

## Evaluation of weed flora changes in Portugal in a 10 year basis

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### Abstract

The aim of this study was to assess the composition of weed communities starting from a rainfed farming system (1997), to the early period of transformation in agricultural systems with the adoption of irrigation (2007). The assessments were made within a 10-year interval and the floristic surveys were conducted in the same georeferenced plots. In 1997, the surveys were mostly performed in winter cereals (wheat, oat, barley), while in 2007, considering the same georeferenced plots, there were winter cereals and also irrigated olive groves, parcels of fallow, pasture and pine forest. Weed flora was determined by means of relative frequency, abundance and weed infestation degree. The effect of time was also evaluated by applying the methodology of variance analysis on the values of Shannon-Wiener Index. Canonical Correspondence Analysis was used to complement this information only for 2007 to know how the flora was distributed by the different cultures. Our results revealed that in both cases weed flora was of high diversity (229 and 264 species in 1997 and 2007, respectively), with the most representative families being always the same, namely Asteraceae, Poaceae and Fabaceae. The number of weeds that could be of concern for the farmers revealed to be relatively low. Our study confirmed that despite of the intensification of the agricultural production system, biodiversity increased over time. In both years, *Lolium rigidum* was present at high densities.

**Keywords:** Agroecosystems, weed infestation degree, *Lolium rigidum*, Mediterranean climate, plant biodiversity, multivariate analysis.

### Introduction

Out of a large number only a few hundred weed species are important in agro-ecosystems. The ability of these weeds to compete with crops for water, light and nutrients is among the most important factors which determine the potential reduction of crop productivity. The potential losses of crop production caused by weeds can reach a global average of 38% (Vidal, 2010). The mechanisms available to weeds include - but not limited to - seed dormancy of different duration, high seed germination rate, environmental plasticity, high seedling growth and reproductive capacity, short life cycle, self-compatibility, efficient and well organized methods of seed dispersal, allelopathy and tolerance to abiotic and biotic stresses (Baker, 1974). Furthermore, weeds are able to survive and grow in different ecological habitats, with their occurrence being very sensitive to changes in agricultural practices (Fried et al., 2010; Petit et al., 2011). New agricultural management practices lead to new balances and such information is considered to be of major importance (José-María et al., 2010; Sans et al., 2013). Ecological services that this type of vegetation provides to the agro-ecosystems are not negligible. Wild plants often protect soil against water and wind erosion, which is especially important on slopes, where many perennial crops are planted. Natural vegetation also protects soil from mechanical compaction, while many of the weeds have

medicinal and/or other value (Bilalis et al., 2014). In general, more intensive agriculture leads to lower biodiversity and reduces the environmental sustainability (Storkey and Westbury 2007; Stoate et al., 2009; José-María et al., 2010; Sans et al., 2013). Consequently, the maintenance of biodiversity in agro-ecosystems is considered to be of great importance in modern agriculture, based on economic, ecological and social parameters (Firbank et al., 2008). This kind of integrated weed management requires a detailed knowledge of the weed species (Johnson and Kent, 2002; Karar et al., 2005; Monteiro et al., 2012) and becomes relevant since it is one of the key steps for the development of sustainable weed management models.

The greatest European irrigation project, covering about 120000 ha of irrigated land is included into the *Alqueva* Multi-purpose Undertaking, located in south of Portugal, and has been accomplished in 2015 (EDIA, 2016). From its beginning in 2006, there were profound changes in traditional farming systems in the region, traditionally based in rainfed cereal crops and, consequently, there has been diversification of cultural farmer's options. These changes are also linked to the adoption of a more intensive production model, which impact on natural resources should be well known in order to ensure the sustainable use of these new irrigated areas. Therefore, a very important issue is to evaluate the effect of

time on composition of weed community, and its level of occurrence, in order to achieve an efficient weed management taking into account the ecological services that weeds render to the agro-ecosystems.

There are relatively few studies about weed flora based on long-term changes. Some of them are cited below and consider different periods of time and different perspectives which include changes in weed flora in arable fields (Table 1). The aim of this study was to assess the composition of weed communities and its occurrence in a 10-year interval considering 2007 the early stage of the transformations in agricultural systems that is taking place in south of Portugal. This study is the first of its kind carried out in Portugal, and it was conducted in order to evaluate the effect of time on weed species composition as the first part of a larger study which aims to study long-term changes (20-year interval). In the spring of 2017 new surveys will be done in the same geo-referenced places with the irrigation project in full operation.

