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Weeds in organic fertility-building leys

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1 Weeds in Organic Fertility-Building Leys: Aspects of Species
2 Richness and Weed Management

3

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27 **Abstract**

28 Legume-based leys (perennial sod crops) are an important component of fertility
29 management in organic rotations in many parts of Europe. Despite their importance,
30 however, relatively little is known about how these leys affect weed communities or how the
31 specific composition of leys may contribute to weed management. To determine whether the
32 choice of plant species in the ley affects weeds, we conducted replicated field trials at six
33 locations in the UK over 24 months, measuring weed cover and biomass in plots sown with
34 monocultures of 12 legume and 4 grass species and in plots sown with a mixture of 10
35 legume species and 4 grass species. Additionally, we monitored weed communities in leys on
36 21 organic farms across the UK either sown with a mixture of the project species or the
37 farmers' own species mix. In total, 63 weed species were found on the farms, with the
38 annuals *Stellaria media*, *Sonchus arvensis*, and *Veronica persica* being the most frequent
39 species in the first year after establishment of the ley, while *Stellaria media* and the two
40 perennials *Ranunculus repens* and *Taraxacum officinale* dominated the weed spectrum in the
41 second year. Our study shows that organic leys constitute an important element of farm
42 biodiversity. In both replicated and on-farm trials, weed cover and species richness were
43 significantly lower in the second than in the first year, owing to lower presence of annual
44 weeds in year two. In monocultures, meadow pea (*Lathyrus pratensis*) was a poor competitor
45 against weeds, and a significant increase in the proportion of weed biomass was observed
46 over time, due to poor recovery of meadow pea after mowing. For red clover (*Trifolium*
47 *pratense*), we observed the lowest proportion of weed biomass in total biomass among the
48 tested legume species. Crop biomass and weed biomass were negatively correlated across
49 species. Residuals from the linear regression between crop biomass and weed biomass
50 indicated that at similar levels of crop biomass, grasses had lower weed levels than legumes.

51 We conclude that choice of crop species is an important tool for weed management in leys.

52

53

54 **Keywords:** clover, conservation, grass, legume, rotation, soil fertility, species richness, weed
55 community

56

57 **Introduction**

58 In agricultural production, nitrogen is a key nutrient for achieving acceptable yields and crop
59 quality [1]. Due to globally rising costs of mineral nitrogen fertilizer and concerns over the
60 negative environmental impact of anthropogenic nitrogen [2, 3], agricultural policy makers,
61 farmers and scientists are increasingly paying attention to the use of leguminous plants as an
62 alternative source of nitrogen [4, 5]. Through their symbiosis with rhizobacteria, legumes are
63 able to fix atmospheric nitrogen [6] and convert it to a form that is readily available to plants
64 [7]. After incorporating (e.g. ploughing) legumes into the soil, nitrogen accumulated in the
65 plants' above-ground and below-ground residues is broken down by microbial activity and
66 released for uptake by the following crop [8]. This use of legumes for fertility-building in the
67 rotation is common in a variety of farming systems, e.g. where the use of mineral nitrogen
68 fertilizer is considered to be too expensive, or, as in organic agriculture, where it is not
69 permitted [9, 10]. Both grain legumes and forage legumes are used for fertility building.
70 Because of its function as the main nutrient provider, the use of forage legumes in the
71 rotation, which in Europe is frequently referred to as the ley phase, is of central importance
72 for certain organic (and also increasingly non-organic) farming systems.

73 In Western and Central Europe, organic farmers most frequently use grass-clover mixes for
74 their leys, with white clover (*Trifolium repens*) and red clover (*T. pratense*) being popular

75 legume species, and perennial ryegrass (*Lolium perenne*) and Italian ryegrass (*L. multiflorum*)
76 as commonly chosen grass species [10]. Frequently, these leys are grazed or cut for silage or
77 hay and incorporated into the soil by ploughing before sowing the next crop [11]. Depending
78 on various factors such as climate and soil conditions, the suitability of the land for arable
79 production and the presence of livestock on the farm, the ley phase on organic farms can vary
80 in duration from short term (1-1.5 years) to longer term (around 5 years), but typically the ley
81 is maintained for about 1.5 to 3 years [10, 12].

82 A key requirement for high ley performance (e.g. as measured by above-ground biomass
83 cumulated over time), and the subsequent provision of nitrogen to the following crops is
84 successful establishment of the ley [13]. Ideally, plants need to cover the ground quickly and
85 establish well in a range of environmental conditions. However, according to a consultation
86 of UK organic farmers conducted before the start of this study, white and red clover can be
87 difficult to establish, especially under dry conditions [14]. During the establishment period,
88 weeds can play an important antagonistic role by competing with the sown legumes for light,
89 nutrients and water [15, 16]. Also, annual weeds that exploit the space left by poor ley
90 establishment are more likely to contribute to the weed seed bank in the soil and may
91 therefore become a problem later, in the crop following the ley. For these reasons, the ability
92 to outcompete weeds, either through a high competitive ability and vigour or through
93 allelopathy, is a desirable trait in legume species for use in leys.

94 At the same time, the lack of tillage during the ley phase means that an important tool for
95 weed control in organic farming, namely the mechanical destruction and burying of weeds
96 [17], is not available. Also, lack of tillage means that weeds are not stimulated to germinate,
97 so that weed seeds remain in the seed bank. On the other hand, leys can be repeatedly mown
98 or grazed during the ley phase, which provides an alternative tool for weed management [18].
99 Using multiple species with complementary growth habits in a ley has the potential to further

100 enhance weed suppression by exploiting differences in functional traits [19, 20]. For
101 example, a fast growing early species that covers the ground quickly would complement a
102 species that is taller and more competitive later in the season. Interestingly, leys appear to
103 have the potential to increase weed seed numbers in the seed bank while simultaneously
104 reducing weed emergence in the following crop; in a study on weeds in a wheat (*Triticum*
105 *aestivum*) crop after lucerne (*Medicago sativa*)-grass leys or after potatoes (*Solanum*
106 *tuberosum*) in Southern Germany, higher numbers of weed seeds in the seed bank were found
107 after the ley than after potatoes, but a lower number of weeds emerged in the wheat following
108 the ley [21]. However, careful management is necessary to prevent the build-up of perennial
109 weeds such as docks (*Rumex spp.*) and creeping thistle (*Cirsium arvense*) in leys [22-24].
110 Such species pose a potential problem not only for ley performance but also for subsequent
111 crops and can pose a serious threat to productivity of organic crops [23, 25].

112 Despite the potentially negative effects of annual and perennial weeds in leys, the weed flora
113 may simultaneously contribute to the farm's biodiversity [26, 27]. Weeds provide vital
114 resources for invertebrates and other wildlife [28-31], thereby also helping to regulate pest
115 populations in agro-ecosystems [32]. In addition, some weed species in leys can be a source
116 of mineral nutrients for livestock [33]. Thus, weeds can be seen to provide a range of
117 ecosystem services. However, these same services may also be provided by the crop,
118 especially if multiple crop species in a ley are used. For example, including species with a
119 variety of flowering times would extend the period of nectar and pollen provision [34].

120 Ecological research on the function and diversity weeds in organic farming systems has so far
121 mainly concentrated on weeds occurring in arable crops [35, 36]. Where research has
122 investigated the weed suppression by various small-seeded legume species, the focus has
123 mostly been on the use these legumes as short term cover crops [37, 38]. In contrast, current
124 knowledge about weed diversity and weed control in organic rotational leys is limited. As

125 part of a larger study on optimizing ley composition and management [39] we monitored the
126 dynamics of weed communities in replicated and on-farm trials at multiple locations
127 throughout the UK.

128 Specifically, we asked: (1) Which legume and grass species typically used in legume-based
129 leys show the highest competitive ability against weeds? (2) Which are the dominant weed
130 species in typical organically managed leys in the UK? (3) What is the typical species
131 richness of weeds (as measured by species richness) in organically managed leys? (4) Does
132 crop species richness in the ley affect weed cover and weed species richness?

133

134 **Material and Methods**

135 *Overview*

136 The study was conducted over two years, starting in spring 2009 and consisted of two main
137 experimental series. In series I, we set up replicated field trials at six sites across the UK,
138 evaluating various legume and grass species in monocultures and in a multi-species mixture
139 of legumes and grasses ([Tables 1 and 2](#)).

