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DISTANT (INTERSPECIFIC) HYBRIDIZATION OF SUNFLOWERS IN THE U.S.S.R.

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Many biologists and plant breeders have worked on interspecific hybridization in the genus Helianthus in the USSR and abroad.

Studies on this problem were begun seventy years ago. In 1896 Cowell crossed H. decapetalus L. with H. petiolaris Nutt. Theilung in 1913 and Suiton in 1914 reported obtaining hybrids of H. annuus L. x H. rigidus Dest. In 1916 T. D. Cockerell crossed H. annuus L. with H. decapetalus and H. tuberosus L. and obtained "false hybrids". In 1932 S. Wagner obtained hybrids by crossing H. cucumerifolius T. et G. with various perennial sunflower species.

Interspecific hybridization in the USSR was practised by F. A. Saziperov, and E. M. Platchek, F. A. Saziperov crossed H. annuus L. with H. argophyllus T. et G. The objective of all these experiments was to investigate interspecific hybridization from the genetical point of view.

Soviet investigators N. A. Schibrya, G. S. Shkrebtienko and I. I. Marchenko were already considering interspecific hybridization as one of the methods of practical plant breeding. They set the objectives of improving topinambur (H. tuberosus) by using sunflower, to increase the resistance of sunflowers to rust and broomrape (Orobanche cumana Wallr.), and to create a perennial sunflower producing tops (for forage), tubers, and seeds. Somewhat later I. I, Marchenko used the perennial tuberous species H. macrophyllus Willd. and H. subcanescens Gray in crosses to obtain annual sunflowers resistant to broomrape and rust.

In 1955 I. Georgieva-Todorova (Bulgaria) crossed the sunflower variety 3-18 with H. laetiflorus (Ed: H. lactiflorus in text) and H. rigidus in studies on the inheritance of agronomically valuable characters. The American workers Heiser (1945, 1947, 1951) and Long (1955, 1957, and 1959) have made interspecific crosses between wild species of the genus Helianthus on a rather large scale. Their work is taxonomic in nature.

At VNIIMK (USSR) work on interspecific hybridization of sunflower was begun by V. S. Pustovoit in 1935 with the aim of producing rust-resistant varieties. The annual form of Texas sunflower (H. lenticularis Dougl.) was crossed with VNIIMK varieties 3519, 1646, 6540, and 8931.

As a result of this relatively small scale work, hybrids were produced which in the variety trials were highly resistant to rust, and gave oil yields per hectare equal to good "zoned" (Ed: "licenced" in Canadian terminology) sunflower varieties.

In 1949 the physiologist A. Y. Panchenko undertook research on methods of vegetative hybridization of sunflowers with the objective of producing hybrids of cultivated sunflowers with perennial species possessing resistance to rust. He obtained hybrids between H. tuberosus L. and sunflower by the sexual method with a preliminary vegetative "approach" (Ed: by grafting).

Similar crosses were made by V. S. Pustovoit in 1950 to obtain hybrids resistant to broomrape and rust. This work of Panchenko and Pustovoit was stopped because the hybrids lost their resistance to a new fungus disease - downy mildew.

Fertile hybrids obtained at the Institute in 1958 between H. tuberosus and the sunflower variety VNIIMK 8931 were distributed to the following scientific research institutions: the All-Union Research Institute of Genetics and "Selection" (Ed: "Plant Breeding" in Canadian terminology) in the name of Yuriev; the Odessa Genetical Institute; Don Research Station of VNIIMK; Armavir Substation of VNIIMK; Stavropol Agricultural Research Institute; Krasnodar Agricultural Research Institute; and the Saratov Agricultural Research Institute of the South East. All these institutions are developing this work along various practical plant breeding lines. Interesting results were obtained at the Ukraine Institute in the name of Yuriev (V. G. Volf and I. V. Gretchko) in selecting interspecific hybrids for resistance to Sclerotinia libertiana. At present the Institute has F7 hybrids resistant to Sclerotinia.

We began to work on interspecific hybridization in 1955 because of the appearance on sunflowers of a new disease, downy mildew (Plasmopara helianthi Novot.), which we mistakingly referred to earlier as Plasmopara halstedii Berlese et de Toni. The mycologist N. S. Novotelnova has shown by her work that the heterogeneous species P. halstedii includes a specialized species with a restricted geographic distribution, narrower than of the species on Compositae, restricted to species of the genus Helianthus, with specific infection types and characteristic morphology of the sporangiophores. Within this species, which infects only species of the genus Helianthus there are three biological forms. One has annual mycelium and is specialized on the annual species of Helianthus; a second has perennial mycelium and is specialized on perennial species; the third also infects the perennial species, but differs from the second form in sporangiophore morphology.

The composite species P. halstedii, first described by Farlow in 1883, is American in origin and is polyphagoris, attacking 64 plant species. The geographic range of the new species P. helianthi Novot. is fairly strictly limited in latitude between 30 and 50 degrees north, whereas the composite species P. halstedii occurs over a much wider geographic range on Compositae (Novotelnova 1962).

