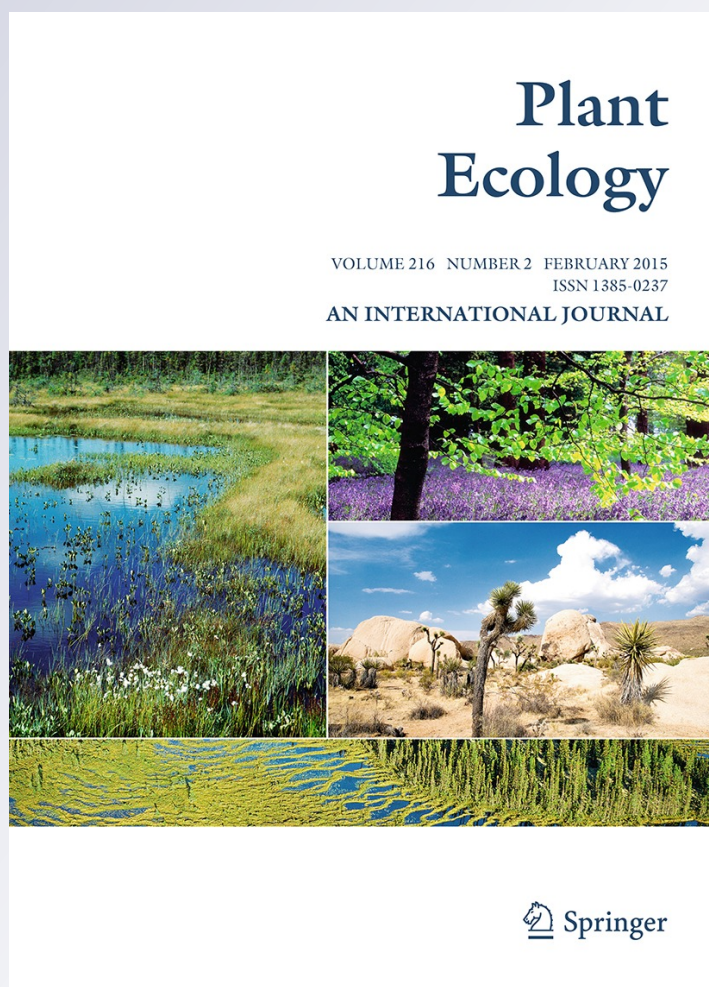*Do climatic and habitat conditions affect the reproductive success of an invasive tree species? An assessment of the phenology of Acacia longifolia in Portugal*

Patrícia Fernandes, Cristina Antunes, Otília Correia & Cristina Máguas

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Do climatic and habitat conditions affect the reproductive success of an invasive tree species? An assessment of the phenology of *Acacia longifolia* in Portugal

Patrícia Fernandes · Cristina Antunes · Otília Correia · Cristina Máguas

Abstract Plant phenological events are some of the most sensitive indicators of how plant species respond to favourable or stressful conditions. The evaluation of the flowering phenology of invasive plant species is particularly relevant, mainly due to its crucial importance in determining plant reproductive success and the outcome of invasion. We studied the phenology of *Acacia longifolia*, an aggressive, invasive plant species in the Mediterranean basin. We measured its vegetative growth and reproductive traits, specifically flowering phenophases and fruit production, under different climatic conditions (mesic and xeric Mediterranean climates), and in two different habitats (pine forest and open area). All the measured phenological phases began earlier at the xeric site than at the mesic site; this was particularly evident when comparing reproductive phenophases. Flowering dates were significantly associated with air temperature, with early peak flowering dates linked to increases in air temperature. The number of fruiting flowers per inflorescence in *A. longifolia* trees was higher at the mesic site, mainly in the pine forest plot, and the number of aborted fruits was notably lower than in the xeric plots. The presence of a pine forest at the mesic site strongly influenced the flowering phenology of *A. longifolia* and resulted in the highest reproductive success and the lowest branch growth rate. Our results demonstrate that a combination of climate and forest structure can cause pronounced differences in phenology and reproductive success of *A. longifolia*. These data can help to understand the variations in invasive rates of *A. longifolia* across the Mediterranean basin.

Keywords Plant phenology · Invasive plants · Maritime pine forest · Dunes

Introduction

Phenology is concerned with the timing of events and in general can be defined as the study of the relationships between temporal occurrences of biological events and climatic variables (Dahlgren et al. 2007; Matesanz et al. 2010; Post and Stenseth 1999; US/IBP Phenology Committee 1972). Plant phenological events are some of the most sensitive indicators of how plant species respond to climate and favourable or stressful conditions, and have been a key topic in botany (e.g. Jackson 1966; Lieth 1974; Rathcke and Lacey 1985).

Recently, plant phenology has emerged as an important focus of ecological research, mainly in the context of global climate change, leading to a new perspective on plant responses to environmental conditions (Dahlgren et al. 2007; Schwartz 1999; Sekhwela

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and Yates 2007). This renewed interest is partly due to findings that earlier flowering is strongly related with warmer springs (Fitter 1995; Miller-Rushing et al. 2008; Sparks and Carey 1995; Sparks et al. 2000).

As climate variations affect the phenology of native plant species, they also directly affect the success of introduced species (Castro-Díez et al. 2003; Godoy et al. 2009a, 2009b; Walther et al. 2009). Such evaluation is unquestionably important, although little attention has been paid to the phenological response of invasive species under specific macro- and micro-climatic conditions. Considering the importance of phenological events, this could be essential to understanding the influence of local environment on the spatial distribution and invasive potential of non-native plant species, in order to improve predictions of invasiveness related with global changes.

