The decline and conservation of bumblebees

D. Goulson*, G.C. Lye, B. Darvill

School of Biological & Environmental Sciences, University of Stirling, Stirling, FK9 4LA, UK;

Email: Dave.Goulson@stir.ac.uk

*Corresponding author

Running title: Conservation of bumblebees

Key Words Hymenoptera, *Bombus*, rarity, population structure, habitat loss

Contents

ABSTRACT

Declines in bumblebee species in the last 60 years are well documented in Europe, where they are primarily driven by habitat loss and declines in floral abundance and diversity resulting from agricultural intensification. Impacts of habitat degradation and fragmentation are likely to be compounded by the social nature of bumblebees and their largely monogamous breeding system which renders their effective population size low. Hence populations are susceptible to stochastic extinction events and inbreeding. In North America, catastrophic declines of some bumblebee species since the 1990s are probably attributable to the accidental introduction of a non-native parasite from Europe, a result of global trade in domesticated bumblebee colonies used for pollination of greenhouse crops. Given the importance of bumblebees as pollinators of crops and wildflowers, it is vital that steps be taken to prevent further declines. Suggested measures include tight regulation of commercial bumblebee use and targeted use of agri-environment schemes to enhance floristic diversity in agricultural landscapes.

INTRODUCTION

The world bumblebee (*Bombus*) fauna consists of approximately 250 known species, largely confined to temperate, alpine and arctic zones of the northern hemisphere (99). There is mounting evidence that many bumblebee species have declined in recent decades, particularly in developed regions such as Western Europe and North America (reviewed in 34, 54, see also 88). The most comprehensive records available are from the UK, where between 1970 and 1974, data were collected from 10km squares comprising most of the British Isles (1). These have been compared with a considerable body of 'pre 1960' records (98). The comparison revealed a dramatic decline in the distributions of many species. More recent data suggest that this decline has continued, with three of the 25 UK species having gone extinct and a further eight species having undergone major range declines (34). The most severely affected species tend to be those with long tongues associated with deep perennial flowers (40). Similar patterns are evident in Europe. In a review of declines in bumblebees of 11 central and western European countries, Kosior et al. (54) describe extinctions of 13 species in at least one country between 1950 and 2000. Four species (*B. armeniacus*, *B. cullumanus, B. serrisquama, B. sidemii*) went extinct throughout the entire region.

There are no equivalent baseline data for bumblebees in North America and there is debate as to whether or not they are suffering similar long-term declines. For example, Golick and Ellis (33) found little variation in the Nebraska bumblebee fauna between 1962 and 2000. In contrast, there is strong evidence for precipitous declines of some North American bumblebee species. *B. franklini* is endemic to a small area in the west of the US, and declined rapidly since 1998. Recent searches found none at many former strongholds, and it is now thought to be extinct at many or all of them (87). *B. occidentalis* is native to the west of North

America and was once the commonest bumblebee here, but since the late 1990’s it has declined dramatically, and is now extremely rare (88, 89). *B. affinis* and *B. terricola*, both eastern North American species, are suffering similar declines, as have *B. sonorus* and *B. pensylvanicus* (88). *B. ashtoni*, a social parasite of *B. affinis* and *B. terricola*, also appears to be suffering losses, presumably in response to decline of its hosts (101).

CONSEQUENCES OF DECLINES IN BUMBLEBEE POPULATIONS

A large number of wild plants are pollinated predominantly or exclusively by bumblebees, sometimes by particular species of bumblebee (34). Most bumblebees are generalist pollinators and most insect-pollinated plants use multiple pollinators (91), so it could be argued that pollination networks are buffered against the loss of a few pollinator species. However, a recent study simulating the effects of removal of individual pollinators from pollination networks demonstrated that removal of highly linked pollinators such as bumblebees produced the greatest rate of decline in plant species diversity (59). Reduced pollination services can be particularly detrimental when plants are already scarce and threatened directly by the same changes in land use that threaten the bees (34).

Aside from the implications for conservation, there are good financial reasons for conserving bumblebees. The yields of many field, fruit and seed crops are enhanced by bumblebee visitation (36). For example, field beans in Europe are largely pollinated by longer tongued species such as *B. pascuorum* and *B. hortorum*, without which, yields are poor (28). In the US there is an ongoing decline in managed honeybee populations due to disease, misuse of pesticides, loss of subsidies and dangers associated with invading Africanized honeybees (55). The value of crop pollination by honeybees in the US has been estimated at between 5 and 14 billion dollars per year, but beekeeping has diminished by around 50% over the last 50 years (55). This has given rise to concerns over the future of insect-pollinated crops such as cucumber, pumpkin, watermelon, blueberry and cranberry (18, 55, 73). At sufficient densities, bumblebees pollinate many of these crops efficiently, often more so than honeybees (81). However, the impoverished bumblebee communities often associated with agricultural landscapes may be insufficient to replace the services currently provided by honeybees.

CAUSES OF BUMBLEBEE DECLINES

Reductions in floral resources

Most researchers are convinced that the primary cause of bumblebee declines in Western Europe is the intensification of farming practices, particularly during the latter half of the 20th century (34, 37). In the UK, a self-sufficiency drive in the wake of the Second World War led to a number of major changes. Permanent unimproved grassland was once highly valued for grazing and hay production but the development of cheap artificial fertilizers and new fast-growing grass varieties meant that farmers could improve productivity by ploughing up ancient grasslands. Hay meadows gave way to monocultures of grasses which are grazed or cut for silage. Between 1932 and 1984 over 90% of unimproved lowland grassland was lost in the UK (46). Grants were introduced to grub out hedgerows, to plough and re-seed pasture and to drain marshy areas. This led to a steady decline in the area of unfarmed land and of unimproved farmland.

In North America, agricultural intensification has caused similar loss and fragmentation of natural and semi-natural habitat and an associated loss in biodiversity, at least in some regions (97). For example in Iowa, 85% of the land area was once prairie grassland which provides good bumblebee habitat, but less than 0.1% now remains. The remainder of the land is largely covered with monocultures of crops or by urban areas (44).

There is evidence to suggest that bumblebee forage plants have suffered disproportionate declines. A recent study in the UK found that of 97 preferred bumblebee forage species, 71% have suffered range restrictions, and 76% have declined in abundance over the past eighty years, exceeding declines of non-forage species (11).

On farmland, the crops themselves may provide an abundance of food during their brief flowering periods. Leguminous crops (notably clovers, *Trifolium* spp.) used to be an important part of crop rotations in much of Europe, and these are highly preferred food sources, particularly for long-tongued bumblebee species (40). Since the introduction of cheap artificial fertilizers, rotations involving legumes have been almost entirely abandoned, and it has been argued that this is one of the primary factors driving the decline of long-tongued bumblebees (38, 72). It has been suggested that flowering crops such as oilseed rape contribute substantially to supporting bumblebee populations in arable landscapes (92). However, in order for bumblebee colonies to thrive, a continuous succession of flowers is required from April until August, and crops alone are unlikely to provide this. Farms must contain areas of wildflowers if they are to support bumblebee populations.

