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1) Electrophoretic classification of selected *G. max* plant introductions and named cultivars in the late maturity groups.

Over the last few years, we have been classifying various *G. max* and *G. soja* lines based on their electrophoretic zymogram patterns. In the 1982 Soybean Genetics Newsletter (Gorman et al., 1982), we published a table compiling electrophoretic profiles for most of the named soybean cultivars in the early maturity groups (000-IV). Listed on the table below is a similar compilation of electrophoretic profiles (zymogram types for 12 enzyme systems) for 60 *G. max* PIs (10 originating from Northeast China, 10 from Central China, 20 from Korea, and 20 from Japan) as well as 90 named cultivars in the late maturity groups (V-VIII). Descriptions of these zymogram types and/or information on their inheritance patterns have been published previously (Gorman, 1983; Gorman et al., 1983; Kiang and Gorman, 1983; Gorman et al., 1982; Kiang, 1981; Gorman and Kiang, 1978). More than one variable locus has been identified in either *G. max* or *G. soja* for several of the enymzes (ADH, Am, Dia, LAP, IDH, PGD, PGM and TO) and all of the zymograms, except MPI, represent the products of more than one enzymatic locus. Thus, most of the zymogram types represent multiple loci phenotypes. Each number in the body of the table represents the zymogram type observed for the particular enzyme (columns) and cultivar or PI (rows). The enzyme abbreviations used were: ADH for alcohol dehydrogenase, Am for amylase, AP for acid phosphatase, Dia for diaphorase, GPD for glucose-6-phosphate dehydrogenase, IDH for isocitrate dehydrogenase, LAP for leucine amino peptidase, MPI for mannose-6-phosphate isomerase, PGD for phosphogluconate dehydrogenase, PGI for phosphoglucose isomerase, PGM for phosphoglucomutase and TO for tetrazolium oxidase. The initial electrophoretic screening consisted of an examination of five seeds (electrophoresed in four different gels) from each cultivar or PI, tested for all 12 enzymes. If all of the enzymes were not satisfactorily resolved, or when unusual results (i.e., mixed zymogram types) were obtained, additional seeds were tested in subsequent electrophoretic runs. Thus, each number in the body of the table represents the observations made on a minimum of five seeds. When two or more numbers are listed, the cultivar or PI had a mixture of these zymogram types. Cultivars and PIs were considered to have a mixed zymogram only when seeds were classified into two or more zymogram types on repeated electrophoretic runs. When only one or two seeds from a single electrophoretic run showed a different zymogram from the majority of seeds for that line, these seeds were considered atypical and the cultivar or PI was not considered mixed. It was felt that one or two unreplicated seeds with abnormal zymograms were more likely the result of scoring or seed handling mistakes by us, rather than line impurities. PI seeds were obtained from Dr. R. L. Bernard (USDA-ARS at the University of Illinois), while seeds from the late maturity cultivars were obtained from Dr. T. C. Kilen (USDA-ARS at Stoneville, MS).

