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**The Lack of Enzymic Browning in Wild Potato Species,
Series Longipedicellata,
and their Crossability with *Solanum tuberosum***

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The Lack of Enzymic Browning in Wild Potato Species, Series Longipedicellata, and their Crossability with *Solanum tuberosum*

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With 7 tables

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Abstract

The enzymic browning of tubers of several species accessions and hybrids was evaluated using cut tubers. The tubers of some Mexican wild potato species, Series Longipedicellata, and in particular *Solanum hjertingii* ($2n = 48$), do not blacken on wounding. Attempts were made to transfer this character to cultivated potatoes, and over 1600 pollinations were made directly between *S. hjertingii* and cultivated potatoes, or indirectly through bridging species, in order to evaluate the utilization of this species in potato breeding. *S. hjertingii* could be crossed with other species in Series Longipedicellata, with *S. acaule*, *S. demissum* and cultivated diploid species, but direct crosses with *S. tuberosum* were virtually impossible. No enzymic browning was observed in some hybrid progeny.

Key words: *Solanum hjertingii* — *S. tuberosum*, Series Longipedicellata potato germplasm — enzymic browning — pre-breeding selection

The discolouration of potato tuber flesh from external and internal bruising caused by mechanical damage is a major cause of losses to producers and processors and of complaints by the consumer. The cause of this enzymatic browning of cut surfaces, external bruising and blackspot caused by internal bruising is well understood and is due to the oxidation of phenolic substrates such as tyrosine and possibly chlorogenic acid by the enzyme polyphenol-oxidase to dark melanin pigments (MUNETTA 1977, GRAY and HUGHES 1978). It is assumed that the enzyme and substrate are held separately *in vivo*, and that on injury this separation breaks down (RHODES and WOOLVERTON 1978).

It is not clear why certain tubers, particularly of different varieties are more prone to internal bruising than others. There is, however, variation in

the expression of tuber discolouration on wounding in different species of the tuber-bearing Solanums. FIRBAS (1961) carried out a survey of 39 wild species of potato, from which he found that *S. hjertingii*, a tetraploid Mexican species (Series Longipedicellata) showed no discolouration when cut and grated. In another Longipedicellata species, *S. polytrichon*, discolouration was only very slight.

Some attempts at crossing *S. hjertingii* with *S. tuberosum* have been reported, but with no success (SAMS et al. 1977), although the successful introduction of the non-blackening character into *S. tuberosum* from *S. hjertingii* has been reported from Denmark (FOLDØ, pers. comm.). The crossability of the two species was not considered problematical, rather it was the difficulty of quality and yield improvement following hybridisation.

A large germplasm collection of wild potato species is held at the University of Birmingham. The work reported here formed part of a wider study on enzymic browning in wild potatoes (WOODWARDS 1982). Several accessions of *S. hjertingii* and other Longipedicellata species as well as *S. tuberosum* cultivars were screened for enzymic browning. A crossing programme was initiated to introduce the non-blackening character from wild species into *S. tuberosum*. In addition, the potential of bridging species was assessed for the transference of the character, in view of the reported problems of direct crosses between *S. hjertingii* and *S. tuberosum*.

Materials and Methods

All material was grown in a insect proof glasshouse or screenhouse. The species are listed in *Table 1*.

For visual assessment of enzymic browning five tubers were used from a bulked sample from each accession and progeny of each cross, where there were sufficient tubers. Tubers were cut in half, washed in tap water to remove surface starch and debris, and then left in open petri dishes for 5 hours before scoring discolouration on a 1—5 scale (*Table 2*). A value was assigned for the 10 half tubers as a group, rather than scoring each tuber piece individually.

For crossability studies self-fertile clones were emasculated 24 hours prior to pollination, before anthesis had occurred. Self-incompatible species were not emasculated. In initial pollinations, mixed pollen of five clones from the same accession was used to pollinate plants from a different accession. Seed produced was bulked for each set of pollinations using the same parental clones. From this bulked seed, a sample was grown and hybrid plants selected for subsequent single clone pollinations.

