

1 **Order of arrival promotes coexistence via spatial niche preemption by the weak**
2 **competitor**

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17 **Running title:** Niche preemption facilitates coexistence

18

19 **Keywords:** Order of arrival, Priority effects, Modern coexistence theory, Spatial segregation,
20 *Tetranychus urticae* and *T. evansi*, Herbivores, Niche modification, Competitive ability

21

22 **Article type:** Letter

23

24 **Number of words in abstract:** 150

25

26 **Number of words main text:** 5006

27

28 **Number of references:** 66

29

30 **Number of figures:** 3

31

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36 **Authorship statement:** IF, OG and SM designed the experiment, IF, AM and RCP collected
37 data, IF and OG performed modelling work and analysed data. IF and SM wrote the first
38 draft of the manuscript, and all authors contributed substantially to revisions.

39

40 **Data accessibility statement:** Data and scripts for data analyses will be deposited in a public
41 repository upon acceptance.

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43

44 **Abstract**

45 Historical contingency, such as the order of species arrival, can modify competitive outcomes
46 via niche modification or preemption. However, how these mechanisms ultimately modify
47 stabilising niche and average fitness differences remains largely unknown. By experimentally
48 assembling two congeneric spider mite species feeding on tomato plants during two
49 generations, we show that order of arrival interacts with species' competitive ability to
50 determine competitive outcomes. Contrary to expectations, we did not observe that order of
51 arrival cause priority effects. In fact, coexistence was predicted when the inferior competitor
52 (*Tetranychus urticae*) arrived first. In that case, *T. urticae* colonized the preferred feeding
53 stratum (leaves) of *T. evansi* leading to spatial niche preemption, which equalized fitness but
54 also increased niche differences, driving community assembly to a close-to-neutrality scenario.
55 Our study demonstrates how the spatial context of competitive interactions interact with
56 species competitive ability to influence the effect of order of arrival on species coexistence.

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61 **Introduction**

62 Priority effects are broadly defined as the process by which historical contingencies in
63 community assembly (e.g. order and/or timing of arrival) change the outcome of interspecific
64 interactions (Chase 2003; Fukami 2015). Priority effects can be inhibitory if earlier arrival by
65 one species inhibits the growth of the species arriving subsequently (Chase 2003; Fukami 2015;
66 Ke & Letten 2018). Such effects are expected to result in alternative stable states and hamper
67 coexistence (Fukami 2015; Ke & Letten 2018). In turn, facilitative priority effects occur when
68 population growth is higher when individuals arrive after the settlement of a first species.
69 Facilitative effects are expected to foster coexistence (Fukami 2015), but they have been less
70 often identified in natural communities (Queijeiro-Bolaños *et al.* 2017; Clay *et al.* 2019;
71 Halliday *et al.* 2020).

72 Two major mechanisms are predicted to cause priority effects: niche preemption, in which
73 early colonizers reduce the amount of resource available to late colonizers, and niche
74 modification, in which the species arriving first modifies the environment, thereby inhibiting
75 or facilitating later colonization (Kardol *et al.* 2013; Vannette & Fukami 2014; Fukami 2015;
76 Delory *et al.* 2019, 2021; Grainger *et al.* 2019). For example, niche preemption in plant
77 communities was found to be strong in environments with high nutrient supply, as early
78 arriving plants grew quickly and prevented growth of later colonizers by depleting space and
79 light (Kardol *et al.* 2013). Also, previous colonization by different plant communities modified
80 the soil metabolome and inhibited population growth of forb, but not grass species, via
81 decreased root foraging (Delory *et al.* 2021). Although distinguishing among niche preemption
82 and modification is not always possible (Grainger *et al.* 2018; Boyle *et al.* 2021), recent
83 advances from coexistence theory can serve as a powerful approach to better understand the
84 importance of historical contingencies for species coexistence. Yet the combination of these
85 theoretical tools has seldom been applied in empirical settings.

86 Modern coexistence theory posits that the long-term persistence of competing species (i.e.,
87 species coexistence) can be attained by two non-mutually exclusive mechanisms: (i) equalizing
88 mechanisms that reduce average fitness differences, and therefore, dominance between species
89 and (ii) stabilising mechanisms, which tend to stabilise the interaction between competitors by
90 increasing the strength of intraspecific competition relative to interspecific competition
91 (Chesson 2000). Therefore, species will stably coexist if the stabilising niche differences,
92 leading to negative frequency dependence, are larger than differences in fitness between
93 competitors. Otherwise the species with higher fitness will eventually dominate the community
94 (Chesson 2000; Barabás *et al.* 2018; Spaak & De Laender 2021). Under this framework,
95 priority effects are strictly defined as the case in which the population dynamics of interacting
96 species are governed by positive frequency dependence (i.e., via negative niche differences),
97 which leads to the dominance of the early-arriving species (Ke & Letten 2018; Grainger *et al.*
98 2019; Spaak & De Laender 2021). Hence, species cannot coexist locally unless there is spatial
99 variability in the order of arrival. However, despite the fact that recent theory offers predictions
100 on the outcome of coexistence in systems with historical contingencies, empirical tests are
101 conspicuously lacking (but see Cardinaux *et al.* 2018; Grainger *et al.* 2019; Song *et al.* 2020).
102 Therefore, there is as yet scarce knowledge of which species traits interact with historical
103 contingencies to determine outcomes of interspecific interactions.