Zymogram Types

PI or Cultivar	Enzymes											
	ADH	Am	TO	AP	LAP	PGD	GPD	PGM	Dia	MPI	IDH	PGI
Northeast China												
103414	2	1	1	2	1	1	1	2	1	2	6	1
103415	1	2	1	3	1	2	1	2	4	1	3	1
103419	1	1	1	1	1	1,2	2	2	1	1	7	1
135589	1	1	1	3	1	2	1	2	2	2	2	1
135590	1	1	1	2	1	2	1	2	2	2	1	1
232987	1	1	1	1		2	1	2	2	2	1	1
232988	1	1	1	3		2	1	2	2	2	1	1
232989	1	1	1	3		2	1	2	2	2	1	1
232990	1	1	1	3		2	1	2	2	2	1	1
232991	1	1	1	3		1	1	2	2	2	2	1
Central China												
103080	1	2	1	3		1	1	1	1	2	1	1
103088	1	1	1	3		1	1	2	2	2	1	1
103091	1	2	1	2	1	1	1	1	1	2	1	1
123577	1	1	1	2	1	1	2	2	2	1	3	1
158765	1	1	1	2	1	1	1	1	3	3	3	1
253650A	1	1	1	2	1	1	2	1	2	1	7	1
253650B	1	1	1	1	1	1	1	2	4	2	5	1
253651A	1	1	1	2	1	1		1	1	2	3	1
253651B	1	2	1	1	1	1	2	1	2	2	7	1
253653A	1	1	1	2	1	1	1	2	1	2	3	1
Korea												
157395	1,3	1	1	2	1	1	2	2	2	2	7	1
157396	1	1	1	2	1	1	2	1	2	2	7	1
157396	1	1	1	2	1	1	1	2	2	2	7	1
157398	3	1	1	3	1	1,2	1	1	2	2	7	1
157401	1	1	1	1	1	1	2	2	1	2	3	3
157402	1	1	1	2	1	1	1	2	2	2	7	1
157404	1	1	1	2	1	2	2	2	2	2	7	1
157405	1	2	1	2	1	1,2	1	2	2	2	7	1
157408	1	1	1	2	1	1	2	1	2	2	7	1
157409	1	1	1	2	1	2	1	1	2	5	7	1
157410	1	2	1	1	1	1	1	1	1	2	7	1
157414	3	1	1	2	1	1	2	1	1	2	7	1
157416	1	1	1	2	1	1	2	2	1	2	3	1
157417	1	1	1	2	1	2	1	2	1	1	7	1
157419	1	1	1	2	1	2	1	1	1	1	7	1

Zymogram Types

PI or Cultivar	Enzymes											
	ADH	Am	TO	AP	LAP	PGD	GPD	PGM	Dia	MPI	IDH	PGI
157421	1	2	1	3	1	1	1	2	2	2	7	1
157424	1	2	1	2	1	1	1	2	1	2	7	1
157428	1	2	1	2	1	1	2	2	1	2	7	1
157429	1	2	1	1	1	1	2	1	2	3	7	1
157431	1	1	1	2	1	1	2	1	2	2	7	1
Japan												
124871	1	2	1	3	1	2	1	1	1	1	7	1
181531	1	1	1	2	1	2	2	2	1	5	7	1
181532	1	1	1	2	1	2		2	1	5	7	1
181533	1	1	1	2	1	1	2	2	4	2	3	1
181534	1	1	1	2		1	1	2	1	1	7	1
181535	3	1	1	2	1	1	1	2	2	2	6	1
181536	1	1	1	3	1	2	1	1	2	2	7	1
181537	1	1	1	2	1	1		1	2	2	7	1
181538		1	1	2	1	1	2	2	4	1	3	1
181539		1	1	2	1	1	2	1	4	2	3	1
181540	1	1	1	2	1	2		1	2	5	7	1
181541	1	1	1	2	1	2	1	1	2	5	7	1
181542		1	1	2	1	2		1	8	5	7	1
181548		1	1	2	1	2		2	2	1	7	1
181549		1	1	2	1	2		1	1	5	5	1
181550	3	1	1	2	1	2	2	1	2	1	8	1
181551	1	1	1	2	1	1	2	1	2	2	5	1
181552	1	1	1	2	1	2	2	2	1	1	7	1
181553	1	1	1	2	1	1	2	1	2	5	7	1
181554	1	1	1	2	1	1	2	1	2	5	7	1
Southern Maturity												
Acadian	1	1	1	2	1	1	2	2	2	3	3	1
Arisoy	1	1	1	2	1	1	2	2	2	3	3	1
Arksoy	1	1	1	2	1	1	1	1	2	2	3	1
Armredo	1	1	1	2	1	1	1	2	3	2	3	
Avoyells	1	1	1	2	1	1	2	2	1	2	5	1
Barchet	1	1	1	2	1	1	2	2	1	2	3	1
Bedford	1	1	1	2	1	1	2	1	1	2	5	1
Biloxi	1	1	1	2	1	1	1	2	1	2	3	1
Bosier	1	1	1	2	1			1	1	2	5	
Bragg	1	1	1	2	1	1	2	1	1	2	5	1
Centennial	1	1	1	2	1	1	2	1	2	2	5	1
Charlee	1	1	1	2	1	1	1	1	1	3	1	1
Cherokee	1	1	1	2	1	1	2	2	1	3	3	1
Creole	1	1	1	2	1	1	1	2	2	3	3	1
Clemson	1	1	1	2	1	1	1	2	2	1	1,3	1