Results

Enzymic browning

Several accessions of *S. hjertingii* were identified which did not blacken when tubers were cut (*Table 3*). Two accessions of *S. polytrichon* and *S. stoloniferum* were also non-blackening, but tubers of *S. fendleri* blackened completely upon wounding. Tubers of three cultivars of *S. tuberosum* had an un-

Tab. 1 Tuber-bearing *Solanum* species used in this study

Species	Ploidy	Species abbreviation
Series Longipedicellata		
<i>S. fendleri</i>	4x	fen
<i>S. hjertingii</i>	4x	hjt
<i>S. polytrichon</i>	4x	plt
<i>S. papita</i>	4x	pta
<i>S. stoloniferum</i>	4x	sto
Series Tuberosa		
<i>S. tuberosum</i>		
ssp. <i>tuberosum</i>	4x	tbr
ssp. <i>andigena</i>	4x	adg
<i>neotuberosum</i>	4x	neo
<i>S. goniocalyx</i>	2x	gon
<i>S. phureja</i>	2x	phu
<i>S. stenotomum</i>	2x	stn
<i>S. verrucosum</i>	2x	ver
Series Pinnatisecta		
<i>S. cardiophyllum</i> ssp.		
<i>ehrenbergii</i>	2x	ehr
<i>S. jamesii</i>	2x	jam
<i>S. pinnatisectum</i>	2x	pnt
Series Acaulia		
<i>S. acaule</i>	4x	acl
Series Demissa		
<i>S. demissum</i>	6x	dem

Tab. 2 Scale used for visual assessment of enzymic browning in tubers

Score	Description
1	No discolouration
2	Faint discolouration at edges of tuber (approximately 10% of cut surface)
3	Dark discolouration at edges of tuber (approximately 10% of cut surface, but unacceptable)
4	Discolouration of 50–60% of cut tuber surface
5	Dark discolouration over 90% of cut tuber surface

Tab. 3 The discolouration of cut tuber surfaces after 5 hours, based on a scale of 1–5, in wild and cultivated potato species and interspecific hybrids

Species	Accession	Discolouration Score
Longipedicellata		
hjt	B371	1
	B372	2
	B373	1
	B376	3
	B371 X B372	1
	B372 X B376	2
	B373 X B372	2
	B375 X B376	1
	(B375 X B376) X B372	1
	fen	B354
plt	B553	2
	2521	2
pta	B382	1
	B392	2
sto	B399	1
	B508	3
Tuberosa		
phu	B2297	3
adg	B1478	4
	B1595	4
	B2621	4
tbr	'Desiree'	3
	'Duke of York'	2
	'Majestic'	2
	'Pentland Crown'	4
	'Pentland Dell'	3
Interspecific Hybrids		
hjt x fen	B371 X B354	4
	B372 X B354	4
fen x hjt	B354 X B371	1
	B354 X B376	3
hjt x plt	B371 X B553	5
	B376 X 2521	1
plt x hjt	B553 X B371	3
	B553 X B376	2
pta x hjt	B379 X B371	2
	B379 X B376	2
hjt x sto	B371 X B508	2
	B376 X B508	2
sto x hjt	B508 X B376	4
	B393 X B371	1
	B393 X B376	2
acl x hjt	B1889 X B376	2
hjt x phu	B376 X CIP701954	1
pta x neo	B382 X 77B/45	1
(sto x hjt) x gon	(B508 X B376) X CIP702494	1
(sto x hjt) x neo	(B399 X B371) X 77M/149	1

acceptable level of discolouration, but tubers of *S. tuberosum* 'Duke of York' and 'Majestic' had tuber discolouration scores as low as some of the *S. hjertingii* accessions.

In many of the hybrids between *S. hjertingii* and potential bridging species or cultivated diploid species, discolouration scores were low (Table 3). In crosses between *S. fendleri* B 354 and *S. hjertingii* B 371, hybrid progeny had a discolouration score of 1 when the latter was the male parent, but in reciprocal crosses, discolouration scores were high. The non-blackening character was transferred to hybrids with *S. phureja* from both *S. hjertingii* and *S. papita*. No hybrids between *S. hjertingii* and *S. tuberosum* were formed.

