1 **Performance variation of common ragweed (*Ambrosia artemisiifolia* L.) across invasion levels in Western**

2 **Europe**

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15

16   **Abstract**

17   The occurrence of an invasive plant across a continent is generally not homogeneous; typically, some areas are

18   highly invaded whereas others show moderate or low invasion levels. This situation might be a snapshot of an

19   ongoing spread, but it could also remain stable under the pressure of factors that constrain the invasion. Among

20   those factors, plant performance variation among invasion levels can explain an invasion slowdown. However,

21   few studies have investigated the large-scale variation of invasive plant performance in the field. *Ambrosia*

22   *artemisiifolia* L. in Western Europe represents a good opportunity to address this issue, with areas of high,

23   moderate and low invasion levels being documented across a ca. 1000 km transect. In this study, we compared in

24   situ plant performance-related traits in 12 populations from areas of contrasting invasion levels. We also tested

25   whether performance-related traits were influenced by the intra-and inter-specific competition, by the local

26   climatic conditions or by latitude (a proxy for growing season length). Overall, we did not find differences in

27   performance-related traits across invasion levels, and intra-and inter-specific competition had low effects on

28   plant performance. This study highlights the fact that *A. artemisiifolia* individuals express similar performance

29   across invasion levels, even beyond what can be considered the present invasion front. Further research has to

30   expand this study northwards, and assess other factors that could constrain the invasion in order to highlight if

31   the species invasion northward is constrained or if it has the potential to invade new areas.

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1 Introduction

The occurrence of an invasive plant in a range where it has been introduced can vary greatly (Hengeveld and Haeck, 1982; Lawton, 1993), and this is true at the landscape (Bradley and Mustard, 2006), regional (Guillerm et al., 1990; Müller, 2004) and continent (Welk, 2004) levels. The reasons for the variations in patterns can be multiple, including the dynamic of introduction and the invasion pathways; the human actions to control the spread; and the spatial variation in environmental conditions. Typically, at the continent level, such variations are common, with areas of i) high invasion levels characterized by high population occurrences (Orivel et al., 2009) and often situated in the relative vicinity of the initial introductions (see e.g. Allard, 1943); ii) moderate invasion levels, such as at invasion fronts and/or in areas of recent colonization (Eckert et al., 2008; Lawton, 1993); and iii) low invasion levels, where populations are scarce and often considered casual or recently naturalized. This situation might present a snapshot of an ongoing process of invasion, i.e., an inexorable colonization of the whole continent. In this case, the pattern of population dispersion across the continent would mainly be explained by the dynamic of introduction and the invasion pathways (Guillerm et al., 1990; Lachmuth et al., 2010; Monty and Mahy, 2010). However, this situation could persist over time if it can be explained by environmental conditions and/or demographic processes that constrain invasion (Arim et al., 2006). This latter case corresponds to what is generally observed in native plant species that have reached their distribution equilibrium (Villellas et al., 2013), which is governed by different ecological and evolutionary processes (reviewed in Sexton et al., 2009). In the case of invasive species, the invaded range expansion could be limited by genetic constrains that impediment local adaptation, dispersal limitations to suitable sites, or a too harsh environment to allow survival beyond the species range (Alexander and Edwards, 2010; Arim et al., 2006; Hargreaves et al., 2014; Sexton et al., 2009; Thouvenot et al., 2013). The limiting action of one or several of these processes could be indicated by a plant performance reduction at the edge of the species range, for example in response to biotic (e.g., inter-or intra-specific competition) or abiotic (e.g., too short growing season length, limiting precipitation or temperature) factors. However, this hypothesis has not been explicitly tested for invasive species to date.

The variation in the in situ performance of invasive plant populations can help to understand the constraints on the spread dynamic and therefore anticipate the future of the invasion process. For example, the performance of *Verbascum thapsus* L. populations were studied on a large scale by Alba and Hufbauer (2012), and they found that the main competitive interaction that limited *V. thapsus* depended on regional precipitation, which paved the way for additional studies on the species invasion (Seipel et al., 2014). Other large-scale studies have allowed to

63   disentangle the factors influencing the invasion process (see e.g. Jakobs et al., 2004; Erfmeier & Bruelheide

64   2004). However, few data are available about large-scale performance variation for most plant invaders.

