INTERRELATION OF FATTY ACIDS IN OIL OF WILD ANNUAL SUNFLOWER (HELIANTHUS ANNUUS L.).

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## Abstract

Wild sunflower germplasm is the basic genetic material of cultivated sunflower. Wild sunflower species continue to contribute specific characteristics for sunflower improvement. Use of wild species in breeding programs may cause problems because of the unknown interrelations of the fatty acids. The interrelations of four fatty acids (palmitic 16:0; Stearic 18:0; oleic 18:1; and linoleic 18:2) of 35 wild annual sunflower genotypes grown at a common location were examined weekly for 14 weeks (flowering dates) during the season. The number of weeks that a genotype flowered was dependent upon the genotype. Interrelations of fatty acids from each of the 14 flowering dates, as well as an average over all weeks, were examined. There was a significant positive relationship between palmitic and stearic acids for each week, as well as the average over all weeks (r=0.561). Palmitic and oleic acids showed a negative relationship which was significant in about half of the weeks examined, but was significant over all weeks (r=-0.422). The relation of palmitic and linoleic acids was variable. In some weeks (especially the early part of the season) it was positive. Later, it was generally negative. The relation between these two acids was significant and positive (r=0.311) over all weeks. Stearic acid vs. oleic acid showed a significant negative relation for all weeks and over all weeks (r=-0.644). Stearic vs. linoleic acid had a negative relationship early in the season, but later in the season it was positive, but not significant for most of the weeks. Overall correlation of these two acids was significant (r=0.521). Oleic vs. linoleic acids showed a high significant negative relationship, being the highest early in the season, and lowest at the end of the season. The correlation over all weeks was r=-0.989. Interrelations of fatty acids of wild annual sunflower does not follow the same pattern as commercial Hybrid 894, except for oleic vs. linoleic acid and palmitic vs. stearic. The other fatty acids of wild and commercial sunflower had similar correlation coefficients but different signs.

### Introduction

Wild sunflower germplasm besides constituting the basic genetic stock from which cultivated sunflower originated has contributed specific characteristics for sunflower (Helianthus annuus L.) improvement (Gimenez and Fick, 1975; Leclercq, 1969; Putt and Sackston, 1957). Since wild H. annuus and other sunflower species appear to possess considerable variability for most economic characteristics, the use of wild material in breeding programs has the potential for markedly changing quality characteristics of sunflower oil (Dorrell and Whelan, 1978; Thompson et al., 1981).

Sunflower oil of cultivated sunflower usually contains a total of 85-91% linoleic and oleic acids and 9-12% saturated fatty acids (palmitic and stearic) (Kinman, 1972). Other minor fatty acids are present in small amounts. In wild annual sunflower, oil usually contains a total of 88-94% linoleic and oleic acids and 5-9% saturated acids (Seiler, 1983).

Interrelation of fatty acids has been examined in cultivated sunflower to a limited extent by Putt et al. (1969) and Zimmerman and Fick (1973). Putt et al. (1969) reported a strong negative association of oleic and linoleic

acids. Both these acids showed some association with stearic acid. Palmitic acid showed no consistent association with any of the other three acids. Zimmerman and Fick (1973) reported similar results except for higher correlations between stearic and oleic acids. No information is available about the interrelation of fatty acids in wild annual sunflower and how they may react when included in commercial breeding programs. The objective of the present study was to examine the interrelations of the four major fatty acids—palmitic, stearic, cleic, and linoleic acids—over the flowering period of several genotypes of wild annual sunflowers grown at a common location.

## Materials and Metbods

The 35 populations used in this study were collected from wild populations (genotypes) of <u>H. annus</u> throughout the United States from 1977 through 1980. The genotypes examined were obtained from a region ranging from 26 to 46° N latitude and 81 to 122° W longitude. All populations were planted 19 May 1981 at Bushland, Texas, which is about 35° N latitude and 102° W longitude. 'Hybrid 894' was planted on six different dates to compare development of a commercial hybrid to the wild genotypes under approximately the same environmental conditions. All plots were irrigated to maintain maximum plant growth. Not all flowering dates of the wilds were represented by a flowering date in the commercial hybrid, but a representative sample was present.

