

# EVALUATION OF THE GERM PLASM COLLECTION OF SAFFLOWER, *CARTHAMUS TINCTORIUS* L. V. DISTRIBUTION AND REGIONAL DIVERGENCE FOR MORPHOLOGICAL CHARACTERS

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## SUMMARY

For many centuries, safflower, *Carthamus tinctorius* L., was grown for the dye extracted from its flowers and as a minor oilseed crop. Only in recent decades has it become an oilseed crop of importance in some areas. In the traditional mode of cultivation, in small plots, there were many small populations in which various selection pressures could operate. The world germ plasm collection of safflower with nearly 2000 lines originating in about 30 countries, was evaluated for divergence by studying the distributions of various expressions (states) of 13 morphological characters (descriptors). The traits were: late rosette leaves' margin; upper stem leaves' margin, shape and color; branches' location and angle; primary head shape; outer involucre bracts' cross section, attitude, spines' length, number and location; corolla color. Divergence due to human and/or natural selection was found for some of the traits and ten centers of cultivation were designated. Geographical sources for desirable traits were identified.

## INTRODUCTION

Safflower, *Carthamus tinctorius* L., was grown until recent decades primarily for the dye extracted from its dried florets but it has also been a minor oilseed crop (HANELT, 1961; KNOWLES, 1955; WEISS, 1971). Since 1945 safflower has assumed growing importance as an oil crop in its traditional areas of cultivation and in the western parts of the USA and Mexico, Australia and elsewhere (KNOWLES, 1955; WEISS, 1971). KUPSOW (1932) was the first to study the range of variability in cultivated safflower and the wild *Carthamus* species. He concluded that safflower had two centers of variability: Ethiopia and Afghanistan. HANELT (1961) considered the Afghanistan-India area a primary center of variability and the Ethiopia-Egypt area a secondary center. Extensive studies and collections were made by KNOWLES in 1958 and 1965 in the old centers of safflower production (KNOWLES, 1969) and thus the germ plasm collection with about 2000 lines was established. KNOWLES (1969) found from field studies and observations in the nursery that divergence has taken place in safflower and suggested the concept of 'Centers of Culture' with a predominant type in each. Consequently, many of the introductions from each center resemble one another. KNOWLES (1969) defined seven such centers: a) Far East; b) India-Pakistan; c) Middle East; d) Egypt; e) Sudan; f) Ethiopia; g) Europe.

In 1966, a USDA P. L. 480 project was initiated to study the entire germ plasm collection in Israel. It was designed to evaluate the whole collection for the first time for resistance to insect pests and to some diseases as well as for regional divergence,

and to identify geographical centers of desirable traits (ASHRI, 1973). Divergence was found for certain yield components (ASHRI et al., 1974), oil content (ASHRI et al., in press) and disease reaction (ASHRI, 1971a). In the present paper, the extent of divergence and the distribution of 13 morphological traits will be examined. The importance of making germ plasm collections and their utilization has been generally recognized (FRANKEL, 1970; KONZAK & DIETZ, 1969; KRULL & BORLAUG, 1970).

The genetic control of many of the traits discussed in this study is not known, those investigated are controlled by one or a few genes only. The margin of the rosette leaves is determined by a single gene (ASHRI & EFRON, 1964). The location of the branches on the main axis is greatly affected by stand density and the environment as well as by genotype (BEECH, 1969; KNOWLES, 1958; WEISS, 1971). The angle of the side branches which can vary widely, 15°–70° in Indian varieties (CHAVAN, 1961) is controlled by one gene (LEON & KNOWLES, 1964) or by two additive genes (ASHRI & EFRON, 1964). Head shape was not studied but head width was: wide heads were reported especially from the Afghanistan area (KNOWLES, 1958; KUPSOW, 1932; WEISS, 1971). Inheritance of spininess was reported to be monogenic (ASHRI & EFRON, 1964; CLAASSEN, 1952) but probably with modifying genes (CLAASSEN, 1952). Corolla color is affected by four genes at least (CLAASSEN, 1952; WEISS, 1971).

#### MATERIALS AND METHODS

The germ plasm collection, containing nearly 2000 lines originating from all the safflower growing areas in the world (Table 1), was supplied by Dr P. F. Knowles, University of California, Davis, CA., via the USDA. Some duplications were probably present in the collection as often happens in collections of this type.

The lines were studied in 1968 and 1969 at the Experimental Farm, Volcani Center, Beit-Dagan, Israel. Since one of the goals of the study was to screen for insect resistance (ASHRI, 1971b) the nurseries were planted very late: on May 14–17, 1968 and May 2, 1969. The nurseries were sprinkler-irrigated according to need.

