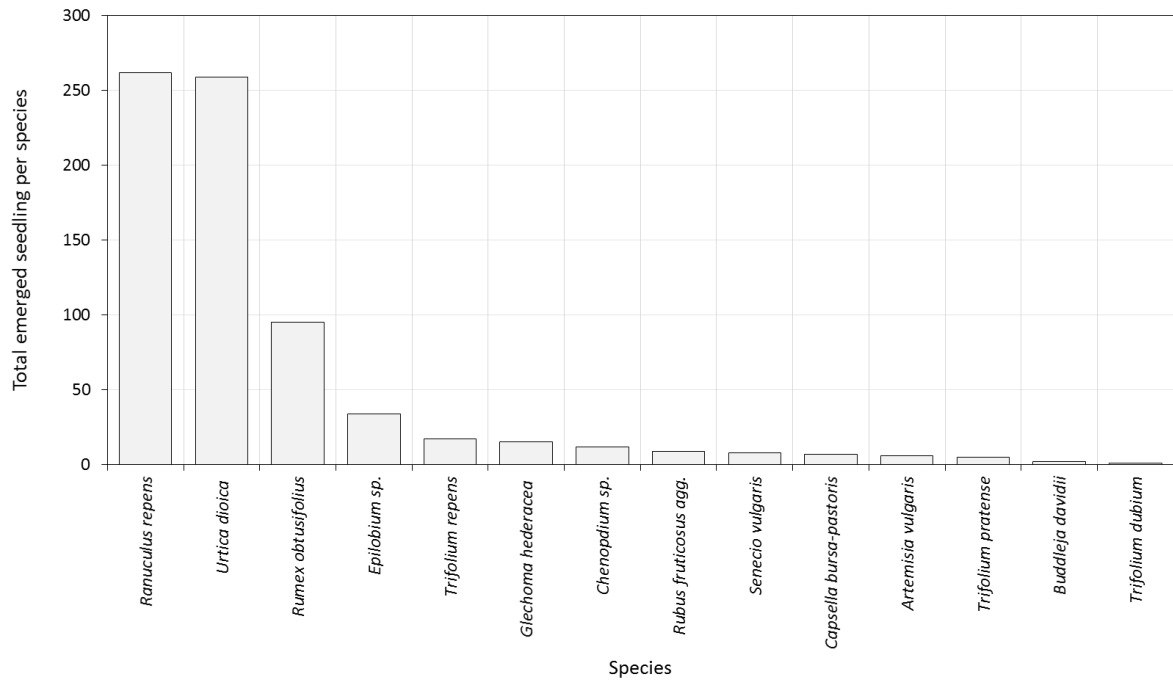
**Table 5.1:** The three treatment regimes used within the germination trial. The average light reading (of ten readings) for each location within the woodland, and the average light reading (ten readings) within the seed trays after the shading mesh had been applied. The Standard Error of the Mean (SEM, n=10) has been present by each average.

Treatment	Representative woodland location	Average light readings within the woodland ( $\text{mmol m}^{-2}\text{S}^{-1}$ )	Average light readings for each treatment ( $\text{mmol m}^{-2}\text{S}^{-1}$ )	Percent light penetrating the seed trays
<b>Open space reading</b>		<b>1000</b>	<b>850</b>	<b>100%</b>
T1	<b>Central</b> dense cover	10 ( $\pm 0.62$ )	14 ( $\pm 0.69$ )	<5%
T2	<b>Ride</b> patchy cover	168 ( $\pm 1.93$ )	152 ( $\pm 1.94$ )	15-20%
T3	<b>Edge</b> light shading	400 ( $\pm 1.58$ )	378 ( $\pm 1.85$ )	40-45%

## 5.4 Results

### 5.4.1 Determining the current seed bank quality within the plantation woodland.

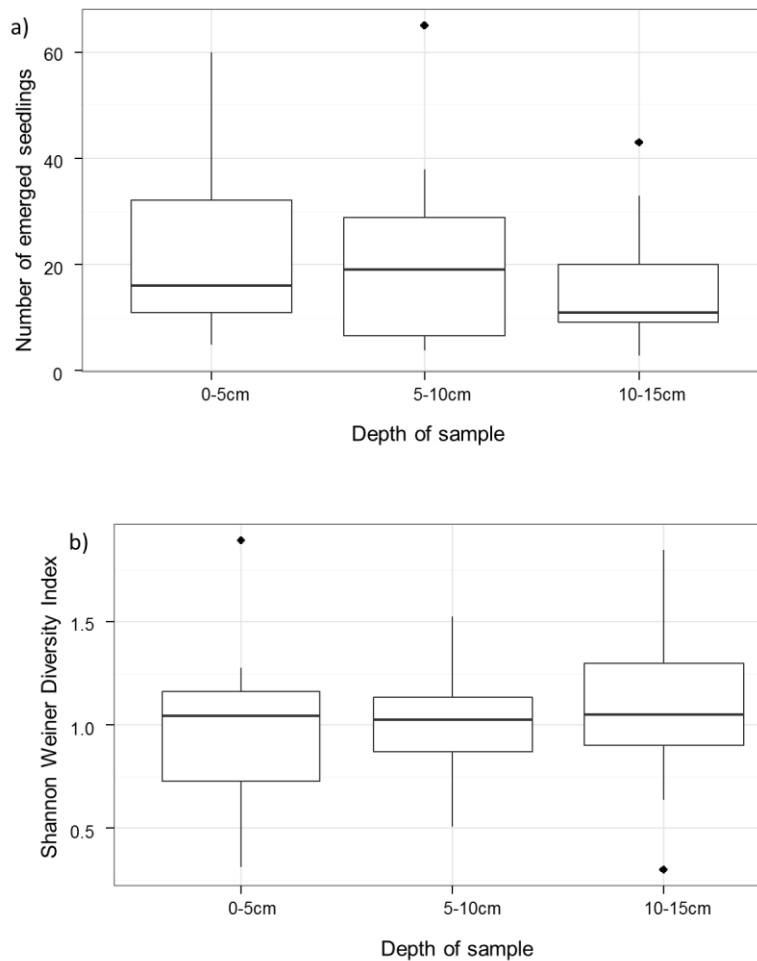
A total of 726 herb seedlings emerged within the 36 trays over the 18 month trial. However, only 14 flowering species were recorded of which *R. repens* and *U. dioica* seedlings dominated, mirroring the above ground flora currently observed within the woodland (Figure 5.1).



**Figure 5.1:** Bar graph showing the total abundance of dicotyledon seedlings recorded over the 18 month seedbank study. The soil samples were collected at three depths (0-5 cm, 5-10 cm, and 10-15 cm) from the centre of the woodland at the Alcan Smelter, Northumberland UK.

**5.4.2 Investigating the effects of depth on natural seedbank composition.**

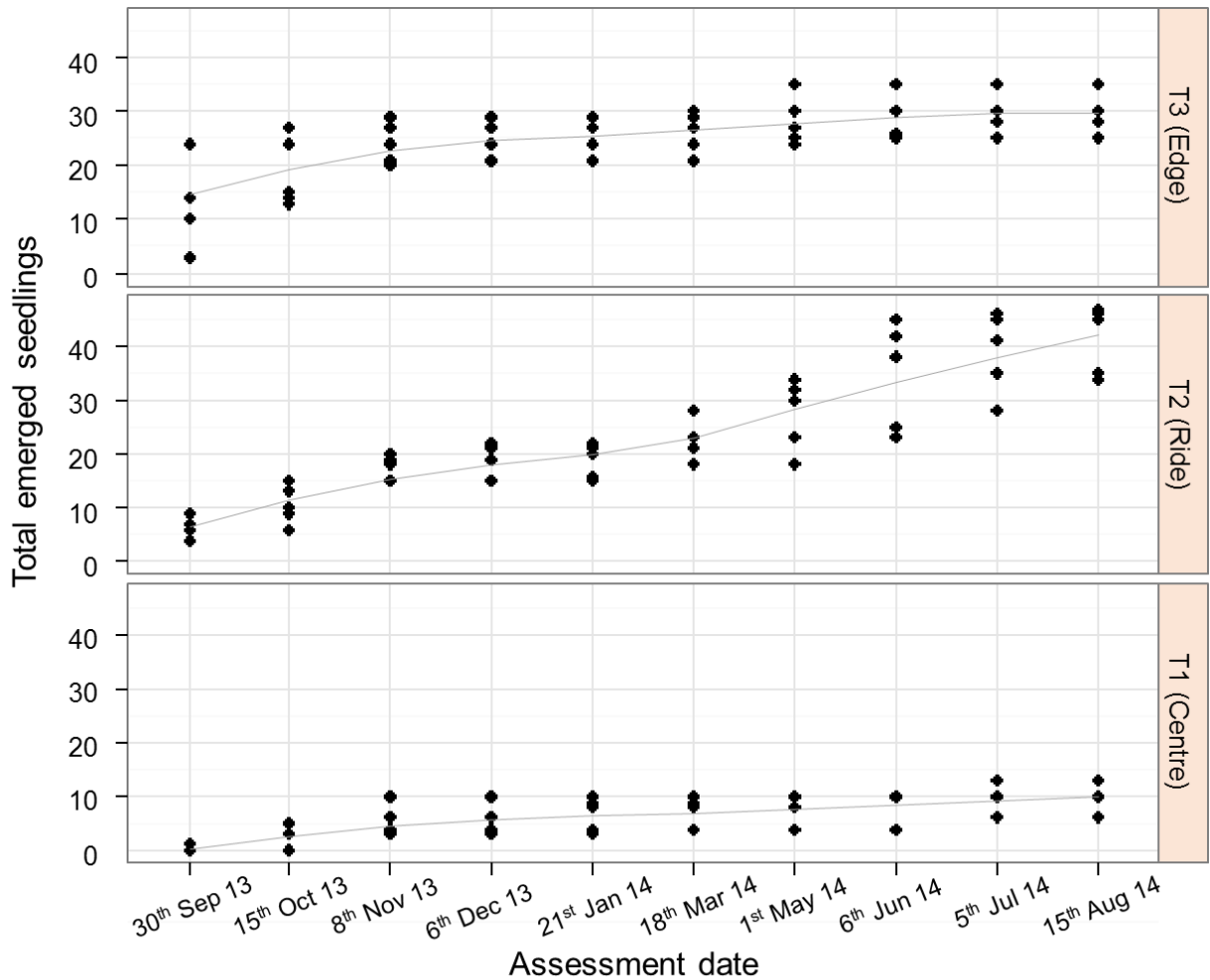
The seedling abundance within the soil samples collected decreased with depth (Figure 5.2a). Although there was a slight increase in Shannon-Weiner Diversity index recorded at 10-15 cm compared to both 0-5 cm and 5-10 cm (Figure 5.2b), these trends were not significant (Kruskal-Wallis,  $\chi^2 = 1.165$ , d.f =3,  $P = 0.558$ , and ANOVA,  $F = 0.051$ , d.f = 3,  $P = 0.951$ , respectively).



**Figure 5.2:** Whiskered boxplot showing; (a) seedlings per depth and (b) seedling diversity (Shannon Weiner Diversity) per depth. The box plot is constructed from the median, and upper and lower quartiles. Whiskers show the maximum and minimum data points recorded, excluding any outliers. Outliers within the data set are identified by black dots.

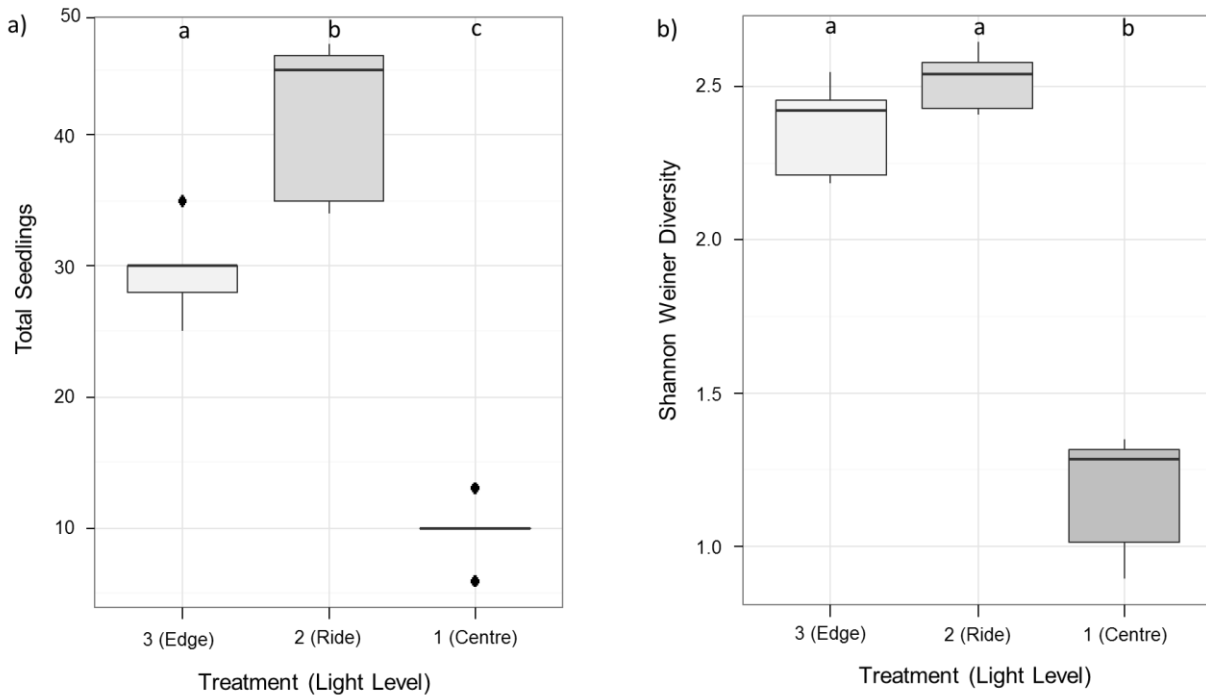
#### **5.4.3 Determining the effectiveness of a seeding regime within the plantation woodland.**

Germination of seedlings within seed trays was successful at all three light levels (Figure 5.3). However, the rate of germination fluctuated, with germination reduced between December and January. The seedling emergence within the ‘Edge’ treatments (T3) also appeared to slow in May, despite the other two treatments, ‘Ride’ (T2) and ‘Central’ (T1) showing an increase at this time.



**Figure 5.3:** Scatterplot with loess smooth curve (calculated within R package) showing the cumulative increase in total seedling emergence for each of the three light level treatments (T1, T2 and T3) over the 12 month germination trial.

Light levels had a significant effect on both the total seedling abundance (ANOVA,  $F=63.96$ ,  $d.f = 2$ ,  $P < 0.001$ ) and the Shannon Weiner Diversity Index (Kruskal-Wallis,  $\chi^2 = 10.22$ ,  $d.f = 2$ ,  $P < 0.01$ ) for each treatment. Following a Tukey's Higher Significant Difference (HSD), the Post hoc comparison tests showed that treatment 'T1' (Centre) showed a significant reduction in both germination rate (total seedlings) (Figure 5.4a), and Shannon Weiner Diversity (Figure 5.4b) in comparison to both treatment 'T3' (Edge) and 'T2' (Ride). Although the total number of germinated seedlings was significantly lower within T3 (Edge) compared to T2 (Ride), the Shannon Weiner Diversity Index was comparable between these treatments.



**Figure 5.4:** Box plot showing; a) total number of emerged seedlings and b) the Shannon Wiener Diversity Index for each light intensity treatment within the germination trial [T1 (Centre), T2 (Ride) and T3 (Edge)]. The box plot is constructed from the median, and upper and lower quartiles. Whiskers show the maximum and minimum data points recorded, excluding any outliers. Outliers within the data set are identified by black dots. Letters represent significant differences following; a) Tukey's HSD comparison tests and b) Pairwise Wilcoxon comparison test (P-adjusted, Bonferonni).

## 5.5 Discussion

This study assessed the quality of the seedbank available within the plantation woodland at the Alcan aluminium smelter, together with the germination success of a seedmix when applied under light levels representative of the current conditions. These results offer guidance towards possible management strategies for improving the understorey in this habitat, with the aim of increasing invertebrate diversity across the site. Although the study confirmed that the natural seedbank within the woodland would currently not be sufficient to diversify the ground flora; if light levels were increased, the application of a shaded woodland flower mix could significantly increase the biological value of the woodland ground flora.

The former land-use of a site can heavily influence the diversity within a woodland seedbank (Bossuyt and Hermy 2001). Within this study, the current ground flora within the open ride areas supported grassland species such as *Lotus corniculatus*,

*Dactylorhiza fuchsia* and *Trifolium pratense* suggesting that the former land-use could have been as meadow pasture, the seedbank of such pasture may well remain throughout the current plantation woodland. It was this assumption that guided the trial towards a management plan based on natural regeneration of the ground flora by manipulating the available levels of light reaching the understorey. However, results showed that the seedbank was dominated by *U. dioica*, *R. fruticosus* and *R. repens* and that a seedmix in addition to improved light conditions would therefore be needed to achieve a diverse understorey at this site. Although the seedbank was less diverse than expected, many factors can affect the accuracy of seedbank trials, including seed viability following long periods of dormancy (McDonald et al. 1996). Alternatively, the conditions provided within the seed trays themselves may not have been suitable for the growing conditions of all seed to germinate (Van der Valk et al. 1992, Bossuyt and Hermy 2001). Even if the study had shown a desirable seedbank composition, success of natural seedbank regeneration to a rich woodland ground flora from dormant seeds alone has previously been shown to be problematic (Bossuyt and Hermy 2001, Baeten et al. 2009). Interestingly, Saatkamp et al. (2009) questions the accuracy of all seed bank analysis for these reasons, despite the methodologies routine usage in current studies (Granstrom 1988, Cohen et al. 2004, Erenler et al. 2010).

