Transfer of High Seed Protein to High-Yielding Soybean Cultivars[1]

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ABSTRACT

An effective breeding strategy is needed for the development of high-protein, high-yielding soybean [*Glycine max* (L.) Merr.] cultivars. This study was conducted to evaluate a backcross strategy for transferring genes for high seed protein from a low-yielding plant introduction to high-yielding lines with average protein content. Pando, a plant introduction with low yield and about 480 g kg^{-1} of seed protein was crossed to three high-yielding lines with an average protein content of about 400 g kg^{-1}. Selection for high protein was practiced between two generations of backcrossing to the high-yielding parent. After two backcrosses, 95 random BC_2F_2-derived lines from each population were evaluated for seed yield, protein, and oil content in Iowa. None of the BC_2F_2-derived lines equaled the protein content of Pando, but an average of 72% of the lines from the three populations had significantly higher protein than the recurrent parent. On average, about 19% of the BC_2F_2-derived lines with a protein content significantly higher than the recurrent parent were not significantly different ($\alpha = 0.05$) from the recurrent parent in seed yield. The results indicated that when a low-yielding, high-protein donor parent is utilized, selection for high protein between two backcross generations would effectively increase the seed protein in the backcross progeny above that of the high-yielding recurrent parent and still result in some progeny having a seed yield not significantly different from that parent.

Additional index words: ***Glycine max*** **(L.) Merr., Seed yield, Oil percentage, Protein percentage, Plant breeding, Backcross method.**

CULTIVARS of soybean [*Glycine max* (L.) Merr.] with above average seed protein concentrations of about 450 g kg^{-1} on a dry-weight basis are preferred for some human foods, such as tofu. High-protein cultivars have been released periodically in the northern USA, but they typically yield less than commercial cultivars.

One possible strategy for developing high-protein, high-yielding cultivars is to transfer genes responsible for high protein content into high-yielding cultivars by backcrossing. Plant introductions are available that have 480 to 500 g kg^{-1} protein, but they are much lower in yield than current cultivars. Cianzio and Fehr (1982) evaluated the segregation for seed composition in BC_0, BC_1, and BC_2 populations in which the donor parents were plant introductions with 492 to 494 g protein kg^{-1} and the recurrent parents were high-yielding cultivars with 396 to 412 g protein kg^{-1}. They observed that in the absence of selection for seed composition between backcross generations, the mean protein content of the populations decreased with each backcross generation. The lines with the highest protein in the BC_2 populations had 52 to 70 g kg^{-1} lower protein than the high-protein donor parents and 26 to 30 g kg^{-1} higher protein than the recurrent parents. They suggested that selection among and within F_2-derived lines before each backcross generation might permit the development of lines with higher protein than they were able to recover without selection.

The relationship between yield and protein content in backcross populations was studied by Hartwig and Hinson (1972). The seed yield of the donor parent used in their study was 75% that of the recurrent parent, and the protein content of the donor parent was 112% that of the recurrent parent. They evaluated progenies of one BC_1F_1 and one BC_2F_1 plant whose segregation for seed composition resembled that of the BC_0 population. They obtained significantly negative correlations between seed yield and protein content among BC_1- derived lines. These correlations became nonsignificant after the second backcross. They concluded that when low-yielding germplasm is used as the donor parent, selection within BC_2 populations is more likely to be effective for identifying high-protein, high-yielding segregates than selection within BC_0 or BC_1 populations.

The studies of Cianzio and Fehr (1982) and Hartwig and Hinson (1972) collectively provided guidelines for developing a backcross strategy for the transfer of genes for high protein, but no empirical study has been conducted to evaluate the effectiveness of such a strategy. The objective of this study was to evaluate the seed yield and protein concentration of BC_2F_2-derived lines developed in a backcross program designed to transfer genes for high protein from a low-yielding plant introduction to a high-yielding cultivar, with selection for protein content practiced between backcross generations.

MATERIALS AND METHODS

The parents used in this study were the plant introduction Pando, 'Weber', and two experimental lines, A76-202015 and A76-304020. Pando was chosen because it was one of the donor parents used by Cianzio and Fehr (1982). It has about 480 g protein kg^{-1} and a seed yield about 85% less than that of the three high-yielding parents. The high-yielding parents represent the three maturity groups grown in Iowa. Weber of Maturity Group I, A76-202015 of Maturity Group II, and A76-304020 of Maturity Group III ranked first in their respective maturity groups in the 1977 Uniform Soybean Yield Tests, Northern States.

