Can novel seed mixes provide a more diverse, abundant, earlier, and longer-lasting floral resource for bees than current mixes?

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Can novel seed mixes provide a more diverse, abundant, earlier, and longer-lasting floral resource for bees than current mixes?

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Abstract

Several agri-environment schemes aim to improve pollinator diversity and abundance, including the sowing of wildflower areas. These seed mixes are often either low in floral diversity and target few pollinator species (mainly social bees), or high in floral diversity but with limited evidence of good establishment of the component species. In order to support a greater diversity of wild bees in farmland, we need more diverse seed mixes, containing species shown to support a wide diversity of insect pollinators, with good establishment and long flowering periods. Here we trialled two typical seed mixes, a low-diversity Fabaceae-heavy mix (FAB) and a more diverse wildflower mix (WF), against two novel wildflower mixes, one based on literature sources (LT), and one based on first-hand surveys of pollinator attraction to flowers growing on a wildflower farm (WB). Both new mixes were focussed on plants attractive to wild bee species. Replicated field plots were set up on two farms and monitored over three years. Our novel wildflower mixes had higher floral diversity and abundance than the FAB mix, and began flowering earlier, reaching their floral peak before the FAB mix, potentially providing forage for a broader range of pollinators or those with earlier flight seasons. The high floral abundance in LT and WB was driven by annuals in the first year, and then multiple perennials in the second and third year. We identified five perennials from four families (\textit{Daucus carota}, \textit{Leucanthemum vulgare}, \textit{Geranium pyrenaicum}, \textit{Lotus corniculatus} and \textit{Trifolium hybridum}) that established well on both farms, are known to be attractive to a diversity of bee species, and thus could be considered as providing a more taxonomically diverse base for creating future mixes. However, the mixes provided few floral resources in April (needed by early-flying wild bees), and more research is required in this area.

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Introduction

Landscape simplification and loss of semi-natural habitat as a result of agricultural intensification have resulted in reduced farmland biodiversity (Foley et al., 2005). To rectify this loss, land-sharing conservation and restoration efforts through agri-environment schemes (AES) have been implemented through much of the Western world (Harmon-Threatt & Hendrix, 2015; Howlett et al., 2021; Lindenmayer et al., 2012; Scheper et al., 2015). Pollinating insects are one of the many taxa that have suffered declines in both abundance and diversity due to agricultural intensification (Goulson, Nicholls, Botias, & Rotheray, 2015;...
Potts et al., 2010), and there are several AES options targeted towards improving pollinator numbers. A favoured option is the delivery of wildflower areas, usually found along field margins, and typically achieved through the sowing of a wildflower seed mix (Hardman, Norris, Nevard, Hughes, & Potts, 2016).

Fabaceae-based wildflower mixes are found throughout Europe, and have been available in the UK for over 15 years through AES (DEFRA, 2005, 2018). Early research trialling this mix showed it to be particularly good at attracting long-tongued bumblebee species., including some rare species (Bombus ruderatus, B. muscorum, and B. ruderarius) (Carvell, Meek, Pywell, Goulson, & Nowakowski, 2007; Carvell, Westrich, Meek, Pywell, & Nowakowski, 2006; Heard et al., 2007; Pywell et al., 2012). It was generally considered a success: it was cheap to sow (~€130/ha compared to €435-1232/ha for a more diverse wildflower mix) and was easy to establish and manage. It typically comprised of four legume species, all of which flowered during the trials, and it attracted ~10x greater bumblebee abundance than a grass-only mix (Carvell et al., 2007; Pywell et al., 2006). However, studies have also indicated a reduced flowering season compared to other wildflower mixes (Carvell et al., 2007), and that the vast majority of wild bee species use alternative floral resources growing on farms (Wood, Holland, & Goulson, 2015, 2017).

We can draw some comparisons between typical wildflower mixes used across Europe. As well as the low diversity, Fabaceae-heavy mix in the UK, there is also a more diverse mix available of 10-15 species containing a wider range of flower families (DEFRA, 2018). These two options mirror those available in The Netherlands, described by Kleijn et al. (2018): a Fabaceae-focused mix targeting long-tongued specialist insects, and a second mix with a high proportion of Apiaceae species to target a wider range of shorter-tongued species. While federal states in Germany have no common recommendation, the least diverse wildflower mix in Hessen, for example, is composed of ~60% Fabaceae spp., whereas a wildflower bee-targeted mix contains a diverse range of 50 species. Ten of the most commonly selected wildflower species for the seed mixes used in Hessen include four species regularly chosen for mixes across Northern Europe (Trifolium pratense, Onobrychis viciifolia, Daucus carota, and Malva spp.) (Warzecha, Diekötter, Wolters, & Jauker, 2018). Finally, Switzerland has a single mix for floristically enhancing farmland (Pfiffner & Wyss, 2003). The basic version, comprised of 24 species, contained six species commonly included in wildflower mixes in the UK and across Europe (Haaland & Bersier, 2011).