Zymogram Types

PI or Cultivar	Enzymes											
	ADH	Am	TO	AP	LAP	PGD	GPD	PGM	Dia	MPI	IDH	PGI
Cobb	2	1	1	2	1	1	1	1	2	3	5	1
Coke Stewart	1	1	1	2	1	1	2	2	1	1	1	1
Columbus	1	1	1	2	1	1	1	2	1	2	5	1
Dare	1	1	1	2	1	1	1,2	1	2	2	7	1
Dortschoy	1	1	1	2	1	1	1,2	2	1,2	2	5,7	1
Davis	3	1	1	2	1	1	1	1	2	2	7	1
Delsta	1	1	1	2	1	2	2	2	1	1	2	1
Delsoy	2	1	1	3	1	1	1	2	2	1	7	1
Dixie	1	1	1	1	1	1	1	2	2	2	3	
Dyer	1	1	1	2	1	1	2	1	1,2	2	7	1
Easycook	3	1	1	1	1	1	1	1	1	2	3	1
Essex	3	1	1	2	1	1	2	1	1	2	5,7	
Forrest	1	1	1	2	1	1	2	1	1	2	5	1
Gaton	1	1	1	2	1	1	1	2	1	3	3	1
Georgian	1	1	1	2	1	1	1	2	2	2	3	1
Hampton 266	1	1	1	2	1	2	2	1	1	1	5	1
Haberlandt	1	1	1	3	1	1	1	1	2	2	7	1
Hardee	3	1	1	2	1	1	1	1	2	2	1	1
Harvell	1	1	1	2	1	2	1	2	7	2	7	1
Hayseed	1	1	1	2	1	1	1	1	2	2	1	1
Hinn	1	1	1	2	1	1	1	2	1	2	1	1
Hollybrook	1	1	1	2	1	2	1	1	2	2	7	1
Hood	1	1	1	2	1	1	1	2	2	2	7	1
Hutton	1	1	1	2	1	1	2	1	1	2	7	1
Imp'd. Pelican		1	1	1	1	1	1	2	1	3	3	1
Jackson	2	1	1	2	1	2	1	2	1	3	5	1
Jew 45	1	1	1	2	1	2	1	2	1	1	5	1
Jupiter	1	1	1	2	1	1	2	1	1	2	7	1
Kino	1	1	1	2	1	1	1	1	1	2	7	1
La Green	1,2	1	1	2	1	1	2	2	1,2	1	3	1
Laredo	1	1	1	2	1	1	1	2	4	2	1	1
Lee	1	1	1	2	1	1	2	1	2	2	5	1
Lee 68	1	1	1	2	1	1	1	1	1	2	1,5	1
Lee 74	1	1	1	2	1	1	1,2	1	1,2	2	1,5	1
Luthy	1	1	1	3	1	1	2	1	8	1	7	1
Mack	1	1	1	2	1	1	1	1	2	2	5	1
Magnolia	1	1	1	2	1	1	1	1	2	2	3	1
Majors	1	1	1	2	1	1	1	2	1	1	3	1
Mamloxi	1	1	1	2	1	1	2	2	1	3	2	1
Mammoth Yellow	1	1	1	2	1	1	2	3	1	2	1,3	1
Mamredo	1	1	1	2	1	1	1	2	2	2	5	
Manotan 6640	1	1	1	2	1	2	2	2	1	3	2	1
Missoy	1	1	1	2	1	1	1	1	1	3	1	1
Monetta	1	1	1	2	1	1	1	1	2	3	1	1
Nanda	1	1	1	2	1	2	1	2	1	1	5	1

