

## Tuber yield and quality of 4x–2x (FDR) potato progenies derived from the wild diploid species *Solanum berthaultii* and *Solanum tarijense*

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With 2 tables

Received May 30, 2002/Accepted November 5, 2002

Communicated by F. Salamini

### Abstract

Field experiments were carried out in order to evaluate 4x–2x families derived from crosses between elite 4x potato cultivars and 2x *Tuberosum-Solanum tarijense* and 2x *Tuberosum-Solanum berthaultii* clones. Three traits were assessed: total tuber yield (TTY), haulm maturity (HM) and general tuber appearance (GTA). The degree of heterosis of these hybrid families was evaluated by comparison with the respective 2x and 4x parents as well as with seven 4x cultivars. The parental haploid species hybrids derived from *S. berthaultii* and *S. tarijense* combined two or more positive horticultural characteristics. Expressed as yield percentage of the 4x parents, the TTY of the families ranged from 53% to 246%. For TTY, the best 4x–2x hybrid family ranked better than seven out of nine elite 4x cultivars. Some families had GTA scores in the range of the highly selected 4x cultivars. The families, however, were generally later maturing than the 4x parent group. Specific combining ability for TTY and GTA were the only two significant sources of variation observed in this genetic material. Parent–offspring correlation coefficients were low for all traits, and indicated that parental performance would not be informative at either ploidy level. These results parallel previous investigations with distinct haploid species hybrids where a 4x–2x breeding scheme was found to be an effective strategy for increasing progeny TTY over the 4x parents. However, the high degree of heterosis for TTY along with good GTA scores observed in certain cross combinations derived from these unadapted 2x species was a somewhat surprising result. Thus far, the importance of these two South American wild potato species, from the potato-breeding standpoint, has been limited to the fact that they are natural reservoirs of major genes controlling resistance against important diseases and insects. However, the level of performance of some 4x–2x families in comparative assays with elite cultivars suggests the unanticipated possibility of introgressing genetic factors from *S. berthaultii* and *S. tarijense* with positive effects on quantitative traits of horticultural importance along with these major resistance genes.

**Key words:** *Solanum berthaultii*; — *Solanum tarijense* — 2n-gametes — unilateral sexual polyploidization

*Solanum berthaultii* and *S. tarijense* are two diploid (2x) wild potato species endemic to South America, with predominant geographical distribution in central and southern areas of Bolivia and the northern region of Argentina (Hawkes 1990). To date, the importance of *S. berthaultii* and *S. tarijense*, from the potato-breeding standpoint has been limited to the fact that they are natural reservoirs of genes controlling resistance against major potato diseases and pests (for review see Ortiz 1998). Some accessions of *S. berthaultii* with trichome-mediated resistance to aphids also have a positive effect of

decreasing the incidence of some potato viruses (Lapointe et al. 1987). However, reports showing the relevance of this germplasm for improvement of agronomic traits other than disease and pest resistance are still scarce.

The introgression of the 2x *Solanum* germplasm into the cultivated potato has been possible via 4x–2x crosses with unilateral sexual polyploidization (USP) (Peloquin et al. 1989, 1999). However, the majority of the breeding work conducted thus far with 4x–2x crosses has been focused on the germplasm derived from a single 2x species, the cultivated *S. phureja*. Therefore, only a limited fraction of the immense 2x *Solanum* germplasm has been effectively exploited via the USP breeding scheme (Tai 1994). The first set of field trials using 4x hybrid families derived from haploid *Tuberosum-S. berthaultii* or haploid *Tuberosum-S. tarijense* hybrids indicated the high potential value of this germplasm for introgressing important yield and quality traits into the 4x potato gene pool (Darmo and Peloquin 1991). In the present work, more information is provided on the performance of *S. berthaultii*- and *S. tarijense*-derived germplasm as staminate (male) parents in 4x–2x crosses.