Downy mildew of sunflower was first reported in Europe in the 1940's (Yugoslavia, Bulgaria, Rumania, Hungary, Poland, and other countries), and spread by various routes to the Soviet Union. Shortly after appearing in the Soviet Union (1950) the new fungus spread to the main sunflower regions: Northern Caucasus, Moldavia, and Ukraine. In recent years downy mildew has occurred sporadically in the Rostov and Voronezh Regions. Since 1951 there have been epiphytotics of downy mildew in the Kuban almost every year, with infection as high as 70%. The only effective control method available so far is crop rotation. Unfortunately this does not permit sowing sunflowers oftener than once in 10 years on the same field, restricting the crop to 10% of the available area.

First symptoms of the disease can be seen when the scond pair of true leaves appears. Growth of affected plants is retarded and their leaves appear mottled. If soil moisture and relative humidity of the air are sufficiently high, a white cottony growth appears on the underside of infected leaves. Petioles and stems of diseased plants are thickened, internodes are shortened, the heads do not hang down but remain horizontal. Seeds usually do not form in the diseased heads, or else remain very thin. Germinability of such seeds is sharply reduced. Studies on the response to artificial inoculation with downy mildew of 44 sunflower varieties licensed at various times in the Soviet Union showed that all were infected 100%. Argentine varieties (No. 21395 and 213562) and the later generations of rust-resistant interspecific hybrids (H. annuus x H. ruderalis) were equally susceptible. Selection for many years from the best VNIIMK sunflower varieties for resistance to downy mildew proved ineffective.

The absence of immunity to downy mildew in the cultivated sunflower varieties (H. annuus) made us turn to the wild species of the genus Helianthus. The genus Helianthus is a very large and polymorphous one of American origin. Bentham and Hooker described 50 species; according to Watson there are about 108 species; Cockerell reported about 180 species, and the Russian worker F. A. Saziperov reported about 264 species.

It is now believed (P. M. Zhukovski) that the genus Helianthus has a divided geographical distribution. Most of the species (about 50) are concentrated in North America (From Canada, thru USA to Mexico inclusive). Another group consisting of 17 species is located in South America, in the Andes, from South Columbia to Bolivia. Heiser considers Helianthus to be of diphyletic origin; the South American and the North American species groups originated independently, probably, from different sections of the genus Viguiera. Many species are now excluded from the genus Helianthus (about 41 species) and have been transferred to the genera Tithonia, Verbesina and others.

We used 26 wild sunflower species of North American origin to obtain hybrids resistant to downy mildew (Table 1).

The 26 North American species are ranked by their degree of ploidy. Most of them are perennial.

Phytopathological evaluation showed that only 11 species were completely resistant to downy mildew when inoculated by A. Y. Panchenko's method. Fifteen species were resistant to broomrape when inoculated by V. S. Pustovoit's method. Twenty-five species were resistant to rust under conditions of natural infection, and 8 species combine immunity to all three diseases. All species have an armored layer in the seed and are not affected by the sunflower moth (Homoeosoma nebulella). Inoculation of 20 perennial species with Sclerotinia by I. V. Grechka's method showed that 18 were totally immune to this fungus. Only two annual species, H. argophyllus and H. lenticularis were attacked by Sclerotinia, with 60% and 20% infection respectively.

Table 1 List of Wild Species of Genus Helianthus of North American Origin in the Nursery of VNIIMK

	Species	2n	Habit	Synonyms
1.	H. tuberosus v. purpurellus L.	102	perennial	
2.	H. macrophyllus Willd.	102	perennial	H. willdenovianus
3.	H. rigidus (Cass) Desv.	102	perennial	Thll. H. subromboides Ryd
4.	H. subcanescens Gray	102	perennial	
5.	H. tomentosus Nichx	68	perennial	
6.	H. lactiflorus Pers	68	perennial	
7.	H. scaberimus Ell	68	perennial	
8.	H. divaricatus L.	68	perennial	
9.	H. mollis Sam	34	perennial	
10.	H. gigantheus L.	34	perennial	
11.	H. grosse-serratus Martens	34	perennial	
12.	H. maximilliani Schrad	34	perennial	
13.	H. Nuttalli T. et G.	34	perennial	
14.	H. demmovi	34	perennial	
15.	H. microcephallus T. et G.	34	perennial	
16.	H. tracheliflorus Miller	34	perennial	
17.	H. californicus	34	perennial	
18.	H. multiflorus Hook	34	perennial	
19.	H. argialis D.C.	34	perennial	
20.	H. debilis Nutt.	34	annual	
21.	H. lenticularis Doug.	34	annual	
22.	H. argophyllus T. et G.	34	annual	
23.	H. petiolaris Nutt.	34	annual	
24.	H. mollis	34	perennial	Samples with wrong
25.	H. divaricatus	34	perennial	determination of
26.	H. rigidus	68	perennial	species
27.	H. gigantheus	34	perennial	
28.	Unidentified	34	perennial	1 bush

It must be pointed out that we succeeded in obtaining various degrees of infection of wild species with downy mildew only by artificial inoculation of the seedlings with zoospores by A. Y. Panchenko's method. Under conditions of natural epiphytotics none of the 24 perennial species was infected by mildew during 12 years of observation in field plots. Under field conditions at Krasnodar, during 12 years of observation, they also remained free of infection with the following diseases: Verticillium wilt (Verticillium dahliae Kleb), gray rot of above-ground tissues (Botrytis cinerea Pers.), dry rot of the heads (Rhizopus microsporus Namm.), bacterial infection (Bacterium sp.), and white rust (Cystopus tragopogonis Shred.). (Table 2).