65   *Ambrosia artemisiifolia* L. (Asteraceae) is described as one of the most damaging invasive plants in Europe

66   because of the impacts of its pollen on human health (Kazinczi et al., 2008; Laaidi et al., 2003; Smith et al.,

67   2013), and anticipating the future of its invasion is therefore crucial. The current spatial invasion pattern has

68   been derived from pollen maps (Skjøth et al., 2013; Smith et al., 2013) and validated by field observations

69   (Bullock et al., 2010; Fumanal et al., 2008; Martin et al., 2008; Verloove, 2006). The pattern clearly shows

70   multiple foci of very high invasion levels as well as areas that are almost free of the species, and between these

71   two extremes, areas of moderate invasion levels are found. This situation has been observed for years and does

72   not seem to change rapidly. Although some authors showed that *A. artemisiifolia* invasion could be limited by

73   climatic factors (Allard, 1945; Chapman et al., 2014; Deen et al., 1998; Leiblein and Lösch, 2011), it is currently

74   unclear whether this species will continue to spread, or whether the species' area of distribution is in equilibrium.

75   In this study, we aim to test whether a performance variation of *A. artemisiifolia* plants across invasion levels is

76   responsible for an invasion slowdown in Western Europe. To do so, we first defined three invasion level based

77   on atmospheric pollen concentration. Then, we compared growth and reproductive plant performance in areas of

78   different invasion levels, i.e., the highly invaded region of Rhône-Alpes; two areas of moderate invasion: one in

79   the north and one in the south of the region of Rhône-Alpes; and Belgium and the south of the Netherlands,

80   where the species is rare and no invasion has been documented (de Weger et al., 2009; Martin et al., 2008). In

81   addition, we tested if the plant performance was influenced by the intra-and inter-specific competition, by the

82   local climatic conditions or by latitude (a proxy for growing season length).

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84   **2 Material and methods**

85   **2.1 Study system**

86   *A. artemisiifolia* is an annual plant that is native to North America and that was introduced in Europe in the 19th

87   century, most likely in seed lots (Chauvel et al., 2006). It invades both spring crops and open disturbed habitats

88   such as wastelands, roadsides and riverbanks (Bassett and Crompton, 1975; Chauvel et al., 2006; Thibaudon et

89   al., 2004). Separate from its economic impact on crop yields, this wind-pollinated plant is causing a health crisis

90   because its pollen is a strong allergen that causes hay fever, rhinitis and asthma (Thibaudon et al., 2004).

91   There are three main regions in Europe that are highly invaded: the southeast of France (the Rhône Valley) in

92   Western Europe, the Po region in northern Italy and the Carpathian Basin in Eastern Europe (Csontos et al.,

2010; Kazinczi et al., 2008; Mandrioli et al., 1998; Smith et al., 2013). In France, the colonization occurred through multiple introductions throughout the country (Chauvel et al., 2006; Genton et al., 2005). The species is highly problematic in the Rhône Valley (Thibaudon et al., 2004) and has recently expanded into both the Provence-Alpes-Côte-d'Azur and Burgundy regions (Chauvel and Cadet, 2011). North of Burgundy, the species becomes much rarer, but casual populations are described (Just, 2014). Farther north in Belgium, although the presence of the species has been recorded since 1883 (Martin et al., 2008), it is repeatedly described as casual and non-naturalized (Lambinon et al., 2004; Verloove, 2006). The situation is similar in the Netherlands (de Weger et al., 2009). In Germany, the species is described as well established in the southern regions but as remaining casual in the north (Brandes and Nitzsche, 2007). Farther north in Scandinavia, occurrences of the species were found, but the species does not seem presently naturalized (Dahl et al., 1999).

