

## Preventing botanical contamination risk of sunflower hybrid seed in the Valle Bonaerense del Río Colorado, Argentina

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### ABSTRACT

River Colorado Valley in southern Buenos Aires province (VBRC) is the main sunflower seed production area in Argentina, with over 8000 hectares, standing for 85% of the controlled fields over the country. The wild annual species *Helianthus annuus* and *H. petiolaris* naturalized in Argentina can pollinate crop plants giving fertile progeny. As in the US and in Europe, the presence of these species limits seed production because they represent a pollen contamination source. However, at present none of them have invaded the VBRC area. An ecological characterization was performed in order to assess the invasiveness of the VBRC. To test agro-ecological dissimilarity, habitats of both wild species in Argentina and the center of origin were compared using macro-abiotic (climatic and soil taxa), micro-abiotic (soil composition), and biotic (plant community) variables. Ecological conditions were found not to restrict the establishment of wild *Helianthus* invaders in the VBRC. An active alertness was initiated, comprising an exhaustive cleaning of machinery coming from other regions, roguing for off-type plants within seed production fields, to avoid annual *Helianthus* species culture for ornamental purposes, and removal of all feral forms of annual *Helianthus* within the protected region.

**Key words:** ferality – *Helianthus annuus* – *H. petiolaris* – invasiveness – pollination – wild sunflower.

### INTRODUCTION

The potential of the River Colorado Valley in southern Buenos Aires province (VBRC) for sunflower production has been known since the 1980s (Hernandez and Orioli, 1982; Rivas et al., 1987) however it was not profitable until some crop constraints - such as parakeet (*Cyanoliseus patagonus*) damage - were overcome through an increment of acreage and early sowing. The valley comprises ca. 90,000 hectares with gravitational watering devoted to onion and forage crops. Sunflower seed production comprises 10% of valley acreage that constitutes the main sunflower seed production area in Argentina, this being more than 85% of the controlled fields over the country (INASE, 2007).

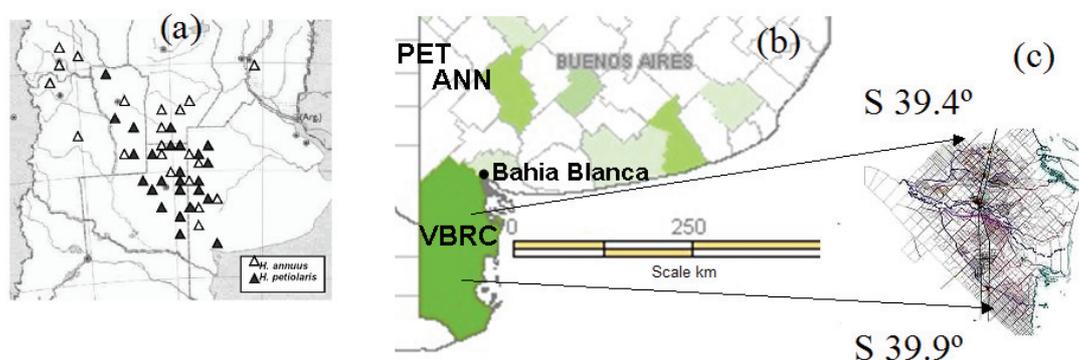
Two wild annual species, *Helianthus annuus* and *H. petiolaris* have established themselves in Argentina on a transitional area between the Pampean steppe and Espinal phyto-geographical provinces (Burkart et al., 1999). *H. petiolaris* invaded sandy soils, usually degraded by wind erosion, whereas wild *H. annuus* has spread on more fine-textured soils, with hydric constraints such as flooding and salinity (Cantamutto et al., 2008). Their migration followed land moving routes, probably facilitated by transportation and machinery although the location of *H. annuus* populations in the irrigated areas of Mendoza and San Juan (Poverene et al., 2002) seems to demonstrate that seeds have moved through the irrigation channels. Stable populations of both species were located 200 km north from the VBRC (Fig. 1) but isolated plants were frequently observed in the surroundings of Bahía Blanca harbor complex, at 100 km of distance from the valley (unpublished data).