Crossability studies

A total of 1631 pollinations was made. The number of pollinations for each cross was not equal, as this was determined by the availability of flowers.

There was a relatively low seed set in intraspecific crosses of *S. hjertingii* (Table 4). In crosses with *S. tuberosum* ssp. *andigena* and ssp. *tuberosum*, as well as "neotuberosum", as females, virtually no seeds were formed, and most fruits were parthenocarpic. Reciprocal crosses with *S. hjertingii* as the male parent failed completely. In the $4x \times 2x$ crosses with cultivated diploid species, the percentage of parthenocarpic fruits was low, and several seeds per fruit were formed.

Tab. 4 Intra-specific crossability of *Solanum hjertingii*, and its crossability with tetraploid cultigens

Cross	Pollinations	Fruit	F/P*	% Parthenocarpic fruit	S/F	S/P
<i>4x x 4x</i>						
hjt x hjt	14	10	0.71	20	31.00	22.14
hjt x adg	36	7	0.19	57	1.14	0.22
adg x hjt	5	0	0	0	0	0
hjt x tbr	76	19	0.25	95	0.05	0.01
tbr x hjt	21	0	0	0	0	0
hjt x neo	19	7	0.37	86	0.57	0.21
<i>4x x 2x</i>						
hjt x gon	11	10	0.91	40	4.70	4.27
hjt x phu	17	7	0.41	0	1.71	0.71
hjt x stn	11	8	0.73	0	6.75	4.91

F/P = Fruits/Pollination; S/F = Seeds/Fruit; S/P = Seeds/Pollination

All crosses with other tetraploid species in Series Longipedicellata were successful, although there were reciprocal differences in crosses between *S. hjertingii* and *S. fendleri* and *S. papita* (Table 5). In most of the crosses, seed set was as high as in the intraspecific crosses of *S. hjertingii*.

Tab. 5 Crossability of *Solanum hjertingii* with potential bridging species

Cross	Pollinations	Fruit	F/P*	% Parthenocarpic fruit	S/F	S/P
Longipedicellata						
hjt x fen	33	9	0.27	22	35.11	9.58
fen x hjt	69	23	0.33	0	60.56	20.19
hjt x plt	29	12	0.41	0	50.67	20.97
plt x hjt	108	63	0.58	2	62.86	36.67
hjt x pta	14	2	0.14	50	29.50	4.21
pta x hjt	64	39	0.61	3	82.33	50.17
hjt x sto	34	20	0.59	0	47.25	27.79
sto x hjt	88	55	0.63	0	54.40	34.00
Pinnatisecta						
hjt x jam	5	1	0.20	0	40.00	10.00
jam x hjt	38	1	0.03	100	0	0
hjt x pnt	8	0	0	0	0	0
pnt x hjt	4	0	0	0	0	0
ehr x hjt	5	0	0	0	0	0
Acaulia						
hjt x acl	27	0	0	0	0	0
acl x hjt	64	23	0.36	0	90.08	32.37
Demissa						
hjt x dem	40	0	0	0	0	0
dem x hjt	147	99	0.67	14	24.69	16.63
Tuberosa						
hjt x ver	19	5	0.26	0	26.80	7.05
ver x hjt	3	0	0	0	0	0

In crosses of *S. hjertingii* with species in the other Series Pinnatisecta, Acaulia, Demissa and Tuberosa, there was evidence of considerable unilateral incompatibility. Most pollinations with diploids of Series Pinnatisecta failed, except for crosses with *S. jamesii*, and only then when it was the male parent. A similar situation was observed in the crosses with *S. verrucosum* (Series Tuberosa), and seeds were produced when the diploid species was the pollen parent in this $4x \times 2x$ cross (Table 5). In crosses with *S. acaule* and *S. demissum*, success was obtained only when *S. hjertingii* was the male parent.