Ten flowering heads per genotype per week (referred to as flowering date) were sibbed or interpollinated. Heads were bagged to prevent open pollination and seed shattering. Some genotypes flowered for as little as three weeks while others flowered over a 14-week period. Seed samples were collected 28 days after flowering (approximately physiological maturity). The number of samples per genotype varied with the length of the flowering period for each genotype.

Fatty acid concentration of solvent extracted oil from 20 to 40 seeds/sample was determined using methyl esterification and gas-liquid chromatography (GLC) (Christie, 1973). Fatty acid concentration was determined on three heads per population per week. Fatty acid peaks were identified by comparing sample peaks with peaks of fatty acid methyl esters standards. An electronic digital integrator was used to calculate the total area of peaks and the area of each fatty acid peak was expressed as a percentage of the total area.

Data for the analysis were an average of the three heads per genotype per flowering date. Data were further grouped by flowering date; each flowering date contained all the genotypes that had heads-flowering at that time. This resulted in varying numbers of genotypes at each flowering date. Phenotypic correlations (based on genotype means) were calculated for the four major fatty acids-palmitic, stearic, oleic, and linoleic-for each flowering date. Ten plants of Hybrid 894, from each date of planting were analyzed for the four major fatty acids and correlation coefficients were calculated for each major fatty acid for each planting date of the hybrid.

# Results and Discussion

Average fatty acid concentration of oil, in seeds of different genotypes and grouped by flowering date grown at a common location, are given in Table 1. The chronological development of the four major fatty acids were similar to previous studies (Seiler, 1982; Seiler, 1983). Fatty acid concentrations of Hybrid 894 were similar to developmental patterns of 'Hybrid 896' (Seiler,

1983), except that oleic and linoleic acid concentrations were lower and higher, respectively, in the present study (Table 2).

Table 1. Average fatty acid concentration of oil of 35 wild annual sunflower genotypes grouped by flowering date.

Flowering	Number	Avera	Linoleic/ Oleic			
date	of genotypes	Palmitic	Stearic	Oleic	Limoleic	ratio
	·		·	%		
15 July	5	4.8	2.4	30.2	59.5	2.0
22 July	15	4.8	2.4	29.5	60.9	2.0
29 July	25	4.8	2.5	28.2	61.2	2.2
5 Aug.	28	4.9	2.5	26.1	£3.8	2.4
12 Aug.	28	5.0	2.5	24.2	55.9	2.7
19 Aug.	33	5.0	2.6	22.2	٤7.7	3.0
26 Aug.	35	5.1	2.6	19.2	71.4	3.7
2 Sept.	31	5.2	2.7	17.5	73.5	4.2
9 Sept.	30	5.3	2.8	15.4	75.2	4.9
16 Sept.	<u>2</u> 6	5.4	2.9	15.0	76.0	5.1
23 Sept.	20	5.4	3.0	14.1	76.2	5.4
30 Sept.	10	5.6	3.2	13.8	76.5	5.5
7 Oct.	8	5.7	3.4	12.6	77.1	6.1
14 Oct.	5	5.6	3.7	12.1	.77.8	6.4
Overall me	an	5.2	2.8	20.0	70.2	3.5

Table 2. Average fatty acid concentration of Hybrid 894 by flowering date.

Flowering	Averag	Linoleic/ Oleic			
date	Palmitic	Stearic	Oleic	Linoleic	ratio
			%		
15 July	5.0	1.9	36.1	48.9	1.4
28 July	5.2	2.1	34.5	53.9	1.6
10 Aug.	5.4	2.0	30.3	57.7	1.9
24 Aug.	5.6	2.6	21.9	61.6	2 <b>.</b> 8
15 Sept.	4.9	2.8	14.4	72.4	5.0
6 Oct.	5.1	3.3	13.3	73,2	5.5
Overall mean	5.2	2.5	25.1	61.3	2.4

The most frequent flowering date was 26 August when all genotypes were flowering (Table 1). The majority of the genotypes flowered between 29 July and 23 September and averaged 8 weeks per genotype.

