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Project Title: Utilization of Somaclonal Variants in a Backcrossing Program for Trichome-mediated Insect Resistance in Potato (*Solanum spp.*)

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The objectives of this research are: 1) to determine the stability of good trichome traits and continued capability of *S. tuberosum*-backcrossed somaclones to produce a high percentage of plants with trichomes type B with droplets; 2) to compare the expression of trichome traits and behaviour of somaclones derived from diploid crosses and tetraploid advanced hybrids, when they are backcrossed to *S. tuberosum* materials; and 3) to generate gene reassortment in the somaclones through backcrossing and select plants with good yield and agronomic traits and at the same time with high level of trichome traits which are associated with insect resistance.

A. Somaciones from Tetraploid Hybrids

Last year we reported that in two evaluations conducted, the same set of families - M502, M509, M514, M515 had high percentage of their progenies with B droplets while the other four controls - M501 (hybrid), M503, M504, and M505 had only a few or none of their progenies with droplets (Table 1). It was also observed that progenies from the first set (with higher B droplets) had better browning or better PPO activity (lower MEBA values) compared to the control set (with low or no B droplets).

Selected clones from each family which had droplets on their type B trichomes and at the same time had good browning were used as female parents,

backcrossed to either cv. Atlantic and/or the clone Q155-3. Selfing was also done. Progenies from these second backcrosses (BC2) were evaluated in the fall of 1989 for the presence of B droplets and among the four families with high B droplets, only the clone M514-1 backcrossed to Q155-3 maintained a high % of B droplets in its progenies; the rest had none or very few. Selfed progenies of all the clones of M502, M509, M514 and M515 however, produced a significantly higher percentage of their progenies with B droplets (73-100%). In the control set (M501, M503, M504 and M505), none or very few of the selfed and backcrossed progenies had B droplets. In terms of vine type, clones from the set without the B droplets were generally more of the Tuberosum-type while those with B droplets resembled more their wild parent, *S. berthaultii*. Third backcrossing (BC3) was made on selected progenies from BC2 (second backcross) and evaluation of these progenies will be done this fall of 1991.

About thirty five selected clones from the first evaluation in April, 1989 were planted in the field last summer of 1990. From these, ten clones were selected mainly for their tuber quality, vine type and their browning potentials. Six of these clones were used in the potato project's hybridization during the winter of 1990-1991. These were backcrossed to cv. Atlantic, Q155-3 or LB (late blight) bulk. Progenies of these second backcross (BC2) were evaluated for the presence of B droplets (Table 2). In this set, only the clone M502-6 backcrossed to Atlantic or Q155-3 had a significantly high percentage of its progenies with B droplets. Clones M509-3 and M515-1 backcrossed to LB Bulk had also high percentage of their progenies with B droplets. The results also show that using the cv. Atlantic or the clone Q155-3 as male parent does not seem to affect the expression of trichomes type B with droplets.

Rooted cuttings of selected 136 clones from the second batch of seeds evaluated in the spring of 1990 were planted in the field during the summer of 1990.

Out of the 136 clones with high B droplets, 40 clones were selected based on their tuber quality (shallow eyes, big tubers and bright skin were selected) and browning potentials (MEBA score). These clones together with some from the control set (M501, M503 and M504) were analyzed for their total glycoalkaloid (TGA) content. Results show that higher total glycoalkaloids, particularly chaconine and solanine were generally higher in clones with the B droplets as compared to those with low/without B droplets (control), although a very limited number of clones from the control set was included in the test (Table 3). Two tubers of each of these clones were allowed to sprout and planted as parent materials for the crossing work done during the winter. Table 4 shows the successful backcrosses and selfings that produced seeds from these crossing work. Evaluation of these progenies will be done this fall of 1991.