104 For the herbivore communities, such traits can be the spatial distribution of consumers
105 associated with resource use. Indeed, niche preemption may arise in this system, as herbivores
106 generally have preferred plant strata and the first arriving species may monopolize that resource
107 (Grainger *et al.* 2018; Godinho *et al.* 2020a). Moreover, herbivores often induce defences on
108 the plants they colonize, which is expected to entail niche modification for species arriving
109 later (Erb *et al.* 2011; Moreira *et al.* 2015; Stam *et al.* 2017). Additionally, some other herbivore
110 species are known to down-regulate plant defences, improving the performance of later

111 colonizers (Sarmiento *et al.* 2011a; Godinho *et al.* 2016), thereby potentially causing facilitative
112 priority effects. Overall, given the spatial heterogeneity of the environment that herbivores
113 experience (e.g., variation in leaf quality within and between plants), effects of the order of
114 arrival on species coexistence are expected to be prevalent in herbivore communities (Utsumi
115 *et al.* 2010; Erb *et al.* 2011; Moreira *et al.* 2015; Stam *et al.* 2017, 2018; Godinho *et al.* 2020a).
116 But what type of competitive outcome we should expect is unclear. Order of arrival has been
117 a traditional link to priority effects, yet the interaction of the chronology of community
118 assembly with the type of impact of species have on the environment (e.g. where they growth
119 and how they modify the habitat) can result in diverse outcomes, from competitive exclusion
120 to species coexistence. Applying the modern coexistence framework to this open question can
121 shed light on the proximate mechanisms that allow for species to coexist under varied historical
122 contingencies.

123 Here shed new light on the drivers of competitive outcomes by combining theoretical and
124 empirical tools to experimentally investigate the mechanisms through which order of arrival
125 affects species coexistence. We use as a model system, the two closely-related competing
126 herbivorous species, the spider mites *Tetranychus urticae* and *T. evansi* (Alzate *et al.* 2020).
127 *Tetranychus evansi* generally outcompetes *T. urticae* on tomato plants (Sarmiento *et al.* 2011b;
128 Orsucci *et al.* 2017; Alzate *et al.* 2020), although both species are also commonly observed to
129 co-occur on the same location (Ferragut *et al.* 2013). Niche modification is expected to be at
130 play in this system, because the two species interact with plant defences, albeit differently: *T.*
131 *urticae* induces tomato defences, whereas *T. evansi* suppresses them (Sarmiento *et al.* 2011a;
132 Alba *et al.* 2015). This asymmetrical niche modification is predicted to facilitate coexistence
133 by hampering growth of the stronger competitor and favouring growth of the inferior one, when
134 they arrive on plants colonized by the other species. Moreover, niche preemption may occur,
135 as both *T. evansi* and *T. urticae* prefer the upper, more nutritious leaves of tomato plants, where

136 their performance is higher (Godinho *et al.* 2020a). Thus, early-arriving species could occupy
137 the preferred niche and displace the other species to lower, less optimal, plant strata. We tested
138 this, by performing a series of multi-generational experiments where we varied order of arrival
139 and measured space use by the two competing species. We then applied modern coexistence
140 theory framework to unravel the conditions favouring coexistence or potentially leading to
141 priority effects.

142

143 **Material and Methods**

144 **Model system, species characteristics, and maintenance of experimental populations**

145 *Tetranychus urticae* is a generalist herbivore that feeds on many economically important crops
146 (Helle & Sabelis 1985; Grbić *et al.* 2011; Sousa *et al.* 2019), whereas *T. evansi* is a solanaceous
147 specialist that has recently invaded Europe from South America (Boubou *et al.* 2012). Both
148 species colonize tomato plants, although *T. urticae* may shift to other hosts if *T. evansi* is
149 present (Ferragut *et al.* 2013).

150 All experiments were performed with outbred populations of *T. urticae* and *T. evansi*
151 spider mites, formed via controlled crosses among four *T. evansi* and three *T. urticae*
152 populations collected in different locations in Portugal (Godinho *et al.* 2020b). Before the
153 experiment, spider mite populations were maintained in boxes containing leaves detached from
154 five-week-old tomato plants (*Solanum lycopersicum*, var MoneyMaker), with their petiole in
155 a small pot containing water, under controlled conditions (25 °C, 68% of humidity, photoperiod
156 of 16 hours light/8 hours dark). Twice a week, overexploited leaves were removed, and water
157 and new tomato leaves were added. Before infestation, tomato plants were kept in a climatic
158 chamber at 25°C, with a photoperiod of 16 hours light/8 hours dark with 75% humidity and
159 watered three times per week.

160 To ensure that females used in the experiments were of similar age, we created cohorts of
161 mated *T. urticae* and *T. evansi* females for each block. To this aim, females were placed during
162 48h in petri dishes (14.5 cm diameter) with a layer of wet cotton and two freshly cut tomato
163 leaves. One week later, another tomato leaf was added. Petri dishes were watered twice per
164 week. In the experiment, we used females with 13-15 days of age.

165

166 **Theoretical approach for predicting competitive outcomes: quantifying niche and fitness**
167 **differences**

168 Data collected in the experiments were used to parameterize a mathematical model from which
169 niche and average fitness differences can be quantified as well as the predictions of competitive
170 outcomes. We assume that the population dynamics in our experiment can be described by a
171 Beverton-Holt function (Hart *et al.* 2018):

172
$$(1) N_{i,t+1} = \frac{\lambda_i * N_{i,t}}{(1 + \alpha_{ii} * N_{i,t} + \alpha_{ij} * N_{j,t})}$$

173 Where $N_{i,t+1}$ is the number of individuals of species i in the next generation, λ_i the growth
174 rate of species i in absence of competitors, α_{ii} the intraspecific competitive ability, α_{ij} the
175 interspecific competitive ability and $N_{i,t}$, $N_{j,t}$ the number of individuals of species i and j in the
176 current generation, respectively. In this model, we assume that spider mites do not have a
177 dormant stage. This means that λ_i represents only the fraction of eggs that hatch and become
178 female individuals that grow and reproduce in the next generation.

