Evaluation of Achenes of *Helianthus verticillatus*, an Endemic Sunflower of the Southeast US for Oil Concentration and Fatty Acid Composition

Gerald J. Seiler* and Tom J. Gulya

USDA-ARS, Northern Crop Science Laboratory, Fargo, ND

Abstract

The narrow genetic base of cultivated sunflower has been broadened by the infusion of genes from the wild species which have provided a continued source of desirable agronomic traits. The genus *Helianthus* comprises 51 species, 14 annual and 37 perennial, all native to North America. Whorled sunflower, *Helianthus verticillatus* Small, is a diploid perennial species first collected in western Tennessee in 1898 and not seen again until it was rediscovered a century later in 1998 near the original site. It occurs in low moist prairie soils in Alabama, Georgia, and Tennessee. Unfortunately, no achenes of this rediscovered species were previously available for evaluation for oil concentration and fatty acid composition for sunflower improvement. The objective of this study was to collect achenes from as many populations of *Helianthus verticillatus* as possible for the USDA-ARS, National Plant Germplasm System wild sunflower germplasm collection and evaluate the achenes for oil concentration and fatty acid composition. The exploration covered 1200 miles in the states of Alabama, Georgia, and Tennessee during October, 2003. Two populations were collected from Georgia and Tennessee with population size varying from 100 to 250 plants. These collections represent the first available populations in the wild sunflower germplasm collection housed at the USDA-ARS North Central Regional Plant Introduction Station, Ames, Iowa. The achene oil concentration averaged 323 g/kg, while the linoleic fatty acid concentration was 749 g/kg, significantly higher than normally observed for populations of wild sunflower growing at southern latitudes. This is the first report of oil concentration and fatty acid composition for this species. The collection of populations of whorled sunflower will assure their preservation in the wild sunflower genebank and their use for future improvement of cultivated sunflower.

Introduction

Sunflower (*Helianthus annuus* L.) oil has the potential to be improved for industrial and nutritional purposes through selection and breeding. The narrow genetic base of cultivated sunflower has been broadened by the infusion of genes from wild species resulting in a continuous improvement of agronomic and economic traits in cultivated sunflower (Jan and Seiler, 2007). The genus *Helianthus* consists of 51 species and 19 subspecies with 14 annual and 37 perennial species (Schilling, 2006). While a few populations of some wild sunflower species have been collected and evaluated for oil concentration and fatty acid composition, many remain to be evaluated to fully characterize the available genetic diversity. There is an urgent need to collect species that are endemic to limited geographic areas that may be at risk of being eliminated by the activities of man. One of these species is diploid perennial whorled sunflower, *H. verticillatus* Small, which was described over 100 years ago (Small, 1898), but was not rediscovered or recollected until recently in western Tennessee (Mathews et al., 2002). It occurs in low wet prairie openings, moist roadsides, and edges of woods where the limestone soil is deep and poorly drained in Georgia, Alabama, and Tennessee. Recent emphasis on the fatty acid composition of sunflower achenes has increased interest in using wild species in breeding programs to enhance oleic or linoleic acid, or to reduce saturated fatty acids, especially palmitic and stearic acids, but information about oil concentration and fatty acid composition is lacking for a number of rare and threatened species. The objective of the study was to evaluate achene oil of *H. verticillatus* populations for composition of four major fatty acids: palmitic (16:0), stearic (18:0), oleic (18:1), and linoleic (18:2).

Materials and Methods

Plant materials

Populations of wild sunflowers were collected between 17 and 28 October, 2003. The expedition covered a distance of 1,200 miles in three states: Alabama, Georgia, and Tennessee. Details of this exploration can be found in Gulya et al. (2007). Heads of the wild sunflowers were collected from 50 to 250 plants within each population and bulked into a single sample. Each sample represented an isolated, open-pollinated segregating population. Specific site information obtained from local botanists, generalized distribution maps, and federal and state agencies was used to locate populations. Population size (number and extent), GPS coordinates, elevation, habitat, soil type, soil sample, seed set per head, and the presence of diseases, insects, and other wild sunflower species were recorded for each population. Achene samples were sent to the USDA-ARS North Central Regional Plant Introduction Station, Ames, Iowa, where they are maintained and distributed. All populations were collected from the broad distributional range of the species.