The differences recorded between the species composition of the seedbank and the associated plant community can be influential to future management decisions, and can also indicate whether woodland or grassland have been planted over ancient woodland sites or heathlands for example. Granstrom et al. (1988) identified high densities of *Calluna vulgaris*, *Carex pilulifera* and *Juncus sp.* seedlings within soil samples collected at conifer woodlands in Sweden, indicating that the woodland was planted on open heathland. In this example and others (Erenler et al. 2010), the poor correspondence between above and below ground species composition can provide valuable evidence to justify ecological restoration. In this study, although the seedbank composition was not that which was predicted (based on the evidence in the surrounding ride areas), it was representative of the current understorey flora of the woodland, and as such the results are likely to be an accurate representation of the understorey vegetation which would develop if measures were undertaken to improve light levels within the woodland. It was for this reason that the application of a suitable seedmix would be advantageous.

Light penetration through the canopy is critical for understorey invertebrates and the respective flora (Sparks et al. 1996, Plue et al. 2013). To assess this at the study site, the effect light levels had on seedling germination was assessed. A greater abundance of flowering species were recorded within treatment T2 (Ride), which had a mid-range light level. Interestingly, it appeared that the abundance of grasses recorded within the 'T3' treatment (Edge) which had the highest light availability of all treatments, may have actually restricted wildflower germination. Therefore, it is important to ensure that light levels are dappled, managed through structured clearing and shrub planting to ensure that the highest germination rate can be achieved to allow diversification of the sward.

Finally, the plantation is located within an area dominated by agriculture; a restoration management plan here could support invertebrate populations, provide refuge for wildlife, and enhance connectivity between habitat parcels within the local landscape (Godefroid and Koedam 2003, Bailey 2007). The site remains considered as 'plantation woodland', however no timber production is associated with the site as the woodland was planted as a screen. Therefore phased removal of trees, and tree planting of native species in keeping with the local landscape could be successfully implemented to improve the structural composition, light levels and tree species diversity within the woodland (Lindenmayer and Hobbs 2004, Anderson 1996). This would in turn enhance the overall ecological value of the woodland within the surrounding landscape.

## **5.6 Conclusions**

The investigative trial undertaken at the Alcan Aluminium smelter introduces two methods of classifying the status of the woodland understorey to formulate a protocol to value woodland understorey habitat and prescribe further management options. Following assessment of the seedbank and light levels the woodland was considered to be of low ecological value to flower-visiting invertebrates, and considerable enhancements to the structure of the woodland (other than simple removal of trees and addition of seed mixes) would be required for an enhancement in biodiversity. Providing evidence backed reporting could be a method of establishing ecological management plans within habitat patches such as this, and further work to identify other factors which determine the ecological status of woodland habitats could help form an assessment and mitigation strategy.



### **5.6.1 Future management recommendations**

To improve the light levels penetrating the understorey, gradual tree felling would be an important aspect for future management within the woodland. The phased removal of approximately 50% of the hybrid poplar would be suggested to replicate light levels currently observed within the woodland ride. Removing blocks of unfavourable trees such as *P.nigra 'italica'* would create structural diversity and differing light levels across the site. The development of an additional ride would also aid structural diversity within the ground flora, encouraging the development of a scrub layer within the woodland. Native tree planting is recommended, however species would need to be representative of the local area, and suitable for the soil composition on site. The trees should be planted in staggered cohorts to increase age diversity, structural diversity and maintain light levels in the woodland. The development of a scrub layer should be encouraged including species such as *Ilex sp.*, *Corylus sp.*, and *Crataegus monogyna* to further improve the structural diversity within the woodland.

## Chapter 6

### **Importance of hedgerows for flower-visiting invertebrates: a comparison of species poor and species rich hedgerows**

#### **6.1 Abstract**

Hedgerows are historic structures, providing botanical diversity and heterogeneity in agricultural and urban dominated landscapes. Valuable habitat for birds, mammals and invertebrates, hedgerows provide linear structures allowing connectivity between habitats. The importance of hedge structure for flower-visiting invertebrates was assessed by recording invertebrate diversity found within pan traps located on six hedgerows in the area around an industrial site. Following a hedgerow survey to determine the number of woody native species within the hedgerows, three species poor and three species rich hedgerows were selected for further survey. Results show that there were significant differences in abundance, species richness and Shannon Weiner diversity between the different hedgerow types. A far greater richness and abundance was found in species rich hedgerows compared to those classed as species poor. This result strongly supports the need for planting/maintaining species rich hedges to aid in increasing flower-visiting invertebrates within specific areas and also allowing for connectivity to occur between other habitats.

## 6.2 Introduction

Hedgerows are important features within the agricultural landscape; enhancing structural heterogeneity, botanical diversity, and providing valuable breeding habitat for birds, small mammals and invertebrates (Burel 1996, Hinsley and Bellamy 2000, Gelling et al. 2007). Hedges evolved with agricultural practises, providing a method of containing livestock, identifying boundaries, and preventing soil erosion (Pollard et al. 1974). Now these landscape features provide valuable linear habitat for flora and fauna; creating a network of pathways across agricultural pastures, emphasising the importance of these features within a modified landscape. Bird and small mammal dependences have been positively highlighted (Tattersall et al. 2002, Whittingham et al. 2009, Boughey et al. 2011), however the value to flower-visiting invertebrates is not as well documented (Maudsley 2000). Studies targeting Hymenoptera and Lepidoptera dominate the literature despite being a small percentage of total invertebrates in agricultural environments (Dover and Sparks 2000, Maudsley 2000, Hannon and Sisk 2009), however Dover et al. (2000) also showed a positive relationship between hedgerows and butterfly abundance. Interestingly, some studies have also reported differences in invertebrate diversity and/or abundance between hedge types. For example, green lane hedges have been reported as having higher *Bombus sp* and Lepidoptera activity (Dover et al. 2000, Croxton et al. 2002) compared to arable hedges, a result which indicates that the surrounding vegetation may also be important in supporting invertebrates (Maudsley 2000).

The intensification of farming methods in the 20<sup>th</sup> century, and the ever increasing demand for higher crop production has resulted in the need to increase field area (Pollard et al. 1974, Petit et al. 2003). Dover et al. (2000) summarised that between 1946 and 1990, the UK alone had lost over 351,000km of hedgerows within the agricultural environment. A substantial amount of attention has since been directed to counteract this trend of biodiversity loss within agricultural environments; however, the focus has been towards the enhancement of wildflower margins within agricultural landscapes (Carvell et al. 2004, Pywell et al. 2005, Carvell et al. 2007). Limiting pesticide and herbicide use, restricting cutting regimes and providing approximately 2 metre strips of flower-rich native grassland along field margins are becoming a more common sight. These prescriptions are an option within UK agri-environment schemes, however their success has been the focus of much debate (Pywell et al. 2006, Whittingham 2006, Konvicka et al. 2007). Facilitating forage availability, these flower

rich margins have boosted invertebrate abundance within an area where large declines had previously been recorded (Meek et al. 2002, Carvell et al. 2004, Pywell et al. 2006). These flower margin enhancement schemes provide nectar and pollen rich plants for invertebrates, providing forage and improving invertebrate diversity in one location (Vaughan et al. 2007). They do not, however necessarily improve the overall habitat for invertebrates as other fundamental needs such as shelter and breeding sites which are not accounted for (Haaland et al. 2011). Furthermore some seed mixes currently contain non-native plant species which could affect foraging behaviour in pollinating species (Chittka and Schürkens 2001, Graves and Shapiro 2003). In contrast to flower rich margins, considerably less research has been focused on the importance of hedgerows with regard to biodiversity. Hedgerows, offer shelter, a food source and breeding sites addressing the fundamental needs of flower-visiting invertebrates, working together with a native flower margin increasing nectar resource, the combination could significantly improve invertebrate diversity throughout the countryside. Furthermore, one might also expect the hedge type to play an important role in diversity and abundance. Relict hedgerows are likely to support fewer invertebrate species than managed hedges, a result of reduced structural complexity and plant diversity (Maudsley 2000, Croxton et al. 2004). These studies emphasise the importance of maintenance and restoration of hedgerows, both on a structural and botanical level (Benton et al. 2003).

Within this study, six hedges were identified and monitored over the invertebrate flight period between May and September (Appendix 4, Figure A4.1). The project aim is to establish the value of two hedgerow types by assessing the invertebrate communities inhabiting, species rich and species poor hedgerows. Three hypotheses were formulated i) species rich hedgerows would support a significantly higher abundance of flower-visiting invertebrates than species poor hedgerows, ii) species rich hedgerows would support significantly higher species diversity of flower-visiting invertebrates compared to species poor hedgerows and iii) species rich hedgerows would host a significantly higher abundance of nectar feeding species and pollen collecting invertebrate guilds, than predator and parasitic guilds.

### 6.3 Study Design

A total of six hedgerows were selected from two specified groups (n=3, species poor and n=3, species rich, regarding their botanical assemblages). Within each hedgerow three sample points (Subsamples) were positioned 15 metres apart, each sample point (subsample) supported three pan traps (section 6.3.1). The hedgerows considered species poor (SP) were in the region of 40 years old and hedges considered species rich (SR), were planted under an agri-environment scheme within the last ten years. Typically, the species poor hedges (SP) were dominated by *Crataegus monogyna* (90%) with a small amount of *Fagus sylvatica* (5%), and *Rubus fruticosus* present (5%). The species rich hedges (SR) were also dominated by *C. monogyna* (60%), but also supported additional *Rosa canina* (5%), *Prunus spinosa* (30%), *Acer campestre* (5%) *F. sylvatica* (5%) and *Ilex aquifolium* (5%).

All hedgerows were approximately 1.5km apart to ensure they were independent from each other, and all hedges selected for the trial had no natural or artificial wildflower border, they were either cropped with wheat or improved grazing pasture to avoid bias from wildflower species. All hedges were managed in the same way, cut back in autumn, and left for the entire year. All SP hedges were in need of restoration in places, therefore stretches of hedge were selected where there were no gaps, and the hedge was fully intact, for a comparable distance to the SR hedges.

#### 6.3.1 Invertebrate Sampling

Each of the six hedgerows used within the trial supported three sample points (subsamples) spaced 15 metres apart (Appendix 4, Figure A4.1b). At each point Three pan traps (17cm diameter and 6cm depth) were placed at each sample point, 1m apart in a line along the hedgerow at approximately 1m height, level with the vegetation (Tuell and Isaacs 2009). Wooden posts with brackets and wire were used to secure the pans in place during sampling. There were three subsamples on each hedgerow totalling nine traps per hedgerow, and 54 pans in total. Blue, yellow and white UV reflective plastic bowls were used to account for colour preference by certain invertebrates (Wilson et al. 2008, Scriven et al. 2013). These colours were used as they have been successfully used to attract invertebrates from a variety of taxa, and they are similar to natural flower colouration (Moroń et al. 2008, Westphal et al. 2008, Scriven et al. 2013). Traps were filled to the three quarter line with water, to which several drops of unscented dishwashing detergent (Ecover-Zero) were added to reduce the surface tension.

Traps were exposed for a period of 30 hours (traps set before 10.00 and collected after 16.00 the following day, to prevent bias between sampling times) every two weeks during May – September 2013 (n=11 samples) during weather conditions considered favourable for flying invertebrates (when temperatures were above 17 °C, it was neither raining nor likely to rain, and wind speeds were less than five on the Beaufort scale). On collection, the specimens were transferred into glass vials, labelled and preserved in 70% ethanol. All invertebrate species considered to be a flower-visitor; shown to have a distinct taxonomic group or function e.g a predator utilising plants or important pollination were identified to family level and where possible to genus and species using a dichotomous key (Unwin 1984, Chinery 2005, Ball et al. 2013). Individuals were recorded from Hymenoptera including Ichneumonidae, Tenthredinidae, and Apidae, Lepidoptera, Diptera specifically, Syrphidae, Bombyliidae, Empididae, Scathophagidae, Asilidae and Tachinidae (Larson et al. 2001). Coleoptera and other dipteran families were excluded from the study.

### **6.3.2 Statistical analysis**

The data collected from each subsample (n=3 traps) was combined to give three psudeoreplicates per hedgerow. These values were averaged and used as a single data points per hedge per survey (n=11). All data analysis was completed within the R program (R Core Team 2013). The mean abundance (average number of individuals), the Shannon Weiner Diversity and the species richness (number of different species present) at each site were compared using Kruskal-Wallis, as a result of non-conformation to normality and heterogeneity. To assess the community matrix, Principle Component Analysis (PCA) was conducted on the data; scatter plots provided an insight into the influential species in the dataset. PCA provides a good visual indicator of significant variances within the dataset, allowing the identification of key trends by removing some of the 'noise within the data, however, PCA can be limited, as the method relies on linear assumptions, and is not scale inherent. Ordination plots can also distort data creating a horseshoe or arch effect, where the second axis is an arched function of the first axis, caused by unimodal distribution or long gradients within PCA. Further SIMPER analysis on the data from a Bray Curtis dissimilarity matrix, highlighted the abundance and importance of specific invertebrate species. Multivariate PERMANOVA (Manhattan distance matrices) was used to determine a significant difference between species poor and species rich hedgerows

(Anderson et al. 2005); the homogeneity of distribution was assessed using the “betadisper” function within the vegan package (Oksanen et al. 2013).

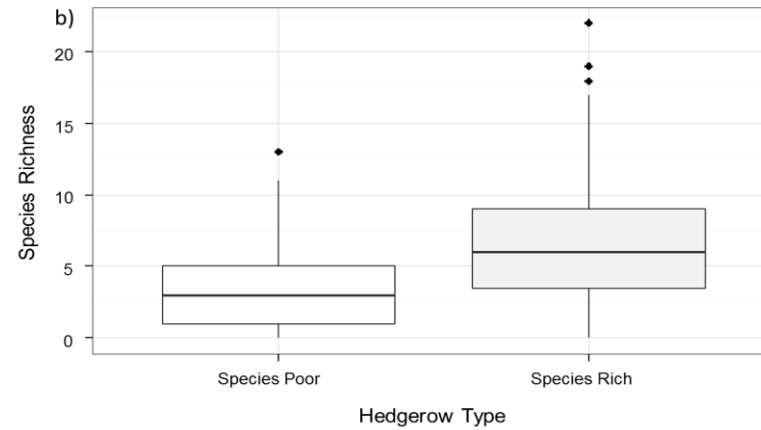
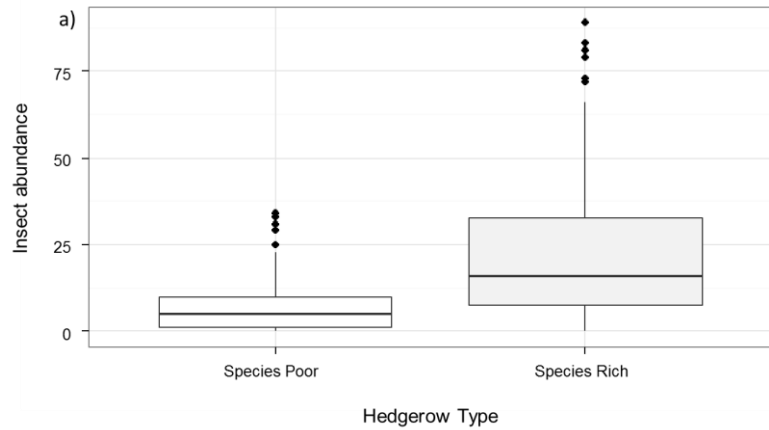
## **6.4 Results**

Over the course of the trial (11 visits, traps set every 14 days throughout May – September 2013), 3042 flower-visiting invertebrates were collected in total, 2273 individuals were recorded within species rich (SR) hedges, and 769 individuals were recorded within species poor (SP) hedges (Species list available Appendix 4, Table A4.1).

