

**RESUMEN**

El girasol silvestre *Helianthus annuus* ssp. *annuus* es originario de América del Norte y se ha naturalizado en la Argentina estableciéndose actualmente en siete provincias. Constituye un germoplasma adaptado a condiciones locales, útil para programas de conservación y de mejoramiento del cultivo. Comparte su territorio con el girasol cultivado y con otro silvestre, *H. petiolaris*, con los que experimenta procesos de flujo génico. Esta tesis propone caracterizar las poblaciones silvestres locales mediante diferentes herramientas moleculares. Se espera conocer la estructura genética de las poblaciones silvestres y obtener información que permita interpretar el proceso de colonización y la interacción cultivo-silvestre. Diez poblaciones y seis líneas cultivadas se estudiaron utilizando secuencias entre repeticiones de secuencia simple (ISSR) y repeticiones de secuencia simple (SSR). Se obtuvieron bandas y alelos únicos del silvestre, ninguno del cultivo. Las poblaciones mostraron una variabilidad molecular 60% mayor que el cultivo. El silvestre local mantuvo aproximadamente 70% de la diversidad observada en el centro de origen, sugiriendo un significativo potencial para la adaptación. Se encontró que 20% de la variabilidad genética total reside entre poblaciones. La baja correlación entre la distancia genética y geográfica indicaría múltiples eventos de introducción del silvestre en la Argentina y/o nuevas recombinaciones. La baja correlación entre ISSR y SSR a nivel de individuo y población descarta la existencia de grupos evolutivos divergentes. Las poblaciones variaron en sus niveles de diversidad, pudiéndose determinar los sitios que reúnen los valores más altos en las medidas estimadas y que por tanto podrían ser objeto de evaluaciones en caracteres agronómicos de interés. Se compararon medidas de distancias calculadas con los coeficientes de similitud de Jaccard, Sorensen-Dice, Ochiai, Anderberg, Simple Matching, Rogers y Tanimoto, Ochiai II y Russel y Rao. Los siguientes grupos llevan a resultados similares en cuanto a las relaciones entre individuos, tanto para ISSR como SSR: i) Jaccard, Sorensen-Dice y Ochiai; ii) Simple Matching, Rogers y Tanimoto. Ambos grupos presentaron una alta correlación. Para un análisis global de los resultados obtenidos con ISSR y SSR se definió una nueva distancia basada en relaciones geométricas entre las distancias originales que resultó apropiada para agrupar matrices que presentan

baja correlación. El análisis con nueve genes asociados a respuestas a estrés biótico y abiótico en seis poblaciones silvestres indicó que las mismas presentan mayor variabilidad que el cultivado, conteniendo numerosos alelos únicos. No se encontraron alelos únicos del cultivo. Los niveles de diversidad genética fueron similares entre loci génicos y SSR. Las poblaciones presentaron estructura genética y alelos únicos de cada población. La elevada diversidad alélica encontrada para genes relacionados con la defensa frente a patógenos y a resistencia a sequia y salinidad podría incluir variantes agronómicamente ventajosas. Se estudiaron plantas androestériles de una población silvestre mediante marcadores diseñados en base a secuencias mitocondriales; loci SSR nucleares ligados al carácter y cruza con líneas cultivadas. Se encontraron plantas androestériles tipo CMS PET1 y otras con nuevos mecanismos. La nueva forma de androesterilidad fue estable e incluyó genes restauradores. Se hallaron genes de restauración para CMS-PET1 diferentes de *Rfl*. Estos materiales son útiles para incrementar la diversidad genética del cultivo. El hallazgo de plantas silvestres con CMS PET1 sugiere flujo génico desde el cultivo *vía* semilla. Las poblaciones silvestres locales conforman un sistema dinámico, enriquecido por las interacciones con el cultivo y los silvestres emparentados. Aunque la introducción en nuestro país es reciente, el sistema representa una oportunidad de estudio del proceso de evolución en condiciones diferentes a las del centro de origen.

**ABSTRACT**

The wild sunflower *Helianthus annuus* ssp. *annuus* is native to North America and has become naturalized in Argentina and established currently in seven provinces. It constitutes a germplasm adapted to local conditions, useful for conservation programs and crop improvement. It shares its territory with the cultivated sunflower and other wild species, *H. petiolaris*. Gene flow and hybridization occur among them. The aim of this thesis was to characterize the local wild populations by different molecular markers. We expect to assess the wild genetic variability and to obtain information for understand the colonization process and the crop-wild interaction. Ten populations and six cultivated lines were studied using inter simple sequences repeats (ISSR) and simple sequence repeats (SSR). Unique bands or alleles were obtained for the wild but no crop private alleles were found. Molecular variability in wild populations was approximately 60% higher than in inbred lines. Local wild sunflowers kept considerable diversity levels in comparison with populations in the centre of origin (approximately 70%) and therefore they might possess a potential for adaptive evolutionary change. We found 20% of genetic variation among populations. The low correlation between genetic and geographic distance may indicate multiple independent introductions of *H. annuus* in Argentina or the occurrence or novel genetic recombination. The low correlation between ISSR and SSR markers at the individual and population levels rule out the existence of divergent evolutionary groups. Markers analysis allowed to identify populations with highest genetic variability. Distance measures calculated from the similarity coefficients of Jaccard, Sorensen-Dice, Ochiai, Anderberg, Simple Matching, Rogers and Tanimoto, Ochiai II and Russel and Rao were compared. The following groups showed a similar relations between individuals, for both ISSR and SSR: i) Jaccard, Sorensen-Dice and Ochiai ii) Simple Matching, Rogers and Tanimoto. Both groups were highly correlated. For overall analysis of the results obtained with ISSR and SSR a new distance was defined that showed to be appropriate to combine matrices with low correlation. The analysis of nine genes associated to biotic and abiotic stress on six wild populations indicated that they have a higher variability than the

cultivated lines, containing many unique alleles, but none unique alleles were found in the crop. The levels of genetic diversity were similar between gene loci and SSR. The populations presented genetic differences and unique alleles for each population were found. The high allelic diversity for genes related to pathogens defense and resistance to drought and salinity could involved superior phenotypes. We studied male-sterile plants from one wild population using markers designed from mitochondrial sequences, SSR nuclear loci linked to the character and crosses with cultivated lines. Male-sterile plants CMS-PET1 type and plants with no described mechanisms were found. The new form of male sterility was stable and included restorer genes. Restore genes CMS-PET1 different from *Rfl* were found. These materials are useful for increasing the genetic diversity of the crop. Finding wild plants with CMS PET1 suggests gene flow from the crop to the wild via seed. Local wild populations are a dynamic system, enriched by interactions with the crop and wild related. Although the introduction in our country is recent, the system represents an opportunity for study the evolution process in different conditions of the centre of origin.

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