Particularly relevant is the evaluation of the flowering phenology of invasive plant species, due to its crucial importance in determining plant reproductive success and invasion success (Godoy et al. 2009a, 2009b; Gibson et al. 2011). For instance, one important trait linked with invasive success is an appropriate flowering phenology (i.e. the occurrence at the right point in time), which is key to improving fruit development and seed production ((Lake and Leishman 2004; Pysek and Richardson 2007).

The Mediterranean basin, including Portugal, has been severely affected by invasions of introduced plant species (Groves and di Castri 1991), for example Australian acacias (Marchante et al. 2003). The typical Mediterranean climate is characterized by cool, wet winters and hot summers with high irradiance and little or no precipitation, which subject plants to the distinctive Mediterranean summer drought stress (Di Castri et al. 1981). This marked seasonality strongly influences plant phenology (Castro-Díez et al. 2003; Milla et al. 2010). Australian acacias produce long-lived inflorescences (Stone et al. 2003), and in non-native habitats their flowerings tend to occur earlier than in most native species, reducing their competition for pollinators (Gibson et al. 2011). Acacias do not appear to rely on anemophilous pollination (Fleming et al. 2007; Gibson et al. 2011). Indeed, the release of floral scents that precedes pollen release, as well as the attractive appearance of the dense mass of inflorescences suggests biotic pollination (Stone et al. 2003). Insects are considered to be the main pollinators of most acacias studied to date (Stone et al. 2003).

Acacia longifolia (Andrews) Willd. is one of the most aggressive invasive plant species in Portuguese dune ecosystems, having negative impacts on native biodiversity (Hellmann et al. 2011; Marchante et al. 2003) and community structure (Rascher et al. 2011; Rascher et al. 2012). At the beginning of the 20th century, *A. longifolia* was deliberately introduced on the Portuguese coast, mainly to stabilize open sand dunes and control their erosion (Marchante 2011), but rapidly spread to adjacent forest areas, mainly *Pinus pinaster* plantations of high economical and biological value.

The aim of this study was to identify the influence of climate (mesic and xeric) on the timing of the phenophases of *A. longifolia*, and the relative importance of habitat structure (pine forest and open area) on the species' vegetative growth and reproductive traits, specifically flowering phenophases and fruit production, in order to understand the influence of local environment on the invasive potential of this non-native plant species. These reproductive traits are crucially important to the establishment and spread of invasive plant species (Godoy et al. 2009a; Gibson et al. 2011), and are subject to selective pressures of resource availability and climatic conditions (Elzinga et al. 2007). On the basis of the above information, we expected a delay in the timing of phenophases under mesic conditions. We also expected *A. longifolia* plants to display higher vegetative growth and reproductive fitness under mesic conditions.

Materials and methods

Study sites

Field sites were established along the Atlantic coast in the north (Osso da Baleia, 40°0′N, 8°54′W, 32 m altitude, 180 km north of Lisbon) and south (38°15′N, 8°45′W, altitude 50 m, 70 km south of Lisbon) of Portugal. The climate at both study locations can be classified as Mediterranean-type with an oceanic influence due to the proximity of the Atlantic Ocean. Average (30 year average 1961–1991) yearly precipitation is 841 mm at the north site (nearest meteorological station 40°39.7′N, 8°44.1′W). The south site is drier having average yearly precipitation of 573 mm (nearest meteorological station 38°22.3′N, 8°27′W). At both sites the precipitation occurs predominantly from autumn to early-spring (October to April). Summers

are dry with 50 mm of rain during summer months (June to August) at the north site and only 26 mm at south site. Furthermore, the north site is cooler (mean annual temperature 14.4 °C, mean maximum temperature for the hottest month 22.3 °C) than the south (mean annual temperature 16.3 °C, mean maximum temperature for the hottest month 30.8 °C). In this study, north site was classified as mesic and south site as xeric. Both sites are interior secondary dune ecosystems occupied by endemic xerophytic shrub and *Pinus pinaster* Aiton (Maritime Pines) forests. Both sites are also occupied by invasive *A. longifolia*.

Two adjacent sample plots of 800 m^2 (40 m × 20 m) were established at each study site: one with *A. longifolia* trees under pine overstory (forested plot); and another with acacias and no pine overstory (open plot). Each site had isolated populations of *A. longifolia* (characteristic of the early stages of invasion), growing as single small trees in both the forested and open plots. The number of *A. longifolia* trees in the sampled plots ranged from 31 at the south site to 45 at the north site. At the south site, *A. longifolia* trees had an average basal area at ground level of 0.0046 m^2 and an average crown projected area of 8.4 m^2. The corresponding values for the *A. longifolia* trees sampled at the north site were 0.005 and 7.9 m^2, respectively. The main characteristics of the overstory (*P. pinaster*) in the forested plots are presented in Table 1.

Meteorological measurements

Climate conditions were recorded during the study period, from January 2010 to May 2011, using an automatic weather station at each site, located approximately 15 m from each study site (without overstory plants). Air temperature, relative humidity (CS-215 Temperature and Relative Humidity Probe, Campbell Scientific, Logan, UT, USA) and rainfall (ARG100 Rain gauge; Campbell Scientific, Logan, UT, USA) were automatically stored every 60 min in Campbell Scientific dataloggers (CR1000, CR10X) and multiplexers (AM416, AM16/32). At both sites, daily mean air temperatures and cumulative rainfall were calculated. In both years and at both sites, daily values of temperature (mean, maximum and minimum) (T, °C), relative humidity (RH, %), vapour pressure deficit (VPD, Pa KPa^{-1}) and rainfall (mm) were averaged (T, RH, VPD) or summed (rainfall) for the period between 1 January and the peak flowering date of the sampled *A. longifolia* trees. VPD at each weather station was calculated from air temperature and air relative humidity values.

