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Inbreeding and true seed in tetrasomic potato. I. Selfing and open pollination in Andean landraces (*Solanum tuberosum* Gp. *Andigena*)

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Abstract True potato seed (TPS) may be an alternative method of potato production in developing countries. A breeding method for the sexual propagation of this vegetatively propagated crop should consider the development of parental lines and the type of cultivar to be released. Open-pollinated (OP) cultivars seem to be an inexpensive procedure to produce potato from true seed. However, OP progenies are the result of selfing and outcrossing in male-fertile tetraploid potatoes. The aim of the present research was to establish the effect of inbreeding and open pollination in TPS. Ten *Andigena* clones were used as parental material to derive hybrid (S_0), inbred (S_1 and S_2), and open-pollinated (OP_1 and OP_2) generations. Significant differences among generations were found for pollen production, pollen viability (as determined by its stainability with aceto-carmine glycerol), number of flowers and berries plant^{-1}, number of seeds berry^{-1}, weight of 1000 seeds, and tuber yield plant^{-1}. The parental populations were significantly different for most of the traits, but not for flower production and berry weight. The interaction of population × generation was significant for pollen and seed production as well as for weight for 1000 seeds. All the traits evaluated except seed weight showed a strong inbreeding depression, while the OP progenies had intermediate values between the S_0 and the S_1. This demonstrates that open pollination in potatoes is not exclusively the product of selfing; it also results from outcrossing.

Key words Synthetic cultivars · True potato seed

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Introduction

Research on true potato seed (TPS) conducted by the International Potato Center (CIP) in cooperation with public and private institutes around the world has received a great deal of attention since 1977 (Mendoza 1985). Several thousand TPS progenies are distributed annually to cooperators worldwide. However, some basic questions remain unanswered. Is TPS the way in which potato should be produced in developing countries? What should breeders do to develop TPS cultivars? Who will produce TPS? How will TPS be obtained? These and other questions must be answered before potato production from true seed can become a reality.

Basic research has been conducted to determine the extent and genetic nature of the variability for several agronomic and production traits relevant to TPS (Thompson et al. 1983; Thompson and Mendoza 1984). However, information on the effect of inbreeding is scarce on the basis of the performance of open-pollinated (OP) TPS progenies. Most inbreeding studies in Andean tetraploid potato published to-date have not continued beyond the S_1 generation (Atlin 1985 b). Hence, the objective of the present paper was to determine the effect of inbreeding on reproductive and agronomic traits and to explain the performance of OP true potato seed progenies in Andean potatoes (*Solanum tuberosum* Gp. *Andigena*).

Materials and methods

Ten tetraploid *Andigena* clones from the tuber-bearing *Solanum* gene bank held at CIP were selected at random as parental material. The clones' codes are: CIP 700127, CIP 700396, CIP 700397, CIP 700575, CIP 701349, HUA 385, HUA 404, HUA 475, OCH 5298, and OCH 8334. They were initially collected in farmer's fields of the South American Andes.

Single plants from each clone were artificially crossed with a bulk of unrelated pollen to produce the S_0 generation. The S_0 plants were selfed in order to obtain the S_1 generation. OP berries were collected from each *Andigena* clone to obtain the OP_1 generation. The S_2 resulted from selfing S_1 plants, whereas the OP_2 was produced by harvesting OP berries of OP_1 plants.

This experiment included five generations (S_0, S_1, S_2, OP_1, and OP_2) for each of the ten *Andigena* clones (hereafter, populations). The field evaluation was carried out in CIP's Andean highland station at Huancayo (altitude 3200 m) during 1984–1985.

Seeds of the crosses belonging to the same generation and population were mixed and sown in the greenhouse nursery. After 8 weeks the seedlings were transplanted to the field. The experimental field layout was a randomized complete block design with four replications. The treatment combinations (generation × population) were arranged following a split-plot design. The main plot was the population (according to the original parental clone) and the subplot was the generation. Each plot consisted of 40 plants. The distance between rows was 90 cm, with 30 cm between plants within the same row.

The following traits were evaluated: pollen production (mg flower^{-1}) and stainability (%), number of flowers and berries plant^{-1}, number of seeds berry^{-1}, weight of berries and 1000 seeds, and tuber yield (kg plant^{-1}). Pollen grains were stained with acetocarmine glycerol jelly (Marks 1954) to determine their viability.