Uncropped areas of farmland, such as hedgerows, field margins and borders of streams may provide flowers throughout the season, and therefore support greater numbers of foraging bumblebees than cultivated areas (3, 56). However, these areas will be adequate only if there are enough of them, and if they have not been degraded by drift of herbicides and fertilizers. Insufficient flower-rich uncropped areas may lead to gaps in the succession of flowering plants during which bumblebee colonies may starve and die. With a decline in bees, the plants that they pollinate set less seed, resulting in less forage for the bees in subsequent years (63). The process by which mutually-dependent species drive each other to extinction is known as an 'extinction vortex'. We do not as yet know whether this process is really occurring, but it is clear that farmland provides less food for bees than it once would have done.

Loss of nest sites

In addition to floral resources, bumblebees need suitable nesting sites, the precise requirements for which vary between species (50). The carder bees (*Thoracobombus*) such as *B. pascuorum* tend to nest in dense grassy tussocks while other species such as *B. terrestris* nest underground in cavities. Both groups often use abandoned rodent nests. The loss of hedgerows and of unimproved pastures is likely to have reduced availability of nest sites for both above and below-ground nesting bumblebee species (3). Those species that nest above ground frequently have their nests destroyed by farm machinery, particularly by cutting for hay or silage. The scarcity of weeds and field-margin flowers on modern intensive farms means that there are less seeds, and therefore less food for voles and mice. Lower populations of these mammals will lead to fewer nest sites for both above and below-ground nesting bumblebee species.

In California, even on organically managed farms, the presence of bumblebees, in this case *B. vosnesenskii* and *B. californicus*, depends on proximity to areas of natural habitat in which the bees can nest (55). A study in Sweden found that field boundaries within 100m of a semi-natural grassland area contained a greater abundance and diversity of foraging bumblebees than similar sites > 1 km from such habitat. However, bumblebee abundance was found to be significantly lower in the semi-natural grasslands themselves suggesting that these sites were used primarily for nesting (62). Similarly, Greenleaf and Kremen (42) found that tomato fields in northern California obtain high visitation rates from *B. vosnesenskii* only when they were positioned within 300m of a patch of natural habitat and if at least 40% of the land within a 2100m radius of the farm was natural habitat.

There is some evidence for a paucity of suitable nest sites in urban areas. Bumblebee abundance in urban parks in San Francisco has been found to be positively correlated with the number of rodent holes (58) suggesting that nest sites may be a limiting factor.

Pesticides

Pesticide risk assessments are routinely carried out for honeybees, but the results of these are probably not directly applicable to bumblebees (86). For example, to avoid honeybees, pyrethroids are commonly applied to flowering oilseed rape in the early morning or evening, when bumblebees are often active. Laboratory and field-based bioassays appropriate to bumblebees have been developed in response to the growing use of bumblebees for the pollination of greenhouse crops, but these are not widely used and few toxicological data are available (85). Almost all tests conducted so far have been on *B. terrestris,* and suggest that toxicity is similar to that found in honeybees. There are three possible routes for exposure, through: direct contact with sprays (on flowering crops or adjacent wild flowers); contact with contaminated foliage; uptake of chemicals in nectar. The latter is most likely with systemic insecticides. Tests with dimethoate and carbofuran suggest that these chemicals are selectively transported into the nectar where they can reach high concentrations (17). Given the large volume of nectar consumed by bumblebees, this could prove to be the most important route of exposure.

When colonies are large it is likely that they can tolerate the loss of some of their workers. However, in the spring when queens are foraging, and subsequently when nests are small and contain just a few workers, mortality may have a more significant effect (85). Thus spring applications of pesticides are of particular concern.

Despite risk assessments, widespread poisoning of honeybees has been reported (27). Such effects are obvious in domestic hives where dead bees are ejected and form piles by the nest. It seems probable that pesticides would have similar effects on bumblebees but they are unlikely to be noticed in most situations. In Canada, the use of the insecticide fenitrothion in forests led to a decline in yield of nearby *Vaccinium* crops due to a reduction in abundance of bumblebee pollinators (21). In the UK, bumblebee deaths have been reported following applications of dimethoate or alphacypermethrin to flowering oilseed rape, and of λ-cyhalothrin to field beans (85, 86).

A growing appreciation of the damaging effects of broad-spectrum pesticides has led to the development of a new generation of more target-specific compounds. EU, US and Canadian law now demand that oral and acute toxicity tests are carried out on honeybees prior to the registration of any new pesticide (61). However, there is no obligation to study sub-lethal effects on any bees, or to look at specific effects on bumblebees. Some of these substances cause no mortality in bumblebees if used appropriately (26, 76), but there is evidence that supplementary trials for non-lethal effects are necessary. For example, spinosad is a commonly used insect neurotoxin which, based on studies of honeybees, has been deemed harmless to bees. However, it has recently been shown that bumblebee larvae fed with pollen containing this pesticide give rise to workers with reduced foraging efficiency (61). Mommaerts *et al.* (60) screened eight chitin synthesis inhibitors currently registered as pesticides and found that although no lethal effect could be found on adults, the use of these pesticides has strong effects on colony growth and the development of larvae. Diflubenzuron and teflubenzuron were found to be the most harmful to bumblebees, greatly reducing reproductive output at concentrations far below the recommended field concentrations. In summary, it is likely that many pesticides currently in use do impact on bumblebee populations, but hard data are largely lacking.

Impacts of non-native bees

It has been argued that the most immediate threat to bumblebees in the US, unlike in Western Europe, is the spread of disease due to widespread trafficking of commercial bumblebee hives (88). Commercial bumblebee hives are used for greenhouse pollination all over the world, including; Israel, Korea, Japan, North America and Europe (35). In the US, colonies of *B. impatiens* and *B. occidentalis* have been commercially reared since the early 1990s for the pollination of greenhouse crops such as tomatoes (93) and sweet peppers (78). These colonies have been found to have a greater parasite load than wild colonies with an elevated prevalence of the bumblebee specific protozoan pathogens *Crithidia bombi* and *Nosema bombi*, and of the tracheal mite *Locustacarus buchneri* (12). These parasites have detrimental effects on colony survival and reproduction and/or the foraging efficiency of individual workers (5, 29, 65).

There is little doubt that there are greenhouse escapes. In Japan, feral colonies of the non-native *B. terrestris* are now common (47), and a recent study in Canada found that 73% of pollen carried by workers returning to commercial colonies originated from plants outside the greenhouse (94). Consequently, there is a high likelihood of interaction between wild and commercially reared bees at flowers, providing conditions for ‘pathogen spillover’ from the commercial population to wild populations. Significant increases in the prevalence of *C. bombi* and *N. bombi* have been found in wild bumblebee populations near to commercial greenhouses, compared to wild populations elsewhere (12). In 1998, a *N. bombi* outbreak was reported in bumblebee production facilities in North America. This was thought to be a result of the importation of infected European *B. terrestris* colonies into Mexico in 1995 and 1996 (101). Similarly, *C. bombi* has only been detected in the US since use of commercially reared bumblebees began and it is suspected that this parasite is not native to the US (101).

The introduction may have occurred as a result of the shipment of queens of *B. occidentalis* to Europe for commercial rearing before re-importation into the US in the early 1990s (12, 101). Exposure to a non-native pathogen is a likely cause of the catastrophic declines in *B. terricola*, *B. affinis, B. franklini* and *B. occidentalis* (87, 88, 93). However, we have a very poor understanding of the relative susceptibilities of bumblebee species to parasites, or of the natural distributions of these parasites, and work is urgently needed in this area. It is clear that tight controls are needed on transport of domesticated bumblebee hives.