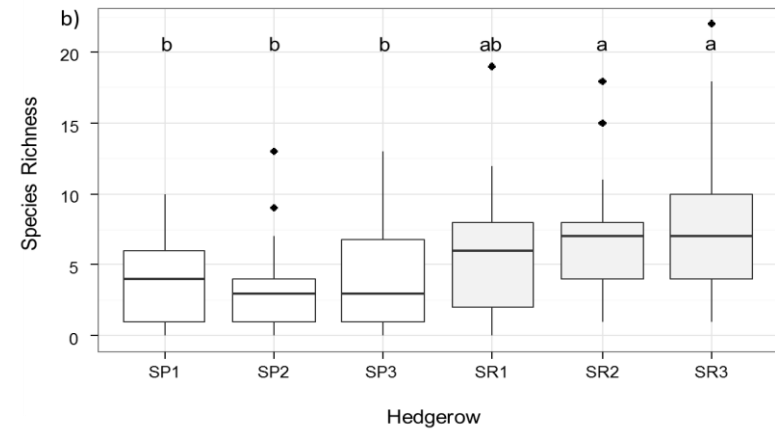
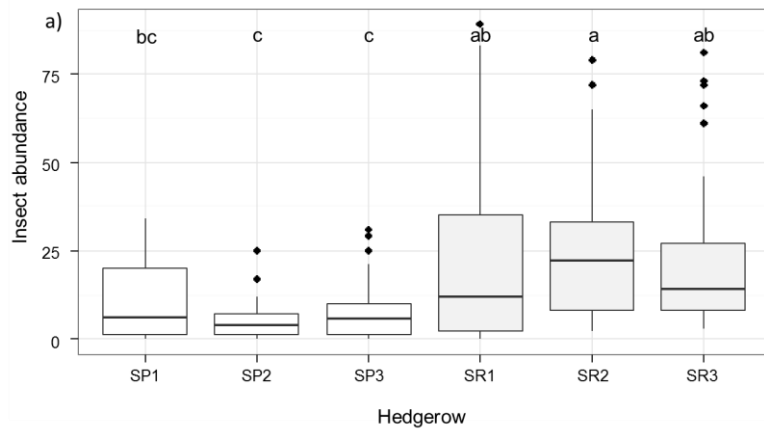
### ***6.4.1 Total invertebrate abundance and species richness between species rich and species poor hedgerows.***

Total flower-visiting invertebrate abundance was significantly different between the SR and SP hedges (Kruskal-Wallis,  $\chi^2 = 43.82$ , d.f. = 5,  $P < 0.0001$ ), SR hedges had a higher flower-visiting invertebrate abundance compared to SP hedges (Figure 6.1a). There was a substantial amount of variation in invertebrate abundance between replicates (n=3) within both hedge types (Figure 6.2a). Pairwise Wilcoxon comparisons (P-adjusted, Bonferonni) showed hedgerow SR2 had a significantly higher invertebrate abundance compared to all SP hedgerows. SP1 however, was comparable to two of the SR hedgerows, despite a lower average invertebrate abundance.

Results were similar for invertebrate species richness for the two hedgerow classifications (Figure 6.1b), SR hedgerows had a significantly higher invertebrate species richness when compared to SP hedgerows (Kruskal-Wallis,  $\chi^2 = 30.11$ , d.f. = 5,  $P < 0.0001$ ). Again, average site species richness varied between hedgerow replicates. Pairwise Wilcoxon comparison tests (P-adjusted, Bonferonni) showed two SR hedgerows had significantly higher species richness compared to all SP hedgerows (Figure 6.2b).



**Figure 6.1:** Whiskered boxplot showing the average; a) invertebrate abundance and b) species richness between the two Hedgerow classifications. The box plot is constructed from the median, and upper and lower quartiles. Whiskers show the maximum and minimum data points recorded, excluding any outliers. Outliers in the data are identified by black dots.

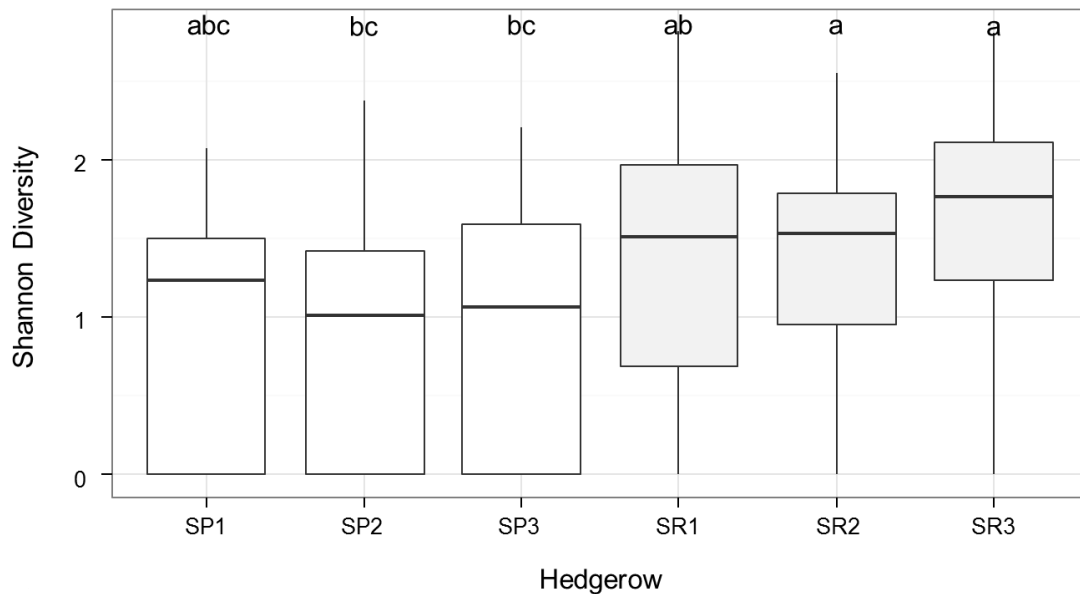


**Figure 6.2:** Whiskered Boxplot showing the average; a) invertebrate abundance and b) species richness of the six surveyed. The white boxes represent SP hedges and grey SR hedges. The box plot is constructed from the median, and upper and lower quartiles. Whiskers show the maximum and minimum data points recorded, excluding any outliers. Outliers in the data are identified by black dots. Letters represent differences following Pairwise Wilcoxon comparison test, P-adjusted, Bonferonni corrected.



#### 6.4.2 Differences in diversity of species between hedgerow classifications

Shannon Weiner diversity was significantly different between the two hedgerow types (Kruskal-Wallis,  $\chi^2 = 24.08$ , d.f. = 5,  $P < 0.001$ ). Pairwise Wilcoxon comparison tests showed that the two species rich hedgerows SR2, and SR3 had the highest species diversity. Interestingly, site SP1 (which had the highest invertebrate abundance for an SP hedge type) had a comparable Shannon Weiner diversity score to all the SR hedgerows (Figure 6.3).

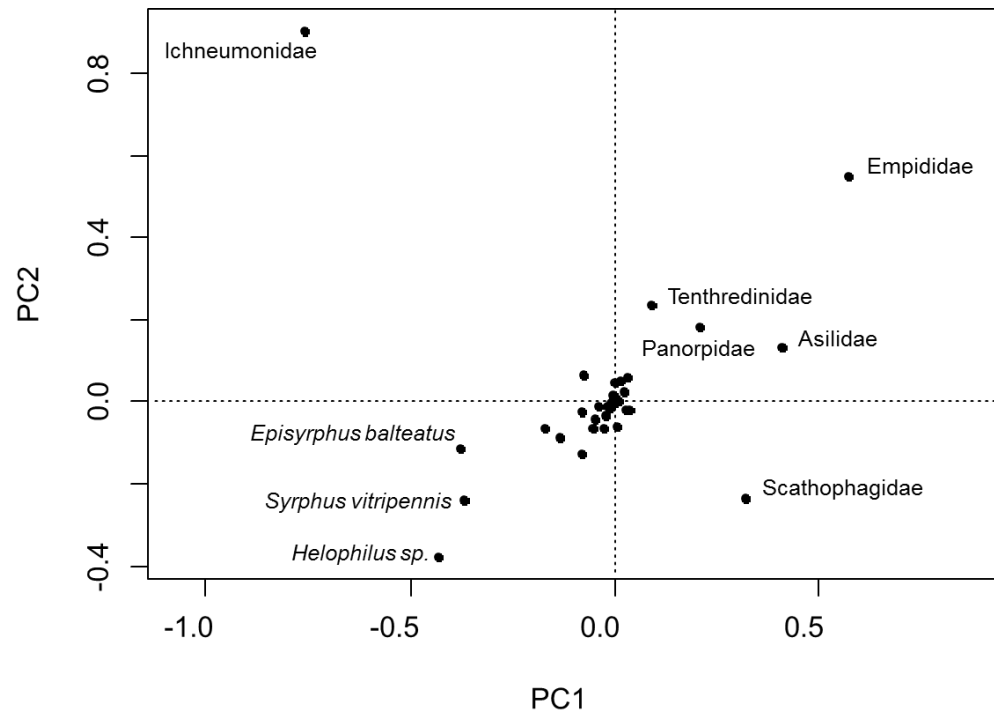


**Figure 6.3:** Whiskered boxplot showing Shannon diversity for each of the six hedgerows. The white boxes represent the SP hedgerows and grey SR hedgerows. The box plot is constructed from the median, and upper and lower quartiles. Whiskers show the maximum and minimum data points recorded, excluding any outliers. Letters represent differences following Pairwise Wilcoxon test,  $P$  adjusted, Bonferonni corrected.

#### 6.4.3 Differences in community structure between species poor and species rich hedgerows

Following PCA analysis it becomes clear that the Parasitic, Nectar Feeding and Predator guilds were promoting the main differences in invertebrate assemblages between sites (Figure 6.4). To partition the Manhattan distance of SP and SR hedgerows, PERMANOVA analysis was conducted and highlighted a significant difference between the invertebrate communities (PERMANOVA,  $R^2 = 0.0258$ , d.f. = 1,  $P < 0.01$ ). SIMPER analysis allowed identification of the invertebrates causing the

greatest effect on the community structure. The abundance and proportion of each identified invertebrate were calculated (Table 6.1). SIMPER analysis showed that 30 of the invertebrate taxa recorded (equalling 74% of the total community) had a higher abundance within the SR hedgerows and the remaining 11 taxa (26% of the total community) were found in higher densities within the SP hedgerows, three of these were species of Syrphidae (Table 6.1).



**Figure 6.4:** Scatterplot of PCA1 and PCA2 following analysis of the invertebrate communities. Each point represents one of the invertebrate classifications given during survey and PCA assumes that large variance within the data equals a low covariance and high importance. The data points showing the greatest variance within the data have been labelled accordingly and appear to cluster in the flower-visiting invertebrate guilds.

**Table 6.1:** Abundance data for SP and SR hedgerows, and the ordered cumulative contribution for each invertebrate following SIMPER analysis on a Bray Curtis dissimilarity matrix. Shaded boxes represent the site type with the highest abundance of each particular invertebrate. Any species with a contribution of less than 1% following SIMPER has been removed from this table, therefore 90% contribution of species is presented below, but 100% was used during assessment.

Invertebrate	SP	SR	Cumulative contribution
Ichneumonidae	<b>0.513</b>	0.440	0.118
Empididae	<b>0.202</b>	0.195	0.211
<i>Helophilus sp.</i>	0.143	<b>0.250</b>	0.304
Asilidae	0.090	<b>0.144</b>	0.378
Tenthredinidae	0.118	<b>0.138</b>	0.440
Scathophagidae	0.067	<b>0.112</b>	0.499
<i>Syrphus vitripennis</i>	<b>0.123</b>	0.078	0.556
<i>Bombus terrestris/lucorum</i>	0.091	<b>0.118</b>	0.607
<i>Episyphus balteatus</i>	<b>0.110</b>	0.084	0.655
<i>Bombus lapidarius</i>	0.075	<b>0.096</b>	0.697
<i>Platycheirus sp.</i>	<b>0.081</b>	0.069	0.737
Panorpidae	0.019	<b>0.100</b>	0.775
<i>Rhingia campestris</i>	0.044	<b>0.066</b>	0.803
<i>Melanostoma sp.</i>	0.025	<b>0.052</b>	0.825
<i>Eupeodes corolla</i>	0.025	<b>0.048</b>	0.845
<i>Eristalis arbustorum</i>	0.000	<b>0.056</b>	0.864
<i>Bombus pascorum</i>	0.009	<b>0.054</b>	0.882
<i>Eristalis tenax</i>	0.008	<b>0.040</b>	0.897
<i>Melangyna sp.</i>	0.016	<b>0.023</b>	0.912

## 6.5 Discussion

Both hedgerow types were utilised by a number of flower-visiting invertebrates throughout the study. The results showed that the three species rich hedges supported a greater diversity and abundance of foraging invertebrates compared to the three older species poor hedgerows. However, it would be expected that SR hedges would have a greater diversity of flowering species providing more forage resources for invertebrates during the survey period (Maudsley 2000). Flower densities recorded within and in close proximity of the hedgerows were low across all sites, and despite there being no flowering plants in bloom within any of the hedges during the survey period, flower-visiting invertebrates including bumblebees were observed occupy all hedgerows. Furthermore, invertebrates were also observed to fly along the length of some of the hedgerows, which suggests certain species of invertebrates may be utilising hedgerows for navigation or commuting between foraging grounds and refuge, a theory which has been recorded for Syrphidae specifically (Haenke et al. 2014).

Both SP and SR hedgerows were visited by invertebrates considered important for pollination such as *Bombus sp.* and *Apis mellifera*. Although, species rich hedges had a higher invertebrate abundance, the importance of species poor hedgerows within the landscape should not be overlooked (Maudsley 2000, Boughey et al. 2011). All hedgerows provide connective habitat which can facilitate and support the dispersal of invertebrates and other species within the wider landscape, despite the composition of woody shrubs (Hinsley and Bellamy 2000, Davies and Pullin 2007, Staley et al. 2012). Furthermore, if land adjacent to hedgerows is managed to support an uncultivated 2 metre margin, which consists of wildflowers of differing heights and flowerhead shapes, the ecological value of the habitat can be increased further (Defra 2007). Within this study, higher numbers of predatory invertebrates such as Ichneumonidae, and nectar feeders such as *Syrphus vitripennis* and *Episyrphus balteatus* were observed within species poor hedgerows compared to the species rich hedgerows. However, groups of pollinators including *Bombus sp.* were also recorded across species poor hedgerows, emphasising that both types of hedgerow are important to flower-visiting invertebrates within the landscape context.

Previous studies have shown that forage availability, structural diversity and botanical diversity are all important to support a diverse invertebrate community (Carvell 2002, Croxton et al. 2002, Sjödin et al. 2008). Such features are easily achievable through the management of hedgerows. The role of hedgerows as boundary features should

be maintained, restored and re-established throughout the countryside because a single hedgerow can support a variety of taxa, enhancing the local biodiversity within the target area.

## **6.6 Conclusions**

Although both hedgerow types were utilised by flower-visiting invertebrates, species rich hedges supported a greater abundance, richness and diversity of invertebrate species than species poor hedgerows showing a higher value as connective habitat within the wider landscape.

### **6.6.1 *Future management strategies***

During late August and September many young birds have fledged their nest and hedge cutting is routinely implemented at this time. However, this can be a crucial time for foraging invertebrates and birds. The most common hedgerow cutting practice among farmers in England is annual trimming; a reduced frequency such as cutting once every 2-3 years, can save money and benefit the environment. However, adverse effects on crop yield have been recorded as a response to increased shading from uncut hedges (Staley et al 2012), so although 3 year cycles would be ideal for invertebrates, machinery and crops may be affected, therefore a compromise of 2 years would be recommended.

Restoration of hedgerows which have become dilapidated is clearly an effective method of improving diversification of flower-visiting invertebrates at any given site. The introduction of species such as Dogrose, Blackthorn, Beech, and Dogwood into current gaps will improve the hedge structure, and increase the height of the hedge to the desirable 1.5-2 metres. Additional planting of Meadowsweet and other species along hedge bottoms would also improve the habitat for invertebrates.

## Chapter 7

### Testing the efficacy of different substrates within artificial nests as a tool to attract flower-visiting invertebrates

#### 7.1 Abstract

Populations of pollinating invertebrates have seen a sharp decline over the past decade. Such a reduction in flower-visiting invertebrate diversity can have a significant effect on plant reproduction success, affecting both crop production and wildflower populations. Habitat fragmentation is one of the most severe causes of such invertebrate decline, reducing the availability of suitable habitat for forage, refuge, and reproduction. Although it remains difficult to reduce or even reverse such a decline, providing artificial habitat specifically for invertebrates via artificial nests could be a low cost method to manage the decline within a localised area.

Bees from the family Megachilidae have been recorded to successfully inhabit artificial nests, however the success of this tool for increasing abundance of other invertebrates and the viability of such artificial nests over time remains unclear. To address these questions, twelve artificial nests were positioned during April 2012, across four different locations at the Alcan Aluminium Smelter, Northumberland, UK. During October 2012, 2013, and 2014, one nest was collected from each site and taken back to the laboratory for analysis. Results showed a significant increase in both trap occupation and species diversity when artificial nests remained in position over a longer period of time, suggesting that artificial nests could increase flower-visiting invertebrate diversity within the local area. However, similar to previously reported results, our study showed a bias towards bees from the family Megachilidae, and showed that a mixture of substrate choices should be available for flower-visiting invertebrates, and that the technique should be integrated with other management strategies to increase invertebrate diversity throughout the site.

## 7.2 Introduction

The decline of European flower-visiting invertebrates, bees in particular, has been well documented (Biesmeijer 2006, Goulson et al. 2008b, Potts et al. 2010). A combination of factors have been associated with the decline; however, agricultural intensification and habitat fragmentation are considered the primary causes (Potts et al. 2010). The reduction in flower-visiting invertebrate abundance could have significant implications for entire ecosystems (Hooper et al. 2005, Tilman et al. 2006, Winfree et al. 2008) as pollinating invertebrates are essential in maintaining terrestrial habitats due to their role in plant reproduction. Continued declines in invertebrate abundance could severely impact plant development, inhibiting crop success, forage availability, and wildflower populations nationwide (Fontaine et al. 2006, Carvell et al. 2007, Westphal et al. 2008, Potts et al. 2009).