179

180 From this model, niche overlap (ρ) is defined as follows (see details in Chesson 2012;
181 Godoy & Levine 2014).

182
$$(2) \rho = \sqrt{\frac{\alpha_{ij} \alpha_{ji}}{\alpha_{jj} \alpha_{ii}}}$$

183 This formula reflects the average degree to which species limit individuals of their own
 184 species relative to heterospecific competitors. If species limit individuals of their own species
 185 more strongly than competitors (α_{jj} , α_{ii} , are much greater than α_{ij} , α_{ji}), then niche overlap
 186 will be low, favouring coexistence. Alternatively, if species limit individuals of their own
 187 species and those of their competitor equally, niche overlap equals one, hampering stable
 188 coexistence. With ρ defining niche overlap between a pair of species, stabilising niche
 189 differences are expressed as $1-\rho$.

190 Average fitness differences ($\frac{\kappa_j}{\kappa_i}$) (Chesson 2012; Godoy & Levine 2014) are defined as:

$$191 \quad (3) \quad \frac{\kappa_j}{\kappa_i} = \frac{\lambda_j - 1}{\lambda_i - 1} * \frac{\sqrt{\alpha_{ij} * \alpha_{ii}}}{\sqrt{\alpha_{ji} * \alpha_{jj}}}$$

192 The greater the ratio, ($\frac{\kappa_j}{\kappa_i}$), the greater the fitness advantage of species j over i . If this ratio
 193 is one, species are equivalent competitors. With niche overlap and average fitness defined in
 194 eqn. 2 and 3 respectively, we can determine the coexistence condition. Specifically,
 195 coexistence requires both species to invade when rare (Chesson 2012), which is satisfied when
 196 (Godoy & Levine 2014):

$$197 \quad (4) \quad \rho < \frac{\kappa_j}{\kappa_i} < \frac{1}{\rho}$$

198 Stable coexistence is possible whenever species have either large niche differences
 199 (corresponding to small niche overlap) that overcome large average fitness differences, or at
 200 the other extreme, via an a close-to-neutral scenario (Scheffer *et al.* 2018), where, even with
 201 weak niche differences, small fitness differences stabilise the interaction between competitors.
 202 If competitors are not predicted to coexist, we can pinpoint if this is due to competitive
 203 exclusion (when fitness differences are larger than niche differences) or due to priority effects,
 204 leading to alternative states when niche differences are negative (Fukami & Nakajima 2011;
 205

206 Ke & Letten 2018). We used data from the fully parameterized models (see below) to generate
207 these predictions.

208

209 **Experiments**

210 We performed a series of experiments in which we either manipulated the order of arrival and
211 relative frequency (i.e., relative initial abundance with a constant density of 20 individuals), or
212 the initial density of each of the two species.

213 In the first experiment, both species were introduced simultaneously using the following
214 proportions of *T. evansi* / *T. urticae*: 1:19; 10:10 and 19:1, along with the single-species
215 controls (20:0 and 0:20). To manipulate the order of arrival, we introduced (i) 10 *T. evansi*
216 females 48h before 10 *T. urticae* females and vice versa and (ii) 19 *T. evansi* females 48h
217 before 1 *T. urticae* female and vice versa. The experiment was done in two blocks, one week
218 apart. Each block contained five boxes of each experimental treatment (nine treatments, with
219 ten boxes per treatment), each with a pot filled with water and two freshly cut tomato leaves
220 from five-week-old tomato plants. Leaf pairs consisted of leaves 2 and 4 or 3 and 5 (leaf
221 number indicates leaf age: leaves 2 and 5 correspond to the oldest and youngest leaves,
222 respectively). This was done to ensure that each box contained a younger and an older leaf,
223 since both species have a preference for younger (upper) leaves (Godinho *et al.* 2020a). Adult
224 females were distributed by the two leaves, following the treatments described above. After
225 one generation (circa 14 days), two more leaves were added to ensure enough resources for the
226 second mite generation. Boxes that initially received the leaf pair 2-4, received leaves 3-5 and
227 vice versa. After two generations, we counted the number of adult females of each species per
228 leaflet and leaf.

229 In the second experiment we also estimated the growth rate of each species by counting
230 the number of adult females obtained from the progeny of a single *T. urticae* or *T. evansi* female

231 ovipositing for 48h in two overlapping 18mm leaf disks, replicated 18 times. These disks were
232 placed in square petri dishes with a layer of wet cotton and were watered every two days. The
233 number of adult females produced was assessed after one generation.

234

235 **Data Analyses**

236 Effect of order of arrival and initial frequency on species abundance

237 To test the impact of order of arrival and frequency and their interaction on the proportion of
238 adult females of each species after two generations, we performed the following general linear
239 mixed model (lme4 package, Bates et al. 2015), using the binomial family:

$$240 \quad (5) Y = \text{Frequency} + \text{Order} + \text{Frequency} \times \text{Order} + \text{Block} + \varepsilon$$

241 Where Y corresponds to the combination of two vectors with the number of *T. evansi* and
242 *T. urticae* females after two generations, Frequency (fixed factor) to the initial ratio between
243 the two species, Order (fixed factor) to the order of arrival, Block (random factor) to whether
244 the experiment was performed on week one or two, and ε to the residual error. Additionally,
245 we ran the same model as above, but merging Frequency, Order and their interaction in a single
246 factor (Treatment, 7 levels). This allowed performing *a priori* contrasts to compare between
247 different orders of arrival and frequencies for each species, since our experimental design was
248 not orthogonal. To compare the effect of different orders of arrival, we performed contrasts
249 between the treatments with same initial frequency but different orders of arrival. To compare
250 the effect of frequency, we performed contrasts between treatments with same order of arrival
251 but different initial frequencies. Contrasts were done using testInteractions fromphia package
252 (Rosario-Martinez 2015) and were then corrected for multiple comparisons using FDR
253 correction (Benjamini & Yekutieli 2001).

254

255 Effects of order of arrival on coexistence

256 We used a maximum likelihood approach to estimate the growth rate in absence of competitors
257 (λ) and the intra and interspecific competitive interactions (the α 's) for each species. To ensure
258 model convergence, we took a nested perspective in which the results of the first model were
259 used as priors for the subsequent models which increased in complexity (Matías et al. 2018).
260 That is, estimates obtained from model 6A were used as priors for model 6B, and those
261 obtained from model 6B were used as priors for model 6C.