2.2 Population selection

Atmospheric concentrations of *Ambrosia* spp. pollen are monitored across Europe by a network of sites using volumetric spore traps (European Aeroallergen Network, 2013; Smith et al., 2013). Since *A. artemisiifolia* is the only species of its genus occurring in high density populations in Western Europe (Chauvel et al., 2006), the contribution of the other species of the genus can be considered to be negligible. Under normal weather conditions, pollen concentrations are highly dependent on the number of *A. artemisiifolia* plants within a distance of about 30 km (Fumanal et al., 2007a; Skjøth et al., 2010). The pollen density database allows the creation of a pollen density map (Skjøth et al., 2013; Smith et al., 2013), that can be used as a spatial assessment of the density of *A. artemisiifolia* populations (Skjøth et al., 2010, 2013; Smith et al., 2013). The annual amount of pollen measured within an area can thus be considered as a good estimate of its invasion level (Skjøth et al., 2010).

Based on this pollen density map and the relevant literature on the species invasion in Western Europe (Bullock et al., 2010; Chauvel et al., 2006; de Weger et al., 2009; Fumanal et al., 2008; Genton et al., 2005; Lambinon et al., 2004; Martin et al., 2008; Verloove, 2006), we considered three contrasted invasion levels:

I) High invasion level, where the pollen index is above 500 pollen grains.m^{-3}.year^{-1}. This level corresponds to the most highly invaded area in Western Europe: the Rhône-Alpes region (Fig. 1). In this area, the species has been well established for more than 120 years, and new populations are regularly found (Fumanal et al., 2008; Genton et al., 2005);

II) Moderate invasion level, where the pollen index ranges from 100 to 500 pollen grains.m^{-3}.y^{-1}. This level is found north and south of the species invaded range: the Burgundy region (northern border; Fig. 1) and the Languedoc-Roussillon region (southern border; Fig. 1) where populations have been established for about 60 years (Chauvel and Cadet, 2011) with a lower occurrence;

III) Low invasion level, where the pollen index is under 20 pollen grains.m^{-3}.y^{-1}. This level corresponds to the area beyond the species current invaded area (Fig.1). In the context of this work, this area is represented by the Belgium and the Netherlands (southern part). In these regions, the species is rare and described as non-naturalized in the literature (Lambinon et al., 2004; Martin et al., 2008; Verloove, 2006).

For the low and the high invasion level, we considered three populations. For the moderate invasion level, we considered three populations in each of the distinct geographical areas it covers (Fig. 1). The 12 populations were growing in ruderal habitats such as riverbanks, shorelines, pebble beds and roadsides. The populations contained at least 50 individuals growing on a given site and were at least 40 kilometers apart. We only selected unmanaged populations, i.e., without traces of mowing or herbicide application on the growing plants, to avoid bias in trait measurements. The locations, invasion levels, annual precipitation and annual mean temperatures in the vicinities of the 12 sampled populations (Meteo Belgique 2014; Meteo France 2014) are presented in Table 1.

2.3 Data collection

Two measurement campaigns were performed in the 12 populations. The first one took place in August 2013 in order to evaluate the growth and ecophysiological traits of 20 individuals per population. The second campaign took place in November 2013, when seeds had reached maturity, in order to measure the reproductive traits of 25 different individuals. Particular attention was paid to monitoring plant maturity in order to know when to launch the second campaign in the different regions to ensure the collection of mature seeds before they dispersed.

Plant selection was made along transects. We laid the longest possible transect in the population and divided it into 20 (first campaign) or 25 (second campaign) equal segments. At each segment start, we selected the nearest plant for measurements.

