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► **To cite this version:**

Mark Szenteczki, Adrienne L Godschalx, Andrea Galmán, Anahí Espíndola, Marc Gibernau, et al.. Spatial and temporal heterogeneity in pollinator communities maintains within-species floral odour variation. *Oikos*, 2021, 130 (9), pp.1487-1499. 10.1111/oik.08445 . hal-03335599

**HAL Id: hal-03335599**

**<https://hal.science/hal-03335599>**

Submitted on 6 Sep 2021

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**Spatial and temporal heterogeneity in pollinator communities maintains within-species floral odour variation**

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1

2 **ABSTRACT**

3 Flowering plants emit complex bouquets of volatile organic compounds (VOCs) to mediate  
4 interactions with their pollinators. These bouquets are undoubtedly influenced by pollinator-  
5 mediated selection, particularly in deceptively-pollinated species that rely on chemical  
6 mimicry. However, many uncertainties remain regarding how spatially and temporally  
7 heterogeneous pollinators affect the diversity and distribution of floral odour variation. Here,  
8 we characterized and compared the floral odours of ten populations of deceptively-pollinated  
9 *Arum maculatum* (Araceae), and inter-annual and decadal variation in pollinator attraction

10 within these populations. Additionally, we transplanted individuals from all sampled  
11 populations to two common garden sites dominated by different pollinator species (*Psychoda*  
12 *phalaenoides* or *Psycha grisescens*), and compared pollinator attraction rates to investigate  
13 whether populations maintained odour blends adapted to a specific pollinator. We identified  
14 high within- and among-population variation in a common blend of VOCs found across the  
15 range of *A. maculatum*. We also observed shifts in pollinator community composition within  
16 several populations over 1-2 years, as well as over the past decade. Common garden  
17 experiments further revealed that transplanted inflorescences generally attracted the dominant  
18 local pollinator species in both transplant sites. However, one population (Forêt du Gâvre,  
19 France) appears to exclusively attract *P. grisescens*, even when transplanted to a *P.*  
20 *phalaenoides*-dominated site. Together, our results suggest that maintaining diverse floral  
21 odour bouquets within populations may be advantageous when pollinator communities vary  
22 over short timescales. We propose that temporally-replicated ecological data are one potential  
23 key to understanding variation in complex traits such as floral odour, and in some cases may  
24 reveal resiliency to shifting pollinator communities.

25

26 **KEYWORDS** balancing selection, evolutionary ecology, floral volatiles, plant-pollinator  
27 interactions, reciprocal transplant, temporal variation

28

## 29 **1. INTRODUCTION**

30 Widely distributed species are subject to an array of eco-evolutionary settings across their  
31 range, due to variation in abiotic conditions and biotic interactions through space and time.  
32 These mosaics of selection are known to influence the traits that underpin diverse plant-  
33 herbivore and plant-pollinator interactions, affecting the evolutionary trajectories of all species  
34 involved (Levin 2000; Hendry 2017). Plant-pollinator interactions appear to be a particularly

35 important mechanism underlying the comparatively rapid rate of speciation in flowering plants  
36 (van der Niet & Johnson 2012; Hernández-Hernández & Wiens 2020); this macroevolutionary  
37 pattern is the result of microevolutionary processes acting on floral traits at the intraspecific  
38 level (Herrera et al. 2006). Understanding the evolutionary and functional ecology of complex  
39 floral traits therefore requires data on how plant-pollinator interactions vary through both space  
40 and time (Waser & Ollerton 2006). However, characterizing complex trait variation and biotic  
41 interactions across wide geographic ranges (e.g. Friberg et al. 2019) and through time (e.g.  
42 Fishbein & Venable 1996) requires substantial effort. As a result, few studies have  
43 simultaneously investigated the impact of spatial and temporal variation in pollinators on  
44 intraspecific floral trait variation.

45

46 As the dominant pollinators across all terrestrial ecosystems, insects exert key selective  
47 pressures on many floral traits. They may be attracted or repelled by odour (Whitehead &  
48 Peakall 2009), colour (Goyret et al. 2007; du Plessis et al. 2018), morphology (Ibanez et al.  
49 2010, Murúa & Espíndola 2015), nectar (Parachnowitsch et al. 2019), pollen (Dobson &  
50 Bergström 2000), or the multimodal expression of several of these traits (reviewed in Junker  
51 & Parachnowitsch 2015). Here, we focus on floral odour bouquets, which are often complex  
52 blends of many volatile organic compounds (VOCs) that mediate diverse interactions between  
53 plants, insects, and microbes (Holopainen 2004; Raguso 2008).

54

55 Recent studies on plant-pollinator interactions have identified many cases of pollinator-driven  
56 evolution of floral odour (Chess et al. 2008; Klahre et al. 2011; Breitkopf et al. 2013; Peter &  
57 Johnson 2014; Gross et al. 2016). However, a review by Delle-Vedove et al. (2017) found that  
58 floral odour variation often cannot be explained by pollinator-mediated selection alone. Several  
59 factors may explain this discrepancy, including trade-offs between pollinator attraction and

60 chemical defence against herbivory (Schiestl et al. 2014), the nature of the interaction (*i.e.*  
61 deceptive pollination; Renner 2006), biochemical and energetic limitations (Delle-Vedove et  
62 al. 2011), phylogenetic constraints in the biosynthetic pathways for VOC production (Raguso  
63 et al. 2006), gene flow (Svensson et al. 2005), or genetic drift (Suinyuy et al. 2012). Another  
64 factor that may maintain floral odour variation within populations is temporal heterogeneity in  
65 pollinator community composition, as a result of pollinator declines (Yuan et al. 2009;  
66 Thomann et al. 2013; IPBES 2016; Zattara & Aizen 2021) or shifts in pollinator phenology  
67 (Burkle & Runyon 2019). However, the impact of temporally heterogeneous pollinators on  
68 floral odour is not yet clearly understood, because most of the aforementioned studies relied  
69 on VOC and pollinator data collected at a single timepoint. Here, we aim to address this gap in  
70 our knowledge, by examining how spatio-temporal variation in pollination interactions  
71 influences floral odour variation in *Arum maculatum* L. (Araceae).

72

73 *Arum maculatum* inflorescences (Figure 1) are pollinated deceptively, by emitting complex  
74 VOC blends which are known to vary within and among populations in England (Kite 1995;  
75 Kite et al. 1998; Diaz & Kite 2002), France (Chartier et al. 2011; Chartier et al. 2013), and  
76 across the Alps (Gfrerer et al. 2021). The floral odour of *A. maculatum* is similar to dung or  
77 decomposing organic matter (Lack & Diaz 1991), mimicking the natural brood sites of their  
78 main pollinators, the moth flies *Psychoda phalaenoides* L. and *Psycha grisescens* Tonnoir  
79 (Diptera: Psychodidae). Since pollinators are temporarily trapped in a specialized floral  
80 chamber (Bröderbauer et al. 2013) until the day after anthesis (Gibernau et al. 2004), it is  
81 possible to collect accurate quantitative data on pollinators attracted by individual  
82 inflorescences. Furthermore, pollinators appear to be attracted by VOCs alone (Dormer 1960;  
83 Urru et al. 2011), meaning that an inflorescence's reproductive success should be closely tied  
84 to its unique floral odour. Since *A. maculatum* floral odour is known to vary, and VOCs such

85 as indole, 2-heptanone, and p-cresol are attractive to *Psychoda* species (Kite et al. 1998), we  
86 chose to focus on floral odour in this study. Specifically, we investigated whether VOC  
87 variation is maintained within populations due to temporally variable pollinator communities,  
88 or across the species distribution due to spatial divergence in pollinator communities.

89

90 Floral odour variation may be maintained within populations through balancing selection  
91 (Delph & Kelly 2013) and/or phenotypic plasticity (Callaway et al. 2003; Majetic et al. 2009;  
92 Campbell et al. 2019). In the case of *A. maculatum*, balancing selection may occur if relative  
93 abundances of *P. phalaenoides* and *P. grisescens* vary temporally within populations (e.g.  
94 Schemske & Horvitz 1989), or if pollinator learning or avoidance to escape deception (Ayasse  
95 et al. 2000; Baguette et al. 2020) leads to frequency-dependent selection. In either case, we  
96 would expect to observe high floral odour variation within populations of *A. maculatum* across  
97 Europe. Phenotypic plasticity under variable abiotic and biotic conditions may also promote  
98 the maintenance of trait variation (Gulisija et al. 2016). If we observe shifts in VOC emissions  
99 when *A. maculatum* inflorescences are transplanted to non-native conditions, we may conclude  
100 that plasticity also contributes to the maintenance of floral odour variation.