140 In series II, the same multi-species mixture was sown on 21 organic farms in the UK as non-
141 replicated 0.5 ha strips alongside farmer-chosen control leys ([Table 3, Figure 1](#)). In the
142 following text we call the series I trials “replicated trials” and the series II trials “on-farm
143 trials”. In both series, trials were performed only once per site. Therefore, effects of year-to-
144 year variation (e.g. effects of yearly differences in weather on weed emergence in the
145 establishment phase of the ley) cannot be analysed. However, although effects of the age of
146 the ley and the study year cannot be separated, this was at least partly compensated for by
147 including a large number of trial sites in the study.

148

149 **Table 1:** Legume and grass species included in the trials: Latin and common name, variety, seeding rate (kg/ha), seed weight (Thousand Kernel
 150 Weight, TKW in g) and seeding rate in the monoculture plots and in the All Species Mix (ASM).

| Abbreviation | Latin name | Common name | Variety | Inoculum* | Seeding rate (kg/ha) | | |
|--------------|------------------------------------|-------------------------|------------------------|-----------|----------------------|------|---------|
| | | | | | Monoculture | ASM | TKW (g) |
| AC | <i>Trifolium hybridum</i> L. | Alsike clover | Dawn | C | 10 | 1.25 | 0.7 |
| BT | <i>Lotus corniculatus</i> L. | Birdsfoot trefoil | San Gabrielle | - | 12 | 2.50 | 1.2 |
| BM | <i>Medicago lupulina</i> L. | Black medic | Virgo Pajberg | L | 15 | 2.50 | 1.6 |
| CC | <i>Trifolium incarnatum</i> L. | Crimson clover | Coutea | - | 18 | 2.25 | 3.1 |
| IR | <i>Lolium multiflorum</i> Lam. | Italian ryegrass | Teana | - | 33 | 1.00 | 2.9 |
| LT | <i>Lotus pedunculatus</i> Cav. | Large birdsfoot trefoil | Maku | - | 12 | 2.50 | 1.0 |
| LU | <i>Medicago sativa</i> L. | Lucerne | La Bella de Campagnola | L | 20 | 2.50 | 2.4 |
| MF | <i>Festuca pratensis</i> Huds. | Meadow fescue | Rossa | - | 25 | 1.25 | 2.1 |
| MP | <i>Lathyrus pratensis</i> L. | Meadow Pea | no specified variety | V | 75 | 3.25 | 153.0 |
| PR | <i>Lolium perenne</i> L. | Perennial ryegrass | Orion | - | 33 | 2.50 | 2.0 |
| RC | <i>Trifolium pratense</i> L. | Red clover | Merviot | C | 18 | 2.50 | 1.8 |
| SF | <i>Onobrychis viciifolia</i> Scop. | Sainfoin | Esparsette | - | 80 | 5.00 | 19.2 |
| TY | <i>Phleum pratense</i> L. | Timothy | Dolina | - | 10 | 0.50 | 0.3 |
| WC | <i>Trifolium repens</i> L. | White clover | Riesling | C | 10 | 1.50 | 0.5 |
| SC | <i>Melilotus alba</i> Medik. | White sweet clover | no specified variety | L | 18 | - | 2.3 |
| WV | <i>Vicia sativa</i> L. | Winter vetch | English Vetch | V | 100 | - | 41.0 |

151 *Inoculum: Inoculation prior to sowing with Clover inoculum (C), Lucerne inoculum (L) and Vetch inoculum (V). Details see text.

152

153 **Table 2:** Details of replicated trials: locations, plot sizes, sowing dates and pre-crops; *taken from one quadrat (50 x 50 cm) per plot; ** taken
 154 from three quadrats (each 50 x 50 cm) per plot

| Site | Barrington Park | Duchy (Rosewarne) | IBERS Aberystwyth | Rothamsted | SAC Aberdeen | Wakelyns Agroforestry |
|--------------------------|-----------------|-------------------|-------------------|-------------|---------------|-----------------------|
| Abbreviation | B | D | I | R | S | W |
| North coordinate | 51°49'52.2" | 50°13'38.2" | 52°25'48.1" | 51°48'38.6" | 57°11'05.6" | 52°21'36.7" |
| West coordinate | 1°40'12.3" | 5°18'23.0" | 4°01'22.1" | 0°22'02.4" | 2°12'45.1" | -1°21'09.2" |
| Altitude (m) | 150 | 42 | 29 | 114 | 109 | 51 |
| Plot width (m) | 1.5 | 1.5 | 2.0 | 2.0 | 1.5 | 1.2 |
| Plot length (m) | 10 | 5 | 8 | 5 | 12 | 10 |
| Sowing (date 2009) | 20 Apr | 24 Apr | 23 Apr | 15 Apr | 13 May | 29 Apr |
| First mowing (date 2009) | 24 Jun | 14 Jul | 20 Jul | 05 Aug | 23 Jul | 27 Jul |
| Previous crop | winter barley | fallow | winter oats | fallow | spring barley | potatoes |
| Biomass sampling dates | | | | | | |
| 2009* | - | 18 Aug | 1 Sep | 5 Oct | 20 Aug | 24 Aug |
| 2010* (1) | - | 20 Apr | - | 15 Apr | 13 May | 28 Apr |
| 2010* (2) | - | 18 May | 21 Sep | 13 May | 11 Jun | 28 May |
| 2011** | - | 13-18 Apr | Mar | - | Apr | Apr |

155

156 **Table 3:** Details of participatory trials: Geographic coordinates and soil properties

| Farm Nr. | Coord. North | Coord. West | Elevation (m) | Soil Texture^a | Sand (%) | Silt (%) | Clay (%) | Soil pH | P (mg/L) | K (mg/L) | Mg (mg/L) | SOM (%) |
|-----------------|---------------------|--------------------|----------------------|---------------------------------|-----------------|-----------------|-----------------|----------------|-----------------|-----------------|------------------|----------------|
| 1 | 52°21'36.71" | -1°21'9.24" | 51 | C | 22 | 20 | 58 | 7.4 | 31.6 | 122 | 58 | ND |
| 2 | 52°37'50.17" | -0°20'42.67" | 1 | C | 28 | 31 | 41 | 7.6 | 38.2 | 441 | 424 | ND |
| 3 | 52°8'28.18" | 0°2'57.15" | 45 | CL | 43 | 22 | 35 | 8.2 | 16.8 | 247 | 61 | ND |
| 4 | 52°31'17.36" | 0°9'46.39" | 0 | CL | 39 | 33 | 28 | 6.7 | 34.2 | 201 | 103 | ND |
| 5 | 51°29'47.91" | 1°3'30.22" | 52 | CL | 41 | 40 | 19 | 6 | 33.6 | 77 | 63 | 2.6 |
| 6 | 51°27'1.65" | 1°9'39.6" | 99 | CL | 46 | 33 | 21 | 7.2 | 31.4 | 185 | 51 | 3.3 |
| 7 | 52°22'1.61" | 1°24'47.37" | 73 | C | 42 | 21 | 37 | 6.6 | 30.4 | 336 | 108 | ND |
| 8 | 51°31'5.7" | 1°27'25.92" | 162 | CL | 32 | 42 | 26 | 8 | 21.0 | 110 | 35 | 8.2 |
| 9 | 51°18'56.26" | 1°31'9.32" | 170 | CL | 29 | 42 | 29 | 7.6 | 28.4 | 123 | 42 | 3.8 |
| 10 | 51°22'49.14" | 1°32'3.67" | 125 | CL | 43 | 38 | 19 | 7.4 | 47.4 | 134 | 44 | 3.4 |
| 11 | 51°26'28.01" | 1°54'5.71" | 164 | SL | 16 | 61 | 23 | 7.1 | 20.4 | 95 | 53 | 2.6 |
| 12 | 51°43'56.32" | 1°56'21.42" | 135 | SC | 18 | 36 | 46 | 7.7 | 17.2 | 224 | 71 | 3.6 |
| 13 | 57°16'52.58" | 2°7'56.92" | 97 | SaL | 45 | 39 | 16 | 5.5 | 34.0 | 213 | 77 | 8.0 |
| 14 | 57°11'5.6" | 2°12'45.13" | 109 | SaL | 58 | 29 | 13 | 5.8 | 94.2 | 179 | 171 | 7.8 |
| 15 | 57°33'3.04" | 2°18'0.48" | 120 | CL | 38 | 41 | 21 | 5.7 | 18.0 | 103 | 80 | 8.3 |
| 16 | 57°18'38.38" | 2°18'29.9" | 194 | CL | 43 | 38 | 19 | 6.2 | 30.4 | 212 | 90 | 9.2 |
| 17 | 57°40'16.47" | 3°16'30.66" | 20 | LSa | 77 | 16 | 6 | 6.3 | 34.0 | 110 | 73 | 2.4 |
| 18 | 53°0'38.65" | 3°38'48.06" | 309 | SL | 7 | 58 | 35 | 4.9 | 21.2 | 131 | 65 | ND |
| 19 | 52°37'45.57" | 4°5'1.99" | 56 | SaL | 77 | 12 | 11 | 6.2 | 16.2 | 89 | 161 | ND |
| 20 | 52°2'44.28" | 4°35'59.37" | 70 | SC | 7 | 47 | 46 | 4.9 | 19.2 | 67 | 62 | ND |
| 21 | 51°48'22.52" | 5°4'5.39" | 85 | CL | 32 | 41 | 27 | 5.9 | 18.4 | 170 | 121 | 6.5 |