Multiple factors play a role in attracting pollinators to a flower strip, such as the floral abundance and species richness (Balzan, Boci, & Moonen, 2014; Carvell et al., 2007; Haaland, Naisbit, & Bersier, 2011), the plant species in flower and their functional traits (Nichols, Goulson, & Holland, 2019; Sutter, Jeanneret, Bartaual, Boci, & Albrecht, 2017; Uyttenbroeck, Piqueray, Hatt, Mahy, & Monty, 2017; Warzecha et al., 2018), and alternative floral resources available at the landscape-scale (Heard et al., 2007). Ideally, such factors must be taken into consideration when formulating a wildflower mix. We have focused on plant species that could provide an earlier flowering season, and more consistent floral abundance in subsequent years. Our mixes were created from selecting wildflower species that have the potential to attract a broader range of wild bee species.

The aim of the study was to:

a) evaluate these new mixes against currently used seed mixes, in order to identify:

b) how well the new mixes performed in terms of floral richness, abundance, length of flowering season, and longevity in comparison to existing ones;

i) which plant species provided the highest floral abundance and longest flowering season when sown in mixes;

ii) the extent to which local factors (soil type, previous management history) affected the resulting species composition; and

iii) a list of species that could be recommended for inclusion in future AES.

Materials and methods

Creating wildflower mixes

We created two novel wildflower mixes using similar processes (Fig. 1). Step 1: Initially we used the existing literature to select wildflowers that were shown to be key resources for a high abundance and diversity of solitary bees within agricultural settings (Howlett et al., 2021) (Table 1). The creation of this list led us to realise how many of the species available for wildflower mixes have insufficient visitation data to species level in the literature. Therefore, our second approach for creating a novel mix was through primary research on a wildflower farm (Nichols et al., 2019). Again, the wildflowers that attracted the highest diversity of wild bees were selected. Step 2: We organised the wildflowers into categories according to their flowering phenology: late-spring, early-summer, mid-summer, and late-summer to ensure we had at least 1-2 species flowering within each category (Williams et al., 2015). Step 3: Replace species that could not be added to a wildflower mix due to availability or “weed” concerns (e.g. Crepis capillaris, Hypochaeris radicata, Sonchus arvensis, Conopodium majus, and Odontites verus). For example, in our mixes, Crepis capillaris and Sonchus arvensis were replaced by Leontodon hispidus. Step 4: Our final literature-based mix (LT) contained 13 species, and our primary research-based wild bee mix (WB) contained twelve wildflower species. There was just under 50% overlap between the species selected for LT and WB mixes (Table 1). LT contains typical wildflower mix species found across Europe, such as
Centaurea nigra, Daucus carota, Achillea millefolium, and Trifolium pratense, whereas WB contains some species referenced less often in the literature, such as Geranium pratense, Geranium pyrenaicum, and Anthyllis vulneraria (Table 1). Both mixes then had four annual cornfield species added to act as a "nursery" in the first year. Cornfield annuals are species that historically filled arable fields before the introduction of herbicides. They do well in fertile soil, and include Poppy (Papaver rhoeas), Corn marigold (Glebionis segetum), Cornflower (Centaura cyanus), and Corncockle (Agrostemma githago). They have the potential to provide plenty of floral cover, helping to reduce weed competition (Emorsgate Seeds, 2021), as well as support a high bee abundance and richness during the first year (Twerski, Albrecht, Fründ, Moosner, & Fischer, 2022). This is not currently standard practice when sowing wildflower seed mixes on farmland in the UK, and we wanted to evaluate the benefit of doing so.

Table 1 shows the overlap between species selected for our mixes and those already included in mixes in other European countries.

**Survey methods**

The study was conducted in a single field on two farms: Church Farm, Oxfordshire (51° 38’ 14.4348” N, 1° 11’ 5.4528” W) and Lee Farm, West Sussex (50° 53’ 0.2112” N, 0° 28’ 24.1464” W). The farms were of different soil type and the fields had received different management prior to our experiment commencing. Church Farm has freely-draining, base-rich loamy soil of high fertility, and the field had recently been in production, with suspected continual over-fertilisation. It also had a high abundance of aggressive weeds (e.g. Cirsium vulgare, Alopecurus myosuroides). Lee Farm had lime-rich loamy soils over chalk, the field had been out of production for 2-3 years, and any natural regeneration had been cut and ploughed yearly, breaking up any perennial thistles and encouraging annuals. Seed mixes were sown in 2018 on 4th and 5th September respectively. Both sites were cultivated and sprayed-off using glyphosate herbicide as ground preparation to aid establishment. The ground was then rolled, seeds were broadcast sown by hand, and then rolled again to ensure sufficient seed-to-soil contact.

Seed mixes were sown in 20 x 5m contiguous plots. Our treatment consisted of four different seed mixes: Fabaceae-focused mix of six species (FAB), typical wildflower mix of 12 species (WF), a new literature-based mix of 17 species (LT), and a research-based wild bee mix of 15 species (WB); and a fallow (control). From here on we will refer to them as FAB, WF, LT, WB. Details of all four mixes and the amounts of wildflower seeds that were sown can be found in Appendix A: Table 1. Each treatment was replicated 5 times on each farm, therefore each farm had 25 20 x 5m plots. Seed mixes were allocated to plots using a Latin-square-type design to ensure no mix was next to itself. Plots were then marked by a GPS device to store and re-find the plot locations for surveys.