Historically, wild bees were the primary pollinators of crops and wild plants (Winfree et al. 2008), contributing to around 35% of crop production worldwide (Klein et al. 2007). However, as a result of increasing demand for pollination services by farmers, hives of *Apis mellifera*, and *Bombus terrestris* sp. are now routinely used to ensure crop and flower yield. *A. mellifera* have been managed commercially for crop pollination and honey production for centuries (Kremen et al. 2002), a result which has directly increased the density of pollinating invertebrates throughout the landscape. Domesticated *B. terrestris* sp. however, have only recently been introduced, and although their use is predominately restricted to within glasshouses, commercial populations have been used within poly-tunnels and open fields to enhance yields of soft fruit crops (Velthuis and Van Doorn 2006). Despite the positive role in improving crop yield on a local scale (Corbet et al. 1991), the introduction of commercial pollinator hives could pose a significant risk to wild invertebrate populations (Steffan-Dewenter and Tscharntke 2000). The increase in commercially reared bee colonies across Europe has resulted in increased competition for forage resources within target areas. Furthermore, close contact and uncontrolled transportation has resulted in these commercially reared bee species being exposed to high parasite loads and infectious pathogens (Fürst et al. 2014, Graystock et al. 2014). Parasites and pathogens are easily transferred from one invertebrate to another (Durrer and Schmid-Hempel 1994), highlighting concern for native invertebrate populations which may occupy similar habitats. An alternative to increasing usage of non-native pollinating species is to encourage an increase in the diversity and abundance of native pollinating species, by

improving habitat for flower-visiting invertebrates within a close vicinity to the target area.

Loss of suitable habitat has been identified as one of the key drivers of the observed invertebrate decline (Potts et al. 2010). Invertebrates require suitable nesting sites and food resources within their foraging range to ensure successful reproduction, and species survival (Goodell 2003, Klein et al. 2004). Providing artificial nests for flower-visiting invertebrates within close proximity to suitable forage resources could improve species abundance within agricultural landscapes. Providing temporary breeding habitat in this form could be used to enhance agricultural areas, while long-term habitat improvements such as wildflower seeding, and hedgerow regeneration are undertaken (Chapter 4 and 5, Maudsley 2000, Carvell 2002, Pywell et al. 2006).

Artificial nests are being produced commercially and are often encouraged by local conservation charities as a feature to attract bees and other invertebrates into urban areas. However, the effectiveness of these nests as a tool for invertebrate conservation still remains unclear (Dicks et al. 2010). The production of artificial nests has allowed researchers to successfully monitor invertebrate population sizes, brood success and environmental change within targeted areas (Kevan et al. 1990, Gathmann et al. 1994, Westrich 1996), yet, the successful uptake of artificial nests appears species dependent. Artificial bumblebee nests for example have had a low success rate (Lye et al. 2011, Lye et al. 2012), whereas positive results have been recorded for above ground nesting solitary bees and wasps (Gaston et al. 2005).

The long term benefits of artificial nests as a method of enhancing local populations also remains unclear. Some studies have indicated that solitary bee uptake increases with length of exposure to artificial nest (Gathmann and Tschardtke 1997, Breeze et al. 2011), yet, a small replicated trial within Maine, USA, found a variable increase between bee species and trial replicates (Stubbs et al. 1997). Tschardtke et al. (1998) also found that increasing the number of traps within an area did not always have a positive effect on bee abundance, questioning the effectiveness of this method, however it is likely that surrounding environment will be more influential to artificial nest success (Gathmann et al. 1994) rather than the number of artificial nests available.

The success of artificial nests for flower-visiting invertebrates is relatively understudied in comparison to Apidae, therefore this trial aims to observe the uptake of flower-visiting invertebrates within artificial nests over a three-year period. Specifically, three



hypotheses were investigated i) the number of occupied stems within each nest will significantly increase with exposure time, ii) there will be no significant differences between the three substrate types utilised, and iii) there will be a significant difference between species and recorded stem diameter.

### 7.3 Study Design

#### 7.3.1 Study site

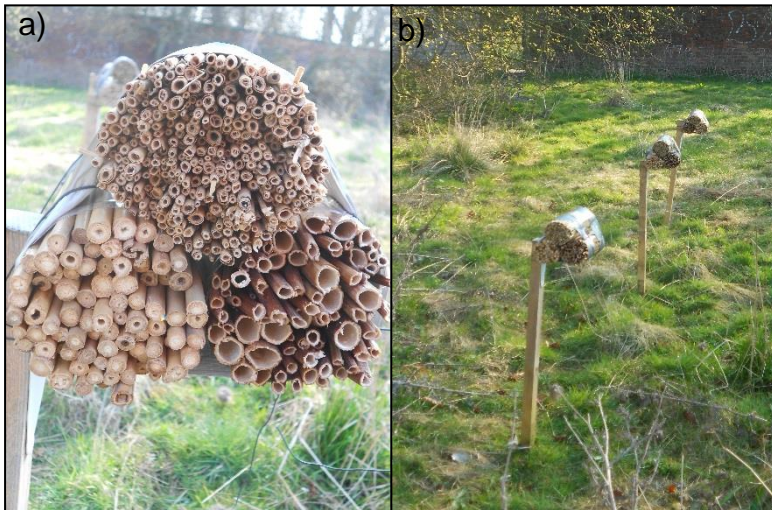
The study was conducted on the Aluminium smelter site of RioTinto Alcan, Lynemouth, UK (55.2016° N, 1.5396° W). Covering 82.7 hectares the site is typical of a working industrial site, with intensively managed grassland and shrub borders around offices, access roads, car parks, and production units. The buffer zone around the boundary provides a fragment of valuable habitat for wildlife within landscape dominated by agriculture.

#### 7.3.2 Artificial nest design

Each artificial nest was constructed from three plastic tubes (length 16cm, diameter 7cm), each filled with a different nesting substrate (Table 7.1). The three substrate tubes were bound together to produce one artificial nest block (Figure 7.1).

**Table 7.1:** Details relating to the internal stem diameter and number of stems per substrate material per artificial nest set out at the Alcan smelter to monitor flower-visiting invertebrate uptake.

Substrate	Stem size range (mm)	Average number of stems per bundle
Reed stem	2-5	335
Japanese Knotweed stem	2-10	77
Bamboo canes	4-8	73



**Figure 7.1:** An example of the artificial nest used within the experiment; a) shows the three substrates (clockwise, from left to right) bamboo, reed and Japanese knotweed, and b) One replicate of the experiment (The vegetation in this area was known to reach approximately 1m in height)

### **7.3.3 Sampling**

Artificial nests were set out during April 2012; three nest blocks (each block consisting of the three substrate types – Figure 7.1a) were positioned at each of four locations across the site (Table 7.2). One of the traps was collected from each location in October 2012, the second in 2013 until the final traps were collected in October 2014. The traps were positioned on individual posts at approximately 1-metre-high, and 3 metres apart (The exact height was dependent on the surrounding vegetation). The traps were positioned in direct sunlight and were oriented in a south-westerly direction. The traps were positioned at a slight angle (entrance lower) to allow drainage from heavy rainfall.

### **7.3.4 Nest analysis**

After collection, the nests were disassembled and each stem containing brood cells or evidence of occupation was placed within individual sealed polypropylene perforated film. Stems were stored at +4°C within a chilled cabinet during assessment. For each occupied stem the internal diameter (mm) was measured using Vernier callipers, the nest construction (leaf, mud or no substrate), and the number of chambers was recorded. For 2013, and 2014 nests it was recorded if the stem was currently unoccupied indicating a previous years nest (However, because the year of occupation could not be confirmed this data was removed during analysis).

**Table 7.2:** The location and dominant flowering plants within each of the four artificial nest sites within the grounds at the Alcan Smelter, Lynemouth, UK.

Site Name	Location (British National Grid)	Dominant flowering species recorded
Woodhorn Grange	NZ29350 BNG89685	Tall grasses, <i>Cirsium arvensis</i> , <i>Leucanthemum vulgare</i> , <i>Prunella vulgaris</i> , <i>Centaurea nigra</i>
Flower-Rich Grassland	NZ29350 BNG89685	<i>Lotus corniculatus</i> , <i>Trifolium repens</i> , <i>Leucanthemum vulgare</i> , <i>Prunella vulgaris</i> , <i>Centaurea nigra</i>
Pond	NZ29384 BNG89278	<i>Centaurea nigra</i> , <i>Cirsium arvensis</i> Tall grasses
Mown Grassland	NZ29674 BNG89205	Tall grasses, <i>Cirsium arvensis</i> , <i>Taraxacum officinale</i> , <i>Trifolium repens</i>

### 7.3.5 Identification

The type of nest constructed was the main indicator of species present, however where inhabitants did hatch, they were identified to genus level where possible. Megachilidae bees such as Leafcutter and Mason bees, are expected to use the nests as they are known to readily accept artificial nesting materials (Krombein 1967, Torchio 1990) (Figure 7.2). However, any flower-visiting invertebrate found using the artificial nests was recorded throughout the trial period.



**Figure 7.2:** Images of the nest cells of leafcutter bees found within Japanese knotweed stems; a) shows the sealed stems, b) leaf cutter prior to larvae hatching, c) larvae hatched and feeding on leaves provided, d) the individual leaf entombed egg sacs which are found stacked together within a single stem e) the egg inside each sac, and f) pollen stored around the egg for when the larvae emerge. **Scales = 5mm.**

### **7.3.6 Statistical Analysis**

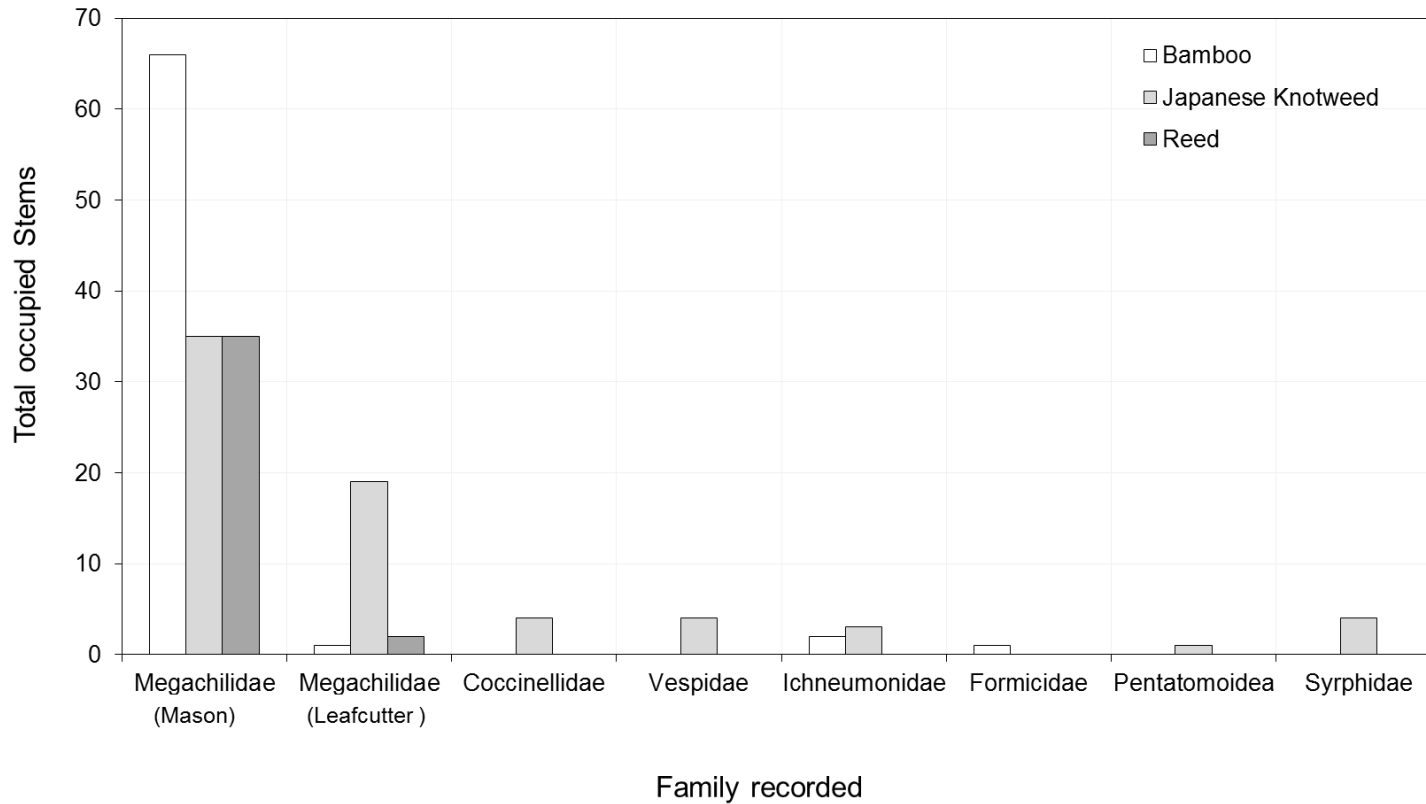
All statistical analyses were completed within the R statistical and programming environment, version 3.0.2 (R Core Team 2013). To assess the uptake of stems between years an Analysis of Variance (ANOVA) was utilised, followed by Tukey's Higher Significance Difference (HSD) tests to establish any significant differences between the three substrate types and year of exposure. Where data did not conform to a normal distribution Kruskal-Wallis, followed by pairwise Wilcoxon tests (P-adjusted Bonferroni) was used as an alternative.

The number of invertebrates other than from the family Megachilidae was low. This prevented further statistical analysis on the full dataset. For this reason, the Chi squared test for association (Ennos 2012) between invertebrates and substrate type was only conducted on the two types of Megachilidae bees found (Leafcutter and mason). The impacts on other families are discussed within the text, but cannot be fully assessed due to the low.

## **7.4 Results**

### **7.4.1 The uptake of nest sites by flower-visiting invertebrates**

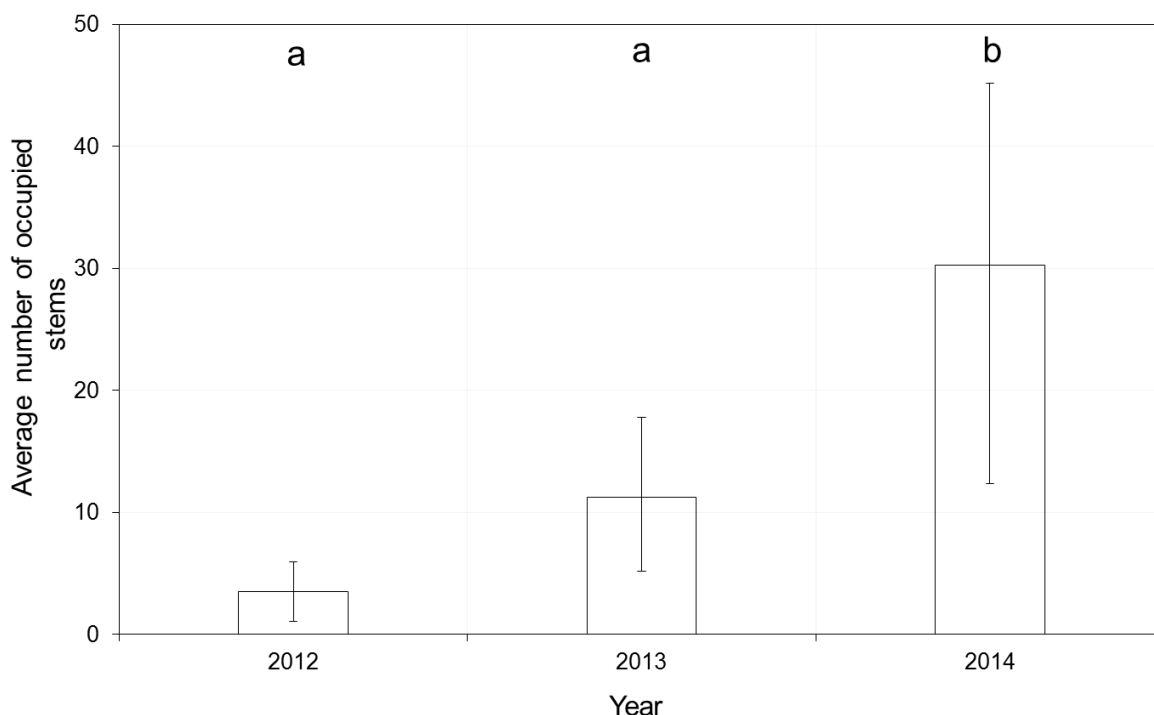
Over the three-year trial period 180 stems were occupied by flower-visiting invertebrates. Megachilidae bees were the most frequently recorded family, with 168 occupied stems totalling 93% of the total occupied stems recorded (Figure 7.3). The highest number of occupied stems was recorded within Bamboo stems, which were predominantly used by the Megachilidae bees. The greatest species diversity was recorded within Japanese Knotweed stems, attracting a greater number of different families; 6 out of the 7 families utilised Japanese knotweed stems during the trial compared to 3 out of 7 and 1 out of 7 for Bamboo and Reed respectively.