262
$$6A) N_{t+1} = \lambda * N_t$$

263
$$6B) N_{i,t+1} = \frac{\lambda * N_t}{(1 + \alpha * N_t)}$$

264
$$6C) N_{i,t+1} = \frac{\lambda_i * N_{i,t}}{(1 + \alpha_{ii} * N_{i,t} + \alpha_{ij} * N_{j,t})}$$

265 The initial model (6A) considers only the intrinsic growth rate in the absence of
266 interactions (λ). This model was parameterized using estimates from the experiment with
267 single *T. urticae* or *T. evansi* female. Model 6B adds an α parameter, which accounts for the
268 overall effect of competition, and finally, model 6C separates this overall competitive effect
269 into intra and interspecific components.

270

271 *Effect of order or arrival and initial frequency on leaf occupancy and aggregation*

272 To test if coexistence vs. exclusion outcomes could be explained by niche preemption due to
273 changes in leaf occupancies, we compared occupancy patterns of each species across the four
274 leaves. For the single species treatment, we tested if the number of females differed across
275 leaves (model 7). For the double species treatment, we tested if the order of arrival or initial
276 frequency, or their interaction changed mite distribution across leaves (model 8). For that, we
277 compared the number of females in each leaf to the distribution of the single species treatment.

278 We applied the following binomial models, with Leaf and/or Treatment and their
279 interaction as fixed factors, for the control (model 7) and experimental (model 8) treatments:

280
$$(7) Y = \text{Leaf} + \varepsilon$$

281
$$(8) Y = \text{Leaf} + \text{Treatment} + \text{Leaf} \times \text{Treatment} + \varepsilon$$

282 where Y corresponds to the combination of two vectors with the number of *T. evansi* (or
283 *T. urticae*) females on each leaf per box and the total number of individuals on each box that
284 were not on that leaf. For the double treatment, a posteriori contrasts were done between each
285 treatment and the corresponding single species treatment. The initial fitting with Block as a
286 random factor, indicated no variance in this factor, thus we fitted only fixed factors.

287 Since *T. evansi* suppresses defences locally, it is expected that *T. urticae* aggregates with
288 it (Sato *et al.* 2016). To test if aggregation changed with order of arrival or initial frequency,
289 we calculated the Checkerboard score (C-score) (Gotelli & Rhode 2002) per replicate. The C-
290 score quantifies species co-occurrence, measuring the extent to which they segregate or
291 aggregate across environments (Gotelli & Rhode 2002). The bipartite package (Dormann *et al.*
292 2008) normalizes the C-score between 0 (no aggregation) and 1 (aggregation), allowing
293 comparisons between treatments. To calculate the C-score per leaf, we created a presence-
294 absence matrix per leaflet and leaf for each box. We then applied the following general linear
295 mixed model to test for differences in aggregation between treatments:

296
$$(9) Y = \text{Treatment} + \text{Block} + \varepsilon$$

297 Where Y is the computed C-score and Treatment is a fixed factor. Contrasts were
298 performed between initial frequency and order of arrival, applying FDR correction for multiple
299 comparisons, as described above.

300 All analyses were done using R (R Core Team 2021) using package “cyr” (García-Callejas
301 *et al.* 2020), and plots were done using ggplot2 (Wickham 2016) and cowplot (Wilke 2020)
302 packages. Data and scripts are available in the github repository:
303 https://github.com/irfragata/priority_effects.

304

305 **Results**

306 Effect of order of arrival and initial frequency on species abundance

307 The number of individuals of each species on tomato plants were affected by the order of arrival
308 ($\chi^2 = 298.93$, $df = 2$, $p\text{-value} < 0.0001$), their initial frequency ($\chi^2 = 568.12$, $df = 2$, $p\text{-value} <$
309 0.0001), and the interaction among these factors ($\chi^2 = 188.61$, $df = 2$, $p\text{-value} < 0.0001$).
310 Specifically, the abundance of *T. evansi* females after two generations was higher when this
311 species arrived first or simultaneously with *T. urticae*, independently of initial frequencies.
312 However, the additional advantage provided by arriving first was much larger in the equal
313 frequency treatment, as contrasts comparing order of arrival were highly significant for equal
314 frequencies (10:10), but not for the high *T. evansi* frequency (19:1) (Table S1, Fig.1). The
315 abundance of *T. urticae* after two generations was also affected by initial frequency and order
316 of arrival. Indeed, the final number of *T. urticae* females was higher when this species arrived
317 first and was at high initial frequency, than in the equal frequency treatment (Table S1, Fig. 1).
318 Overall, these results confirm that *T. evansi* is a superior competitor as expected from previous
319 field and laboratory observations (Sarmiento *et al.* 2011b; Ferragut *et al.* 2013; Alzate *et al.*
320 2020).

321

322 Effect of order of arrival on coexistence

323 The order of arrival modified the outcome of competition between the two species.
324 *Tetranychus evansi* (the superior competitor) excluded *T. urticae* when it arrived first or at the
325 same time. Under this exclusion scenario, the rate of competitive exclusion is expected to be
326 faster when *T. evansi* arrived first due to an increase in fitness differences and due to a decrease
327 in niche differences (Fig 2). Coexistence was only possible when *T. urticae* arrived first (Fig.
328 2). This outcome was due to small fitness and niche differences among competitors, leading to
329 a quasi-neutral scenario. Specifically, when *T. urticae* arrived first, we observe almost similar

330 strengths of intra- and interspecific interactions among species (Fig S1A), combined with an
331 increase in the intrinsic growth rate of *T. urticae* (in comparison to other orders of arrival, Fig
332 S1B). Contrary to expectations and previous studies, order of arrival was not associated with
333 priority effects.