In only one year, 1965, one species (H. demmovi) was affected by Fusarium root rot (Fusarium sp.), and one (H. maximiliani) showed a leaf mosaic attributed to genetic factors.

Table 2 Phytopathologic Characteristics of Wild Species of the Genus *Helianthus*

Species	Chromo-some No.	Infection: Under artificial inoculation: Broomrape, No. of flowering stalks/plant	Sclero-tinia (%)	Downy mildew (%)	Infection: Under natural infection: Downy mildew (%)	Rust (%)	White Rust (%) (Cystopus)
1. H. tuberosus L.	102	0	0	0	0	0	0
2. H. Subcanescens (gray) Wats.	102	0	0	0	0	0	0
3. H. macrophyllus Willd.	102	0.56	0	0	0	0	0
4. H. rigidus (coss) Desv.	102	0	0	0	0	0	0
5. H. tomentosus Michx.	68	1.9	0	14	0	0	0
6. H. lactiflorus Pers.	68	1.3	0	19	0	0	0
7. H. Scaberimus Ell.	68	0	0	22	0	0	0
8. H. divaricatus L.	68	0	0	4.5	0	0	0
9. H. gigantheus L.	34	0.11	0	11.3	0	0	0
10. H. maximiliani Schrad.	34	0.5	0	0	0	0	0
11. H. Nuttalli T. et G.	34	0.4	0	0	0	0	0
12. H. mollis Lam.	34	0	0	6.6	0	0	0
13. H. grosse - serratus Mart.	34	0.45	0	41	0	0	0
14. H. tracheliflorus Miller.	34	0.06	0	0	0	0	0
15. H. californicus	34	0	0	0	0	0	0
16. H. multiflorus Hoox.	34	0.07	0	2	0	0	0
17. H. argophyllus T. et G.	34	0	60	100	40	0	0
18. H. lenticularis Dougl.	34	5.2	20	100	65	10	0
19. H. argialis D. C.	34	0	0	8	0	0	0
20. H. petiolaris Nutt.	34	0	0	100	28	0	0
21. H. microcephallus T. et G.	34	0	0	0	0	0	0
22. H. demmovi	34	0	0	0	0	0	0
23. H. mollis (Italy)	34	0	0	6.5	0	0	0
24. H. debilis Nutt.	34	0	0	100	56	0	0
25. H. divaricatus (Italy)	34	0	0	0	0	0	0
26. H. rigidus (Italy)	68	0	0	0	0	0	0
27. H. annuus N 8931 VNIIMK	34	0	0.70	100	0.80	100	from 10 to 100

enabled us to test more than 25,000 hybrids, wild species, and varieties of sunflowers in the greenhouse during the winter of 1964-1965, and to sow in the nurseries only highly resistant hybrids.

Artificial inoculation with broomrape in the field and in the greenhouse was done by Academician V. S. Pustovoit's method. Since 1965 all parental materials for crossing the hybrids of all combinations of species, and all generations of crosses have been tested for Sclerotinia wilt resistance using the mycelial inoculation method suggested by I. V. Grechka. This method gave excellent results in inoculations on very large populations. Rust infections were rated under conditions of natural infection using the Melchers and Parker scale as modified by V. I. R.

Use of the above methods of artificial inoculation and rating enabled us to study thoroughly the resistance to a whole range of diseases of our source materials, and to set up a well-planned program of breeding for disease resistance.

The F_1 hybrids of topinambur with sunflower, 300 of them, segregated markedly for all plant characters. The hybrids were tentatively grouped into three categories: 1) Plants of the topinambur type; 2) plants of the sunflower type; 3) plants of a new type. All the F_1 hybrids were completely immune to rust, broomrape, downy mildew, and sunflower moth. Most of the hybrids (96%) were perennial forms with very striking heterosis in both number and size of tubers. All the hybrids flowered profusely, but average fertility did not exceed 22%. Seed production was also low. All the F_1 hybrids showed a very large heterotic effect: They grew up to 5 meters tall, produced very many leaves, produced from 1 to 500 flowers per plant and from 1 to 4.5 kilos of tubers per plant.

Cytological studies of the topinambur sunflower hybrids showed that allotetraploid forms ($2n = 68$) appeared in the F_1 generation, resulting in a significant number of viable gametes, so that F_1 hybrids are fairly fertile.

Hybrids of perennial habit were of secondary interest to us. Our objective was to obtain annual forms like the cultivated sunflower, with immunity to a series of fungus diseases, above all to downy mildew.