During the first campaign, the 20 plants per population were cut at ground level. Three fully developed leaves without damage were randomly picked from each plant to measure the specific leaf area (SLA). The leaves were scanned, then oven-dried for 2 days at 80°C, and finally weighed to the nearest 10^{-4} g (using XA105 Mettler Toledo®, Viroflay, France). SLA is an important trait regulating and controlling plant functions such as carbon

assimilation and carbon allocation (Poorter and Nagel, 2000; Poorter, 1999; Wilson et al., 1999). SLA is known to be impacted by soil nutrient content, irradiance, and water availability (Meziane and Shipley, 1999; Sánchez-Gómez et al., 2013; Yousfi et al., 2015). For example, low irradiance can induce in the resource allocating strategy to enhance light interception for photosynthesis. This could be primarily achieved by increasing SLA (Poorter and Nagel, 2000). Poorter (1999) indicated that SLA may be one of the key traits determining the maintenance of a species' growth rate under low levels of irradiance. Area measurements were taken using ImageJ (National Institutes of Health, USA). The rest of the plants were also oven-dried for 2 days at 80°C and weighed to the nearest 10^{-4} g to determine their above ground biomass (AGB) as a proxy for growth. To evaluate the impact of the competition exerted by the local flora on SLA and AGB, we estimated the percentage of the vegetation cover to the nearest five percent around each plant in a 1x2 m quadrat. The quadrat was positioned in order to have the considered plant at the quadrat center. We calculated the mean vegetation height based on the height measurement at 6 randomly chosen points and then calculated inter-specific competition index as the vegetation cover multiplied by the mean vegetation height. High population density, through intra-specific competition, may also have a negative impact on performance (Creed et al., 1996). To take into account this possible influence on SLA and AGB, the distances to the three nearest neighbors were measured for each considered plant, and the mean of the 3 values was used as the average nearest-neighbors distance.

In the second campaign, the 25 plants per population were also cut at ground level and oven-dried for two days at 80°C. To assess the seed number, i.e., the number of seeds per plant, dried plants were lightly hand crushed to detach the seeds. The resulting material was spread on a sieve with a 1 mm mesh width. The seeds were separated from the vegetative material by blowing air through the sieve at increasing speed, and then the seeds were weighted to the nearest 10^{-2} g, and counted using a Contador (Pfeuffer®, Kitzingen, Germany). On a subset of 30 seeds from each plant, we performed a pressure resistance test following Guillemin and Chauvel (2011) in order to estimate the empty seed proportion. The biomass of the plants was also measured following the same method as that used for the first campaign in order to calculate the reproductive effort, as the ratio of the seeds biomass to the sum of the seed and above ground biomasses.

To take climate variation into account across the sampled range, we collected monthly temperature means and monthly rainfall during the actual growing season (from March to November 2013) from the weather station that was closest to each population (Meteo Belgique 2014; Meteo France 2014; Table 1). The influence of the latitude was also tested because it can have an impact on performance through the length of the growing season, and this impact might not be detected using only local climatic data (Kollmann and Bañuelos, 2004).

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2.4 Statistical analysis

To explore the effect of invasion level, population, inter-specific competition index, and average nearest-neighbors distance on AGB and SLA, we used a mixed model analysis of covariance (ANCOVA). We included invasion level as a fixed effect, inter-specific competition index, and average nearest-neighbors distance as covariates, and population nested within invasion level as a random effect. To explore the effect of invasion level and population on seed number, empty seed proportion, and reproductive effort, we used a mixed model analysis of variance (ANOVA). We included invasion level as a fixed effect, and population nested within invasion level as a random effect. The AGB, SLA, and seed number were log-transformed, and the empty seed proportion was arcsine-square root transformed to improve homoscedasticity.

In contrast to inter-specific competition index or average nearest-neighbors distance, climatic data were related to the entire population and therefore could not be included as a covariate. We therefore performed a principal component analysis (PCA) to summarize all local climatic data, and we stored the first axis in a new variable (PCAclim, which explained 53% of the variance). For each performance-related trait, we calculated the mean for each population. We made linear regressions between PCAclim and the population means of each performance-related trait. Then, we conducted another PCA that summarized the mean performance-related traits, and we also stored the scores in a new variable (PCAperf, which explained 46% of the variance). We made a linear regression between PCAperf and PCAclim to test whether overall performance was correlated to local climatic data. Finally, we performed a last linear regression between PCAperf and latitude to test if the overall performance was correlated to this variable. All statistical analyses were conducted using the statistical software Minitab® ver. 16.2.2 (Minitab Inc., State College, PA, USA).