The annual *Helianthus* species established in Argentina can pollinate crop plants giving fertile progeny (Poverene et al., 2004; Ureta et al., 2008). *H. annuus* (Deines et al., 2004; Rosales-Robles et al., 2005) and *H. petiolaris* (Grichar et al., 2004) are also crop weeds. In the US, the presence of these species in roadsides, slopes, and fireguards limits seed production because they are a pollen contamination source (Anfinrud, 1997). Wild contaminant and off-type seed imported from invaded areas has been suggested as being the origin of European wild sunflower populations (Bervillé et al., 2005; Vischi et al., 2006; Muller et al., 2007) and a study of their expansion trend has been recommended (Stankovic-Kalezic et al., 2007).

The use of beehives to improve seed production is frequent in the VBRC, though this practice represents a threat to seed purity because bees often visit wild flowers in the vicinity, outside crop fields (Andrada et al., 2004). Given that most inbred lines used for commercial hybrids production flower at the same time as wild invaders, undesired pollination risk in the VBRC would be very high.

There are a few known cases where it has been possible to anticipate and avoid a biological invasion. A proper environmental management is proposed herein with prevention rules in order to characterize the VBRC and prevent the establishment of the two annual *Helianthus* species already present in other Argentine regions. This would place a severe constraint on sunflower seed production, threatened by wild sunflower gene flow. It would not only affect seed inspection by increasing seed lots and field rejection, but also would be a risk for other regions in the country not yet invaded, due to contaminated sunflower hybrid seed usage, which could originate new feral populations.

To test agro-ecological dissimilarity, habitats of both wild species in Argentina and the center of origin were compared using three analysis scales. If available habitats in the VBRC measured by each scale were different to those colonized habitats in other regions, the null hypothesis of similar environment would be rejected, and there would be no risk of invasion. If not, it would have to be accepted that the two wild *Helianthus* species could invade the VBRC and the employment of prevention rules would be worthwhile.



**Fig. 1.** Distribution (a) and nearest stable populations of *Helianthus petiolaris* (PET) and wild *H. annuus* (ANN) (b) with respect to the VBRC region (c). Shaded counties in Buenos Aires province are devoted to sunflower seed production in Argentina (Maps from Poverene et al., 2002, INASE and CORFO).

## MATERIALS AND METHODS

Argentinean habitats were characterized through a representative sample of nine *H. annuus* and 13 *H. petiolaris* stable populations, geo-referenced by Poverene et al. (2002). The macro-abiotic variables altitude, annual rainfall, hottest month mean temperature, coldest month mean temperature, and soil taxa were obtained from De Fina (1992) and INTA (1990). Micro-abiotic conditions were estimated by soil composition within each habitat, according to methods described in Cantamutto et al. (2008). Plant community was characterized through a semi-quantitative technique in February 2007. Data were collected in each collection site from 10 points on a uniformly spaced grid coordinate system. At each grid point (a 2 m<sup>2</sup> circle), abundance was visually qualified from 0 (absent) to 5 (very abundant).

Climate variables in the center of origin corresponded to 49 *H. annuus* and 19 *H. petiolaris* populations registered in the GRIN-USDA ([www.ars-grin.gov2/cgi-bin/npgs/html](http://www.ars-grin.gov2/cgi-bin/npgs/html)) representative of the different states where germplasm has been collected in the Northern Hemisphere. Climate parameters were taken from the [www.worldclimate.com](http://www.worldclimate.com) site, for series of more than 20 years obtained from meteorological stations located  $4.3 \pm 3.9$  km and  $8.4 \pm 6.5$  km far away from *H. annuus* and *H. petiolaris* populations, respectively.

Macro-abiotic climate variables for VBRC were estimated in four localities (Mayor Buratovich, Pedro Luro, Hilario Ascasubi, and Villalonga) using De Fina (1992). Parameters of soil taxa and soil composition of representative cartographic units were taken from INTA (1990). Plant community on roadsides was estimated as previously described, in 10 points over a transect between Mayor Buratovich and Villalonga (ca. 60 km) in March 2007.