B. Somaclones From Diploid Hybrids (F1)

To determine how an F1 diploid hybrid would respond to culture in contrast to the more advanced tetraploid lines used earlier, Hopper (1989) regenerated plants from F1 hybrids produced by a cross between a dihaploid *S. tuberosum* line and *S. berthaultii*. About 150 regenerated plants were used for this particular study to determine whether somaclones from diploid hybrids will behave in the same way as the tetraploid advanced hybrid materials when backcrossed with *S. tuberosum*. Regenerated plantlets from in vitro were rooted and transplanted in the glasshouse for evaluations and for crossing work. Each somaclone was evaluated for its vine type, browning potentials and the presence of droplets on trichomes type B. Thirty five somaclones which were either diploid or tetraploid were selected and backcrossed to *Tuberosum* clones (cv. Atlantic, Q155-3 and HH1-9, a diploid clone previously used as parent in the production of these hybrids). Crossing was done at the diploid and tetraploid levels because both diploid and tetraploid plants were

recovered (Table 5). All the plants flowered but successful backcrossing and selfing was obtained only from a few somaclones (Table 6). Progeny evaluations of these materials will be done in the fall of 1991.

Table 1. Sets of families evaluated for the presence of droplets in trichomes type B.

		1988*	1989	1990
Set I (High B droplets)				
1. M 502	4001 x Atlantic	44	40	25
2. M 509	4029 x Atlantic	25	22	35
3. M 514	4038 x Atlantic	33	32	36
4. M 515	4040 x Atlantic	39	40	27
Set II (Low B droplets)				
5. M 501	F743-4 x Atlantic	3	0	5
6. M 503	4002 x Atlantic	2	0	1
7. M 504	4004 x Atlantic	2	3	0
8. M 505	4009 x Atlantic	2	0	2
* Lentini (1989)				

Table 2. Evaluation of selected BC2 for the presence of droplets in trichomes type B.

Pedigree	Percentage of plants	
	with B droplets	
M502-6 x Atlantic		31
M502-6 x Q155-3		28
M509-3 x LB Bulk		25
M514-1 x LB Bulk		19
M514-1 X Q155-3		16
K88-60 x M514-1		0
K88-380 x M514-1		6
M514-7 x Atlantic		0
M514-7 x Q155-3		0
M514-7 x LB Bulk		0
M515-1 x Atlantic		0
M515-1 x Q155-3		0
M515-1 x LB Bulk		21
M515-7 x Atlantic		6
M515-7 x Q155-3		6
M515-7 x LB Bulk		0

Table 3. Total glycoalkaloid content of selected clones from BC2 evaluated last May 1991

Clone	Total Glycoalkaloids (mg/100g)			Solanine
	Solasonine	Solamargine	Chaconine	
Set I High B droplets				
M502-101	1.30	1.60	20.80	33.40
M502-102	4.40	4.00	35.70	25.30
M502-103	1.30	0.40	20.00	23.90
M502-104	0.00	0.00	31.00	30.80
M502-105	1.80	0.00	5.30	4.40
M502-106	47.00	18.90	0.00	0.00
M502-107	1.60	1.50	2.80	2.50
M502-108	0.00	0.00	19.10	16.90
M509-101	0.00	0.00	22.50	25.60
M509-102	24.80	20.70	0.80	0.00
M509-103	3.40	3.20	21.20	17.90
M509-104	2.50	0.70	18.00	18.20
M509-105	1.60	1.10	33.20	24.60
M509-106	0.00	0.00	22.90	17.80
M509-107	0.00	0.00	17.70	19.10
M509-108	0.00	0.00	21.50	24.50
M509-109	1.40	2.40	29.10	24.30
M509-110	0.00	0.00	4.60	6.90
M514-101	0.00	0.00	17.20	17.50
M514-102	0.00	0.00	39.70	29.20
M514-103	5.20	4.20	46.10	36.80
M514-104	4.30	3.40	53.60	36.20
M514-105	4.60	0.00	31.30	20.30
M514-106	2.90	3.20	21.10	22.20
M514-107	1.10	0.40	11.40	9.30
M514-108	3.50	3.20	29.70	28.60
M514-109	2.50	2.20	17.90	19.90
M514-110	0.00	0.00	32.30	29.70
M514-111	0.00	0.00	12.10	10.00
M515-101	2.20	1.00	39.70	24.50
M515-102	2.10	0.70	26.30	25.30
M515-103	3.00	1.50	34.50	29.50
M515-104	0.00	0.00	11.50	16.40
M515-105	7.70	6.70	47.80	34.60
M515-106	0.00	0.00	32.10	29.60
M515-107	0.00	0.00	12.90	0.00
Set II Low B droplets				
M501-4	2.00	1.50	38.80	20.20
M501-5	0.00	0.00	21.20	22.50
M503-3	21.40	23.30	0.00	0.00
M503-4	0.80	1.40	8.70	15.60
M504-1	0.00	0.00	3.60	6.70