The traits (descriptors) for study were selected following studies of the germ plasm collection in 1966 and 1967 and consultations with other safflower investigators. The various expressions (states) of the traits were scored visually using criteria developed as above. There were differences between traits in the numbers of lines scored due to variation between years, death of plants later in the season etc. When a line was heterogeneous the score of the majority of the plants was entered. Some of these mixtures could represent initial heterogeneity but many could also result from out-crossing during seed increase at Davis (KNOWLES, 1969).

All the traits were studied in 1968 except for the color of the upper stem leaves and the location of the branches which were recorded in 1969. The traits studied, their expressions and their computer codes were as follows:

##### *Late rosette leaves*

Margin: 1-Entire to slightly serrate, 2-Partly lobed, 3-Deeply lobed.

##### *Upper stem leaves.*

Margin: 1-Entire, 2-Dentate or serrate, 3-Lobed.

Shape: 1-Ovate. 2-Oblong. 3-Lanceolate. 4-Linear.

Color: 1-Light green (yellowish tinge). 2-Dark green (bluish tinge).

*Branches on main axis.*

Location: 1-Basal primarily. 2-Upper half of plant. 3-Base to apex.

Angle: 1-Appressed, 15°–20°. 2-Intermediate, 20°–60°. 3-Spreading, 60°–90°. 4-Drooping.

*Primary head shape before flowering.*

1-Cone shaped to oval. 2-Wide.

*Outer involucrel bracts (= OIB).*

Attitude: 1-Appressed to head. 2-Patent.

Cross section: 1-Flat. 2-Grooved.

Location of spines: 1-None. 2-Tip only. 3-Tip and few basal. 4-Tip and few apical. 5-Tip and all along margins. 6-Margin only.

Spines' length: 1-Short. 2-Medium. 3-Long.

Spines' number: 1-None. 2-Few. 3-Many.

Corolla color types were classified according to KNOWLES (1958) (Nos. 1–8) with an additional category (No. 9) as follows:

<i>Code</i>	<i>Bud</i>	<i>Fresh flower</i>	<i>Dry flower</i>
1	white	white	gray-white
2	light-yellow	light yellow	gray-white
3	light-yellow	light orange base	orange base
4	yellow	yellow	yellow
5	yellow	yellow	light orange base
6	yellow	yellow	orange
7	deep red	reddish orange	deep red
8	yellow, base and apex of corolla lobes		
	orange	as bud	orange
9	pale yellow	pale yellow	pale yellow

## RESULTS AND DISCUSSION

The germ plasm collection safflower contains much variability for the morphological traits (Table 1, 2 and 3) as it has for yield components (ASHRI et al., 1974), oil content (ASHRI et al., in press) and disease reaction (ASHRI, 1971a). Several traits showed considerable variation in most of the regional gene pools, namely: shape of the upper stem leaves; primary head shape; attitude of outer involucrel bracts; spines' length, number and location and corolla color (Table 1, 2 and 3). Considerable variation, but limited to certain regions was found for upper stem leaves' margin and color (Table 1). Interestingly, those lines which had dentate or serrate upper stem leaves were also dark green. There were however, some morphological traits for which the collection was very uniform, as it was for the alcohol dehydrogenase allozymes (EFRON et al., 1973). Such traits were the margin of the late rosette leaves, location and angle of branches on the main stem and cross section of the outer involucrel bracts (Table 1 and 2).

Table 1. Frequency distributions by countries of origin: The number of lines with different types of rosette leaves' margin; upper stem leaves' margin, shape and color; branches' location and angle.

