

# COMPARISON OF OIL QUALITY CHARACTERISTICS OF ACHENES FROM ORIGINAL AND REGENERATED POPULATIONS OF WILD SUNFLOWER SPECIES

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## SUMMARY

One goal of plant genetic resources conservation is to preserve the genetic integrity of individual populations. Since oil quality is environmentally influenced and evaluation for this characteristic is usually performed on oil from achenes from the original populations of wild sunflowers, a study was conducted to compare oil quality of the original populations with populations regenerated for accession maintenance. Five annual species, *Helianthus annuus* L., *H. debilis* Nutt., *H. petiolaris* Nutt., *H. praecox* Englem. & Gray, and *H. niveus* (Benth.) Brandegees, and four perennial species, *H. decapetalus* L., *H. divaricatus* L., *H. giganteus* L., and *H. nuttallii* T. & G. were evaluated for four major fatty acids, palmitic, stearic, oleic, and linoleic acids. There were significant differences between the original and the regenerated populations for palmitic, stearic, and oleic acids in *H. annuus*. In *H. debilis* and *H. praecox*, there were significant differences in oleic and linoleic acids, but not in palmitic or stearic acids. In *H. niveus*, there were significant differences in stearic and oleic acids. Original and regenerated populations of *Helianthus petiolaris* did not differ in oil quality. For the perennial species, only *H. nuttallii* had a significant difference in oleic and linoleic acids. It appears that for certain fatty acids in some species caution will need to be exercised when assessing the potential oil quality of original populations and those regenerated for genebank maintenance.

## Introduction

Plant genetic resources management and conservation comprise several phases including acquisition and maintenance of genetic integrity in germplasm collections (Chang, 1985). The acquisition of germplasm through exploration or exchange is the initial step in the germplasm conservation process. Monitoring and protecting germplasm using *in situ* collections while preserving its original genetic integrity is a priority of germplasm curation (Bretting and Widrechner, 1995). The *Helianthus* germplasm collection maintained at the USDA, Agricultural Research Service, North Central Regional Plant Introduction Station, Ames, Iowa (NCRPIS) includes 50 species (Brothers, 1996). Populations of several wild species have been characterized for fatty acid composition (Seiler, 1985, 1994, 1999; DeHaro and Fernandez-Martinez, 1991; Thompson et. al., 1981; Dorrell and Whelan, 1978). Fatty acid composition is known to be environmentally influenced (Seiler, 1982, 1983, 1986). Since fatty acid analysis is usually

performed on the original populations, the objective of this study was to compare the fatty acid composition of the oil obtained from achenes of original wild species populations to oil obtained from populations regenerated for genebank maintenance purposes.

## Materials and Methods

**Plant Materials.** Achenes from the original populations of 68 annual *Helianthus* populations [54 *H. annuus* L., four *H. debilis* Nutt., five *H. petiolaris* Nutt., three *H. praecox* Englem. & Gray, and two *H. niveus* (Benth.) Brandegees ] and four perennial species (two populations each of *H. decapetalus* L., *H. divaricatus* L., *H. giganteus* L., and *H. nuttallii* T. & G.) were analyzed for fatty acid composition. Achenes were stored at 5 °C and low humidity (<20 %) until analyzed. Achenes were collected from at least 25 individual plants in the original populations that occurred in a variety of habitats. The original populations were isolated, open-pollinated populations. Achenes of the regenerated populations were obtained from plants grown in a common environment under cages using bees as pollinators at the NCRPIS (Iowa). Fatty acid composition of oil from achenes of the regenerated populations was determined and compared to the fatty acid composition of the original populations collected from their natural habitats.

**Fatty Acid Composition Analysis.** Fatty acid composition was determined using a 10 to 20 achene sample. A small portion of the pulverized sample (10 to 20 mg) was transferred to a disposable filter column and eluted with 3.5 ml of diethyl ether. The oil in the diethyl ether solution was converted to methyl esters using an organic-catalyzed transesterification of the triacylglycerols by the addition of 200 µl of tetramethylammonium hydroxide (10 % in methanol), followed by vortexing (Metcalf and Wang, 1981). After 30 minutes, water was gently added to the reaction mixture, and the upper diethyl ether layer was transferred to a glass vial and capped. The sample was injected into a Hewlett-Packard 5890<sup>1</sup> gas chromatograph containing a DB-23 capillary column (25 m x 0.25 mm, J&W Scientific<sup>1</sup>), which was held at 190 °C for 5 min, then programmed to 220 °C at 10 °C per min, held at 220 °C for one min, then programmed to 240°C at 20 °C per minute, and finally held at 240 °C for 0.5 min, for a total time of 10.5 min. The detector was a flame ionization detector (FID). The fatty acid standard used contained the following acids which typically occur in sunflower oil: myristic (14:0), palmitic (16:0), stearic (18:0), oleic (18:1), linoleic (18:2), linolenic (18:3), arachidic (20:0), behenic (22:0), and lignoceric (24:0). Fatty acid peaks were identified by comparing the fatty acid methyl ester peaks and retention times of standards with sample peaks. An electronic integrator was used to calculate the total area under the peaks; the area of each fatty acid peak was expressed as a percentage of the total area. Fatty acids were determined from two samples per population. The peaks of interest in the present study were the saturated palmitic and stearic acids, mono-unsaturated oleic acid, and polyunsaturated linoleic acid. An analysis of variance was used to statistically compare the mean fatty acid values of the original and regenerated populations.

