

HYBRIDS BETWEEN THE GENERA *Helianthus* AND *Tithonia* AND THEIR STUDY

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SUMMARY

An intergeneric hybridization is conducted with species from the genera *Tithonia* and *Helianthus*. Hybrid plants F₁ – F₄ between *H. annuus* and *T. rotundifolia* are produced. The inheritance of characters in F₁ is intermediary and specific depending on the forms used. Plants are produced in next generations with valuable characters which could be useful in breeding. A presence of restoring genes of fertility is found in *T. rotundifolia* for the cytoplasms of *H. petiolaris* and *H. argophyllus*. On the basis of these genes new restorers are developed for these two cytoplasms.

Key words: Sunflower, interspecific hybrids, *Helianthus*, *Tithonia*

INTRODUCTION

Till now eleven species of the genus *Tithonia* are known but the chromosome number is found in few of these (Heiser et al., 1969; Fedorov, 1969; Rogers et al., 1982). According to Heiser et al. (1969) and Schilling and Jansen (1989), a close relation exists between the genera *Helianthus*, *Tithonia*, *Viguiera* and some others. It is determined by the study of Rogers et al. (1982) that some species of the genus *Tithonia* contain specific organic compounds among which natural rubber, about 2%. We found almost a complete resistance of species from the genus *Tithonia* to basic diseases in sunflower as mildew, Phomopsis and Sclerotinia. The seeds of these species contain also some quantity of oil.

These original characteristics of the genus *Tithonia* give an idea of its possibilities as a donor of different characters for sunflower.

Although the genera *Tithonia* and *Viguiera* are closely related with the genus *Helianthus*, the reports for hybridization between these are very few. Heiser et al. (1969) informed of crossing these genera but only the crosses between four species of *Helianthus* and *Viguiera porteri* proved to be successful. Hybridization was also carried out with species of *Tithonia* which failed. Regardless of the equal chromosome number of the sunflower with that of species from *Tithonia* ($2n = 34$) determined by Heiser (1948), Delay (1951) and Turner & King (1964), the hybridization between these is difficult because of considerable morphological and genetic differences.

The present study has the aim to find out possibilities for hybridization between *Helianthus* and *Tithonia* and the use of the latter as a genetic source of new characters for improvement of the cultivated sunflower.

MATERIAL AND METHODS

As a female parent from *H. annuus* the following varieties and lines are used:

- Variety Peredovik, maintained through group pollination;
- Line 3004 with cytoplasm of *H. petiolaris*;
- Line HA-89 with cytoplasm of *H. argophyllus*;
- The variety Peredovik is used as pollinator.

Three species from genus *Tithonia* are used:

- *T. rotundifolia* -567 and *T. rotundifolia* -119, used as male and female parents;
- *T. tagetiflora* -401 and *T. speciosa* -1373, used only as a female parent.

The florets of the mother inflorescences from the variety Peredovik and the three species of *Tithonia* are previously castrated. Treatment by chemical agents is not conducted. Fresh pollen is placed three times on the stigmas from each castrated or male sterile inflorescence. The seeds from the crosses were naturally developed and after maturing were planted under greenhouse conditions. After the second leaf stage the plants were sown in field. Some of the plants were isolated during flowering, while the others were exposed to open pollination. The seed set was determined as a percentage from all florets developed in the inflorescence.

Pollination was made by pollen from the F₁ hybrids between *Helianthus* and *Tithonia* against some male sterile lines to check possible restoring ability from the side of *Tithonia*. The plants produced (testcross) were self-pollinated to verify the ability for seed formation.