Factorial analyses for split-plot designs were performed to analyze the experiment. A least-significant difference (LSD) test at the 0.05 significance level was used to compare family, generation, and family × generation means. Regression analyses between the inbreeding coefficient (independent variable X) and the traits evaluated (dependent variable Y) were carried out to determine the inbreeding effect on each family. A negative and significant slope was considered as an indication of inbreeding depression.

Results

Significant differences were detected between the parental populations for pollen production and viability, number of berries plant^{-1}, number of seeds berry^{-1}, weight to 1000 seeds, and tuber yield (Table 1). Similarly, the generations were significantly different for all the traits evaluated except berry weight. The interaction of population × generation was significant only for pollen production, number of seeds berry^{-1}, and weight of 1000 seeds.

Table 2 indicates the inbreeding effect and generation means for the reproductive traits and tuber yield. A strong and significant inbreeding effect (least value for advanced inbreeding generation) was detected for pollen production and viability; flower, berry, and seed production; and tuber yield. Surprisingly, although we thought seed weight would be affected negatively by inbreeding, it increased significantly in the advanced inbred generations.

Inbreeding depression significantly affected pollen production, viability, and berry and seed set in almost all the *Andigena* clones (Table 3). Tuber yield and berry weight were affected by inbreeding depression in 40% of the clones, whereas flowering was affected in only two *Andigena* clones. Some clones were affected dramatically by inbreeding depression in almost all the traits evaluated (such as OCH 8334), whereas other

Table 1 Analysis of variance of different inbreeding generations of ten parental *Andigena* clones

Source of variation	Degrees of freedom	Pollen production	Pollen viability	Flowers plant^{-1}	Berries plant^{-1}	Tuber yield plant^{-1}
Replications	3	1.4**	122.3	203.6	122.7	55 866.7
Clones (C)	9	1.8**	1162.2**	192.8	303.2*	190 742.6**
Error (a)	27	0.2	55.2	97.9	100.8	38 162.6
Generations (G)	4	3.1*	3736.3**	162.6**	2486.2**	336 940.4**
C × G interaction	36	0.2**	120.8	42.6	87.2**	21 104.3
Error (b)	120	0.1	111.5	31.4	39.5	18 732.2
Mean		1.4	52.4	17.9	16.4	683.5
Coefficient of variation (%)		23.6	20.2	31.3	38.4	20.0

* and ** indicate significance at the 0.05 and 0.01 levels

Table 2 Inbreeding effect on reproductive and agronomic traits of ten *Andigena* clones

Generation	Pollen production (g)	Pollen viability (%)	Flowers plant^{-1} (#)	Berries plant^{-1} (#)	Berry weight (g)	Tuber yield plant^{-1} (g)
S_0	1.75	64.57	19.09	27.74	8.4	799
S_1	1.35	49.67	16.94	13.59	7.2	696
S_2	0.99	38.04	15.51	5.93	7.0	579
OP_1	1.52	55.38	17.43	18.04	8.2	740
OP_2	1.40	54.18	20.72	16.49	8.8	605
$LSD_{0.05}$	0.21	6.61	3.51	3.93	2.6	86

Table 3 Inbreeding depression for six traits in ten *Andigena* clones; + indicates that inbreeding depression negatively affects phenotype as measured by a significant and negative regression coefficient

Clone code	Pollen production	Pollen viability	Flowers $plant^{-1}$	Berries $plant^{-1}$	Berry weight	Tuber yield $plant^{-1}$
OCH 5298	+	+		+		+
700396	+	+		+	+	+
701349		+		+		
700397		+		+	+	
HUA 404	+	+		+	+	
HUA 475	+	+		+		+
HUA 385				+		
700575	+	+				
OCH 8334	+	+	+	+	+	+
700127	+		+	+		
Effect of inbreeding (%)	70	80	20	90	40	40

clones (such as HUA 385) decreased their berry and seed set, traits exhibiting severe inbreeding depression, only in the advanced inbreeding generations.

Discussion

Inbreeding depression

Inbred progenies showed lower pollen viability and production, and berry and seed set, than hybrids and open-pollinated progenies. Golmirzaie et al. (1987) obtained similar results using a hybrid TPS population with a wide genetic background. Therefore, a concomitant reduction for the above traits and for tuber yield with an increase in the inbreeding coefficient (e.g. from $S_0 = 0$ to $S_1 = 0.167$ and $S_2 = 0.306$) was observed in the present experiment. This clearly supports the hypothesis that potato is a species severely affected by inbreeding depression (Mendoza and Haynes 1973).