There are other dangers associated with commercial trafficking of bumblebees. *B. terrestris* is now naturalized in Japan and there are concerns regarding possible competitive effects of this species on native bumblebees. Studies have shown that *B. terrestris* has four times the reproductive output of native species (57) and that there are considerable overlaps in forage use and timing of foraging (47). There are similar concerns in the UK where around 10,000 colonies of a south-eastern European subspecies *Bombus terrestris dalmitinus* are imported each year (49). Britain has an endemic subspecies of this bee, *Bombus terrestris audax*. Evidence suggests that there are dangers to the British subspecies in the form of parasite transmission (48) or out-competition, particularly since the introduced subspecies has superior foraging efficiency and reproductive rate (49). Also, *B. terrestris dalmitinus* and *B. terrestris audax* readily interbreed, so the native subspecies could be lost through introgression (48).

In addition to non-native bumblebee species, native pollinator communities in many parts of the globe also have to contend with other introduced bee species, most notably honeybees (*Apis mellifera*). These natives of Europe, Africa and the Middle East have been introduced by man to almost every country in the world. Their impacts are reviewed by Goulson (35). Recent studies suggest that honeybees can have negative effects on bumblebees. Walther-Hellwig *et al.* (90) found that short-tongued bumblebees avoided areas of forage close to honeybee hives, while carder (*Thoracobombus*) bumblebees switched to foraging later in the day and were displaced from their preferred foodplant. Thomson (83) experimentally introduced honeybees and found that proximity to hives significantly reduced the foraging rates and reproductive success of *B. occidentalis* colonies. In eastern US, Thomson (84) found a strong overlap between the foraging preferences of the two species, which peaked at the end of the season when floral resources were scarce, corresponding with a negative relationship between honeybee and bumblebee abundance.

There is increasing evidence that the spread of natural enemies of bumblebee colonies is being aided by honeybees. Honeybees can act as vectors for the bumblebee specific *C. bombi* via flowers (74). The African honeybee parasite *Aethina tumida* (small hive beetle) recently invaded North America, Egypt, Australia and Europe, and attacks *B. impatiens* colonies where it causes considerable damage (79). Deformed wing virus, a viral honeybee pathogen, has been found in commercial colonies of *B. terrestris*, transmitted between the two species as a result of the practice of placing honeybees with queens to induce colony founding (30). However it has also been found in a wild colony of *B. pascuorum* which had been robbing a managed honeybee hive (30). This virus appears to have higher virulence to bumblebees than to honeybees and the findings raise important questions about transmission and cross-infectivity between bumblebees and honeybees.

Habitat fragmentation and population structure

As a consequence of the various factors discussed above, populations of a number of bumblebee species have become increasingly small, fragmented and separated from one another by large distances. In the UK, where distributions are best known, declines appear to have followed a characteristic pattern. The last bumblebee species to disappear from the UK (*B. subterraneus*), was once widespread across southern England, but declined rapidly in the years after World War II. By the 1980's the few remaining populations were small and isolated, surviving on habitat islands (nature reserves) that had escaped agricultural intensification. However, these populations subsequently disappeared despite the apparent suitability and protected status of the remaining habitat (34). The species was last recorded at Dungeness National Nature Reserve in 1988. Several other UK species such as *B. distinguendus* and *B. sylvarum* are in the late stages of a similar process, and are likely to go extinct in the near future. Why do isolated populations go extinct? Understanding the consequences of the fragmentation of remnant populations of bumblebees is of great importance to conservationists, given the current distributions of many rare species.

Small populations of all taxa are inherently more vulnerable to local extinctions due to environmental and demographic stochasticity (25). If these populations form part of a broader metapopulation then regional extinctions can be balanced by subsequent recolonisation, but if fragmentation is severe then extinct patches may never be repopulated. In addition, a functioning metapopulation ensures that dispersal maintains genetic cohesion. However, if habitat fragmentation results in the isolation of populations, then they may face an additional extinction threat through inbreeding (25). There are a number of reasons why bumblebees may be particularly badly affected by habitat fragmentation. It is the effective population size (N_e), rather than the census population size (N_c) which determines the rate of genetic drift in a population, and N_e may be several orders of magnitude lower than N_c. In bumblebees, as in many other social insects, N_e depends on the number of successful colonies. The N_e contributed by an individual colony depends on the number of egg-laying queens and the number of males they have mated with, but (unlike many other hymenopterans) bumblebee colonies are all founded by a single queen, and the vast majority of species are monoandrous (23, 75). Furthermore, as a consequence of haplodiploidy, their N_e is equal to the number of successful nests x 1.5, not x 2 as would be the case for a diplo-diploid organism. It seems therefore that population sizes of bumblebees may be low, even relative to other social insects, making them particularly susceptible to the loss of genetic diversity.

To date, relatively few studies have attempted to determine the consequences of inbreeding in bumblebees. Gerloff & Schmid-Hempel (32) found significant reductions in colony foundation success and hibernation success in response to brother-sister mating. Little evidence of inbreeding depression was found for reproductive output or cumulative fitness, as was the case for Duchateau *et al.* (19), but in both instances nests were reared in the lab and fed *ad libitum.* However, Beekman et al. (4) found that inbred queens laid fewer eggs. Gerloff *et al.* (31) found no evidence for a reduced encapsulation (immune) response. A recent meta-analysis concluded that, although haplo-diploid insects suffer less from inbreeding than diploid insects (perhaps due to purging of non-sex limited recessive alleles in haploid males (66)), substantial inbreeding depression does occur (43).

An additional cost may be imposed on inbred populations of many hymenopteran species as a result of their haplo-diploid sex determination mechanism. The mechanism centers on a polyallelic sex-determining locus, and has important consequences for small populations (13). Individuals heterozygous at this locus develop into females, and homozygous (or hemizygous) individuals develop into males. As populations diminish in size, genetic drift will lead to a reduction in the number of sex alleles in the population, increasing the probability of a ‘matched mating’. A queen that mates with a male who shares one of her sex determining alleles will produce a colony in which 50% of her workforce are diploid males. In honeybees and ants, diploid male larvae are consumed by the workers, which minimizes their cost, but in bumblebees they are reared to adulthood (19). Bumblebee diploid males are viable but effectively sterile (but see 2), and therefore represent a considerable cost to the colony best viewed as 50% worker mortality (66).

Diploid males represent a clear example of inbreeding depression, and have been detected in numerous wild populations of hymenopterans (e.g. 102, 80). Their frequency has been proposed as an indicator of population fitness (104) and recent modeling work has shown that diploid male production, where present, may initiate a rapid extinction vortex (103). However, until recently, diploid male production had not been detected in naturally occurring populations of bumblebees.

Given the potentially serious consequences of inbreeding in bumblebees, it is essential that we understand its prevalence within wild bumblebee populations. The development of several highly variable microsatellite markers for bumblebees (22) has facilitated the assessment of their population structure. Initial studies focused largely on two abundant and widespread European species, *B. terrestris* and *B. pascuorum*. In *B. terrestris,* there appears to be little population substructuring within mainland Europe, suggesting that dispersal is frequent and that there are no substantial isolating barriers between populations (24). However, populations on various Mediterranean islands and Tenerife (Canary Islands) were distinct (24, 96). In *B. pascuorum*, populations throughout most of mainland Europe are similar, but differ markedly from those found south of the Alps in Italy (68, 95). More recently Shao *et al.* (77) compared seven mainland and island populations of *B. ignitus* in Asia and similarly found that mainland populations were genetically similar, but distant offshore populations had significantly differentiated. It seems therefore that genetic structuring is observed when populations are separated by appreciable barriers, like mountain ranges or large stretches of water.