Surveys were conducted from April to August in 2019, 2020 and 2021. They were conducted every 2-3 weeks, providing a total of eight survey periods each year. Farms were surveyed on two separate days close to one another for each of the eight survey periods. Surveys were missed on a farm if they would occur immediately after a cut, since nothing would be in flower. The AES guidelines recommend regular cutting in the first year to help with plant establishment (DEFRA, 2018). After strips are established, cutting management changes depending on specific AES. To simplify matters, all plots on a farm were managed the same way, but cutting on each farm was conducted according to need. For example, Church Farm was cut in May 2019 when thistles began to take over the plots, and then in June during both 2020 and 2021 when plants started to collapse under their own weight. Lee Farm was cut in July in both 2019 and 2020 when growth began to collapse. Both farms were also then cut during the autumn months in 2019 and 2020 once the majority of plants had seeded and growth could be removed. In total, Church farm was surveyed 7 times each in 2019, 2020 and 2021; and Lee farm 8 times in 2019 and 2021, and 7 times in 2020. During each survey, each plot

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**Fig. 1. Flow-chart depicting our methodological approach.** This same process was used to select species for both our literature-based mix (LT) and our wild-bee based mix (WB) from our own primary research.
### Table 1. Wildflowers in mixes and their characteristics.

Presence in mix indicated by ‘x’ for literature-based mix (LT), research-based wild bee mix (WB), typical Fabaceae-focused mix (FAB), and/or typical wildflower mix (WF). Characteristics include the ‘Months’ in flower indicated by numbering (Jan-Dec = 1-12); the ‘Insect groups’ recorded as foraging upon species and the ‘No. of spp.’ shows the ratio of bumblebees to solitary bees (BB/SB), both of which evidence was gathered from the DoPI database (DoPI). ‘Included in European mixes’ lists European countries species is included in a mix for AES purposes. ‘Literature evidence of wild bees’ keys the evidence used for selecting species in the LT mix; and ‘Research evidence of wild bees’ indicates the number of wild bee species found to use wildflower species.

<table>
<thead>
<tr>
<th>Wildflower species</th>
<th>LT</th>
<th>WB</th>
<th>FAB</th>
<th>WF</th>
<th>Months</th>
<th>Insect groups (from DoPI)</th>
<th>No. of spp. (BB/SB) (from DoPI)</th>
<th>Included in European mixes</th>
<th>Literature evidence of wild bees</th>
<th>Research evidence of wild bees</th>
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<tr>
<td>Centaurea nigra</td>
<td>x</td>
<td>x</td>
<td>x</td>
<td>x</td>
<td>7-9</td>
<td>BB BB SB HF</td>
<td>13/13</td>
<td>Be; Ge; Ne; Sz</td>
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<td>-</td>
</tr>
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<td>Daucus carota</td>
<td>x</td>
<td>x</td>
<td>x</td>
<td>x</td>
<td>6-7</td>
<td>BB BB SB HF W</td>
<td>2/15</td>
<td>Be; Ge; Ne; Sz</td>
<td>β</td>
<td>7</td>
</tr>
<tr>
<td>Malva moschata</td>
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<td>x</td>
<td>x</td>
<td>x</td>
<td>7-8</td>
<td>BB</td>
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<td></td>
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<td>x</td>
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<td>-</td>
<td>Ge; Sz</td>
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<td>-</td>
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<td>Ge; Sz</td>
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<td>Ne</td>
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<td></td>
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<td>BB BB HF</td>
<td>6/6</td>
<td>-</td>
<td>β</td>
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<td>x</td>
<td></td>
<td></td>
<td>6-8</td>
<td>BB SB HF B</td>
<td>3/3</td>
<td>Ne; Sz</td>
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<td>x</td>
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<td>BB SB HF F F B B</td>
<td>2/23</td>
<td></td>
<td>δ</td>
<td>10</td>
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<td>x</td>
<td></td>
<td></td>
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<td>BB BB SB HF F W BF B</td>
<td>1/7</td>
<td>-</td>
<td>δ</td>
<td>7</td>
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<tr>
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<td>x</td>
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<td>HB BB HF F W B</td>
<td>0/9</td>
<td></td>
<td>α δ</td>
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<td>x</td>
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<td>HB BB BB W</td>
<td>1/2</td>
<td>Be; Ge; Sz</td>
<td>α β δ</td>
<td>-</td>
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<td>x</td>
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<td>5/28</td>
<td>Ge</td>
<td>α δ</td>
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<td></td>
<td>HB BB SB HF F W</td>
<td>5/1</td>
<td></td>
<td>δ</td>
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<td>15/5</td>
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<td>Ge</td>
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<td>12/4</td>
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<td>BB BF*</td>
<td>5/0</td>
<td>Ge; Ne; Sz</td>
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<td>-</td>
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<td>Pastinaca sativa</td>
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<td>Ge; Ne; Sz</td>
<td>-</td>
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<td>2/3</td>
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<td>BB</td>
<td>5/0</td>
<td>Ne</td>
<td>-</td>
<td>-</td>
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</tbody>
</table>