RESULTS AND DISCUSSION

A. Biological and morphological characteristics of the hybrids.

Hybridization between sunflower and *T. rotundifolia* is conducted comparatively easily. The seed set produced from seeds (1,31% and 2,20%) shows the pollen function rate from *Tithonia* on the stigma of the line HA-89 and the variety Peredovik. Only one seed is obtained by the line 3004. These differences indicate that there is a genetic control of crossability (Table 1). In the reciprocal cross and the use of the *Tithonia* species as a mother parent the seeds produced are considerable more numerous (up to 32,6%) but the seeds are nonviable. One plant is produced from 119 seeds of the cross *T. rotundifolia* -567 x Peredovic and this plant died before bud stage. The conclusion from this is that the incompatibility between the two genera is increased significantly in the cytoplasm of *Tithonia*. Probably, an intermediate species has to be used to overcome this incompatibility. It could be also supposed the cytoplasm of *T. rotundifolia*, *T. tagetiflora* and *T. speciosa* are depressive towards the cultivated sunflower. On the other side the genome of *T. rotundifolia* interacts rather good with the cytoplasm of sunflower. Although the crossing pattern is not complete, these conclusions were confirmed during the three years of crossing and the development of hybrid plants. The formation of a portion of hybrid plants from the three groups of crosses confirms also these conclusions. For the plants from the cross Peredovic x *T. rotundifolia* -567 it is specific that after 3-4 true leaf pairs deformations occur in subsequent leaves. The final leaves are developed only as petioles with traces of leaf blades. The inflorescence reaches bud formation and then it dies.

Sixteen of 21 plants developed in that manner. The growth of the remaining five plants is normal, inflorescences are formed and after selfpollination seeds are produced in three of these. These characteristics show the heterogeneity of the parents regarding the compatibility of their genetic material in plant formation. In the variety Peredovik the heterogeneity is higher in comparison with the lines as in the use of male sterile lines all hybrids plants developed normally and uniformly.

It is typical that the hybrid plants produced from the crosses of *T. rotundifolia* -119 and male sterile lines HA-89 and 3004 are different by phenotype, especially in stem branching. In the cross 3004 x *T. rotundifolia* -119 the plant is not branched with green leaves developed naturally and the whole plant is covered densely by short white hairs. The plants from the cross (ms)-HA-89 x *T. rotundifolia* -119 are uniform and very branched with dark-green leaves. There is anthocyanin staining on the stem, branches and leaf petioles. Whole plants are also densely covered by short white hairs. The difference in the stem branching in the second cross could be explained by the presence of genetic material of the male sterile line HA-89 incorporated in sterile cytoplasm of *H. argophyllus* and at the moment of crossing it is in BC₄. This species is very branched and its recessive genes interact complementary with the genes for branching from *Tithonia* as a result of which the plants in F₁ are branched. The *Tithonia* genes for branching are expressed in F₂ in crosses (ms) - 3004 x *T. rotundifolia* -119 and Peredovik x *T. rotundifolia* -567. From the second cross branched plants were produced also in F₃. As a result of selection these genes were eliminated in the generation of F₄. It was produced in 1990 and all plants were non-branched. Typical for this type of branching is its location at the stem base.

Table 1. Results of hybridization between *H. annuus* and three species of the genus *Tithonia*

Crosses in years	Pollinated inflorescences			Seeds produced, No	Seed set, %	Hybrid plants produced		
	Number	With seeds				Number	%	Reached the flowering stage, No
		No	%					
1986 - 1987								
<i>H.annuus</i> -Peredovik x <i>T.rotundifolia</i> -567	2	2	100	83	2,20	21	25,3	5
<i>T.rotundifolia</i> -567 x <i>H.annuus</i> -Peredovik	8	6	75	119	15,02	1	0,84	0
<i>T.tagetiflora</i> -401 x <i>H.annuus</i> -Peredovik	4	3	75	30	8,40	0	0	0
<i>T. speciosa</i> -1373 x <i>H.annuus</i> -Peredovik	4	4	100	108	21,95	0	0	0
1988 - 1989								
<i>H.annuus</i> -(ms)-3004 x <i>T.rotundifolia</i> -119	1	1	100	1	0,08	1	100	1
1989 - 1990								
<i>H.annuus</i> -(ms)-HA-89 x <i>T.rotundifolia</i> -119	2	1	50	19	1,31	19	100	19
<i>T.rotundifolia</i> -119 x <i>H.annuus</i> -Peredovik	1	1	100	43	32,57	0	0	0

The leaves of *T. rotundifolia* -119 and F₁ hybrids are less in size than those of sunflower. Special features of the leaf blades and petioles of the species (a) and the hybrid (b) can be seen in Figure 1. Inflorescences of these plants are presented in Figure 2. The inheritance of these characters is intermediate or it is equal to the higher values.