101 Alternatively, if pollinators only vary spatially, they may exert divergent selective pressures  
102 on floral VOCs, leading to locally-adapted bouquets in populations with different pollinators  
103 (Leimu & Fischer 2008, Gervasi & Schiestl 2017; Suinyuy et al. 2018; Sayers et al. 2020).

104 Previous research has shown that *P. phalaenoides* and *P. grisescens* are respectively trapped  
105 by inflorescences in the northern/central and southern/western portions of *A. maculatum*'s  
106 distribution (Espíndola et al. 2010) — except in the Atlantic fringe of France, where  
107 inflorescences trap *P. grisescens*. Furthermore, the population genetic structure of *A.*  
108 *maculatum* roughly aligns with this pattern (Espíndola & Alvarez 2011). Under spatially  
109 varying selection, we may predict that distinct *A. maculatum* floral VOCs or phenological

110 adaptations will be maintained in these two pollinator backgrounds. If these variations are local  
111 adaptations, we would expect to observe decreased pollinator attraction efficiency when  
112 inflorescences are transplanted to non-native pollinator backgrounds.

113

114 In this study, we investigated whether variation in *A. maculatum* floral odour was consistent  
115 with the patterns predicted under either of the two scenarios described above. To this aim, we  
116 surveyed variation in floral odour and pollinator attraction across Europe, and transplanted  
117 individuals from all sampled populations to two common garden sites dominated by either *P.*  
118 *phalaenoides* or *P. grisescens*. By selecting populations where pollinator communities were  
119 first surveyed a decade ago (Espíndola et al. 2010), we were further able to assess temporal  
120 heterogeneity in pollinators at both the inter-annual (*i.e.* our surveys between 2017-2019), and  
121 decadal scales, with the aim of understanding how temporally heterogeneous pollinators affect  
122 intraspecific floral odour variation. Our specific objectives were to (1) characterize and  
123 compare floral odour variation within and among populations of *A. maculatum*, (2) determine  
124 whether the pollinators trapped by *A. maculatum* vary spatially and/or temporally, and (3) use  
125 these results to infer potential drivers of floral VOC variation, namely pollinator-driven  
126 balancing selection and local adaptation.

127

## 128 **2. MATERIALS AND METHODS**

### 129 *2.1 – Natural history & pollination ecology of Arum maculatum*

130 *Arum maculatum* is a common European flowering plant, which is usually found in shaded  
131 areas of deciduous woodlands, particularly beech woods (Brown 2000). Its pollination cycle  
132 takes place over two days (reviewed in Gibernau et al. 2004). On the evening of the first day,  
133 the appendix is thermogenic and emits a dung-like odour to attract dipteran pollinators; heat  
134 alone is not sufficient to attract pollinators (Prime 1960), but likely aids in the dispersal of

135 VOCs such as indole, p-cresol, and 2-heptanone, which are attractive to *Psychoda* species when  
136 used in scented traps (Kite et al. 1998). Notably, the widely distributed *Psychoda phalaenoides*  
137 and *Psycha grisescens* are both trapped by *A. maculatum* across most of Europe, together  
138 representing 87.49% of all insects found within floral chambers (Espíndola et al. 2010).  
139 *Psychoda setigera*, *P. albipennis*, and *P. trinodulosa* are also infrequently observed within  
140 inflorescences, together representing 0.46% of trapped insects in Espíndola et al. (2010). Other  
141 infrequently attracted dipterans include Chironomidae (*Smittia pratorum*), Ceratopogonidae,  
142 and Sphaeroceridae (Brachycera) (Rohacek et al. 1990; Diaz & Kite 2002; Espindola et al.  
143 2010).

144

145 Psychodidae are also known to be efficient carriers of *Arum* pollen (Albre et al. 2003; Diaz &  
146 Kite 2002); one individual can carry 50-150 grains of pollen, and a significant fruit set can  
147 develop from a single trapped pollinator (Lack & Diaz 1991). Based on their similar behaviours  
148 and morphologies, the above-mentioned *Psychoda* species are all presumably efficient  
149 pollinators of *A. maculatum*. However, antennal sensilla are known to vary among Psychodidae  
150 (Faucheux & Gibernau 2011), and species may therefore vary in their responses to VOCs.  
151 Although *A. maculatum* do not provide any nutritive rewards, this should not be detrimental to  
152 trapped *P. phalaenoides* and *P. grisescens*, as they do not feed during their short adult lifespan  
153 (P. Withers, cited in Lack & Diaz 1991). The conservation status of *P. phalaenoides* and *P.*  
154 *grisescens* has not been assessed; however, *Psychoda* populations are known to be large and  
155 dense (up to thousands of individuals per m<sup>2</sup>; Arshad & Moh Leng 1991) due to their reliance  
156 on modern agriculture and human activities (e.g. manure and decomposing organic matter) for  
157 reproduction (Satchell 1947; Vaillant 1971). Peak *P. phalaenoides* abundances also appear to  
158 correspond with the peak flowering period of *A. maculatum* (Ollerton & Diaz 1999).

159

## 160 2.2 – *Field sampling sites*

161 We sampled ten populations of *A. maculatum* (Figure 1 & Appendix S1, Table S1), including  
162 three in France (Forêt du Gâvre, Conteville, and Chaumont), two in Switzerland (Neuchâtel  
163 and Cortaillod), one in Italy (Montese), one in Croatia (Visuč), two in Serbia (Gostilje and  
164 Sokobanja), and one in Bulgaria (Chiflik). This sampling covers the majority of the *A.*  
165 *maculatum* species distribution range (Appendix S1, Figure S1). During the typical flowering  
166 period of *A. maculatum* (April – May), we conducted field sampling each year between 2017  
167 and 2019, visiting each site in at least two years during this timeframe.

168

## 169 2.3 – *Common garden experiment sites*

170 We selected the Swiss population of Neuchâtel and the French population Forêt du Gâvre (on  
171 the Atlantic fringe) as our common garden sites, since they were respectively dominated by  
172 *Psychoda phalaenoides* and *Psycha griseascens*. We conducted common garden experiments in  
173 Neuchâtel in 2018 and 2019, and in Forêt du Gâvre in 2019. These two sites experienced a  
174 relatively similar climate; over the course of our sampling in 2019, the mean temperature and  
175 humidity in Neuchâtel at the time of sampling were 15.8°C and 61% respectively, and in Forêt  
176 du Gâvre, the mean temperature and humidity were 17.2°C and 50% respectively. Five to ten  
177 *A. maculatum* individuals with unopened inflorescences were potted in soil from their native  
178 habitat, and then transplanted from all ten populations listed above to the two common garden  
179 sites. Inflorescences from Neuchâtel and Forêt du Gâvre were also reciprocally transplanted as  
180 part of this experiment. The total numbers of inflorescences sampled *in situ* and in both  
181 common garden sites are given in Appendix S1, Figure S1.

182

## 183 2.4 – *Floral odour and pollinator sampling methods*

184 During both the field surveys and common garden experiments, we collected dynamic  
185 headspace VOCs using identical methods, and analysed them using gas chromatography  
186 coupled to mass spectrometry (GC-MS; full details in Appendix S2 - Supplementary Methods).  
187 Briefly, we collected VOCs from *A. maculatum* inflorescences undergoing anthesis in the early  
188 evening on polydimethylsiloxane (PDMS) coated Twister® stir bars (Gerstel: Mülheim an der  
189 Ruhr, Germany), at an air flow rate of 200 mL min<sup>-1</sup> for 30 minutes. Twisters® were kept on  
190 ice in sealed glass containers until GC-MS analyses, where volatiles were thermally desorbed  
191 and separated on a HP-5MS column. During the morning following VOC sampling, we  
192 collected all insects trapped within inflorescences and preserved them in 70% ethanol until  
193 identification (full identification methods detailed in Appendix S2 - Supplementary Methods).

