



# Using fitness parameters to evaluate three oilseed *Brassicaceae* species as potential oil crops in two contrasting environments

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## ABSTRACT

*Thlaspi arvense* and *Camelina sativa* have gained considerable attention as biofuel crops. But in some areas, these species, including *C. microcarpa*, are becoming rare arable plants because of agricultural intensification. Including them as crops could guarantee their conservation in agricultural systems. The fitness (*i.e.*, vegetative and reproductive growth) of two populations of *T. arvense*, Teruel (Spain) and Minnesota (USA), and one for each *C. sativa* (Minnesota) and *C. microcarpa* (Teruel) was studied under different climates (semi-arid Mediterranean and continental temperate) over two seasons. Fitness of all species and populations was highly affected by the final plant density and the climatic conditions at each site and season. *T. arvense* from Spain and *C. microcarpa* showed more constant equilibrium in resource distribution patterns than those species or populations with American origin. The density variation between species, populations and seasons, which varied between 4 and 500 pl/m<sup>2</sup>, indicate the importance of emergence for crop establishment of these species. Moreover, results showed that environmental conditions significantly affected crop development, *i.e.* plants were always higher in Spain than in the USA, indicating that mild winter sites may be better suited for their production than others. The establishment of *T. arvense* as a crop in a semi-arid region like that of North-eastern Spain is unlikely without irrigation, while *C. sativa* could be a good option as a rotational crop regardless of irrigation. In the case of *C. microcarpa*, although it could be cultivated, its final densities (hence germination and emergence) must be improved in order to establish it as a crop.

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## 1. Introduction

Field pennycress (*Thlaspi arvense* L.) and camelina (*Camelina sativa* L. Crantz) are worldwide weeds of great interest due to their high oilseed contents (Zanetti et al., 2013) and potential use as biofuel feedstocks (Moser et al., 2009), as well as other industrial applications such as paints and varnish production (Walsh, 2007). Furthermore, camelina has applications for food use (Abramovic and Abram, 2006). Both species' growth characteristics make them suitable as winter crops in Mediterranean climates (Zanetti et al., 2013), where they could be rotated with irrigated crops, as proposed for other species by Meza et al. (2008), or double cropped as they are in other countries (Gesch et al., 2014; Groeneveld and Klein, 2014; Johnson et al., 2015). *T. arvense* can also be used as a cover-crop to sequester excess soil nitrogen, reduce erosion, provide spring cover, and suppress other weeds (Dorn et al., 2013;

Johnson et al., 2015). Recently, considerable research has been done in North-America to introduce *C. sativa* and *T. arvense* as viable crops. *C. sativa* has been demonstrated to be a successful crop in semi-arid areas of the USA (Pavlista et al., 2016; Schillinger et al., 2012; McVay and Khan, 2011), and could be an alternative crop for other semi-arid regions in Europe. In this sense, their introduction as a rotational crop in semi-arid areas, such as in the Ebro basin in North-eastern Spain, could be used within an integrated crop management strategy for both oilseed production and weed control. The potential adaptation of these crop species to different sowing dates and grass herbicides could help control many weed species, some of which are herbicide resistant.

Conversely, in many European areas where *T. arvense* and *C. sativa* are not used as a crop, they are rare arable plants, understood arable plant as a species that can only maintain viable populations in crop fields, similar to *Camelina microcarpa* Andr. ex DC, which is as a wild relative of *C. sativa*. In North-eastern Spain, for example, high agricultural inputs and the establishment of conservation tillage systems have reduced their populations together with other arable species to such low levels that they have disappeared in

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many areas (Cirujeda et al., 2011). For this reason, the introduction of *T. arvense* and *Camelina* species as crops could guarantee the conservation of these rare arable plants in fields, which may lead to other benefits, such as increased crop diversity in agroecosystems. Moreover, these species can contribute to an increase of pollen and nectar resources to support the nutritional needs of pollinators (Eberle et al., 2015; Groeneveld and Klein, 2014), especially in late winter and early spring when few other flowers are available.

The fitness of a species is used to evaluate the effect of different environments on its growth and development (García et al., 2015). Performance traits in plants, which refer to vegetative biomass, reproductive output and survival (Violle et al., 2007), can reflect their suitability to be cropped in a certain environment, which affects the growth and the dormancy of their offspring (Hume, 1994). Therefore, the objective of this work was to compare the fitness of two wild populations of *T. arvense* originating from contrasting climates, i.e. Mediterranean climate (Teruel, Spain) versus a continental temperate climate (Morris, MN, USA), and one *C. sativa* and one *C. microcarpa* population, originating in Minnesota and Teruel, respectively. The ultimate goal was to use the fitness parameters measured to evaluate the suitability of these genotypes for potential oilseed production in the two contrasting environments, and furthermore, to determine the importance of the origin of the seeds for crop production. It was hypothesized that a given genotype (i.e., species/population) will perform best in the region (i.e., environment) of the origin of its seed. All populations were grown in both Lleida, Spain (semiarid Mediterranean climate) and Morris, USA (continental temperate climate) to compare their fitness and make evaluations.