**Figure 7.3:** Bar graph showing the total number of occupied stems recorded for each substrate (Japanese Knotweed, Bamboo and Reed) within the artificial nests. The total occupied stems were calculated from the three traps, ordered by the family within the highest abundance (This graph show the total number of occupied stems including old /unoccupied nests, which were excluded from further analysis as the exact age of the nest could not be determined).

#### **7.4.2 Does occupancy within artificial nests increase with exposure length?**

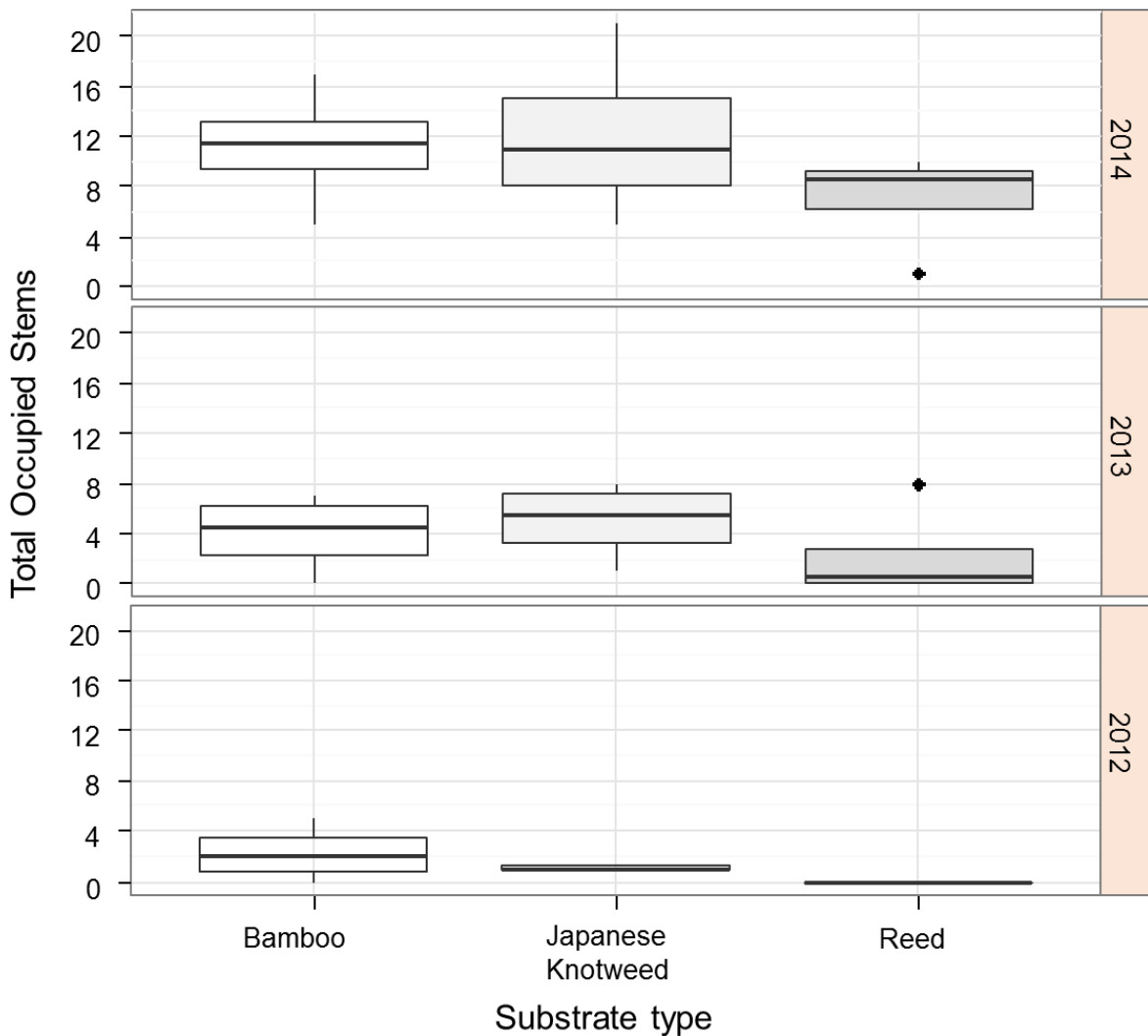
Occupancy of stems significantly increased with year (ANOVA,  $F = 8.783$ ,  $d.f=2$ ,  $P < 0.01$ ). Tukey's Higher Significant Difference (HSD) comparisons showed that 2014 had a significantly higher number of occupied stems compared to years 2012 and 2013. Despite an apparent increase in the number of occupied stems in 2013 compared to 2012, the difference was not significant, likely due to the variation between replicates (Figure 7.4).



**Figure 7.4:** Bar graph showing the increase of stem uptake by invertebrates over the exposure period. Error bars are calculated from the Standard Error of the Mean (SEM,  $n=4$ ), and letters represent significant differences following Tukey's HSD comparison tests,  $P$ -adjusted, Bonferroni corrected.

#### **7.4.3 Is there a preference for nesting substrate by flower-visiting invertebrates?**

All three substrates were utilised by flower-visiting invertebrates across all three sample years. Uptake was low within the first year (2012) with less than 5 stems of each substrate utilised. During years 2013 and 2014 the number of occupied stems for Bamboo and Japanese Knotweed increased at a comparable rate (Figure 7.5). Both were visited by a larger proportion of flower-visiting invertebrates compared to the Reed stems, despite this apparent difference it was not significant (Kruskal-Wallis  $\chi^2 = 18.681$ ,  $d.f = 15$ ,  $P = 0.23$ ).



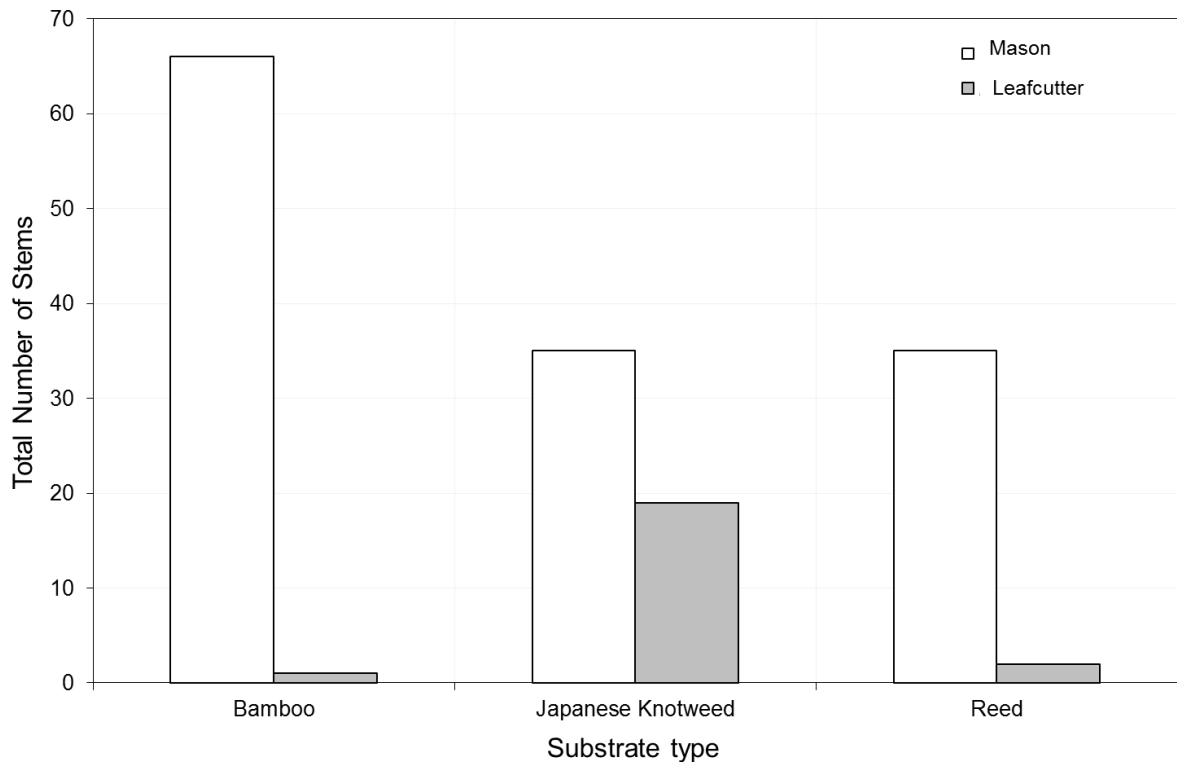
**Figure 7.5:** Whiskered Boxplot showing the total occupied stems for each substrate type (Bamboo, Japanese Knotweed and Reed) during each assessment year. The box plot is constructed from the median, and upper and lower quartiles. Whiskers show the maximum and minimum data points recorded, excluding any outliers. Outliers in the data are identified by black dots.

#### **7.4.4 Is there a substrate preference by Megachilidae bee species?**

Although there was no significant difference in total occupancy recorded between substrate types, a difference in the Megachilidae associated with Bamboo and Japanese knotweed stems was observed (Figure 7.6). The Mason bees appeared to occupy all substrate types, showing a higher occupancy within bamboo stems, whereas Leafcutter bees were recorded in higher numbers with Japanese Knotweed. Following Chi-squared test for association a significantly greater proportion of Mason were recorded occupying Bamboo stems compared to Leafcutter ( $\chi^2 = 18.56$ , d.f = 1,



P < 0.001). A result which may indicate that substrate choice within artificial nests could influence their success.

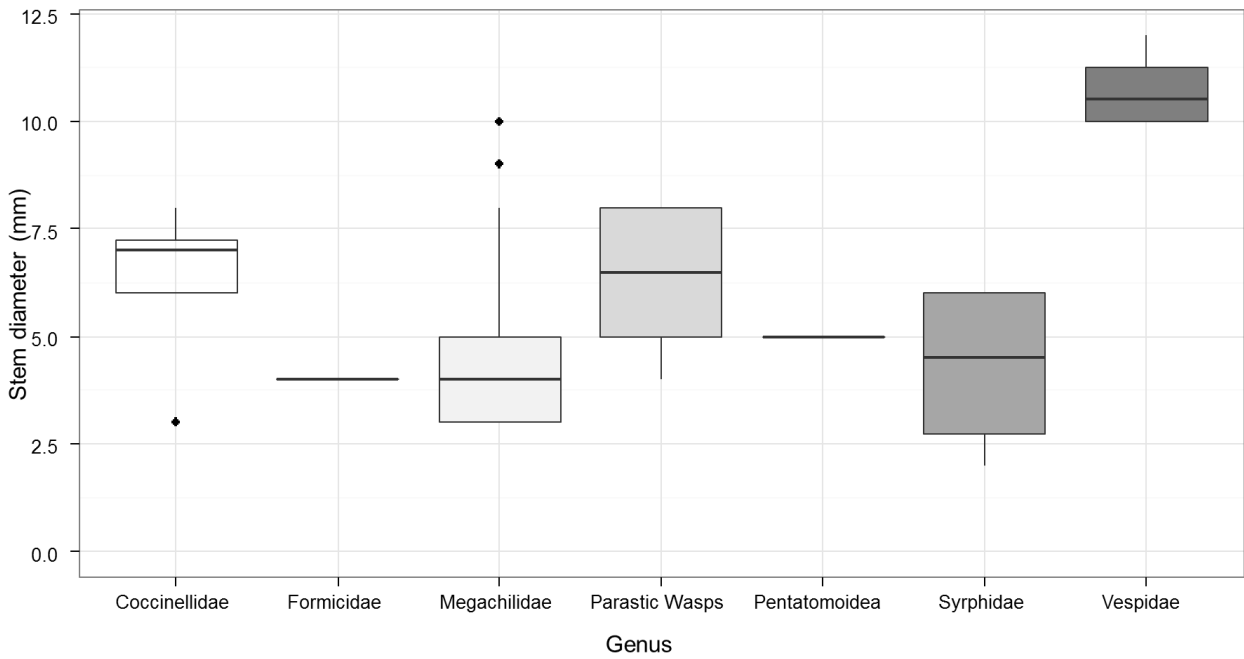


**Figure 7.6:** Bar graph comparing the total number of Mason. and Leafcutter solitary bees utilising the three different substrate types provided within the artificial nests located around the Alcan Smelter.

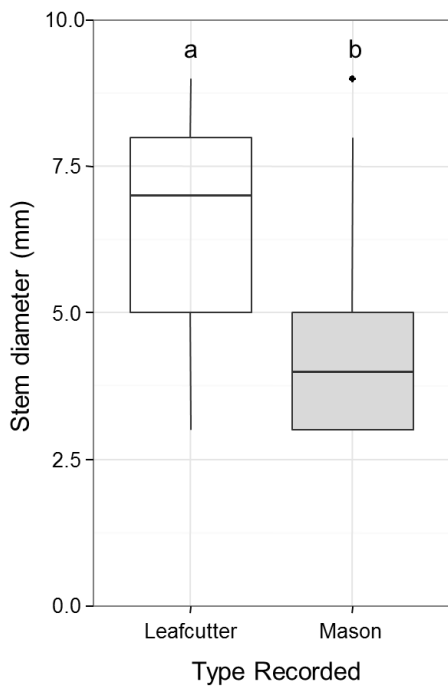
#### ***7.4.5 Is there a stem size preference by different groups of flower-visiting invertebrates?***

The stem diameter was recorded for each occupied stem recorded within the artificial nest. Vespidae appeared to show a preference for larger stem diameters than any of the other groups (Figure 7.7). A stem diameter between 3.5mm and 8mm appeared to be the most frequently used by flower-visiting invertebrates within this trial.

Interestingly, a variation of preference between stem diameters was recorded between the two genera of Megachilidae bees recorded during the study. The average diameter of stems utilised by Mason bees was 4mm compared to 7mm for the Leafcutter (Figure 7.8). This preference between different genera was significant (Kruskal-Wallis  $\chi^2=19.96$ , d.f = 1, P < 0.001), showing that stem size availability could impact the success of artificial nest traps as a tool for conservation.



**Figure 7.7:** Whiskered boxplot comparing the stem diameter used by the different groups of invertebrates recorded during the trial. The box plot is constructed from the median, and upper and lower quartiles. Whiskers show the maximum and minimum data points recorded, excluding any outliers. Outliers in the data are identified by black dots.



**Figure 7.8:** Whiskered Boxplot showing the difference between stem diameter choice between Megachilidae genera. The box plot is constructed from the median, and upper and lower quartiles. Whiskers show the maximum and minimum data points recorded, excluding any outliers. Outliers in the data are identified by black dots, P-adjusted, Bonferroni corrected.

## 7.5 Discussion

The use of artificial nests by flower-visiting invertebrates increased across the trial period. This was particularly the case for bees from the family Megachilidae. The first year nests (those put out in 2012) saw a particularly low uptake by invertebrates, however the increase in numbers between years 2013 and 2014 was significant, indicating that uptake within artificial nests could provide a method of habitat enhancement for certain groups of flower-visiting invertebrates. Interestingly, this study highlighted that the overall success of uptake within the artificial nests depended on the length of time the nest was deployed. There are other factors which could also increase usage over time such as learning, pheromones, and weathering, however this should now be taken into account when utilising this management tool, coupled positioning within a suitable location, consideration of forage resources, exposure to sunlight (Gaston et al. 2005), and the proximity of the nests to other natural nest sites and suitable habitat.

The high uptake by Megachilidae bees within artificial nests was not unexpected as this has been shown before (Krombein 1967), however a preference within the family was highlighted with a higher abundance of Mason compared to Leafcutter bees recorded over the trial period. There was also a significant difference in the size/type of stem being used by these two genera of bee; Mason averaging 4mm and showing a preference for Bamboo stems, whilst Leafcutter averaging 7mm and showing a preference for Japanese knotweed stems. Although, substrate type appears to be the main contributor to variation in diversity and abundance, there may however be other reasons. For example, the number of larger stems within the bamboo sections of the nests was limited as the overall size of the artificial nest was standardised rather than the number of stems present. Furthermore, the Japanese knotweed stems had a greater range of diameters, allowing both species to utilise the substrate effectively. However, this in turn could have also increased competition between the two genera and resulted in less optimum sized stems being present for the Leafcutter to nest build.

While other studies have shown that drilled wooden blocks can be more successful for the uptake of Megachilidae (Wilkaniec and Giejdasz 2003), we aimed to provide hibernation and breeding habitat for a broader range of flower-visiting invertebrates, and stems allow for a standardised nesting resource to be easily reproduced and replicated, and have become common practice for research groups (Tscharntke et al. 1998, Steffan-Dewenter 2002). Stem diameters ranging between 2 and 10 mm have

been thought to be the optimum diameter to encourage the highest diversity of invertebrates (Tschardt et al. 1998, Morón et al. 2008). Within this study, Japanese Knotweed stem diameters ranged from 2-10mm, and Bamboo stems 4-8mm and these two substrates hosted the highest abundance of invertebrates. The uptake of reed stems was significantly lower than the other two substrates, however reed stems had an internal diameter of 5mm or less. Similar to the other studies, it appears that the difference in uptake is correlated with size, as the stems were smaller in diameter in the reeds compared to the other two substrates. Interestingly, a study by Gathmann et al., (1994) reported that 5mm reed stems hosted the highest abundance of Megachilidae in their study undertaken in Kralruhe, Germany. This is in direct contrast to our study. One possible explanation for this difference may be that in our study a greater substrate choice was offered to invertebrates with a greater number of reed stems smaller than 5mm stem diameter which could have resulted in a preference to the other substrates, or variation in the size and species of bee recorded.