Phenological measurements

In 2010, we selected isolated plants of *A. longifolia* at each study site (eight plants from each open plot and eight plants from each forested plot, thus a total of 32 plants) for observation of phenological phases and phenological measurements. The selected plants in this study had similar basal areas (0.0049 m^2 at the south site and 0.0053 m^2 at the north site, $p > 0.05$), similar canopy covers (7.4 and 8.2 m^2 at the south and the north sites, $p > 0.05$) and mean plant height per site ranged from 4.5 m in north site plants to 5.1 m in south site. This variation in plant height among study plots is not significant ($p > 0.05$).

Table 1 Main characteristics of *Pinus pinaster* tress at forested plots

Maritime pine characteristics	North site	South site	Significance
Mean canopy cover (m^2 tree^{-1})	7.43 (4.31)	5.17 (2.42)	***
Mean basal area (m^2 tree^{-1})	0.05 (0.03)	0.03 (0.01)	**
Total density (nº trees ha^{-1})	1,400	630	–
Total canopy cover (m^2 ha^{-1})	10,402	3,257	–
Total basal area (m^2 ha^{-1})	70.0	18.9	–

Values of canopy cover (m^2 tree^{-1}) and mean basal area (m^2 tree^{-1}) correspond to the average canopy cover and average basal area from all maritime pines present in each forested plot (800 m^2), 50 trees at the south site and 112 trees at the north site; numbers in brackets represent standard deviations. Total canopy cover and total basal area correspond to the total canopy projected area (m^2) of those same trees

Values with asterisks are significantly different between north and south sites (** $p < 0.01$; *** $p < 0.001$, according to Student's *t* test)

Phenological studies were conducted during 16 months, from February 2010 to May 2011. Observations were made approximately every 10–15 days in the periods with more intense phenological activity, such as flowering and fruit development. In the other months, with lower phenological activity and few changes, observations were made every 4 weeks. The following phenological events in each selected *A. longifolia* plant were recorded: vegetative growth (VG), inflorescence bud formation (IBF), inflorescence flowering (IF), fruit ripening (FR), fruit growth (FG). Each phenological phase was considered present in the plant when displayed by more than 10 % of the plant crown (adapted from Castro-Díez and Montserrat-Martí 1998). The inflorescence flowering phase was only considered to occur when open fresh flowers were visible. In each plant, the start and end dates of the phenological phases were determined by interpolation between the dates when more than 10 % of the crown plant displayed the specific phenophase and that when less than 10 % of the plant crown displayed the same specific phenophase. The semi-quantitative method adapted by Montserrat-Martí and Pérez-Rontomé (2002) was used to quantify the frequency of the phenological phases according to two frequency classes: I—specific phenophase in more than 50 % of the plants; II—the same in 25–50 % of plants. A phenological diagram was drawn, to synthetically show the frequency of occurrence of phenophases along the study period.

Flowering phases (start, peak and end) were registered during both study years, with the exception of the start of flowering in 2010. We defined the peak flowering date as that when approximately 90 % of the inflorescences in the plant had open fresh flowers. We also calculated the time between start of flowering and end of flowering to provide an estimate of flowering duration in each plant. In order to understand the relationship between peak flowering date and microclimatic conditions, each meteorological variable (T, RH, VPD and rainfall) was subjected to simple linear regression analysis. For the purposes of analysis, peak flowering dates were converted to ordinal dates (day 1 = January 1). Data for each plant were plotted as the reciprocal of the ordinal date of peak flowering against the average mean temperature. From the regression equation, base temperature (T_{base}) was determined as the intercept with the temperature axis, and thermal time (Tt) obtained from the slope (Tt = 1/slope).

Additionally, three branches were selected from each *A. longifolia* plant (terminal branches facing different directions) for quantification of branch elongation, inflorescence and fruit production. Branch length was measured using a steel tape (error ± 1 mm), and relative growth rate (RGR, expressed as $cm^{-1} d^{-1}$) was determined: $RGR = [(\log(Fg) - \log(Ig))/N] * 1000$ ($10^{-3} cm^{-1} d^{-1}$), where Fg is the final length, Ig is the initial length; N is the interval between measurements, in days. We registered the number of inflorescences with open fresh flowers per marked branch during the flowering period. To best visualize the temporal flowering phases, the data for each plot were fitted with a Gaussian curve. This curve-fitting shifted the dates of start of flowering, peak flowering or end of flowering by 3 or 4 days, forwards or backwards, at most. The number of fruits was also monitored on the same branches. Reproductive success was estimated as the ratio between the maximum number of inflorescences with open flowers and the maximum number of fruits on each branch. The number of aborted fruits was assessed by comparing the total number of fruits produced with the total number of fruits matured per branch; we considered mature fruits to be those which were brown and developed seeds.

Data analysis

Differences in Maritime Pine characteristics and *A. longifolia* study plants between north and south sites were discerned using the Student's *t* test. Differences in the fruit set, number of fruits per branch length and percentage of aborted fruits between north and south sites and between forested and open plots within each site were tested using the one-way Analysis of variance (ANOVA). The ANOVA assumptions of normality and homogeneity were tested using the Shapiro–Wilk test and Levene's test (Zar 1999). For linear regressions, the Pearson correlation coefficient was calculated. All tests were performed with the Statistica 11.0 package (StatSoft, Tulsa, OK, USA).