334

335 Effect of order of arrival and initial frequency on leaf occupancy and aggregation

336 To explore whether changes in coexistence outcomes were driven by a shift in leaf occupancy,
337 we tested how initial frequency and order of arrival affected the proportion of females of the
338 two species on each leaf (Fig 3 A, C, Fig S2) vs. the occupancy in the single species treatments
339 (Fig 3B, D). When *T. evansi* was alone, it reached a consistently higher abundances on leaves
340 3 and 4 (Table S2A, Fig 3B), whereas *T. urticae* was less abundant on leaf 2 (the oldest leaf)
341 in comparison to all others (Table S2A, Fig 3D). In treatments where *T. urticae* arrived first,
342 significantly fewer *T. evansi* females were found on leaf 4 (Fig 3, Fig S2A, Table S2B). This
343 pattern was observed for leaf 3 when *T. urticae* started with higher frequency and both species
344 arrived at the same time. When *T. evansi* arrived first or started at higher frequency, we
345 observed fewer changes on its own leaf occupancy (Fig S2A). The distribution of *T. urticae*
346 showed a slight shift when it arrived first, with a reduction on the prevalence of leaves 2 and 5
347 and slightly higher occupation of leaves 3 and 4 (Fig. S2B, Table S2B). When *T. evansi* started
348 at high frequency, there was also a shift in *T. urticae* distribution, with a lower occupancy of
349 leaves 2 and 5 (Fig S2B).

350 Spatial aggregation significantly differed among treatments ($\chi^2 = 18.186$, $df = 6$, p -value
351 = 0.01279), being higher in treatments with similar initial densities (cf. Fig S3 with Fig 1, Table
352 S3). We observed a significant difference in C-score (the degree of spatial aggregation) with
353 higher aggregation when both species arrived at the same time and had equal frequency, and a

354 lower aggregation when both species arrived at the same time and *T. evansi* started at higher
355 frequency (Table S3). Order of arrival did not change the C-score (Fig. S3, Table S3).

356

357 **Discussion**

358 This study shows that order of arrival interacts with the competitive ability to determine the
359 probability of coexistence between congeneric species that share common resources such as
360 food and/or space. When both species arrive at the same time or the superior competitor
361 (*Tetranychus evansi*) arrived first, *T. urticae* was predicted to be excluded. Coexistence was
362 only predicted when the inferior competitor (*T. urticae*) was the first species colonizing the
363 habitat. Analyses of leaf occupancy show that these competition outcomes are linked to a
364 spatial niche preemption process in which *T. evansi* was displaced from its preferred food
365 stratum when *T. urticae* arrived first. As a result of this complex interaction between order of
366 arrival, species competitive ability, and spatial occupancy, we observed a particular
367 configuration that allows species coexistence: both species increased niche differences and
368 reduced fitness differences to the extent that they can coexist despite small niche differences.
369 These multiple lines of evidence challenge current paradigms on the direct connection between
370 order of arrival and priority effects.

371 We found that *T. evansi* had higher competitive ability and growth rate, and often excluded
372 *T. urticae* (Fig 1, 2). This is in line with laboratory observations showing that *T. evansi*
373 outcompeted *T. urticae* on tomato plants (Sarmiento *et al.* 2011b; Alzate *et al.* 2020, but see
374 Orsucci *et al.* 2017) and with field observations showing a reduction in the prevalence and a
375 shift in host use in *T. urticae* upon invasion by *T. evansi* (Ferragut *et al.* (2013). Still, these two
376 species can co-occur in the field in the same plant species (Ferragut *et al.* 2013; Orsucci *et al.*
377 2017; Zélé *et al.* 2018). The advantage created by the earlier arrival of *T. urticae*, and associated
378 reduction in interspecific competition by *T. evansi*, could be one of the possible mechanisms

379 fostering coexistence of the two species in nature. Indeed, *T. urticae* can withstand colder
380 temperatures than *T. evansi* (Gotoh *et al.* 2010; Khodayari *et al.* 2013; Riahi *et al.* 2013; White
381 *et al.* 2018), hence it is expected to arrive first in the season. Field surveys that sample both
382 species in the same location across seasons are needed to further explore this hypothesis.

383 Historical contingencies emerging from order of arrival can happen through two main
384 mechanisms: niche modification or niche preemption (Fukami 2015). In our system, niche
385 modification may arise via interactions between spider mites and plant defences. As *T. evansi*
386 suppresses plant defences, *T. urticae* could benefit from an early colonization from its
387 competitor (Alba *et al.* 2015; de Oliveira *et al.* 2016, 2017; Godinho *et al.* 2020a). However,
388 we observe competitive exclusion when *T. evansi* arrives first, suggesting that if *T. urticae*
389 gained this benefit, it was not enough to outcompete *T. evansi*. In turn, niche preemption can
390 occur through monopolization of nutrients or space, which can be particularly important in
391 intraguild competitive interactions (Grainger *et al.* 2018; Holditch & Smith 2020). In this study,
392 resource depletion could not explain the differences we observe in competitive outcomes, since
393 order of arrival did not have a large impact on the growth rate of both species. However, we
394 observed a shift in the leaf occupancy pattern of *T. evansi* females when *T. urticae* arrived first.
395 This displacement of *T. evansi* from the preferred food stratum (i.e., younger, more nutritious
396 leaves) by early-arriving *T. urticae* can explain the decreased competitive ability of the superior
397 competitor. Thus, our results indicate that variation in species performance driven by habitat
398 quality heterogeneity (Orians *et al.* 2000; Orians & Jones 2001) combines with order of arrival
399 to generate niche preemption, providing a mechanism for the two herbivores to coexist.