157 ^a C: Clay; CL: Clay Loam; SC: Silty Clay; SL: Silty Loam; SaL: Sandy Loam; LSa: Loamy Sand; SOM: Soil organic matter; ND: Not
 158 determined

159

160 *Species selection and composition of species mixture for use in field trials*

161 Leys can be sown with mixtures of different plant species, which may provide insurance
162 against the failure of individual species. In addition, mixing species is a way to combine
163 desirable species-specific traits. To compose optimal species mixtures, a useful criterion for
164 species selection is the functional complementarity of the different species [32, 40-42], with
165 the aim of minimizing functional redundancy.

166 According to this idea, we collected data on the ecological and agronomic traits of 22 legume
167 species and five grass species from the literature [for details see 43]. To assess
168 complementarity, a principal component analysis (PCA) was conducted on traits of the 22
169 legume species (maximum height, flowering time, seed size, rooting depth, productivity,
170 establishment and competitive ability, [see supplementary material of reference 43]). The
171 distance of individual species from each other in the PCA bi-plot was considered to be an
172 indicator of functional divergence and potential for complementarity, in terms of coexistence
173 and delivering multiple ecosystem functions when grown together in a mixture. Additional
174 selection criteria included agronomic and practical aspects such as frost tolerance, resistance
175 to grazing and seed availability of the species in the UK.

176 As a result of this selection process we chose a subset of four grass species and twelve
177 legume species with functionally complementary properties for the replicated and on-farm
178 trials (Table 1). Further details of the selection process, as well as the identity of the non-
179 selected species are given elsewhere [44]. All four selected grass species, as well as ten of the
180 twelve tested legume species, were combined in an ‘All Species Mixture’ (ASM) (Table 1),
181 which was tested in both the replicated and on-farm trials. Two species (*M. albus* and *V.*
182 *sativa*) were not included in the ASM because of concerns by the participating farmers about
183 potential detrimental effects of these species on animal health or agronomic management.
184 Seed densities of the monocultures were chosen according to general recommendations for

185 the UK [45]. The average plant density in the monocultures was 1180.5 plants m⁻², whereas
186 the total plant density in the ASM was 1811.1 plants m⁻². The different densities mean that
187 diversity or species richness effects cannot be separated from density effects in this study. In
188 farming practice, however, density in species mixtures often exceeds the densities of their
189 components [46, 47, but see 48]. This is because on the one hand, an additive mixture is
190 frequently considered to be impracticable as its density is too high and causes too much
191 competition among plants, especially when including a large number of species in the
192 mixture. On the other hand, a substitutive mixture may not make full use of the larger
193 resource space available to the mixture. The relative seed rates of species in the mixture were
194 chosen on a number of criteria including expected productivity, seed cost, and seed
195 availability.

196 *Replicated field trials*

197 In the replicated field trials we evaluated 18 treatments. In total, twelve legume species and
198 four grass species were each grown singly as monocultures. In addition, two treatments were
199 reserved for the ASM, which was grown both with and without *Rhizobium* inoculation (see
200 below). At all six trial locations, the experiments were sown in spring 2009 (Table 2). All
201 trials were laid out as single-factor randomized complete block designs with three
202 replications.

203 Following common practice, and to remove the possibility of any differences being due to
204 lack of natural inoculum at sites, seed lots of the four clover species, *V. sativa*, *M. sativa* and
205 one of the ASM treatments were inoculated with rhizobial preparations before sowing (Table
206 1), with 1 % (w/w) substrate per total seed weight. No suitable commercial inoculants could
207 be obtained for the other legume species prior to sowing. The locations, plot sizes and sowing
208 dates are listed in Table 2. Trial sites were distributed over a large geographical area within

209 the UK. All trial sites were mown two times per year at 5-10 cm height, with the first mowing
210 date after establishment in 2009 being between late June and early August (Table 1).

211 *On-farm trials*

212 In addition to the replicated trials, the inoculated ASM was sown by 21 organic farmers
213 across the UK, including sites in East England, South England, North East Scotland and
214 Wales. A further 13 sites were also included in the study, but data could not be included in
215 the analysis because of incompleteness (e.g. sampling only in one of the two study years).
216 Seed of the ASM was provided for a 0.5 ha strip which was sown by the farmers next to or
217 within a control ley (Figure 1). Most of the 21 farmers sowed the leys in spring 2009, while
218 some delayed sowing until later in 2009 for reasons of rotational planning (Table 3). On each
219 farm, the management for the ASM and the control ley were identical (Table 4), but ley
220 management differed among farms. The species composition and seed rates of the control ley
221 were chosen by each farmer individually and differed greatly in the species richness of the
222 sown mixtures (Table 4). On 16 of the 21 farms white clover was included in the control ley.



223

224 **Figure 1:** Photograph of an on-farm trial at Wakelyns Agroforestry, Suffolk, taken in the
 225 summer of 2010. On the left, slightly paler, the control ley (white clover-chicory-black medic
 226 mix), on the right the All Species Mix (ASM). A different site on the same farm was also
 227 used for replicated experiments.

228

229 **Table 4:** Management details for on-farm trials

| Farm Nr. | Sowing month | Mowing | Grazing | Sown species in control ley |
|----------|--------------|--------|---------|---|
| 1 | April | Yes | none | AC, BM, CH, WC |
| 2 | April | Yes | none | RC |
| 3 | April | Yes | none | AC, LU, PR, RC, WC |
| 4 | May | Yes | none | AC, BM, WC |
| 5 | July | Yes | none | CC, LU, RC, WC |
| 6 | April | No | S | BT, CF, MF, RG, WC |
| 7 | April | Yes | none | LU |
| 8 | April | NA | S | RG, RC, WC |
| 9 | April | No | S | RG, WC |
| 10 | April | No | S | BM, BT, CF, PR, RC, WC |
| 11 | April | Yes | C | AC, BM, BT, CC, CF, CH, MF, PR, RC, RG, SB, SF, WC, YW* |
| 12 | June | NA | S | BT, IR/PR, WC |
| 13 | April | No | C | PR, RC, TY, WC |
| 14 | April | no | S | PR, RC, WC |
| 15 | April | no | C | PR, TY, WC |
| 16 | May | no | S | RC, PR |
| 17 | April | yes | none | RC, PR |
| 18 | May | yes | S | WC |
| 19 | April | yes | C & S | RC, WC |
| 20 | April | yes | C | IR, RC |
| 21 | May | yes | C & S | WC |

230 Mowing: NA: No information available

231 Grazing: S: Sheep, C: Cattle

232 Species abbreviations CH: Chicory (*Cichorium intybus* L.); SB: Salad burnet (*Sanguisorba*
 233 *minor* Scop.). RG: Ryegrass (*Lolium spec.* L.). YW: Yarrow (*Achillea millefolium* L.); other
 234 species abbreviations are the same as Table 1

235 * This complex mix contained two additional species that could not be identified

236

237

238 *Weed cover assessments*

239 Weed and crop species were assessed for percentage cover several times during the trial

240 duration, using 0.25 m² sectioned quadrats. Within the replicated trials (series I), visual cover

241 assessments were carried out at one of the sites only (Barrington Park), by estimating
242 percentage ground cover five times over the trial period in two quadrats per plot.

243 In the on-farm trials (series II), all weed and crop cover assessments were carried out with a
244 0.25 m² sectioned quadrat. On each farm, cover was assessed in four locations within each
245 treatment, i.e. both in the ASM strip and in an adjacent strip of the control ley, resulting in
246 eight assessment points per farm and date. Sampling locations were chosen randomly but at
247 least 10 m were left between any two assessment points. Assessments were performed twice
248 per farm: in 2009 several weeks after sowing (i.e. late spring in most cases) and in the
249 following year at a similar time in the growing season. Although this method, with a
250 relatively small total sampling area per farm and low temporal sampling frequency, did not
251 allow us to build complete species lists for each trial area, it provided information about the
252 most frequent weed species.