Results

General climate characteristics

Seasonality was evident at both sites, with lower air temperatures and higher precipitation in winter

(November to February) than during spring and summer (March to August) (Fig. 1). The lowest mean temperatures were registered in January: 10.1 and 11.3 °C in 2010, and 11.9 and 10.18 °C in 2011 at the north and south sites, respectively. Total annual rainfall in 2010 was 1096 mm at the north site and 762 mm at the south site. From January 2011 to May 2011, the total recorded rainfall was 449 mm at the north site and 278 mm at the south site (Fig. 1). The spring rain events were short periods of precipitation, particularity at the south site, in contrast to autumn and winter rainfall (data not shown). Summer 2010 was a period with the highest temperatures and almost no precipitation (Fig. 1). On average, the highest temperatures were recorded in July (north site 22.9 °C; south site 28.2 °C), when the precipitation was less than 1 mm at the south site and 2 mm at the north site.

General phenological patterns

The seasonal patterns of phenophases at both sites (north and south site) are presented in Fig. 2. The period between summer and beginning of winter is the most inactive period of the year for the measured

phenological phases. *A. longifolia* invested in branch growth mainly during spring (March–May), with more favourable temperatures (mean air temperatures above 14 °C at both sites) and high water availability. There was no vegetative growth at either site during the summer months (drought season). After the vegetative growth period, *A. longifolia* started to form buds of inflorescences. Although initiated during the summer, their development was delayed until winter, and flowering occurred from late winter to beginning of spring, being about 2 weeks later at the north site. The end of flowering and first display of fruits occurred in March, when the mean air temperature was 1 °C higher than in the previous 2 months, and the mean maximum air temperature was 2 °C higher. Floral development and vegetative growth tended to occur sequentially (Fig. 2). Fruit growth occurred mainly in mid-spring—early summer, after flowering. The difference in the timing of phenophases between sites was more evident in the case of reproductive events than in vegetative growth. All reproductive phenophases took place later at the north site. Fruit growth and maturation occurred around one month earlier at the south site than at north site.

Fig. 1 Daily mean temperature and total monthly rainfall of the north (Osso da Baleia) and south (Pinheiro da Cruz) study sites, recorded by meteorological stations approximately 500 m from each site, from January 2010 to May 2011. South site presents higher temperature and lower precipitation values which increase dryness
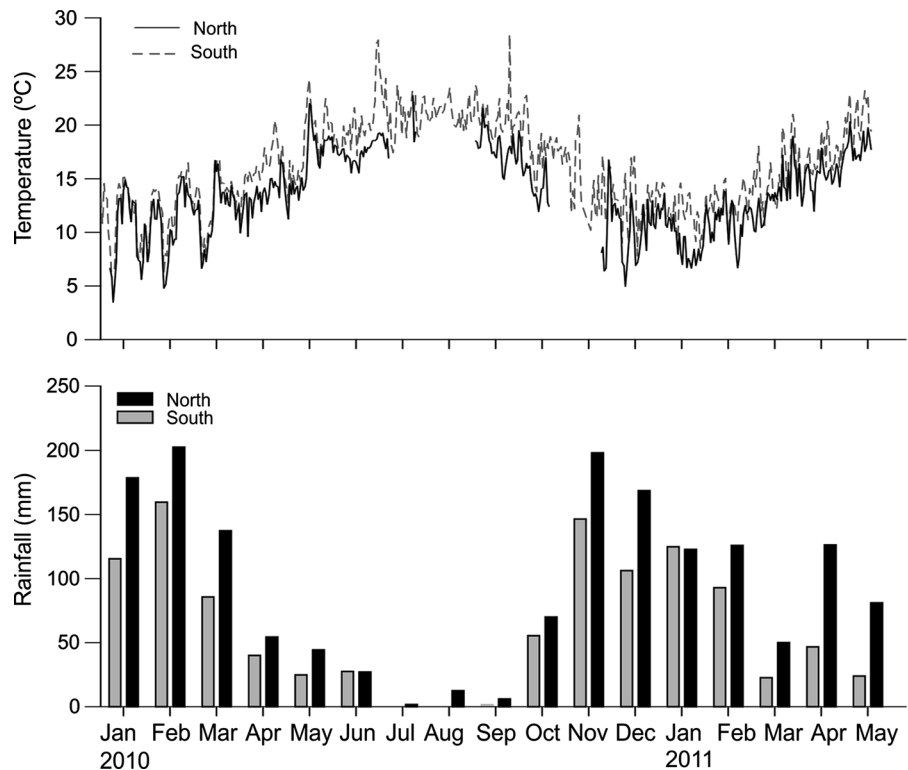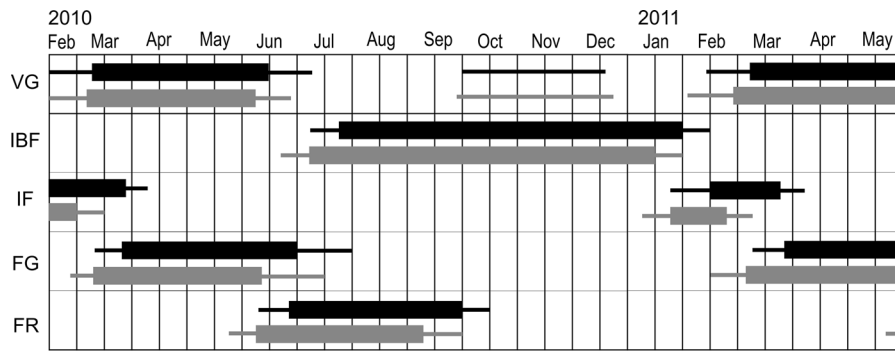
Fig. 2 Phenophase diagram of *A. longifolia* at each study site from March 2010 to May 2011. *VG* vegetative growth, *IBF* inflorescence bud formation, *IF* inflorescence flowering, *FG* fruits growth, *FR* fruits ripening. Frequencies are indicated by filled bars (I: >50 % of the population) and lines (II: 25–50 % of the population). Results are displayed in black for the north site and in grey for the south site

Vegetative growth rate

The vegetative growth rates of *A. longifolia* differed not only between the two sites, but also between plots within each site (Fig. 3). At the north site, the plants in the forested plot had lower branch relative growth rates (RGR) than those in the open plot ($p < 0.001$), whereas there were no significant differences ($p = 0.774$) between the open and the forested plots at the south site. We observed significant differences in *A. longifolia* branch RGR between the north and south sites ($p < 0.0001$ comparing forested plots; and $p < 0.01$ comparing open plots).