An analysis of reproductive traits provided important information. The inbred progenies produced less pollen, with lower quality, than the hybrid progenies. This was not surprising because both pollen traits are mainly under the control of genes with non-additive effects (Ortiz and Peloquin 1994).

The results obtained with open-pollinated progenies in this experiment can be explained by the genetic structure of the populations. OP progenies are developed by selfing and outcrossing due to the action of wind and natural pollinators such as bees of the genus *Bombus* (Atlin 1985 a). Therefore, the inbred progenies contributed little to the gamete pool. This resulted in less inbreeding than expected in OP progenies.

Open-pollinated true potato seed and synthetic cultivars

The use of open-pollinated true potato seed may represent an inexpensive way to produce potato in developing countries. However, the yield potential of OP progenies should be less than that of hybrid TPS progenies. This problem may be the consequence of higher inbreeding in OP progenies than in hybrids from self-pollination. Indeed, the results indicated that the yield of OP progenies was in-between that of the hybrids (S_0) and inbred progenies (S_1 or S_2). Moreover, it seems that advanced OP generations derived by obtaining TPS from individual clones followed a similar trend as selfed generations, i.e. $OP_1 > S_1 > OP_2 > S_2$.

Synthetic cultivars mixing different numbers of parents (more than two) could be an alternative mode of potato production from TPS (Atlin 1985 a; Ortiz 1999). The advanced generation of a synthetic cultivar must reduce the average inbreeding depression. Otherwise, selection or roguing of weak seedlings at early stages in the nursery may help eliminate some inbred progenies from the inbred-hybrid OP mixtures (Atlin 1985 b; Ortiz 1997). Initial results (Golmirzaie and Mendoza 1986; Golmirzaie 1987; Atlin and Wiersema 1988) indicated that both solutions for using OP progenies are feasible in potato.

References

Atlin G (1985 a) Inbreeding in TPS progenies: implications for seed production strategy. In: Page OT (ed) Present and future strategies for potato breeding and improvement. International Potato Center, Lima, Perú, pp 71–85

Atlin G (1985 b) Farmer maintenance of TPS varieties. In Page OT (ed) Innovative methods for propagating potatoes. International Potato Center, Lima, Perú, pp 39–62

Atlin G, Wiersema SG (1988) Selection against inbred seedlings in mixtures of inbreed lines and hybrid true potato seed. Potato Res 31 : 105–112

Golmirzaie AM (1987) Performance of true potato seed synthetic populations (abstract). Am Potato J 64 : 440

Golmirzaie AM, Mendoza HA (1996) Effect of early selection for seedling vigor on an open pollinated TPS (abstract). Am Potato J 63 : 246

Golmirzaie AM, Bretschneider K, Ortiz R (1987) Inbreeding effect of reproduction and agronomic characters on different true potato

seed generations. In: Foldo NE (ed) Abstracts of Conference Papers and Posters, 10th Triennial Conference of the European Association for Potato Research, Aalborg, Denmark, pp 294–296
Marks GE (1954) An acetocarmine glycerol jelly for use in pollen fertility counts. Stain Technol 29 : 277
Mendoza HA (1985) Selection of uniform progenies: implications for breeding and seed production strategy. In: Page OT (ed) Innovative methods for propagating potatoes. International Potato Center, Lima, Perú, pp 87–98
Mendoza HA, Haynes FL (1973) Some aspects of breeding and inbreeding potatoes. Am Potato J 50 : 216–222
Ortiz R (1997) Breeding for potato production from true seed. Plant Breed Abstracts 67 : 1355–1360
Ortiz R (1999) Potato breeding via ploidy manipulations. Plant Breed Rev 16 : 15–86
Ortiz R, Peloquin SJ (1994) Effect of sporophytic heterozygosity on the male gametophyte of the tetraploid potato (*Solanum tuberosum*). Ann Bot 73 : 61–64
Thompson PG, Mendoza HA (1984) Genetic variance estimates in a heterogeneous potato population propagated from true seed (TPS). Am Potato J 61 : 697–702
Thompson PG, Mendoza HA, Plaisted RL (1983) Estimation of genetic parameters for characters related to potato propagation by true seed (TPS) in an *Andigena* population. Am Potato J 60 : 393–401