### Materials and Methods

Field experiments were conducted to evaluate the performance of 4x–2x families using as staminate parents the wild diploid species *S. berthaultii* and *S. tarijense*. Controlled crosses were made employing the 2x parental clones 'TTH 100-1' and 'TTH 100-2' (obtained from 'Merrimack' haploid *Tuberosum-S. tarijense*) and the clone 'TBH 94-1' (selected from 'Merrimack' haploid *Tuberosum-S. berthaultii* crosses). The 4x female parents were a random sample of long-day ('Belladonna') and short-day ('Chiquita') elite cultivars. These three 2x *Tuberosum-S. tarijense* and *Tuberosum-S. berthaultii* hybrids were then hybridized (in a complete factorial design) as males to 'Belladonna' and 'Chiquita'. Tuber seeds were produced under glasshouse conditions. Three tubers per plant from each of the 20 plants obtained in each cross combination were used to constitute three family bags (with one tuber per genotype per bag). Tubers of the 4x–2x families were treated twice with Rindite (1 mg/kg for 3 days) to break dormancy. The seed tubers of the 4x parental and a set of standard cultivars were field-grown and were larger than the 4x–2x family tubers. Field assays were set up in a sandy soil at Hancock (WI, USA) in a randomized complete block design (RCBD). The assay was divided into three experiments. The six families derived from the complete 4x–2x factorial crosses were evaluated in experiment 1 (E1) with three replications (20 hills per plot). The 4x cultivars (used as parents) as well as a set of seven commercial 4x cultivars ('Atlantic', 'Spunta', 'Claustar', 'Baronesa',

‘Aracy’, ‘Elvira’ and ‘DTO-21’) were evaluated in experiment 2 (E2). The 2x clones (used as parents) were evaluated in experiment 3 (E3). Experiments E2 and E3 were conducted in adjacent plots (i.e. in the same experimental field as E1) in a RCBD with two replications (four-hill plots). The ‘unbalanced’ plot size of the parents and cultivars was employed because the genetic material in E2 and E3 represented a group of selected clones. The 4x–2x families, however, are composed of a group of 20 full-sib clones (i.e. all 20 clones are genetically distinct from each other). All subexperiments had identical fertilization and cultural management. Plants were spaced 0.91 m between rows and 0.31 m between hills in a row. The total duration of the assays was 110 days from planting to harvesting (June–October). Families were assessed for total tuber yield (TTY), expressed in kg per hill (using only tubers over 25 mm diameter), haulm maturity (HM) scoring (made 77 days from planting) and general tuber appearance (GTA) (evaluated at harvesting). The analysis of variance (ANOVA) was performed under the assumption that the 4x and 2x parents were random samples of populations of 4x and 2x clones. Variation among families from the factorial mating was partitioned into the sources due to males (male general combining ability, GCA), females (female GCA) and male–female interactions (specific combining ability, SCA). Tests of significance for males and females were performed by F-test comparison to their respective male–female interaction mean square. Female–male interactions were performed by testing their mean squares against the error mean squares. Data from the parental clones, cultivars, and haploid-species evaluations were used to obtain useful comparisons, to estimate of the degree of heterosis of the 4x–2x families, and to estimate parent mean-family mean correlation.

Results

The ANOVA indicated a significant difference among families for TTY (Table 1). The families outyielded the two 4x parents by 13.3%. Families with higher TTY than the 4x parent were observed in all families derived from ‘Belladona’ but no high-parent heterosis was observed with families derived from ‘Chiquita’. The highest-yielding 4x–2x family (‘Belladona’ × ‘TTH 100-1’) had yield identical to ‘Chiquita’ (that was the second best yielding 4x clone in the E2). Expressed as yield percentage of the 4x parent, the family TTY ranged from 52.8% to 246%. The families outyielded four out of seven standard cultivars. However, the 4x–2x family mean yield (0.85 kg/hill) was identical to the 2x parent group mean yield. The best 4x–2x family (‘Belladona’ × ‘TTH 100-1’) ranked better or similar to six out seven cultivars. Differences for HM scores among families were not significant (Table 2). When considered individually, only two families had HM scoring close to the corresponding 4x parents. Differences among