194

#### 195 *2.5 – Characterizing and comparing floral odour variation across the range of A. maculatum*

196 We compared total VOC emissions within and among populations of field-sampled  
197 inflorescences (*i.e.* VOCs sampled in their native habitats) using Bray-Curtis distance matrices,  
198 and visualized inter-individual variation using nonmetric multidimensional scaling (NMDS).  
199 Then, to test for a significant effect of population (fixed effect factor) on the entire VOC matrix,  
200 we performed pairwise permutational multivariate analysis of variance (PERMANOVA, Bray-  
201 Curtis distance,  $n = 999$  permutations) with Bonferroni correction using the *adonis* function in  
202 the R v.3.6.1 (R Core Team 2019) package *vegan* (Oksanen et al. 2019).

203

#### 204 *2.6 – Characterizing temporal variation in pollinator community composition*

205 We investigated inter-annual variation in pollinator community composition during our study  
206 (*i.e.* 2017 – 2019) by calculating Bray-Curtis distance matrices, comparing the mean quantities  
207 of pollinators trapped per inflorescence for all populations with *in situ* pollinator data collected  
208 in two or more years. We then visualized the result using nonmetric multidimensional scaling

209 (NMDS) ordinations. Next, we repeated this process to investigate shifts in the dominant  
210 pollinators trapped by *A. maculatum* over the past decade. Using Bray-Curtis distance matrices  
211 and NMDS ordinations, we compared the mean quantities of 1) Psychodidae species only, and  
212 2) all insect families trapped per inflorescence in our study, with those collected approximately  
213 a decade ago (2006-2008; Espíndola et al. 2010).

214

### 215 2.7 – Identifying VOCs associated with species-specific pollinator attraction

216 Since we collected paired VOCs and pollinator data for each sampled inflorescence, we were  
217 able to correlate these two matrices and identify candidate compounds associated with the  
218 attraction of either *P. phalaenoides* or *P. grisescens*. To this aim, we used the Random Forest  
219 implementation in the R package *randomForest* (Liaw & Wiener 2002), with permutation  
220 importance enabled (*n tree* = 500, *m try* = 8; optimized using the *tuneRF* function). We then  
221 calculated conditional feature contributions and identified combinations of compounds which  
222 had the greatest influence on the predictive strength of the classifier, using the Python package  
223 *TreeInterpreter* (Saabas 2019). As a measure of plasticity in these candidate compounds when  
224 flowering in different habitats (*i.e.* transplant effects), we calculated and plotted population  
225 standard scores of these compounds for both *in situ* and common garden samples. We then  
226 used Mann-Whitney *U*-tests to test whether populations with sufficient sample sizes ( $n \geq 8$ )  
227 emitted different quantities of candidate compounds when transplanted to common garden  
228 sites.

229

### 230 2.8 – Testing for local adaptation to pollinators using common garden experiments

231 We began by visualizing geographic patterns in mean pollinator attraction for each population  
232 *in situ*, and following transplants to both common garden sites. Then, following the ‘local vs.  
233 foreign’ definition of local adaptation (Kawecki & Ebert 2004), we tested the hypotheses that:

234 in the Neuchâtel common garden, transplanted *A. maculatum* inflorescences which attract *P.*  
235 *phalaenoides* in their native population should catch 1) more *P. phalaenoides* and/or 2) more  
236 pollinators in total than inflorescences which attract *P. griseascens* in their native population.  
237 We expected to observe the inverse result in the Forêt du Gâvre common garden (*i.e.*  
238 transplanted inflorescences that attract *P. griseascens* in their native population should perform  
239 better on average in Forêt du Gâvre). These expectations were tested using a two-way ANOVA  
240 on log+1 transformed pollinator counts, including ‘native pollinator’ (*i.e.* *P. phalaenoides*- or  
241 *P. griseascens*-dominated origin) and ‘common garden location’ (*i.e.* whether the common  
242 garden site was dominated by *P. phalaenoides* or *P. griseascens*) and their interaction as fixed  
243 factors. Here, a significant interaction would indicate local adaptation, which could be  
244 confirmed using contrasts comparing trait values between native and transplant sites. Finally,  
245 we analysed ‘deme × habitat’ interactions (Kawecki & Ebert 2004) for mean attraction rates of  
246 *P. phalaenoides*, *P. griseascens*, and all insects. Here, deme and habitat respectively referred to  
247 an *A. maculatum* population and its local pollinator community conditions. In this analysis,  
248 data were subset based on each inflorescence’s native pollinator and common garden location,  
249 as described above.

### 250 **3. RESULTS**

#### 251 *3.1 – Floral odour is highly variable within and among A. maculatum populations*

252 After filtering out compounds present in blank samples, we retained 18 *A. maculatum* floral  
253 VOCs present in relative abundances above 1% (Table 1). All of the major compounds we  
254 identified (e.g. indole, p-cresol, 2-heptanone,  $\beta$ -citronellene, and three unnamed  
255 sesquiterpenes) had been previously reported in studies of *A. maculatum* floral odour (Diaz &  
256 Kite 2002; Chartier et al. 2013; Marotz-Clausen et al. 2018). We observed substantial variation  
257 in floral odour both within (Figure 1) and among populations (Figure 2).

258

259 3.2 – *Pollinator communities are variable at both annual & decadal scales*

260 Our sampling allowed us to compare inter-annual (*i.e.* 2017/18 vs. 2019) variation in  
261 Psychodidae trapped by *A. maculatum* in six populations. The dominant Psychodidae species  
262 trapped by inflorescences appear to have shifted in three of these populations (Conteville, FR,  
263 Neuchâtel, CH, and Visuć, HRV) over this time period (Figure 3).

264

265 We also observed temporal shifts in the average abundances of trapped Psychodidae in several  
266 populations over the past decade (*i.e.* compared to Espíndola et al. 2010). The relative  
267 compositions of the complete pollinator communities trapped by *A. maculatum* appears to have  
268 shifted in five out of six populations where comparisons could be made; only Forêt du Gâvre  
269 remained consistent over the past decade (Appendix S1, Figure S2). We also observed apparent  
270 shifts in the dominant Psychodidae pollinator trapped within inflorescences in four out of six  
271 populations over the past decade, in Chaumont FR, Conteville FR, Gostilje SRB, and Visuć,  
272 HRV (Table 2 & Appendix S1, Figure S2).

273

274

275 3.3 – *Low population-level VOC divergence coincides with temporally variable pollinators*

276 PERMANOVA pairwise contrasts did not identify any pairs of populations with significant  
277 divergence in their proportional emissions of VOCs sampled in their native habitats, after  
278 correction for multiple testing. This lack of population-level differentiation in VOC blends is  
279 evident when visualizing Bray-Curtis similarities between field-sampled individuals (Figure  
280 2), and appears to coincide with the temporal heterogeneity in pollinator communities  
281 identified in the previous section (see Figure 3 and Appendix S1, Figure S2).

282

283 3.4 – *Arum maculatum populations typically are not locally adapted to specific pollinators*

284 Through Random Forest analyses (Appendix S1, Figure S3), we identified one compound  
285 positively associated with *P. phalaenoides* attraction ( $\beta$ -humulene), and three compounds  
286 positively associated with *P. griseascens* attraction (unnamed sesquiterpenes RI 1470 and 1681,  
287 and  $\alpha$ -selinene). After evaluating combined feature contributions within the Random Forest  
288 classifier, we found that its predictive strength was most strongly influenced by unnamed  
289 sesquiterpene (RI 1681) alone. Other strong combinations included unnamed sesquiterpene RI  
290 1681 paired with unnamed sesquiterpene RI 1470 or  $\alpha$ -selinene, as well as  $\beta$ -humulene alone,  
291 mirroring our initial results (full *Treeinterpreter* results in Appendix S1, Table S2).

292

293 The four candidate compounds we identified ( $\beta$ -humulene, unnamed sesquiterpenes RI 1470  
294 and 1681, and  $\alpha$ -selinene) were widely observed and not restricted to a specific region or  
295 pollinator background. Average emissions of these compounds remained relatively consistent  
296 between *in situ* collections and samples transplanted to Neuchâtel, though some appeared to  
297 increase after inflorescences were transplanted to Forêt du Gâvre (see Appendix S1, Figure  
298 S4). We only observed a significant transplant effect in one compound (an increase in unnamed  
299 sesquiterpene RI 1470) within one population, Neuchâtel (Mann-Whitney tests; full result in  
300 Appendix S1, Table S3).