Zymogram Types

PI or Cultivar	Enzymes											
	ADH	Am	TO[a]	AP	LAP[b]	PGD	GPD	PGM	Dia	MPI	IDH	PGI
Nansemond	1	1	1	2	1	2	1	2	7	2	7	1
Nela	3	1	1	1	1	2	1	2	2	1	8	1
Old Dominion	1	1	1	2	1	1	1	2	2	2	1	
Palmetto	1	1	1	2	1	1	1	2	1	3	1	1
Pickett	1	1	1,2	2	1	1	2	1	1	2	5	1
Pickett 71	1	1	1		1	1	2	1	1	2	5	1
Pine Delta Perfection	1	1	1	2	1	1	1	2	2	2	3	1
Pluto	1	1	1	2	1	1	1	2	1	2	1	1
Pochahontas	1	1	1	2	1	1	1	1	1	1	5	1
Ralsoy	1	1	1	2	1	1	1	1	2	2	3	1
Roanoke	2	1	1	2	1	2	1	1	2	2	5	1
Rokusun	1	1	1	2	1	2	1	1	1	1	5,6	1
Rose Non-Pop	1	2	1	3	1	1	2	1	2	2	7	1
S-100	2	1	1	2	1	1	2	1	1	2	5	1
Seminole	1	1	1	2	1	1	1	2	1,2	2	3	1
Semmes	1	1	1	2	1	2	2	1	1	2	7	1
Tanner	1	1	1	2	1	1	1	2	1	3	3	1
Tarheel Black	1	1	1	2	1	1	1	2	1	2	3	1
Tenn Non-Pop	2	1	1	2	1	2	1	2	2	1	5	1
Tokyo	1	1	1	2	1	2	1	2	2	2	7	1
Tracy and Tracy-M	1	1	1	2	1	2	2	1	2	2	7	1
Volstate	3	1	1	2	1	2	1	1	2	2	5	1
White Biloxi	1	1	1	2	1	1	2	2	1	3	3	1
Wood Yellow	1	1	1	2	1	1,2	1	2	1	2	6	1
Yelnanda	1	1	1	2	1	1	1	2	1	1	4	1
Yelredo	1	1	1	2	1	1	1	2	1	1	4	1
York	1	1	1	2	1	1	1	1	2	2	5	1
Total number of seeds scored:	847	750	849	797	788	1033	810	788	908	700	1113	750
Total number of mixed lines:	2	0	1	0	0	3	3	0	5	0	7	0
Total number of atypical seeds:	2	0	0	2	0	6	2	2	3	2	8	0

[a]Only the variable locus To_4 was scored in this table (Type-1 vs. Type-2 TO zymograms), an additional variant TO zymogram (Type-3) was not tested for.

[b]Only the variable locus Lap_1 was scored in this table (zymogram types 1, 2 or 3), an additional LAP variable locus (Lap_2) was not scored for.

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2) Linkage of electrophoretic loci.

