

Inheritance of Three Electrophoretically Determined Protein Bands in Potato (Solanum tuberosum L.)*

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Summary. A modified polyacrylamide gel electrophoresis technique is employed to resolve proteins for use as biochemical gene markers in potato. Dominant, duplicate dominant and complementary gene action are three modes of inheritance that adequately explain the segregation of three respective protein bands in two generations of crossing within diploid Phureja × haploid Tuberosum families.

Key words: PAGE — Proteins — Biochemical gene markers — Inheritance — Solanum tuberosum L.

Introduction

The presence and position of protein bands separated by polyacrylamide gel electrophoresis (PAGE) from potato tuber extract are known to be genetically determined (Stegemann 1975). Changes in climate or soil, treatment with growth regulators, size of mature tubers, fertilization practices or several months of storage of mature tubers do not alter the banding pattern of an individual cultivar (Stegemann 1975; Zacharius et al. 1971; Zwartz 1967). PAGE of potato proteins from mature tubers has been suggested as a tool to determine biochemical gene markers (Desborough and Peloquin 1966; Stegemann and Loescheke 1977). Such markers could be helpful in furthering our understanding of potato genetics. In this study PAGE was used to investigate the inheritance of three prominent protein bands from the tubers of an experimental population of diploid x haploid potatoes (Solanum tuberosum L.).

Materials and Methods

Potato clones were derived from four and five generations of intercrossing three diploid Phujera X haploid Tuberosum clones: US-W5278.1, US-W5336.6, US-W5340.1 (see Desborough and Weiser 1972, for parentage). The fourth generation consisted of twelve families containing a total of 178 individuals, while the fifth generation consisted of 10 families and a total of 141 individuals (see Table 1). The small numbers reflect the reduced vigor of these plants due to inbreeding depression and possibly some virus infection.

Tuber proteins were extracted as described by Desborough and Peloquin (1966) with the addition of 1 ml 30% (v/v) DMSO to the macerated tuber. 74% polyacrylamide slab gels, 0.75 mm thick, with a pH of 4.3 were formed in a Hoefer Scientific Instruments apparatus according to the formulations in Table 2. Twenty-five μ l of tuber extract was applied to each sample slot. Proteins were separated after 1½ hours of 60 mAmps current per gel slab. The protein bands were stained with 0.5% (w/v) Aniline Blue-Black in 74% (v/v) acetic acid.

Results and Discussion

About thirty different bands can be resolved with this method; any one sample may contain up to sixteen bands (Fig. 1). Three of the bands segregating in these samples were usually darkly stained and sharply defined, and so were chosen for the inheritance study. These were numbered 12, 13 and 15. On the basis of the frequencies observed for each band in the F_3 parents and the F_4 progenies, models were proposed for the inheritance of these three bands. Subsequently, the F_5 progenies were examined to determine whether the proposed models were valid.

In five families, where neither parent had the band, fully half the progeny possessed band 12 (Table 3). The simplest model to explain such segregation is complementary gene action. This model postulates two complementary, independently segregating genes, each with one dominant and one recessive allele. Both genes must

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Table 1. Partial pedigree of potato clones studied

F_3 parents	Identification Number of F ₄ progeny family	n	F ₄ parents	Identification Number of F ₅ progeny family	n
US-W9302-49 × US-W9302-48	193	8	206-62 × 202-10	137	5
US-W9302-49 × US-W9305-29	202	6	206-62 × 196-19	133	17
US-W9324-51 × US-W9323-73	205	7	209-6 × 196-19	144	27
US-W9324-51 × US-W9305-7	197	24	209-13 × 196-19	147	13
US-W9305-31 × US-W9305-7	203	6	209-10 × 196-19	146	16
US-W9324-39 × US-W9305-7	196	18	209-10 × 207-14	138	15
US-W9324-39 X US-W9325-4	206	56	$207-23 \times 202-2$	141	7
US-W9324-39 X US-W9325-24	207	25	$208-2 \times 202-2$	142	28
US-W9324-39 X US-W9325-1	208	3	208-2 × 197-11	134	2
US-W9324-39 X US-W9325-7	209	13	206-18 × 193-8	135	11
US-W9302-31 X US-W9302-7	195	8			
US-W9302-31 × US-W9324-59	204	4			

have a dominant allele in order for band 12 to be expressed. When either gene is homozygous recessive, band 12 is absent. Table 3 shows the frequencies of phenotypes and the proposed genotypes for band 12. On the basis of the P-values from chi-square tests of this model, (Table 3), the two complementary genes suffice to explain the observed banding in generations 4 and 5.

The most straightforward model to explain the inheritance of band 13 is control by a single dominant gene causing expression of the band. Table 4 shows the frequencies of phenotypes and the proposed genotypes for band 13. P-values from chi-square tests (Table 4) indicate this simple model adequately explains the data in the two generations.