Table 1: Analysis of variance of total tuber yield (TTY), haulm maturity (HM) and general tuber appearance (GTA) of 4x families from 4x–2x crosses involving haploid *Tuberosum-Solanum tarijense* and haploid *Tuberosum-Solanum berthaultii* hybrids. The experiment was conducted at Hancock, Wisconsin, USA

| Source of variation <sup>1</sup> | df | Mean squares       |       |         |
|----------------------------------|----|--------------------|-------|---------|
|                                  |    | TTY                | HM    | GTA     |
| Blocks                           | 2  | 0.055              | 0.389 | 0.514   |
| 4x–2x families                   | 5  | 0.080**            | 0.222 | 0.289** |
| Males (GCA)                      | 2  | 0.121              | 0.389 | 0.389   |
| Females (GCA)                    | 1  | 0.095              | 0.000 | 0.222   |
| Male × female (SCA)              | 2  | 0.032 <sup>+</sup> | 0.167 | 0.222*  |
| Error                            | 10 | 0.009              | 0.122 | 0.047   |

<sup>+</sup>, \*, \*\* Significant at  $P = 0.10$ ,  $P = 0.05$  and  $P = 0.01$ , respectively.  
<sup>1</sup> GCA, general combining ability; SCA, specific combining ability.

Table 2: Means of total tuber yield (TTY), haulm maturity (HM) and general tuber appearance (GTA) of families derived from 4x–2x crosses involving *Tuberosum-Solanum tarijense* (‘TTH 100-1’ and ‘TTH 100-2’) and *Tuberosum-Solanum berthaultii* (‘TBH 94-1’) hybrids, parental 4x cultivars and a set of standard 4x clones/cultivars. The experiment was conducted at Hancock, Wisconsin, USA

| Genotype                 | Origin <sup>1</sup> | Means |                 |                  |
|--------------------------|---------------------|-------|-----------------|------------------|
|                          |                     | TTY   | HM <sup>2</sup> | GTA <sup>3</sup> |
| <b>2x clones</b>         |                     |       |                 |                  |
| ‘TBH 94-1’               | UW-Madison          | 0.89  | 4.0             | 2.75             |
| ‘TTH 100-1’              | UW-Madison          | 0.70  | 3.5             | 3.00             |
| ‘TTH 100-2’              | UW-Madison          | 0.97  | 4.0             | 3.75             |
| LSD <sub>0.05</sub>      | —                   | ns    | ns              | ns               |
| <b>4x cultivars</b>      |                     |       |                 |                  |
| ‘Belladona’              | Germany             | 0.43  | 4.5             | 2.75             |
| ‘Chiquita’               | Brazil              | 1.06  | 3.5             | 3.75             |
| ‘Atlantic’               | USA                 | 1.04  | 3.0             | 3.25             |
| ‘Spunta’                 | Holland             | 0.76  | 3.0             | 3.75             |
| ‘Claustar’               | France              | 0.73  | 2.5             | 4.00             |
| ‘Baronesa’               | Brazil              | 0.65  | 3.5             | 2.50             |
| ‘Aracy’                  | Brazil              | 1.32  | 4.0             | 4.00             |
| ‘DTO-21’                 | USA                 | 1.05  | 3.0             | 3.25             |
| ‘Elvira’                 | Germany             | 0.64  | 3.5             | 3.50             |
| LSD <sub>0.05</sub>      | —                   | 0.49  | ns              | 0.83             |
| <b>4x–2x families</b>    |                     |       |                 |                  |
| ‘Belladona’ × ‘TBH94-1’  | UW-Madison          | 0.85  | 4.67            | 3.50             |
| ‘Belladona’ × ‘TTH100-1’ | UW-Madison          | 1.06  | 4.67            | 3.67             |
| ‘Belladona’ × ‘TTH100-2’ | UW-Madison          | 0.86  | 5.00            | 3.50             |
| ‘Chiquita’ × ‘TBH94-1’   | UW-Madison          | 0.56  | 5.00            | 2.83             |
| ‘Chiquita’ × ‘TTH100-1’  | UW-Madison          | 0.92  | 4.33            | 3.67             |
| ‘Chiquita’ × ‘TTH100-2’  | UW-Madison          | 0.86  | 5.00            | 3.50             |
| LSD <sub>0.05</sub>      | —                   | 0.22  | ns              | 0.48             |