Insect groups key: HB = honeybee; BB = bumblebee; SB = solitary bee; HF = hoverfly; F = other fly; W = wasp (including solitary and parasitoid); BF = butterfly (*indicates moth); B = beetle

Included in European mixes: Be = Belgium (Ouvrard et al., 2018); Ne = The Netherlands (Kleijn et al., 2018; Kohler, Verhulst, Van Klink, & Kleijn, 2008); Sz = Switzerland (Haaland & Bersier, 2011); Ge = Germany (Denys & Tschamtke, 2002; in a minimum of two mixes from Warzecha et al., 2018)

Literature evidence key:

α: Popular wildflower foraged upon (>5% of visitations) by solitary bees (Wood et al., 2015).

β: Solitary bee pollen load analysis indicating >10% pollen collected from wildflower species (Wood et al., 2017).

δ: Throughout the season, wildflower plants that received >10% of visitations by solitary bees (Wood et al., 2017).

Research evidence species calculated from Nichols et al. (2019).
was walked centrally lengthways and flower species seen 2 m either side were noted and the estimated abundance of open flowers in the plot recorded (Campbell, Wilby, Sutton, & Wackers, 2017; Wood et al., 2017). A maximum of 10 minutes was allowed for each plot. ‘Flower’ was defined as either a single flower, flowers on an umbel or spike, or a capitulum (Heard et al., 2007).

Statistical analysis

All data analysis was handled in R (R Core Team, 2020). Data for 2019, 2020 and 2021 were analysed and are reported separately. Linear Mixed Models and GLMMs were built using the lme4 package (Bates, Maechler, Bolker, & Walker, 2015), zero-inflated models using the glmmTMB package (Brooks et al., 2017), and all figures were created using ggplot2 (Wickham, 2016). An ANOVA was then performed on each model and its null, reported as X² values, and post-hoc Tukey tests were conducted to see where the significance lay where appropriate.

First, we considered the abundance of sown vs. spontaneous species in each mix during each year. To assess floral abundance, we summed flower counts for all plant species in each plot (replicate), for each survey, per farm. A zero-inflated model with negative binomial family was built with ‘floral abundance’ as response variable, and ‘species origin’ (sown or spontaneous) as predictor variables, and then a ‘treatment’, ‘survey period’ and ‘replicate’ nested within ‘farm’ as random variables.

Next, the total and sown floral abundance and species richness of the treatments were considered. Total floral abundance was calculated as described above. ‘Floral abundance’ was then log (N + 1) transformed to normalise the data and fitted with an LMM, with ‘treatment’ and ‘survey period’ as predictor variables, and ‘replicate’ nested within ‘farm’ as a random variable. To assess floral richness, the total number of species recorded in flower were summed per plot in the same way as for abundance. A zero-inflation model was built using a Poisson family. ‘Floral richness’ was used as the response variable, ‘treatment’ and ‘survey period’ as predictor variables, and ‘replicate’ nested within ‘farm’ as a random variable. ‘Row ID’ was also included as a random variable to improve model fit.

Next, we looked at just the sown species in each plot. The intention here was to examine the success of the sown flowers. Abundance and richness were calculated in the same way as above, but only for the flowers sown in the respective plots. Fallow plots were removed for this analysis. A zero-inflation model using a negative binomial family was built with ‘floral abundance’ as the response variable, ‘treatment’ and ‘survey period’ as predictor variable, and ‘replicate’ nested within ‘farm’ as a random variable. ‘Floral richness’ was modelled with a GLMM using a Poisson family, with ‘treatment’ and ‘survey period’ as response variable, and ‘replicate’ nested within ‘farm’ as a random variable.

Finally, we wanted to determine if the floral abundance differed between farms for total species and just sown species in either year. First, all species identified in 2019 across both farms had their total abundances calculated for each farm, and Wilcoxon rank sum test conducted. The same was then done for 2020 and 2021. Next, just the sown species were identified and abundances calculated, and Wilcoxon rank sum tests conducted comparing both farms, first for 2019, then 2020 and 2021.

Results

Overall floral richness and abundance of treatments

Over the three years, 356,935 flower units were counted, and 79 different wildflower species from 21 families were identified. Of the 31 sown species, 28 were recorded flowering. Over the three years, the AES mixes (FAB and WF) had successful germination and flowering of all species included in their mixes. However, LT and WB were missing two species each: Pulsatilla dysenterica and Sinapis arvensis from LT; Convolvulus arvensis and Sinapis arvensis from WB.