## Results and discussion

### Weed surveys

Weed surveys were conducted in south Portugal - *Alentejo* (Fig. 1), in two years, 1997 and 2007. In 2007, about 10% of the studied sites started to have irrigated crops, especially olives.

### Weed vegetation composition and diversity

In 1997, 229 species were identified and distributed over 40 families, increasing their number in 2007, where 264 species were identified and distributed among 43 families. Our results are in accordance with previous studies conducted in the Mediterranean region, verifying always high weed diversity: 175 species in 138 surveys conducted by Cirujeda et al. (2011) in Spain and 278 species in 86 surveys in Greece (Damanakis, 1983). These numbers are significantly higher compared with those found in the north and central Europe (Holzner and Immonen, 1982; Chancellor and Froud-William, 1984; Sutcliffe and Kay 2000; Salonen et al., 2001). The botanical families with greater representation were the same in both periods, and also with similar representation (of about 50%): Asteraceae followed by Poaceae and Fabaceae. These families have been the most common ones in previous studies carried out in the central region of Portugal on vineyard (Monteiro et al., 2012) and maize (Calha et al., 2014).

In 1997, the following seven species were identified in more than 50% of the surveys: *Avena sterilis*, *Anagallis arvensis*, *Convolvulus arvensis*, *Lolium rigidum*, *Phalaris minor*, *Polygonum aviculare* and *Raphanus raphanistrum*. In 2007, only three species were identified, namely *Avena sterilis*, *Lolium rigidum* and *Phalaris minor*. These findings are similar to those observed for the most frequent species identified in Spain by Cirujeda et al. (2011), namely *L. rigidum* and *A. sterilis*. According to Ramôa et al. (2015), soil texture and soil phosphorus content had a significant effect on the species distribution. In particular, *L. rigidum* seems to be associated with medium soil texture and *A. sterilis* also showed a preference for medium and fine soil texture soils. The distribution of *P. minor* was not related to the soil texture but it showed preference for soils with medium phosphorus content.

As shown in Table 2, *L. rigidum* was the only species with a very high weed infestation degree in both study periods.

In 1997, a total of the following eight species showed a high level of weed infestation degree: *A. arvensis*, *Galium aparine*, *Lolium temulentum*, *Oxalis pes-caprae*, *P. minor*, *P. aviculare*, *Ranunculus trilobus* and *R. raphanistrum*. In 2007, the following 14 species showed high infestation degree: *Agrostis pourretii*, *A. sterilis*, *Carthamus lanatus*, *Chamaemelum mixtum*, *Crepis vesicaria*, *Cynodon dactylon*, *Bromus hordeaceus*, *Galium aparine*, *Hirschfeldia incana*, *Juncus bufonius*, *Leontodon taraxacoides*, *Stipa capensis*, *Trifolium campestre* and *Vulpia ciliata*. Regarding *L. rigidum*, it has been found in several other places as a very common and abundant species (Recasens et al., 1996; Cirujeda et al., 2011; Borger et al., 2012; Monteiro et al., 2012). Armengot et al. (2011) indicated that species like *L. rigidum* are closely linked to intensified agricultural practices. According to Sans et al. (2013), the high pressure of herbicides can lead to resistant species, reduction of the dicotyledonous and increase of the monocotyledonous species, such as *L. rigidum*, *A. sterilis* and *P. minor*. Another noteworthy finding is related to the small number of problematic species (which could be considered as weeds of agronomic importance), compared to the total number of identified species. This fact is in accordance with previous findings of Borger et al. (2012) in Australia.

### Trend over time

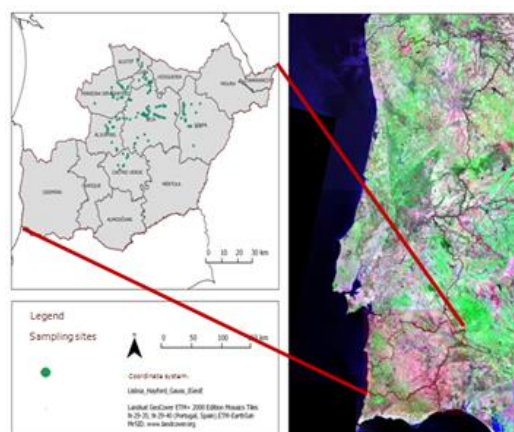
Table 3 shows the statistically significant increase of the Shannon index over time, which confirms the higher plant biodiversity in 2007 compared with 1997.