Duplicate dominant genes were proposed for control of band 15 due to the 15:1 and 7:1 ratios observed (Table 5). This model postulates two independently segre-

Table 2. Formulations for slab PAGE at pH 4,3 as suggested by Peloquin et al. 1975

Stock Solutions
A: 30.0 gm Acrylamide, 0.8 gm bis-acrylamide, H ₂ O to 100 ml
B: 48 ml 1 N KOH, 17.2 ml glacial acetic acid, H ₂ O to 200 ml
C: 48 ml 1 N KOH, 2.87 ml glacial acetic acid, H ₂ O to 200 ml

	Running gel	Stacking gel
A:	10.0 ml	1.5 ml
В:	10.0 ml	_
C:	_	2.5 ml
H ₂ O:	19.6 ml	5.9 ml
10% Ammonium persulfate:	0.2 ml	0.1 ml
TEMED:	0.2 ml	5.0 µ1

Reservoir buffer: 31.2 gm β -alanine, 8.0 ml glacial acetic acid, H_2 O to 10 liters.

gating genes each with one dominant and one recessive allele. Band 15 appears when either dominant allele is present. Both genes must be homozygous recessive for band 15 to be absent. Table 5 indicates frequencies of phenotypes and the proposed genotypes for band 15. Again, the P-values from chi-square tests of this simplest possible model show significant agreement between expected and observed banding patterns for both generations 4 and 5.

These three bands appear to have rather simple inheri-

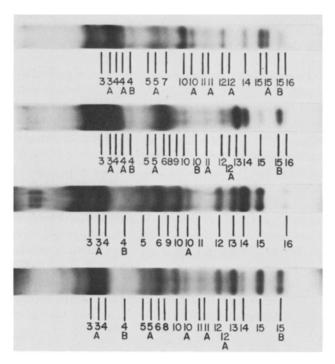


Fig. 1. Sample gel of tuber extracts showing numbering system employed. Bands 12, 13, and 15 chosen for study

Table 3. Model for the inheritance of band 12. Complementary gene action

	Parents with band	Parenta genoty propos	pes		Progeny families	Theoretical distribution of band 12 in the progeny	Observed	Expected	P from X ² test
4	neither	AAbb	X	aaBb	196, 203, 206, 207, 208	1 present : 1 absent	60 : 48	54 : 54	0.25
	neither	bb	Х	bb	202	all absent	0:6	0:6	_
Generation	one	AABb	Χ	aaBB	204	all present	4:0	4:0	_
ıer	one	AABb	Х	aaBb	197, 209	3 present : 1 absent	27:10	28:9	0.75
Ger	one	AABb	Х	bb	193, 205	1 present : 1 absent	8:7	7.5:7.5	0.90 - 0.75
J	both	AABb	X	AaBb	195	3 present : 1 absent	5:3	6:2	0.50 - 0.25
	neither	Aabb	X	AAbb	141	all absent	0:7	0:7	
n 5	one	Aabb	Х	AaBb	133	3 present : 5 absent	7:10	6:11	0.50 - 0.75
tio	one	Aabb	Χ	AABb	135	1 present : 1 absent	6:5	5.5:5.5	0.75
era	one	AaBb	Χ	AAbb	137, 142	1 present : 1 absent	20:13	16.5:16.5	0.25 - 0.10
Generation	one	AaBB	Χ	Aabb	144, 146, 147	3 present: 1 absent	40:16	42:14	0.75 - 0.50
	both	AaBB	Χ	AaBb	134, 138	3 present : 1 absent	14:3	13:4	0.75 - 0.50

Table 4. Model for the inheritance of band 13. Single dominant gene action

	Parents with band	Parental genotypes proposed	Progeny families	Theoretical distribution of band 13 in the progeny	Observed	Expected	P from X ² test
4	neither	dd × dd	193	all absent	0:8	0:8	_
Generation	one	$Dd \times dd$	197, 202, 207, 206, 209, 205	1 present : 1 absent	74:57	65.5 : 65.5	0.25 - 0.10
Gene	both	Dd × Dd	195, 196, 204, 208, 203	3 present : 1 absent	31:9	30 : 10	0.75 - 0.50
- ~	neither	dd × dd	137	all absent	0:5	0:5	_
	one	$Dd \times dd$	135, 138, 141, 142	1 present : 1 absent	33:28	30.5 : 30.5	0.50
Generation	one	$DD \times dd$	133, 144, 146, 147	all present	73:0	73:0	-
	both	$Dd \times Dd$	134	3 present: 1 absent	1:1	1.5:0.5	0.50 - 0.25

Table 5. Model for the inheritance of band 15. Duplicate dominant gene action

	Parents with band	Parental genotypes proposed	Progeny families	Theoretical distribution of band 15 in the progeny	Observed	Expected	P from X ² test
	one	Eeff × eeff	193, 202	1 present : 1 absent	6:8	7:7	0.75 - 0.50
Generation 4	both	$EE\times E$ or $F-$	195, 196, 206, 207, 208, 209	all present	123:0	123:0	_
era	both	$EeFf \times EeFf$	197	15 present : 1 absent	22:2	22.5:1.5	0.75 - 0.50
en	both	$EeFf \times Eeff$	203, 205	7 present : 1 absent	11:2	11:2	_
G	both	$Eeff \times eeFf$	204	3 present : 1 absent	2:2	3:1	0.25
	one	Eeff × eeff	137	1 present : 1 absent	2:3	2.5 : 2.5	0.75 - 0.50
'n 5	both	$EE \times$	133, 134, 138,	all present	97:0	97:0	_
ţį		Eor F -	141, 144, 146,	-			
Generation			147				
	both	$EeFf \times Eeff$	135	7 present : 1 absent	9:2	10:1	0.50 - 0.25
	both	$Eeff \times eeFf$	142	3 present : 1 absent	22:6	21:7	0.75 - 0.50

tance patterns which may make them useful as gene markers. Further investigation is needed to determine whether other protein bands might also prove good markers. It should be feasible to establish linkage groups of biochemical gene markers to be used in conjunction with the pigmentation markers now known, in the hope of more fully characterizing the potato genome.

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