The crossability of *S. tuberosum* with several species in three series was assessed (Table 6). In most of the crosses between *S. tuberosum* and other tetraploids in Series Longipedicellata, seed set was much higher than in crosses between *S. tuberosum* and *S. hjertingii*, with a couple of exceptions. Nevertheless in absolute terms, seed set was low. Crosses between *S. tuberosum* and *S. acaule* and *S. demissum* were only successful when the cultigen was the male parent. In general, the crossability of *S. tuberosum* with these species was higher than that observed in crosses of *S. hjertingii* with the same species.

A further evaluation was made of the crossability of *S. hjertingii* hybrids and *S. tuberosum* (Table 7). Seed set in all crosses was very low, but somewhat higher in the $4x \times 2x$ crosses between hybrids and cultivated diploids.

Tab. 6 Crossability of *Solanum tuberosum* with potential bridging species

Cross	Pollinations	Fruit	F/P*	% Parthenocarpic fruit	S/F	S/P
Longipedicellata						
fen x tbr	8	2	0.25	0	9.50	2.38
pta x tbr	7	5	0.71	20	19.40	13.86
tbr x pta	2	0	0	0	0	0
plt x tbr	33	3	0.09	67	4.00	0.36
sto x tbr	27	5	0.19	40	13.60	2.52
fen x adg	15	5	0.33	20	10.20	3.40
plt x adg	18	4	0.22	75	0.25	0.06
adg x plt	2	0	0	0	0	0
pta x adg	8	5	0.63	60	0.20	0.13
sto x adg	7	2	0.29	0	9.00	2.57
Acaulia						
acl x tbr	33	14	0.42	0	106.36	45.12
tbr x acl	2	0	0	0	0	0
acl x adg	10	2	0.20	0	74.50	14.90
Demissa						
dem x tbr	61	40	0.66	3	37.63	24.67
tbr x dem	2	0	0	0	0	0
dem x adg	8	6	0.75	0	31.17	23.38
adg x dem	1	0	0	0	0	0

Tab. 7 Crossability of bridging species hybrids with tetraploid and diploid cultigens

Cross	Pollinations	Fruit	F/P*	% Parthenocarpic fruit	S/F	S/P
(hjt x fen) x tbr	17	7	0.41	86	0.14	0.06
(fen x hjt) x tbr	3	3	1.00	100	0	0
(hjt x plt) x tbr	4	3	0.75	67	1.00	0.75
(plt x hjt) x tbr	8	8	1.00	75	5.63	5.63
(pta x hjt) x tbr	21	17	0.81	41	4.06	3.29
(hjt x sto) x tbr	15	9	0.60	78	2.78	1.67
(sto x hjt) x tbr	39	27	0.69	100	0	0
(hjt x fen) x neo	25	14	0.56	86	1.50	0.84
(plt x hjt) x neo	11	7	0.64	100	0	0
(pta x hjt) x neo	4	2	0.50	0	3.50	1.75
(sto x hjt) x neo	27	10	0.37	10	15.90	5.89
(hjt x sto) x gon	7	1	0.14	0	9.00	1.28
(sto x hjt) x gon	7	5	0.71	0	20.80	14.86
(hjt x fen) x phu	4	4	1.00	0	37.50	37.50
(sto x hjt) x phu	3	3	1.00	0	13.33	13.33
(hjt x sto) x stn	5	1	0.20	0	30.00	6.00
(sto x hjt) x stn	5	3	0.60	0	13.33	8.00
(acl x hjt) x tbr	9	2	0.22	0	2.00	0.44
(dem x tbr) x hjt	164	36	0.22	75	1.69	0.37

Discussion

The visual assessment of wounded tubers was a reliable guide to the expression of enzymic browning in parental stocks and hybrids, and had the advantage of being quick and easy to use. Most tests for tuber discolouration are based on a form of visual assessment of cut tubers (DRAPER et al. 1979), grated tuber tissue (FIRBAS 1961), or the more refined technique of using a pendulum system whereby surface damage, depth of bruise and extent of discolouration can be assessed at the same time (MASS 1966, HUGHES et al. 1975). Attempts at correlating the degree of enzymic browning with variation in polyphenoloxidase isozymes have not been successful (WOODWARDS 1982).