In our studies of the inheritance of various electrophoretic variants, we have examined F_2 segregation data from many crosses (see Gorman, 1983, for a listing). Many of these crosses were segregating for multiple loci, allowing linkage data to be collected. Table 1 represents a summary of the linkage patterns we have observed between the listed electrophoretic loci (see Gorman and Kiang, 1978; Kiang, 1981; Gorman et al., 1983, concerning the establishment of these loci). Only the locus pairs for which we had data from a minimum of 95 F_2 seeds were included in Table 1, while just those pairs that showed an independent segregation pattern based on greater than 300 F_2 seeds, or a conclusive linkage pattern, were listed without question marks. The question marks were used to indicate insufficient data to firmly establish a linked pair or to detect the possibility of weak linkage. The product method (Immer and Henderson, 1943) was used to calculate F_2 recombination percentages and standard errors. To facilitate the use of the product method, those loci showing a codominant segregation pattern had their heterozygous class bulked with one of their homozygous classes. Sixty-six gene pairs had F_2 segregation ratios consistent with independent assortment, but in 45 of those pairs the number of seeds tested was too small to be able to eliminate the possibility of weak linkage. The segregation data and appropriate χ^2 values for those 7 gene pairs found to have a recombination fraction 2 or more standard errors below 0.5 are listed on Table 2. Three of these gene pairs, Adh_1 with Adh_4 (Gorman and Kiang, 1978), *Ap* with Lap_1 and *Pgd* with *Pgi*, showed conclusive linkage results. The establishment of possible linkage of Am_3 with Lap_1 and of Idh_1 with Lap_1 was clouded by the significant deviation (from 3:1) observed in their single-locus ratios. The data for the *Ap* with Idh_1 and *Ap* with Idh_3 gene pairs are consistent with weak linkages, but the number of F_2 seeds tested was too

small to detect a significant deviation from independence with a χ^2 test. However, since the *Ap* and Lap_1 loci are clearly linked and since both the Idh_1 with Lap_1 and *Ap* with Idh_1 gene pairs gave indications of linkage, it seems likely that *Ap*, Lap_1 and Idh_1 belong in the same linkage group. Hildebrand et al. (1980) reported that the *Ti* and *Ap* loci were linked, belonging to linkage group 9. Therefore, Lap_1 can be placed in linkage group 9, with Idh_1 likely also belonging to this group. Since Am_3 showed independent segregation with *Ap*, Idh_1, and *Ti* (Gorman, 1983; Hildebrand et al., 1980; Orf and Hymowitz, 1977), its membership in this group is questionable. The linkage groups to which the Adh_1 with Adh_4 and the *Pgd* with *Pgi* gene pairs belong is not known.

Table 1. Linkage relationships between electrophoretic loci

	Loci														
	Adh_4	Am_3	*Ap*	Di_1	Di_2	Di_3	*Gpd*	Idh_1	Idh_2	Idh_3	Lap_1	*Mpi*	*Pgd*	*Pgi*	Pgm_1
Adh_1	L	I?[a]		I?			I?	I?	I?		I?	L?			
Am_3			I?	I	I?		I?	I	I?	I	?	I?			I
Ap				I?				L?		L?	L		I?	I?	I?
Di_1					I?	I?	I	I?	I	I?	I	I?	I?		I?
Di_2						I?	I?	I?	I?	I	I?	I?	I?		I?
Di_3										I?		I?	I?		
Gpd								I?	I	I	I?	I	I		I
Idh_1									I	I?	L?	I?	I?		I?
Idh_2										I	I	I?			I
Idh_3											I?	I?	I?		I
Lap_1												I?			I
Mpi													I		I
Pgd														L	I
Pgi															

[a]Only gene pairs with 95 or more segregating F_2 seeds were included in the table, while only gene pairs with greater than 300 segregating F_2 seeds or conclusive linkage data were typed without a question mark. I stands for independent assortment, while L stands for linked loci.

Table 2. Examination of the gene pairs with a recombination fraction of 2 or more S.E. below 0.5

Gene Pair A : B	Phase	Phenotypic class A	B	C	D	χ^2(3:1) Locus A	χ^2(3:1) Locus B	χ^2(9:3:3:1) Loci A & B	Product/Method %R	S.E.	Conclusion
Adh_1 : *Adh_4*	C	315	3	6	122	3.25	2.17	484.0**	2	1%	Linked
Am_3 : *Lap_1*	R	562	192	168	34	7.6**	0.95	13.81**	43	3%	Questionable
Ap : *Idh_1*	R	65	37	32	7	0.49	2.00	7.10*	36	6%	Possible, but inconclusive
	C	19	4	5	3						
Ap : *Idh_3*	R	29	7	13	1	0.09	3.60	3.14	38	6%	Possible, but inconclusive
	C	53	10	15	6						
Ap : *Lap_1*	R	452	202	214	12	0.21	0.21	60.21**	23	3%	Linked
Idh_1 : *Lap_1*	C	185	53	70	42	9.17**	0.85	21.90**	40	4%	Possible, but inconclusive
Pgd : *Pgi*	C	112	14	19	27	0.28	0.12	42.80**	21	4%	Linked

*Significant χ^2 deviation at the 0.1 level. **Significant χ^2 deviation at the 0.01 level.