## Results and Discussion

*Helianthus annuus* is the widest ranging annual species in the genus. It is also the most

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frequently represented wild species; hence a large number of *H. annuus* populations were analyzed. Oil from achenes of 54 original and regenerated populations was analyzed for fatty acid composition (Table 1).

Table 1. Average fatty acid composition of oil from original and regenerated wild *Helianthus* populations.

Species	Fatty Acids (%)			
	Palmitic	Stearic	Oleic	Linoleic
<b>ANNUAL</b>				
<i>H. annuus</i> (n=54)				
Original	5.9	3.8	16.9	68.4
Regenerated	5.2*	3.0*	21.1*	67.6
<i>H. debilis</i> (n=4)				
Original	8.1	4.3	23.3	61.6
Regenerated	7.2	3.3	14.8*	73.0*
<i>H. niveus</i> (n=2)				
Original	8.8	5.5	31.5	52.9
Regenerated	8.7	4.1*	35.1*	49.6
<i>H. petiolaris</i> (n=5)				
Original	5.7	2.9	20.1	65.9
Regenerated	5.2	2.7	18.3	70.7
<i>H. praecox</i> (n=3)				
Original	7.5	4.3	29.6	55.8
Regenerated	6.8	4.5	17.3*	69.1*
<b>PERENNIAL</b>				
<i>H. decapetalus</i> (n=2)				
Original	5.2	2.4	27.1	61.9
Regenerated	5.5	3.0	27.2	61.0
<i>H. divaricatus</i> (n=2)				
Original	5.2	3.1	21.5	66.7
Regenerated	5.5	2.8	15.6	74.3
<i>H. giganteus</i> (n=2)				
Original	5.7	2.0	12.0	76.8

Regenerated	5.4	1.7	12.0	78.8
<i>H. nuttallii</i> (n=2)				
Original	4.8	2.9	14.8	75.5
Regenerated	4.8	2.4	24.1*	65.4*
Cultivated sunflower ( <i>H. annuus</i> )	7.0	5.0	16.0	70.0

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\* = Indicates a significant difference between the original and regenerated populations at the 0.05 significance level.

There was a significant decrease in palmitic and stearic acids, an increase in oleic acid, and no change in linoleic acid for the regenerated populations of *H. annuus*, compared to the original populations. A previous study comparing original and regenerated sibbed *H. annuus* populations found differences in stearic, palmitic, and linoleic acids, but not for oleic acid (Seiler, 1983). The inconsistencies between the two studies could be due to the different environments in which the populations were regenerated (Iowa vs. Texas). The fatty acid composition of a typical cultivated sunflower oil was included in Table 1 for comparison, but was not included in the statistical analysis. The present results indicate that both original and regenerated populations should be evaluated to obtain an accurate assessment of the fatty acids for selection and breeding. Also, information about the regenerated populations will need to be kept separate in the germplasm database for future comparisons. Fatty acid composition is influenced by environmental factors, especially temperature, which probably was responsible for the variability between the original and regenerated populations in this study.

Regeneration of *H. debilis* and *H. praecox* populations decreased oleic acid and a significantly increased linoleic acid, but had no significant effect upon palmitic and stearic acid concentrations (Table 1). This relationship is expected due to the high negative correlation between oleic and linoleic acids in sunflower oil (Seiler, 1986). Based on this limited data, both the original and regenerated populations should be evaluated to determine oleic and linoleic acids if these were the fatty acids of interest in a breeding program.

Regenerating *H. niveus* populations decreased the concentration of stearic acid, and increased oleic acid (Table 1). Too few populations were available to obtain a clear understanding of the relationship between the original and regenerated populations of this species.

No significant differences were observed for the populations of *H. petiolaris* for the major fatty acids (Table 1). Due to the limited number of *H. petiolaris* populations observed, these results should be interpreted with caution.

Among the perennial species, only *H. nuttallii* increased in oleic acid and decreased in linoleic acid when regenerated, opposite to what was observed in *H. debilis* and *H. praecox*. Since only a few populations of each perennial species were available for analysis, statistical comparison

can be made, but should be cautiously interpreted until additional information is available.

## Conclusions

Results indicated that *H. annuus* had significant differences between the original and regenerated populations for palmitic, stearic, and oleic acids. There were an adequate number of populations to make a reasonable inference about these populations. Annual species *H. debilis* and *H. praecox* had significant differences between the original and regenerated populations for oleic and linoleic acids. It appears that selection for specific fatty acids in certain species will require analyzing both the original and regenerated populations before determining which populations should be utilized in a selection and breeding program. An insufficient number of populations of wild perennial species was available to obtain a clear understanding of the relationship between fatty acids of the original and regenerated populations. More perennial species populations will need to be examined to provide useful information for selection of fatty acids in a breeding program.

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