Oil and fatty acids analysis

Achenes were stored at 5°C and low humidity (<20%) until analyzed for fatty acid composition (expressed as g/kg on a dry weight basis). Oil concentration was analyzed using nuclear magnetic resonance. Fatty acid composition was determined using a gas chromatograph on oil extracted from a 10-achene sample. The extracted oil was converted to methyl esters using an organic-catalyzed transesterification method. Fatty acids analyzed included the following acids: palmitic (16:0), stearic (18:0), oleic (18:1), and linoleic (18:2). Fatty acid composition was determined from two samples per population.

Results and Discussion

Two populations of *H. verticillatus*, one from Tennessee and one from Alabama were collected (Table 1 and Figure 1). The general habitat and phenotype of *H.* verticillatus is shown in Figures 2-4. Oil concentration in the achenes averaged 323 g/kg (Table 2). This was the first report of oil concentration for this species. Wild perennial *Helianthus* populations generally have oil concentrations of 250 to 350 g/kg (Seiler, 1985), which is much lower than cultivated sunflower with 440-480 g/kg (Robertson et al., 1979).

The average oleic acid concentration was 138 g/kg in *H. verticillatus* (Table 2). This was accompanied by a high linoleic acid concentration of 749 g/kg. The fact that both populations of *H. verticillatus* had linoleic concentrations > 720 g/kg indicates that this trait should have a genetic basis because it is relatively stable in the different populations over a range of environments. High linoleic sunflower oil with >700 g/kg is preferred for the production of soft margarine (DeHaro and Fernandez-Martinez, 1991).

The linoleic fatty acid concentration observed in the *H. verticillatus* populations was unusually high for southern latitudes. High temperatures during flowering, achene filling, and maturation favor a low linoleic acid concentration and a high oleic acid concentration (Seiler, 1986). Generally, the cooler northern latitudes have higher concentrations of linoleic acid in the oil than the warmer southern latitudes (DeHaro and Fernandez-Martinez, 1991). A lower concentration of 500 g/kg of linoleic acid is more typical of the concentration expected in warmer southern latitudes.

The concentration of palmitic and stearic acids in *H. verticillatus* averaged 57 g/kg and 26 g/kg, respectively. This is a reduction of 30% compared to these fatty acids in commercial sunflower oil. The lower saturated fatty acid profile of *H. verticillatus* has the potential to reduce saturated fatty acids in commercial sunflower oil.

Table 1. Location of *H. verticillatus* populations collected in Tennessee and Alabama.

Site no.	Species	Collection Date	GPS – N	GPS- W	Alt (ft.)	State	County
2391	*H. verticillatus*	10/18/03	35.48489	88.71247	440	TN	Madison
2425	*H. verticillatus*	10/26/03	34.12869	85.46075	699	AL	Cherokee

Table 2. Oil concentration and fatty acid composition of *Helianthus verticillatus* populations collected from Tennessee and Alabama.

Population (Location)	Oil content	Fatty acid composition			
		Palmitic (16:0)	Stearic (18:0)	Oleic (18:1)	Linoleic (18:2)
	g/kg				
2391 (TN)	346.0	55.1	24.9	153.6	733.0
2425 (AL)	301.5	58.1	26.7	123.0	766.5
Mean±SE	323.4±12.9	56.6±1.1	25.8±0.6	138.3±9.0	749.3±9.8

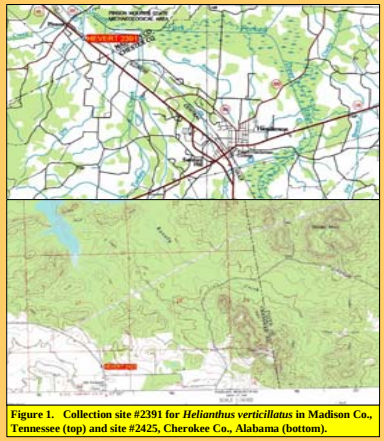

Figure 1. Collection site #2391 for *Helianthus verticillatus* in Madison Co., Tennessee (top) and site #2425, Cherokee Co., Alabama (bottom).