F_2 hybrids from backcrosses with sunflowers were completely sterile because of the formation of allotriploids ($2n = 51$). We overcame the sterility of the F_2 (backcrossing H. tuberosus x 8931 hybrids with sunflower) by using temperature shocks when the hybrids were undergoing meiotic division. This was done as follows: By adjusting sowing dates the flowering of the F_1 hybrids was timed for the first decade (10 days) of September when there are sharp diurnal changes in air temperature in the Krasnodar area. Starting in August and the beginning of September, we made backcrosses with sunflower on the H. tuberosus x 8931 hybrids, with meiosis occurring when temperature shocks were to be expected. By comparing the occurrence of fertile hybrids with meteorological data, it can be shown that for successful completion of meiosis it is essential that temperatures vary from a minimum between $2.2^{\circ} - 4.9^{\circ}C$, to a maximum between $33^{\circ} - 34.7^{\circ}C$, repreated at intervals of 7 to 9 days.

Unfortunately conditions favorable for overcoming the sterility of hybrids by this method do not occur very often in the Krasnodar area. In an 8-year period only three years (1957, 1962 and 1963) were favorable for this purpose (Table 4). It was therefore necessary to find some dependable method to overcome failure of crosses, and sterility of crosses between wild species of Helianthus with the sunflower.

Table 4 September Temperature Regime in Krasnodar Favouring Spontaneous Production of Highly Fertile Interspecific Hybrids.

Year	Mean temp. for 10-day periods 1 - 10	11 - 20	21 - 30	Mean monthly temp.	Mean temp. at 1 p.m.	Mean minimum temp.	Absolute minimum	Mean maximum	Absolute maximum	No of days with temperature 30°
1957	23.6	20.8	16.0	20.2	25.3	14.3	4.9	27.1	33.5	9
1962	20.2	16.4	20.2	18.9	25.0	12.1	4.1	26.8	34.7	7
1963	20.5	20.8	15.6	19.0	24.2	13.1	2.1	25.9	33.0	7

To accomplish this we used the method worked out by Michurin of preliminary vegetative approach (Ed: grafting) of the partners in the cross, followed by pollination with a mixture of pollen from the male parent. This method enabled us to make reciprocal crosses with wild species regardless of their degree of ploidy, and significantly increased the fertility of the hybrids without any further special treatment.

The use of Michurin's method of overcoming incompatibility and sterility of hybrids in "distant" crossing of wild species of Helianthus with sunflower (H. annuus) enabled us to obtain 18 fertile hybrid combinations with various wild species, of which 6 combinations are now in advanced generations (F_4 to F_{10}) (Table 5). Thirteen of these combinations were achieved by sexual crossing after preliminary "vegetative approach" by grafting.

The obtaining of later generations of fertile hybrids by crossing different wild species of Helianthus with sunflower varieties allowed us to synthesize di-specific and tri-specific hybrids possessing new biological properties and strong heterosis.

Cytological analyses of highly fertile hybrids showed that they all are allodiploid forms (2n = 34) resulting from a reduced parthenogenesis stimulated by temperature shocks. According to S. S. Zamotailov who made the cytological studies, the gametes of sunflower did not take part in the fertilization of the hybrids. Cytological studies of early generations of twelve hybrid combinations from crossing autohexaploid and autotetraploid species of the genus Helianthus with sunflower varieties showed that the occurrence of fertile hybrid forms of early generations is always related to the depolyploidization process resulting in allodiploid forms. Evidently the diploid state is an optimal ploidy level for the hybrids of the sunflower with wild species.

Table 5 Fertile Interspecific Hybrids at VNIIMK in 1966.

Hybrid source	Year of cross-ing	2n of parents	Gener-ation	Method of producing fertile hybrids
H. mollis x Peredovik	1963	34x34	F_2	Sexual crossing with preliminary vegetative approach
H. lenticularis x Peredovik	1963	34x34	F_2	Sexual crossing
H. divaricatus x Peredovik	1963	68x34	F_2	Sexual crossing with preliminary vegetative approach
H. scaberrimus x Peredovik	1962	68x34	F_3	"
H. multiflorus x Peredovik	1962	34x34	F_3	"
H. nuttalli x Peredovik	1962	34x34	F_3	"
H. giganteus x Peredovik	1962	34x34	F_3	"
Peredovik x H. Nuttalli	1962	34x34	F_3	"
Peredovik x H. argialis	1962	34x34	F_3	"
H. argophyllus x Peredovik	1962	34x34	F_3	Sexual crossing
8931 x H. scaberrimus	1961	34x68	F_4	"
8931 x H. lactiflorus	1961	34x68	F_4	Vegetative approach (side grafting)
H. rigidus x Smena	1961	102x34	F_4	"
8931 x H. tomentosus	1960	34x68	F_5	"
H. tomentosus x 8931	1959	68x34	F_6	Thermal shocks
8931 x H. rigidus	1958	34x102	F_7	Vegetative approach
H. macrophyllus x 8931	1957	102x34	F_8	Thermal shocks
H. tuberosus x 8931	1956	102x34	F_9	"

F_2 hybrids (H. tuberosus x 8931) appeared totally immune to broomrape, downy mildew, rust, and sunflower moth. Segregation for other characters occurred. Ninety-five percent of the hybrids were annual forms.