400 Order of arrival is a major determinant of community assembly across diverse taxa, from
401 microbes to plants (Chase 2003; Erb *et al.* 2011; Kardol *et al.* 2013; Stam *et al.* 2017; Grainger
402 *et al.* 2018, 2019; Clay *et al.* 2019, 2020; Halliday *et al.* 2020). Most of these studies show that
403 early colonizers inhibit growth and decrease performance of late arriving species, especially in

404 those that occupy very similar niches (Fargione *et al.* 2003; Vannette & Fukami 2014; Delory
405 *et al.* 2019, 2021; Grainger *et al.* 2019), although very few concern herbivorous species
406 competing for the same niche (e.g. Grainger *et al.* 2018; Holditch & Smith 2020). Nevertheless,
407 other studies show that order of arrival does not always affect community assembly (e.g.
408 Delory *et al.* 2021) or that initial colonizers may facilitate later colonization of other species
409 (e.g. Queijeiro-Bolaños *et al.* 2017; Delory *et al.* 2019). Here, we show that coexistence is
410 promoted by early colonization by the inferior competitor species, which reduces the
411 competitive ability of the superior competitor and increases the fitness of the inferior
412 competitor. Through fully parameterized models from modern coexistence theory, we found
413 that niche preemption increases niche differences and strongly reduces fitness differences,
414 leading to an equalising effect that allows species to coexist. Our study adds a novel perspective
415 to the growing body of evidence that historical contingencies shape ecological communities,
416 by showing that the probability of coexistence of two competing herbivores changes due to an
417 interaction between order of arrival and species competitive ability.

418 Priority effects were recently incorporated into modern coexistence theory (Ke & Letten
419 2018; Spaak & De Laender 2021), but to our knowledge, this is one of the very few studies
420 that empirically tested the impact of changes in order of arrival on species coexistence so far,
421 using this theoretical framework. Grainger *et al.* (2019) observed that positive frequency
422 dependence, due to strong priority effects, arose from changes in order or arrival in yeast
423 species feeding on floral nectars. In contrast, our results show that order of arrival did not lead
424 to priority effects caused by positive frequency dependent alternative states. Instead, when *T.*
425 *evansi* arrived first, it excluded *T. urticae* due to a large increase in fitness differences and a
426 reduction in stabilising niche differences. However, when *T. urticae*, arrived first, coexistence
427 was predicted, via an increase in niche differences and a decrease in fitness differences, making
428 the competition between the two species more neutral. These results suggests that, in the event

429 of a small environmental perturbation that produces changes in niche or fitness differences, the
430 two studied species might not be able to coexist, compared to a scenario in which species
431 coexist via strong niche differences. Framing priority effects in the modern coexistence theory
432 (Ke & Letten 2018) is undoubtedly an important step to mechanistically understand how order
433 of arrival affects community assembly processes. However, here we show that order of arrival
434 does not always affect competitive outcomes via priority effects; instead, it can lead to
435 coexistence via niche preemption by the inferior competitor. Thus, our results show that order
436 of arrival can produce a wide range of competitive outcomes from coexistence to competitive
437 exclusion due to positive and negative frequency dependence. Therefore, it is urgent that
438 ecologists widen the scope of the multiple outcomes that historical contingency can produce
439 on species coexistence.

440 Most empirical and theoretical studies emphasize the inhibitory nature of niche preemption
441 (Fargione *et al.* 2003; Fukami 2015; Vieira *et al.* 2018; Delory *et al.* 2019), with the early
442 arriving species outcompeting the other. However, recent theory suggests that, in a resource
443 competition model of two species, niche preemption by the inferior competitor could facilitate
444 coexistence under a trade-off between order of arrival and the resource levels of zero net growth
445 (R^*) (Qi *et al.* 2021). Our study is, to the best of our knowledge, the first empirical study
446 showing that niche preemption by the weaker competitor facilitates coexistence. This striking
447 change in the outcome of competitive interactions emerge mostly due to a decrease in fitness
448 differences coupled with the decrease in niche overlap. This suggests that even small
449 differences in order of arrival can be sufficient for the monopolization of a resources in plant-
450 herbivore interactions, which may suffice to allow coexistence between competitor species.
451 Therefore, our results demonstrate how small temporal differences percolate into small spatial
452 heterogeneities, fostering coexistence and the maintenance of diversity.

453

454

455 **Acknowledgments:** This work was financed by an ERC (European Research Council)
456 consolidator grant COMPCON, GA 725419 attributed to SM and by FCT (Fundação para
457 Ciência e Tecnologia) with the Junior researcher contract (CEECIND/02616/2018) attributed
458 to IF. RC-P is supported by grant #2020/11953-2 São Paulo Research Foundation (FAPESP)
459 and grant R-2011-37572 Instituto Serrapilheira. OG acknowledges financial support provided
460 by the Spanish Ministry of Economy and Competitiveness (MINECO) and by the European
461 Social Fund through the Ramón y Cajal Program (RYC-2017-23666). AM was funded by
462 National Science Centre, Poland (grant no. 2018/28/T/NZ8/00060) and Excellence Initiative -
463 Research University programme (support for the internationalization of the Adam Mickiewicz
464 University PhD students, no. 003/13/UAM/0018).

465

466 **Competing interests:** Authors declare no competing interests.

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636 Figure 1 – Proportion of spider mites *Tetranychus evansi* females (y-axis) depending on initial
637 frequency (number of initial females *T. evansi*: *T. urticae*, x-axis) and order of arrival (same
638 time vs. *T. evansi* or *T. urticae* arriving 48h before its competitor) after two generations.
639 *Tetranychus evansi* is the better competitor overall (ratio above 0.5), unless *T. urticae* arrives
640 first or is at higher initial frequency. A posteriori contrasts show a strong effect of order of
641 arrival in the proportion of females of the two species (Suppl. Table 1B). Initial frequency also
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643 than *T. evansi* (Suppl. Table 1B). Boxplots represent median and quartiles of the 10 boxes
644 within treatment.