There was a significant difference between floral abundance of sown and spontaneous species in 2019 (GLMM: X² = 139.7, p <0.001), 2020 (GLMM: X² = 89.7, p <0.001), and 2021 (GLMM: X² = 97.9, p <0.001). Sown species made up 68.8%, 78.7% and 76.2% (WF, LT and WB respectively) of the total floral abundance recorded over the three years, compared with 35.5% for the FAB. Despite WF, LT and WB all having similar number of sown species, only LT and WB plots showed reduced spontaneous floral abundance in 2019 (Fig. 2). Overall, sown abundance was greater in all mixes during 2020 and 2021 compared to 2019, spontaneous floral abundance showed overall reduction after 2019, and all mixes had lower spontaneous floral abundance than the Fallow plots in 2020 and 2021. Throughout the experiment, the FAB mix had the lowest sown floral abundance and highest spontaneous floral abundance of all seed mixes.

Total floral richness increased in each treatment from 2019 to 2020 (Table 2), and then decreased in all treatments in 2021, except WB. In both 2019 and 2021, treatment had a significant effect on total floral richness, WB had a significantly higher floral richness than the Fallow in 2019, and LT in 2021. There was no significant effect of treatment on floral abundance in 2019, but WB had greater floral abundance than Fallow in 2020, and both WB and LT had greater floral abundance than FAB in 2021.

Sown floral richness was significantly lower in FAB and WF than LT and WB in 2019, and despite an overall increase in sown richness, FAB remained significantly lower than the other mixes in 2020 and 2021 (Table 2). Sown floral
abundance increased for all mixes from 2019 to 2020, and again in WF and LT in 2021, but decreased in FAB and WB. LT and WB had significantly greater sown floral abundance than FAB and WF in 2019, and remained significantly greater than FAB in 2020 and 2021. By 2021, WF had the highest sown floral richness and abundance compared to the other mixes, though not significantly so; and the mean sown floral abundance of FAB was >50% lower than the other mixes.

Seasonal patterns of the mixes

Survey period had a significant effect on floral abundance and richness, for all species and just the sown species in all three years (Table 2). There was particularly high floral abundance in the first survey period, late April in 2019, largely due to profuse flowering of some fast-growing ruderal species from the seedbank: Capsella bursa-pastoris, Cerastium fontanum, Fumaria officinalis, Lamium purpureum, Senecio vulgaris, Sherardia arvensis, and Stellaria media. Consequently, we will focus on the seasonal patterns of the sown species.

There was an overall increase across the season in sown floral richness for all mixes each year (Fig. 3), with Fallow plots providing greater richness than sown species in the early season surveys, and FAB producing consistently lower sown richness across all three years for the majority of the season, only matching the other mixes at the end of the season.

Species responsible for seasonal patterns

Looking more closely at the species’ abundances, the high floral abundance of LT and WF in the first year was primarily driven by the cornfield annuals (Fig. 4), particularly Papaver rhoeas (47.7% and 48.8% of sown abundance, respectively), Glebionis segetum (13.2% and 23.2% respectively) and Centaurea cyanus (17.1% and 11.6% respectively). However, as we would expect, in 2020 and 2021 the annuals declined and were replaced by perennials.

Having established that species abundance was driven in large by cornfield annuals in the first year, we wanted to identify the key species in the subsequent years. Considering only the ten species that reached the highest average abundance per plot in 2020 and 2021 resulted in a list of twelve species across seven families (Anthyllis vulneraria, Daucus carota, Geranium pyrenaicum, Leontodon hispidus, Leucanthemum vulgare, Lotus corniculatus, Malva moschata,
**Table 2. Floral abundance and richness summary.** Mean floral richness and abundance per plot for the whole season, averaged across both farms, for each year reported for each mix (± SE) (see Appendix A: Table 3 for floral abundance and richness for each survey period). Results of GLMM ANOVA for effect of treatment, and effect of survey period, presented as $X^2$ value, $X^2$ degrees of freedom ($X^2$ df), and significance (NS, not significant: *, $p < 0.05$; **, $p < 0.01$; ***, $p < 0.001$), with post-hoc Tukey pairwise comparisons for the treatments designated by lettering.

<table>
<thead>
<tr>
<th>Year</th>
<th>Treatment</th>
<th>Fallow</th>
<th>FAB</th>
<th>WF</th>
<th>LT</th>
<th>WB</th>
<th>$X^2$</th>
<th>$X^2$ df</th>
<th>Sign.</th>
<th>Survey Period</th>
<th>$X^2$</th>
<th>$X^2$ df</th>
<th>Sign.</th>
</tr>
</thead>
<tbody>
<tr>
<td>2019 Total fl. richness</td>
<td>13.2 a</td>
<td>15.9 ab</td>
<td>16.8 ab</td>
<td>16.0 ab</td>
<td>17.2 b</td>
<td>12.9</td>
<td>4</td>
<td>**</td>
<td>58.1</td>
<td>7</td>
<td>***</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Total fl. abundance</td>
<td>2091.2 ±1.4</td>
<td>2401.2 ±1.6</td>
<td>2183.8 ±1.2</td>
<td>2645.4 ±1.4</td>
<td>749.9</td>
<td>1.6</td>
<td>NS</td>
<td>61.4</td>
<td>7</td>
<td>***</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sown fl. richness</td>
<td>2.6 a</td>
<td>3.3 a</td>
<td>6.4 b</td>
<td>6.9 b</td>
<td>7.01</td>
<td>3</td>
<td>***</td>
<td>264.7</td>
<td>7</td>
<td>***</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sown fl. abundance</td>
<td>287.4 a</td>
<td>210.8 a</td>
<td>1095.8 b</td>
<td>1482.3 c</td>
<td>60.1</td>
<td>3</td>
<td>***</td>
<td>174.9</td>
<td>7</td>
<td>***</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