In 1959 hybrids were sown in a specially infested field of 1.5 ha., using the "pairs method" with the male parent, 8931, as the control variety. The F_3 hybrids (backcrossed twice with sunflower pollen) segregated for reaction to the three diseases. The F_3 hybrids proved susceptible to broomrape; this was quite unexpected as the maternal parent H. tuberosus was immune and the parental parent 8931 was highly resistant. Apparently there was an interaction of complementary dominant genes none of which individually controlled susceptibility to broomrape. Breeding sunflowers for resistance to rust and sunflowers is hampered by the development of these diseases after flowering of the hybrids, so that susceptible plants are discarded after pollination has been done, with pollen from plants of varying degrees of resistance. F_3 plants were rogued three times, removing all individuals susceptible to downy mildew, rust, broomrape, and such undesirable agronomic characters as late maturity, excessive height, branching, etc. Plants remaining in the nursery were all uninfected and of desirable sunflower type, and were used for subsequent breeding work.

To avoid loss of resistance to mildew as a result of excessive backcrossing to the sunflower parent, crossing was essentially intrahybrid from the F_3. Crosses of the F_3 material were made in one of four ways: Group pollination of the best lines with a mixture of their pollen; paired crosses between the best plants; backcrosses of the F_3 hybrids with the best sunflower varieties; and open pollination in plots of the best hybrids after severe roguing. The three first methods were used on plants protected with paper bags.

By 1960, after complex work with F_3 hybrids, 200 out of 1200 families in the F_4 were totally immune to downy mildew. The degree of infection of 400 families was 1-5%, and the others were discarded susceptible. All hybrid families were divided into three groups according to their response to downy mildew: 1. Immune group consisting of families totally unaffected by downy mildew and with completely infected controls; 2. Group of resistant families affected not more than 1%; 3. Group of weakly susceptible families (the disease did not exceed 5%). All families with more than 5% infection were discarded. Further breeding was conducted within the above groups.

Evaluation of the hybrids for resistance to broomrape was done in infested plots in the field, and in the greenhouse by a vegetative method in winter. Controls in these trials were the variety A-41, susceptible to all broomrape races of group B, and the sunflower variety 8931 resistant to them. In three years, by crossing resistant plants with subsequent repeated individual selection, we succeeded in getting about 200 familes resistant to broomrape.

Selection of hybrids resistant to sunflower rust was made in the same nurseries under conditions of natural epiphytotics. Three-year selection among interspecific hybrids resulted in forms resistant to sunflower rust. Evaluation of F_5 hybrids for reaction to various diseases showed that an appreciable number of families possessed "group resistance" to sunflower rust, broomrape, and downy mildew.

The study of interspecific hybrid for resistance to Sclerotinia is of special interest. This fungus has multiplied to such an extent in the last 10 years that epiphytotics now occur annually, infecting up to 40 to 70% of the sunflower heads on tremendous acreages, reducing the yield and the quality of the seed for sowing.

Sclerotinia is particularly widespread in the foothills of the Northern Caucasus, the Central Black Soil region, Moldavia, Ukraine, the Altai and Krasnoyarsk provinces, eastern Kazakhstan, and other districts.

It was not possible to select for resistance to Sclerotinia because this fungus does not occur in Krasnodar. Development by I. V. Grechka of a method of artificial inoculation of sunflower with Sclerotinia made it possible for us to evaluate the resistance of our breeding materials, and interspecific hybrids of all generations and all combinations.

We found that the 11 best varieties of sunflower commercially grown in the Soviet Union on an area of about 4 million hectares were completely susceptible to this fungus. These varieties were VNIIMK 8931, VNIIMK 6540, VNIIMK 8883, Peredovik, Smena, Armavirski 3497, 3495, Mayak, Enissey, Armaviretz and Kubanetz.

F_1 and F_2 interspecific hybrids from crosses of 9 perennial species with sunflowers were highly resistant to this fungus, but as a result of segregation the hybrids are increasingly affected in the F_3 and later generations. Inoculation of 300 lines of F_9 (H. tuberosus x 8931) with a group immunity to broomrape, sunflower rust, downy mildew and sunflower moth revealed that only 5 lines were totally resistant; the rest had various degrees of resistance, and 12 were completely susceptible to Sclerotinia.

The occurrence of families resistant to Sclerotinia in the hybrids makes it possible to plan a breeding program for sunflower varieties with resistance to a group of diseases including this important one.

Thus the potential immunity of interspecific hybrids is increasing significantly as the result of inheritance from topinambur and other wild sunflower species of resistance to a whole range of diseases which we did not consider when we started this work. Most of the hybrids are resistant, not only to the diseases mentioned above, but also to Verticillium wilt (Verticillium dahliae Kl), bacterial disease (Bacterium sp.), and are partially resistant to Rhizopus rot (Rhizopus microsporus) and white rust (Cystopus tragopogonis Shred).