Until recently, studying the population genetics of rare bee species was extremely difficult, as lethal sampling was necessary. Work in this area was greatly aided by the development of a non-lethal DNA sampling technique (45), and this has recently been applied to studies of fragmented populations of rare species: *B. muscorum* (16), *B. sylvarum* (20) and

B. distinguendus (6). All three studies found significant population structuring. For example in *B. muscorum,* all populations >10 km apart were significantly differentiated, as were some populations just 3km apart. Low frequencies of diploid males were found in 3 of the 16 studied populations. Ellis *et al.* (20) used microsatellite markers to group workers into sisterhoods and so estimated the number of colonies (and hence N_e) in populations of *B. sylvarum*, a species which is highly endangered in the UK. Estimates of N_e were very low (range 21-72) suggesting that, if isolated, these populations are very vulnerable to loss of genetic diversity through drift. Indeed, significant differentiation was found between all populations suggesting that they are genetically isolated. Diploid males were found at one of the 6 sample sites. It is important to exercise caution when making comparisons between species based on a small number of microsatellite markers. However, in all three rare species, genetic diversity (allelic richness and heterozygosity) was reduced compared to common species, and island populations showed further reductions (Table 1).

We do not as yet have unequivocal evidence that inbreeding plays a major role in driving small, isolated populations of bumblebees to extinction, but it seems likely. If reductions in the genetic diversity of neutral markers found in rare species are indicative of reductions in the diversity of functional genes, then there will be concomitant consequences for population fitness and evolutionary potential. If fragmented populations of rare bumblebee species are suffering from reduced fitness through inbreeding then we must take steps to conserve what genetic diversity remains. Management strategies in vertebrates routinely consider genetic factors, and we may need to adopt similar measures in the management of rare bumblebee populations.

Why do some bumblebee species remain common?

Some bumblebee species appear to have been largely unaffected by habitat loss, fragmentation and degradation. In much of Europe, six species are widespread and common (*B. terrestris, B. lucorum, B. lapidarius, B. pratorum, B. hortorum* and *B. pascuorum*). How do these species differ from those that have declined? Based on studies of forage use, Goulson et al. (39, 40) argue that the rare species tend to be long tongued and have narrower diets, with a very large proportion of the pollen they collect being from Fabaceae (many of which have deep flowers). These bumblebee species are associated with Fabaceae-rich unimproved grasslands, a habitat which has been very largely eradicated in western Europe. In contrast, the common species tend to have broad foraging preferences and readily encompass non-native garden plants and mass-flowering crops in their diets (41). Williams (100) recently showed that rare and declining species in Britain tend to have small geographic ranges within Europe. He suggests that these species may have more specific habitat associations or climatic requirements, which render them more susceptible to environmental change. Of course these two explanations are mutually compatible since a species with a narrow diet is also likely to have specific habitat requirements. However, current data suggest that most bumblebee species are not strongly associated with particular habitat types (39). For example, prior to its extinction in Britain, *B. subterraneus* occurred in habitats as diverse as shingle, saltmarshes, sand dunes, and calcareous and neutral unimproved meadows. Although some of the rarer species do appear to exist in very specific habitats, historical records show that most once existed across a much wider range of biotopes (39).

In the UK there seems to be a broad correlation between rarity and emergence time, with rare species tending to emerge later (40). Bumblebee species differ greatly in the time of year at which queens emerge from hibernation; the earliest species emerge in February, while other species do not appear until late May. Where nesting habitat is scarce, those species in which queens emerge early in the season may be able to monopolize available nest sites, reducing the chances of colony founding for later emerging queens. In urban parks in San Francisco, *B. vosnesenskii* is the most abundant species, and is also the earliest emerging of the community of bumblebees found there (58). Rodent holes were found to limit bumblebee abundance and it is suggested that the earliest emerging species monopolizes nest sites.

In recent years it has become apparent that there are major differences between bumblebee species in their foraging range (15, 53). Species such as *B. terrestris* and *B. lapidarius* have been found to forage further afield than so-called “doorstep foragers” such as *B. pascuorum*, *B. sylvarum*, *B. ruderarius* and *B. muscorum*. It is perhaps significant that the former two species remain ubiquitous in much of Europe, while three of the four doorstep foragers have declined. A larger foraging range would give a greater chance of colony survival in areas where the average density of floral resources is low or where resources are highly patchy. Intensively farmed arable landscapes with occasional fields of mass-flowering crops provide just such a landscape, and it is probably no coincidence that *B. terrestris* and *B. lapidarius* are among the species most commonly recruited in large numbers to such crops (39).

CONSERVING BUMBLEBEES

Enhancing bumblebee diversity in farmland

A major cause of bumblebee declines is undoubtedly loss of habitat to intensive farming. However, there are moves to reverse this trend in Europe and North America where there is a growing emphasis on combining the goals of agriculture and conservation (52, 64). Subsidies are currently available in many countries for agri-environment schemes that promote biodiversity, including replanting of hedgerows, leaving land fallow, sowing wildflower strips, and restoring flower-rich grassland. Most of the management options promote floral abundance and diversity. It has been found that a 6m wide field margin kept free of crops and agrochemicals may contain six times as many flowering plants and ten times as many flowers than the equivalent cropped area (51). The effects of UK field margin management options on bumblebee communities have been the focus of many studies in recent years.

The most valuable form of field margin management for bumblebees has been found to be the sowing of either wildflowers or a pollen and nectar mix consisting of agricultural cultivars of legume species (7, 9, 10, 70, 71). Carvell et al. (10) found that the pollen and nectar mixture produced the highest flower abundance with a succession of forage plants flowering over the 3-year trial period. The wildflower mixture produced few flowers in the first year but flower abundance increased over the three years as the mixture became established. Both treatments led to an increase in bumblebee species richness and abundance, and in the third year the wildflower mix was as valuable as the pollen and nectar mixture. Once established, the wildflower mix should persist for up to ten years, while the agricultural cultivars in the pollen and nectar mix are likely to need re-sowing within 5 years (69).

Long-term set-aside (lasting five years or more) can also produce the mid-successional communities preferred by bumblebees (63). However, the perennial plants vital to such communities are poorly represented in the seed banks in agricultural land, thus establishment of such species may be slow. To maintain mid-successional communities, intermediate levels of disturbance such as mowing, cutting or seasonal grazing may be required. Carvell (8) conducted a study to assess the benefits of different grassland management regimes on bumblebee populations in the UK. Grazing during the autumn and winter months was found to provide excellent bumblebee habitat, but without it coarse grasses became dominant. However, heavy grazing during the spring and summer is detrimental since most flowers are eaten. An alternative is rotational grazing through the spring and summer so that there are always some parts of the farm providing forage for bees. In general, grazing by cattle seems to be more favorable to bumblebees than grazing by sheep, and low summer grazing densities are preferable (8).