Table 3. Total glycoalkaloid content of selected clones from BC2 evaluated last May 1990.

Clone	Total Glycoalkaloids (mg/100g)			Solanine
	Solasonine	Solamargine	Chaconine	
M504-2	0.00	0.00	3.30	4.00
M504-3	0.00	0.00	8.20	5.10
M504-4	0.60	0.40	11.50	11.00
M504-5	0.00	0.00	19.60	23.20

Table 4. Successful second and third backcrosses done during the winter of 1990-1991.

Clone	Male Parent		Selfed
	Q155-3	ATLANTIC	
A. SECOND BACKCROSS			
M502-101	X	X	
M502-102		X	
M502-104	X	X	
M502-105	X	X	
M502-107	X	X	x
M509-101		X	
M509-102		X	
M509-103	X	X	x
M509-105	X	X	x
M509-110	X	X	
M509-106		X	x
M509-107		X	
M509-108		X	
M509-109	X	X	
M514-101		X	
M514-102	X		x
M514-103	X		
M514-104		X	x
M514-105	X	X	
M514-106	X	X	x
M514-107	X		x
M514-108	X		
M514-109			x
M514-111	X	X	x
M514-126		X	x
M515-101	X	X	x
M515-103	X	x	
M515-104		x	
M515-106		X	x
M515-109		X	
M501-101	X	x	x
M501-102		x	
M501-103	X	x	x
M501-104	X	x	
M501-105	X		
M501-106	X		
M501-107	X	x	
M501-108		x	
M501-109	X	x	
M501-110		x	x
M501-111	X		

Table 4. Successful second and third backcrosses done during the winter of 1990-1991.

A. SECOND BACKCROSS	Q155-3	ATLANTIC	
M503-103	X		
M503-104			x
M504-101	X	x	x
M504-102	X	x	
M504-103		x	
M504-104	X		x
M504-105	X	x	x
M505-101		x	
M505-104	X	x	x
M505-105			x
B. THIRD BACKCROSS			
N312-3	X	x	
N312-14	X	x	
N312-17		x	
N312-28		x	
N312-32	X	x	
N312-37	X	x	
N312-38	X	x	
N312-45	X	x	
N312-46	X	x	x
N308-1	X	x	
N308-2	X	x	
N308-4		x	
N308-5	X	x	
N308-7	X		
N314-5	X	x	
N314-9		x	
N314-15		x	
N314-17	X	x	
N304-1	X	x	
N304-3		x	
N304-6	X	x	
N304-9		x	
N304-10	X	x	
N305-9		x	
N306-1	X	x	
N306-3		x	
N306-4	X	x	
N317-6	X	x	

Table 5. Results of the evaluation of somaclones from diploid hybrids (F1).