Development of BC_2 populations was initiated when Pando was crossed as female to each of the high-yielding parents in the field at Ames, IA, during the summer of 1978. In each cross, one F_1 seed was harvested from each of six plants. The six F_1 seed from each cross were planted at the Iowa State University-University of Puerto Rico soybean nursery at Isabela, PR, in November 1978, and F_2 seed were harvested. The F_2 seed of each cross were planted in Puerto Rico in February 1979, and 200 random F_2 plants per cross were harvested individually.

Twenty F_3 seed from each of the 200 F_2 plants were analyzed for protein content by near-infrared analysis at the

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USDA Northern Regional Research Center at Peoria, IL. Some 50 F_2 plants were selected from the 200 plants in each cross based on high protein content. The progeny of these selected plants were grown as F_2-derived lines in the F_3 generation at Ames in 1979 in a randomized complete-block design with two replications. The plots were single rows 1.5 m long, with a row spacing of 85 cm, and a planting rate of 30 seed per plot. One seed from each of 20 random plants of each plot was bulked. These bulked plot samples were analyzed for protein content, and the 10 F_2-derived lines with the highest protein averaged across replications were selected from the 50 lines in each cross. Twenty F_3 plants from each selected line were threshed individually, and their seed were analyzed for protein and oil content. The 10 F_3 plants from each cross with the highest protein content were chosen as parents for the first backcross.

The progeny of the 10 selected F_3 plants (F_3-derived lines) of each cross were mated as females to their respective recurrent parent in the field at Ames in 1980. One BC_1F_1 seed was harvested from each of four random plants of each F_3-derived line. The four BC_1F_1 seed from each F_3-derived line were planted in Puerto Rico in November 1980, and each BC_1F_1 plant was harvested individually. A seed sample from each BC_1F_1 plant was analyzed for protein content, and the plant with the highest protein content from each of the 10 lines was selected. The BC_1F_2 seed from each selected plant were sown in Puerto Rico during February 1981, and 20 random plants were harvested individually from each of the 10 progeny rows within each cross.

A sample of BC_1F_3 seed from each of the 200 BC_1F_2 plants was analyzed for protein content, and 50 plants with the highest protein were selected from each cross. Progeny of the 50 BC_1F_2 plants (BC_1F_2-derived lines) were evaluated in a yield trial at Ames in 1981, using the same field plot techniques as for the BC_0F_2-derived lines in 1979. The BC_1F_3 plants of each BC_1F_2-derived line were harvested individually, a bulk sample of seed from each plot was analyzed for protein content, and the 25 lines (out of 50) with the highest protein were selected within each cross. Each selected BC_1F_2-derived line in the BC_1F_4 generation was further evaluated for protein composition in Puerto Rico during November 1981. The lines were grown in two replications of a randomized complete-block design. The plots were 60 cm long, with a row spacing of 60 cm, and a planting rate of 12 seed per plot. A bulk seed sample harvested from each plot was analyzed for protein content. The 10 BC_1F_2-derived lines (out of 25) with the highest protein were selected for each cross. The progeny of 10 individual BC_1F_3 plants from each of the selected BC_1F_2-derived lines were evaluated in Puerto Rico in January 1982, using the same field plot techniques as in the November 1981 planting. A bulk seed sample was harvested from each plot, the sample was evaluated for protein content, and the BC_1F_3-derived line with the highest protein from each of the 10 BC_1F_2 families was selected for use as a parent in the second backcross.

The 10 BC_1F_3-derived lines from each cross were mated as females to their respective recurrent parent. One BC_2F_1 seed was harvested from each of six random plants of each BC_1F_3-derived line. The BC_2F_1 seed obtained at Ames in 1982 were planted in Puerto Rico in November 1982, each plant was harvested individually, and its seed were analyzed for protein content. The BC_2F_1 plant with the highest protein from each of the 10 BC_1F_3-derived lines was identified, its BC_2F_2 seed were planted in Puerto Rico in February 1983, and 20 random plants were harvested individually.

The progeny of the 200 BC_2F_2 plants (BC_2F_2-derived lines) of each cross were grown at Ames in 1983 in unreplicated hill plots. The single-hill plots were spaced 1 × 1 m and were planted with 12 seed. For each cross, 95 lines with maturity similar to the recurrent parent were threshed individually in bulk.