Studies of forage use by bumblebees suggest that it is not necessarily important to provide a great diversity of flowers (38, 40). In coastal scrubland in California, five plant species were found to account for between 80 and 93% of bumblebee visits over a three year study period (84). Similarly, in studies of 15 bumblebee species across a broad range of habitats in the UK, 80% of all pollen collecting visits were to just 11 plant species (40). Carvell et al. (10) found that 92% of visits were to just six flowering plants in managed field margins and that a diverse sown wildflower field margin option consisting of eighteen herb species was no more beneficial than a simple sown wildflower option consisting of only three herbaceous species. Few, well chosen forage species provide suitable resources for a diverse bumblebee community.

Bumblebees not only require a suitable source of forage, but also nest and hibernation sites. A popular agri-environment scheme in the UK is the sowing of field margins with tussocky grasses (71). These habitats attract the small mammals whose abandoned holes are used by bumblebees for nest sites (82), so it is likely that this form of management is of value to bumblebees. Carvell *et al.* (9) found that field margins sown with a 'split' treatment consisting of a mixture of tussocky grasses and wildflower mix attracted almost as many bumblebees as margins sown solely with the wildflower seed suggesting that it is possible to provide both forage and nesting habitat in small areas.

The importance of urban areas

In the US, 2.2 million acres of farmland and open space are converted into urban areas every year (58). Although conservationists would generally view this negatively, there is evidence that gardens and urban parks are particular strongholds for some species of bumblebee. Urban parks in San Francisco were found to have higher mean abundances and equal diversities of bumblebees, compared to nearby 'wilder' areas. Abundance of bumblebees was partly explained by the openness of the matrix surrounding the park, suggesting that suburban gardens were also playing a role (58). In the UK, young nests of *B. terrestris* placed in suburban gardens grew more quickly and attained a larger size than nests placed in arable farmland (41). It is likely that gardens provide favorable habitat for several bumblebee species as a result of the density, variety and continuity of flowers that they provide (41). However, many commonly used garden plants are unsuitable for bumblebees. Artificial selection has often resulted in modern flower varieties which provide little or no reward, or which are inaccessible to insects (14). Similarly, some exotic plants, such as those pollinated by hummingbirds, provide rewards that are inaccessible to native species. It is clear that urban gardens can provide a refuge for several bumblebee species but encouraging gardeners to choose their plants appropriately could be particularly beneficial.

CONCLUSIONS

Widespread declines of bumblebee species threaten pollination services to both wildflowers and crops. It is clear from studies of population structure that most bumblebee species cannot be conserved by managing small protected 'islands' of habitat within a 'sea' of unsuitable, intensively farmed land. Large areas of suitable habitat are needed to support viable populations in the long term. Also, studies of foraging range indicate that bumblebees exploit forage patches at a landscape scale, so that the scale of management must be appropriate.

An integrated approach across large areas or several farms is more likely to succeed than localized efforts. Where small, isolated populations of rare species remain in habitat fragments, targeting the adjacent farms for uptake of suitable agri-environment schemes could increase the population size and so reduce the likelihood of stochastic extinction events and inbreeding. Similarly, such schemes could be used to provide linkage between habitat islands.

Unimproved flower-rich grassland is one of the most important habitats for bumblebees, but has been largely lost to agriculture in Western Europe and North America. Restoration of areas of this habitat will boost bumblebee populations and has been shown to provide improved pollination services on nearby farmed land. Substantial benefits could also be obtained by reintroducing clover (e.g. *Trifolium pratense*) ley crops into rotations, since this is a key forage source for many declining bumblebee species. This would also reduce dependency on artificial fertilizers.

Pesticide poisoning is likely to have contributed to bumblebee declines, and the current risk assessments of the dangers of pesticides to honeybees are inadequate for bumblebees. In addition there is a clear need to assess sub-lethal effects of pesticides on bumblebees.

In the US, recent declines in several bumblebee species have been linked to increases in the commercialization of bumblebees for greenhouse pollination and associated introductions of parasites. There are already restrictions in place on the importation and movement of bumblebees in Canada, Mexico and the US but there are calls for increased restrictions on transportation of bees and for stricter quarantine and monitoring systems (101).

Finally, long-term monitoring of bumblebee populations is required in order to build up a picture of the current status of bumblebee species and to establish baselines to which future studies can refer. This is particularly important in areas such as North America where declines of some species have been documented but the extent is poorly quantified.

SUMMARY POINTS

1. Many bumblebee species have declined in western Europe and North America
2. Declines are largely attributable to agricultural intensification, and in North America probably to introduction of parasites with commercial bumblebee nests
3. Bumblebees have a low effective population size, rendering them prone to stochastic extinctions and inbreeding
4. Conservation measures must be coordinated and deployed at a landscape scale to be effective
5. Improved safety testing of pesticides is needed
6. Tight controls on the international trade in commercial bumblebee colonies are required to prevent further introductions of non-native parasites

UNRESOLVED ISSUES

1. Baseline surveys of bumblebee abundance and distributions are lacking for most countries, so that quantifying population change is difficult
2. It is not yet clear whether inbreeding depression is playing a major role in declines of bumblebees
3. Agricultural intensification continues apace in developing countries in Eastern Europe and Asia, and unless lessons are learned from developed countries then further declines in bumblebees are inevitable
4. Basic aspects of the ecology of many species are not known, so that conservation measures are poorly informed

LITERATURE CITED

1. Alford DV. 1980. *Atlas of the Bumblebees of the British Isles.* Institute of Terrestrial Ecology Publications, No. 30, UK
2. Ayabe T, Hoshiba H, Ono M. 2004. Cytological evidence for triploid males and females in the bumblebee, *Bombus terrestris*. *Chromosome Res.* 12:215-223
3. Banaszak J. 1992. Strategy for conservation of wild bees in an agricultural landscape. *Agric. Ecosyst. Environ.* 40:179-192
4. Beekman M, van Stratum P, Veerman A. 1999. Selection for non-diapause in the bumblebee Bombus terrestris, with notes on the effect of inbreeding. *Entomol. Exp. Appl.* 93:69-75
5. Brown MJF, Schmid-Hempel R, Schmid-Hempel P. 2003. Strong context-dependent virulence in a host-parasite system: reconciling genetic evidence with theory. *J. Anim. Ecol.* 72:994-1002
6. Bourke AFG, Hammond RL. 2002. *Genetics of the scarce bumble bee, Bombus distinguendus, and nonlethal sampling of DNA from bumble bees*. A Report for the RSPB, January 2002.
7. Carreck NL, Williams IH. 2002. Food for insect pollinators on farmland: insect visits to flowers of annual seed mixtures. *J. Insect Conserv.* 6:13-23
8. Carvell C. 2002. Habitat use and conservation of bumblebees (*Bombus* spp.) under different grassland management regimes. *Biol. Conserv.* 103:33-49
9. Carvell C, Meek WR, Pywell RF, Nowakowski M. 2004. The response of bumblebees to successional change in newly created arable field margins. *Biol. Conserv.* 118:327-339
10. Carvell C, Meek WR, Pywell RF, Goulson D, Nowakowski M. 2007. Comparing the efficiency of agri-environment schemes to enhance bumble bee abundance and diversity on arable field margins. *J. Appl. Ecol.* 44:29-40

11. Carvell C, Roy DB, Smart SM, Pywell RF, Preston CD, Goulson D. 2006. Declines in forage availability for bumblebees at a national scale. *Biol. Conserv.* 132, 481-489

12. Colla SR, Otterstatter MC, Gegear RJ, Thomson JD. 2006. Plight of the bumble bee: Pathogen spillover from commercial to wild populations. *Biol. Conserv.* 129:461-467