301

302 In both common garden sites, we observed that transplanted inflorescences typically attracted  
303 the dominant local pollinator species as efficiently as native inflorescences (Figure 1). While  
304 common garden location had a significant effect on the quantity of *P. phalaenoides* and *P.*  
305 *griseascens* caught by *A. maculatum* (*i.e.* the two transplant sites were dominated by different  
306 Psychodidae species), no native pollinator  $\times$  common garden location interaction effect was  
307 observed (2-way ANOVA,  $\text{Pr}( > F ) > 0.05$ ; full results in Appendix S1, Table S4). Together,  
308 these results suggest that *A. maculatum* populations are generally not locally adapted to a single

309 pollinator species (Figure 4), with notable exceptions in Forêt du Gâvre and the two Serbian  
310 populations Gostilje and Sokobanja, which will be discussed below.

311

312 While we do not find general indications of populations being locally adapted to specific  
313 pollinators, not all inflorescences were equally attractive to all pollinator species. One  
314 population (Forêt du Gâvre) continued to exclusively attract their native pollinator *P.*  
315 *grisescens* when transplanted to the Neuchâtel common garden (see Figure 1). Remarkably,  
316 this exclusive attraction of *P. grisescens* was maintained even though *P. phalaenoides* was also  
317 present (and trapped by inflorescences from other populations) at the time when inflorescences  
318 from Forêt du Gâvre opened (see Appendix S1, Figure S5a). Additionally, transplanted  
319 inflorescences occasionally attracted the ‘non-dominant’ Psychodidae species in both common  
320 garden sites, and a third *Psychoda* species (*Psychoda trinodulosa*) was also identified within  
321 inflorescences in Croatia and Serbia during our field surveys. *Psychoda trinodulosa* was also  
322 observed in the Neuchâtel common garden, but not in Forêt du Gâvre; inflorescences from both  
323 Serbian populations (Gostilje and Sokobanja) continued to occasionally attract *P. trinodulosa*  
324 when transplanted to the Neuchâtel common garden (Figure 1).

325

#### 326 **4. DISCUSSION**

327 In this study, we identified substantial within-and among- population variation in *A. maculatum*  
328 floral odour (Figure 1 and Figure 2) and shifts in pollinator community composition in several  
329 populations at both the inter-annual (Figure 3) and decadal scales (Appendix S1, Figure S2).  
330 Although some *A. maculatum* populations continued to attract their native pollinators in  
331 transplant sites (e.g. Forêt du Gâvre and Serbian populations), most populations did not  
332 exclusively attract *P. phalaenoides* or *P. grisescens* when transplanted to ‘foreign’ pollinator  
333 backgrounds (Figure 4). Since this result does not appear to be due to transplant effects /

334 phenotypic plasticity (Appendix S1, Table S3), this leaves temporally heterogeneous pollinator  
335 communities as a potential mechanism underlying the maintenance of high within-population  
336 variation in floral odour.

337

#### 338 4.1 – *The influence of temporally variable pollinators on A. maculatum floral odour*

339 Overall, it appears that local pollinator availability at the time of anthesis plays a key role in  
340 the quantity and composition of pollinators trapped within *A. maculatum* inflorescences.  
341 Previous studies on *A. maculatum* pollination in England (Diaz & Kite 2002) and France  
342 (Chartier et al. 2013) arrived at a similar conclusion. The difference in sex ratios, particularly  
343 that no male *P. phalaenoides* or *P. trinodulosa* are attracted, may be due to differences in  
344 copulation timing among *Psychoda* species (*i.e.* immediately after adult emergence), and  
345 parthenogenetic reproduction (F. Vaillant, cited in Albre et al. 2003), or brood-site specific  
346 VOCs which are only attractive to females of these two species. While we were unable to  
347 reliably collect data on background pollinator communities as part of our sampling design,  
348 Diaz & Kite (2002) found that Psychodidae species caught on sticky traps mirrored those found  
349 within *A. maculatum* and *A. italicum* inflorescences. Therefore, our data should still allow us  
350 to infer whether the patterns in pollinator attraction we observed within each population were  
351 a consequence of balancing selection or local adaptation.

352

353 Balancing selection may maintain variation within populations through several selective  
354 regimes, including relaxed selection, negative frequency-dependent selection due to pollinator  
355 learning, or environmental heterogeneity (Delph & Kelly 2013). While relaxed selection on  
356 floral odour has been observed in some angiosperms (Salzmann et al. 2007), this is unlikely to  
357 be the case for deceptively pollinated *A. maculatum*, given that pollinator attraction is driven  
358 by VOC emissions alone (Dormer 1960; Lack & Diaz 1991). Pollinator learning has been

359 shown to maintain polymorphism in floral colour (Gigord et al. 2001) and odour (Ayasse et al.  
360 2000), but this is also less likely to occur in the case of *A. maculatum*. Psychodidae likely  
361 experience weak and inconsistent selective pressures caused by deception, due to their large  
362 population sizes and short generation times (Prime 1960; Lachmann et al. 2000), and  
363 potentially sex-specific attraction in some cases. Araceae also exploit pre-existing VOC  
364 detection abilities in their pollinators (Schiestl & Dötterl 2012), which likely contributes to  
365 their persistent inability to distinguish deceptive inflorescences (Renner 2006). This leaves  
366 temporal heterogeneity in pollinator communities as the mechanism most likely contributing  
367 to the maintenance of diverse floral odour bouquets in *A. maculatum*.

368

369 In this study, we demonstrated that pollinator communities can vary within populations over  
370 relatively short time periods (*i.e.* over the years- to decade-scale) across most of the range of  
371 *A. maculatum*. Temporally heterogeneous pollinators have long been known to influence floral  
372 traits (Schemske & Horvitz 1989), but to our knowledge, have not been investigated as a  
373 mechanism underlying floral odour variation. Further research on temporal variation in  
374 pollinators may provide clarity in other cases where floral trait variation in deceptively  
375 pollinated species cannot be explained by pollinator learning (e.g. Pellegrino et al. 2005;  
376 Jersáková et al. 2006). Temporally variable pollinators may have also contributed to the finding  
377 that, contrary to expectations, deceptively pollinated species generally do not maintain more  
378 variable floral odours than rewarding species (Ackerman et al. 2011; Delle-Vedove et al. 2017).

379

380 Currently, it is not known whether the phenologies of *P. phalaenoides* and *P. grisescens* are  
381 influenced by environmental variation; our data indicate that at least in Forêt du Gâvre, *P.*  
382 *grisescens* emerges slightly earlier than *P. phalaenoides* (Appendix S1, Figure S5b). A recent  
383 environmental DNA survey of cow dung (Sigsgaard et al. 2020) also identified *P.*

384 *phalaenoides*, *P. trinodulosa*, and *P. grisescens* all within a single site in Mols Bjerger,  
385 Denmark. At the time the sample was taken (June 2019; *i.e.* later than our field surveys), *P.*  
386 *phalaenoides* overwhelmingly predominated over all other insects in terms of read counts (*i.e.*  
387 abundance). Taken together, these results corroborate two key points: all of the major  
388 Psychodidae pollinators of *A. maculatum* are present across the range of *A. maculatum* (Ježek  
389 et al. 2018), and their relative abundances may vary temporally over the course of the flowering  
390 period.

391

392 If environmental variation influences pollinator phenology, then plasticity in floral odour based  
393 on the same environmental cues might enhance pollinator attraction. With one exception in a  
394 single population (*i.e.* unnamed sesquiterpene RI 1470 emitted by inflorescences from  
395 Neuchâtel), we did not observe significant shifts in the four candidate compounds linked to  
396 species-specific pollinator attraction following transplants to common gardens (*i.e.* phenotypic  
397 plasticity). However, as we transplanted mature inflorescences, some environmental effects  
398 may have already been in place prior to transplanting (Wund 2012). Growing *A. maculatum*  
399 for several generations in common garden sites would give a more accurate result and eliminate  
400 maternal effects, but this was not feasible within the scope of our study, as *A. maculatum*  
401 requires at least two years (and sometimes longer) from germination to flowering (Brown  
402 2000). Given the central role of VOCs in *A. maculatum* pollination (Kite 1995; Kite et al,  
403 1998), it is unlikely that the high variation in floral odour we observed is the result of plasticity  
404 alone. Further research tracking the VOC emissions of the same individual over multiple  
405 flowering events would provide greater resolution into environmental influences on floral  
406 odour variation.