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3) Inheritance of a second leucine aminopeptidase locus and tests of its linkage with other loci.

In dry soybean seed, only one leucine amino peptidase (LAP) anodal band was observed by acrylamide slab gel electrophoresis (Gorman et al., 1983). This band gradually declined in intensity and disappeared in all tissues about 10-12 days after germination. Three mobility variants (Rf's 0.59, 0.53, and 0.58) were observed in the band, which was controlled by a single locus (Gorman et al., 1983; Kiang and Gorman, 1983; Gorman, 1983).

A second LAP anodal band (Rf 0.80) was detected in all tissue of both *G. max* and *G. soja* plants about 8 days after germination. Cotyledons from 12- to 15-day-old seedlings were used to screen for the second LAP variant by acrylamide slab gel electrophoresis.

One activity variant and three mobility variants of the second LAP band were observed among 400 *G. max* cultivars and 140 *G. soja* accessions examined. The null activity variant found in cultivar 'Jefferson' did not show enzyme activity in any tissue. Based on the progeny of the crosses between 'Amsoy' and Jefferson, Jefferson x Amsoy and Jefferson and 'Wilson', genetic analysis indicated that the null was recessive to the active allele, and the F_2 segregated into a 3:1 ratio (Table 1). Since there was no difference between reciprocal crosses, it was controlled by a nuclear gene. This second LAP locus is designated *Lap2* to distinguish it from the first LAP locus (*Lap1*), and the null allele is designated as *lap2*.

Table 1. The F_2 data segregating for leucine aminopeptidase activity at the *Lap2* locus in soybean

Cross	Zymogram: Genotype:	Present *Lap2*	Null *lap2 lap2*	χ^2 (3:1)	P
Jefferson x Amsoy *lap2 lap2* x *Lap2 Lap2*		98	28	0.519	0.6
Amsoy x Jefferson *Lap2 Lap2* x *lap2 lap2*		105	33	0.093	0.9
Total		203	61	0.505	0.6

The F_2 dihybrids segregation data were used to test linkage relationships by the maximum likelihood method (Allard, 1956). The results showed that the *Lap1* and *Lap2* loci were unlinked (Table 1A), and the *Am3* locus (Gorman and Kiang, 1978) was found to be inherited independently of the *Lap2* locus (Table 2B). The F_2 data from dihybrids involving the *Lap2* and the hypocotyl color trait were examined. The F_2 data showed that the purple hypocotyl color was dominant to the green, and segregated into a 3:1 ratio. The hypocotyl color is controlled by the gene pair (W_1 w_1) that also controls flower color by pleiotropic effect (Bernard and Weiss, 1983; Palmer and Payne, 1979). The linkage test indicated that the *Lap2* locus was linked with the hypocotyl color locus W_1 with 40.9±4.0 map units between them (Table 2C). Thus, *Lap2* would belong to linkage group 8 (Palmer and Kaul, 1983). The estimate is based on a sample size of 268 plants, and a larger sample size is needed to make a more accurate estimate.

Table 2. The F_2 data from soybean dihybrids segregating for several gene pairs

	Gene pair	Cross	Phase	Phenotype frequency a	b	c	d	χ^2 (9:3:3:1)
A.	*Lap1-Lap2*	Jefferson x Wilson	R	214	80	87	31	3.758
B.	*Am3-Lap2*	Amsoy x Jefferson	R	107	31	32	11	0.638
C.	*Lap2-*W_1	Amsoy x Jefferson	R	154	50	39	25	6.653

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