3 Results

We found high variability in all of the performance-related traits (Fig. 2). AGB averaged $2.44 \pm 3.89\ 10^{-1}$ g (mean $\pm$ SE), SLA averaged $29.2 \pm 6.00\ 10^{-1}$ mm².mg^{-1}, seed number averaged 227 ± 18.4, reproductive effort averaged $28.6 \pm 8.19\ 10^{-1}$ %, and empty seed proportion averaged $7.79 \pm 6.85\ 10^{-1}$ %.

The ANCOVA showed that AGB was not significantly different among invasion levels ($F_{2,\ 9} = 0.00$; $P = 0.998$), although it differed significantly among populations ($F_{9,\ 220} = 5.41$; $P < 0.001$). The same result occurred with SLA: there were no significant differences among invasion levels ($F_{2,\ 9} = 2.06$; $P = 0.184$) but significant differences among populations ($F_{9,\ 220} = 20.56$; $P < 0.001$). Inter-specific competition index did not impact AGB

212   ($F_{1, 220} = 0.34$; P = 0.560), but it positively impacted SLA ($F_{1, 220} = 45.50$; P < 0.001). Average nearest-neighbors

213   distance did not impact AGB ($F_{1, 220} = 0.27$; P = 0.601) neither, but it negatively impacted SLA ($F_{1, 220} = 4.88$; P

214   = 0.028). The mean Average nearest-neighbors distance of each population is given in Table 1. The ANOVA

215   showed similar results; we did not find any differences between invasion levels for seed number ($F_{2, 9} = 0.270$; P

216   = 0.768), reproductive effort ($F_{2, 9} = 0.800$; P = 0.481), or empty seed proportion ($F_{2, 9} = 2.18$; P = 0.169). The

217   differences between populations remained highly significant for all performance-related traits: seed number, $F_{9, 220} = 9.33$ (P < 0.001); reproductive effort, $F_{9, 220} = 6.35$ (P < 0.001); and empty seed proportion, $F_{9, 220} = 6.06$ (P

219   < 0.001).

220   The results of the regressions between PCAclim and each performance-related trait showed no significant

221   correlations: AGB, R^2 =13.4% (P = 0.242); SLA, R^2 =4.3% (P = 0.523); seed number, R^2 =3.9% (P = 0.537);

222   reproductive effort, R^2 =4.7% (P = 0.497); and empty seed proportion, R^2 =23.9% (P = 0.107). The same result

223   occurred when we summarized the performance-related traits using the PCA: the regression between PCAperf

224   and PCAclim did not show a significant correlation ($R^2 = 7.4\%$; P = 0.392). Finally, the regression between

225   PCAperf and latitude was also not significant ($R^2 = 5.1\%$; P = 0.480).

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227   **4 Discussion**

228   In Western Europe, the invasion of *A. artemisiifolia* is well described, with multiple foci of very high invasion

229   levels, moderate invasion levels, and low invasion levels in areas that are almost free of the species but where the

230   presence of rare populations are documented (Martin et al., 2008). In this study, we attempted to determine

231   whether this pattern, which does not appear to change rapidly, could be explained by declining plant

232   performance in areas of lower invasion levels. In contrast to many studies that investigate the varying

233   performance of a species in ex situ conditions, e.g., in common gardens, we chose to perform an in situ survey

234   that allowed us to study phenotypic variation under real conditions. The results showed that the performance-

235   related traits were similarly high across all investigated areas (Fig. 2) and that there were no significant

236   differences across invasion levels; the hypothesis of a plant performance variation across invasion levels

237   responsible for an invasion slowdown in Western Europe is thus rejected.