Seed of the 95 BC_2F_2-derived lines and the recurrent parent of each cross was increased at Ames in 1984. The lines and recurrent parents were evaluated for seed yield, protein content, and oil content in 1985. The plots were two rows 5 m long, with 70 cm between rows within a plot, and 1 m between rows of adjacent plots. The planting rate was 30 seed per meter of row. The plots were end-trimmed to 3 m before harvest. The Weber population was planted at Ames and Manson, the A76-202015 population at Ames and Marshalltown, and the A76-304020 population at Stuart and Ottumwa, IA. Data for seed yield was based on the weight of the harvested sample after drying at 40°C for 2 days. Protein and oil content were measured with a near-infrared analyzer and expressed on a dry weight basis. The analyses were conducted in cooperation with C.R. Hurburgh at Iowa State University.

The changes in yield and seed composition during backcrossing were assessed for each cross. The recurrent parent, donor parent, the 10 F_3-derived lines used as parents for the first backcross, and the 10 BC_1F_3-derived lines used as parents for the second backcross were grown in replicated tests in 1984 and 1985 at Ames. Two replications of single-hill plots spaced 1 × 1 m were used each year.

Standard analysis of variance procedures were used to summarize the data from the experiments. The BC_2F_2-derived lines and the locations were considered random effects. For the analysis of the 1984 and 1985 data involving the donor and recurrent parents and the BC_0F_3 and BC_1F_3 parents of each backcross, entries were considered a fixed effect and years a random effect.

The least significant difference (LSD) was calculated for those traits that had significant mean squares for entries in the analysis of variance. For comparing means of individual BC_2F_2-derived lines, the LSD value was calculated using the equation $LSD = t_{df,\ 0.05}\ (2EMS/n)^{1/2}$, where EMS= error mean squares used for estimating the significance of the line effect and n = number of values used in computing a line mean. For comparing means of the 10 F_3-derived lines used as parents of the first backcross with the 10 BC_1F_3- derived lines used as parents of the second backcross, the LSD was calculated using the equation $LSD = t_{df,\ 0.05}\ (2EMS/n)^{1/2}$, where EMS = error term used for estimating the significance of the generation effect and n = number of values used in computing the means of the 10 F_3- or 10 BC_1F_3-derived lines. For comparing means of the donor or recurrent parents with those of the 10 F_3- or BC_1F_3-derived lines, the LSD was calculated using the equation $LSD = t_{df,\ 0.05}\ [EMS(1/n1 + 1/n2)]^{1/2}$, where EMS = error term used for estimating the significance of the generation effect, $n1$ = number of values used in computing the mean of the 10 F_3-or BC_1F_3-derived lines and $n2$ = number of values used in computing the mean of the donor or recurrent parent.

RESULTS

In all three crosses, the population means were significantly lower in seed yield and oil content and higher in protein content than those of their recurrent parent (Table 1). Two backcrosses were adequate to recover high-yielding lines. On average, about 37% of the BC_2F_2-derived lines in the three populations were judged to be not significantly different in yield from their recurrent parent (Table 2). The highest-yielding line in each population had 12 to 14 g kg^{-1} higher

Table 1. Mean performance of 95 BC_2F_2-derived lines, the highest protein line, the highest-yielding line, and the recurrent parent for each of the three populations averaged over two locations.

Entry	Character		
	Yield	Protein	Oil
	g m^{-2}	g kg^{-1}	
Weber population			
Population mean	229*	379*	239*
Highest protein	258	422	246
Highest yielding	274	373	250
Recurrent parent	267	361	248
$LSD_{0.05}$†	39	20	9
A76-202015 population			
Population mean	247*	390*	231*
Highest protein	249	433	238
Highest yielding	303	376	236
Recurrent parent	287	362	244
$LSD_{0.05}$†	30	16	8
A76-304020 population			
Population mean	294*	437*	208*
Highest protein	267	462	198
Highest yielding	336	427	212
Recurrent parent	328	413	220
$LSD_{0.05}$†	25	10	4

* Population mean significantly ($\alpha = 0.05$) different from the recurrent parent.

† Least significant difference ($\alpha = 0.05$) to compare the highest-protein line and the highest-yielding line with the recurrent parent.

Table 2. Percentage of 95 BC_2F_2-derived lines from each of the three populations not significantly ($\alpha = 0.05$) different from the recurrent parent in yield (Yield-NS), significantly higher in protein (Protein-S), and significantly higher in protein while not significantly different in yield (Yield-NS, Protein-S) averaged over two locations.

