

of-freedom analysis compared one set of treatments against another, whereas the regression analysis measured the relationship between 2 variables, in this case the percentage germination of the seeds and respective yields. This could mean that compensated-rate seeding may not always solve the problem of yield maximization with 'Florida-Sweet', particularly with seed of lower germination percentages.

The relationship of range (estimated from the 20-cm plant spacing) as independent variable showed a poor fit with few exceptions. Those of some importance are given in Table 2.

Effect on ear characteristics. Ear quality was unaffected detrimentally by Difolatan-benomyl added to thiram-Captan as seed protectants (7). Compensated-rate seeding did not affect ear quality to the point of unmarketability but the relationship of the absolute deviation from 20-cm plant spacing (inde-

pendent variable) on ear diameter and percentage of U.S. Fancy ears was significant (Table 2). Both observed tendencies are debatable because, in the case of diameter, the limit of unmarketability was not reached and, in the case of percentage of Fancy ears in the fall test, all were below the desirable Florida U.S. Fancy grade. This might indicate that growing conditions during the fall had a greater effect than compensated-rate seeding on the percentage of U.S. Fancy ears.

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Screening for Chilling Resistance in Tomato Seedlings

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Abstract. Seedlings of a high-altitude form of the wild tomato *Lycopersicon hirsutum* Humb. & Bonpl. were able to develop the first true leaf when grown at a night temperature of 0°C (16 hours) and a day temperature of 20° (8 hours). Cultivars of *L. esculentum* Mill. developed poorly or not at all beyond the cotyledon stage under these conditions. The species were crossed and some selections from the second backcross to *L. esculentum* also were able to develop first true leaves when exposed to the low night temperatures. These gave progeny which approached or equalled the performance of the wild species, indicating that this cold resistance can be transferred from *L. hirsutum* to *L. esculentum* using low night temperature as a nondestructive screening technique.

The tomato does not grow well when mean night temperatures fall much below 10°C, and for optimum development, night temperatures should approach 18°. This is one aspect of "chilling sensitivity" in the tomato, and it is probably composed of many different responses to low temperature (2, 5). The wild species *L. hirsutum* Humb. & Bonpl. grows over a wide range of altitudes in Ecuador and Peru (7), and chilling resistance is

generally greater in accessions from high altitudes (6). The 2 species are easily crossed, providing that *L. esculentum* is used as the seed parent (4) and, therefore, attempts were made to transfer chilling resistance from an accession of *L. hirsutum* originally from Alta Fortaleza at 3100 m in Ancash, Peru (LA 1363, Tomato Genetics Stock Centre, Dept. Vegetable Crops, Univ. of California, Davis, CA 95616).

Development of hybrid progeny using the survival test at 0°C. Hybrids between *L. esculentum* and *L. hirsutum* had been previously selected for chilling resistance as follows. The F₁ hybrid *L. esculentum* 'Rutgers' × *L. hirsutum* LA 1363 was very vigorous, but largely infertile as a seed parent, and the few F₂ plants were sterile. However, the F₁ hybrid was a good pollen parent and was therefore backcrossed to *L. esculentum* 'Rut-

gers'. Pollen was collected from the resulting backcross population and pooled before pollination, so that sibbing was random. The progeny from sibbed pollinations was exposed at the cotyledon stage to 0° in darkness for 7 days (6). This "survival test" selection, which was applied at the appearance of the first true leaf, killed most seedlings of *L. esculentum* cultivars, but most seedlings of high altitude *L. hirsutum* survived with varying amounts of damage (6). The 10% of hybrid progeny least damaged by the survival test was grown and their pooled pollen was backcrossed to *L. esculentum* 'Tiny Tim'. The resulting 2nd backcross was sib-pollinated as before and 10% of the progeny selected by the survival test. The progeny of this selection were used for the subsequent night chill selection.

Disadvantages of the survival test. The survival of seedling tomatoes after chilling at 0°C depends on other factors as well as on their chilling resistance. For instance, it is difficult to control pathogenic fungi after chilling, and therefore survival can be influenced by differences in genetic resistance to such pathogens. Nongenetic variation also influences survival (3, 5). When seedlings are chilled from 4 hr after dark they require about twice the exposure to 0° to kill them as when they are chilled from dawn (3). For these reasons, a nondestructive test was devised which is not influenced by pathogens or by diurnal variations in chilling resistance.

Night chill selection method. This method exaggerated the nocturnal chilling which is characteristic of the tropical mountain climate to which *L. hirsutum* LA 1363 is native. Aseptic conditions were used throughout.

Seeds were surface-sterilized in 2.5% sodium hypochlorite for 20 min (*L. esculentum* and hybrids) or for 30 min (*L. hirsutum*) and washed thoroughly with sterile water. Seeds were germinated in the dark at 25°C in sterile Petri dishes on filter papers wet with Hoaglands solution (1). When the radicles were