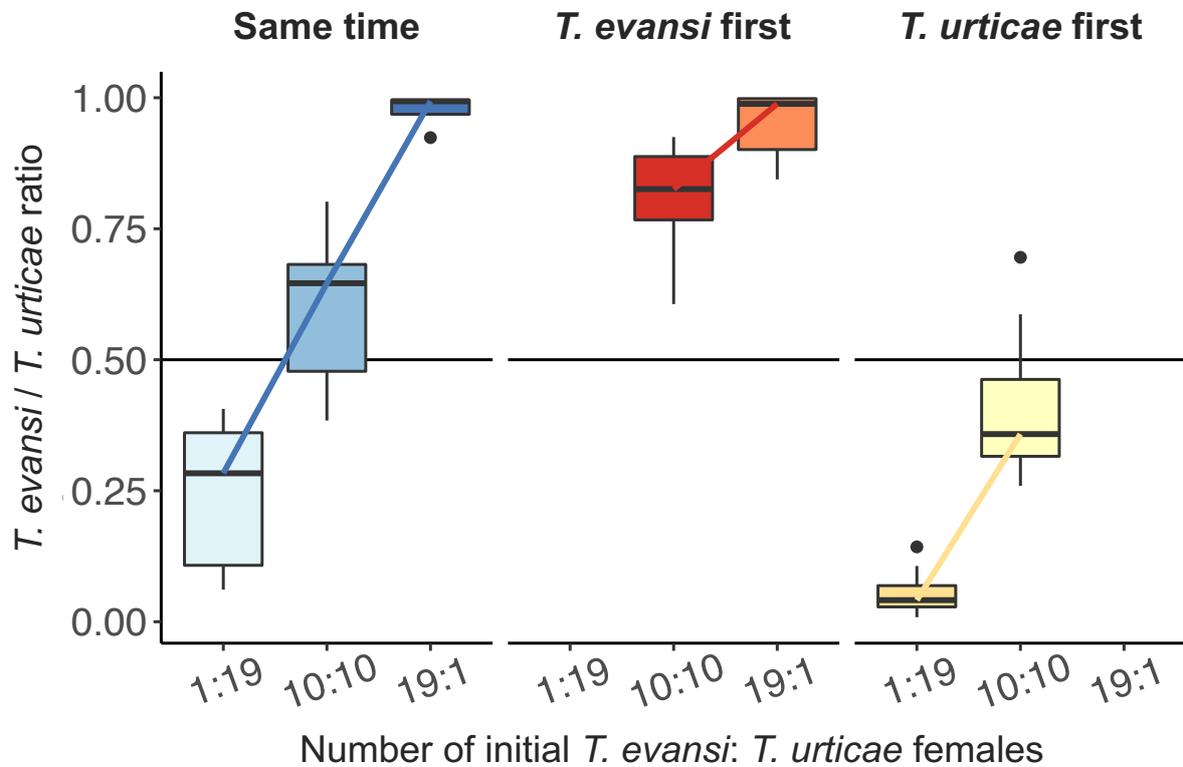
645
646 Figure 2 – Relationship between average fitness differences ($\frac{\kappa_j}{\kappa_i}$, y-axis) and stabilising niche
647 differences ($1 - \rho$, x-axis) for different orders of arrival (*Tetranychus evansi* first – red, same
648 time – blue, *T. urticae* first – yellow). Plotting average fitness differences against niche
649 differences allows mapping different competitive outcomes predicted by modern coexistence
650 theory (Chesson 2000; Ke & Letten 2018; Spaak & De Laender 2021). The coexistence
651 condition (eq. 4) and its inverse, represented by the two solid black lines, allow defining the
652 space in which species can coexist due to negative frequency dependence or enter alternative
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654 smaller than zero respectively. Otherwise, the species with higher fitness will exclude the other.
655 In our case, the only scenario in which species are predicted to coexist is when *T. urticae* arrives
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657 maximum likelihood estimates. For the other two cases, it is predicted that the superior
658 competitor *T. evansi* will exclude *T. urticae*.