This study has illustrated that substrate type is an important factor to consider when constructing and choosing artificial nests for flower-visiting invertebrates. Interestingly, our results support those of others (Dicks et al. 2010), whereby bamboo stems were the most frequented substrate type. This substrate has been so successful it is often the most common substrate found in commercially available artificial nests (also known as bee hotels). Interestingly, the two most common genera which were found to nest in this study showed significant preferences, with Leafcutter being lowest in bamboo and Mason highest. This result contrasts findings by Sihag (1993), where bamboo stems were not utilised within artificial nests. These difference in findings may simply be a result of geographical location, as Sihag's study was undertaken in India, where the abundance, diversity and nesting preferences of bees from the Megachilidae are different to the UK (Abrol 2011).

Finally, the overall aim of positioning the artificial nests across the site was to assess the success of this method as a means of increasing the diversity of flower-visiting invertebrates in suitable habitat. Although, invertebrates from 7 families were found to occupy these artificial nests, species of solitary bee from Megachilidae did appear to be the most frequent inhabitants, suggesting that this particular management strategy has the potential support numbers, and potentially increase numbers and diversity of this group in particular. That said, the value of the artificial nest to support Vespidae and Coleoptera should not be overlooked. Despite, all the artificial nests being within

the site boundary (82 hectares), located within similar habitat types under the same conditions; variation was recorded in relation to uptake success. Such variation in uptake appeared to correlate with flower density at the site as discussed in Chapter 2. Combined, this suggests that floral density and diversity are also an important consideration for invertebrates during nest selection (Gathmann et al. 1994, Scriven et al. 2013), suggesting that positioning of artificial nests is an equally important decision along with substrate type when choosing to use this management technique.

### **7.5.1 Future direction**

Further research into the optimum range of stem sizes and substrate preferences to benefit target groups would be beneficial to the construction of artificial nest in the future. In addition sweep net data before and after artificial nests are laid, and within the local landscape would help to understand the true impact of the artificial nests for conservation purposes (Stubbs et al. 1997). Furthermore, monitoring the behavioural ecology of invertebrates utilising the artificial nest would be beneficial. The use of human observation or camera trapping technology could monitor the movement, construction and species interactions, either inter species or parasitoid within and around the nest allowing further understanding of the benefits and problems associated with this technique.

## **7.6 Conclusions**

Flower-visiting invertebrates successfully inhabited the artificial nests positioning within the Alcan Smelter, however numbers of invertebrate families other than Megachilidae were low. Results showed that the greatest diversity of species were found within the Japanese knotweed stems which supported a larger stem diameter than the other nesting substrates, but in terms of abundance bamboo was significantly higher. The results are encouraging, however further research is required to establish the future role of artificial nests to boost local flower-invertebrate populations, and provide a tool for conservation purposes.

## Chapter 8 General Discussion

### 8.1 General Discussion

Research into the causes and consequences of pollinator decline have dominated the literature since Biesmeijer et al. (2006) published an evidence based paper detailing a significant decline in pollinator assemblages pre and post 1980. Although widespread pollinator decline and its implications for the economy had been discussed prior to this time (Kremen and Ricketts 2000, Roubik 2001), Biesmeijer et al's landmark paper evidenced the significance of the problem, opening new avenues of research aimed primarily at identifying the primary causes of decline, the species of particular concern and the methods which could be available for mitigation strategies. However, despite considerable advances in the understanding of invertebrate behaviour and forage preferences (Jha and Vandermeer 2009, Rands and Whitney 2010), coupled with increased conservation efforts through the introduction of flower rich arable margins (Carvell et al. 2007, Lye et al. 2011, Pywell et al. 2011b, Feltham et al. 2015), and planting wildflower mixes (Haaland et al. 2011, Blackmore and Goulson 2014) invertebrate fauna continues to face rapid decline.

In an attempt to truly mitigate this observed decline in both number and diversity of invertebrate species, habitat management aimed at protecting invertebrates is required. For this to occur, we first need to a) address appropriate target habitats and species, and b) improve the connectivity of patches of suitable habitat to enhance distribution of species on a landscape scale (Bennett 1999, Donald and Evans 2006). This thesis increases our understanding of habitat management specifically within an industrial area; an area of habitat which would normally be deemed of low ecological value, and provides management prescriptions which could rapidly improve biodiversity at the target site in a cost effective manner; a method which could also be utilised on specific sites nationwide. The principal findings and outcomes of the thesis (Table 8.1) confirm that changes to intensive management regimes on sites such as the Alcan Smelter in Northumberland, can enhance invertebrate number and diversity within the local area (Chapter 3, 4, and 6). The methods discussed within the thesis suggest that an increase in local species diversity is possible, and therefore have the potential to reduce, at least in some part, the continued invertebrate decline (Biesmeijer et al. 2006). By guiding future management of urban areas, industrial areas, and agricultural environments on a wider scale, the extent of invertebrate

decline could be impacted (Kleijn et al. 2006, Kleijn et al. 2011, Potts et al. 2011, Baldock et al. 2015, Threlfall et al. 2015).

Encompassing a variety of flower-visiting invertebrates, including parasites, predators, nectar feeders and pollinators; the forage preferences of invertebrate guilds were studied to inform the enhancement of biodiversity through habitat management. Studying a broad range of invertebrates allowed assessment of interactions between different taxa and the available forage plants, rather than focusing on a single target group such as bumblebees, hoverflies or butterflies (Moeller 2004, Baldock et al. 2015). Incorporating all flower-visiting invertebrates allows a wider understanding of the implications of any management scheme to enhance biodiversity, allowing us to ensure that any management prescriptions assigned, benefit all species at a landscape level, and most importantly ensure that it is complementary to habitat features present or evidenced on site.

The study area for this thesis was focused on the Alcan Aluminium Smelter in Northumberland, UK. Despite being an active industrial site; the individual habitat types within the buffer zone of the smelter were similar to those found within many urban and agricultural situations, allowing the findings to be easily adapted and used in other scenarios on a national level (Kearns and Inouye 1997). Furthermore, aside from the disturbance attributed to industrial activity, the mosaic of habitats on site including tall ruderal herbs, scrub, colonised bare ground and woodland could all be considered equivalent to habitat found within Brownfield sites (land previously used for commercial or industrial purposes). Brownfield sites are classified as a BAP priority habitat as a result of the open mosaic habitat which hosts a diverse assemblage of invertebrates (Alker et al. 2000, Small et al. 2002, Eyre et al. 2003); if managed to support and enhance biodiversity, an active industrial area, which would have previously been considered to be of low ecological value, can also be of significant value to wildlife within the local landscape.

**Table 8.1:** Summary of main hypotheses, results and findings associated with the thesis

	<b>Key hypotheses</b>	<b>Key Results</b>	<b>Significance:</b>
<b>Chapter 2</b>	<p>Habitat type / site would have a significant effect on the abundance and diversity of flower-visiting invertebrates recorded,</p> <p>Flower density will have a significant positive effect on flower-visiting invertebrate assemblages</p> <p>Flowering plant diversity will have a significant positive effect on flower-visiting invertebrate assemblages.</p>	<p>REJECT - Habitat type / site was only found to have a significant effect on invertebrate species richness</p> <p>ACCEPT- Flower density was highlighted as a significant factor with regard to invertebrate assemblage, and showed a significant correlation between Shannon Wiener Diversity of invertebrates and flower density</p> <p>REJECT- Flowering plant diversity was not identified as a significant factor for flower-visiting guild assemblage and no correlation of flower diversity with insect diversity</p>	<p>This study highlights the importance of data interpretation to determine management objectives, and recommends analysing the community structure and identifying the dominant species prior to undertaking any land management scheme.</p> <p>Flower density attracted invertebrates to the site indicating the value of large wildflower areas within a management plan targeting invertebrate conservation.</p>



<p style="text-align: center;"><b>Chapter 3</b></p>	<p>Removing grass trimmings from experimental plots will result in a higher flower density</p>	<p>REJECT - The removal or not, of grass trimmings was not a significant variable with regards to increasing flower density</p>	<p>The timing of flowering grassland is important so as not to affect the attractiveness of the site and affect seed deposition.</p>
	<p>Plots subjected to the 2M regime over the flowering period will support a significantly higher herb cover and support greater suppression of grasses</p>	<p>ACCEPT- Plots maintained under the 2M regime had a significantly higher percentage cover of flowering species</p>	<p>Results show that in the short term the 1M regime appears to attract the greater number of invertebrates, but over a longer time period the benefits of grass suppression within the 2M regime could further improve grassland attractiveness.</p>
	<p>Flower density will have a significant effect on invertebrate abundance and richness.</p>	<p>ACCEPT- Flower density was a significant factor when predicting invertebrate abundance</p>	<p>The 1M regime is considered standard practice to maintain grassland diversity, but over a long time period the 2M regime could provide a better grassland composition, which should be considered during implementation of ecological management plans for areas of grassland.</p>
	<p>Plots subjected to the 1M regime will support a significantly higher flower-visiting invertebrate diversity.</p>	<p>ACCEPT- A significant difference in invertebrate diversity was observed within 1M regime plots, however, the 2M regime saw an increase in invertebrate diversity during the second year</p>	

<b>Chapter 4</b>	<p>Flower visiting invertebrate abundance and diversity will be significantly higher at nature reserve grasslands and sites managed with wildflower mix</p>	<p>REJECT – The grassland at Alcan was found to support higher invertebrate abundance than the two seeded sites but was comparable to the two nature reserve grasslands.</p>	<p>Comparison results provide evidence based backing that the amendments to mowing regimes were effective and comparable to the wider landscape.</p>
	<p>Flower density and flowering plant diversity will be significantly higher at nature reserve grasslands and sites managed with wildflower mix</p>	<p>REJECT – The grassland at Alcan supported significantly higher flower density and plant diversity compared to the other four sites.</p>	<p>Although sites have individual abiotic and biotic conditions, the process has identified species of importance within the area and should be further incorporated into management schemes, future landscaping proposals and/or public open space.</p>
	<p>Ecological network analysis can be used to highlight flowering plant species of importance to guide future management strategies.</p>	<p>ACCEPT – Species of importance highlighted within the network analysis corresponded with field observations suggesting that the information gain from the network could be used to influence management decisions within the local area</p>	<p>Ecological networks could prove a valuable tool in determining the optimal wildflower mix to support pollinators in different geographical locations and situations.</p>

<b>Chapter 5</b>	<p>Seed diversity would be significantly higher in samples collected from 10-15cm depth compared to the surface layer (0-5cm)</p> <p>Light levels representative of the ride habitat (T2) will significantly increase germination rate and plant species diversity of a shaded woodland mix.</p>	<p>REJECT – Trends recorded in relation to an increase of plant species diversity, and abundance with depth were not significant.</p> <p>ACCEPT- Germination of seedlings within T2 plots saw a significantly higher number of seedlings compared to the other treatments. In terms of plant species diversity, the plots were comparable to the T3 edge plots</p>	<p>Following assessment of the seedbank and light levels associated with the woodland was considered to be of low ecological value to flower-visiting invertebrates, and considerable enhancements to the structure of the woodland would be required for an enhancement in biodiversity. Providing evidence backed reporting could be a method of establishing ecological management plans within habitat patches such as this, and further work to identify other factors which determine the ecological status of woodland habitats could help form an assessment and mitigation strategy.</p>
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<b>Chapter 6</b>	<p>Species rich (SR) hedgerows support a significantly higher abundance of flower-visiting invertebrates than species poor (SP) hedgerows</p>	<p>ACCEPT- SR hedgerows had a higher flower-visiting invertebrate abundance compared to SP hedgerows</p>	<p>SR hedgerows appear to support a greater diversity of invertebrates providing greater value as wildlife corridors and habitat patches themselves.</p>
	<p>SR hedgerows support significantly higher species diversity of flower-visiting invertebrates compared to SP hedgerows</p>	<p>ACCEPT- Shannon Weiner diversity was significantly different between the 2 hedge types</p>	<p>Long-term management and protection to SR hedgerows should be conducted to secure biological diversity, work to enhance the plant species diversity within species-poor hedgerows should also be encouraged to increase levels of biodiversity within these hedges</p>
	<p>SR hedgerows would host a significantly higher abundance of nectar feeding species and pollen collecting invertebrate guilds, than predator and parasitic guilds</p>	<p>ACCEPT- Significant differences was found between the invertebrate communities associated with the 2 hedge types, with SP supporting more members of the parasitic and predatory guilds.</p>	

**Chapter 7**

The number of occupied stems within each nest will significantly increase with exposure time

There will be no significant differences between the three substrate types utilised

ACCEPT- Occupancy within the artificial nests showed a significant increase over the three year period.

REJECT- Mason bees showed a significant preference for bamboo canes

Results showed that the greatest diversity of invertebrate species were found within the Japanese knotweed stems. These stems had larger stem diameters than the other nesting substrates, potentially explaining this difference.

Further research into the optimum range of stem diameters and substrate preferences of other insects should be conducted, specifically targeting known beneficial invertebrate species or those showing regional or national decline.

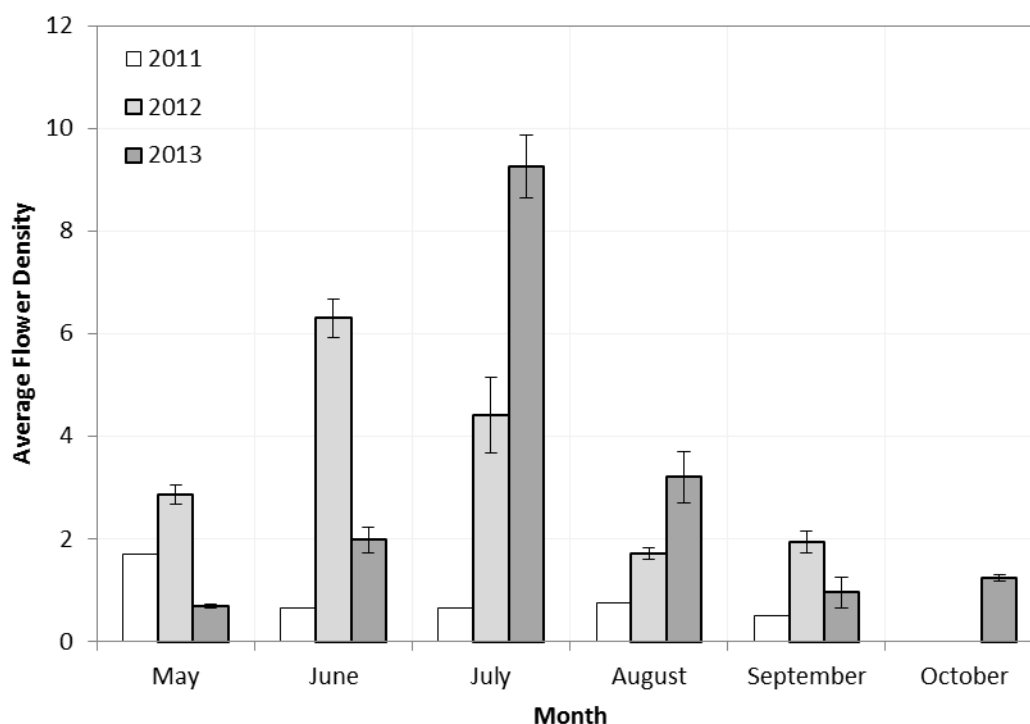
Survey of the local landscape for invertebrates before and after artificial nests are laid should be conducted and would allow greater understanding with regard to the true impact of the artificial nests for conservation purposes.

An understanding of the site context is an important factor when considering a site's potential to respond positively to management techniques which enhance biodiversity, and its relative importance within the local landscape. The connectivity between habitat patches can almost be more important in some cases, than the composition of the habitat itself (Bennett 1999, Saura and Torné 2009, Saura and Rubio 2010). An area of land which can be managed to benefit biodiversity, which is well-connected across the landscape could have more value in the maintenance of the wider environment than the preservation of isolated rarer habitat types which have statutory designations for their botanical composition or declining habitat types (Donald and Evans 2006). For instance, the location of the smelter within an agriculturally dominated landscape, provides an increase in structural diversity and increased forage resource within an otherwise monoculture dominated landscape (Benton et al. 2003, Rands and Whitney 2010). The connectivity of the site via linear features including hedgerows, trees, and ditches allows dispersal of species to Local Nature Reserves including woodland and country parks, the RAMSAR Coastline, and SSSI wetlands, ponds and grasslands. The importance of connectivity between favourable habitat patches should not be underestimated. Future management should focus on facilitating and encouraging flower-visiting invertebrate dispersal via the careful management of the site and these existing pathways of dispersal could further enhance the biodiversity into the wider landscape (Tscharntke et al. 2005).

When establishing habitat management and mitigation strategies for a site, a thorough assessment of the current condition for the target species should be undertaken (Carvell et al. 2011). This assessment allows analysis post planned intervention to determine any positive or negative effects. In this instance Chapter 2 identified potential areas within the site to target management and provided valuable baseline data with regard to flower density and current flower-visiting invertebrate assemblages. Results showed that the flowering period within the grassland at the smelter was relatively short, with low flower density recorded within late summer (between September and October), a time which is important for foraging pollinators (Memmott et al. 2007). This is a particularly important fact when you consider that Chapter 2 also illustrated that flower density was more important to invertebrate foragers than plant species diversity, with a higher abundance of flower-visiting invertebrates foraging in areas of high flower density over flower diversity (Scriven et al. 2013), a finding supported by others (Potts et al. 2003, Ghazoul 2006, Dicks et al. 2015).

