

COLLABORATIVE MEXICO/UNITED STATES INITIATIVE TO BREED FREEZE TOLERANT FRUIT AND FORAGE OPUNTIA VARIETIES.

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Introduction

Texas and northern Mexico are marginal for Opuntia fruit production due to their susceptibility to 15-year record freezing temperatures that may reach - 12° C (10° F). In previous reports of the Texas Prickly Pear Council (Borrego-Escalante et al., 1990; Felker et al., 1991; Nobel, 1990) we have discussed problems and opportunities specific to Kingsville, Texas and Saltillo, Mexico for overcoming the freeze sensitivity of Opuntia fruit producing clones.

Recognizing the benefits of freeze-hardy fruit producing Opuntia clones for both northern Mexico and Texas, we have formed a team project, with modest funding from the U.S. Department of Agriculture, to develop more freeze hardy Opuntia fruit varieties. This manuscript will integrate previous work by these groups and outline the strategies which are now being undertaken to develop freeze hardy fruit varieties.

Literature Review

Texas A&I University germplasm collection

In 1984, with National Science Foundation funding, a germplasm collection of Opuntias was established at Texas A&I University that have been used in other parts of the world for either fruit, vegetable, or forage production.

From this collection, a highly desirable vegetable type, clone 1308, was identified from the genus Nopalea. This clone had almost no glochids or spines, produced small, tender cladodes all year round when temperatures exceeded 25° C and had very low mucus content. The very low frost tolerance of this clone was not important since it was grown under plastic to enhance its growth rate in the winter.

Highly productive spineless Opuntia clones with potential for forage production were identified in this trial. Of especial interest was the Brazilian forage clone known as "Palma redonda" since it was significantly higher in protein (11-12 %) for several age classes than other clones in this field trial (Gregory and Felker, 1991).

Most of the clones evaluated for fruit in the Texas trial were of Mexican origin, although there were a few clones from Chile, S. Africa, Brazil and Algiers. The Brazilian and Mexican clones had the greatest production in the first 5 years of this study. The Brazilian clones 1270 and 1271 and the Mexican clones 1280, 1294, and 1301 appeared especially promising for abundant fruit production at young ages. Clone 1270 was the only clone to produce a second fruit crop in the fall. This delayed fruit production could be an important economic factor to produce fruit in the "off-peak" season when prices

would be higher. While the Chilean clones produced little fruit, one of these clones (1321) had the fruit with the greatest sugar content of 13.7%.

Several clones such as 1270, 1271, 1280, 1287 and 1288 had consistently high sugar content over all 3 years of this study. Other clones, i.e. 1292, 1294 and 1301 failed to produce high sugar content fruit consistently in all 3 seasons, or had generally low sugar content fruits. While the analytical data indicate that clones 1270, 1271, 1280, 1287 and 1288 all had similarly high sugar contents of 11 to 13 %, it is Felker's opinion that clones 1287 and 1288 tasted sweeter than the other clones. In addition, the yellow fruited 1287 possesses a "fruity" flavor.

These clones exhibited considerable variation with respect to fruit color. The Chilean fruits typically are a lime-green color and the Brazilian clones had yellow to yellow/orange fruits. The Mexican varieties exhibited a great deal of variation with some being red, orange, and lime-green color. The color of the inner pulp and juice was not always identical to the outer peel. This occurred mainly in cultivars which had an orange pulp and a red to yellow peel. The fruit of the Mexican clone 1288 is a light-lime green but is listed here as white, since this is the literal translation of their most common name "blanca" in Mexico.

The Texas A&I collection experienced major damage from freezes of - 9° C in 1984/1985 (Russell and Felker, 1987), -12° C in 1989/1990 and - 7° C in 1990/1991. In the winter of 1989/1990 all of the clones mentioned above froze to ground level, but some of them resprouted from the base. None of the cold hardy clones reported by Borrego et al. (1990) were present in the field during the 1989 freeze. The clones of most northern origin, which all froze to the ground, were from San Luis Potosi, which is about 270 km south of Saltillo.