Figure 2. Developmental stages of *H. verticillatus*: flower (left) and vegetative plant (right).

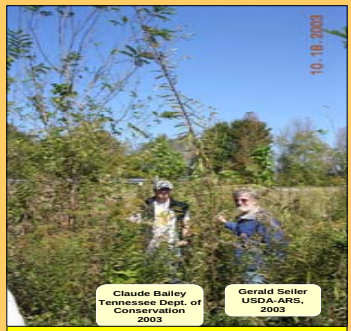

Figure 3. *Helianthus verticillatus* is very tall, up to 12 feet (left, center) dwarfing Claude Bailey (left), Tennessee Department of Conservation and Gerald Seiler (right), USDA-ARS, Fargo, North Dakota in a moist disturbed roadside ditch near a stream at Site #2391, Madison Co., TN.

Figure 4. *Helianthus verticillatus* site #2425 post-flowering along edge of woods in a moist roadside ditch, Cherokee Co., Alabama.

Conclusions

The average oil concentration of 323 g/kg is on the high end for a wild perennial sunflower species. The average linoleic acid concentration of 749 g/kg is higher than expected for populations grown in southern latitudes. The concentration of palmitic and stearic acids in *H. verticillatus* averaged 57 g/kg and 26 g/kg, respectively, which is a reduction of 30% compared to commercial sunflower oil. The lower saturated fatty acid profile has the potential to reduce saturated fatty acids in commercial sunflower oil. High linoleic acid concentration will provide a source for increasing this fatty acid in sunflower oil. Further research will be needed to determine the inheritance of the fatty acid composition and oil concentration.

References

DeHaro, A., and J. Fernandez-Martinez. 1991. Evaluation of wild sunflower (*Helianthus*) species for high content and stability of linoleic acid in seed oil. J. Agric. Sci. 116:359-367.

Gulya, T.J., G.J. Seiler, G. Kong, and L.F. Marek. 2007. Exploration and collection of rare *Helianthus* species from the southeastern United States. Helia 30(46):13-24.

Jan, C.C., and G.J. Seiler. 2007. Sunflower. p. 103-165. In: R.J. Singh (ed.), Genetic Resources, Chromosome Engineering, and Crop Improvement. Vol. 4, Oilseed Crops, CRC Press, NY.

Mathews, J.F., J.R. Allison, R.T. Ware, C. Nordman, C. 2002. *Helianthus verticillatus* Small rediscovered and redescribed. Castanea 67:13-24.

Robertson, J.A., W.H. Morrison, R.L.Wilson. 1979. Effects of planting location and temperature on oil content and fatty acid composition of sunflower seeds. USDA, Agricultural Research Services Results, Southern Series No. 3, Washington, D.C.

Schilling, E. E. 2006. *Helianthus.* In: Flora of North America Editorial Committee (eds.), Flora of North America North of Mexico. New York and Oxford. Vol. 21, pp. 141-169.

Seiler, G. J. 1985. Evaluation of seeds of sunflower species for several chemical and morphological characteristics. Crop Sci. 25:183-187.

Seiler, G.J. 1986. Analysis of the relationships of environmental factors with seed oil and fatty acid concentrations of wild annual sunflower. Field Crops Res. 15:57-72.

Small, J.K., 1898. Studies in the botany of the southeastern United States- XIV. Hitherto undecided species. Bull. Torr. Bot. Club. 25:465-482.