253 *Weed identification*

254 In most cases, weeds were identified to species level. Where this was not possible, individual
255 plants were assigned to a species group. For example, docks (*Rumex spp.*), could not always
256 be assigned to *R. crispus* L., *R. obtusifolius* L. or the hybrid *R. crispus* x *obtusifolius*.
257 Therefore, all docks were summarized under *Rumex spp.* However, where differentiation was
258 possible, *R. obtusifolius* was the most dominant taxon. Volunteer crops, such as potato
259 (*Solanum tuberosum* L.), wheat (*Triticum aestivum* L.) and oats (*Avena sativa* L.), which
260 were encountered in weed assessments were excluded from further data analysis.

261 *Weed and crop biomass measurements*

262 In the replicated trials, above ground biomass samples were taken in 2009, 2010 and 2011 on
263 five of the six trial sites (Table 2). Quadrats for sampling biomass had a size of 0.50 x 0.50
264 cm and were randomly placed within plots; along the length of the plots, the outer 1 m were

265 avoided for sampling to minimize edge effects. Sampling quadrats were aligned diagonally in
266 the plot. Sampling was performed on one sampling quadrat per plot (2009, 2010) or three
267 quadrats per plot (2011). While the samples were still fresh, weeds were manually separated
268 from crops and the weed and crop fractions were separately dried at 80 °C until sample
269 weights were constant. The timing of sampling in 2011 was chosen to reflect the situation
270 directly prior to incorporation of the ley into the soil.

271 *Soil sampling and other environmental variables*

272 Immediately prior to sowing in 2009, soil samples were taken on all trial sites, including the
273 on-farm trials. Soil samples were collected across the field with a soil corer to a depth of 15
274 to 20 cm (i.e. the typical depth of ploughing in the study area) and then bulked into a single
275 composite sample. Individual corer samples were obtained on each trial field when walking
276 the field in a W-shape with sampling points 2 to 4 m apart.

277 The samples (> 300 g) were air dried and analysed at Natural Resource Management Ltd
278 (Bracknell, UK) analytical laboratories. Samples were analysed for soil texture (percentage
279 sand, silt and clay) using pipette sedimentation. Textural classes followed the UK
280 Classification (Sand 2.00-0.063 mm, Silt 0.063-0.002 mm, Clay < 0.002 mm). Soil organic
281 matter was determined using the wet oxidation Walkley Black colorimetric method. Plant
282 available P was determined according to Olsen at 20 °C; plant available K was extracted
283 using 1 M NH₄NO₃ and K concentration was determined by flame photometry. Available Mg
284 was extracted using 1 M NH₄NO₃ and Mg concentration was determined using AAS.

285 Geographic coordinates (latitude, longitude and altitude; [Table 3](#)) of all sites were obtained
286 from publicly available digital maps. Management data such as sowing and cutting dates
287 were requested from the participating farmers.

288 *Statistical analysis*

289 All statistical analyses were performed with the programme R, version 2.14.1 [49].

290 **(1) Weed cover in All Species Mix and in monocultures**

291 We compared the cover in the ASM with the average cover from all component
292 monocultures, either weighted or not weighted by the respective seed density in the ASM.
293 The weighted average of weed cover was calculated as follows. If s_i is the seed rate of species
294 i (in g m^{-2}) in the ASM; and w_i is the weight per seed for species i (in g); then $n_i = s_i/w_i$ is the
295 number of sown plants per m^2 of species i within the ASM. The relative proportion p_i of the
296 species i in the ASM can then be defined as $p_i = n_i / \sum_i n_i$. If c_i is the weed cover in plots of
297 crop species i (in %), the average weed cover c_w across the monocultures of all species that
298 constitute the ASM, weighted by the proportion of species within the ASM is $c_w = \sum_i c_i p_i$,
299 whereas the unweighted average of the weed cover is $c_u = (\sum_i c_i)/m$, where m is the total
300 number of species in the ASM. Proportions of individual species within the ASM (measured
301 by the relative number of sown plants) were relatively high for white clover (0.166), large
302 birdsfoot trefoil (0.138) and birdsfoot trefoil (0.115), and relatively low for meadow pea
303 (0.001), sainfoin (0.014) and Italian ryegrass (0.019). The weighting by the relative seed
304 density in the ASM was performed to account for the unequal proportions of individual
305 species in the mixture. Specifically, assuming that the effects of individual species on weeds
306 increases with their proportion in the mixture, the expected weed cover in the ASM (in the
307 absence of any effects of diversity or absolute seed density) would be equal to the
308 proportional weed cover values in all constituent monocultures, i.e. c_w . Differences in weed
309 cover between ASM and the unweighted or weighted average of the monoculture were tested
310 with linear mixed effects models using days after sowing as continuous random effect.
311 Because this analysis revealed significant time x treatment interactions, treatment effects
312 were analysed for each time separately with one-factorial analyses of variance. Block effects
313 were non-significant in all cases of this analysis and were removed from the model.

314 Normality of model residuals was checked with the Shapiro-Wilks test. No significant
315 deviations from normality occurred in the weed cover data in the replicate trial.

316 **(2) Weed cover in All Species Mix compared to control ley on farms**

317 In the on-farm trials, weed cover data were analysed with analysis of variance to test
318 differences between ASM and control ley. However, weed cover data from 2009 and 2010
319 was found to be significantly non-normal ($P < 0.001$). Since non-normality of the 2009 data
320 could not be removed by (logarithmic) data transformation, a non-parametric sign test was
321 applied to data of both years. This test assesses the significance of the direction of the
322 difference between ASM and control ley. In addition, the 2010 weed cover data was log-
323 transformed and the transformed data subjected to an analysis of variance.

324 **(3) Weed biomass and crop biomass in different legume and grass monocultures**

325 Weed biomass and crop biomass in the replicated trials was analysed in the following way.
326 To account for strong site effects in weed and crop biomass, we first calculated for each plot
327 the relative differences (in weed biomass and crop biomass) between individual plot data and
328 site means, i.e. for weeds $W^*_{s,b,i} = (W_{s,b,i} - W_s) / W_s$ 100%, where W^* is the relative difference
329 in weed biomass from the site mean for species i at site s in block b ; $W_{s,b,i}$ is the absolute
330 weed biomass for species i at site s in block b and W_s is the site mean of absolute weed
331 biomass across all species and blocks. Analogous calculations were performed for crop
332 biomass to determine relative crop biomass as $C^*_{s,b,i} = (C_{s,b,i} - C_s) / C_s$ 100%. Further, to
333 determine the relationship between relative weed biomass W^* and relative crop biomass C^* ,
334 we performed a linear regression of W^*_i against C^*_i across species; in order to avoid inflation
335 of degrees of freedom and to account for non-independence of data within sites, values of
336 $C^*_{s,b,i}$ and $W^*_{s,b,i}$ were averaged across sites and blocks for each species prior to the analysis
337 of linear regression. In a subsequent analysis, residuals of individual species values from the

338 linear regression function of W^* against C^* were tested for significance based on a mixed-
339 effects model with site as a random factor, using the *lme* function in R.

340 To compare the various species with regard to, $K_i = W_i/(C_i+W_i)$, i.e. the proportion of weed
341 biomass in total above-ground biomass, the data from all sites was analysed with a linear
342 mixed-effects model with site as a random factor followed by Dunnett's test to separate
343 means of individual species from the means of a set control species; these control species
344 were chosen as white clover for the legume species and perennial ryegrass for the grass
345 species, because these species had been found to be most commonly used by the organic
346 farmers participating in the study (Table 4).

347 **(4) Change in the proportion of weed biomass over time**

348 The temporal change of the proportion K_{is} of weed biomass in total biomass was analysed by
349 comparing K_{is} from the last biomass sampling date against the first date (2011 vs. 2009). For
350 each species, the absolute difference in K_{is} between the two dates was tested for the direction
351 and significance of change by a two-tailed t-test against zero, based on a mixed effects model
352 with site and block within site as random factors, using the *lme* function in R. To make
353 comparisons among legume species, white clover was considered as a control and the
354 difference between this species and all other legume species was tested with a multiple
355 (many-to-one) comparisons test after Dunnett; the same test was employed to test the
356 difference between perennial ryegrass and the other grass species.