It must be emphasized that inheritance of disease resistance in sunflower is complex, and differs for every disease. Resistance is not inherited as a unit to all diseases.

The agronomic properties of later generation hybrids (oil content, hull percentage, weight per volume, and 1000 kernel weight) show intermediate inheritance. Most plants have a reduced oil content and an increased hull percentage. Trying to speed the process of making the hybrids agronomically acceptable by crossing them with high oil yielding sunflower varieties resulted in increasing their oil content greatly, and almost total loss of resistance to downy mildew.

Some families obtained by intra-hybrid crossing, selected on the basis of laboratory and biochemical tests, were found to have significant resistance to downy mildew, and to be as good as the best sunflower varieties in a whole range of agronomic characters. (Table 6).

In the 1965 elite breeding nursery 500 families were selected with resistance to downy mildew equal to that of the wild parent. Germinating seeds were not infected even when exposed to any inoculum of 250,000 zoospores per cc (counts were made with a Goryayev counting chamber). Nearly 400 families of highly resistant hybrids had high oil yield, high weight per 1000 seeds, low hull percentage, and high weight per volume. (Table 7).

A preliminary evaluation of progenies of 300 lines by Academician V. S. Pustovoit's system, showed that in the first-year nursery a series of families combined immunity to a group of diseases with high seed yield. They exceeded the control by 820 to 1105 kilos/ha., or increases of 25.4 to 36.6%. (Table 8).

Table 6 F_9 Hybrids of H. tuberosus x 8931 Outstanding in Oil Content of Seeds - VNIIMK 1965

Analysis No.	% infection of downy mildew	% infection of rust	% seeds with armored layer	% of husk	Absolute weight	% oil in kernel dry basis	% oil in seed dry basis
5533	3	0	100	17	80	61.5	52.1
212	0	0	100	19	61	62.2	51.5
82	1	0	100	20	60	62.2	51.0
1738	1	0	100	23	77	62.9	50.1
736	2	0	100	19	57	61.2	50.9
537	2	0	100	19	57	61.4	50.6
270	2	0	100	25	49	65.2	50.4
235	2	0	100	25	57	65.2	50.4
Check average N 8931 VNIIMK	100	100	100	21	66.7	63.1	51.3

Table 7 F_9 Hybrids of H. tuberosus x 8931 Outstanding for Agronomic Characters and Disease Resistance - VNIIMK, Krasnodar, 1965

Analysis No.	% infection of downy mildew	% infection of rust	% infection of broom-rape	% of seeds with armored layer	% of husk	Absolute weight /1000 seeds	% oil in kernel dry basis	% oil in seed dry basis
1963	0	0	0	100	21	64	61.8	50.3
178	0	0	0	100	20	54	61.2	50.2
2016	0	0	0	100	21	79	61.6	49.9
154	0	0	0	100	23	65	63.4	49.9
207	0	0	0	100	21	65	61.3	49.6
Check average N 8931	100	100	0	100	20	66.7	63.1	51.3

Table 8 F_9 Hybrids of H. tuberosus x 8931 Outstanding in Seed Yield and Immunity in the First-Year Nursery Test

Elite No.	Hybrid Source	% plants downy mildew	Volume weight g/l	1000 seeds (g)	Seed yield 100kg/ha.	Yield gain with reference to check centner/ha	Yield % of check
28899	H. tuberosus x 8931	0	459.0	71.4	40.8	11.0	136.6
	Check - 8931	100	390.4	80.8	29.8		
28796	H. tuberosus x 8931	0	441.6	70.7	42.5	9.9	130.0
	Check - 8931	100	424.4	78.5	32.6		
28960	H. tuberosus x 8931	0	478.0	63.0	38.2	8.5	128.0
	Check - 8931	100	390.4	80.8	29.8		
28949	H. tuberosus x 8931	0	448.0	68.2	40.4	8.2	125.4
	Check - 8931	100	389.0	72.4	32.2		

In 1965, after 5 years selection for productiveness, 30% of the heads in the elite breeding nursery had from 1,900 to 4,500 seeds per head. Weight per 1000 seeds was significantly higher than that of good sunflower varieties. (Table 9).

Although oil content of the seeds of most of the hybrids is lower than that of the control (VNIIMK 8931), the yield of oil per hectare of a whole series of hybrids is significantly higher than that of the control (by 119 to 384 kg/ha.). (Table 10).

In spite of great difficulties encountered in the breeding process, "distant hybridization " of the cultivated sunflower with wild species of Helianthus has permitted us to obtain plants of the cultivated sunflower type, with "group immunity" to broomrape, rust, downy mildew, and sunflower moth; with great possibilities of incorporating resistance to other diseases; and with sharply increased seed yields, and yield of oil per hectare.