On a landscape scale it would be expected that, to encourage greater invertebrate species diversity, a variety of flowering species would be required to provide forage for a greater number of species (Potts et al. 2003, Fründ et al. 2010). It is likely that the large blocks of colour from a high density of flowering plants attracts invertebrates in the first instance, rather than the overall attractiveness of a specific plant species being present or not (Benton et al. 2003). Other studies support this theory and have illustrated that large monocultures such as oil seed rape (*Brassica napus*) are attractive to pollinating species and can significantly influence their forage behaviour within agricultural landscapes (Westphal et al. 2003, Mänd et al. 2010, Rands and Whitney 2010). To further understand how to enhance biodiversity within the study area, the availability of flowering plants and their diversity should be enhanced within grassland areas allowing and extension of the flowering season (Mouradov et al. 2002) a topic addressed within Chapters 3 and 4.

In these next two Chapters different management techniques were implemented and compared with the aim of enhancing biodiversity in comparison to the base line levels recorded in Chapter 2. Chapter 3 introduced a mowing regime trial at the site, reducing the mowing intensity within the experimental plots, with results showing a significant increase in flower density and species diversity. A similar reduced intensity method is commonly employed within hay meadow environments, and is encouraged within flower rich grasslands to maintain flowering plant density and diversity, but also control the dominance of grassland species within the sward (Moog et al. 2002, Weiner et al. 2011). Within this experiment, the reduction in mowing intensity removed the constraint on the flowering plants allowing them to mature, flower and most importantly set seed to maintain the diversity in the future. Furthermore, species diversity within the sward increased in plots which were mown less regularly, suggesting that the seedbank was of high quality increasing the number of species over the trial period. Compared to the baseline data collected in Chapter 2, it is clear that the changes in grassland management improved the flower density recorded over the study period of three years (Figure 8.1), results are mirrored in agricultural grazing pastures showing an effective method of enhancing such habitat for flower-visiting invertebrates (Carvell 2002, Potts et al. 2009).



**Figure 8.1:** Bar graph to show the change in flower density within the area of flower rich grassland at the Alcan smelter, Lynemouth, UK, over the entire study period. The 2011 data was from a single site (Chapter 2), however the data does form a base line allowing comparison following management undertaken within Chapter 3. Error bars (2012 and 2013) show the Standard Error of the Mean (SEM, n=5).

Although a reduction in mowing intensity provided a significant increase in botanical and invertebrate diversity at the Alcan site, other sites may need more management to achieve or sustain an observable increase in flower density (Landres et al. 1999, Carvell 2002). Nevertheless, as a low cost method for improving sward diversity, and increasing flower density a change in mowing frequency can be very effective. If this is then combined with a greater understanding of the current seedbank quality at any given site, and the abundance and species composition of above ground dicotyledon plants already present the success of this technique can be improved further. It should also be noted, that the effectiveness of any such management can be influenced by the local, regional and national weather conditions at the time of implementation. For example, in this study during 2013, a higher than average amount of rainfall fell on the site, which appeared to result in the plants flowering later on in the year. Interestingly, during this same season, flowering appeared to be extended, reaching through until October (Figure 8.1), again most likely linked to the level of rainfall. This reflects results found in an earlier study by McCall and Primack (1992). However, that said, rainfall



was not recorded during this study, which prevents an evidence based cause for this observed trend.

The grassland experimentally manipulated at the Alcan smelter was comparable to areas declared as 'of local interest' in terms of the abundance and diversity of flower-visiting invertebrates, and flower density. In some instances, the factors assessed outperformed the other sites of which some had been involved in a flower-rich seeding program to enhance pollinator habitat within the county (Chapter 4). During 2013, we compared the grassland sward at the smelter site to grassland within two local wildlife sites and plots associated within the Northumberland Wildlife Trust's, Growing Wild, nectar and pollen rich seeding program. All sites were located within 15 km of the Alcan smelter, and supported a species rich grassland sward of similar composition. Interestingly, the species diversity recorded of both plants and invertebrates was comparable, if not marginally better within the grassland at the Alcan smelter compared to the other four sites. Furthermore, in terms of ecological stability (following plant – pollinator networks) the Alcan site supported a broader range of invertebrate species which were observed to forage on an equally broader range of plants compared to the other sites, where certain flower-visiting invertebrates were becoming dependant on one or two species of flowering plants (Chapter 4, Bosch et al. 2009, Morris et al. 2014). The stability of the habitat to support invertebrates becomes significant when planning an introduction of flower seed mixes; as it was the sites with the seeding applications which had the lowest habitat stability and flower density (Chapter 4), a result which could be accredited to poor establishment conditions or an inaccurate composition of seeds for the specific site (Landres et al. 1999, Mouradov et al. 2002).

Chapter 4 also highlights the importance of achieving the correct mix of flowering plants for nectar and pollen seed mixes. Specifically, the choice of plants and the post-application management utilised, such as mowing frequency and timing of the initial sowing. With this in mind, additional research should be conducted with the aim of ensuring that the appropriate flowering plants are supplied within the seed mixes which are applied in different conditions. It could be considered more appropriate to develop a seed-mix specific to each application site, following assessment of the current grassland sward condition in the first instance. Chapter 4 has emphasised that further research and a greater understanding of the effects of seed mix application and

development is required, particularly when selecting nectar/flower-rich seeding mixes for future agri-environmental field margins and roadside verges, an increasingly common feature, of which the true benefit to invertebrate conservation is currently under debate (Haaland et al. 2011, Feltham et al. 2015)

In Chapter 5, the focus of the work moved away from grassland to the plantation woodland which had been highlighted as relatively species poor in Chapter 2. The aim was to assess which management methods would be most suitable to improve the species diversity within the woodland. Interestingly, although, the ecological value of the woodland was classified as low, and supported a low species diversity (both plants and invertebrates), the habitat was noted as being important for parasitic insect species such as members of the family Ichneumonidae (Chapter 2). However, the connectivity of the woodland to the wider environment, and the proximity to the already improved open grassland, and arable fields suggested that this site had far greater potential to support a much more diverse invertebrate assemblage (Steffan-Dewenter et al. 2002, Bailey 2007). Although the initial intention was to illustrate that with relatively little management this site could be enhanced, a detailed survey of the existing seedbank highlighted that if the trees were simply thinned, the woodland would not be able to develop a significantly diverse understory because the abundance and diversity of seeds within the natural seedbank was low. Therefore, a woodland seed mix would be the ideal management strategy, however, a clearing strategy for trees would need to be established to allow germination and future colonisation of the understory (Barkham 1992). With this in mind, three different shading scenarios were replicated within seed trays using light levels comparable to the current level of canopy cover, and the light levels recorded under the ride, central canopy and woodland edge of the plantation. As expected, germination of seedlings was considerably reduced in the current level of canopy shading scenarios, indicating that further works such as gradual thinning to increase light penetration would be required to allow the seed mix to grow and develop (Mitchell and Woodward 1988, Atkinson et al. 2015). Such a study would allow the effects of felling and gradual thinning of trees within the woodland to be monitored and assessed as to whether or not a positive effect of this management strategy can be seen with relation to increasing certain groups of invertebrates (Woodcock et al. 2003, Atkinson et al. 2015).

Successful connectivity of habitat fragments across the landscape is important to achieve an increase of biodiversity (Crooks and Sanjayan 2006, Donald and Evans 2006, Bailey 2007). Specifically, the Alcan smelter currently benefits from a good structure of mature hedgerows, which were shown to support a diverse assemblage of invertebrate species (Chapter 2 and 6). In Chapter 6, both species poor and species rich hedgerows were considered important to invertebrate foragers, with the latter hosting a greater abundance of species overall (Chapter 6, Maudsley 2000, Hannon and Sisk 2009, Nicholls and Altieri 2013, M'Gonigle et al. 2015). This was despite the low flower density recorded, which reinforces the importance of hedgerows for navigation, and protection from predators (Wolton et al. 2013) rather than solely as food source. Future work to understand the importance of hedgerows as a forage resource, breeding habitat and navigation tool for flower-visiting invertebrates would benefit future management of hedgerows, particularly within agricultural situations. For example, if species-rich hedgerows support a greater number of flower-visiting invertebrates including pollinators, parasites and predators (as suggested by this study), then management would enhance the value of the hedgerows within the local landscape, providing natural predators for common agricultural pests, and an increase in native pollinating species (Boughey et al. 2011, Wolton et al. 2013).

The features discussed so far, work together to improve the dispersal of invertebrate species into and away from the site, and improve forage availability, inevitably improving the site for flower-visiting invertebrates (Carvell et al. 2007, Heard et al. 2007, Pywell et al. 2011a). However, another method to enhance biodiversity is to improve the provision of suitable hibernation or breeding habitat for invertebrates (Wray and Elle 2015). Within Chapter 7 artificial nest sites were introduced across the Alcan site, and results showed that they were successfully utilised by groups of flower-visiting invertebrates; particularly by bees from the family Megachilidae. As would be expected, the uptake of stems increased significantly with time (Gathmann and Tscharrntke 1997, Breeze et al. 2011), indicating that the artificial nests can and should be considered effective at providing suitable breeding and hibernation habitat (Chapter 7). The artificial nests were utilised by a number of invertebrate families, suggesting that either, there is a lack of suitable nesting/hibernation habitat within the local area, or that invertebrates were presented with an easier nesting option. The location of the nests within areas of open grassland dominated by flowering plants suggests that positioning of the artificial nests is the reason for success and should be considered

when implementing this management strategy in future areas (Gathmann and Tschardtke 1997, Tschardtke et al. 1998, Potts et al. 2003). In this instance, the grassland habitat presented limited opportunities for nesting, and therefore the addition of artificial nests within good foraging habitat has resulted in successful uptake. That said, to truly understand the impact of the artificial nests as a tool for conservation, an assessment of the local foraging population should be included, in addition to continued monitoring within areas supporting artificial nests and those without. This would establish whether the nests significantly boost adult numbers in proximity to the artificial nests, or whether invertebrates are changing areas.

Interestingly, although uptake did increase with time, the proportion of stems occupied was low compared to the number of stems available per artificial nest. However, having a larger number of stems available allows the artificial nests to be active for a longer period of time, potentially increasing the functionality of the nests as a result of weathering, pheromones or behavioural learning, but can also provide refuge for invertebrates while the establishment of suitable natural nesting habitat can be made. Conversely, it has been suggested that artificial nests could provide an easy target for predatory invertebrates (MacIvor and Packer 2015), and the longer the artificial nests are in place, the less desirable the nests will become. Therefore, as a temporary measure, artificial nests could boost local invertebrate populations, however increasing the provision of natural alternatives such as deadwood and flowering plants with hollow stems (Apiaceae) within the same area is also of value.

### **8.1.1 Conclusions**

The results of all the experimental studies included in this thesis show a positive effect of small-scale management strategies on the diversity of plants and invertebrates on site at the Alcan smelter. Furthermore, they importantly identify areas of future research to continue and maintain this success. It is undeniable that implementing these management prescriptions will benefit any given site, improving biodiversity and making the site a valuable part of the local landscape. Addressing the importance of appropriate management techniques to improve biodiversity, this thesis shows the speed in which recovery of degraded habitat can take place when the right management prescriptions are found. Furthermore, the overall study supports the need

for evidence based management, developed principally by a land manager who understands the impacts and importance of habitat and species interactions.

As stated at the beginning, the Alcan smelter hosts a variety of habitats, together forming a mosaic which is valuable to invertebrate conservation. The more transitions between habitats, the more niches are available to invertebrates, inevitably enhancing biodiversity. With this in mind, it is difficult not to conclude that the site of the Rio Tinto Alcan Smelter is extremely beneficial to invertebrate assemblages, not only because of the habitats currently available on site, but also from the connection with the local landscape. It is therefore assumed that under the correct management, many other fragments of habitat within industrial, or residential estates could also provide valuable habitat parcels which can be integrated and connected to enhance biodiversity on a national scale.

## **8.2 Specific management prescriptions for the Aluminium smelter**

Reversing the effects of biodiversity loss is an ambitious goal, but one which is frequently highlighted as conservation target within LBAP's or Management Plans (Watts and Selman 2004). To reverse biodiversity loss on a national scale, identification of habitat fragments within the landscape, which can enhance connectivity, is essential. Communicating between landowners to establish and maintain habitat features is also required on a national scale, along with the provision of funding for enhancing fragmented habitats, rather than just supporting designated sites. This latter issue needs to be raised, as although the preservation of rare habitat is important, it is the provision of connectivity, breeding habitat, foraging resources and refuge which is essential for success (Bennett 1999, Crooks and Sanjayan 2006). Therefore, the more urban and industrial habitats managed and integrated into the local landscape the closer the target of improving biodiversity can become (McGill 2015).

### **8.2.1 Woodland management targets**

A well-structured woodland can support the richest invertebrate fauna, across a number of ecological niches including bare ground, leaf litter, deadwood, bark and canopy. As such woodlands can support many herbivorous and predatory invertebrate species (Bailey 2007). Further work should be undertaken within the plantation woodland at the site, primarily gradual tree clearance as this will provide open space

allowing clearings or rides where light can penetrate and support shrubs and wildflowers, and facilitate the planting of young native trees and scrub (Smith et al. 2007, Harmer et al. 2010, Fuentes-Montemayor et al. 2012, Atkinson et al. 2015). This task will help to diversify the age of the woodland and add structural diversity, features which are currently lacking within the site (Chapter 5). Once clearance has been undertaken, the introduction of a shaded wildflower mix, and 3-4 year old shrubs such as Hawthorn, Holly, Blackthorn, or Hazel would help to increase structural diversity within the woodland (Kirby 1992). Once some of the non-native trees are removed, native tree planting will help shape the future structure of the woodland. Oak and Willow are two important species for invertebrates (Kirby 1992), but a mix of other broadleaved species including Field Maple, and Ash should also be encouraged. Additionally, the woodland margins should also be enhanced, encouraging tall herbs, and bramble scrub which would provide valuable habitat for invertebrates between more intensively managed environments, providing a gradual transition from grassland to scrub to woodland which would be of great ecological value (Dangerfield et al. 2003), and in some cases would improve connections with hedgerows..

### **8.2.2 Hedgerow management targets**

The most common downfall of hedgerows within the agricultural environment is over management (Burel 1996, Maudsley 2000), often reducing hedgerows to narrow strips of low thin scrub managed by mechanical felling, which prevents integration into the adjoining habitat (Defra 2007). Hedgerows of high value to invertebrates should be thick and dense, and managed on a two to three year cycle to allow growth of fruit, flowers and dense cover (Staley et al. 2012). Hedgerows within a network should be managed on a rotation system to prevent all hedgerows being managed at the same time, this has the added benefit of increasing structural diversity, and preventing loss of habitat throughout the landscape (Amy et al. 2015). The transition between habitat types should also be as gradual as possible, with field margins of at least 2 metres between crops or grassland supporting scrambling plants such as bramble, and tall herbs such as Hogweed, Hemp Agrimony, Yarrow and Meadowsweet (Dangerfield et al. 2003, Defra 2007, Nicholls and Altieri 2013). This will provide invertebrates with suitable refuge from management practices and provide foraging habitat in close proximity (Kirby 1992, Defra 2007). Restoration of defunct hedgerows should also be addressed, gap filling should be undertaken with locally dominant native species during early winter. The hedgerows around the Alcan smelter were Hawthorn dominant (even

the species-rich hedgerows), therefore gap filling should be undertaken with additional species such as Dogrose, Elder, Blackthorn, Holly and Field Maple to reduce the dominance of Hawthorn and further promote biological diversity. The addition of trees along hedgerows would also be advantageous as suggested by the study by Staley et al. (2015).

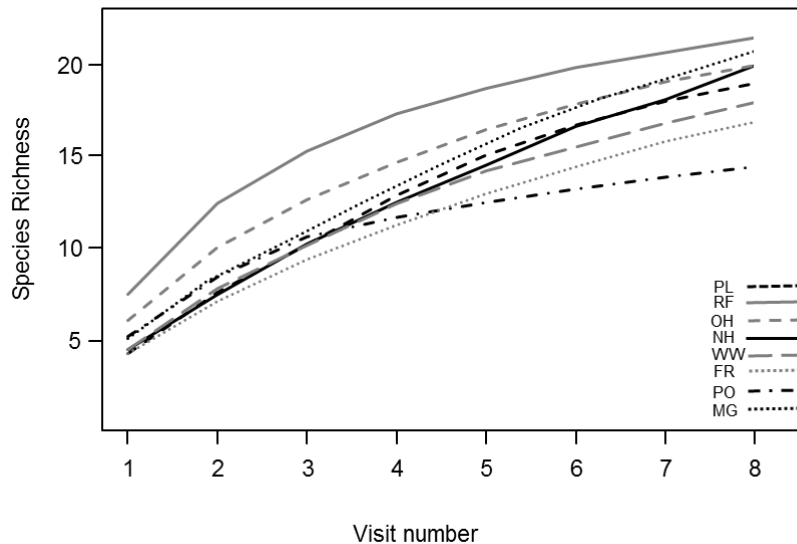
### **8.2.3 Grassland management targets**

Management within grassland habitats is essential to maintain a species diverse sward. Within this thesis the effects of different management techniques have been clearly observed, and show that a reduction of mowing intensity at the Alcan site significantly increases grassland sward diversity. This mowing regime should be maintained, and implemented in other areas of the site to further promote grassland management for biodiversity. The introduction of mid to late summer flowering species such as *Centaurea nigra* and *Malva moschata*, could improve the longevity of forage resources, in particular during the reproductive stages of bumblebees (Westphal et al. 2009). Towards the end of the season the new queens need to gather substantial resources in preparation for hibernation, ensuring forage at this time is essential for preventing species decline (Memmott et al. 2010). The thesis also highlighted that both cut methods had their own advantages; Buri (2013) highlights the importance of uncut patches within grassland as refuge for insects, in addition to the importance of delaying the first cut as long as possible, which is supported by our findings. It is also well documented that unmanaged grassland is more suitable for Carabidae, Orthoptera and Lepidoptera larvae. However, in the long term, management by grazing or mowing is essential for the success of flowering grassland. Therefore, a regime which incorporates a mixture of cut timings and cut lengths on a rotation system with the cuttings removed would be the ideal method to enhance grassland biodiversity.