357 **(5) Weed floristic similarity between study years**

358 Weed floristic similarity between the two study years, based on presence vs. absence of
359 individual species in each of the two years, was compared using Jaccard's index with
360 confidence intervals given by Real [50]; Jaccard's index ranges from 0 (no similarity) to 1
361 (maximal similarity). For individual species, the change from the first to the second study

362 year in the number of farms or quadrats on which the species was found to be present was
363 tested for significance with χ^2 tests protected with a Bonferroni correction for multiple
364 testing.

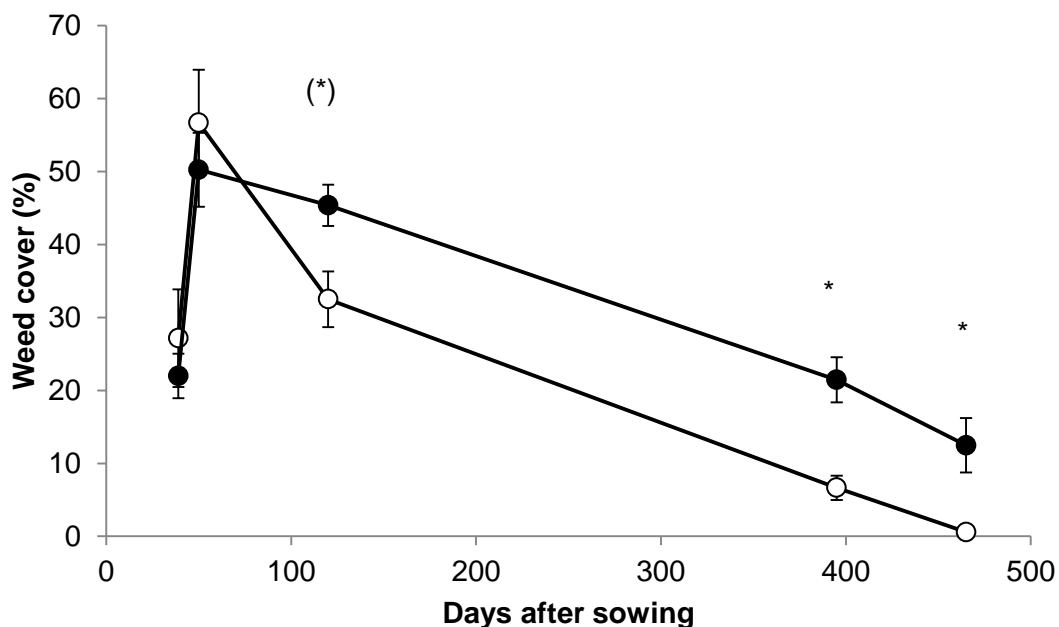
365 Results

366 Weed cover in All Species Mix and in monocultures

367 Weed cover at the Barrington Park site rose sharply in the first two months of the trial and
368 then declined gradually over the remaining duration of the trial (Figure 2). At the two later
369 assessments, weed cover in the ASM (c_{ASM}) was significantly lower than in the weighted
370 average c_w of the component species. The comparison between weed cover in the ASM and
371 the unweighted average c_u of the weed cover in the monoculture yielded similar results, with
372 c_{ASM} being significantly lower than c_u at the last three assessment dates.

373

374 Figure 2



375

376

377 **Figure 2:** Development of estimated weed cover (%) over time in a complex species mixture
378 of grasses and legumes (All Species Mix, ASM, open circles); and in the average of the
379 ASM's component species when grown in monocultures (weighted by relative plant density
380 in the ASM, filled circles); average over three replicates and standard errors (error bars); (*):
381 $P < 0.1$; *: $P < 0.05$ (t-test).

382

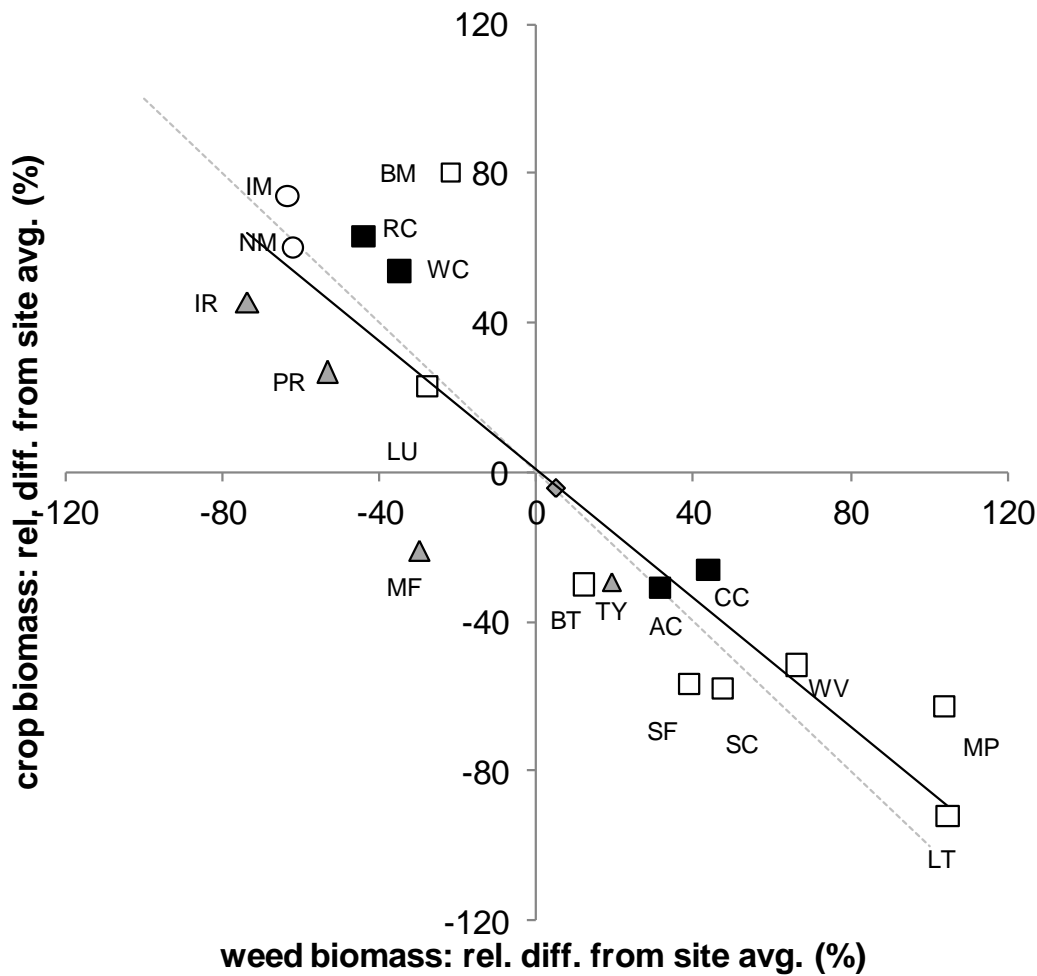
383 However, it was not possible to separate the weed reducing effect of increased plant density
384 in the ASM from effects of species richness, e.g. through increased weed suppression due to
385 complementarity of growth habits of the component species.

386 In the on-farm trials, average weed cover was 10.6 % in year 1 and 5.1% in year 2. Weed
387 cover was not significantly different between ASM and Control ley in either of the two trial
388 years following a sign test; also, no significant difference between ASM and control ley was
389 found for log-transformed weed cover data from 2010, following analysis of variance.

390

391 *Weed suppression by different crop species*

392 The legume species with the strongest weed suppression was red clover (**Figure 4**). For this
393 species, the proportion of weeds in total biomass at the first sampling was $28.3 \% \pm 9.9 \%$
394 across sites. Averaged across all legumes, the weed proportion in total biomass at the first
395 sampling was $56.0 \% \pm 7.6 \%$; for the grasses, this value was at $33.2\% \pm 7.0 \%$. There was a
396 strong and highly significant negative relationship between above ground crop biomass and
397 weed biomass across species (**Figure 4**; Adjusted $R^2 = 0.78$, $P < 0.001$, $df = 16$).



399
400

401 **Figure 3:** Relationship between weed biomass and crop biomass in early autumn 2009, both
 402 expressed as relative difference (in %) of species values from respective site averages. Filled
 403 squares: Clover species (*Trifolium* spec.); open squares: other legume species; grey triangles:
 404 grass species; open circles: All Species Mixtures (ASM); Grey diamond: average of
 405 monocultures (only ASM components); Black line: linear regression through all points;
 406 broken line: $y = -x$. Mean of five sites (all except Barrington Park). IM: Inoculated All
 407 Species Mixture; NM: Non-inoculated All Species Mixture; other abbreviations see Table 1.