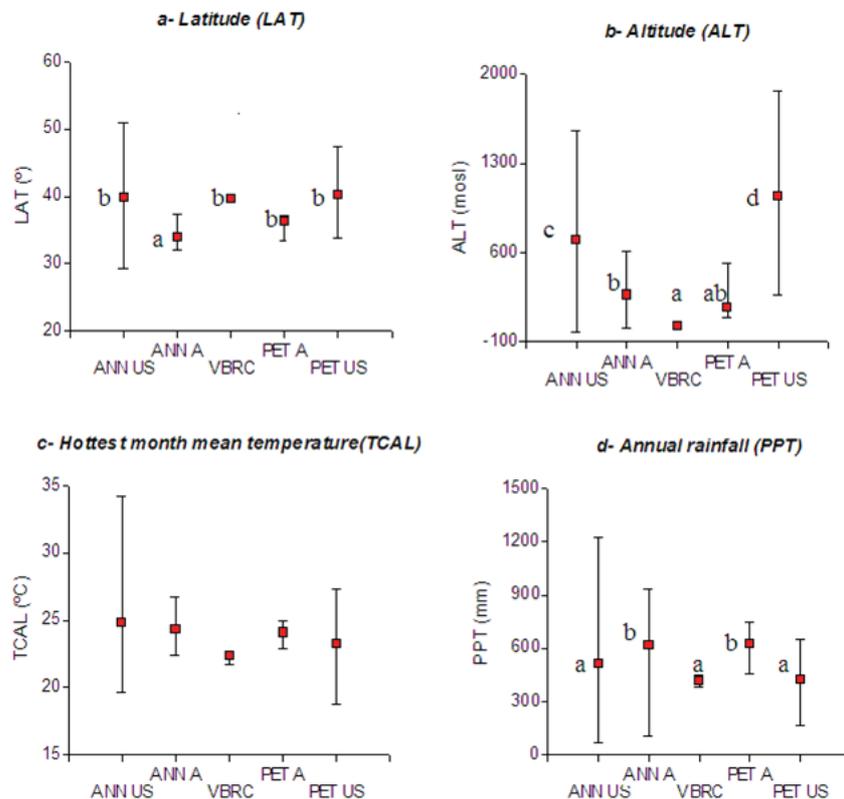
On the macro and micro-abiotic scales, variables were compared through the non-parametric Kruskal-Wallis test and graphical analysis. Soil similarity was estimated through the frequency of *H. annuus* and *H. petiolaris* population occurrence in soil units (taxa) also present in the VBRC. On the biotic scale, plant community of Argentine habitats where *H. petiolaris* and *H. annuus* have established

themselves, was compared to that of the VBRC through contingency tables. All the analyses were performed with the InfoStat (2002) statistics package.