The CCA analysis shows the distribution pattern of biodiversity and its relationship to the different crops. The values of the inflation factor of variance (VIF), taking into consideration all the variables involved, are shown in Table 4. The results shown in Table 4 revealed no statistical correlations between the variable crops. The pine forest revealed a value very close to 1, indicating that it was an independent variable. Canonical Correspondence Analysis (CCA) revealed significant community differences among weed crop's species (Table 5 and Fig. 2). Nearly 10.5% of the species variability was explained by the different crops. The percentages of accumulated variation explained by the species and crops, from the first to the third CCA axes, were 34.4%, 51.9%, 66%, respectively and in all cases statistically significant. This means that the constrained ordination did not explain 34% of the remaining variability. Moreover, ANOVA test also indicated that the extraction of the first two axes revealed significant.

Based on Canonical Correspondence Analysis (Table 5), it was verified that both fallow and pasture flora were positively related to axis 1 while wheat, barley and oat had a negative related with this axis, indicating differences in flora composition. Between the factors that presented significant differences, pasture ( $p < 0.001$ ) had the largest weight ( $R^2 = 0.512$ ). These results revealed clear differences in the flora between rainfed cereals and fallow or pasture areas. In the case of oat there were also some significant differences and studied flora was significantly affected ( $R^2 = 0.236$ ). These differences may be attributed at differences in weed control. Oat has a reduced (or even absent) weed control comparing with wheat and barley (where herbicide application is common), once its production is devoted to livestock feed. In addition, in fallow and pasture areas, no weed control is

**Table 1.** Information about some weed flora studies based on long-term changes.

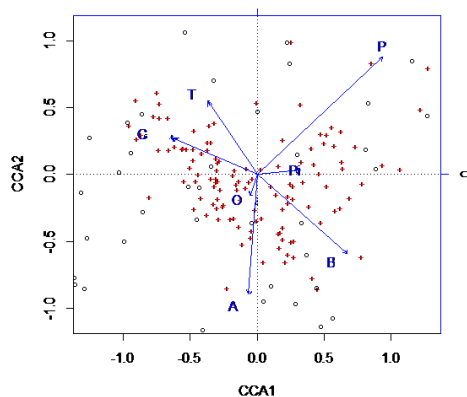
Reference	Year period	Country
Rademacher et al. (1970)	1956 - 1968	Germany
Chancellor (1985)	1969 - 1981	England
Sutcliffe and Kay (2000)	early 1960s - 1997	England
Mas and Verdú (2003)	1997 - 1998 and 200 - 2001	Spain
Lososová et al. (2004)	1954 - 2003	Czech Republic and Slovakia
Baessler and Klotz (2006)	1957 - 2000	Germany
Peter (2007)	1975 - 2004	Switzerland
Firbank et al. (2008)	An overview about pressures of agricultural change on biodiversity	British perspective
Hiltbrunner et al. (2008)	1999 - 2006	Switzerland
Roitman et al. (2008)	1991 - 2004	Brazil
Stoate et al. (2009)	An overview of the ecological status of agricultural systems	European Union: UK, The Netherlands, Boreal and Baltic countries, Portugal, Hungary and Romania,
Fried et al. (2009)	1970 - 2000	France
Andreasen and Streibig (2010)	1967 - 2007	Denmark
Potts et al. (2010)	1968 - 2005	England
Cirujeda et al. (2011)	1976 - 2007	Spain
Plaza et al. (2011)	1986 - 2008	Spain
Wang et al. (2011)	Prior to 1949 - 2010	Australia
Borger et al. (2012)	1997 - 2008	Australia
Salonen et al. (2012)	1997 - 2009	Finland
Fried et al. (2012)	1973, 1976 and 2003 - 2006	France
Kolářová et al. (2013)	2006 - 2008	Czech Republic.
Meyer et al. (2013)	1950s/60s - 2009	Germany
Alignier and Baudry (2015)	1995 - 2013	France