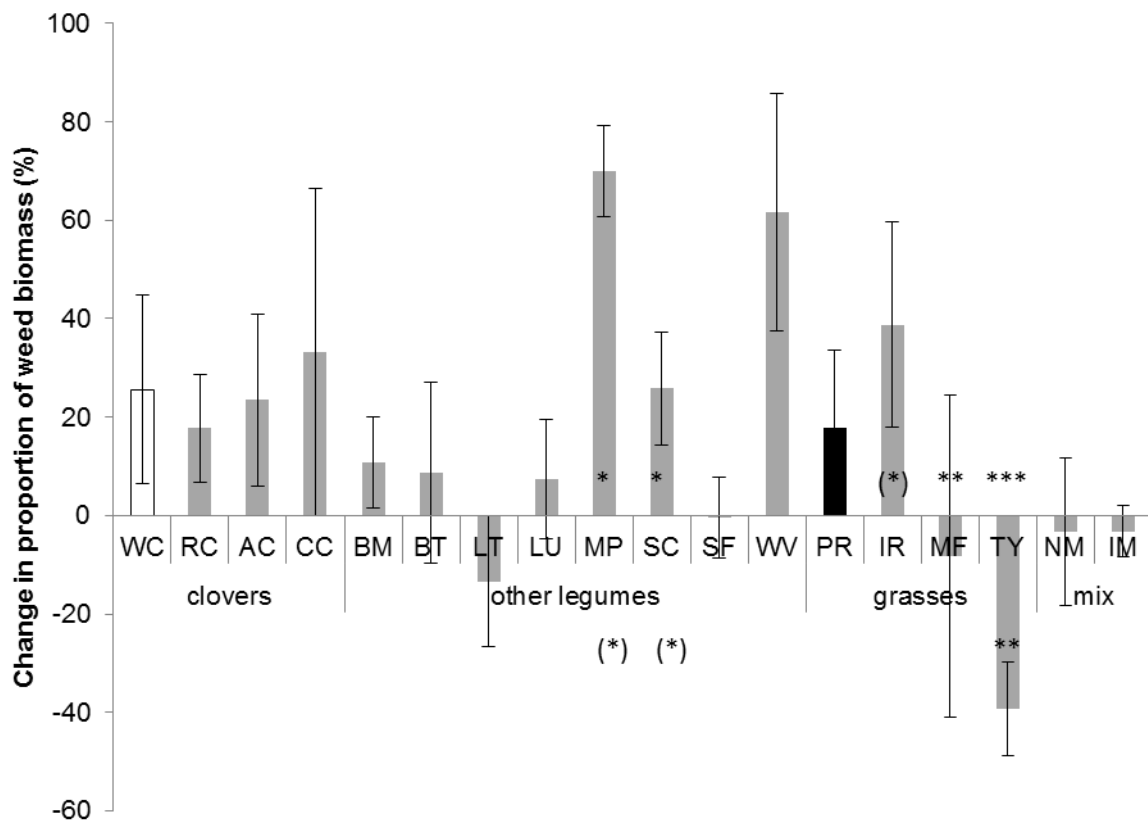
408

409 Interestingly, all four grass species were left of the regression line, i.e. their weed reducing
410 effect was higher than would be expected from their above ground crop biomass. To test the
411 significance of deviations from the regression, a mixed effects model with site as random
412 factor was run, followed by a t-test on the difference between observed values and values
413 estimated from regression line shown in [Figure 3](#). According to this analysis, there was a
414 significantly higher weed suppression ability in grasses than in legumes ($P < 0.001$). When
415 individual species were tested, the deviation from the regression line was only significant for
416 *F. pratensis*, ($P < 0.01$), but overall, *L. multiflorum* had the highest crop biomass and lowest
417 weed biomass ([Figure 3](#)).

418 In most species, the proportion of weed biomass within the total above ground biomass did
419 not significantly change over time, i.e. the absolute temporal change in the weed proportion,
420 over the period of autumn 2009 to spring 2011 was not significantly different from zero
421 ([Figure 4](#)).

422

423 Figure 4



424

425

426 **Figure 4:** Proportion (in %) of weed biomass in total biomass (above ground): Absolute
 427 change from autumn 2009 to spring 2011; means and standard errors across 4 sites. Positive
 428 values mean an increase in the proportion of weed biomass in the total above ground biomass
 429 over time. Significance stars *below* the zero-line indicate whether this temporal change was
 430 significantly different from zero (t-test); stars *above* the zero-line refer to the difference
 431 between white clover (white bar) and the other legume species and or the difference between
 432 perennial ryegrass (black bar) and the other grass species (Dunnett-test). (*): $P < 0.1$; *: $P <$
 433 0.05 ; **: $P < 0.01$; ***: $P < 0.001$. For abbreviations see Table 1. [

434

435 This indicates that the characteristics of species shown in [Figure 3](#) (relative crop biomass and
 436 weed suppression) were mostly consistent over the two years of the study, since the
 437 proportions of weeds in total biomass remained largely constant over time (with the

438 exception of Timothy grass). We observed a nearly significant ($0.05 < P < 0.1$) increase in the
439 proportion of weed biomass over time in only two of the legume monocultures, meadow pea
440 and white sweet clover (Figure 4). Among the grass species, the proportion of weeds in the
441 biomass significantly decreased in Timothy grass from autumn 2009 to spring 2011 ($P <$
442 0.01).

443

444 *Weed community composition in on-farm trials*

445 In total, 63 weed species were recorded in the leys. With a total of 56 weed species found in
446 the first year of the ley, the species richness was twice as large as in the second year, when
447 only 28 species were recorded. Similarly, the number of weed species per farm was higher in
448 the first than in the second year, with 11.9 ± 1.6 and 3.8 ± 0.7 weed species per farm,
449 respectively (average \pm standard error). Floristic similarity between the two study years (2009
450 and 2010), as measured by Jaccard's index on species presence in either of the two years, was
451 found to be 0.344; this was not significantly different from random similarity or dissimilarity
452 according to confidence intervals given by Real [50]. The total number of weed species found
453 on each farm, in both years together, ranged from 3 to 27. Weed species numbers between the
454 first and the second years of the study were uncorrelated across farms (linear model, adjusted
455 $R^2 = 0.08$, $P = 0.14$, $df = 16$), *i.e.* farms with a higher number of weed species in the first year
456 did not necessarily tend to have a higher species number in the second year as well.

457

458 Weed species richness did not correlate with the crop species richness sampled in the ley
459 (Adjusted $R^2 = 0.007$, $P = 0.247$), indicating that increasing the number of species within in a
460 ley mixture does not compromise the conservation of wild farmland plants. Similarly, for

461 both 2009 and 2010, the number of weed species was not significantly different between the
462 ASM and the Control leys.

463 In the first year of the ley (2009), the most frequently encountered weed species were
464 chickweed (*Stellaria media*), sow thistle (*Sonchus arvensis*) and field speedwell (*Veronica*
465 *persica*) (Table 5). In the second year of the ley, almost all annual species decreased in
466 frequency, i.e. the proportion of farms and of quadrats on which they were present decreased
467 over time. Conversely, some perennial species such as dandelion (*Taraxacum officinale* agg.)
468 and creeping thistle (*Cirsium arvense*) increased slightly but non-significantly in frequency.
469 However, *C. arvense*, as well as the other weed species *Rumex spp.* with recognized
470 economic relevance in organic agriculture, were relatively infrequent, being recorded in only
471 9 to 16 out of 168 sampling quadrats (Table 5).