Flowering phenology

The phenological diagram (Fig. 2) shows that flowering started and ended earlier at the more xeric south site than at the north site, with significant differences in the timing as well as the duration of flowering (Table 2). It is also possible to discern the lag in the flowering phase between sites by comparing the number of inflorescences per shoot length (flowering density) during the flowering season (Fig. 4). At the north site, the peak flowering date of *A. longifolia* trees occurred in the first week of March (average of both study years), when the plants at the south site were at the end of their flowering phase (Table 2). At the south site, there were no significant differences in the numbers of inflorescences with open flowers between the open and forested plots. *A. longifolia* trees in the north forested plot maintained a higher number of
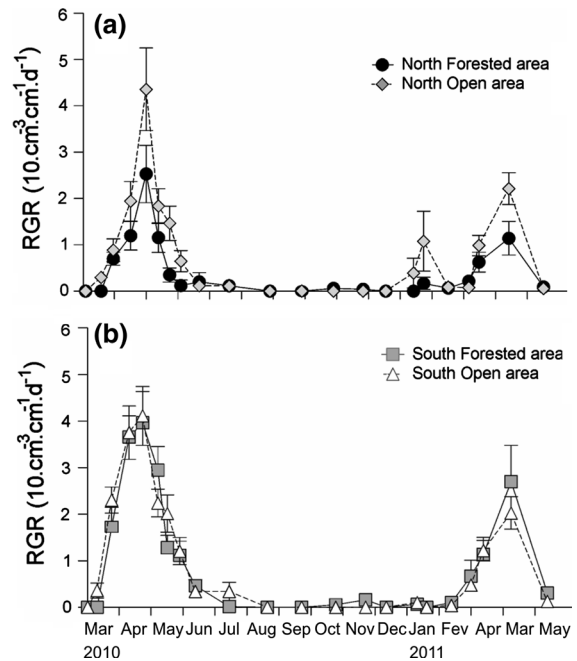

Fig. 3 Branch relative growth rates (RGR) of *A. longifolia*. **a** North site: forested plot and open plot; **b** South site: forested plot and open plot. Mean + SE ($n = 8$)

inflorescences in the period following peak flowering than those in the open plot (Fig. 4).

Influence of climate on flowering phenology

Of the tested climate data, average daily mean temperature was the variable found to be better related, with significant regression, to the peak

Table 2 Mean dates of *A. longifolia* flowering events (start of flowering, peak flowering, end of flowering) and duration of flowering at the study sites in 2010 (peak flowering and end of flowering) and 2011 (start of flowering, peak flowering, end of flowering and duration of flowering)

Site	Start	Peak	End	Duration of flowering
North site	28 January (5.9)	6 March (2.3)	8 April (4.4)	70.5 (6.5)
South site	13 January (3.6)	13 February (2.1)	12 March (1.3)	59.0 (3.3)
Significance	**	***	***	***

Values in parentheses refer to standard deviation ($n = 8$). Flowering duration is given in days. T test significances are indicated: ** $p < 0.01$; *** $p < 0.001$
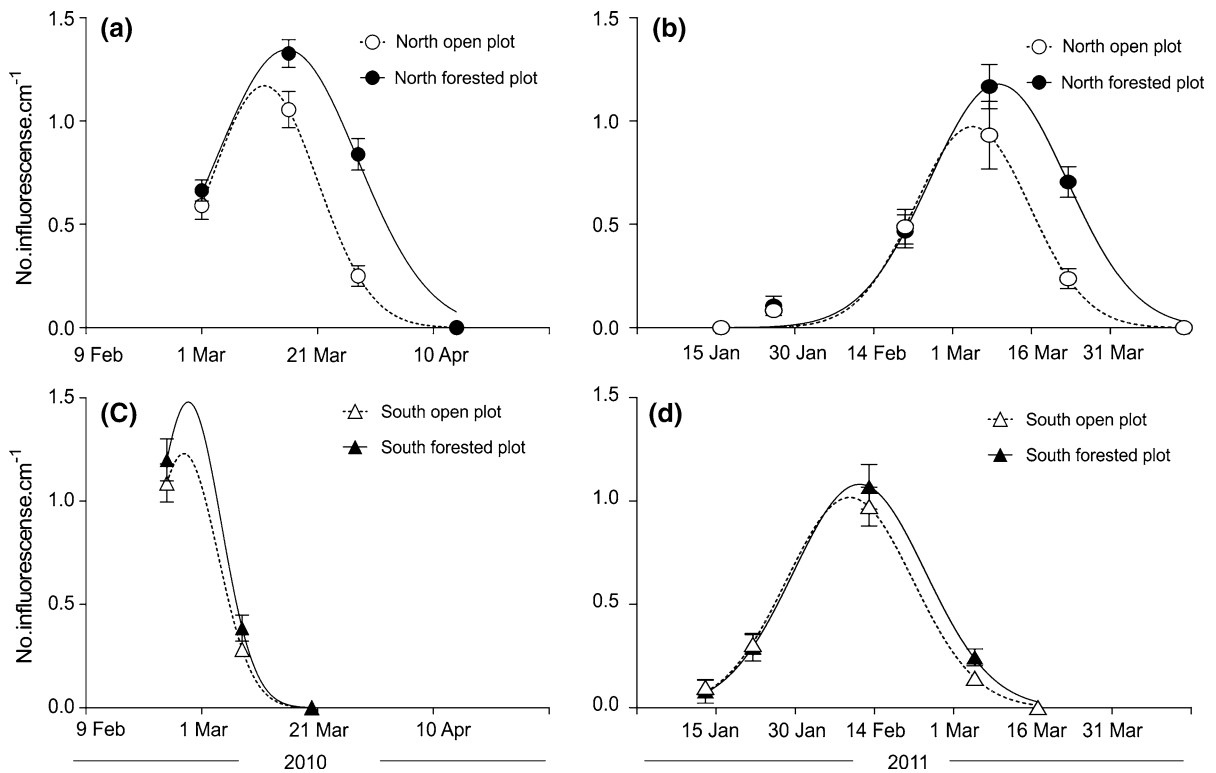