<sup>1</sup> UW-Madison, University of Wisconsin-Madison.  
<sup>2</sup> HM, based on a scale from 1 = senesced to 5 = still flowering.  
<sup>3</sup> GTA, based on a scale from 1 = poor to 5 = excellent.

families for GTA were highly significant (Table 1). Five out of six families had GTA scores equal to or above three (Table 2). The results indicated a significant (at the 10% level) SCA for total tuber yield. For GTA, a significant SCA (at the 5% level) was observed (Table 1). A significant (at the 10% level) 2x-parent-offspring correlation was observed for HM ( $r = 0.79$ ) but not for TTY ( $r = -0.51$ ) and GTA ( $r = 0.21$ ). No significant 4x parent–offspring correlation was observed for HM ( $r = 0.01$ ), TTY ( $r = -0.48$ ) or GTA ( $r = 0.21$ ).

Discussion

The TTY of the families indicates the parental value of haploid *Tuberosum-S. berthaultii* and haploid *Tuberosum-S. tarijense* hybrids. The occurrence of heterotic families TTY parallels the results of previous investigations using USP breeding scheme with Phureja-haploid hybrids as well as with distinct haploid *Tuberosum-S. berthaultii* and haploid *Tuberosum-S. tarijense* hybrids (Darmo and Peloquin 1991, Tai 1994). It is important to mention that the yield potential of the 4x–2x families in the present experiment was probably underestimated because of the lower weight of the greenhouse-grown tubers of the families (15–50 g) compared with those of the field-grown seed tubers of the parental clones (50 g or more). The smaller tuber size of the families compared with the parental group, penalizing the 4x–2x progenies, may give some erroneous estimation about the breeding potential of these crosses. However, some surprisingly high levels of heterosis observed in specific cross combinations could be explained, in part, as being a result of the lack of local adaptation of the 4x parents

used for crossing. The performance of the additional set of 4x cultivars provided, in this regard, useful comparisons, since several families had higher TTY than some more adapted cultivars such as 'Atlantic'.

Genetic variance for tuber yield in potato has been reported to be almost entirely nonadditive with both allelic as well as non-allelic interactions being considered of great importance (Peloquin et al. 1989). Therefore, the increased yield of 4x–2x families over the 4x parent can be explained by the introgression of genetic and allelic diversity; favourable linkages (*sensu* Demarly 1979) or through favourable changes in the patterns of gene expression and/or regulation. Giovannini et al. (1993) observed heterosis for the quantitative expression of a set of isozymes after crossing *S. tuberosum* × *S. tarijense*. Moreover, they observed the disappearance of some parental bands and the appearance of several additional bands, suggesting that this interspecific cross may lead to new patterns of gene expression and genome reorganization. Another plausible explanation for the very good performance for TTY of certain 4x–2x families is the presence of inherent high-yielding factors in the genome of the 2x parental clones.

The average HM of 4x–2x families was higher than the mean HM of the 4x parent group. No significant differences were found among families, indicating that they were uniformly late maturing. No 4x–2x family had HM scores equal or similar to the earlier 4x parent. The 2x parental clones used in this study were very late compared with other haploid *Tuberosum*-diploid species hybrids evaluated. Thus, it seems that selection among haploid *Tuberosum*-*S. berthaultii* and haploid *Tuberosum*-*S. tarijense* hybrids may be needed to obtain 4x–2x family with HM values in the range of commercial 4x cultivars. The selection of superior parents for earliness in haploid *Tuberosum*-*S. berthaultii* and haploid *Tuberosum*-*S. tarijense* has been demonstrated to be effective by Darmono and Peloquin (1991).