Country of origin	Number of lines <sup>1</sup>	Late rosette leaves, margin			Upper stem leaves						Branches								
					margin			shape			color			location <sup>2</sup>			angle <sup>3</sup>		
		1	2	3	1	2	3	1	2	1	2	2	3	1	2	3			
Afghanistan	33	33	0	0	28	3	0	19	12	28	3	4	23	0	31	0			
Australia	18	16	2	0	17	1	0	4	14	17	1	0	18	1	17	0			
Bangladesh	49	37	11	1	27	21	0	13	36	27	21	2	44	3	46	0			
Canada	15	15	0	0	15	0	0	2	13	15	0	0	15	1	14	0			
China	1	1	0	0	0	0	1	0	1	0	0	1	0	0	1	0			
Egypt	375	317	55	3	315	53	0	69	299	315	53	7	348	34	330	4			
Ethiopia	25	25	0	0	16	9	0	0	25	16	9	3	22	0	25	0			
France	4	4	0	0	4	0	0	0	4	4	0	1	3	0	4	0			
India	958	945	13	0	851	107	0	155	803	851	107	18	893	49	882	25			
Iran	119	118	1	0	113	6	0	94	25	113	6	10	93	8	108	3			
Iraq	11	11	0	0	11	0	0	10	1	11	0	0	11	0	11	0			
Israel	21	20	1	0	20	1	0	15	6	20	1	2	19	5	16	0			
Italy	6	6	0	0	6	0	0	0	6	6	0	0	6	1	5	0			
Japan	9	2	1	6	1	3	4	0	9	1	3	3	6	1	8	0			
Jordan	15	15	0	0	15	0	0	9	6	15	0	1	14	1	14	0			
Kashmir	13	7	6	0	9	4	0	1	12	9	4	0	13	0	13	0			
Kenya	9	9	0	0	8	1	0	0	9	8	1	0	8	1	8	0			
Kuwait	19	16	3	0	16	3	0	7	12	16	3	0	19	3	16	0			
Morocco	14	14	0	0	14	0	0	1	13	14	0	1	13	0	14	0			
Pakistan, W.	24	23	1	0	17	7	0	6	18	17	7	1	19	1	23	0			
Philippines	3	3	0	0	3	0	0	0	3	3	0	0	3	0	3	0			
Portugal	65	65	0	0	63	2	0	5	60	63	2	4	60	6	59	0			
Romania	2	2	0	0	2	0	0	1	1	2	0	0	2	1	1	0			
Spain	25	24	1	0	25	0	0	8	17	25	0	1	24	0	25	0			
Sudan	32	30	2	0	30	2	0	0	32	30	2	3	29	0	32	0			
Syria	9	9	0	0	9	0	0	5	4	9	0	0	5	1	8	0			
Turkey	85	84	1	0	76	9	0	56	29	76	9	7	74	3	81	1			
USSR	7	7	0	0	6	0	0	0	6	6	0	1	6	0	6	0			
Total	1966	1858	98	10	1717	232	5	480	1476	1717	232	70	1790	120	1801	33			

<sup>1</sup> Maximum number of lines. Some lines were not scored for certain traits because of delayed germination, death, etc.

<sup>2</sup> There was also one line from India with a score of 1.

<sup>3</sup> There was also one line from India with a score of 4.

For certain traits, the full range of variation including an extreme expression was limited to given areas while variation for less extreme degrees of expression was common in most areas. For instance, lines with partly lobed and entire rosette leaves were found in many countries but deeply lobed leaves were found only in Bangladesh (1 line out of 49), in Egypt (3 out of 375) and Japan (6 out of 9 lines). Similarly, there were only five lines in the entire collection with lobed upper stem leaves: four from Japan and one from China. It is interesting to note that HANELT (1961) considered pinnately divided lower leaves as a primitive characteristics. Yet this trait was most frequent outside the area of origin of safflower, in Japan. Such cases may represent a 'founder effect' where a single original introduction became widespread.

SAFFLOWER EVALUATION

Table 2. Frequency distribution by countries of origin: The number of lines with different types of primary head shape; outer involucre bracts' cross section and attitude; spines' length, number and location.