407

408 Since *A. maculatum* are rhizomatous, variation may also persist within populations for longer  
409 periods of time compared to annual plants. Gene flow between populations may also contribute  
410 to the maintenance of floral odour variation within populations. Pollen dispersal by  
411 Psychodidae is likely limited due to their poor flying abilities and short adult lifespans (Lack  
412 & Diaz 1991), but seed dispersal by frugivorous birds (Snow & Snow 1988) could lead to gene  
413 flow among nearby populations. While there appears to be a strong barrier to gene flow  
414 between populations from north/central Europe, and from Italy and the Balkans (Espíndola &  
415 Alvarez 2011), our data suggest that floral odour variation is widely maintained across this  
416 barrier. This pattern suggests that — at least regionally — balancing selection is at work,  
417 possibly in association with phenotypic plasticity and/or local adaptation in populations with  
418 consistent pollinator communities.

419

#### 420 4.2 – *The influence of spatially variable pollinators on A. maculatum floral odour*

421 The results from our Europe-wide transplant and common garden experiment do not support  
422 the hypothesis that all *A. maculatum* populations are (locally) adapted to attract exclusively *P.*  
423 *phalaenoides* or *P. grisescens*. This contrasts with the patterns found in other wide-ranging  
424 deceptively pollinated species such as the sexually deceptive orchids *Ophrys sphegodes*  
425 (Breitkopf et al. 2013) and *Ophrys insectifera* (Triponez et al. 2013), but aligns with the results  
426 of a previous reciprocal transplant experiment between two *A. maculatum* populations in  
427 France (Chartier et al. 2013). However, we found that the Forêt du Gâvre population in coastal  
428 NW France appears to have lost the ability to attract *P. phalaenoides*, possibly due to local  
429 adaptation and/or genetic drift; the latter process is known to occur at the limits of species  
430 ranges (Geber 2011; Gould et al. 2013). Interestingly, Forêt du Gâvre is also the only  
431 population in this study with little recorded inter-annual and decade-scale variation in  
432 pollinator communities. It is therefore possible that the pollinator attraction patterns we

433 observed are a result of selective pressures imposed by the likely stable population of *P.*  
434 *grisescens* in this site. Further transplants of Forêt du Gâvre inflorescences to additional  
435 populations are needed to test this hypothesis.

436

437 Our results also do not exclude the possibility that individual VOCs are differentially attractive  
438 to certain Psychodidae species. First, we identified four sesquiterpenes that were able to predict  
439 the attraction of *P. phalaenoides* ( $\beta$ -humulene) and *P. grisescens* (unidentified sesquiterpenes  
440 RI 1470 and 1681, and  $\alpha$ -selinene) through Random Forest analyses. Additionally, both  
441 Serbian populations in this study (Gostilje and Sokobanja) attracted a third Psychodidae species  
442 (*P. trinodulosa*) *in situ* and following transplants to the Neuchâtel common garden, suggesting  
443 that there may also be VOCs related to species-specific attraction of *P. trinodulosa*. These  
444 patterns suggest that specific compounds emitted by inflorescences from Serbia may also be  
445 differentially attractive to *P. trinodulosa*. However, this species was too infrequently observed  
446 in our study to make any firm conclusions; further studies of these populations are needed to  
447 address this question.

448 In this study, the VOCs typically associated with Psychodidae brood sites (e.g. indole, p-cresol,  
449 and 2-heptanone) were not correlated with species-specific pollinator attraction. A recent study  
450 by Gfrerer et al. (2021) also found that variation in the aforementioned foetid-smelling VOCs  
451 were not among those which significantly influenced fruit sets in *A. maculatum*. Given that  
452 blends of indole, p-cresol, and 2-heptanone are known to be attractive to *Psychoda* species  
453 (Kite et al. 1998), these compounds appear to be generally attractive to all species. However,  
454 blends of VOCs (e.g. with specific sesquiterpenes) or the ratios at which they are emitted may  
455 be differentially attractive. Behavioural assays, where different pollinator species are presented  
456 with several *A. maculatum* inflorescences or individual VOCs, would be useful in identifying  
457 which compounds or blends elicit general or species-specific responses. Similarly, Gas

458 Chromatography-Electroantennography (GC-EAD; Cork et al. 1990) could be used to identify  
459 all VOCs which elicit a physiological response in different pollinator species.. The *A.*  
460 *maculatum* populations with unique patterns in pollinator attraction identified in this study may  
461 be useful targets for future research testing whether selection is acting on specific compounds  
462 (e.g. Strensmyr et al. 2002; Urru et al. 2010), or on “super-attractive mixtures” (e.g. in *A.*  
463 *palaestinum*; Stökl et al. 2010). In either case, our results still allow us to conclude that variable  
464 floral odour blends are effective in the specialized

465

#### 466 *Conclusion*

467 To date, almost all studies on floral odour and pollinator variation have been carried out at a  
468 single timepoint — possibly contributing to the numerous cases where floral odour diversity  
469 appears to exceed pollinator diversity (Delle-Vedove et al. 2017). As evidenced by the  
470 extensive literature on local adaptation in plants (Leimu & Fischer 2008), spatially varying  
471 selection is an undoubtedly important driver of floral odour divergence, and appears to have  
472 influenced a few *A. maculatum* populations (e.g. Forêt du Gâvre) as well. However, the  
473 variable floral odour bouquets emitted by *A. maculatum* may also represent an important  
474 adaptation to temporally heterogeneous pollinator communities, if the pollinator shifts we  
475 observed have been occurring over longer periods of time. In closing, we advocate for  
476 increased attention on the temporal dimension of pollinator-mediated selection. By  
477 simultaneously studying spatial and temporal variation in selection, we may further our  
478 understanding of how and why flowering plants maintain high diversity in key functional traits  
479 such as floral odour.

480

#### **ACKNOWLEDGEMENTS**

We thank Gregory Roeder for his assistance with the processing of our VOC samples, Jérôme Albre for his assistance in Psychodidae identification, and Alberto Garcia Jimenez and Monica Fleisher for their dedicated assistance during our field sampling. We also thank Laurent Oppliger and colleagues at the Jardin Botanique de Neuchâtel for their support in hosting and maintaining our experimental populations of *Arum* inflorescences. We are grateful to the journal editor Dr. Ignasi Bartomeus, and the two anonymous reviewers for their constructive comments on this manuscript. The project was funded by the Swiss National Science Foundation through grant 31003A\_163334 awarded to N.A and S.R.

481

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## TABLES

**Table 1.** Average proportional emissions (M) of volatile organic compounds (+/- SD) emitted by north/ central populations of *Arum maculatum*. Compounds representing more than 5% of the average total blend are highlighted in bold.