The identification of haploid-species hybrids and 4x–2x families with GTA similar to commercial cultivars was an interesting observation. The utilization of selected 2x parents with shallow eyes and fewer and heavier tubers may be the most plausible explanation for the observed differences between GTA values for these families and haploid-species hybrids when compared with those from Phureja–*Tuberosum* hybrids. As pointed out by Hermundstad and Peloquin (1985), the low number, larger size and shallow eyes of 2x haploid *Tuberosum*-*S. berthaultii* and haploid *Tuberosum*-*S. tarijense* tubers were the major advantages in relation to *S. phureja* germplasm. Results of the E3 confirmed one of the parental clones ('TTH 100–2') had the highest GTA score among a range of haploid-species hybrid clones. The families derived from 'TTH 100–2' (as a group) had an average GTA score of 3.5, which is in the same range of several elite 4x cultivars. Therefore, to maximize improvements, the 2x hybrid parents should be selected for specific GTA components (e.g. eye depth) before making 4x–2x crosses.

These overall results indicated that only SCA variances were significant for GTA. The SCA variances were also greater than the GCA variances for TTY. The degree of heterosis would be dependent upon the parental clone being employed in the crosses. Predominant SCA variances have been also reported for TTY in progenies from the conventional 4x–4x crosses (Killick 1977) as well as 4x–2x crosses with 2x Phureja–haploid *Tuberosum* hybrids as parents (for

review see Buso et al. 1999a). Greater SCA for TTY was also observed in crosses involving a distinct set of haploid *Tuberosum*-*S. berthaultii* or haploid *Tuberosum*-*S. tarijense* hybrids (Darmono and Peloquin 1991). In contrast, several investigations have indicated GCA as being the more important component for TTY in Phureja–haploid *Tuberosum* hybrids (Buso et al. 1999a). These conflicting results may be partly explained by: (1) genetic differences in the 4x and 2x parents; (2) differences in the mechanisms of 2n pollen formation; and (3) the assessments being done under different environmental conditions.

The lack of significant correlations between 4x and 2x parents and the respective offspring for TTY is in agreement with previous results in interploid crosses (De Jong and Tai 1991). This indicates that the tuber yield of the parents *per se* should not be a major criterion when choosing parents for 4x–2x crosses. The same trend was observed for GTA. Good predicting power based upon the 2x parent performance for HM has been reported in distinct sets of 4x–2x progenies evaluated in Europe (Hutten et al. 1996). The present results indicated, however, that parental performance for HM might be dependent upon the set of clones used as parents as well as the environment employed for testing. In these experiments, only a weak correlation (significant at the 10% level) was observed between 2x parent families for HM. This information suggests that HM of the 2x parental clones could be misleading in predicting performance of the 4x–2x families.

Although employing a relatively small sample of 4x–2x families, the overall results indicated that the USP breeding scheme with *S. berthaultii* and *S. tarijense* clones could provide an alternative method of increasing tuber yield of families over elite 4x cultivars, as also demonstrated by Darmono and Peloquin (1991). This germplasm could thus represent new sources of potentially useful alleles for exploitation in 4x–2x crosses. In addition, previous studies indicated that two 2x clones tested here ('TBH 94–1' and 'TTH 100–1') were also the best male parents for yield in 2x–2x crosses (Ortiz and Peloquin 1993). It is important to mention that the analysis reported here was based upon families as a group. Therefore, it will be interesting to compare the performance of some transgressive 4x clones for TTY selected within these 4x–2x families. These 4x clones selected within 4x–2x progenies are the so-called 'diplandrous tetraploid hybrids' or 'DTO' clones. Assays employing some of these hybrids (of *S. phureja* origin) indicated high levels of heterosis over elite cultivars under distinct environmental conditions (Buso et al. 1999b). Similarly, the results given here indicate that high-value genotypes derived from *S. berthaultii* and *S. tarijense* could be readily introgressed into the 4x gene pool via 4x–4x crosses with 'DTO' clones.

These experiments with *S. berthaultii* and *S. tarijense* clones reinforce the notion that prediction of genetic gains based solely upon the phenotype of the wild species can be misleading. The exotic germplasm may have a series of initially unexpected favourable alleles with direct effects on traits of agronomic and horticultural significance (Tanksley and McCouch 1997). In the specific case of potato breeding, the development of useful commercial germplasm is expected to be relatively rapid, especially because *S. tuberosum*-like phenotypes can be readily recovered as previously indicated by interspecific crosses involving closely related wild potato germplasm (Flewelling and Peloquin 1987).

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