13. Cook JM, Crozier RH. 1995. Sex determination and population biology in the Hymenoptera. *TREE* 10:281-286

14. Corbet SA, Bee J, Dasmahapatra K, Gale S, Gorringe E, La Ferla B, Moorhouse T, Trevail A, Van Bergen Y, Vorontsova M. 2001. Native or exotic? Double or single? Evaluating plants for pollinator-friendly gardens. *Ann. Bot.* 87:219-232

15. Darvill B, Knight ME, Goulson D. 2004. Use of genetic markers to quantify bumblebee foraging range and nest density. *Oikos* 107:471-478

16. Darvill B, Ellis JS, Lye GC, Goulson D. 2006. Population structure and inbreeding in a rare and declining bumblebee, *Bombus muscorum* (Hymenoptera: Apidae). *Mol. Ecol.* 15:601-611

17. Davis AR, Shuel RW. 1988. Distribution of carbofuran and dimethoate in flowers and their secretion in nectar as related to nectary vascular supply. *Can. J. Bot.* 66:1248-1255

18. Delaplane KS, Mayer DF 2000. *Crop pollination by bees.* CABI Publishing, Wallingford, UK.

19. Duchateau MJ, Hishiba H, Velthuis HHW. 1994. Diploid males in the bumble bee *Bombus terrestris*. *Entomol. Exp. Appl.* 71:263-269

20. Ellis JS, Knight ME, Darvill B, Goulson D. 2006. Extremely low effective population sizes, genetic structuring and reduced genetic diversity in a threatened bumblebee species, *Bombus sylvarum* (Hymenoptera: Apidae). *Mol. Ecol.* 15:4375-4386

21. Ernst WR, Pearce PA, Pollock TL. 1989. Environmental effects of Fenitrothion use in forestry. Conservation and Protection, Environment Canada, Atlantic Region, Canada.

22. Estoup A, Solignac M, Harry M, Cornuet JM. 1993. Characterization of (GT)n and (CT)n microsatellites in 2 insect species – A*pis mellifera* and *Bombus terrestris*. *Nucleic Acids Research* 21:1427-1431

23. Estoup A, Scholl A, Pouvreau A, Solignac M. 1995. Monandry and polyandry in bumble bees (Hymenoptera, Bombinae) as evidenced by highly variable microsatellites. *Mol. Ecol.* 4:89-93

24. Estoup A, Slignac M, Cornuet J-M, Goudet J, Scholl A. 1996. Genetic differentiation of continental and island populations of *Bombus terrestris* (Hymenoptera: Apidae) in Europe. *Mol. Ecol.* 5:19-31

25. Frankham R, Ballou JD, Briscoe DA. 2002. *Introduction to Conservation Genetics*. Cambridge, UK: Cambridge University Press. 617 pp.

26. Franklin MT, Winston ML, Morandin LA 2004. Effects of clothianidin on *Bombus impatiens* (Hymenoptera: Apidae) colony health and foraging ability. *Ecotoxicology* 97:369-373

27. Free JB, Ferguson AW. 1986. Foraging of bees on oil-seed rape (*Brassica napus* L.) in relation to the flowering of the crop and pest control. *J. Agric. Sci.* 94:151-154

28. Free JB, Williams IH. 1976. Pollination as a factor limiting the yield of field beans (*Vicia faba* L.). *J. Agric. Sci.* 87:395-399

29. Gegear RJ, Otterstatter MC, Thomson JD. 2005. Does parasitic infection impair the ability of bumblebees to learn flower-handling techniques? *Anim. Behav.* 70:209-215

30. Genersch E, Yue C, Fries I, de Miranda JR. 2006. Detection of Deformed Wing Virus, a honey bee viral pathogen, in bumble bees (*Bombus terrestris* and *Bombus pascuorum*) with wing deformities. *J. Invertebr. Pathol.* 91:61-63

31. Gerloff CU, Ottmer BK, Schmid-Hempel P. 2003. Effects of inbreeding on immune response and body size in a social insect, *Bombus terrestris*. *Funct. Ecol.* 17:582-589

32. Gerloff CU, Schmid-Hempel P. 2005. Inbreeding depression and family variation in a social insect, *Bombus terrestris* (Hymenoptera: Apidae). *Oikos* 111:67-80.

33. Golick DA, Ellis MD 2006. An update on the distribution and diversity of *Bombus* in Nebraska (Hymenoptera: Apidae). *J. Kans. Entomol. Soc.* 79:341-347

34. Goulson D. 2003a. *Bumblebees: Behaviour and Ecology*. Oxford, UK: Oxford University Press

35. Goulson D. 2003b. Effects of introduced bees on native ecosystems. *Annu. Rev. Ecol. Evol. Syst.* 34:1-26

36. Goulson D. 2003c. Conserving wild bees for crop pollination. *Int. J. Food, Agric. Environ.* 1:142-144

37. Goulson D. 2003d. The conservation of bumblebees. *Bee World* 84:105-106

38. Goulson D, Darvill B. 2004.Niche overlap and diet breadth in bumblebees; are rare species more specialized in their choice of flowers? *Apidologie* 35:55-63

39. Goulson D, Hanley ME, Darvill B, Ellis JE. 2006. Biotope associations and the decline of bumblebees (*Bombus* spp.). *J. Insect Conserv.* 10:95-103

40. Goulson D, Hanley ME, Darvill B, Ellis JS, Knight ME. 2005. Causes of rarity in bumblebees. *Biol. Conserv.* 122:1-8

41. Goulson D, Hughes WOH, Derwent LC, Stout JC. 2002. Colony growth of the bumblebee, *Bombus terrestris* in improved and conventional agricultural and suburban habitats. *Oecologia* 130:267-273

42. Greenleaf SS, Kremen C. 2006. Wild bee species increase tomato production and respond differently to surrounding land use in Northern California. *Biol. Conserv.* 133:81-87

43. Henter HJ. 2003. Inbreeding depression and haplodiploidy: Experimental measures in a parasitoid and comparisons across diploid and haplodiploid insect taxa. *Evolution* 57:1793-1803

44. Hines HM, Hendrix SD. 2005. Bumble bee (Hymenoptera: Apidae) diversity and abundance in tallgrass prairie patches: effects of local and landscape floral resources. *Environ. Entomol.* 34:1477-1484

45. Holehouse KA, Hammond RL, Bourke AFG 2003. Non-lethal sampling of DNA from bumble bees for conservation genetics. *Insect. Soc.* 50:277-285

46. Howard DC, Watkins JW, Clarke RT, Barnett CL, Stark GJ. 2003. Estimating the extent and change in broad habitats in Great Britain. *J. Environ. Management* 67:219-227

47. Inari N, Nagamitsu T, Kenta T, Goka K, Hiura T. 2005. Spatial and temporal pattern of introduced *Bombus terrestris* abundance in Hokkaido, Japan, and its potential impact on native bumblebees. *Pop. Ecol.* 47:77-82

48. Ings TC, Raine NE, Chittka L. 2005. Mating preference of commercially imported bumblebees (*Bombus terrestris*) in Britain (Hymenoptera: Apidae). *Entomologia Generalis* 28:233-238

49. Ings TC, Wards NL, Chittka L. 2006. Can commercially imported bumble bees out-compete their native conspecifics? *J. Appl. Ecol.* 43:940-948