VOC	RI	Forêt du Gâvre, France (n = 14)			Conteville, France (n = 10)			Chaumont, France (n = 8)			Neuchâtel, Switz- erland (n = 21)			Cortailod, Switz- erland (n = 29)		
		M	SD	Freq	M	SD	Freq	M	SD	Freq	M	SD	Freq	M	SD	Freq
2-heptanone	891	1.09	1.69	50.0	<b>7.74</b>	<b>16.54</b>	<b>40.0</b>	4.68	10.37	25.0	0.14	0.29	28.6	1.12	2.01	41.4
$\beta$ -citronellene	943	3.58	6.02	64.3	0.02	0.07	10.0	2.25	2.28	75.0	<b>6.32</b>	<b>7.17</b>	<b>81.0</b>	3.39	3.74	82.8
cis $\beta$ -ocimene	1037	0.10	0.37	7.1	0.00	0.00	0.0	0.02	0.05	12.5	0.32	0.75	33.3	0.51	1.02	27.6
p-cresol	1076	4.04	7.54	42.9	<b>5.71</b>	<b>10.15</b>	<b>30.0</b>	0.04	0.12	12.5	0.79	2.11	14.3	0.00	0.00	0.0
indole	1289	<b>22.50</b>	<b>15.45</b>	<b>100</b>	<b>19.92</b>	<b>20.11</b>	<b>90.0</b>	<b>15.23</b>	<b>15.61</b>	<b>87.5</b>	<b>39.02</b>	<b>30.26</b>	<b>95.2</b>	<b>46.26</b>	<b>26.81</b>	<b>96.6</b>
skatole	1383	1.89	2.84	50.0	1.44	2.13	50.0	4.21	2.28	87.5	1.59	1.84	57.1	1.62	1.41	69.0
$\alpha$ -copaene	1374	0.02	0.04	28.6	0.01	0.02	20.0	0.25	0.53	25.0	0.02	0.07	14.3	0.48	2.06	20.7
Z-caryophyllene	1405	<b>9.26</b>	<b>11.07</b>	<b>71.4</b>	1.87	4.44	30.0	2.11	2.00	62.5	<b>8.39</b>	<b>7.52</b>	<b>85.7</b>	3.94	4.74	75.9
$\beta$ -caryophyllene	1416	1.48	2.14	50.0	<b>7.87</b>	<b>9.84</b>	<b>70.0</b>	1.92	2.30	62.5	1.37	1.93	47.6	2.82	5.68	72.4
$\alpha$ -humulene	1452	<b>5.15</b>	<b>4.43</b>	<b>71.4</b>	<b>17.55</b>	<b>22.93</b>	<b>70.0</b>	<b>11.32</b>	<b>2.66</b>	<b>100</b>	<b>5.82</b>	<b>5.44</b>	<b>76.2</b>	3.87	3.10	72.4
alloaromadendrene	1459	4.66	3.81	85.7	3.96	4.53	60.0	<b>7.02</b>	<b>4.26</b>	<b>87.5</b>	<b>5.62</b>	<b>4.41</b>	<b>85.7</b>	<b>8.09</b>	<b>9.94</b>	<b>82.8</b>
$\beta$ -humulene	1473	<b>5.46</b>	<b>7.02</b>	<b>64.3</b>	1.24	1.95	40.0	<b>10.13</b>	<b>5.87</b>	<b>87.5</b>	4.97	6.45	85.7	2.72	1.91	82.8
$\alpha$ -selinene	1491	2.06	3.68	35.7	0.35	1.12	10.0	3.97	4.52	62.5	1.66	3.43	28.6	0.43	1.18	17.2
bicyclogermacrene	1493	0.58	1.02	42.9	0.87	2.75	10.0	1.18	1.70	50.0	2.64	4.07	57.1	<b>6.46</b>	<b>9.51</b>	<b>75.9</b>
d-cadinene	1520	0.48	1.09	35.7	1.63	4.66	20.0	<b>9.59</b>	<b>12.96</b>	<b>100</b>	3.75	6.47	57.1	1.55	2.14	58.6
Unnamed sesqui.	1404	0.63	1.21	42.9	0.04	0.14	10.0	0.20	0.37	37.5	4.94	10.48	57.1	2.49	6.65	37.9
Unnamed sesqui.	1470	4.62	6.00	71.4	2.17	3.98	30.0	<b>11.17</b>	<b>5.59</b>	<b>100</b>	3.48	3.27	76.2	3.96	2.82	79.3
Unnamed sesqui.	1681 <sup>^</sup>	<b>32.39</b>	<b>20.75</b>	<b>100</b>	<b>27.61</b>	<b>17.68</b>	<b>80.0</b>	<b>14.71</b>	<b>8.88</b>	<b>100</b>	<b>9.15</b>	<b>14.36</b>	<b>81.0</b>	<b>10.29</b>	<b>10.45</b>	<b>75.9</b>

**Table 1 (cont).** Average proportional emissions (M) of volatile organic compounds (+/- SD) emitted by southern (Italy) and eastern (Balkan) populations of *Arum maculatum*. Compounds representing more than 5% of the average total blend are highlighted in bold.

VOC	RI	Montese, Italy (n = 13)			Visuč, Croatia (n = 6)			Gostilje, Serbia (n = 19)			Sokobanja, Serbia (n = 11)			Chiflik, Bulgaria (n = 3)		
		M	SD	Freq	M	SD	Freq	M	SD	Freq	M	SD	Freq	M	SD	Freq
2-heptanone	891	<b>8.28</b>	<b>14.17</b>	<b>61.5</b>	<b>29.36</b>	<b>27.79</b>	<b>100</b>	0.00	0.00	0.0	0.00	0.00	0.0	0.00	0.00	0.0
β-citronellene	943	3.26	5.49	69.2	<b>8.77</b>	<b>2.18</b>	<b>100</b>	0.65	2.12	21.1	0.05	0.15	9.1	0.00	0.00	0.0
cis β-ocimene	1037	0.21	0.40	30.8	0.32	0.79	16.7	0.01	0.04	5.3	0.00	0.00	0.0	0.00	0.00	0.0
p-cresol	1076	<b>7.58</b>	<b>11.99</b>	<b>61.5</b>	4.34	5.81	66.7	0.00	0.00	0.0	0.00	0.00	0.0	2.85	4.94	33.3
indole	1289	<b>22.38</b>	<b>16.81</b>	<b>92.3</b>	<b>7.99</b>	<b>9.73</b>	<b>83.3</b>	<b>55.41</b>	<b>26.79</b>	<b>100</b>	<b>43.41</b>	<b>37.98</b>	<b>90.9</b>	<b>54.24</b>	<b>27.05</b>	<b>100</b>
skatole	1383	1.46	1.49	61.5	1.07	1.10	66.7	4.34	5.80	73.7	2.92	3.19	63.6	1.84	0.45	100
α-copaene	1374	0.32	0.64	46.2	0.10	0.20	50.0	1.61	4.51	68.4	4.46	10.94	72.7	2.52	2.65	66.7
Z-caryophyllene	1405	<b>12.78</b>	<b>14.31</b>	<b>69.2</b>	<b>22.25</b>	<b>20.66</b>	<b>83.3</b>	4.65	6.29	63.2	<b>9.88</b>	<b>19.53</b>	<b>45.5</b>	<b>6.90</b>	<b>3.86</b>	<b>100</b>
β-caryophyllene	1416	0.88	1.20	38.5	1.96	2.50	50.0	0.54	2.06	15.8	0.73	2.34	18.2	0.00	0.00	0.0
α-humulene	1452	6.25	8.75	76.9	<b>6.11</b>	<b>5.24</b>	<b>100</b>	<b>6.91</b>	<b>10.62</b>	<b>57.9</b>	1.81	3.34	27.3	0.70	1.21	33.3
alloaromadendrene	1459	2.89	2.70	69.2	2.04	1.65	83.3	1.86	3.22	36.8	4.71	5.97	54.5	0.73	1.26	33.3
β-humulene	1473	<b>8.07</b>	<b>13.22</b>	<b>100</b>	0.75	1.09	50.0	1.54	2.77	42.1	2.64	4.09	36.4	0.67	1.16	33.3
α-selinene	1491	0.88	1.55	30.8	0.00	0.00	0.0	1.38	3.12	31.6	0.47	1.11	18.2	0.00	0.00	0.0
bicyclogermacrene	1493	0.43	0.69	38.5	0.00	0.00	0.0	1.02	1.60	47.4	0.97	2.18	27.3	<b>6.95</b>	<b>12.04</b>	<b>33.3</b>
d-cadinene	1520	1.36	2.55	46.2	0.62	0.96	33.3	2.36	8.28	21.1	0.28	0.76	18.2	0.00	0.00	0.0
Unnamed sesqui.	1404	0.47	0.87	30.8	0.00	0.00	0.0	0.99	2.61	42.1	0.74	1.43	27.3	0.00	0.00	0.0
Unnamed sesqui.	1470	3.35	3.61	69.2	4.06	2.17	100	2.99	5.27	52.6	4.97	7.83	36.4	4.00	1.71	100
Unnamed sesqui.	1681 <sup>^</sup>	<b>19.15</b>	<b>18.28</b>	<b>84.6</b>	<b>10.23</b>	<b>12.36</b>	<b>100</b>	<b>13.73</b>	<b>13.14</b>	<b>84.2</b>	<b>21.96</b>	<b>20.18</b>	<b>81.8</b>	<b>18.60</b>	<b>15.25</b>	<b>100</b>

RI = Kovats retention index (<sup>^</sup> = RI 1716 in Diaz & Kite 2002)

Freq = proportion of chromatograms (%) in which the compound was identified in a given population.