In the less severe freeze of 1990, much variability was observed in freeze damage. The spineless O. robusta clone 1241 from South Africa, and the spiny Algerian clone of robusta type 1253 had essentially no damage from the freeze. Other promising cold hardy clones included the spiny Mexican clones 1274 and 1298 and the spineless Chilean clone 1321. Unfortunately, neither the spiny clones 1274 or 1298 have yet set fruit, so it is not possible to ascertain the fruit potential. The Chilean spineless clone 1321 had the greatest frost resistance of all the O. ficus-indica, and while it had the greatest sugar content (13.7%) of all the clones, it only produced several fruit.

Some of the fruit clones with the highest sugar content had low resistance to freezing weather. For example, the spiny Mexican clones 1288 and 1287 that produced high sugar content fruits 3 years in a row had only 19 % and 26 % freeze resistance.

Due to their promising fruit quality, cladodes of 1288 and 1287 were removed from the mother plant after the first night of the disastrous Christmas, 1989 freeze and propagated by tissue culture.

After the freeze of Christmas 1989, a spineless Opuntia clone that suffered no damage from the freeze was discovered by County Agent Buddy Johnson near Crystal City, Texas. This clone survived the -12 C freeze of Christmas 1989 without damage,

when all O. ficus-indica, O. robusta types and other spineless types froze to the ground. This Opuntia appears to be O. ellisiana Griffiths as described by Griffiths (1915). While this clone has outstanding freeze hardiness, its fruits and cladodes are even smaller than the native O. lindheimerii. Quantitative data on its growth rate are not available, but 2-year-old O. ellisiana are only about 60 cm tall. In contrast, clones of cold hardy Opuntia ficus-indica and O. megacantha from the Saltillo collection, in adjacent field plots of the same age from Saltillo, are 2 m tall. This O. ellisiana could be useful material in breeding studies to enhance cold hardiness of fruit and vegetable fodder cactus varieties.

Universidad Autonoma Agraria Antonio Narro germplasm collection

For over 30 years the Universidad Autonoma Agraria Antonio Narro (UAAAN) at Saltillo has been engaged in research to develop cold hardy fruit, forage and vegetable Opuntia clones for the colder regions of northern Mexico. Saltillo is just north of the mountain range that protects Mexico's central plateau (Matehuala, San Luis Potosi and Mexico City) from cold arctic fronts that influence the climate of Texas. Saltillo is at a sufficiently high elevation (1500 m) to be considerably colder than more northern regions in the Rio Grande Valley.

An English version of the description of UAAAN's Opuntia clones, previously reported in Spanish (Borrego-Escalante et al., 1991), is presented in Table 1. Of especial interest with regard to tolerance to freezing weather is the fact that clone AN-V1 was selected from an individual in a plantation west of Saltillo that survived temperatures of -14° C (7° F) to -12° C (10° F) in 1984. The varieties AN-TV1, AN-TV2 and AN-TV3 came from seed in a mass selection program made by Dr. Lorenzo Martinez Medina after the freeze of -16° C (3° F) in 1962.

In March 1991, Borrego-Escalante of UAAAN and Felker of Texas A&I conducted a germplasm exchange of the 15 UAAAN clones and of the best clones at Texas A&I University financed by Mr. John B. Armstrong. The UAAAN clones were immediately planted in the field at Texas A&I University and in December, 1992, after two growing seasons, most of them were 1.5 to 2.0 m tall. There has been no fruit production or freezing weather from the spring of 1992 through December of 1992. In the winter of 1992/1993 these clones were fertilized with 100 kg/ha of N, P, and K.

Cold hardy Opuntia fruit clones obtained at 2,300 m elevation near Saltillo

In order to augment the excellent germplasm of cold hardy Opuntia fruit clones obtained from UAAAN, members of the Texas Prickly Pear Council, including Felker, made a collection trip to the mountains near Saltillo. Dr. Valdemar Gonzalez of the Mexican Department of Agriculture provided assistance in locating good sources of high elevation genetic materials.