The ratio of linoleic:oleic acid in the wild genotypes was lowest at the first flowering date and highest at the last flowering date, 2.0 and 6.4, respectively (Table 1). At the most frequent flowering date, 26 August, the ratio was 3.7. The average ratio was 3.5. The linoleic:oleic acid ratic in Hybrid 894 varied from 1.4 to 5.5 for the first and last flowering dates, respectively. The average ratio was 2.4. Filipescu and Stoenescu (1981) reported an average ratio of 2.8 for 20 hybrids grown at 15 different locations. The steady positive increase in the ratio of wild sunflower is accounted for by the

almost direct negative relationship between oleic and linoleic acids (r=-0.989, P=0.01) (Table 3).

Table 3. Phenotypic correlation coefficients between pairs of fatty acids of 35 wild annual sunflower genotypes grouped by flowering date.

Flowering date	P vs. $S^{1/2}$	P vs. 0	P vs. L	S vs. 0	S vs. L	0 vs. L
15 July.	0.885*	-0.193	0.050	-0.189	-0.018	-0.974*
22 July	0.719**	-0.239	0.093	-0.533*	-0.303	-0.987**
29 July	0.501**	-0.353	0.176	-0.248	-0.050	-0.972**
5 Aug.	0.492*	-0.246	0.038	-0.466*	0.287	-0.969**
12 Aug.	0.668**	-0.384*	0.154	-0.384*	0.152	-0.965**
19 Aug.	0.398*	-0.413*	0.259	-0.354*	0.153	-0.920**
26 Aug.	0.398*	-0.551*	0.365*	-0.469*	0.307	-0.903**
2 Sept.	0.452*	-0.018	-0.269	-0.393*	0.608	-0.917**
9 Sept.	0.428*	-0.153	-0.134	-0.486*	0.209	-0.871**
16 Sept.	0.602**	-0.631*	0.414	-0.475*	0.173	-0.885**
23 Sept.	0.473*	-0.520*	0.154	-0.230	-0.199	-0.883**
	0.638*	-0.041	-0.468	-0.096	-0.541	-0.774**
	0.708*	-0.189	-0.526	-0.199		-0.472
14 Oct.	0.880*	-0.569	-0.046	-0.312*	0.140	-0.626
Overall		•	,	X.		
Correlation	0.561**	-0.422**	0.311**	-0.644**	0.521**	-0.989**
30 Sept. 7 Oct. 14 Oct. Overall	0.638* 0.708* 0.880*	-0.041 -0.189 -0.569	-0.468 -0.526 -0.046	-0.096 -0.199 -0.312*	-0.541 -0.728* 0.140	

<sup>\* =</sup> Significant at P=0.05.

There was a significant positive relationship between palmitic and stearic acids for each flowering date as well as the overall flowering dates (Table 3). This positive relationship was not found in inbred lines and varieties examined by Putt et al. (1969) and Zimmerman and Fick (1973). They found a low but significant negative association between these two acids. In the present study, Hybrid 894 had a low positive significant association over all flowering dates between the two acids (Table 4). Fatty acid concentration of Hybrid 894 from several flowering dates within a season was examined. Some (28 July-24 August) had a significant negative relationship, but over all flowering dates the association was positive and significant. The moderate correlation between palmitic and stearic acids in wild sunflower suggests that selection for one acid may result in the change of the other.

The association between palmitic acid and oleic or linoleic acid was not as high as that of stearic acid, but was significant over all flowering dates for the wild sunflower (Table 3). The palmitic vs. oleic association was negative for every flowering date while palmitic vs. linoleic was variable (i.e. positive at early flowering date, negative at later flowering dates). The moderately low correlations between palmitic and oleic or linoleic acids suggest that the level of palmitic acid might be selected in a breeding program without appreciably altering the level of the oleic and linoleic acids.

The association of palmitic acid and oleic or linoleic acid of Hybrid 894 was very low and not significant and of the opposite sign of that of the wild sunflower (Table 4). The data for Hybrid 894 agree with results of Putt et al. (1969) but not those of Zimmerman and Fick (1973). The data of Zimmerman

 $<sup>\</sup>frac{**}{1}$  = Significant at P=0.001.