**Table 2.** Mean *in-situ* abundances of *Psychoda phalaenoides* and *Psycha grisescens* individuals trapped per *Arum maculatum* inflorescence between 2006-2008 (data from Espíndola et al. 2010), and in our field surveys between 2017-2019. The number of individual inflorescences sampled *in situ* are indicated in parentheses. NAs indicate that no inflorescences were open yet at the time populations were visited.

<b>Poll. Species / Year Population</b>	<b><i>P. phalae- noides</i> (2008)</b>	<b><i>P. grisescens</i> (2008)</b>	<b><i>P. phalae- noides</i> (2019)</b>	<b><i>P. grisescens</i> (2019)</b>
Fôret du Gavre (France)	0 (5)	6.4 (5)	0 (16)	25.7 (16)
Conteville (France)	47.6 (8)	2.3 (8)	0.4 (14)	1.3 (14)
Chaumont (France)	1.6 (6)	0.5 (6)	0 (7)	0.6 (7)
Lausanne/Neuchâtel (Swit- zerland)	3.0 (2)	0 (2)	2.0 (44)	0.6 (44)
Montese (Italy)	0 (5)	2 (5)	NA	NA
Visuč (Croatia)	0.1 (7)	0.6 (7)	1.3 (6)	3.0 (6)
Gostilje (Serbia)	0 (5)	2.4 (5)	0.4 (18)	0 (18)
Sokobanja (Serbia)	0 (5)	0.2 (5)	0 (21)	0.1 (21)
Chiflik (Bulgaria)	0.4 (7)	4.3 (7)	NA	NA

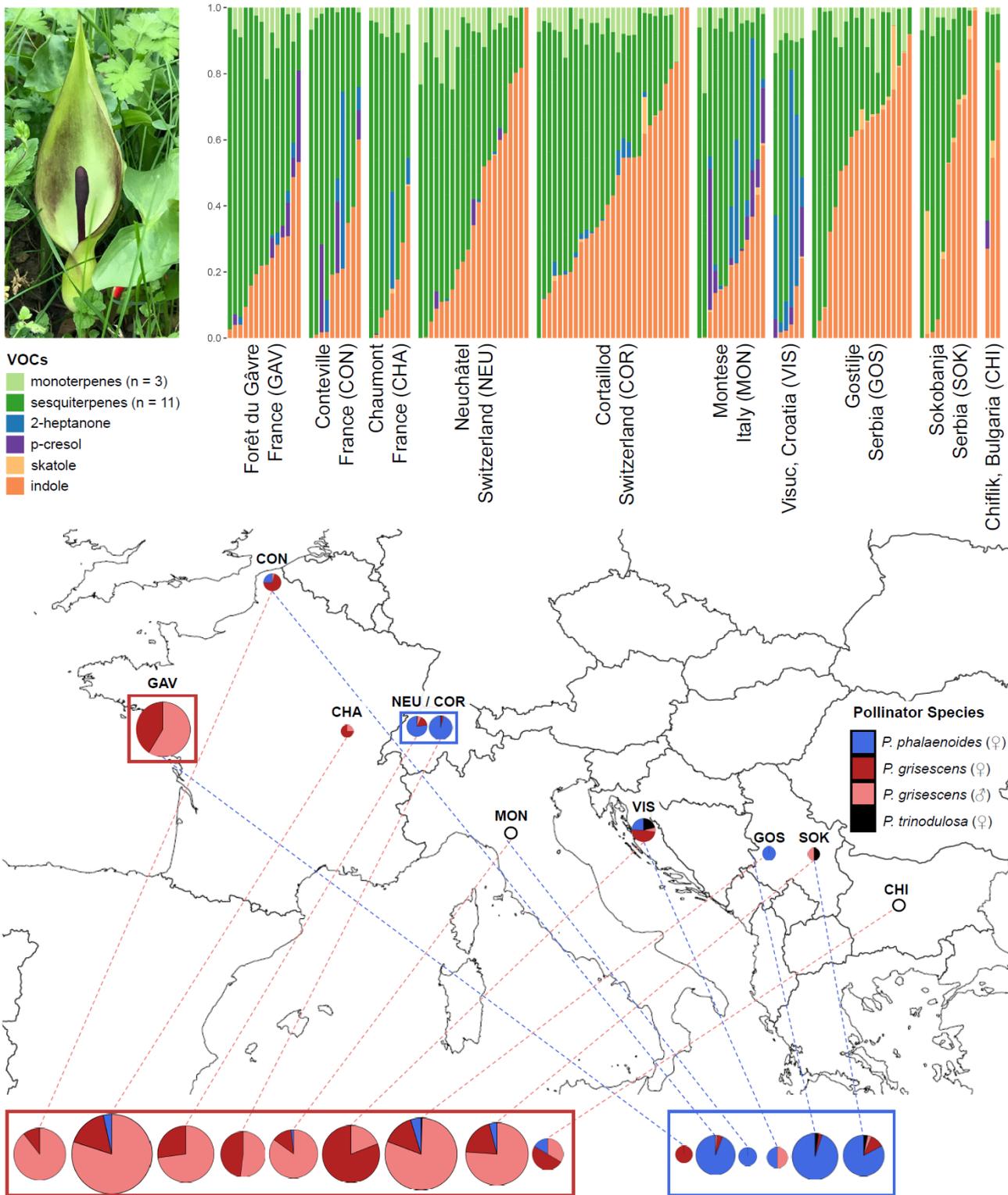
## FIGURE CAPTIONS

**Figure 1.** Europe-wide variation in floral volatile organic compounds (VOCs) and pollinator attraction of *Arum maculatum*. Stacked barplots present the relative quantities of VOCs in two terpene classes (mono- and sesquiterpenes) and four dung-mimicking VOCs emitted by individual inflorescences. Pie charts indicate the composition of Psychodidae trapped by *A. maculatum* inflorescences during field surveys (on map), and following transplants to two common garden sites (below map), scaled to represent mean quantities of Psychodidae per plant. Dotted lines link each field result with corresponding transplant results. *Note: empty chart = no Psychodidae attracted during our surveys.*

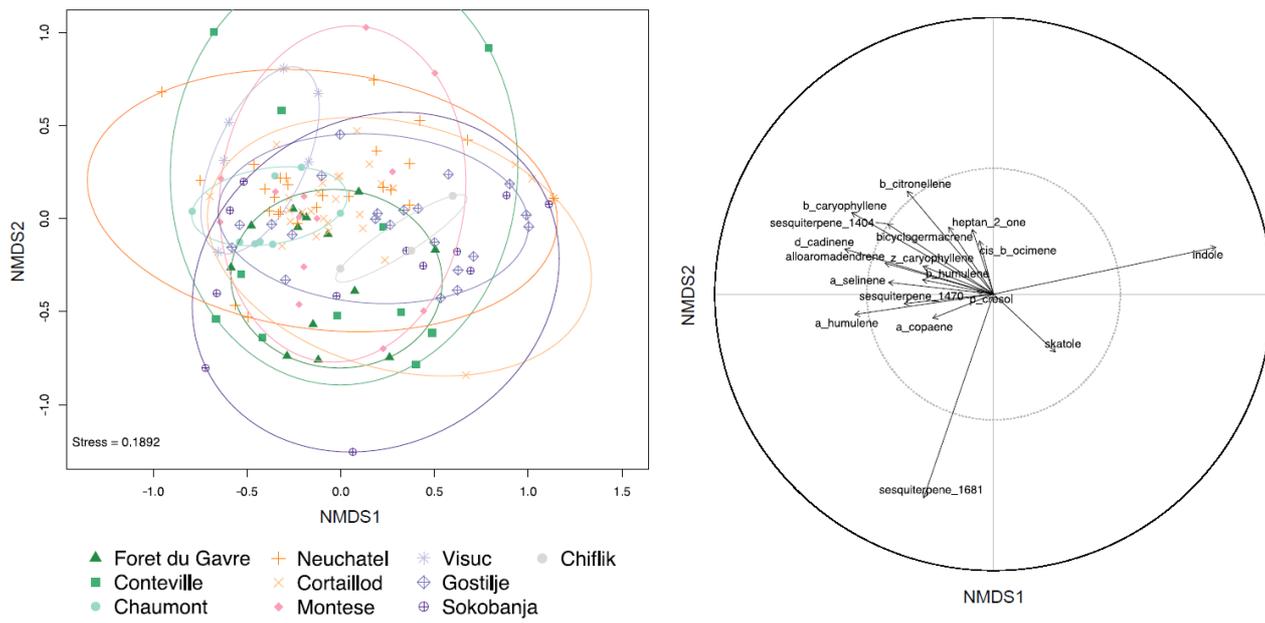
**Figure 2.** A) Multivariate representation of *Arum maculatum* floral volatile organic compound (VOC) emissions, using nonmetric multidimensional scaling (NMDS) of Bray-Curtis distances between individuals. Points and ellipses are coloured according to individuals' population of origin. B) Circle plot visualizing the correlations between all VOCs and the NMDS axes.