At the small village of Escobedo, about 10 km west of the top of the pass of the Saltillo/Matehuala highway, excellent material was located. This site was at about 2,300 m (7,000 ft) and, according to villagers, often experiences temperatures of -12° C (10° F). Even in October during the collection, many of the varieties had fruit, so the varieties could be tasted as well as collected. At Escobedo, cladodes were purchased of each of

the following varieties [numbers in parenthesis are newly assigned Texas A&I accession numbers]: (1) spiny cactus with purple fruit (morado) [1404], (2) spiny cactus with white fruit (blanca) [1401], (3) spiny cactus with yellowish-pink fruit with slight banana taste (chapeada) [1402], (4) a spineless-red fruited variety (pelon) [1406], (5) a spiny red-fruited variety (roja) [1405], (6) a spiny deep-orange-fruited variety called 'amarilla' (normally amarilla is yellow but this one was not) [1403]. About 50 pads were obtained from all the varieties, except for the spineless red-fruited variety for which 200 pads were obtained. A few pads were obtained from Carlos Molina of a very large red-fruited variety [1407].

From Escobedo, we proceeded several Kilometers east to Santa Rita, which is the highest point in the valley and where snow was common. Pinyon pine trees were growing adjacent to the cactus at this location, indicating the cold nature of the climate. Spiny white and red fruit types were found at this location, but only the spiny red type was collected [1408].

We then proceeded to the lowest portion of the valley, to the town of Los Llanos, where due to cold air drainage, the lowest minimum temperatures were obtained. Here pads of a yellow (amarilla) variety [1398], a white variety (blanca) [1399] and red variety [1400] were obtained. The red variety was of the species Q. robusta, and while attractive in having thick round, bluish pads, its large fruits were not sweet. The amarilla variety at this location was a yellow (not orange) variety. Due to the thicker peel of the yellow variety, the grower said it would be a better shipping variety.

While frost had already occurred in October at this location and as snow and temperatures of -12° C were common, there is a possibility that these clones may not be adaptable to Texas conditions due to acclimatization problems. In this mountainous region of Mexico, it gets cold early in the fall and stays cold throughout the winter. In contrast, temperatures in south Texas may be 27° C for several days in December and -12° C several days later.

It was truly amazing to observe such great genetic diversity in Opuntia fruit clones over such a short distance. It is indeed a travesty that this genetic material has not been collected and placed in germplasm banks.

Goals and objectives of the Opuntia hybridization program.

One of us, Barrientos-Perez, has been actively involved in Opuntia breeding since 1958 and has created and released 16 genetically improved Opuntia clones, designated as Copena 1 through 16. Nevertheless, there has never been an emphasis on breeding Opuntia species for the extremely cold regions of northern Mexico/southern United States.

With identification of (1) well-adapted fruit, vegetable and fruit varieties at UAAAN, (2) disease resistant, high-fruit-producing, but freeze sensitive, varieties in Texas, and (3) a thornless Opuntia variety with extreme resistance to freezing weather, all of the pieces of the genetic puzzle are available to create cold-hardy useful varieties.

Our objectives are:

1. To produce more cold hardy Opuntia fruit varieties for northern Mexico and southwestern United States.
2. To produce freeze hardy, spineless, high-protein forage varieties.
3. To examine the genetics of freeze hardiness.
4. To evaluate the UAAAN collection for freeze hardiness in the United States.

Procedures

We will use O. ellisiana as the primary source of cold hardy genes. O. ellisiana will be used as the male and female parent in crosses with other clones. The emasculation technique developed by one of us (FBP) will be used in the crosses. In order to reduce sterility barriers between diverse species, grafts of O. ellisiana onto the intended hybrid parent will be made.