## Appendix A – Supplementary Information - Chapter 2



**Figure AA.1:** OS Map (scaling 1:1500) showing the location of the eight sampling locations to determine flower-visiting invertebrate abundance within the buffer zone of the Alcan smelter, Lynemouth, UK. Flower Rich grassland (FR), Mown Grassland (MG), New Hedge (NH), Old Hedge (OH), Plantation Woodland (PI), Pond (Po), Ridge and Furrow Grassland (RF) and Woodhorn Woodland (WW).



**Figure AA.2:** The cumulative number of species is plotted for the eight visits within each of the eight sampling sites. Flower Rich grassland (FR), Mown Grassland (MG), New Hedge (NH), Old Hedge (OH), Plantation Woodland (PI), Pond (Po), Ridge and Furrow Grassland (RF) and Woodhorn Woodland (WW)



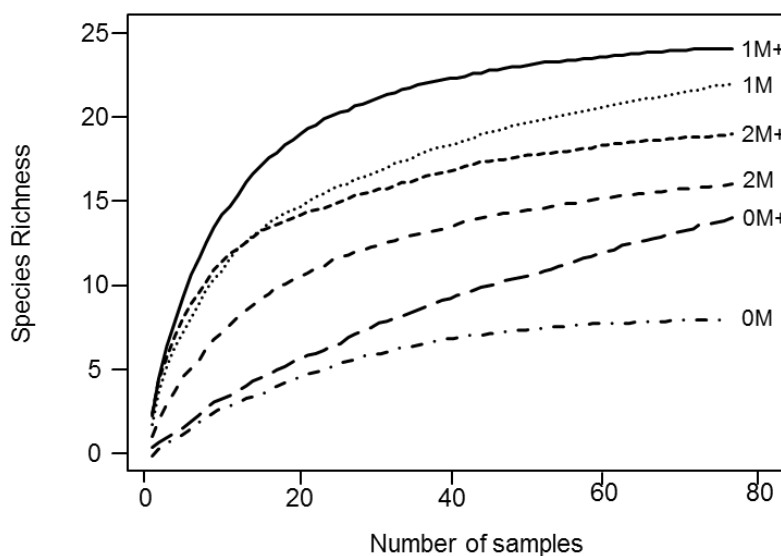
**Table AA.1:** Species list and total abundance recorded across the Alcan site following pan trapping for flower-visiting invertebrate on eight occasions within eight habitat parcels (Flower Rich grassland (FR), Mown Grassland (MG), New Hedge (NH), Old Hedge (OH), Plantation Woodland (PI), Pond (Po), Ridge and Furrow Grassland (RF) and Woodhorn Woodland (WW)).

Recorded Taxa	Total Abundance
<i>Leucozonia sp</i>	1
<i>Lejogaster sp</i>	1
<i>Epistrophe sp</i>	4
<i>Eupeodes sp</i>	29
<i>Helophilus sp</i>	32
<i>Chrysotoxum sp</i>	1
<i>Melanostoma sp</i>	10
<i>Syrphus sp</i>	59
<i>Episyrphus sp</i>	176
<i>Platycheirus sp</i>	35
<i>Sphaerophoria sp</i>	6
<i>Scaeva sp</i>	2
<i>Eristalis sp</i>	19
Panorpidae	2
sciomyzidae	44
Tachinidae	3
Sialidae	1
Emphididae	12
Asilidae	88
Tipulidae	5
Tenthredinidae	86
Chalcidoidea	61
Ichneumonidae	264
Vespidae	10
Chrysididae	2
Megachilidae	17
<i>Bombus pascorum</i>	14
<i>Bombus lucorum / terrestris</i>	33
<i>Bombus Lapidarius</i>	52
<i>Psithyrus silvestris</i>	3
<i>Psithyrus rupestris</i>	4
<i>Apis mellifera</i>	4
Pieridae	27
Kin Nymphalidae	7
Satyridae	1
Hesperiidae	7
Geometridae	2
Noctuidae	10
Coccinellidae	4

## Appendix B – Supplementary Information - Chapter 3



**Figure AB.1:** Map (scaling: 1:1,500). showing layout of experimental plots during the experimental phase of the mowing regime trial scaling. The plots were 3m x 10m and were separated by a 1metre buffer zone. The colours represent the cutting regime undertaken on each plot (0M Frequent cutting, 0M+, Frequent cutting with cuttings removed, 1M one cut mid-July, 1M+ One cut mid-July with cuttings removed, 2M Two cuts June and Sept, and 2M+ Two cuts June and Sept with cuttings removed).

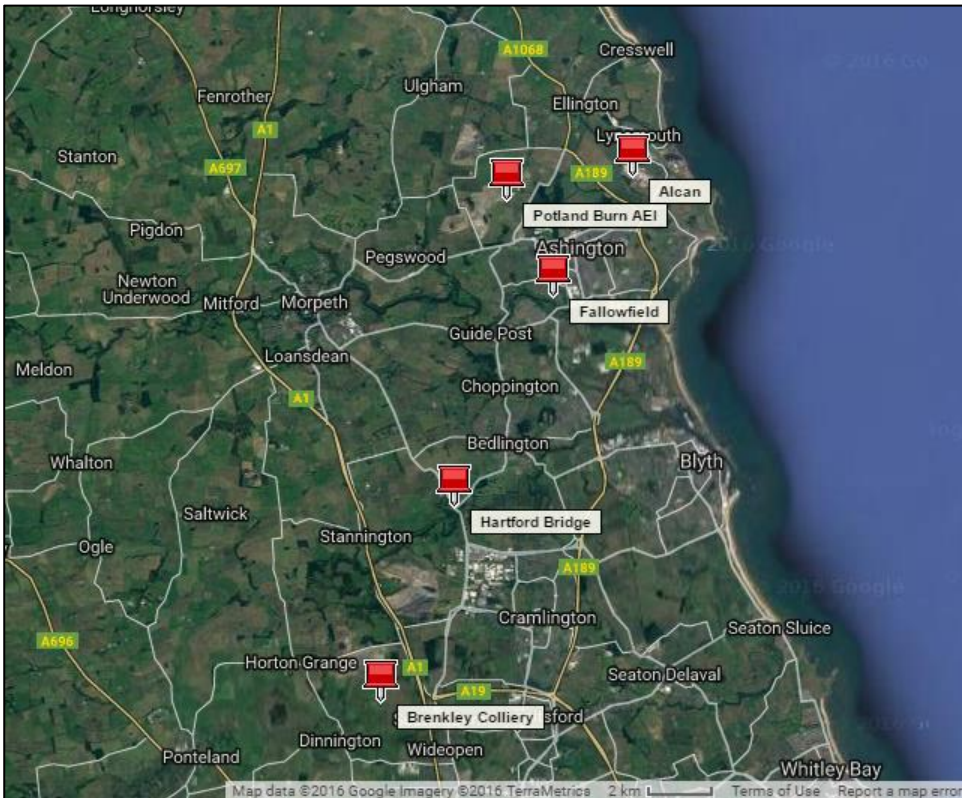


**Figure AB.2:** The cumulative number of species plotted for each of the experimental treatments within the mowing regime trial, undertaken at the Lynemouth Smelter, Alcan UK. (0M Frequent cutting, 0M+, Frequent cutting with cuttings removed, 1M one cut mid-July, 1M+ One cut mid-July with cuttings removed, 2M Two cuts June and Sept, and 2M+ Two cuts June and Sept with cuttings removed).

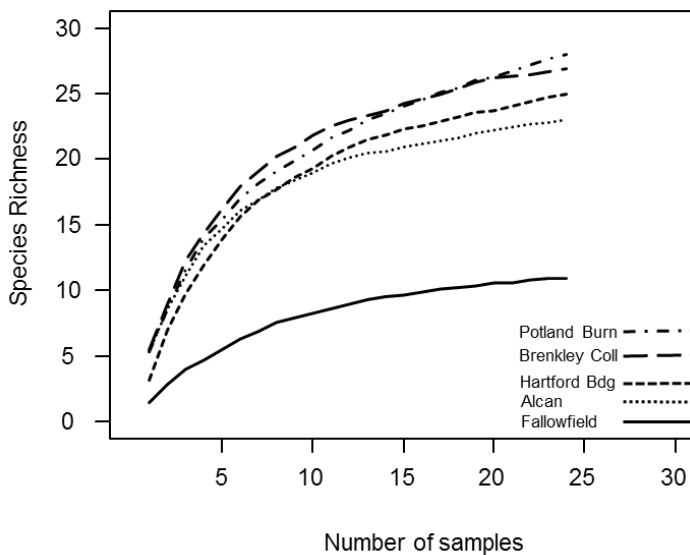
**Table AB.1:** Species list and total abundance of flower-visiting invertebrates recorded during the mowing regime trial, Lynemouth, UK.

<b>Recorded Taxa</b>	<b>Total abundance</b>
<i>Polyommatus icarus</i>	127
<i>Bombus pascorum</i>	57
<i>Bombus lapidarius</i>	199
<i>Eristalis sp.</i>	65
<i>Helophilus sp.</i>	89
Coccinellidae	6
<i>Bombus terrestris / lucorum</i>	58
<i>Episyrphus balteatus</i>	85
<i>Syrphus ribesii</i>	34
Tipulidae	30
Cantharidae	1
micromoth	63
Ichneumonidae	4
<i>Melastoma sp.</i>	13
<i>Pararge aegeria</i>	1
<i>Aglais urticae</i>	8
Orthoptera	25
Tenthredinidae	6
Zygoptera	6
Pieridae	6
<i>Thymelicus sylvestrus</i>	22
<i>Pyronia tithonus</i>	6
<i>Zygaena filipendulae</i>	17
<i>Erynnis tages</i>	1
<i>Eupeodes corollae</i>	7
<i>Platycheirus sp.</i>	16

## Appendix C – Supplementary Information - Chapter 4



**Figure AC.1:** Ariel photograph displaying the location of the five sampling sites used to collect flower-visiting invertebrate and flower density data used within the comparative study (Chapter 4).



**Figure AC.2:** The cumulative number of flower-visiting invertebrate species plotted for each of the 5 sampling locations over the course of the trial undertaken IN Northumberland, UK.

**Table AC.1:** Species list and total abundance of flower-visiting invertebrates recorded at the five sites during the comparative study (Chapter 4).

Taxa Record	Total Abundance				
	Alcan	Brenkley Colliery	Fallowfield	Potland Burn AEI	Hartford Bridge
<i>Helophilus sp.</i>	8	19	4	25	10
<i>Eristalis sp.</i>	15	2	0	9	8
<i>Episyrphus balteatus</i>	26	13	3	37	5
Pipizini	0	7	0	2	4
<i>Syrphus sp.</i>	0	0	0	4	4
<i>Eupeodes sp.</i>	8	4	0	5	2
<i>Melanostoma sp.</i>	0	0	0	0	3
<i>Platycheirus sp.</i>	9	12	0	2	3
<i>Xylota segnis</i>	0	2	0	2	0
<i>B. lapidarius</i>	107	79	6	89	23
<i>B. terrestris / lucorum</i>	36	6	11	64	23
<i>B. pascorum</i>	18	22	26	75	30
<i>B. hortorum</i>	1	0	1	1	8
<i>A. mellifera</i>	0	0	0	0	1
Ichneumonidae	1	4	0	1	3
Tenthredinidae	0	9	0	2	3
Tipulidae	5	18	13	18	9
Empididae	0	0	0	1	0
scathophagidae	2	0	0	0	2
Asilidae	0	0	0	0	1
Micromoth	36	25	8	15	12
<i>Polyommatus icarus</i>	30	14	0	0	0
<i>Erynnis tages</i>	2	5	0	0	0
Peridae	4	11	0	11	2
<i>Pyronia tithonus</i>	4	19	0	10	3
<i>Pararge aegeria</i>	0	1	0	1	0
<i>Zygaena filipendulae</i>	17	6	1	1	0
<i>Aphantopus hyperantus</i>	0	0	1	1	0
<i>Thymelicus sylvestris</i>	6	5	2	4	0
<i>Ochlodes sylvanus</i>	5	4	0	2	1
<i>Aglais urticae</i>	2	0	0	1	1
<i>Maniola jurtina</i>	0	1	0	1	1
<i>Lycaena phlaeas</i>	0	1	0	9	0
<i>Lasiommata megera</i>	0	1	0	18	0
<i>Vanessa cardui</i>	1	0	0	0	0
coccinella sp.	1	1	0	0	2
Cantharidae	0	1	0	0	0

Appendix D – Supplementary Information - Chapter 6



**Figure AD.1:** a) Aerial photograph showing the locations of the three species-poor hedgerows (SP1, SP2, and SP3) and the species-rich hedgerows (SR1, SR2, and SR3) used to determine the value of hedgerows for flower-visiting invertebrates, b) details the positioning of pan traps within three sampling points (Pseudo replicates) along each of the six hedgerows.

**Table AD.1:** Species list and total abundance of flower-visiting invertebrates recorded at the six hedgerows during the one year trial.

Taxa Recorded	Total Abundance					
	SP1	SP2	SP3	SR1	SR2	SR3
Asilidae	12	16	1	181	38	16
Scathophagidae	9	0	18	29	16	30
Empididae	80	23	11	24	125	53
<i>Helophilus sp</i>	26	13	16	80	47	99
<i>Rhingia campestris</i>	1	1	14	15	29	49
<i>Chrysogaster sp</i>	0	0	0	1	2	0
<i>Eupeodes corollae</i>	0	3	5	10	31	13
<i>Syrphus vitripennis</i>	13	6	12	18	12	20
<i>Eristalis tenax</i>	0	0	2	8	3	8
<i>Eristalis arbustorum</i>	0	0	0	1	7	31
<i>Baccha sp</i>	0	3	6	0	2	0
<i>Psyllobora vigintiduopunctata</i>	0	0	0	0	3	0
<i>Pyrophaena granditarsa</i>	0	0	0	0	0	1
<i>Episyrphus balteatus</i>	25	8	44	12	38	55
<i>Eristalis sp</i>	0	1	0	2	5	21
<i>Scaeva sp</i>	0	1	1	0	0	2
<i>Anasimyia SP</i>	0	0	0	0	0	1
<i>Melangyna sp</i>	0	1	4	1	4	2
<i>Chrysotoxum bicinctum</i>	0	0	0	2	0	0
Eumeninae	0	0	0	1	0	0
<i>Tropidia scita</i>	0	0	0	1	1	0
<i>Xylota segnis</i>	1	2	2	3	0	3
<i>Melanostoma sp</i>	0	3	6	11	8	16
<i>Platycheirus sp</i>	10	9	5	19	6	14
<i>Psithyrus sylvestris</i>	1	0	0	2	1	1
<i>Bombus pascorum</i>	2	0	1	15	15	7
<i>Bombus hortorum</i>	0	0	1	2	0	0
<i>Bombus terrestris / lucorum</i>	17	5	8	24	37	58
<i>Bombus lapidarius</i>	15	6	3	20	19	52
<i>Syrphus sp</i>	1	0	2	0	0	1
<i>Apis mellifera</i>	0	0	0	1	0	1
<i>Andrena sp</i>	0	0	0	2	2	1
Ichneumonidae	107	48	57	118	185	93
Tenthredinadae	17	6	14	13	82	59
Panorpidae	1	1	7	1	92	38
Cantharidae	1	0	3	7	1	2
Noctidae	0	2	1	3	3	2
<i>Pieris . rapae</i>	0	0	0	1	0	9
<i>vespula sylvestris</i>	5	2	0	1	1	4
<i>Volucella bombylans</i>	0	0	0	1	0	1
<i>Vespula vulgaris</i>	3	3	0	14	0	1
<i>Philianthus sp</i>	0	0	0	5	3	0
<i>Neoascia podagrica</i>	0	0	2	0	0	2
<i>Halyzia 16-guttata</i>	0	0	0	0	0	1
<i>Pieris napi</i>	1	0	0	2	0	3
<i>Aphantopus hyperantus</i>	2	1	0	1	0	1
<i>Lasiommata megera</i>	0	0	0	0	0	2
<i>Aglais urticae</i>	0	0	0	0	0	1

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