659

660 Figure 3 – Differences between expected and observed leaf occupancy for *Tetranychus evansi*
661 (A) and *T. urticae* (C) for a subset of the experimental treatments (when *T. urticae* arrived first
662 or at the same time as *T. evansi*, note that Figure S2 includes all treatments); leaf occupancy
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664 to the oldest leaf and leaf 5 to the youngest. For each box, we calculated the ratio of females
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666 treatments we calculated the difference between this ratio and the average ratio for the control
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669 we see that *T. evansi* reduces occupancy on leaf 4 when *T. urticae* arrives first and on leaf 3
670 when the two species arrive at the same time. In contrasts, *T. urticae* shows a slight increase in
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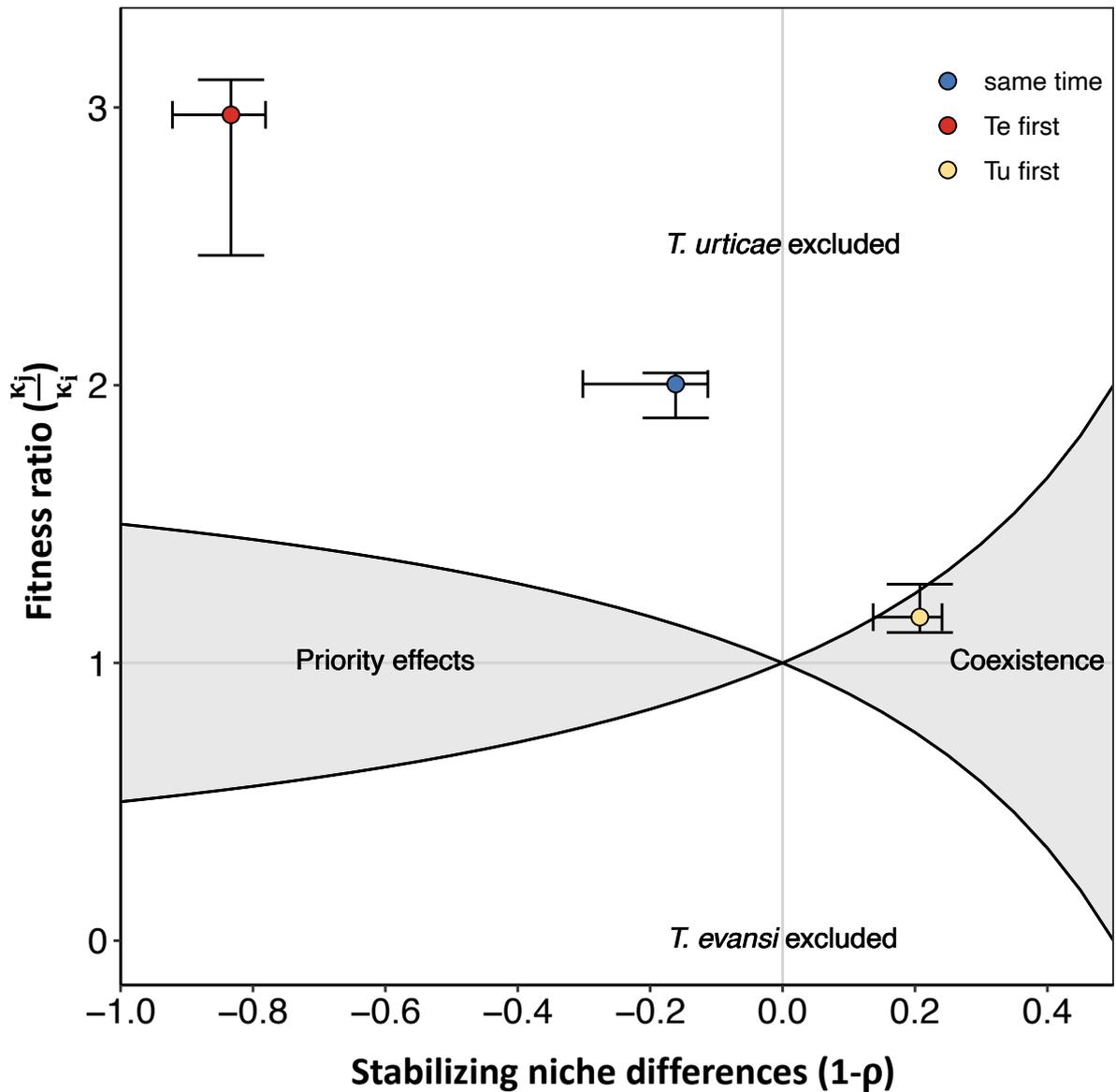
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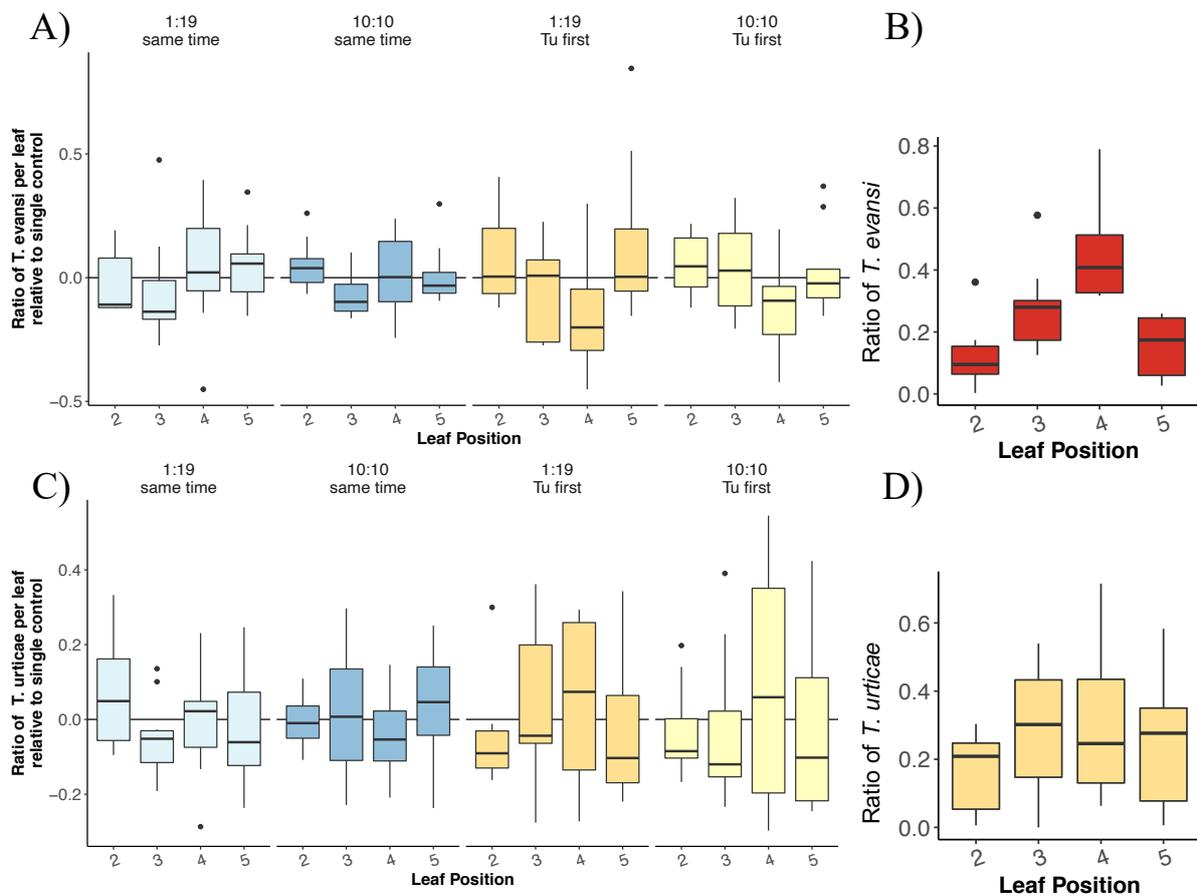
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