Population	Character		
	Yield-NS	Protein-S	Yield-NS Protein-S
	%		
Weber	54	40	15
A76-202015	31	85	22
A76-304020	25	90	19

protein than the recurrent parent (Table 1), but the difference in yield and protein was not statistically significant, except in the A76-304020 population.

Selection for protein between backcross generations was effective in obtaining a high frequency of high-protein lines. On average, about 72% of the BC_2F_2-derived lines were significantly higher in protein content than their recurrent parent (Table 2). The difference in protein content between the highest protein line and the recurrent parent was 61 g kg^{-1} in the Weber population, 71 g kg^{-1} in the A76-202015 population, and 49 g kg^{-1} in the A76-304020 population.

On average, about 19% of the BC_2F_2-derived lines from the three populations were significantly higher in protein and not significantly different in yield from the recurrent parents (Table 2, Fig. 1). Of the lines that were included in this category, the 14 lines in the Weber population averaged 31 g kg^{-1} higher; the 21 lines in the A76-202015 population averaged 24 g kg^{-1} higher; and the 18 lines in the A76-304020 population averaged 21 g kg^{-1} higher protein than their recurrent parents. Although the lines were not significantly different in yield from the recurrent parent, only one line

Table 3. Genotypic correlations between yield, protein content, and oil content for 95 BC_2F_2-derived lines from three populations.

Population	Characters correlated		
	Yield-protein	Yield-oil	Protein-oil
Weber	−0.86	0.83	−1.03
A76-202015	−0.64	0.70	−0.98
A76-304020	−0.54	0.53	−0.98

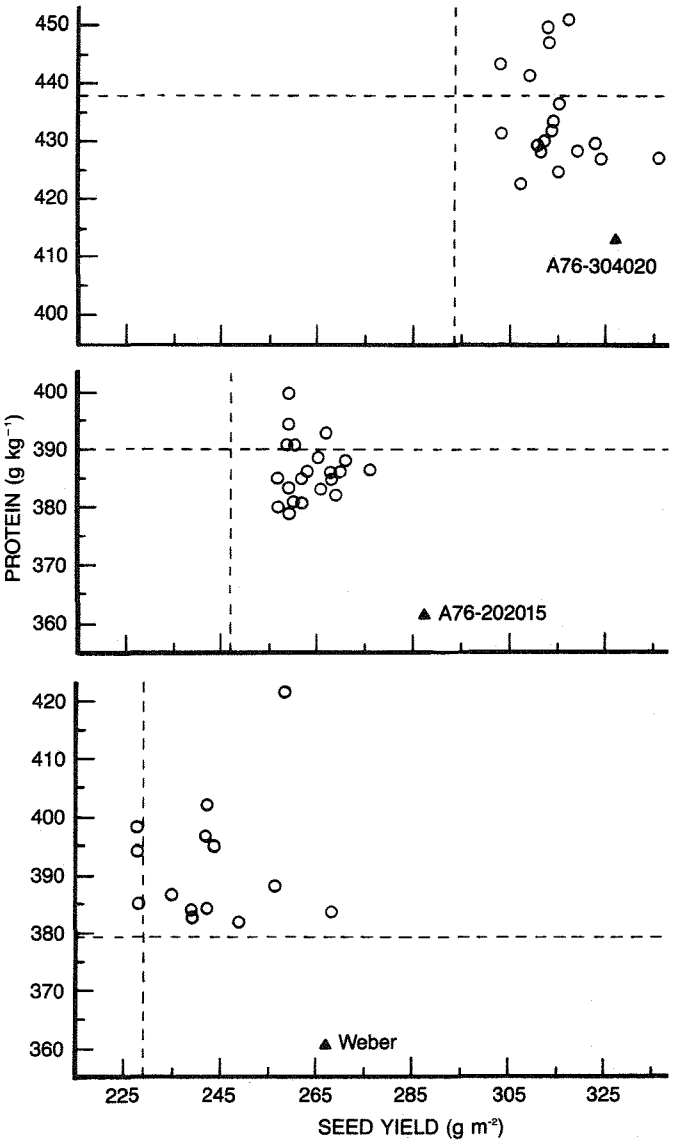

Fig. 1. Protein content plotted as function of yield for the BC_2F_2-derived lines that were not significantly different from the recurrent parent in yield but were significantly higher ($\alpha = 0.05$) than the recurrent parent in protein content, when averaged over two test locations in 1985. Yield and protein content of recurrent parents are represented by the solid triangles. Means of the three populations for yield are represented by the vertical dashed lines and, for protein content, by the horizontal dashed lines.