The basic morphological indexes of parents and hybrids are given in Table 2. Plant height in F₁ approaches the higher parent or it is heterotically expressed. The branching of the stem from *Tithonia* is manifested as a recessive character in the use of a homogeneous and uniform material from sunflower. The hybrids with the line HA-89 are branched because of residual material from *H. argophyllus*. Each branch of first or second order carries an inflorescence. The inflorescence size is specifically inherited. In the hybrids of Peredovik x *T. rotundifolia* -567 the inflorescence diameter is 18,0 and 19,0 cm, similar to that of the variety Peredovik. The hybrids produced between the lines 3004 and HA-89 and *T. rotundifolia* - 119 form smaller inflorescences. The same tendency is found also in relation to the number of tubular florets which depends directly on the size of inflorescence. There are considerable differences between single plants in the same cross. All this shows that the species *T. rotundifolia* is heterogeneous and its gametes possess different potential for combination with sunflower gametes. By the side of the sunflower lines specific differences are also determined regarding the morphological characters and the fertility. These characteristics are of importance for future work on the transfer of some characters from the genus *Tithonia* in sunflower. The transfer of resistance to diseases is of special interest. The resistance, which the genus *Tithonia* possesses, is a very suitable source for tolerance to important and universal diseases in sunflower.

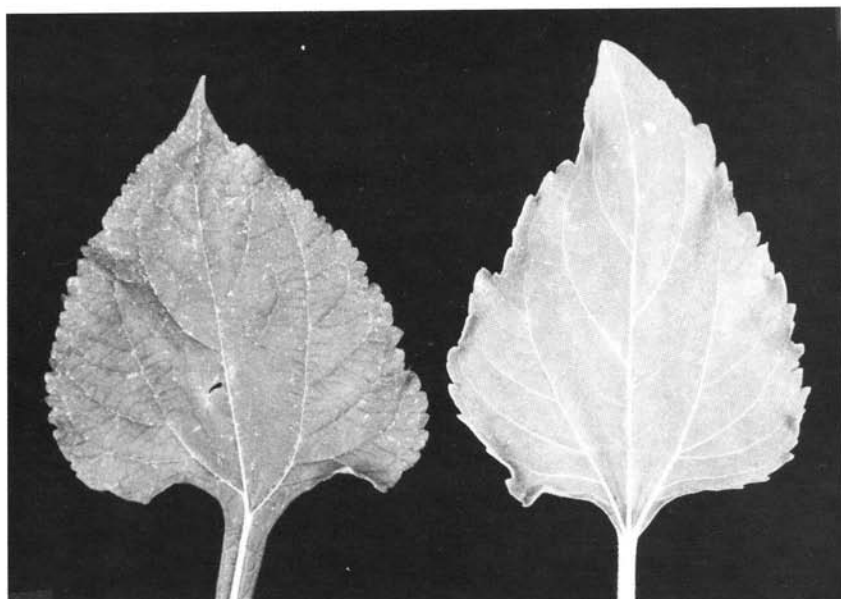


Figure 1. Features of the leaves from *T. rotundifolia*-119 (a) and the F₁ hybrid with the variety Peredovik (b)

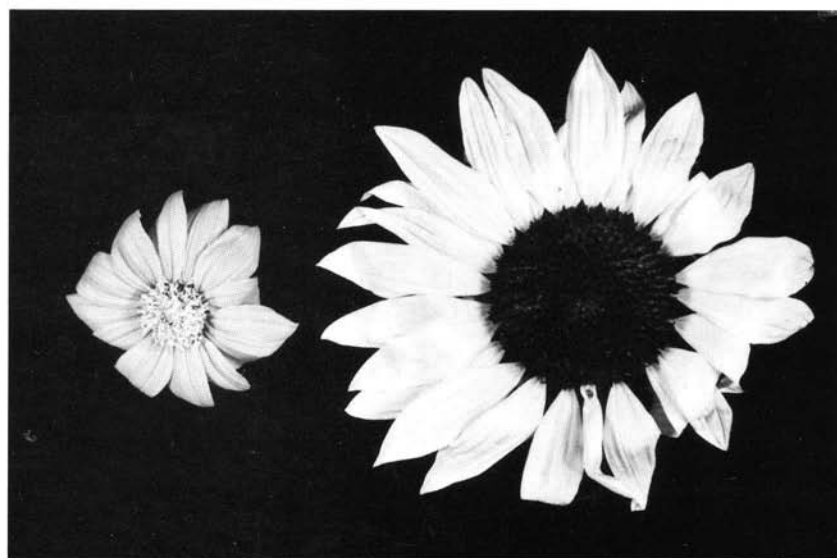


Figure 2. Inflorescence from *T. rotundifolia*-119 (a) and the F_1 hybrid with the variety Peredovik (b)