472

473 **Table 5:** Weed species found in year 1 and 2 of the ley on 21 organic farms: Number of farms
 474 and number of quadrats in which the weed species were present, sorted in descending order
 475 by the number of quadrats in 2009 on which the species was present; (a) species with
 476 presence on a total 10 or more sampling quadrats; (b) species with presence on a total of
 477 fewer than 10 quadrats. For individual species, the change from the first to the second study
 478 year in the number of farms or quadrats on which the species was present was tested for
 479 significance with χ^2 tests protected with a Bonferroni correction for multiple testing (***: $P <$
 480 0.001; **: $P <$ 0.01; *: $P <$ 0.05). No significant effect of sampling year was found for
 481 species listed in (b).

| Species | (a) Presence in 10 or more quadrats | | No. quadrats | | |
|---|-------------------------------------|------|--------------|------|-----|
| | No. farms (out of 21) | | (out of 168) | | |
| | 2009 | 2010 | 2009 | 2010 | |
| <i>Stellaria media</i> (L.) Vill. | 15 | 7 | 82 | 15 | *** |
| <i>Sonchus arvensis</i> L. | 10 | 0 | 38 | 0 | *** |
| <i>Veronica persica</i> Poiret | 8 | 0 | 35 | 0 | *** |
| <i>Persicaria maculosa</i> L. | 11 | 0 * | 33 | 0 | *** |
| <i>Ranunculus repens</i> L. | 9 | 5 | 32 | 13 | |
| <i>Viola arvensis</i> Murray | 9 | 2 | 32 | 7 | ** |
| <i>Spergula arvensis</i> L. | 6 | 0 | 26 | 0 | *** |
| <i>Veronica spec.</i> L. | 5 | 4 | 26 | 6 | * |
| <i>Chenopodium album</i> L. | 8 | 1 | 23 | 2 | ** |
| <i>Poa annua</i> L. | 7 | 0 | 21 | 0 | *** |
| <i>Lamium purpureum</i> L. | 6 | 2 | 20 | 3 | * |
| <i>Myosotis arvensis</i> (L.) Hill | 7 | 1 | 20 | 1 | ** |
| <i>Sinapis arvensis</i> L. | 5 | 4 | 20 | 5 | |
| <i>Anagallis arvensis</i> L. | 5 | 0 | 19 | 0 | ** |
| <i>Tripleurospermum maritimum</i> (L.) Koch | 4 | 0 | 19 | 0 | ** |
| <i>Capsella bursa-pastoris</i> (L.) Medik. | 5 | 0 | 18 | 0 | ** |
| <i>Galeopsis tetrahit</i> L. | 4 | 0 | 17 | 0 | ** |
| <i>Polygonum spec.</i> L. | 4 | 0 | 16 | 0 | ** |
| <i>Rumex spec.</i> L. | 8 | 5 | 16 | 11 | |
| <i>Anthemis arvensis</i> L. | 4 | 2 | 15 | 7 | |
| <i>Convolvulus arvensis</i> L. | 5 | 1 | 15 | 1 | |
| <i>Fallopia convolvulus</i> (L.) Löve | 5 | 0 | 15 | 0 | * |
| <i>Papaver rhoeas</i> L. | 4 | 2 | 14 | 4 | |
| <i>Polygonum aviculare</i> L. | 3 | 0 | 14 | 0 | * |
| <i>Taraxacum officinale</i> F.H. Wigg | 5 | 8 | 13 | 22 | |
| <i>Galium aparine</i> L. | 3 | 1 | 12 | 1 | |
| <i>Cirsium arvense</i> (L.) Scop. | 5 | 2 | 9 | 12 | |
| <i>Cerastium fontanum</i> Baumg. | 2 | 3 | 8 | 5 | |
| <i>Elymus repens</i> (L.) Gould | 1 | 1 | 8 | 3 | |
| <i>Achillea millefolium</i> L. | 2 | 1 | 4 | 6 | |
| <i>Aphanes arvensis</i> L. | 1 | 2 | 3 | 11 | |

482

483

| Species | No. Farms (out of 21) | | No. Quadrats (out of 168) | |
|---|--------------------------|------|------------------------------|------|
| | 2009 | 2010 | 2009 | 2010 |
| <i>Senecio vulgaris</i> L. | 4 | 0 | 9 | 0 |
| <i>Alopecurus myosuroides</i> Huds. | 3 | 0 | 9 | 0 |
| <i>Vicia hirsuta</i> (L.) Gray | 1 | 0 | 8 | 0 |
| <i>Geranium spec.</i> L. | 4 | 1 | 5 | 1 |
| <i>Avena fatua</i> L. | 2 | 0 | 5 | 0 |
| <i>Kickxia elatine</i> (L.) Dumort. | 2 | 0 | 5 | 0 |
| <i>Plantago major</i> L. | 2 | 0 | 5 | 0 |
| <i>Sisymbrium officinale</i> (L.) Scop. | 2 | 0 | 4 | 0 |
| <i>Matricaria recutita</i> L. | 1 | 0 | 4 | 0 |
| <i>Brassica napus</i> L. | 1 | 0 | 3 | 0 |
| <i>Legousia hybrida</i> (L.) Delarbre | 1 | 0 | 3 | 0 |
| <i>Veronica arvensis</i> L. | 2 | 1 | 2 | 6 |
| <i>Mentha arvensis</i> L. | 2 | 0 | 2 | 0 |
| <i>Matricaria discoidea</i> DC | 2 | 0 | 2 | 0 |
| <i>Glebionis segetum</i> (L.) Fourr. | 1 | 0 | 2 | 0 |
| <i>Kickxia spuria</i> (L.) Dumort. | 1 | 0 | 2 | 0 |
| <i>Aethusa cynapium</i> L. | 1 | 0 | 1 | 0 |
| <i>Fumaria officinalis</i> L. | 1 | 0 | 1 | 0 |
| <i>Lactuca serriola</i> L. | 1 | 0 | 1 | 0 |
| <i>Lapsana communis</i> L. | 1 | 0 | 1 | 0 |
| <i>Odontites vernus</i> Dumort. | 1 | 0 | 1 | 0 |
| <i>Poa trivialis</i> L. | 1 | 0 | 1 | 0 |
| <i>Senecio jacobaea</i> L. | 1 | 0 | 1 | 0 |
| <i>Urtica urens</i> L. | 1 | 0 | 1 | 0 |
| <i>Poa spec.</i> L. | 0 | 1 | 0 | 8 |
| <i>Cirsium vulgare</i> (Savi) Ten. | 0 | 2 | 0 | 5 |
| <i>Cichorium intybus</i> L. | 0 | 1 | 0 | 4 |
| <i>Arabidopsis thaliana</i> (L.) Heynh. | 0 | 1 | 0 | 2 |
| <i>Daucus carota</i> L. | 0 | 1 | 0 | 1 |
| <i>Sherardia arvensis</i> L. | 0 | 1 | 0 | 1 |

485

486 **Discussion**

487 Within the context of organic rotations in Europe, this study addresses two contrasting
 488 aspects of weeds in agricultural rotations, namely weed control and weeds as constituents of
 489 farm biodiversity. It highlights, therefore, the potential conflict between agronomic and
 490 biodiversity aspects of agricultural production.

491 *General observations*

492 Overall, we found total weed cover in the range of 5.1-10.6 % in the on-farm trials, which is
 493 comparable to values of total weed cover in grass/clover leys reported in a study on weeds in

494 organic rotations in the North of England [18:Eyre]. In the replicated trial at Barrington Park
495 however, we observed much higher weed cover. It is likely that differences between these
496 observations are due to different sampling times, since there is a large time effect on weed
497 cover (Figure 2).

498 In the replicated trials, crop biomass and weed biomass were inversely related (Figure 4),
499 confirming earlier findings [e.g. 51, 52]. Only one species deviated significantly from the
500 regression between the two parameters relative weed biomass and relative crop biomass;
501 meadow fescue had a lower weed biomass than would be predicted given its crop biomass
502 (Figure 4).

503 This result indicates that crop productivity, measured as above-ground biomass per unit area,
504 is an excellent indicator of competitiveness against weeds. At the same time, this relationship
505 may to some degree suggest functional complementarity between crops and weeds. In
506 monocultures with relatively low crop biomass, weeds filled the gap, thus resulting in
507 relatively high weed biomass. In arable cash crops there is (almost) no complementarity
508 between crops and weeds. In terms of yield as the primary function of the cash crop, weeds
509 make no direct positive contribution; on the contrary, weeds limit yields through competition.
510 Leys with their associated weeds are different in this respect. Many functions are fulfilled by
511 both the ley crop and weed species, e.g. covering the soil and thereby protecting it from
512 erosion, providing plant residues for building up to soil organic matter or supporting
513 pollinators and other beneficial insects. Although some central functions of the sown ley
514 species such as nitrogen fixation are not fulfilled by the majority of weed species, there is at
515 least some degree of functional complementarity between crops and weeds in rotational leys.
516 Apart from this, there is a further important difference between weeds in leys and weeds in
517 arable cash crops. In leys, the time between emergence of weeds and their destruction
518 through mowing is typically shorter than between weed emergence and harvest of arable

519 crops. Therefore, many annual weed species may not have completed their life cycle and set
520 seed before the ley is cut. In fact, the first cut of organic leys is often timed before weeds
521 have produced seed. For these reasons, we suggest that weeds can be tolerated in organic leys
522 to a higher degree than in organic cash crops. However, it is currently unclear where the
523 balance lies between functional complementarity and functional antagonism of sown ley
524 species vs. weeds.