We will hybridize O. ellisiana with the 15 clones in the UAAAN collection, with the Texas A&I fruit producing clones 1288 (tuna blanca) and 1287 (tuna durazno), with the Chilean tuna varieties and with the high-protein-producing forage clone 1270. The progeny of these crosses will be evaluated at Colegio de Postgraduados in Chapingo, at UAAAN in Saltillo, and at Texas A&I University.

In considering a hybridization program, it is important to review the cytogenetics of possible hybrid combinations. The Opuntia wild types are diploid, while most of the commercial fruit types are tetraploid or octaploid hybrids resulting from natural hybridization with the wild types.

Octaploid Opuntia species ($2n=8x=88$) actually have mixed polyploidy (disomic/polysomic). Some genes should reveal disomic inheritance, while genes in other genomes should reveal tetrasomic inheritance. The specificity of pairing in disomic polyploids, which prevents pairing between homeologous chromosomes, greatly influences fertility and genetic segregation. Polysomic polyploids differ from disomic polyploids in having generally reduced fertility. Heterozygosity has been recognized as an element of polyploidy in random mating populations having comparable frequencies, polyploids will be much more heterozygous than diploids. The process of genetic improvement by interspecific hybridization in Opuntia has a lot of possibilities to produce cultivars improved for their processing value with environmental tolerance to herbicides as well as cold tolerance. For example, hybrids which have been made between O. amyclaea and O. ficus-indica species segregate for glyphosate tolerance which, according to Wilkins, (1991), is a non-toxic biodegradable herbicide.

In breeding for cold hardiness it is important to select for fruit color since consumers of European descent in New York, Chicago and eastern United States have historically preferred red fruit, even if the sugar content were the same. The excellent red-fruited variety named "Frieda" that was recently released by one of us (FBP) should have excellent demand.

It is not desirable to hybridize O ellisiana with nopalito varieties since we wish to produce nopalitos year round, and cold-hardy varieties produce minimal growth in the fall. This is evidently the result of a hardening-off mechanism.

Funds for the travel of FBP to Texas A&I to make these crosses have been obtained by the USDA Cooperative State Research Service. Unfortunately, the funds arrived too late in 1992 to make hybrids, but an intensive hybridization program is scheduled for April/May of 1993.

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Table 1. Description of agronomic characteristics of 16 genetically superior *Opuntia* clones in the Universidad Autonoma Agraria Antonio Narro collection.

UAAAN Code	Latin Binomial	Intended use	Fruit color	Cladode shape	Chromosome number
AN-V1	<i>Opuntia ficus-indica</i>	vegetable	white	elongated	88
AN-V2	<i>Opuntia crassa</i>	vegetable	yellow	circular	77
AN-V3	<i>Opuntia crassa</i>	vegetable	red	egg shaped	77
AN-V4	<i>Opuntia crassa</i>	vegetable	red	circular	88
AN-V5	<i>Opuntia megacantha</i>	vegetable	yellow	elongated	88
AN-T1	<i>Opuntia megacantha</i>	fruit	yellow	elongated	66
AN-T2	<i>Opuntia megacantha</i>	fruit	white	egg shaped	88
AN-T3	<i>Opuntia megacantha</i>	fruit	white	egg shaped	88
AN-T4	<i>Opuntia megacantha</i>	fruit	white	egg shaped	66
AN-TV1	<i>Opuntia amyclaea</i>	fruit & veg.	white	elongated	88
AN-TV2	<i>Opuntia amyclaea</i>	fruit & veg.	white	elongated	88
AN-TV3	<i>Opuntia amyclaea</i>	fruit & veg.	white	elongated	88
AN-TV4	<i>Opuntia amyclaea</i>	fruit & veg.	white	elongated	88
AN-TV5	<i>Opuntia crassa</i>	fruit & veg.	red	circular	88
AN-TV6	<i>Opuntia megacantha</i>	fruit & veg.	red	elliptical	66
AN-F1	<i>Opuntia megacantha</i> x <i>O. ficus-indica</i>	fruit & veg.	---	elongated	88