Country of origin	Number of lines <sup>1</sup>	Primary head shape		Outer involucre bracts														
				cross section		attitude		spines' length			number of spines			spines' location				
				1	2	1	2	1	2	3	1	2	3	1	2	3	4	5
Afghanistan	31	3	25	22	9	25	6	25	6	0	0	26	5	0	12	0	0	19
Australia	18	7	11	18	0	14	4	10	7	1	0	13	5	0	2	1	0	15
Bangladesh	49	30	11	49	0	35	14	42	7	0	0	38	11	0	2	0	0	47
Canada	15	13	1	15	0	8	7	3	9	3	0	8	7	0	0	0	0	15
China	1	0	1	1	0	1	0	1	0	0	0	1	0	0	1	0	0	0
Egypt	368	176	179	347	20	239	128	206	125	37	0	255	113	0	63	54	0	251
Ethiopia	25	12	13	25	0	0	23	1	24	0	0	0	25	0	0	1	0	24
France	4	2	2	4	0	4	0	3	0	1	0	3	1	0	0	1	0	3
India	958	766	144	957	1	754	204	334	532	92	7	697	254	2	14	15	1	926
Iran	119	18	93	93	26	105	14	116	3	0	1	116	2	1	72	5	0	41
Iraq	11	3	8	10	1	6	5	6	5	0	0	7	4	0	2	1	0	8
Israel	21	6	15	20	1	15	6	15	6	0	0	17	4	0	8	2	0	11
Italy	6	5	1	6	0	4	2	0	6	0	0	4	2	0	1	1	0	4
Japan	9	2	7	9	0	1	8	1	8	0	0	1	8	0	1	0	0	8
Jordan	15	2	13	14	1	14	1	15	0	0	0	15	0	0	6	4	0	5
Kashmir	13	13	0	13	0	12	1	12	1	0	0	9	4	0	0	0	0	13
Kenya	9	5	3	9	0	3	6	3	3	3	0	4	5	0	0	0	0	9
Kuwait	19	13	6	19	0	16	3	6	8	5	0	14	5	0	2	0	0	17
Morocco	14	3	11	14	0	10	4	6	7	1	0	12	2	0	4	1	0	9
Pakistan W.	24	11	11	24	0	13	11	18	5	1	0	20	4	0	1	2	0	21
Philippines	3	0	3	1	2	3	0	3	0	0	0	3	0	0	1	0	0	2
Portugal	65	31	33	62	3	47	18	34	29	2	0	38	27	0	5	10	0	50
Romania	2	0	2	2	0	2	0	2	0	0	0	2	0	0	0	0	0	2
Spain	25	14	11	25	0	19	6	18	6	1	0	21	4	0	8	3	0	14
Sudan	32	21	11	31	1	20	12	8	17	7	0	19	13	0	3	1	0	28
Syria	9	1	5	9	0	9	0	9	0	0	0	9	0	0	2	7	0	0
Turkey	85	35	46	81	4	59	26	45	36	4	0	70	15	0	31	3	0	51
USSR	7	3	4	2	4	2	4	2	4	0	0	3	3	0	2	1	0	3
Total	1957	1195	670	1882	73	1440	515	942	854	157	8	1425	523	3	243	113	1	1596

<sup>1</sup> Maximum number of lines (see Table 1).

A 'founder effect' may have occurred also in India. KNOWLES (1969) noted the remarkable uniformity of safflower in south-central India where it was grown for oil on 600,000 hectares and speculated that it developed from a single introduction. Earlier studies have shown considerable variation in the safflower of India (see CHAVAN, 1961; HANELT, 1961; KUPSOW, 1932).

Corolla color variation has been studied by HANELT (1961), KNOWLES (1969) and KUPSOW (1932). KUPSOW (1932) who had a limited collection, described four corolla types. He considered the white corolla (type 1 of KNOWLES - 1958 - and the present study) as the rarest. Our studies show that it is rare but still it can be found in many countries. Corolla color variability in lines from India was higher than that reported

Table 3. Percent of the lines in the nine types of corolla color, by countries of origin<sup>1</sup>.

Country of origin	Number of lines	% in corolla type								
		1	2	3	4	5	6	7	8	9
Afghanistan	33	0	0	45	9	21	0	0	15	9
Australia	18	11	0	6	39	17	0	0	28	0
Bangladesh	49	0	0	61	31	4	0	0	4	0
Canada	15	0	0	0	13	87	0	0	0	0
Egypt	375	11	0	4	34	33	0	0	16	2
Ethiopia	25	0	0	12	8	4	0	0	76	0
France	4	0	0	0	0	75	0	0	25	0
Hungary	4	0	0	0	50	50	0	0	0	0
India	958	7	4	25	9	48	3	0.1	5	0.1
Iran	119	1	0	23	13	14	0	0	49	0
Iraq	11	0	0	45	9	27	0	0	18	0
Israel	21	0	0	5	19	38	0	0	38	0
Italy	6	17	0	0	67	17	0	0	0	0
Japan	9	0	0	22	44	11	0	0	22	0
Jordan	15	0	0	7	7	20	0	0	67	0
Kashmir	13	0	0	38	46	15	0	0	0	0
Kenya	9	11	0	0	44	44	0	0	0	0
Kuwait	19	0	0	63	16	0	0	0	21	0
Morocco	14	29	0	0	21	36	0	0	14	0
Pakistan, W.	24	0	4	50	13	21	0	0	0	13
Philippines	3	0	0	33	33	33	0	0	0	0
Portugal	65	0	0	2	2	55	0	0	42	0
Spain	25	4	0	0	0	56	0	0	40	0
Sudan	32	9	3	9	31	38	0	0	9	0
Syria	9	0	0	0	11	11	0	0	78	0
Turkey	85	0	0	16	22	11	0	0	51	0
USSR	7	0	0	0	14	43	0	0	29	14

<sup>1</sup> Countries with 1–2 lines were omitted. Type 5 was found in Algeria, China, Greece, Poland, Romania; type 8 in Lebanon; types 1 and 5 in Argentina.

by KNOWLES (1969). He reported that most of the Indian lines were orange while yellow ones were rare. Apparently the collection studied in the present collection was more extensive since 13% of the lines (125 lines) had yellow (type 4) or light yellow (type 2) corollas.