238   The reproductive performance of the species in the area of low invasion level tends to question its casual status

239   north of France. According to Richardson et al. (2000), the casual status apply to introduced taxa that can

240   reproduce occasionally, but do not form self-replacing populations, and that rely on repeated introduction for

241   their persistence. Naturalized taxa, however, involve alien plants that reproduce consistently, sustain populations

over many life cycles without direct intervention by humans, and do not necessarily invade natural, semi-natural or human-made ecosystems (Richardson et al., 2000). Despite that we did not study the populations dynamic; we found that the species can produce lots of viable seeds in the area of low invasion level. The populations sampled in this area are most likely able to survive from year to year without an anthropogenic intake of seeds. *A. artemisiifolia* thus appears to be naturalized rather than casual in this area.

A single growing season was investigated in this study. In the case of an annual species that produces a large number of long-lasting seeds, a single year of successful fructification can stock the soil seed bank and thus ensure the establishment of the species for a number of years. This is typically the case with *A. artemisiifolia* (Bassett and Crompton, 1975). It is thus relevant to consider a single growing season with successful seed production, since that season is sufficient to establish the population for many years.

We tested the influence of local climate and latitude on the performance-related traits, and we found no significant correlation between local climatic data and performance-related traits, neither individually nor summarized together; overall performance also did not appear to be related to latitude. These results suggest that phenotypic variation in *A. artemisiifolia* does not clearly respond to a climatic or latitudinal pattern in the geographical range we examined. This observation contrasts with the latitudinal and environmental trends that have been found in situ for other invasive species (Kollmann and Bañuelos, 2004; Monty and Mahy, 2009; Parker et al., 2003; Weber and Schmid, 1998). The reason for this absence of response might be a lack of climate-induced genetic structuring in the invaded range we considered. Indeed, the invasion of *A. artemisiifolia* in Western Europe has been facilitated by multiple sources of introduction that have resulted in very high genetic diversity in its populations (Genton et al., 2005). In addition, evidence of active gene flow and population admixture has been documented (Chun et al., 2010).

We found considerable variability in all of the measured performance-related traits (Fig. 2; coefficients of variation: SLA = 31.5; AGB = 244; seed number = 140; empty seed proportion = 152; reproductive effort = 49.7). This high variability could reflect the ability of the species to thrive in various environmental conditions (Willis and Hulme, 2004), which suggests strong phenotypic plasticity, e.g., in response to soil conditions, and which is consistent with other studies on the species (Brandes and Nitzsche, 2006; Fumanal et al., 2007b; Leiblein and Lösch, 2011; Ortmans et al., 2016). However, it is possible that a decrease in performance-related traits has not been detected, given this high variability of the measured traits, and the relatively small number of sampled populations. This plasticity could also explain the lack of clear differentiation in response to climate.

The species is known to be particularly sensitive to competition (Leskovšek et al., 2012). The results showed that average nearest-neighbors distance decreased SLA, which means that intra-specific competition increased SLA. The inter-specific competition also increased SLA. This influence of intra-and inter-specific competition translates a change in resource use strategies and thus suggests an environmental stress for the plants. The competition decreased the irradiance received by *A. artemisiifolia* leaves, which led to an increase of leaf area by biomass unit to compensate (Poorter and Nagel, 2000). The population D is a good example: it had almost not suffered any intra-or inter-specific competition because it occurred on a pebble beach, and had the smallest SLA because full irradiance was available for each plant. Despite this stress due to intra-and inter-specific competition highlighted by SLA, the plants showed an unaltered performance-related trait, AGB, across the study regions. This demonstrates the species' ability to cope with various environmental conditions and stresses without decreasing performance-related traits. These findings on phenotypic plasticity suggest a Jack-of-all-trades strategy (Richards et al., 2006). The impact of intra-and inter-specific competition was only assessed during the first field campaign, so it was not possible to assess its impacts on the other performance-related traits. Further research on these impacts would allow to better explore this strategy. In the area of high invasion level, one can expect populations to show a higher plant density on average than in the other areas (Brussard, 1984). This higher plant density could have had an adverse effect on plant performance, and thus led to an underestimation of the plant performance in high invasion level. This underestimation, in turn, could have concealed a performance reduction in the area of low invasion level. Since the AGB was left unaltered by the intra-specific competition, population density probably did not have significantly impacted plant performance, and this hypothesis is therefore held invalid.