**Fig 1.** Location of the surveys in the Alentejo region – Portugal.

**Table 2.** Weed infestation degree.

Weed infestation degree	1997	2007
Very high	<i>Lolium rigidum</i>	<i>Lolium rigidum</i>
	<i>Anagallis arvensis</i>	<i>Agrostis pourretii</i>
	<i>Galium aparine</i>	<i>Avena sterilis</i>
	<i>Lolium temulentum</i>	<i>Carthamus lanatus</i>
	<i>Oxalis pes-caprae</i>	<i>Chamaemelum mixtum</i>
	<i>Phalaris minor</i>	<i>Crepis vesicaria</i>
	<i>Polygonum aviculare</i>	<i>Cynodon dactylon</i>
High	<i>Ranunculus trilobus</i>	<i>Bromus hordeaceus</i>
	<i>Raphanus raphanistrum</i>	<i>Galium aparine</i>
		<i>Hirschfeldia incana</i>
		<i>Juncus bufonius</i>
		<i>Leontodon taraxacoides</i>
		<i>Stipa capensis</i>
	<i>Trifolium campestre</i>	
	<i>Vulpia ciliata</i>	



**Fig 2.** Triplot graph of CCA. (P: pasture; B: fallow; Pi: pine forest; T: wheat; C: barley; A: oat; O: olive grove).

**Table 3.** Time effect on weed flora.

Time	Shannon-Wiener index*	Standard error	Tukey test	
1997	2.8700	0.0487	b	CV = 16.1%
2007	3.0257	0.0464	a	

\*mean values

**Table 4.** Values of the inflation factor of variance (VIF).

Year	T	A	C	B	O	P	Pi
2007	2.405	2.210	2.131	3.118	1.727	1.623	1.127

T: wheat; A: oat; C: barley; B: fallow; O: olive grove; P: pasture; Pi: pine forest.

**Table 5.** Results of CCA analysis.

	CCA <sub>1</sub> **	CCA <sub>2</sub> **	It	Ic	R <sup>2</sup>	F
Cumulative percentage of variance explained from species × crops (%)						
Wheat	-0.249	0.372			0.105	*
Oat	-0.048	-0.612			0.236	***
Barley	-0.443	0.193			0.116	**
Olive grove	-0.037	-0.115			0.010	ns
Fallow	0.460	-0.406			0.186	***
Pasture	0.643	0.602			0.512	***
Pine forest	0.221	0.024			0.028	ns

Level of significance: \*\*\* 0.001; \*\* 0.01; \* 0.05; ns not significant; CCA<sub>1</sub> and <sub>2</sub>: first and second, canonical axes.

**Table 6.** Criteria for assessing weed infestation degree.

Abundance coefficients	Relative frequency (%)		
	< 25	25 - 50	> 50
<b>1</b>	<b>0</b>	+	
<b>2</b>	+		
<b>3</b>		++	
<b>4</b>			
<b>5</b>		+++	++++

**0** very low; + low; ++ medium; +++ high; ++++ very high.

performed and consequently very diverse weed flora is present, probably because of the rich soil seed bank and the potential inclusion of leguminous species in sown pastures (Travlos, 2010). Olive groves and pine forest had no significant effect on weed diversity due to their low representation compared with the other crops.

Several studies have shown that the intensification of crops, due to the availability of irrigation, leads to a decrease in biodiversity (Storkey and Westbury, 2007; José-María et al., 2010; Plaza et al., 2011; Sans et al., 2013). However, in the present study, an increase of total plant diversity was observed. This could be attributed to the establishment of new and diverse crops and, the low percentage (10%) of irrigated crops in 2007. Besides, the fallow areas were significant in number, which may explain this fact, since they are particularly rich in weed flora (Robleño et al., 2013).

Sown pastures also revealed high floristic richness and abundance by having a large supply of seeds in the soil (Sans et al., 2013).

According to Piazza et al. (2011), it is necessary to conduct new studies on dynamics of vegetation using longer periods of time. This is justified by the high persistence of seeds in soil working as a population buffer against environmental changes (Légère et al., 2005).