PLANT NUMBER	PLOIDY LEVEL	VINE TYPE		MEBA		DROPLETS	
		45 DAP	60 DAP	45 DAP	60 DAP	TYPE B	TYPE A
Controls:							
HH1-9	2n	4	4	98,81	70		
S. ber. PI473331	2n			30	38	present	5
Norland	4n						
F1 Parents:							
2	2n	4	2	79	87		2
3	2n	3	3	47	58	present	5
6		4	3	71	64		
8		3	4	73	83		
10		4	5	57	44		
11	2n	2	2	42,35	54,64		
11	2n	4	3	56,48	35		
12		3	2	61	40		
15		4	2	93	70		
16	2n	3	1	30,35	87,74		
17		4	3	45	73		
18		3	3	51	51		
20		4	4	87	95		
21		4	4	60	68		
Regenerates:							
2.01	2n	2	2	69	67		1
2.02	2n	2	2	88	90		3
2.03	4n	4	3	73	76	present?	5
2.05	2n	4	2	64	89		2
2.06	4n+	3	3	73	55		5
2.07	2n	3	2	84	89		3
2.08	2n	3	2	75	94		1
2.09	2n	4	2	83	79		2
2.10	2n	4	2	65	58		4
2.11	4n	5	4	75	72		3
2.12	2n	3	2	84	82		2
2.13	2n	4	3	92	89		1
2.14	2n	3	2	71	67		3
2.15	4n	5	3	55	49		5
2.16	4n	4	3	65	51		5
2.17	4n	5	4	45	49		5
2.28	2n	4	3	78	82		2
2.29	4n	5	4	55	64		5
2.30	2n	3	3	89	81		1
2.31	4n	3		74	53		3

Table 5. Results of the evaluation of somaclones from diploid hybrids (F1).

PLANT NUMBER	PLOIDY LEVEL	VINE TYPE		MEBA		DROPLETS	
		45 DAP	60 DAP	45 DAP	60 DAP	TYPE B	TYPE A
2.32	4n	4		62	53		4
2.33	4n	5	4	62	48		5
2.34	4n	5	4	67	73		4
2.35	4n	4	3	71	85		3
2.36	4n	3	4	73	61		4
2.37	4n+	5	3	70	61		4
2.38	2n	3	1	92	74		1
2.39	4n	4	3	66	39		4
2.40	4n	4	1	56	69	present	5
2.41	4n	5	3	70	38		5
2.42	4n-	3	3	79	42		5
2.43	4n	3	1	65	58	present	4
2.44	4n-	4	2	67	55		5
2.45	4n	5	3	72	55		4
2.47	4n	3	2	71	44		4
2.48	2n	2	1	82	74		3
2.49	4n	4	3	50	58		5
2.50	2n	2	1	88	82		1
2.52	4n	3	2	68	37		5
2.53	4n	4	3	74	49		4
2.55	4n	4	3	54	37		5
2.56	4n	5	3	81	52		4
2.57	4n	3	2	72	65		3
2.58	4n	5	3	51	48		5
2.59	4n	3	2	72	65		3
2.60	4n	4	3	42	45	present	5
2.64	2n	3	3	65	90		
2.66	2n	3	2	81	77		2
2.67	2n	3	2	64	68		3
2.74	2n	3	2	83	74		2
2.75	2n	3	2	85	76		2
2.76	2n	3	2	82	64		3
2.77	2n	2	1	84	81		2
2.78	2n	2	1	82	72		2
2.80	2n	3	2	99	67		3
2.81	2n	4	3	89	71		2
3.01	2n	4	3	49	48		5
3.02	2n	4	2	56	48	present	5
4.01		2	2	54	55		4
4.04		3	2	31	33		4
4.05		3	2	40	43		5
4.08		2	2	45	45		4
4.09		4	3	49	36		4
4.11	2n	5	2	50	39		3

12

Table 5. Results of the evaluation of somaclones from diploid hybrids (F1).