50. Kells AR, Goulson D. 2003. Preferred nesting sites of bumblebee queens (Hymenoptera: Apidae) in agroecosystems in the UK. *Biol. Conserv.* 109:165-174

51. Kells AR, Holland JM, Goulson D. 2001. The value of uncropped field margins for foraging bumblebees. *J. Insect Conserv.* 5:283-291

52. Kleijn D, Sutherland WJ. 2003. How effective are European agri-environment schemes in conserving and promoting biodiversity? *J. Appl. Ecol.* 40:947-969

53. Knight ME, Martin AP, Bishop S, Osborne JL, Hale RJ, Sanderson A, Goulson D. 2005. An interspecific comparison of foraging range and nest density of four bumblebee (*Bombus*) species. *Mol. Ecol.* 14:1811-1820

54. Kosior A, Celary W, Olejnikzak P, Fijal J, Krol W, Solarz W, Plonka P. 2007. The decline of the bumble bees and cuckoo bees (Hymenoptera: Apidae: Bombini) of Western and Central Europe. *Oryx* 41:79-88

55. Kremen C, Williams NM, Thorp RW 2002. Crop pollination from native bees at risk from agricultural intensification. *PNAS* 99:16812-16816

56. Mänd M, Mänd R, Williams IH. 2002. Bumblebees in the agricultural landscape of Estonia. *Agric. Ecosyst. Environ.* 89:69-76

57. Matsumara C, Nakajima M, Yokoyama J, Waishitini I. 2004. High reproductive ability of an alien bumblebee invader, *Bombus terrestris*, L., in the Hidaka region of southern Hokkaido, Japan. *Jap. J. Conserv. Ecol.* 9;93-102

58. McFrederick QS, LeBuhn G. 2006. Are urban parks refuges for bumble bees? *Biol. Conserv.* 129:372-382

59. Memmott J, Waser NM, Price MV. 2004. Tolerance of pollination networks to species extinctions. *Proceedings of the Royal Society, London. Series B.* 271:2605-2611

60. Mommaerts V, Sterk G, Smagghe G. 2006. Hazards and uptake of chitin synthesis inhibitors in bumblebees *Bombus terrestris*. *Pest Management Sci.* 62:752-758

61. Morandin LA, Winston ML, Franklin MT, Abbott VA. 2005. Lethal and sub-lethal effects of spinosad on bumble bees (*Bombus impatiens* Cresson). *Pest Management Sci.* 61:619-626

62. Ockinger E, Smith HG 2007. Semi-natural grasslands as population sources for pollinating insects in agricultural landscapes. *J. Appl. Ecol.* 44:50-59

63. Osborne JL, Corbet SA 1994. Managing habitats for pollinators in farmland. *Asp. Appl. Biol.* 40:207-215

64. Ovenden GN, Swash ARH, Smallshire D 1998. Agri-environment schemes and their contribution to the conservation of biodiversity in England. *J. Appl. Ecol.* 35:955-960

65. Otterstatter MC, Gegear RJ, Colla S, Thompson JD. 2005. Effects of parasitic mites and protozoa on the flower constancy and foraging rate of bumble bees. *Behav. Ecol. Sociobiol.* 58:383-389

66. Packer L, Owen R. 2001. Population genetic aspects of pollinator decline. *Conserv. Ecol.* 5:4

67. Paxton RJ, Thorén PA, Estoup A, Tengö J. 2001. Queen-worker conflict over male production and sex ratio in a facultatively polyandrous bumble bee, *Bombus hypnorum*: the consequences of nest usurpation. *Mol. Ecol.* 10:2489-2498

68. Pirounakis K, Koulianos S, Schmid-Hempel P. 1998. Genetic variation among European populations of *Bombus pascuorum* (Hymenoptera, Apidae) using mitochondrial DNA sequence data. *Eur. J. Entomol.* 95:27-33

69. Pywell RF, Bullock JM, Hopkins A, Walker KJ, Sparks TH, Burke MJW, Peel S. 2002. Restoration of a species-rich grassland on arable land: assessing the limiting processes using a multi-site experiment. *J. Appl. Ecol.* 39:294-309

70. Pywell RF, Warman EA, Carvell C, Sparks TH, Dicks LV, Bennett D, Wright A, Critchley CNR, Sherwood A. 2005. Providing forage resources for bumblebees in intensively farmed landscapes. *Biol. Conserv.* 121:479-494

71. Pywell RF, Warman EA, Hulmes L, Hulmes S, Nuttall P, Sparks TH, Critchley CNR, Sherwood A. 2006. Effectiveness of new agri-environment schemes in providing foraging resources for bumblebees in intensively farmed landscapes. *Biol. Conserv.* 129:192-206

72. Rasmont P, Mersch P. 1988. Première estimation de la dérive faunique chez les bourdons de la Belgique (Hymenoptera, Apidae). *Annales de la Société Royale zoologique de Belgique* 118:141-147

73. Richards AJ. 2001. Does low biodiversity resulting from modern agricultural practice affect crop pollination and yield? *Ann. Bot.* 88:165-172

74. Ruiz-Gonzalez MX, Brown MJF. 2006. Honey bee and bumblebee trypanosomatids: specificity and potential for transmission. *Ecol. Entomol.* 31:616-622

75. Schmid-Hempel R, Schmid-Hempel P. 2000. Female mating frequencies in *Bombus* spp. from Central Europe. *Insect. Soc.* 47:36-41.

76. Sechser B, Freuler J. 2003. The impact of thiomethoxam on bumble bee broods (*Bombus terrestris* L.) following drip application in covered tomato crops. *J. Pest Sci.* 76:74-77

77. Shao ZY, Mao HX, Fu WJ*.* (2004) Genetic structure of Asian populations of *Bombus ignitus* (Hymenoptera: Apidae). *J. Hered.* 95:46-52

78. Shipp JL, Whitfield GH, Papadopoulos AP 1994. Effectiveness of the bumble bee *Bombus impatiens* Cr. (Hymenoptera: Apidae), as a pollinator of greenhouse sweet pepper. *Scientia Horticulturae* 57:29-39

79. Spiewok S, Neumann P. 2006. Infestation of the commercial bumblebee (*Bombus impatiens*) field colonies by small hive beetles (*Aethina tumida*). *Ecol. Entomol.* 31:623-628

80. Stahlhut JK, Cowan DP. 2004. Inbreeding in a natural population of Euodynerus foraminatus (Hymenoptera: Vespidae), a solitary wasp with single-locus complementary sex determination. *Mol. Ecol.* 13:631-638

81. Stubbs CS, Drummond FA. 2001. *Bombus impatiens* (Hymenoptera: Apidae): An alternative to *Apis mellifera* (Hymenoptera: Apidae) for lowbush blueberry pollination. *J. Econ. Entomol.* 94:609-616

82. Svensson B, Lagerlöf J, Svensson BG. 2000. Habitat preferences of nest-seeking bumble bees (Hymenoptera: Apidae) in an agricultural landscape. *Agric. Ecosyst. Environ.* 77: 247-255.