Table 9 — F_9 Hybrids of *H. tuberosus* x 8931 Outstanding in Agronomic Characters

Plot No.		Percent infection downy mildew	Percent infection rust	% seeds with armored layer	Volume weight g/l	1000 seeds weight g.	Plant height cm	Growing season
30177	F_9/H.tuberosus x 8931	0	0	100	443.0	59.1	184	84
	Check - 8931	100	100	100	418.0	66.9	178	85
29981	F_9/H.tuberosus x 8931	0	0	100	422.8	54.8	190	89
	Check - 8931	100	100	100	406.8	63.3	177	85
30220	F_9/H.tuberosus x 8931	2	0	100	424.6	61.5	165	85
	Check - 8931	100	100	100	418.8	66.9	174	86
29987	F_9/H.tuberosus x 8931	0	0	100	455.2	56.9	175	88
	Check - 8931	100	100	100	434.8	71.0	169	84
30233	F_9/H.tuberosus x 8931	2	0	100	453.6	62.7	174	85
	Check - 8931	100	100	98.9	419.6	61.2	169	86
30275	F_9/H.tuberosus x 8931	2	0	100	460.0	65.5	208	93
	Check - 8931	100	100	100	394.0	62.6	173	86
300075	F_9/H.tuberosus x 8931	0	0	100	450.8	64.5	180	86
	Check - 8931	100	100	100	421.0	73.6	181	86
30227	F_9/H.tuberosus x 8931	0	0	100	448.6	66.6	176	87
	Check - 8931	100	100	100	412.8	62.9	170	86

Table 10 F_9 Hybrids of *H. tuberosus* x 8931 Outstanding Oil Yield per Hectare in Progeny Tests in the First-Year Nursery

Elite No.		Percent of			Seed yield 100 kg/ha.	Oil yield 100 kg/ha.	Oil gain with respect to check kg/ha	%
			oil in kernel dry basis	oil in seed dry basis				
30177	F_9/H. tuberosus x 8931	21.5	61.6	49.68	35.34	15.43	384	33.1
	Check - 8931	21.2	62.6	50.66	26.64	11.59		
29981	F_9/H. tuberosus x 8931	25.4	61.9	47.75	33.99	14.29	311	27.7
	Check -N8931	22.3	63.3	49.80	25.46	11.18		
30220	F_9/H. tuberosus x 8931	22.5	63.4	50.56	35.66	15.89	261	19.6
	Check -N8931	22.5	63.7	50.80	29.69	13.28		
29987	F_9/H. tuberosus x 8931	28.1	53.9	40.27	40.57	14.39	191	15.2
	Check -N8931	22.4	60.5	48.30	30.52	12.48		
30233	F_9/H. tuberosus x 8931	26.5	63.6	48.43	30.00	12.79	154	13.6
	Check -N8931	21.3	63.3	51.97	24.63	11.25		
30275	F_9/H. tuberosus x 8931	32.6	62.5	44.16	33.38	12.98	149	12.9
	Check -N8931	21.6	63.3	50.99	25.55	11.49		
30075	F_9/H. tuberosus x 8931	25.5	60.1	46.31	29.95	12.23	121	10.9
	Check -N8931	22.1	63.3	50.71	24.60	11.02		
30227	F_9/H. tuberosus x 8931	26.8	62.1	47.12	33.42	13.85	119	9.5
	Check -N8931	21.8	63.9	51.36	27.96	12.66		

BREEDING SCHEME

Breeding the interspecific hybrids (H. tuberosus x 8931 VNIIMK) for group immunity to the broomrape, sunflower rust, Plasmopara and Homoesoma.

All work was conducted on a highly infected background.

First Year - Preliminary sexual crosses were made under head bags without emasculation of the H. tuberosus with the high oil-yielding variety VNIIMK 8931. The female parent was H. tuberosus. Allotetraploid forms were obtained ($2n = 68$) in F_1 having a low fertility.

$$2n = \frac{102 + 34}{2} = 68$$

Second Year - To overcome the sterility of the F_2 hybrids, the F_1 hybrids (H. tuberosus x VNIIMK 8931) were backcrossed with a pollen mixture of the male parental form and treated by temperature shocks during their meiotic division.

As a result of the reduced parthenogenesis stimulated by the temperature shocks allotetraploid sterile forms were not obtained as in the preceding experiments, but highly fertile hybrids on a diploid level ($2n = 34$). Without temperature shocks meiosis takes place as follows:

$$2n = \frac{68 + 34}{2} = 51 \text{ totally sterile allotriploids.}$$

With temperature shocks the meiosis takes place as follows:

$$2n = \frac{68}{2} = 34 \text{ - a highly fertile allodiploid}$$

form obtained as a result of a reduced parthenogenesis stimulated by temperature shocks. In the last case the gametes of the sunflower did not take part in the fertilization.

Third Year - Highly fertile F_3 hybrids were obtained by further (backcrosses) saturating crosses of alloploid F_2 generation with the sunflower. Thus, the sunflower pollen took part in the fertilization twice - in F_1 and F_3.

Fourth Year - To prevent the loss of immunity of the hybrids, starting with the F_3 generation the intrahybrid crosses were made of resistant forms thus lengthening the period of breeding the hybrids of the sunflower type.