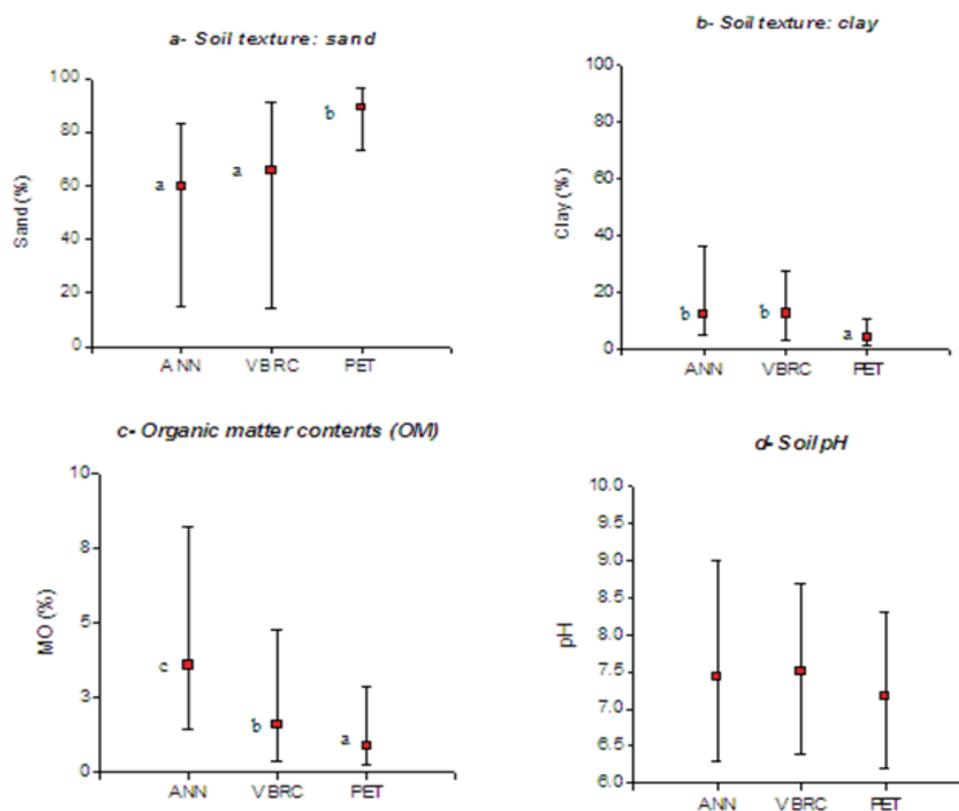
## RESULTS AND DISCUSSION

Macro-environment geographical and climate conditions would not be a restrictive factor for the establishment of wild *Helianthus* invaders in the VBRC. The valley is located at a latitude that almost corresponds to the mean value of the latitudinal distribution range in the US. The VBRC latitude did not differ from those recorded for both species in the center of origin, but it was different from *H. annuus* in Argentina (Fig. 2) probably because the invasion began in the northern part of the country, though it progressed towards the South (Cantamutto et al., 2008). The climate variables did not differentiate the habitats of VBRC and the US either. Although the habitat altitude is different for *H. annuus* environments - given that in the US this species grows even below sea level - the VBRC does not seem to have any altitude restriction.

Soil cartographic units did not show differences on a macro-abiotic scale. There are 15 soil groups in the VBRC, among which Mollisols and Entisols predominate. These orders were found to be closely associated with both annual *Helianthus* populations in Argentina and in the US (Cantamutto et al., 2008). More than one fifth of *H. annuus* and *H. petiolaris* populations in Argentina were associated with soil taxa present in the VBRC. Given that soil taxonomy can be used as an ecosystem processes index (Mann et al., 1999; Bouma, 2003) the taxa associated with Argentinian populations would show that soil genesis processes taking place in the VBRC are similar to those in other invaded regions elsewhere in the country.



**Fig. 2.** Geographical and climatic distribution (mean, range) of *Helianthus annuus* (ANN) and *H. petiolaris* (PET) populations in North America (US) and Argentina (A) compared to that observed in River Colorado Valley in Buenos Aires province (VBRC). Means significantly different are followed by different letters.



**Fig. 3.** Soil physical and chemical composition in *Helianthus annuus* (ANN) and *H. petiolaris* (PET) habitats in Argentina (mean, range) compared to those observed in River Colorado Valley (VBRC) using Kruskal-Wallis test. Means significantly different are followed by different letters.

At a micro-abiotic level, soil physical and chemical composition in the VBRC did not show differences with other annual *Helianthus* micro-habitats in Argentina, except for organic matter content (Fig. 3). However, a canonical discriminant analysis for these parameters showed that 10 out of 22 representative soils in the VBRC corresponded to those where the two invaders had previously established themselves in Argentina (Cantamutto et al., 2007).

On a biotic scale, no differences were found in plant community composition accompanying annual *Helianthus* species in Argentina and the sampled sites in the VBRC. More than two thirds of plants species considered as invaders found in the VBRC were associated with both annual *Helianthus* in their habitats in Argentina (Table 1). The high association with cosmopolite species would point to the invasion capacity of the VBRC by the two annual *Helianthus* species.