83. Thomson DM. 2004. Detecting the effects of introduced species: a case study of competition between *Apis* and *Bombus*. *Oikos* 114:407-418

84. Thomson DM. 2006. Competitive interactions between the invasive European honey bee and native bumble bees. *Ecology* 85:458-470

85. Thompson HM. 2001. Assessing the exposure and toxicity of pesticides to bumblebees (*Bombus* sp.). *Apidologie* 32:305-321

86. Thompson HM, Hunt LV. 1999. Extrapolating from honeybees to bumblebees in pesticide risk assessment. *Ecotoxicology* 8:147-166

87. Thorp RW. 2005. Species profile: *Bombus franklini.* In *Red List of Pollinator Insects of North America.* Eds. Shepherd, M.D., D.M. Vaughan and S.H. Black. The Xerces Society for Invertebrate Conservation.

88. Thorp RW, Shepherd MD. 2005. Profile: Subgenus *Bombus*. In *Red List of Pollinator Insects of North America.* Eds. Shepherd, M.D., D.M. Vaughan and S.H. Black. The Xerces Society for Invertebrate Conservation.

89. Tommasi D, Miro A, Higo HA, Winston ML. 2004. Bee diversity and abundance in an urban setting. *Can. Entomol.* 136:851-869

90. Walther-Hellwig K, Fokul G, Frankl R, Buechler R, Ekschmitt K, Wolters V. 2006. Increased density of honeybee colonies affects foraging bumblebees. *Apidologie* 37:517-532

91. Waser NM, Chittka L, Price MV, Williams N, Ollerton J. 1996. Generalization in pollinator systems and why it matters. *Ecology* 77:1043-1060

92. Westphal C, Steffen-Dewenter I, Tsharntke T. 2003. Mass-flowering crops enhance pollinator densities at a landscape scale. *Ecol. Lett.* 6:961-965

93. Whittington R, Winston ML. 2004. Comparison and examination of *Bombus occidentalis* and *Bombus impatiens* (Hymenoptera: Apidae) in tomato greenhouses. *J. Econ. Entomol.* 97:1384-1389

94. Whittington R, Winston ML, Tucker C, Parachnowitsch AL. 2004. Plant-species identify of pollen collected by bumblebees placed in greenhouse for tomato pollination. *Can. J. Plant Sci.* 84:599-602

95. Widmer A, Schmid-Hempel P. 1999. The population genetic structure of a large temperate pollinator species, *Bombus pascuorum* (Scopoli) (Hymenoptera: Apidae). *Mol. Ecol.* 8:387-398

96. Widmer A, Schmid-Hempel P, Estoup A, Scholl A. 1998. Population genetic structure and colonisation history of *Bombus terrestris* s.l. (Hymenoptera: apidae) from the Canary Islands and Madeira. *Heredity* 81:563-572

97. Wilcove DS, Rothstein D, Dubrow J, Phillips A, Losos E. 1998. Quantifying threats to imperiled species in the United States. *Bioscience* 48:607-615

98. Williams PH. 1982. The distribution and decline of British bumble bees (*Bombus* Latr). *J. Apic. Res.* 21:236-245

99. Williams PH. 1994. Phylogenetic relationships among bumble bees (*Bombus* Latr.): a reappraisal of morphological evidence. *Syst. Entomol.* 19:327-344

100. Williams P. 2005. Does specialization explain rarity and decline among British bumblebees? A response to Goulson *et al*. *Biol. Conserv.* 122:33-43

101. Winter K, Adams L, Thorp R, Inouye D, Day L, Ascher J, Buchmann S. 2006. Importation of non-native bumble bees into North America: potential consequences of using *Bombus terrestris* and other non-native bumble bees for greenhouse crop pollination in

Canada, Mexico and the United States. A white paper of the North American Pollinator Protection Campaign (NAPPC).

102. Zayed A, Packer L. 2001. High levels of diploid male production in a primitively eusocial bee (Hymenoptera: Halictidae). *Heredity* 87:631-636

103. Zayed A, Packer L. 2005. Complementary sex determination substantially increases extinction proneness of haplodiploid populations. *PNAS* 102:10742-10746

104. Zayed A, Roubik DW, Packer L. 2004. Use of diploid male frequency data as an indicator of pollinator decline. *Proc. R. Soc. Lond.* 271: S9-S12

TABLE 1. Genetic diversity estimates for populations of a number of *Bombus* species (mean ± S.E.).

Species	Population	Sample Size	Allelic Richness	H_E	Source
B. ignitus	Beijing, China	33	12.2 ± 1.53*	0.85 ± 0.02	Shao *et al.* 2004
B. ignitus	Nagano, Japan	26	8.22 ± 0.72*	0.83 ± 0.03	Shao *et al.* 2004
B. pascuorum	Landford, UK	183	6.22 ± 1.19*	0.52 ± 0.15	Darvill *et al.*2004
B. pascuorum	Rothamsted, UK	125	5.71 ± 1.01	0.52 ± 0.11	Knight *et al. 2005*
B. pascuorum	Continental Europe	22.7 average	5.49 ± 0.16*	0.56 ± 0.01	Widmer & Schmid-Hempel 1999
B. terrestris	Continental Europe	37.5 average	5.96 ± 0.12*	0.61 ± 0.01	Estoup *et al.* 1996
B. lucorum	Bern, Switzerland	40	7.00 ± 2.00*	0.60 ± 0.12	Estoup *et al.* 1996
B. hypnorum	Various, Sweden	10	6.75 ± 1.03*	0.72 ± 0.14†	Paxton *et al.* 2001
B. sylvarum	Southern UK	25.6 average	3.12 ± 0.10	0.39 ± 0.02	Ellis *et al. 2006*
B. sylvarum	Epenede, France	10	4.00 ± 0.85	0.53 ± 0.09	Ellis *et al. 2006*
B. muscorum	Outer Hebrides, UK	43.8 average	3.22 ± 0.12	0.39 ± 0.01	Darvill *et al.* 2006
B. muscorum	Inner Hebrides, UK	62.7 average	3.21 ± 0.07	0.47 ± 0.01	Darvill *et al.* 2006
B. muscorum	Southern UK	35.5 average	4.01 ± 0.06	0.51 ± 0.01	Darvill *et al.* 2006
B. distinguendus	Scotland, UK	7.75 average	2.63 ± 0.23*	0.42 ± 0.01 †	Bourke & Hammond 2002

* allelic richness was not available, and the average number of alleles per locus is presented. Allelic richness is a normalised measure which takes account of differing sample sizes to give a comparable figure for all populations.

† Expected heterozygosity was not available, and observed heterozygosity is given. These measures are expected to be very similar for populations that are in Hardy-Weinberg equilibrium.

Mini glossary

Diploid male: In inbred populations, bees which are genetically female may instead develop into sterile males if they are homozygous at the sex-determining locus.

Inbreeding: An increase in the frequency of individuals which are homozygous for alleles identical by descent relative to another or the ancestral population.

Inbreeding depression: Reduced fitness that can result from inbreeding, for example when deleterious recessive alleles are expressed.

Extinction vortex: A process of positive feedback whereby declines in abundance render a species less viable, leading to accelerated declines.

Acronyms

No acronyms are used in the text.

Annotations to references

Carvell et al. (2007): A long-term study of the relative merits of different field-margins management techniques for bumblebees.

Colla et al. (2006): Highlights the potentially devastating effects of pathogen spillover from commercial colonies.

Darvill et al. (2006): The first study of the population structure of a rare bumblebee species persisting in fragmented habitat islands

Goulson (2003)b: A comprehensive review of the sometimes conflicting evidence concerning the impacts of non-native bees on native ecosystems.

Thomson (2006): The first convincing demonstration of the negative effects of non-native honeybees on the reproduction of native bumblebees.