PLANT NUMBER	PLOIDY LEVEL	VINE TYPE		MEBA		DROPLETS	
		45 DAP	60 DAP	45 DAP	60 DAP	TYPE B	TYPE A
4.12	2n	2	2	65	46		3
4.14	4n-	5	2	35	54		4
11.04	4n+	5	5	35	34		
11.05	2n	3	3	44	50	present?	5
11.10		4	3	29	37		5
11.11	4n-	3	3	26	61		4
11.16	4n	4	3	29	53		5
11.17	4n	4	4	28	49	present?	5
11.18	2n	3	3	39	42		3
11.19	4n	4	3	35	32		
11.20	4n+	4	3	21	53		5
11.21	4n+	3	4	43	54		
11.22	4n+	4		24	33		
11.24		5	4	38	29	present?	5
11.25	4n	5	4	39	25	present?	5
11.26	4n	4	3	38	41		5
11.27	4n	4	3	35	58		5
11.28	2n	3	2	47	51		1
11.29	2n	3	3	56	43	present?	5
11.30	2n	4	3	45	44		5
11.32	4n	4		48	19		
11.33	4n	5	4	39	25	present	5
11.35	4n	3	4	47	27	present?	4
11.36	4n	4	3	36	55		5
11.37	4n	5	4	37	28	present?	5
11.38	4n	4	3	45	69		5
11.39	2n	2	2	40	73		3
11.40	2n	2	2	42	67		3
11.41	2n	4	4	51	39		3
11.42	2n						
11.43	2n	3	3	37	38		5
11.44	2n	3	3	48	35		5
11R.01	4n	4	4	24	52		5
16.01	2n	4	3	48	69		4
16.02	2n	4		55	80		
16.04	2n	4	2	47	70		5
16.05	2n	4	4	74	64	present	4
16.06	2n	4	3	66	35		4
16.08	2n	4	3	42	72		4
16.10	2n	3	2	53	64		2
16.11	2n	4	3	59	56		4
16.12	2n	3	2	49	52		3
16.15	2n	4	3	53	67		4

Table 5. Results of the evaluation of somaclones from diploid hybrids (F1).

PLANT NUMBER	PLOIDY LEVEL	VINE TYPE		MEBA		DROPLETS	
		45 DAP	60 DAP	45 DAP	60 DAP	TYPE B	TYPE A
16.16	2n	4	3	46	61	present	3
16.17	2n	3	2	61	60		5
16.18	2n	3	3	62	70		2
16.19	2n	4	3	62	55		5
16.20	2n	4	3	52	60		5
16.22	2n	4	3	50	70		4
16.23	2n	4	3	50	55		5
16.24	2n	4		49	54		
16.25	4n	4		57	77		
16.27	4n	4	4	55	47		3
16.28	4n	4	3	28	22		5
16.29	4n	4	2	35	26		5
16.30	4n	4		69	48		
16.31	4n+	4		60	40		
16.33	4n	4		46	38		
16.34	4n	4		46	61		
16.38	4n	4		61	48		
16.39	4n	4		65	46		
16.40	4n	4		53	48		
16.41	4n	4		54	45		
16.42	4n+	5		36	56		
16.43	4n	3		73	22		
16.44	4n	4		67	44		
16.45	4n	4		49	58		
16.46	4n	4		70	54		
MEBA : Modified Enzymic Browning Assay							
Vine Type: 1=berth., 3=intermediate, 5=tbr.							
Type A Droplets: 1=none/very few, 3=some, 5=many							

Table 6. Successful backcrosses and selfs of diploid and tetraploid somaclones.

Clone Number	Male Parent	Selfed
2-15		x
2-17	Q155-3, Atlantic	x
2-32	Atlantic	
2-40	Q155-3, Atlantic	x
2-41	Q155-3, Atlantic	x
2-43	Q155-3, Atlantic	x
2-52		x
2-55	Atlantic	x
2-60		x
11-10	Q155-3	
11-17	Q155-3	x
11-24		x
11-25	Q155-3	x
11-40	Q155-3	
16-29		x

Reciprocal Crosses

Atlantic	2-1
Atlantic	2-15
Atlantic	2-17
Q155-3	2-60
Atlantic	3-1
Atlantic	11-4
Atlantic	11-30
Atlantic	16-8
Atlantic	16-29
Q155-3	16-6