According to our results, the hypothesis of environmental similarity between the VBRC and the *H. annuus* and *H. petiolaris* habitats in Argentina and the US could not be discarded. As a consequence, the valley would be vulnerable to the invasion of both species.

The obtained data would be useful for helping set guidelines for commercial seed production. In order to derive recommendations for handling ecological aspects of invasion risks, the analysis results were disseminated to 24 of the main seed companies responsible for sunflower hybrid seed production in the VBRC, and to centres involved in science and technology within the region. An active alertness was initiated, comprising an exhaustive cleaning of machinery coming from other regions, roguing for off-type plants within seed production fields, to avoid annual *Helianthus* species culture for ornamental purposes, and removal of all feral forms of annual *Helianthus* within the protected region.

**Table 1.** Plant community main species found in disturbed lands at roadsides and channels in the River Colorado Valley (VBRC) also present in wild *Helianthus annuus* and *H. petiolaris* Argentine habitats, grouped by botanic family.

| Plant species  | Habit <sup>1</sup> | Origin <sup>2</sup> | Distribution % <sup>3</sup> |
|--|--------------------|---------------------|-----------------------------|
| Asteraceae   |                    |                     |                             |
| <i>Centaurea solstitialis</i> (L.)                               | A                  | E                   | 50                          |
| <i>Cichorium intybus</i> (L.)                                    | P                  | E                   | 50                          |
| <i>Solidago chilensis</i> (DC.) Baker                            | P                  | N                   | 88                          |
| <i>Hyalis argentea</i> D. Don ex Hook. & Arn.                    | P                  | N                   | 30                          |
| <i>Verbesina encelioides</i> (Cav.) Benth. & Hook. f. ex A. Gray | A                  | N                   | 83                          |
| <i>Xanthium spinosum</i> (L.)                                    | A                  | N                   | 88                          |
| Brassicaceae   |                    |                     |                             |
| <i>Diplotaxis tenuifolia</i> (L.) DC.                            | P                  | E, A                | 54                          |
| Chenopodiaceae   |                    |                     |                             |
| <i>Kochia scoparia</i> (L.) Schard.                              | A                  | E, A                | 50                          |
| <i>Salsola kali</i> L.   | A                  | E, A                | 20                          |
| <i>Chenopodium album</i> (L.)                                    | A                  | E                   | 80                          |
| Convolvulaceae   |                    |                     |                             |
| <i>Convolvulus arvensis</i> (L.)                                 | P                  | E                   | 58                          |
| Fabaceae   |                    |                     |                             |
| <i>Melilotus albus</i> Desr.                                     | B                  | E                   | 75                          |
| Poaceae  |                    |                     |                             |
| <i>Agropyron elongatum</i> (Host) P. Beauv.                      | P                  | E                   | nd                          |
| <i>Cynodon dactylon</i> (L.) Pers.                               | P                  | A                   | 88                          |
| <i>Distichlis spicata</i> (L.) Greene                            | A                  | N                   | 80                          |
| <i>Sorghum halepensis</i> (L.) Pers.                             | P                  | E, A                | 54                          |
| <i>Cenchrus pauciflorus</i> (Benth)                              | A                  | N                   | 63                          |
| <i>Eleusine indica</i> (L.) Gaertn                               | A                  | A                   | 63                          |
| <i>Eragrostis curvula</i> (Schrad.) Nees                         | P                  | F                   | 33                          |
| Solanaceae   |                    |                     |                             |
| <i>Solanum elaeagnifolium</i> (Ort.) Dun.                        | P                  | N                   | 85                          |
| Zygophyllaceae   |                    |                     |                             |
| <i>Tribulus terrestris</i> (L.)                                  | A                  | E                   | 63                          |

<sup>1</sup>A = annual; B = biannual and P = perennial.

<sup>2</sup>E = Europe; A = Asia; F = Africa; N = Native or America

<sup>3</sup>Proportion of invaded provinces in Argentina, nd = not determined

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