| 2020 Total fl. richness | 19.7 | 20.9 | 19.8 | 17.3 | 17.8 | 3.7 | 4 | NS | 192.7 | 7 | *** |
| Total fl. abundance | 1459.2 a ±2.4 | 2111.4 ab ±0.5 | 2144.4 ab ±0.6 | 3098.7 b ±1.8 | 181.1 | 15.9 | 4 | ** | 190.3 | 7 | *** |
| Sown fl. richness | 5.0 a | 9.0 b | 9.6 b | 8.7 b | 42.6 | 3 | *** | 254.98 | 7 | *** |
| Sown fl. abundance | 1356.0 a ±0.3 | 2449.9 bc ±0.4 | 1935.3 b ±0.5 | 2830.8 c ±0.5 | 33.66 | 3 | *** | 209.0 | 7 | *** |
| 2021 Total fl. richness | 18.0 ab ±2.3 | 20.6 ab ±0.8 | 14.2 a ±1.3 | 180.0 | 10.2 | 4 | * | 389.9 | 7 | *** |
| Total fl. abundance | 1497.7 ab ±2.3 | 3436.6 ab ±3.2 | 2846.9 b ±14.9 | 2224.7 b ±367.7 | 97.9 | 3 | *** | 319.6 | 7 | *** |
| Sown fl. richness | 6.0 a | 10.1 b | 7.4 b | 7.8 b | 32.6 | 3 | *** | 305.0 | 7 | *** |
| Sown fl. abundance | 625.5 a | 3252.6 c | 2617.7 c | 1755.9 b | 97.9 | 3 | *** | 319.6 | 7 | *** |

**Ranunculus acris**, **Rhinanthus minor**, **Taraxacum officinale agg.**, **Trifolium hybridum**, and **Trifolium pratense**. Each mix contained at least four of these high abundance species, and was dominated by one or two species (Fig. 5). FAB had a relatively even abundance of **L. corniculatus**, **M. moschata**, **T. hybridum**, and **T. pratense** during 2020, and then became dominated by **L. corniculatus** in 2021. WF had almost equally high numbers of **D. carota** and **L. corniculatus** in 2020, but then became dominated by **L. vulgare** in 2021. LT contained six of the twelve most abundant species during 2020, with **D. carota** and **T. pratense** reaching similarly high abundances, but then also became dominated by **L. vulgare** in 2021. WB was dominated by **G. pyrenaicum** during 2020, and then by **A. vulneraria** in 2021. Although the same species were sown in multiple mixes (e.g. **D. carota**), they had varying abundances. It is important to note that the abundances did not necessarily reflect the seed mix compositions (see Appendix A: Table 1), and are possibly a by-product of within-mix competition.

Species peaked in abundance at different times throughout the season (Appendix A: Table 2). **Lotus corniculatus** and **T. hybridum** provided the mid-season floral abundance in the FAB mix, followed by **M. moschata** and **T. pratense** late-season (Fig. 6). Early- to mid-season floral abundance was driven by **R. acris**, **L. corniculatus**, **R. minor**, and **L. vulgare** in the WF mix, followed by late-season peaks in **D. carota** and **M. moschata**. **Taraxacum officinale agg.** was the only species displaying any notable abundance during late April (found in both the LT and WB mixes). As **T. officinale agg.** started to decline, LT had early- to mid-season peaks in **R. acris**, **T. pratense** and **L. vulgare**, followed by a late-season peak in **D. carota**; whilst WB had early- to mid-season peaks in **G. pyrenaicum** and **A. vulneraria**, also followed by late-season peaks of **D. carota**, and **M. moschata**. **Leontodon hispidus** floral abundance peaks appeared to shift from late-season in 2020 to mid-season in 2021.

We’re also able to select species that, when combined in a seed mix, could provide high abundance floral resources throughout the spring-summer season (Fig. 6): **Taraxacum officinale agg.**, **R. acris**, **G. pyrenaicum**, **L. corniculatus**, and **A. vulneraria** for early- to mid-season; the addition of **L. vulgare** and **T. hybridum** for mid-season; and the inclusion of **D. carota** and **M. moschata** for late-season.

**Differences between Farms**

When we compared farms through community dissimilarity analysis (Appendix A: Fig. 6), it was apparent that they had very different seedbanks. However, the four seed mixes
showed a high level of similarity between farms, suggesting that differences in the seedbanks did not greatly affect establishment of sown species, especially once the perennials established in the second and third year.