## Materials and Methods

### Weed survey and data collection

This project began in 1997 with the objective of knowing the composition of weed communities for a better weed management in rainfed arable crops. At the same time,

*Alqueva* dam was in construction. So, in 1997, the plots were randomly selected taking into account only those with rainfed winter cereal crops and the surveys were made in the future zones of influence of the *Alqueva* Multi-purpose Undertaking basin. At that time, 100 floristic surveys were done, mainly in wheat but also in oat and barley plots. The surveyed plots were all referenced with GPS using the *Quantum GIS* version 1.8.0 program (2011) for setting them in the Administrative Official Map of Portugal - *Beja* district. Ten years later, in 2007, 105 floristic surveys were performed exactly in the same georeferenced places. We found plots with wheat, oat, barley and new ones containing irrigated olive groves, pine forest, fallow and pasture, recently established as a result of the new irrigated areas. The surveys were conducted on April and May, walking around the field in a random way until no new species were detected (Vasconcelos et al., 1999; Cirujeda et al., 2013).

Then, a list of presence and absence of species was created, in which the latin name and the EPPT code (European and Mediterranean Plant Protection Thesaurus) were added. Species' abundance was estimated as proposed by Barralis (1975, 1976) and also used by Monteiro et al. (2012), considering the following abundance coefficients: 1 (abundance less than 1 individual per m<sup>2</sup>), 2 (abundance 1-2 individuals per m<sup>2</sup>), 3 (abundance 3-20 individuals per m<sup>2</sup>), 4 (abundance 21-50 individuals per m<sup>2</sup>), 5 (abundance higher than 50 individuals per m<sup>2</sup>). Weed infestation degree due to the relative frequency and average abundance was determined, according to Monteiro et al. (2012) and is shown in Table 6. These procedures were identical for both years. Table 6 - Criteria for assessing weed infestation degree.

#### Data analysis

In order to assess changes in weed flora over time, the methodology of analysis of variance (ANOVA) was used to the values of Shannon-Wiener Index over time, according to a completely randomized outline. The differences between the average values of the studied factors (time and diversity) and their interaction were considered significant for a significance level of  $p < 0.05$ . Tukey's test for levels of significance  $p = 0.05$  was used in cases that the analysis of variance revealed a significant effect for the analyzed factors or their interaction, in order to identify differences between average values.

Multivariate analysis methodology and particularly Canonical Correspondence Analysis (CCA) was used to complement gathered information. For processing data R-Project for Statistical Computing software was used (R Development Core Team, 2011), with Vegan package (Oksanen et al., 2013). The methodology is described in general terms according to Kindt and Coe (2005) and Oksanen et al. (2013). Canonical Correspondence Analysis (CCA) was used only for the year 2007 to examine flora and the different crops. Before running the program, the species whose relative frequency values were  $\leq 10\%$  were removed from the original file. Only then, the CCA was performed, scaling 2 analyses with all the independent variables (types of crops and other forms of land use) to get the relation with weed flora. The values of the inflation factor of variance (VIF) were calculated in order to eliminate previously independent and highly correlated variables ( $VIF \geq 10$ ).

#### Conclusion

Our survey revealed that the main botanical families that have been maintained over time were the same, namely

Asteraceae, Poaceae and Fabaceae. The numbers of identified *taxa* were always very high, although most species showed low frequency and abundance values. Only a small group of species, in both years, showed levels of frequency and abundance that should concern the farmers. *Lolium rigidum* revealed to have very high degree of infestation in both periods of time. In 2007, weed flora was more abundant in species richness. Gradual changes in farming systems traditionally practiced in the region, driven by the introduction of irrigation are partially responsible for the observed differences. Moreover, new areas of olive groves which are associated with more intensive agriculture techniques and different pressure on the environment have probably created favorable conditions for the emergence of weeds. In addition, the long-term impact because of the EU policies regarding the implementation of the set-aside, subsidized crops (including areas of pasture and fallow) had a great expression in *Alentejo*. Fallows and pastures are particularly rich in weeds, which are integrated in certain areas as greening policies in order to increase biodiversity. Consequently, such surveys should be further conducted and progressive changes in weed flora like the ones observed in the present study ought to be taken into account for the development of an efficient and tailor-made weed management system.

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