Table 2. Phenotypic characteristics of the intergenetic hybrids F_1 , *H. annuus* x *T. rotundifolia*

Crosses in years	Plant height, cm	Branches, No	Leaves, No	Inflorescences, No	Head diameter, cm	Tube-like florets, No	Seeds produced		
							By self-pollination, No	By free pollination, No	%
1987									
<i>H. annuus</i> -Peredovik x <i>T. rotundifolia</i> -567									
1-st plant	180	0	43	1	19,0	1690	39	-	2,3
2-nd plant	185	0	37	1	18,0	1742	191	-	11,0
1989									
<i>H. annuus</i> -(ms)-3004 x <i>T. rotundifolia</i> -119	152	0	29	1	13,0	711	267	-	37,5
1990									
<i>H. annuus</i> -(ms)-HA-89 x <i>T. rotundifolia</i> -119									
1-st plant	175	92	436	92	10,0	636	-	603	94,8
9-th plant	175	77	193	77	12,0	176	-	102	57,9
17-th plant	145	29	69	29	11,0	490	-	211	43,1
18-th plant	148	24	58	24	11,0	484	95	-	19,6
19-th plant	160	34	72	34	7,8	402	30	-	7,5
<i>T. rotundifolia</i> - 119	135	118	230	118	2,9	132	-	71	53,8
Peredovik	190	-	33	1	19,0	1883	-	1743	92,6
L - 3004	115	-	28	1	12,5	1194	-	886	74,2
L - HA-89	100	-	30	1	18,0	1447	-	1132	78,2

B. Fertility and seeds of hybrids

It was underlined that the rate of fertilization of sunflower by pollen from the *Tithonia* species is comparatively high for the systematic remoteness of the two genera. The reason for the differences between the reciprocal crosses could be the larger pollen from *Helianthus* and the shorter stigma of *Tithonia*. The reason for the non-viability of hybrid seed and a part of the plants produced, as well as the variations in hybrid fertility are due to genetic distinctions. Although *Helianthus* and *Tithonia* have equal chromosome number, $2n = 34$, (Heiser, 1948; Delay, 1951) the meiotic partition probably, does not run normally and the self-fertility is decreased. It is evident that forms from the two genera with a very good genetic compatibility could be found which is confirmed by the data in Table 2 for seed quantities produced from single plants.

The seeds from *T. rotundifolia* are shown in Figure 2, also seeds of F_1 hybrid and the line HA-89, produced after open pollination. The seeds of the first hybrid are narrow, long, with protrusions for dispersing. The hybrid seeds have intermediate form and a specific protrusion at the apex. The seeds are well filled as the 1000-seed weight is from 28 g to 45 g. In single plants, the seed size is different (Table 3). Oil content is inherited intermediately. As the data show, the seed oil content of *T. rotundifolia* is 23,7%, 48,6% of the line HA-89; and up to 38% in the hybrid seed of F_1 .

Pollen fertility in F_1 and its ability for self-pollination is especially significant. The plant produced in F_1 from the cross (ms) - 3004 x *T. rotundifolia* -119 during 1989 was selffertile and formed 37,5% seeds in self-pollination. Thirty seeds of these were planted producing 27 plants, of which four were male sterile and the rest formed seeds in a different degree. This indicates that the malesterile line 3004 with cytoplasm from *H. petiolaris* can restore its male sterility from Rf genes of *T. rotundifolia*.