The total floral abundance was significantly different between the two farms in 2019, with Lee Farm recording a total abundance of 119,574 open flowers (n = 8) and Church Farm only recording 8,665 open flowers (n = 7) (W = 1871, p < 0.001), but no significant difference in 2020, with Lee Farm supporting 62,699 open flowers (n = 7) and Church Farm with 53,001 open flowers (n = 7) (W = 2412, p = 0.081). In 2021 the difference remained non-significant, with 68,141 open flowers on Lee Farm (n = 8), and 44,855 open flowers (n = 7) on Church Farm (W = 2448, p = 0.054). Considering just the sown species, again, there was a significant difference in sown floral abundance.

![Fig. 3. Mean sown floral richness (A) and abundance (B) for each mix across the season, per year.](image)

The averages were calculated across the plots and farms for each mix during each survey period, in each year. Floral richness shows a slow increase for all mixes over the season for all three years, whereas floral abundance has quite stark differences between years. (Figure with SE bars in Appendix A: Fig. 1 & 2).

![Fig. 4. Comparison of cornfield annual abundance to other sown species.](image)

Flower type: Cornfield annuals, Others. The abundance averages were calculated for each mix across the plots, farms, and survey periods in each year. A particularly high abundance of cornfield annuals can be seen in LT and WF in 2019. (See Appendix A: Fig. 3 for version with SE bars).
between farms in 2019, with 29,314 open flowers recorded on Lee Farm (n = 8) and 1,162 recorded on Church Farm (n = 7) (W = 259, p = 0.023), but no significant difference in 2020, with 44,927 recorded on Lee Farm (n = 7) and 40,793 on Church Farm (n = 7) (W = 379, p = 0.458), nor in 2021, with 49,212 recorded on Lee Farm (n = 8) and 33,305 on Church Farm (n = 7) (W = 425, p = 0.113). The significantly lower floral abundance on Church Farm in 2019 was likely due to a combination of factors (Appendix A: Fig. 6). Due to these factors, we have focused on the floral abundances of...
the sown flowers in the second year, once mixes were well established. Using the same twelve most abundant species as above, the species broadly performed similarly on the two farms in 2020 (Fig. 7), with five plant species dominating floral abundance at both farms (Daucus carota, L. vulgare, L. corniculatus, G. pyrenaicum and T. hybridum). However, there were notable differences in 2021. Specifically, D. carota disappeared from Church farm, whereas L. vulgare increased from 14.1% to 50.7% of floral abundance (of the twelve most abundant species). Additionally, Lee Farm had a greater percentage of A. vulneraria (15.7%) and R. minor (10.8%) than Church Farm (1.6% and 0.0% respectively) in 2021. Lee Farm and Church Farm both experienced large reductions in T. hybridum abundance (89.4% and 57.0% respectively) from 2020 to 2021.

**Discussion**

Earlier studies have considered or compared wildflower mixes in terms of their attractiveness to pollinators (typically focusing on Bombus spp.), but often with minimal detail on the success of sown species in terms of their presence, abundance, and longevity (though see Mallinger, Franco, Prischmann-Voldseth, & Prasifka, 2019; Ouvrard, Transon, & Jacquemart, 2018; Schmidt, Kirmer, Kiehl, & Tischew, 2020). We conducted this experiment to identify a novel seed mix or mixes that could provide a more diverse, abundant, earlier, and long-lasting floral resource for insect pollinators. By using two farms of different soil types and management histories, we hoped to improve the generality of our findings.

We used our methodological approach to create two novel wild bee-targeting seed mixes, first using the literature (Howlett et al., 2021), and then primary research. We selected species that should have created a long flowering season (Williams et al., 2015). Indeed, our mixes provided floral abundance earlier in the season, with greater sown diversity and longevity than the current Fabaceae-heavy mix (FAB) available.

Overall, the novel mixes, LT and WB, had a higher sown floral richness and abundance than the Fabaceae-heavy mix (FAB), in all three years, which is vital for attracting a broad range of pollinating insects. In particular, sown species surpassed spontaneous growth in the first year, unlike FAB and WF. Although a species-rich seed mix is shown to outcompete the seedbank, resulting in a higher proportion of sown to spontaneous species flowering (Carvell et al., 2006; Van Der Putten et al., 2000), we suggest it was the presence of cornfield annuals in LT and WB acting as a nursery to reduce spontaneous growth in year one (Emorsgate Seeds, 2021). Indeed, annual arable plants can also support a diversity of pollinators (Twerski et al., 2022) provided the frequent cutting required in the first year doesn’t remove the flowers. The annuals allowed the sown species to establish well, providing >75% of total floral abundance from sown species over the three years, similar results to other studies (Schmidt et al., 2020, Ouvrad et al. 2018). Additionally, by the third year, the FAB mix was at <50% of the sown floral abundance of the other mixes, suggesting it had passed its floral peak and would continue to subside (Carvell et al., 2007). This would have benefited from additional survey years to confirm the exact longevity of each of the mixes.

We also aimed to identify a seed mix with an extended flowering season than is currently provided by the current mixes (FAB and WF). As anticipated, the majority of early-season floral richness and abundance was provided by spontaneous species (Fig. 3 – Fallow). Specifically, FAB showed low floral abundance until June, when L.