**Figure 3.** Interannual changes (2017/18 – 2019) in the average composition of Psychodidae pollinators trapped by six *Arum maculatum* populations. [NMDS; Bray-Curtis Distance; overall stress = 0.005]

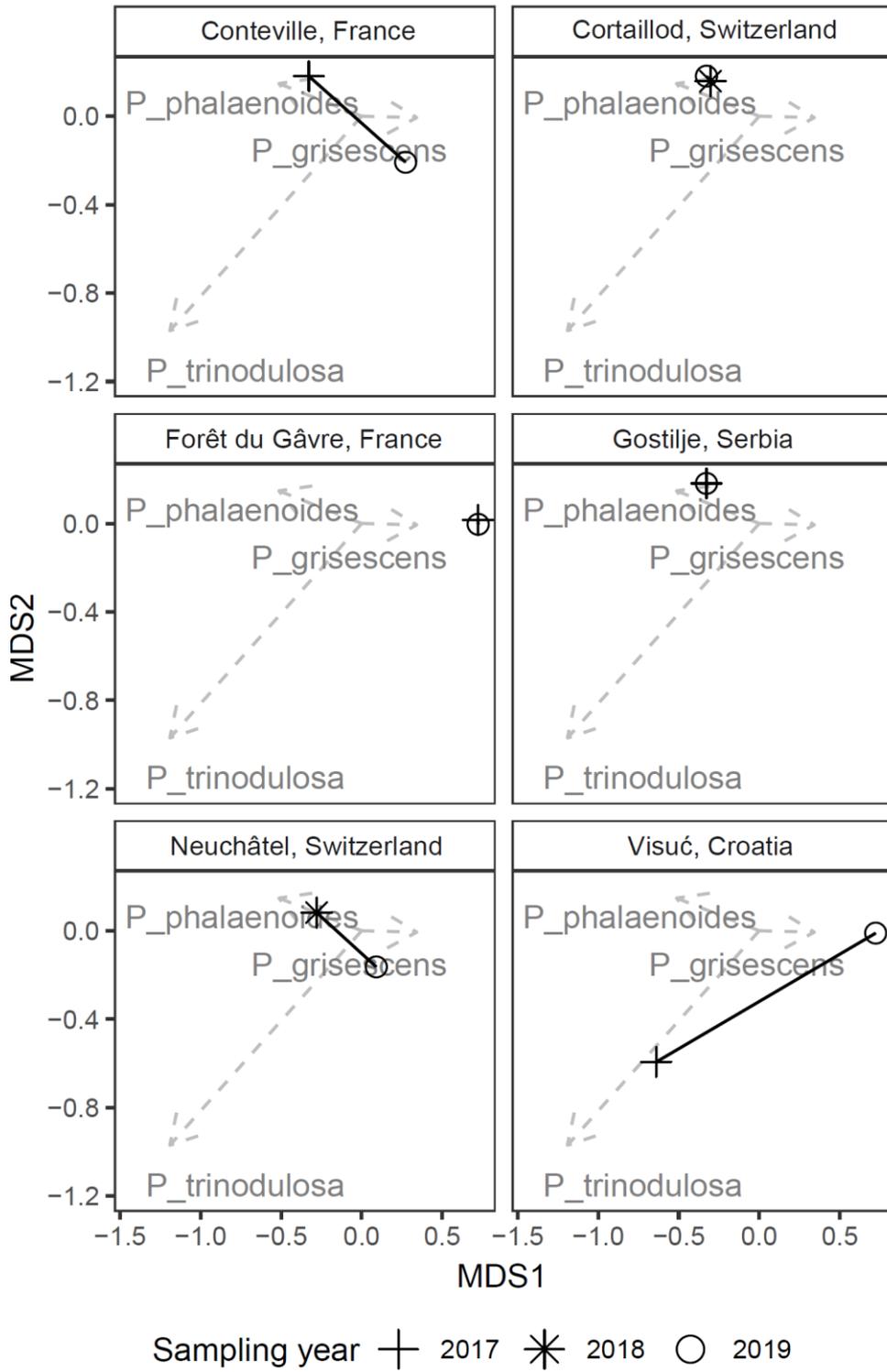
**Figure 4.** Pollinator attraction in the reciprocal transplant experiment. Plots show the mean (+/- SE) numbers of A) *Psychoda phalaenoides*, B) *Psycha grisescens*, and C) all insects trapped by inflorescences, transplanted from either *P. phalaenoides*-dominated populations (filled circles) or *P. grisescens*-dominated populations (hollow circles). No deme × habitat interactions (indicative of local adaptation) were identified.



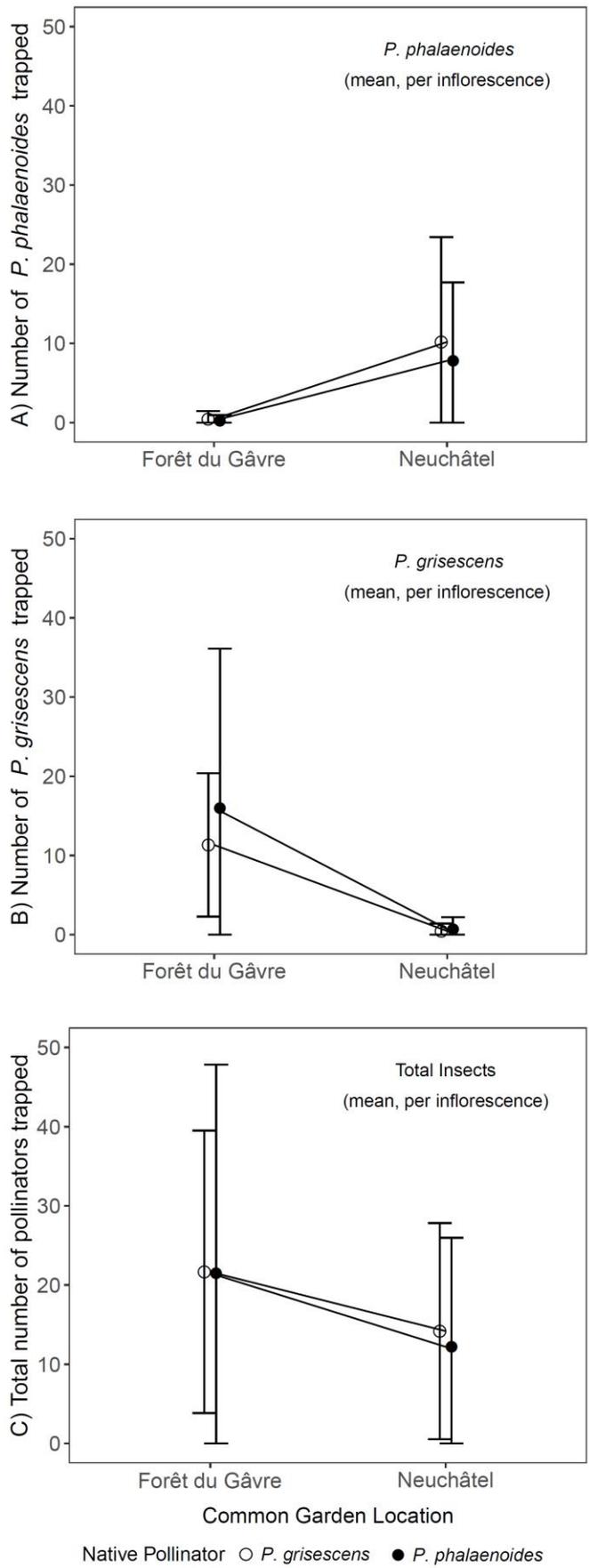
**Fig.1**



**Fig.2**



**Fig.3**



**Fig.4**