Figure 3. Seeds from *T. rotundifolia*-119 (a), F_1 after open pollination (b) and the line HA 89 (c)

Table 3. Characteristics of the seeds from different generations of intergeneric hybrids *H. annuus* x *T. rotundifolia*, 1990

Pro-geny	Crosses and plants	Size of seeds, mm			1000 seed weight, g	Oil content, %
		Length	Width	Thickness		
F ₁	(ms)-HA-89 x <i>T. rotundifolia</i> -119					
	1-st plant	8,4	4,3	2,5	34,4	38,0
	9-th plant	12,2	6,3	4,5	45,5	37,3
F ₂	17-th plant	7,3	4,2	2,2	28,0	36,2
	(ms)-3004 x <i>T. rotundifolia</i> -119					
	1-st plant	11,2	5,2	2,2	48,80	41,3
F ₄	2-nd plant	10,2	5,4	3,4	45,20	42,3
	Peredovik x <i>T. rotundifolia</i> -567					
	1-st plant	9,2	3,2	2,3	24,40	50,1
	2-nd plant	14,2	6,3	3,2	80,0	44,1
Peredovik		12,2	6,2	4,3	68,40	49,4
L - 3004		14,3	5,4	3,7	52,00	41,1
L - HA-89		11,7	5,7	4,4	44,00	48,6
<i>T. rotundifolia</i> -119		7,3	2,3	1,2	1,25	23,7

A high degree of fertility is found in F₁ and in the cross (ms) - HA-89 x *T. rotundifolia* -119 and the line's cytoplasm is from *H. argophyllus*. It is known from our studies that this cytoplasm is different from that one of *H. petiolaris*. The presence of restoring genes in *T. rotundifolia* toward the cytoplasm of *H. argophyllus*, and also to *H. petiolaris*, gives the possibility for new restorers of fertility to be produced in development of hybrid sunflower. All F₁ plants of the cross mentioned above were self-fertile. Two male sterile inflorescences were pollinated by pollen from these plants, respectively with the cytoplasms of *H. petiolaris* and *H. argophyllus* (testcross) and 68 and 106 seeds were produced. This shows that the pollen function in F₁ is normal. The plants produced could be used for developing a new type of fertility restorers.

C. Subsequent generations

In Table 3 data are presented for a part of the progenies in F₂ and F₄ in connection with their breeding value. It can be seen that some of the plants possess valuable breeding indices. Plant No 2 from the cross Peredovik x *T. rotundifolia* -567 in F₄ is remarkable for a high 1000 seed weight - 80 g, and a comparatively high oil content. The combination of this character with other economic indexes could increase considerably the productivity of sunflower lines used in hybrid seed production.

CONCLUSION

As a result of intergeneric hybridization conducted, hybrid plants F₁ - F₄ between *H. annuus* and *T. rotundifolia* are produced. The inheritance of characters in F₁ is intermediate and specific depending on the forms used. Plants are obtained in F₄ possessing valuable single features. A presence of restoring genes for fertility is found in *T. rotundifolia* for the cytoplasms of *H. petiolaris* and *H. argophyllus*. New restorers of these two cytoplasms are developed on the basis of these genes.

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HÍBRIDOS ENTRE EL GENERO *Helianthus* Y *Tithonia* Y SU ESTUDIO

RESUMEN

Se llevó a cabo una hibridación intergenérica entre los géneros *Tithonia* y *Helianthus*. Plantas híbridas F-1 - F-4 entre *H. annuus* y *T. rotundifolia* fueron producidas. La herencia de los caracteres en F-1 fué intermedia y específica dependiendo del material utilizado. En la generación siguiente fueron producidas plantas con caracteres valiosos que podrían ser útiles en la mejora. Se encontraron genes de restauración de la fertilidad en *T. rotundifolia* para un citoplasma de *H. petiolaris*, y *H. argophyllus*. En base a estos genes se están desarrollando nuevos restauradores para estos citoplasmas.

HYBRIDES ENTRE LES GENRES *Helianthus* ET *Tithonia*.

RÉSUMÉ:

Une hybridation intergénérique a été effectuée avec des espèces appartenant aux genres *Helianthus* et *Tithonia*. Des plantes hybrides F1-F4 entre *H. annuus* et *T. rotundifolia* ont été produites. L'héritabilité des caractères dans la F1 est intermédiaire et dépend des formes utilisées. Les plantes produites dans la génération suivante possédaient des caractères intéressants d'un point de vue sélection. La présence de gènes de restauration de fertilité a été détectée chez *T. rotundifolia* pour les cytoplasmes issus de *H. petiolaris* et *H. argophyllus*. Sur la base de ces gènes de nouveaux restoreurs ont été développés pour ces deux cytoplasmes.