**Fig. 7. Proportion of flowering sown species on each farm, each year.** Using the twelve most abundant sown species across 2020 and 2021, a comparison of the total abundance of each species on each farm summed across all plots and surveys, showing different species dominated the farms each year. (See Appendix A: Fig. 7 & 8) for mean abundance of each species with SE bars).
corniculatus and T. hybridum started to peak in floral abundance (Fig. 6). Through the inclusion of T. officinale agg., LT and WB were the only mixes with substantial sown floral abundance in late April. Additionally, the inclusion of R. acris, G. pyrenaicum, and A. vulneraria across WF, LT and WB provided sown floral abundance before June. This earlier flowering season is likely to benefit both social bees and early-flying solitary species, when many insect pollinators rely heavily on volunteer species and/or hedgerow plants for early spring floral resources (Ouvrard et al., 2018; Wood et al., 2015, 2017). Hedgerow management that allows shrubs to flower, such as Crataegus monogyna, Prunus spinosa and Salix spp., along with a diverse hedge base could be a better avenue for supporting earlier-flying species, where up to ten times more species can be found than in a typical flower strip (Holland, Smith, Storkey, Lutman, & Aebischer, 2015). Indeed, field boundaries were identified as the main source of nectar in arable landscapes, and although agri-environment options can be a rich source, they are not implemented often enough at present to have an impact on a national scale (Baude et al., 2016).

Of the twelve most abundant sown perennials, five had similarly high abundances on both farms in 2020 (Fig. 6). Daucus carota, L. vulgare, L. corniculatus, G. pyrenaicum and T. hybridum, though each had varying abundances between mixes. These five species are identified as being particularly important foraging resources for a range of wild bee species, and should be retained in future mixes (Ouvrard et al., 2018). During the third year, floral composition of each mix shifted (Fig. 5), as some species abundances reduced (T. hybridum, T. pratense, D. carota), whilst others increased in abundance (A. vulneraria, R. acris, L. vulgare, L. corniculatus). In particular, FAB became dominated by L. corniculatus (Carvell et al., 2007), whilst WF and LT became dominated by L. vulgare (Blackmore & Goulson, 2014). Table 1 shows four of these five species are already present in many mixes Europe-wide, and all five species have the potential to provide forage for the majority of key pollinating insect groups, as well as target specific wild bees. Lotus corniculatus and T. hybridum attract bumblebee species such as Bombus lapidarius and B. pascuorum (Carvell et al., 2011; Heard et al., 2007), whereas L. vulgare, D. carota and G. pyrenaicum support solitary bee species within Andrenidae, Halictidae, and Colletidae (Falk, 2016; Nichols et al., 2019; Wood et al., 2015, 2017).

However, our study also showed that under basic management (one- to two-yearly cuts), floral richness and abundance can fluctuate significantly from year to year. This can make wildflower strips appear difficult to manage, potentially leading to them being an unpopular AES option (Kleijn et al., 2019). When formulating a seed mix, species should be tailored to the soil type, and seeds should be sourced locally in order for maximum success of the seed mix (Nowakowski & Pywell, 2016). Additionally, farmers should be supported in their AES management by experts, as farmer experience and engagement plays a key role in AES success (McCracken et al., 2015).

Conclusion

Both of the new seed mixes performed well in our trials, providing a longer flowering season and higher diversity of flowers than a simple Fabaceae mix, though not significantly more than the current WF mix. This suggests simply having a mix of at least twelve species from a variety of flower families can create a longer and more abundant flowering season than a low diversity, Fabaceae-focused mix. Five perennial plant species emerged as reliable performers that established well and produced many flowers on both farms by the second year: D. carota, L. vulgare, G. pyrenaicum, L. corniculatus, and T. hybridum. An additional seven species performed well within a certain mix or part of the season: T. officinale agg., M. moschata, A. vulneraria, R. minor, R. acris, L. hispidus and T. pratense. However, it appeared that the Fabaceae spp. were relatively short-lived, along with D. carota, as these reduced significantly in abundance by the third year. Finally, the addition of the cornfield annuals P. rhoesas and G. segetum are worth considering as they provided forage in the first year while not preventing (and perhaps providing a nursery for) establishment of perennials.

Although this study focused on selecting and monitoring plant species that have the potential to support wild bee diversity, it is important to note that many non-bee pollinators are also at risk (Powney et al., 2019), are important pollinators of many crops worldwide (Rader et al., 2016), and future studies should consider the creation of seed mixes that encompass non-bee pollinators as well (Howlett et al., 2021). In addition to selecting plant species that improve insect pollinator abundance and diversity, species that aid in reducing crop pests could also be considered.

Finally, in order to ascertain the true attractiveness of each plant species and the species combinations of each mix, the insect pollinators using the mixes should be identified and quantified. We propose this is considered through a follow-up publication.

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Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.
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Supplementary materials

Supplementary material associated with this article can be found in the online version at doi: 10.1016/j.baae.2022.02.002.

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