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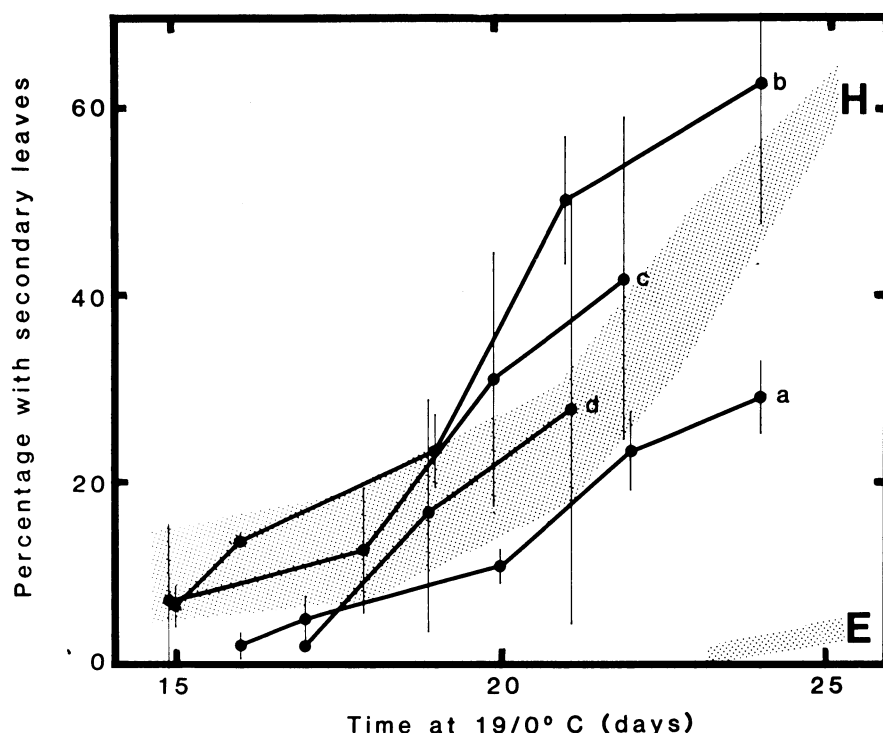


Fig. 1 Cumulative appearance of the first true leaf at 0°C night (16 hr) and 19° day (8 hr) conditions. Shaded (H): *L. hirsutum* LA 1363. Shaded (E): *L. esculentum* 'Tiny Tim', both \pm SE. Solid lines: progeny of the best 5% from selection shown in Table 1, labelled according to mean germination time in days. Letters show time to germination at 25°C: a, 4 days; b, 5 days; c, 6 days; d, 7 days. Bars show SE.

about 5-mm-long, but before the cotyledons had emerged, the seedlings were exposed to 16-hr nights at 0° and 8-hr days at 18 to 21°. The night temperature was controlled by placing melting ice around aluminium plates above and below the seedlings, in an insulated container in a room at 1°. During the day, the seedlings were kept under fluorescent lights (light intensity 60 $\mu\text{E s}^{-1}\text{m}^{-2}$) in a room set at 20°.

Result of night chill selection. The performance of *L. hirsutum* LA 1363 in the test was compared with that of 2 cultivars of *L. esculentum* ('Tiny Tim' and 'Grosse Lisse') and with the hybrids produced as described under development of hybrid progeny. After

15 days of the treatment, most of the *L. hirsutum* seedlings showed the first true leaf, and therefore all seedlings were assessed for this character. Table 1 shows the development of the hybrid progeny in comparison with that of the parent species after 15 days. All cultivars and hybrids were able to expand their cotyledons under these conditions. More than 80% of the *L. hirsutum* seedlings had developed first true leaves, but the 2 *L. esculentum* cultivars showed almost no growth of the first true leaf (growth was assumed to have occurred when the first true leaf had attained a length of 2 mm). In contrast, nearly 40% of the 2nd backcross hybrids showed growth of the first true leaf. The 5% of these

hybrids with the best leaf development were grown and their progeny from sib-pollinations were challenged with the same night chilling conditions, except that the day temperature was lower (19° \pm 1°C). Again, it is clear from Fig. 1 that the *L. hirsutum* parent developed much better under the low night temperatures than did the *L. esculentum* parent. The hybrid progeny varied more in germination time than did the parent species and, therefore, the progeny were separated into 4 groups, each with a similar germination time, and their development was plotted as separate lines in Fig. 1. With the lower day temperature than in the previous selection, overall development was slower. The hybrid progeny, which had been previously selected for low temperature leaf development as shown in Table 1, developed almost as well as the wild species. In contrast, *L. esculentum* 'Tiny Tim' showed very little leaf development under low night temperatures.

Our results show that a night temperature of 0°C interspersed by warm days clearly differentiates a chilling-resistant ecotype of *L. hirsutum* from cultivars of *L. esculentum*. The method does not suffer from interference by pathogens or diurnal variations (3) in the response to chilling. The results with hybrids backcrossed twice to *L. esculentum* suggest that night chilling can be used as a screening method to introduce a specific kind of chilling resistance into *L. esculentum*. This is the ability to develop under low night temperatures, which may help to extend both the seasonal and climatic limits of *L. esculentum*.

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Table 1. Development of first true leaves after 15 days at 0°C night (16 hr) and 20° day (8 hr) of *Lycopersicon* species and 2nd backcross to *L. esculentum*. Each replicate contained between 40 and 60 seedlings.

Species	Cultivar or accession	1st true leaves developed (% range)	No. replications
<i>L. esculentum</i>	Tiny Tim	0-2	4
<i>L. esculentum</i>	Grosse Lisse	0	2
<i>L. hirsutum</i>	LA 1363	83-94	4
Progeny of 2nd backcross to <i>L. esculentum</i>	---	32-68	20