Fig. 4 Inflorescence density per branch (Number of inflorescences with fresh open flowers per branch length (cm)) during the flowering phase of *A. longifolia* at the study sites.

Mean + SE ($n = 24$). **a** North site data in 2010; **b** North site data in 2011; **c** South site data in 2010; **d** South site data in 2011. The lines represent fitted Gaussian curves

flowering date at both sites in both years (Table 3). The peak flowering date was highly related to the average maximum temperature, but not to the average minimum temperature (Table 3). An earlier peak flowering date was associated with an increase in average temperature (T_{mean} and T_{max}) and vapour pressure deficit (VPD), while a later peak flowering date was associated with increases in air relative humidity (RH) and cumulative rainfall (Table 3). The

linear regression between the rate of flowering development (1/days, using the ordinal date of peak flowering) and average daily mean temperature (T_{mean}) shows that T_{mean} alone explained much of the observed variation in the rate of flowering development in both years at both study sites (Fig. 5). Extrapolation of the regression line indicated that T_{base} and Tt required for peak flowering are 6.8 and 231 °C day, respectively.

Table 3 Linear regression analysis of peak flowering date in *A. longifolia* and meteorological variables, between January 1 and peak flowering date

Microclimatic variables	Slope	R^2
Temperature (°C)		
T_{mean}	−0.059	0.73***
T_{max}	−0.031	0.72***
T_{min}	−0.009	0.02 n.s.
Relative humidity (%)		
RH_{mean}	0.239	0.68***
RH_{max}	0.170	0.66***
RH_{min}	0.305	0.31***
Vapour pressure deficit (Pa kPa^{-1})		
VPD_{mean}	−0.007	0.65***
VPD_{max}	−0.026	0.57***
VPD_{min}	−0.041	0.24***
Cumulative rainfall (mm)	7.598	0.65***

Meteorological variables are: average daily mean (T_{mean}), maximum (T_{max}) and minimum (T_{min}) air temperatures; average daily mean (RH_{mean}), maximum (RH_{max}) and minimum (RH_{min}) air relative humidity; average daily mean (VPD_{mean}), maximum (VPD_{max}) and minimum (VPD_{min}) air vapour pressure deficit; and cumulative rainfall. The peak flowering dataset includes all data obtained in 2010–2011 from the four studied plots

Slope indicates an association between the meteorological variable and peak flowering date: positive—earlier; negative—later. *** Significant at $p < 0.001$; n.s. $p > 0.05$

Reproductive success

The proportion of fruiting flowers per inflorescence (Fruit set) in *A. longifolia* was similar in both years at both study sites (Fig. 6a). The number of fruiting flowers per inflorescence in *A. longifolia* trees was significantly higher at the north site, comparing like with like: forested and open plots (Fig. 6a). At the north site, the fruit set in the forested plot was significantly higher than in the open plot, but at the south site it did not significantly differ between plots. The number of mature fruits per branch length displayed similar patterns (Fig. 6b): the differences between north and south forested and open plots in 2010 and 2011 were significant, the open and forested plots were only significantly different at the northern site. The percentage of aborted fruits was significantly higher at the south site, comparing like with like (forested and open plots, in 2010 and 2011), however the open and forested plots were not significantly different at either site (Fig. 6c).
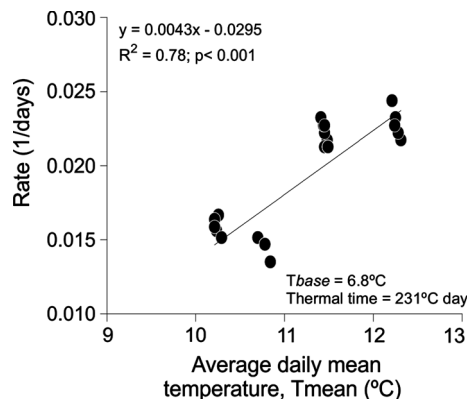

Fig. 5 Influence of average daily mean temperature on rate of flowering development (1/days) until peak flowering date. Each *symbol* represents individual plants of *A. longifolia*. Data include measurements made in 2010 and 2011 in open and forested plots at the north and south study sites; $n = 64$. The *fitted line* represents predicted values for the rate of progress (1/days) from January 1 to peak flowering date according to linear regression. The values above x axis (*lower right corner*) describe the response of rate to temperature, where: T*base* (base temperature) is the intercept with the temperature axis, and Thermal time is obtained from the slope (1/slope)

The linear regression analysis of the fruit set and the date of peak flowering indicated that the flowering date and duration (days) explained a significant amount of the variation in the fruit set (Fig. 7). The fruit set was positively related with peak flowering date, i.e. when the ratio was higher, peak flowering was delayed.