## 2. Materials and methods

### 2.1. General set up and site descriptions

Seeds of *Thlaspi arvense* and *Camelina microcarpa* were harvested at maturity between June and July 2011 and 2012 in Camarillas (40°38'39"N-0°48'35"W, Teruel, Spain) and also in Morris (45°43'36"N-95°49'17"W, Minnesota, USA). *Camelina sativa* (cultivar CO46) was originally provided by the North Dakota State University Extension Service, Fargo, North Dakota, USA. However, seeds used for the study were produced by plants grown in Morris, Minnesota. Seeds were stored dry under laboratory conditions (23–25 °C) from harvest until sown for field studies. Batches of seeds of each Teruel and Morris population were interchanged, so that all populations were sown in both locations each season. Field trials were conducted from autumn to spring in two consecutive seasons (2011–12 and 2012–13) in a commercial cereal field in Almenar (41°46'36"N-0°32'7"E, Spain) and in an experimental field in Morris (as above).

In Morris, seeds were sown on 19 and 18 September respectively in 2011 and 2012, while due to climatic conditions, they were sown on 2 November in 2011 and on 4 October in 2012 in Almenar. Because populations used for the experiment are undomesticated (except *C. sativa*), and to ensure seedling emergence and plant material, seeds were sown at a rate of 1000 seeds per plot at 1 cm depth in 1 m<sup>2</sup> plots, with four replications. Sowing was performed in four lines per plot, each with 250 seeds, to facilitate hand weeding and to simulate crop row conditions. Organizing the seedlings in rows also reduced the potential for volunteer seedlings to enter into the study that might confound results. Emergence of seedlings was determined weekly until May and newly emerged seedlings were identified with coloured wires. Plots were not watered, so that emergence and growth of these species was conditioned mainly by climatic conditions. Both sites were free of *T. arvense* and *Camelina*

species before sowing and there was no historical evidence of their presence in the experimental plot areas.

After each season, in early June in Lleida and in mid-July in Morris, final plant density of each plot was estimated and values of fitness parameters were determined for 10 plants from each plot. At harvest in each experimental site, plants were cut from the soil surface and each plant transported in an individual paper bag to the laboratory where the following measurements were performed: height, vegetative biomass (VB), reproductive biomass (RB), reproductive effort (RE = RB/VB), number of branches per plant (Br/P), number of silicles per plant (Sl/P), seeds per silicles (Sd/Sl), seeds per plant (Sd/P) and the weight of 1000 seeds (only in 2012–13). VB and RB were measured after drying the plants in an oven at 70 °C for 24–48 h. Sd/Po is the mean of 10 randomly chosen silicles per plant. Sd/P was estimated by multiplying the number of silicles per plant and the number of seeds per silicle (= Sl/P × Sd/Sl). The weight of 1000 seeds was not measured in 2011–12 because plots were used in 2012–13 for another experiment and plants were removed before full maturity to avoid seed shedding.

### 2.2. Statistical analysis

For *T. arvense* differences in the final density and the fitness parameters between populations (Spanish vs USA), localities (Almenar vs Morris) and between seasons (2011–12 vs 2012–13) was performed with a three-way ANOVA, previous average of the ten values by repetition for each of the species. If interactions were found between factors, two-way ANOVA was performed to separate the respective factors. If new interactions were found, a new separation of factors was done. In the case of *Camelina*, because they were different species with different origin, no comparison was done between them. Instead, two-way ANOVA was performed in each species, comparing localities and seasons. To satisfy normality and homogeneity of variance, parameters were transformed by the function  $\arcsin(\sqrt{x/100})$  or  $\log(x+1)$  if needed. Statistical analyses were performed with JMPPro11 (SAS Campus Drive, Cary, NC USA). Linear regression analyses were performed between biological characteristics (VB vs. RB and fecundity) with the Sigma Plot program 11.0, using  $\log(x+1)$  transformations of the 40 collected plants in each species and population; a comparison of the slopes of the regressions between sites each season and between seasons each site was performed with JMPPro11 (SAS Campus Drive, Cary, NC USA).

### 2.3. Weather data

Daily rainfall and maximum and minimum air temperatures were obtained from a meteorological station situated 4 km away from Almenar, while in Morris, weather data were obtained from a standard meteorological station located at the experimental site.

## 3. Results

### 3.1. Climatic characteristics of Almenar and Morris

The two sites differed in terms of temperature and precipitation (Table 1). Season 2011–12 was dryer in both sites mainly during the winter period (December–March). It should be noted that all rainfall in Morris in autumn 2012–13 fell after 17 October. Season 2012–13 was colder than 2011–12 in both Almenar and Morris, and temperatures at Morris were also colder than in Almenar for approximately the same periods of time.

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