In F_3 generation rigorous selection was conducted on the hybrids with undesired properties and further hybridization of the best plants took place. Crosses were in pairs, with the mixture of pollen, "saturating" with the sunflower and free cross-pollinations of the best biological types. The first three types of crosses were made using the oil paper bags.

Fifth and Sixth Years - In F_4 and F_5 generations the sorting of families with a group immunity to the broomrape, sunflower rust Plasmopara and Homoeosoma were conducted using the method of intrahybrid crosses of resistant plants with consequent repreated individual selection.

Beginning with the F_4 generation all the selected elite plants were evaluated in field trials in infected conditions and in the greenhouse for their resistance to mildew and broomrape in the conditions of artificial infection. Resistant selected elite plants after laboratory studies of their percentage of husk, armoured layer and absolute seed weight were studied at the biochemical laboratory for oil percentage.

Seventh Year - The best plants with a group immunity, a high oil yield and seed yield and other useful properties were sown in the greenhouse of the selected elite plants, designed as a nursery of directed cross-pollination type. Here in conditions of circumstantial isolation and good field technique the plants were spaced 70 cm by 70 cm with a single plant in the bed with a free-controlled cross pollination. Thus the selected elite plants were grown.

The best plant heads after rigorous selection were threshed individually and after evaluating them in the greenhouse, laboratory and biochemical laboratory were studied according to the scheme of the selection of the Academician V. S. Pustovoit.

COMMENTS BY DR. SACKSTON DURING PRESENTATION OF MME. PUSTOVOIT'S PAPER

They use a check row, two test rows, and a check row. All comparisons are made exclusively on the basis of paired comparisons with the adjacent check. They don't use as many replications as we do but in comparing every test row to an adjacent check they feel that they can pick up very, very small differences. They have a great deal of faith in this. It takes a lot more space than our method of replication with very few checks. All their critical work is done on this pairing of every row with its adjacent check.

Don't take the term 'Michurin' as a challenge. The intent here is to describe a specific grafting technique as proposed by Michurin for some of his work. It is a method of bringing things together in intimate vegetative contact which has apparently facilitated their subsequent work. They used other grafting techniques as well.

Commenting on a slide showing Orobanche cumana. The stalks of sunflower are standing at normal height. The flowering stalks of O. cumana are less than a foot high, have a very tremendous number of flowers and a fantastic load of seeds. The individual seed is smaller than tobacco seed, very, very minute. They ripen about a month and a half before the sunflowers and re-infest the soil. By the time the sunflowers are ripe only the empty flowering stalk or rachis of Orobanche remains.

Commenting on a slide showing the hybrid in the 9th generation. It is in no way distinguishable from the cultivated sunflower. It is a heavy pollen producer; there is pollen all over the upper leaves. The head diameter under good conditions reaches 12 inches. It is a single stem, single-flowered plant. They have managed to get away completely from the branching habit and in every respect - height, maturity - it is a cultivated sunflower.

DISCUSSION

Heiser: May I ask for repetition of the mixture of pollen?

Sackston: The idea of the mixed pollen is the assumption that any given species is a population varying in its fertility and crossability. When pollinating with Helianthus annuus, the cultivated sunflower pollen, they took pollen from perhaps 50 to 100 bagged heads of the variety which they wanted to use as the pollen parent, bulked that pollen and then put it on the tuberosus flowers. It wasn't a question of mixing species. It was just pollen from a group of individuals. When they made the reciprocal cross then again they took the pollen from various bagged heads of only the one species but bulked the pollen from individuals on the assumption that otherwise they might hit an infertile pollen sample. Their cytologists have found that when they use this bulked pollen method that there seems to be competition between the germinated pollen tubes and only the most vigorous and rapidly growing ones actually effect the pollination.

Kinman: Bulk pollination is a better term than mixed, we use this.

Heiser: What is the height of their sunflower?

Sackston: The average height varies from about 140 to 180 centimeters which would be from $4\frac{1}{2}$ to $5\frac{1}{2}$ - 6 feet, in other words not very different from what we have here. Some of the early maturing varieties are only one metre tall or even shorter, in other words they have got to the dwarf.

Orellana, R.G.: What causes bacteriosis?

Sackston: They don't know.

* * * * * *

ADDENDUM

The following cablegram was sent to Academician V. S. Pustovoit, VNIIMK, Krasnodar, Russia, on August 23, 1966.

Academician Pustovoit
VNIIMEMK
Krasnodar, Russia.

With great pleasure I convey this message delivered verbally by Gordon Ross to the Second International Sunflower Conference at the moment Galina Pustovoit completed her report. "As a representative of industry who has worked with sunflowers more than twenty years I must comment. Scientists work hard and encounter many difficulties and disappointments usually with little recognition. This is particularly true of agricultural scientists. We all regret that eminent agricultural scientist Academician Pustovoit was unable to attend to describe his work personally. We extend to him sincere appreciation and acknowledgement of his tremendous contribution. We congratulate him on his achievements and on the excellence of his personal ambassador and representative Galina Pustovoit who impressed us with her very important work."

My personal regards,

Putt.

It has been gratefully acknowledged.