Discussion

In this study, we only partially verified our initial hypothesis that mesic conditions are positively related with enhancement of vegetative growth and reproductive output of an invader plant species (*A. longifolia*). Knowing that the local climate under which plants evolve can determine their phenology (Dahlgren et al. 2007; Godoy et al. 2009b; Marquis 1988) and given the high plasticity of exotic-invasive species (e.g. Máguas et al. 2011), one would expect them to respond to climatic variables by adjusting their phenological phases (i.e. the timing of phenological events), as had been previously demonstrated (Seghieri et al. 2012; Schröder et al. 2014). The above-mentioned findings corroborate our observations that *A. longifolia* displayed different phenological timings in a mesic and in a xeric climate (Fig. 2).

Fig. 6 Mean values (± SD) of **a** Fruit set (fruit/inflorescence ratio), **b** Fruits density (number of fruits per branch length), and **c** proportion of aborted fruits of *A. longifolia*; in forested and open plots at the north and south study sites. Significant differences (*p* < 0.05) between north and south sites are represented by *capital letters* and significant differences (*p* < 0.05) between forested and open plots within each site are represented by *small letters*
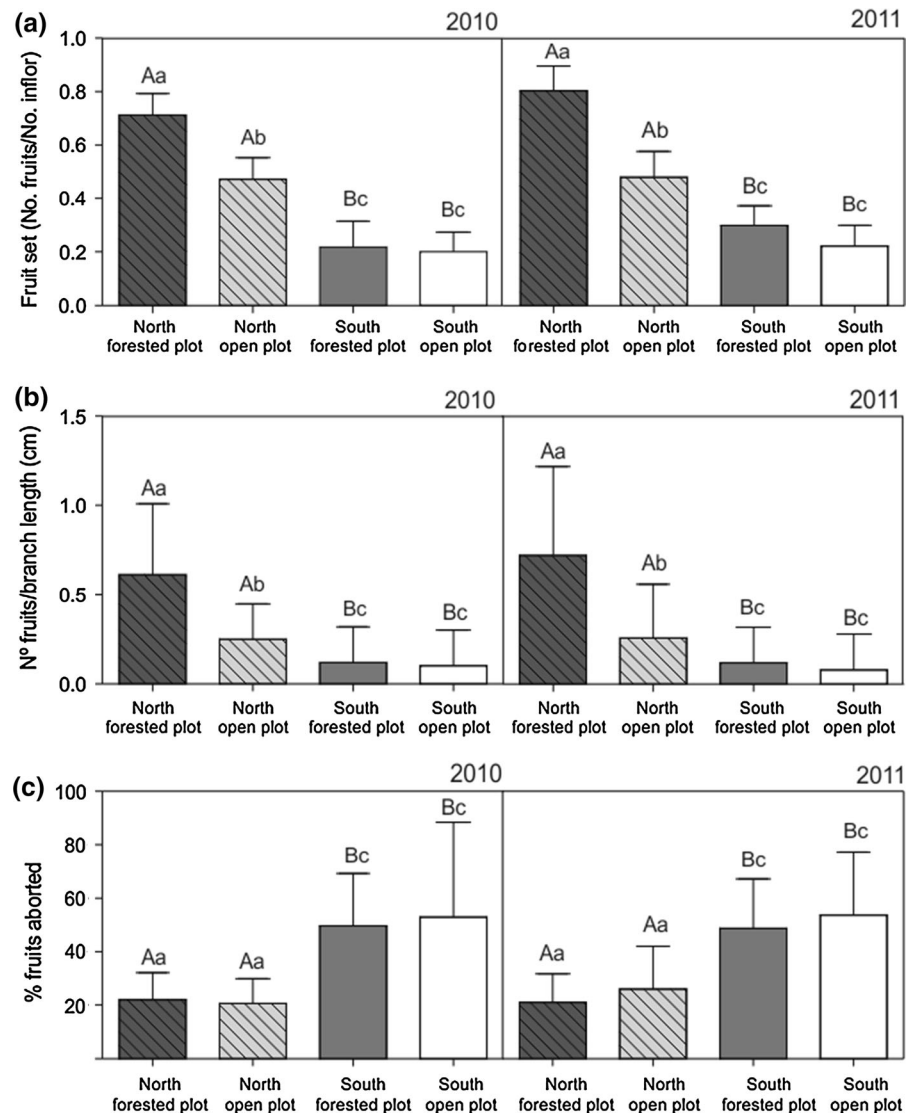
Accordingly, under xeric conditions (Fig. 2, south site), all measured phenological phases occurred earlier than under mesic conditions (Fig. 2, north site) and this was particularly evident when comparing reproductive phenophases (Inflorescence flowering (IF) and fruit growth (FG) and ripening (FR)). Moreover, our results also showed that *A. longifolia* plants under xeric conditions (south site) had a shorter flowering period than those under mesic conditions (north site) (Table 2). These differences could be attributed to the higher temperatures occurring at the south site during the monitoring period. Similar observations have been made in other latitudes and climates, indicating that increased temperatures cause plant phenological events to advance (Lesica and Kittelson 2010; Menzel et al. 2006; Orlandi et al. 2010). The advance in flowering time with increasing temperatures is quite well documented in some species (Fitter and Fitter 2002; Lesica and Kittelson 2010; Orlandi et al. 2010; Sparks et al. 2000). Some studies have indicated that precipitation is also an important factor in flowering phenology (Fox 1990; Lesica and Kittelson 2010; Prieto et al. 2008), and extended blooming is probably restricted by limited precipitation (Bustamante and Búrquez 2008). Once again, our study corroborates these findings: air temperatures
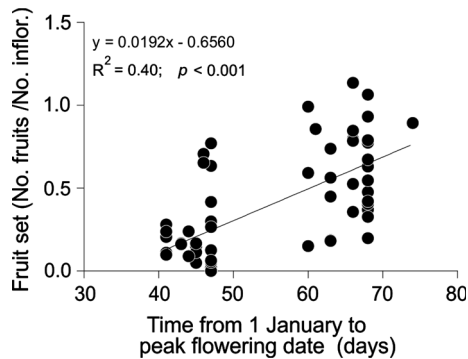
$y = 0.0192x - 0.6560$
$R^2 = 0.40; \quad p < 0.001$