525

526 *Characterisation of individual legume and grass species*

527 In this study, we found that the proportion of weeds and crops in total above-ground biomass
528 did not significantly change between the first and the last sampling time for most species
529 (Figure 5); this result is unexpected because of the asymmetry of competition typically
530 observed in plant communities [53]. With asymmetric competition it would be predicted that
531 proportions of crops or weeds change over time, as the competition dynamics lead to shifts in
532 the proportion of species towards the dominating species. There may be several reasons why
533 our observations do not support the expectations arising from asymmetric competition. First,
534 the sampling effort may have been insufficient to detect significant effects over time.
535 Similarly, the study period may not have been long enough for asymmetric competition to
536 become apparent. Also, in leys competition between crops and weeds may be reset to a
537 certain degree with each cut and with the break in vegetative growth over winter. While
538 spring-germinating annual weed species form a new generation each spring, most legumes
539 tested here are perennials, but they also need to re-grow after winter, or after cutting. In
540 contrast to most of the ley species assessed in this study, in three species we found significant
541 shifts over time in the proportion of weeds, namely white sweet clover and meadow pea
542 (towards an increasing proportion of weeds), as well as timothy grass (towards an increasing
543 proportion of the crop). In the cases of meadow pea and sweet clover, the observed increase

544 in the proportion of weeds was likely due to poor recovery of plant growth following
545 mowing. Large variation across sites (indicated by large standard errors) was observed for
546 crimson clover with respect to the change of weed proportion over time (Figure 4). This
547 species is annual but is able to re-grow from seed; here, shifts over time in the proportion of
548 crops and weeds may reflect variation in the ability of the crop to produce a second
549 generation.

550 Differences observed among species in their competitiveness against weeds may to some
551 extent reflect the intensity of plant breeding efforts. It is indeed reasonable to assume that
552 there is a positive feedback relationship between a species' productivity and the breeding
553 efforts dedicated to it. For instance, both red clover and white clover, in this study found to be
554 the two species with the strongest weed suppression (Figure 4), have received much more
555 attention from breeders than the other legume species trialled here, which can be interpreted
556 both as a reason for and a consequence of the relatively high productivity of white and red
557 clover. Further, this study found that grasses outperformed legumes in terms of weed
558 suppression, which is in line with earlier findings on the smaller weed suppression abilities of
559 legumes in comparison to grasses [e.g. 13, 15].

560 The analysis of the individual legume species also shows that there is a degree of redundancy
561 in the ASM, where some species (such as meadow pea) perform too poorly to warrant an
562 inclusion in ley mixtures. Thus, mixtures with fewer species, but with complementary
563 functions, may optimise weed management (and crop performance) in leys. This has been
564 supported by analyses of potential mixtures with different numbers of the species trialled in
565 this study [43].

566

567 *Weed communities in on-farm trials*

568 This study suggests that several weed species are dominant in organically managed leys
569 typical in the UK and that weed species richness may be higher than previously reported [16].
570 With the dominating *Stellaria media*, *Sonchus arvensis* and *Veronica persica* we found
571 species that are common and typical annual weeds of arable fields in the UK and throughout
572 Western Europe. With their short life cycles they are adapted to high-disturbance regimes.
573 With an average value of 11.9, the number of weed species encountered per farm was slightly
574 greater than in a single-site study investigating the effects of rotations on weeds, where only 9
575 weed species were recorded from a grass/clover ley [18]. Further, our results showed that
576 annual weed species typical for arable fields were dominant in the year of establishment of
577 the ley. In terms of weed communities the start of the ley phase is thus similar to those found
578 in arable crops. On some sites, the ley was, in fact, undersown into cereals. Further, the weed
579 community changed considerably in the second year, towards perennial and grassland
580 species, most probably owing to the cessation of tillage and the repeated cutting, mulching or
581 grazing. This change in community composition from annual to perennial species following
582 the changes in land managed is typical and has been observed in several other studies [e.g.
583 13, 54].

584 However, as pointed out in the Methods section, the sampling strategy for the weed species in
585 the on-farm trials was not designed to generate an exhaustive picture of the weed flora in
586 organic fertility building leys. In particular, because of spatial aggregation in weeds [55], the
587 number of quadrats for sampling in on-farm trials was likely too small to reliably detect all
588 species present on the farms. Therefore, it is likely that the data obtained for species richness
589 on the organic leys underestimate the actual weed species richness [cf. 21]. Similarly, the
590 actual frequency of species on the farms, i.e. the proportion of farms on which a given
591 species is present, is likely to be higher than measured with our sampling method. Further,
592 the methods applied here do not allow us to build a picture of the weed species present in the

593 seed bank. Finally, it is not known to which degree the ley management, e.g. cutting vs.
594 grazing, had an impact on weed communities but this aspect was outside the scope of this
595 study.

596

597 *Ley species mixtures and weeds*

598 Compared to the average of monocultures, the ASM was found to have significantly lower
599 weed cover (Figure 2), and ranked among the best performers with regard to both crop
600 biomass and weed biomass (Figure 4). However, these effects cannot be ascribed to the
601 mixing of species, since diversity effects and density were confounded in this study. Seed
602 density in the ASM was 53.4 % higher than the average seed density of all component
603 monocultures. In the on-farm trials, ASM was not significantly better at controlling weeds
604 than the control leys. However, sowing rates for the control leys were not recorded.
605 Therefore, it remains speculative whether differences in seed densities between ASM and
606 control leys might be a reason for the observed results.

607 Generally, there is evidence that mixing species does help to control weeds, especially when
608 crops are functionally diverse [56]. A study on weeds in short-term grassland showed weed
609 suppression to be higher in mixtures than in monocultures [57]. Weed suppression in annual
610 species mixtures has also been found to be better than in monocultures [47, 58, 59]. Further,
611 because of functional complementarity among different sown species, seed densities in multi-
612 species mixtures may generally be increased above the sowing rates used in respective
613 monocultures or simpler mixtures with a lesser degree of complementarity. Thus, higher plant
614 densities – made possible by mixing multiple species – may then be used as a tool to suppress
615 weeds [60]. At the same time, further research is necessary to separate species richness
616 effects on weeds from the impact of plant density in leys.

617 Our on-farm trials show that weed species richness as a component of farm biodiversity is not
618 significantly reduced when including more crop species in the ley, in contrast to earlier
619 findings [61]. Weed species richness in the ley is more likely to be influenced by the history
620 and landscape features [62] of any particular site. In the first year of establishment, leys may
621 be seen to provide a suitable habitat for arable weeds. For the later stages of the ley, whilst
622 annual weed species decline, the challenge remains to control perennial weeds such as
623 creeping thistle (*Cirsium arvense*) and docks (*Rumex spp.*). However, we speculate that these
624 species are again likely to be mostly influenced by site history (e.g. tillage [63]) and to be
625 relatively unaffected by the choice of species in a ley mixture.

626

627 **Conclusions**

628 In the past, the question of what organic agriculture contributes to the conservation of
629 farmland biodiversity has been researched extensively [64], showing biodiversity benefits of
630 organic farming in comparison with conventional farming [65, 66]. In this debate, little
631 attention has so far been paid to organic leys, despite legume based leys being an essential
632 feature of many organic systems, in particular in Europe. No direct comparison is therefore
633 possible with conventional agriculture, because typically there is no ley phase in current
634 conventional rotations [e.g. 67, 68]. Organic leys add to the diversity on farms by including a
635 range of crop species that are otherwise not cultivated. This study has shown that organic leys
636 harbour a range of wild plant species that further contribute to species richness on the farm.
637 Recent evidence shows that young leys (<1.5 years old) provide a better habitat for spiders
638 than cereal fields [69]. Leys therefore constitute an important element of farm biodiversity.
639 As we have demonstrated, the choice of species in organic leys can be used to optimise weed
640 control. It remains open to which degree the ecological functions provided by weeds may be

641 fulfilled by designing targeted crop mixtures, i.e. by replacing weeds with crops while
642 maintaining their ecological functions. However, it is unlikely that effective protection of rare
643 weed species can be achieved through ley design only. Further research is needed to show
644 how leys can be optimized for multifunctional performance.

645

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652

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