 $<sup>\</sup>frac{1}{2}$  P = Palmitic, S = Stearic, L = Linoleic, and 0 = Oleic.

and F'k (1973) agree with the present data from the wild sunflower (i.e. a low positive association of palmitic vs. linoleic and low negative association of palmitic vs. oleic acid).

Table 4. Simple correlation coefficients between pairs of fatty acids of Hybrid 894 by flowering date.

Flowering date	P vs. s1/	, P vs. 0	P vs. L	S. vs. 0	S vs. L	0 vs. L
15 July 28 July 10 Aug. 24 Aug. 15 Sept. 6 Oct.	0.500 -0.399 -0.637* -0.731* 0.686* 0.459	0.238 0.092 -0.638* 0.336 0.585 0.469	-0.258 -0.108 0.601 0.039 -0.680* -0.681*	0.493 0.514 0.662* 0.588 0.843* 0.195	-0.428 -0.539 -0.665* -0.684* -0.972**	-0.996** -0.982** -0.979** -0.793** -0.978**
Overall Correlation	0.347**	- 0.117	-0.177	0.835**	-0.808**	-0.997**

<sup>\* =</sup> Significant at P=0.05.

The stearic vs. oleic acid association was negative for all flowering dates and significant over all flowering dates for the wild sunflower (Table 3). The overall correlation coefficient between stearic and oleic acid was the second highest reported (r=-0.644), with the oleic vs. Tinoleic acid being the highest (r=-0.989). The overall correlation coefficient of stearic vs. oleic acid for Hybrid 894 was positive and significant (Table 4). The correlation coefficient for Hybrid 894 was higher than previously reported for inbred lines and varieties by Putt et al. (1969) and Zimmerman and Fick (1973), but had the same sign. Oleic acid showed some association with stearic acid in both the wild and cultivated sunflower. Selection for a certain level of oleic acid may result in a change in the level of stearic acid.

The stearic vs. linoleic acid association was negative early in the season, positive at mid-season and negative again toward the end of the season (Table 3). The correlation coefficients were generally low for each flowering date, but the overall correlation was significant and positive for all flowering dates. Hybrid 894 showed a negative association between stearic and linoleic acid for all flowering dates and only half were significant. Overall correlation was highly significant and negative. These data agree with Putt et al. (1969) and Zimmerman and Fick (1973) for inbred lines and varieties. Linoleic acid showed some association with stearic acid in the wild sunflower and Hybrid 894, so that selection for linoleic acid may affect the level of stearic acid.

Oleic and linoleic acids were highly negatively correlated at all flowering dates (Table 3), with the correlations decreasing for the later flowering dates (especially the last three). This may be due in part to the environmental conditions which are not favorable for plant growth that late in the season. The overall correlation was high and negative (r=-0.989) for the wild sunflower. Hybrid 894 also had a large negative correlation between oleic and linoleic acids (Table 4). The large negative correlation is very similar to

 $<sup>^{**}</sup>$  = Significant at P=0.001.

 $<sup>\</sup>frac{1}{P}$  P = Palmitic, S = Stearic, L = Linoleic, and O = Oleic.

that reported for sunflower varieties grown at several different locations (Putt et al., 1969).

### Conclusions

Breeding for different levels of oleic and linoleic acid appears feasible considering the wide range shown by the wild genotypes (Table 1). The generally lower ranges for palmitic and stearic acids indicate that selection for different levels of these acids would be more difficult.

The strong association of oleic and linoleic acid in wild sunflower is similar to that of cultivated sunflower. Selection for different levels of one will inevitably change the levels of the other. Both of these acids showed some association with stearic acid so that selection for either may affect the level of stearic acid. The associations of palmitic acid with linoleic and oleic acids were weak as shown by the low coefficients. Breeding work with these acids would not affect the level of palmitic acid. There was a moderate positive association between palmitic and stearic acids in the present study, which was not reported in any other study. This association would indicate that selection for palmitic acid may affect the level of stearic acid and vice versa.

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