Fig. 7 Relationship between peak flowering date (days) and mean fruit set per plant (represented by the slope of the *fitted line*). Each *symbol* represents individual plants of *A. longifolia*. Data include measurements made in 2010 and 2011 in open and forested plots at the north and south study sites; $n = 64$

(T_{mean} and T_{max}) were found to be the most important meteorological drivers of *A. longifolia* peak flowering date (Table 3). We also found a significant association between the early peak flowering and decrease in relative humidity and cumulative rainfall and increase in VPD (Table 3).

Climatic conditions can also influence the duration of the flowering period of invasive species (Castro-Díez et al. 2003; Yates and Broadhurst 2002). This study showed the strong influence of increasing temperature on accelerating development (i.e. the time to peak flowering was shortened), indicating that the rate of flowering development was accelerated in *A. longifolia* by warmer temperatures (Fig. 5). In this way, the peak flowering date of this invasive species could be predicted on the basis of time–temperature requirements with a specific base temperature (Roberts and Summerfield 1987). Our results suggest that to reach peak flowering, *A. longifolia* must experience a 231 °C day (thermal time) above 6.8 °C (base temperature) (Fig. 5).

In our study, *A. longifolia* plants under mesic conditions (north site) displayed a higher reproductive success (fruits density and fruits set) than those under xeric conditions (Fig. 6a, b). Recent findings indicate that an extended flowering period is a trait correlated with the invasiveness potential of exotic species, thus increasing their competitiveness for pollinators (Raine et al. 2007) and may help mitigate pollinator limitation (Gibson et al. 2011). We did not evaluate plant–insect relations in this study but it is well known that acacia flowers, including *A. longifolia*, do not require a highly

specific relationship with specific pollinators (Stone et al. 2003). As a consequence, *A. longifolia* flowers share many pollinators with co-existing native plant species. At our study plots, *A. longifolia* grows in species-rich assemblages that may potentiate competition for pollinators among native and invasive flowering species (Stone et al. 2003). Thus, we suggest a relationship between extended flowering period and pollination success, and, as a consequence, higher fruit set (reproductive success). Indeed, linear regression analysis demonstrated that the flowering duration can explain a significant amount of variation in the fruits set between xeric and mesic sites (Fig. 7). In addition, the percentage of fruits aborted was notably higher under xeric conditions (south site), which could limit invasiveness under typical drought climates (Fig. 6c). So, mesic conditions seem to favour fruit set and fruit development in *A. longifolia*.

Besides climate, the above-canopy structure limits and strongly influences understory plant species, by decreasing light intensity and temperature and increasing air humidity (Herrera 1997; Kilkenny and Galloway 2008; Newmark 2005). As pointed out in Table 1, the forested plot at south site had sparser trees and lower *P. pinaster* cover than the north forested plot. Under different canopy structures *A. longifolia* presented clear differences in reproductive investment and vegetative growth rate. This suggests that the characteristics of the north forested plot and the microclimatic conditions created by a denser *P. pinaster* canopy had an influence on *A. longifolia*'s flowering, providing a higher proportion of inflorescences with open flowers and extended blooming (Fig. 5). Moreover, the presence of a higher density pine forest at the north site was associated with a higher number of fruits set than in the open plot (Fig. 6a). On the other hand, we observed no significant differences between responses of *A. longifolia* under a more sparsely forested plot and an open plot at the south site (Fig. 6).

Our initial hypothesis, that mesic conditions (north site) confers an advantage in both vegetative and reproduction output fitness to *A. longifolia*, was only partially confirmed. Indeed, the similar RGR in mesic and xeric open plots was surprising (Fig. 3). Knowing that under Mediterranean conditions, drought limits plant growth, RGR would be expected to be lower under xeric conditions than under mesic conditions. However, the bibliography shows that *Acacia* species,

including *A. longifolia,* have a high photosynthetic capacity and exhibit a strong resource allocation and a constant allocation pattern under different conditions, and while some species reduce their water consumption in response to increasing temperatures and water stress, *A. longifolia* maintains high water consumption rates (Werner et al. 2009). The pine forest only influenced *A. longifolia* branch RGR at the north site, where it was lowest (Fig. 3). The growth of the latter species is limited by temperatures below 15 °C and less than 7.5 h exposure to sunlight per day (Milton and Moll 1982). In agreement, we suggest that *A. longifolia* responds with a decrease in branch RGR to more limiting light conditions (higher pine density and cover) and lower temperatures in the north forested plot. Physiological measurements on these plants showed lower carbon assimilation rates at the north forested plot, mainly due to lower photosynthetic photon flux density (Fernandes, unpublished data).

In conclusion, *A. longifolia* cannot maximize growth and reproduction across all environments, so the success of this invasive species is habitat-dependent. Like other *Acacia* species, *A. longifolia* is a nitrogen-fixing plant. This ability has enabled it to invade nutrient poor environments but the rate of invasion (related to reproductive success) is dependent on climate and plant community structure. This study shows that the combination of climate and forest structure caused pronounced differences in the reproductive success of *A. longifolia*. These data can help to understand the different invasive rates across *A. longifolia*'s geographical distribution in Portugal, and could explain its greater and more aggressive invasiveness in the north of Portugal, with mesic conditions and denser forests.

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