Although *S. hjertingii* had been reported as the main source of the non-blackening character, other species in Series Longipedicellata, such as *S. papita* and *S. stoloniferum*, also did not blacken. The fact that two *S. tuberosum* cultivars 'Duke of York' and 'Majestic' had an acceptable low level of discolouration in this study is interesting, because the latter has been reported as a cultivar which showed much enzymic browning (HUGHES and MAPSON 1967, in GRAY and HUGHES 1978). Undoubtedly growing conditions, affected by such factors as soils, rainfall and mineral nutrition, can have a substantial effect on the amount of discolouration in a particular cultivar, even from season to season. Consequently it is difficult to specify cultural practices which will give a consistent quality. We feel therefore that the incorporation of a genetic factor such as that found in *S. hjertingii* for non-blackening will ultimately be of value to potato producers in economic terms, even though the elimination of undesirable characters from wild species may be a slow process.

The utilisation of *S. hjertingii* does present many breeding problems, as the data reported here indicate. The low seed set in intraspecific crosses of *S. hjertingii* can be explained in terms of low pollen fertility (WOODWARDS 1982). The direct incorporation of the non-blackening character from *S. hjertingii* is difficult because of the lack of crossability with tetraploid *S. tuberosum*. Other Longipedicellata species can be crossed slightly more easily with *S. tuberosum*, but even in these the probability of success is not high. The failure of these crosses can be understood by reference to the Endosperm Balance Number hypothesis of JOHNSTON and HANNEMAN (1980) and JOHNSTON et al. (1980). Tetraploid Longipedicellata species appear to be 2EBN, whereas *S. tuberosum* is 4EBN. The apparent anomalous crossing behaviour of *S. polytrichon* (RAMANNA and ABDALLA 1970) and *S. acaule* (HERMSEN 1966), both also 2EBN, with diploid species and the successful production of triploid progeny in $4x \times 2x$ crosses can be explained in terms of this hypothesis. If conventional breeding strategies are to be utilised to transfer the non-blackening character, then bridging species may be one method to facilitate this effort. The use of cultivated diploid species such as *S. phureja*, *S. gonio-calyx* and *S. stenotomum* to form hybrids with *S. hjertingii*, and in which the non-blackening character has been inherited, has several advantages over the choice of wild bridging species, in terms of fewer undesirable agronomic characters. The success of direct crosses might be increased perhaps through

embryo culture, as there is some evidence that fertilisation does occur occasionally.

Although genetic studies were not undertaken on the inheritance of the non-blackening character in *S. hjertingii*, other data from WOODWARDS (1982) suggest that it is controlled by one or few major genes, which could be handled relatively easily in a breeding programme.

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Zusammenfassung

Das Fehlen der enzymatischen Verbräunung in Wildkartoffel-Arten der Serie Longipedicellata, und deren Kreuzbarkeit mit *Solanum tuberosum*

Das enzymatische Verbräunen aufgeschnittener Knollen verschiedener Species und Hybriden wurde untersucht. Die Knollen einiger mexikanischer Wildkartoffel-Species der Reihe Longipedicellata, besonders *Solanum hjertingii* ($2n = 48$), werden nach Verwundung nicht schwarz. Es wurde versucht, diese Eigenschaft auf Kulturkartoffeln zu übertragen. Über 1600 Bestäubungen zwischen *S. hjertingii* und Kulturkartoffeln wurden entweder direkt oder über andere Species als Kreuzungsbrücke durchgeführt. *S. hjertingii* konnte mit anderen Arten der Reihe Longipedicellata, mit *S. acaule*, *S. demissum* und anderen kultivierten diploiden Species gekreuzt werden, aber nicht direkt mit *S. tuberosum*. In einigen Hybriden ist keine Verbräunung festgestellt worden.

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