The different levels of invasion in Western Europe were not explained by the variation in performance-related traits. Thus, one may wonder why the species does not seem to rapidly spread to and colonize northern areas.

The hypothesis tested in this work focused on the plant performance and did not take into account processes occurring at other scales, e.g., population dynamic. Although the seed production was assessed, seed germination and seedling survival, for instance, may differently affect recruitment dynamic in field populations. Other processes than reduced performance in low invasion area could explain the observed invasion pattern in Western Europe.

First, the residence time, the time from the first record in the wild until now, is known to be an important determinant of the geographical range sizes of alien plants species (Williamson et al., 2009). One can hypothesize that the populations in the area of low invasion level, due to their much smaller residence time, had

301 less time to recruit as much individuals and establish as much populations as in the areas of higher invasion

302 levels. However, this hypothesis is unlikely since the first occurrence of the species in the area of low invasion

303 level has been recorded in the late 19th century (Martin et al., 2008), like in the other levels (Chauvel et al.,

304 2006).

305 Genetic constrains on adaptive evolution during range expansion, e.g., low genetic variation, maladaptive gene

306 flow, or genetic correlation, could also explain an invasion slowdown (Alexander and Edwards, 2010; Colautti et

307 al., 2010). However, a very high genetic diversity has been documented in Western Europe for *A. artemisiifolia*

308 (Genton et al., 2005) and to date, no evidence of genetic constrains limiting the range expansion of *A.*

309 *artemisiifolia* has been highlighted.

310 Another possible explanation might be a lack of effective dispersal to suitable sites in areas of low invasion

311 levels. For example, the spread of the species by agricultural machinery might be hampered by the absence of

312 sunflower fields north of Burgundy, because sunflowers are known to be one of the most invaded field crops in

313 France (Genton et al., 2005). If this lack of dispersal is actually the main factor that limits its invasion, the

314 species would therefore be in a lag phase and could show an invasive behavior in the future. Invasion might also

315 accelerate under the pressure of different processes and human activities, such as global warming that allows

316 seed production in news areas, changes in agricultural practices (herbicide withdrawal), the displacement of

317 embankments that are already contaminated with *A. artemisiifolia* seeds, etc.

318 This work showed that the invasion slowdown, if it exists, is not caused by reduced plant performance. In the

319 area of low invasion level, the species was as performant as in the areas of higher invasion levels. This result

320 suggests an invasion potential in Belgium and southern Netherlands if there is no obstacle to the dispersal,

321 germination, and plant survival. Further research that extends this study to the northern countries would be

322 interesting to conduct. In addition, the investigation of the other process that could limit the invasion would help

323 to forecast the future invaded range. In this uncertain situation, early detection and structured monitoring of

324 population establishment should be encouraged in order to avoid future problems related to *A. artemisiifolia*

325 invasion north of its current distribution in Western Europe.

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499

500   **Caption of figures and tables**

501   Table 1. Location of the 12 sampled populations of *Ambrosia artemisiifolia*, with their invasion level, annual

502   precipitation, mean temperature, pollen density index, and the average nearest-neighbors distance.

503

504   Fig. 1. Locations of the 12 sampled populations of *Ambrosia artemisiifolia* in Western Europe. The symbol

505   shapes indicate the invasion levels of the area: discs (●) correspond to low invasion levels, triangles (▲) to

506   moderate levels, and crosses (+) to high levels.

507

508   Fig. 2. Means and standard errors of inter-specific competition index (a), average nearest-neighbors distance (b),

509   and performance-related traits (c-g) of *Ambrosia artemisiifolia* among populations and invasion levels. Low =

510   area of low invasion level; Moderate = area of moderate invasion level; High = area of high invasion level.

511