This study shows that divergence has occurred in safflower and that several regional gene pools – the centers of culture and similarity described by KNOWLES (1969) – have been formed. The distributions of the morphological traits and other attributes (ASHRI, 1973; ASHRI et al., 1974; ASHRI et al., in press) indicate that the Middle East center of KNOWLES (1969) should be divided into three regions. There are differences between the Iran-Afghanistan germ plasm pool and the Near East pool (Israel, Jordan, Syria, Iraq). The Turkish lines should be placed in a separate group because many of them are not endemic and have diverse origins (HANELT, 1961; KNOWLES, personal communication). Also, in view of our findings for the morphological and other traits, Kenya should be added to the list. Thus the major pools recognized are: 1) Far East (poorly represented in the collection); 2) Indian subcontinent; 3) Iran,

Afghanistan; 4) Israel, Jordan, Iraq, Syria; 5) Turkey; 6) Egypt; 7) Sudan; 8) Kenya; 9) Ethiopia; and 10) Morocco, Spain, Portugal, France. Divergence is not always clearcut. Where it was not found it is possible that the characters had little or no selective advantage. It is also possible that classification by countries masked divergence because different phytogeographical regions were included in the same political boundaries. Naturalized recent introductions also contributed to counteract divergence.

The divergence process in safflower would fit well with HARLAN'S conclusion (1970) that cultivated plants have undergone repeated cycles of hybridization and differentiation. Thus, hybrid swarms were created and then through isolation and selection new types were established. Safflower is both self- and cross-pollinated (CLAASSEN, 1950; KNOWLES 1969; WEISS 1971), which facilitates intercrossing, segregation and genetic recombination. The traditional methods of cultivation of safflower satisfied well the requirements for isolation and selection. It was grown by many farmers in small plots, often in rows bordering and 'fencing off' fields of other crops (HANELT, 1961; KNOWLES, 1969; WEISS, 1971). Frequently, the farmers planted their own seeds. Thus, the gene pool was fragmented into many isolated, small, populations in which natural and human selection as well as drift could operate. In India a bushy and spiny plant which could fill sparse stands and resist birds was preferred (KNOWLES, 1969). In Iran, where apparently the above was not necessary, selection was practiced for orange and red flowered, more spineless plants from which it was easier to pick the florets. In Ethiopia on the other hand, an orange colored but spiny type was established.

Safflower satisfied also other requirements of HARLAN'S (1970) model. There was exchange of seeds between the regions by nomads, armies and merchants in ancient times (HANELT, 1961; KNOWLES, 1958; KNOWLES, 1969; WEISS, 1971) and introductions which became naturalized in recent years. Finally, also the other source of variability in HARLAN'S model (1970), i.e. the inflow of genes from related wild species (which may be considered weedy forms) is found in safflower. The species *C. oxyacantha* M.B., *C. palaestinus* EIG and *C. persicus* WILLD. (= *C. flavescens*) which form fertile F<sub>1</sub> and F<sub>2</sub> hybrids with the cultivated species (ASHRI & KNOWLES, 1960; ASHRI & RUDICH, 1965; DESHPANDE, 1952; HANELT, 1961 and 1963) contributed genes to the latter in their respective areas of distribution (ASHRI & RUDICH, 1965; DESHPANDE, 1952; HANELT, 1961; KNOWLES, 1969). The strong and yellow spines of many Indian varieties are reminiscent of those of *C. oxyacantha*.

The associations of the morphological characters with each other and with yield components and oil content were studied by contingency tables (ANON., 1967). Significant  $\chi^2$  values were obtained for most of the associations of the morphological traits, but often it was difficult to discern meaningful trends. Associations with yield, oil content and disease reaction were discussed separately (ASHRI, 1973; ASHRI et al., 1974; ASHRI et al., in press). Briefly, it is noteworthy that correlations of spininess and oil content were small and not significant (ASHRI et al., in press). Furthermore, the number of heads per plant – which is the most important yield component – was significantly correlated with spininess only in the Iranian and Egyptian lines (ASHRI et al., 1974). Plants having more seeds per head tended to have appressed branches and ovate, dark green leaves with few short spines; lines with heavier seeds

tended to be bushy, with oblong, light green leaves, cone-shaped heads and spinier (ASHRI et al., 1974).

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