in the Weber and A76-304020 populations and none of the lines in the A76-202015 population were equal to or higher in yield than the recurrent parent (Fig. 1). Yield and protein content had negative genotypic correlations in the three populations (Table 3). In contrast, there was a positive correlation between yield and oil content. The lines with significantly higher protein than their recurrent parent were generally lower

Table 4. Means and ranges of the three populations for yield, protein content, and oil content of Pando, the recurrent parent, the 10 F_3-derived line parents of the first backcross, and the 10 BC_1F_3-derived line parents of the second backcross averaged over 2 yr.

Entry	Yield		Protein		Oil	
	Mean	Range	Mean	Range	Mean	Range
	g m^{-2}		g kg^{-1}			
Weber population						
Pando	31		470		179	
Parents of BC_1	177	105–212	443	418–462	195	177–217
Parents of BC_2	186	157–232	436	418–450	202	180–220
Recurrent parent	236		388		228	
$LSD_{0.05}$†	13		5		4	
$LSD_{0.05}$‡	30		12		8	
A76-202015 population						
Pando	31		478		177	
Parents of BC_1	126	82–162	466	440–481	179	146–218
Parents of BC_2	213	151–243	436	425–447	196	177–213
Recurrent parent	279		398		220	
$LSD_{0.05}$†	22		8		6	
$LSD_{0.05}$‡	51		19		15	
A76-304020 population						
Pando	28		482		170	
Parents of BC_1	170	94–217	448	423–467	188	146–190
Parents of BC_2	173	109–212	449	440–459	186	164–194
Recurrent parent	219		401		207	
$LSD_{0.05}$†	10		7		4	
$LSD_{0.05}$‡	24		17		9	

† Least significant difference ($\alpha = 0.05$) to compare the mean performance of the parents of the first backcross with that of the parents of the second backcross.

‡ Least significant difference ($\alpha = 0.05$) to compare the mean performance of the parents of the first backcross with that of the donor parent and the parents of the second backcross with that of the recurrent parent.

in oil than the recurrent parent, as reflected by the significant negative correlation between the two traits.

There was a fourfold or greater difference in yield between Pando and the mean of the F_3-derived and BC_1F_3-derived lines used as parents for the first and second backcross in the three populations (Table 4). The mean yields of the parents of the two backcross generations were significantly less than that of the recurrent parent of each population. The mean protein content of the parent lines for the first and second backcrosses were significantly greater than that of the recurrent parent, but significantly less than that of Pando. The differences in yield and protein content between the parents of the first and second backcrosses were not consistent among the three populations, being considerably smaller in the Weber and A76-304020 populations than in the A76-202015 population.

DISCUSSION

The strategy evaluated in this study for the development of high-yielding cultivars with high seed protein involved a low-yielding, high-protein donor parent and high-yielding, average-protein recurrent parents. Two backcross generations were used, as proposed by Hartwig and Hinson (1972), and selection for high protein was practiced between the backcross generations, as suggested by Cianzio and Fehr (1982). The results indicated that two backcrosses were sufficient to recover lines that were not significantly different at the $\alpha = 0.05$ level from the yield of the recurrent parent, even though the yield of the donor parent was only about 15% of that of the recurrent parents. The evaluation of the F_3- and BC_1F_3-derived lines used as parents for the first and second backcrosses suggested that, in some crosses, fewer than two backcrosses may be sufficient. Hartwig and Hinson (1972) based their suggestion for two backcrosses on the significant difference in yield between lines derived from the BC_1 and BC_2 generations in one cross. In our study, there was a significant difference in yield between the F_3- and BC_1F_3-derived lines used as parents in the A76-202015 cross, and none of the BC_1F_3-derived lines used as parents for the BC_2 approached the yield attained by the recurrent parent, which indicated that two backcrosses were minimally necessary (Table 4). In the Weber and A76-304020 crosses, however, there was not a significant difference in mean yield between the parents of the first and second backcrosses, and some of the BC_1F_3-derived lines used as BC_2 parents were not significantly different in yield from the recurrent parent. For these two crosses, yield evaluation of progeny from the BC_1 and, possibly, from the BC_0 population may have been effective in identifying high-yielding lines with high protein. The evaluation for yield of lines from each generation of a backcross program may be advisable instead of yield testing lines only from the BC_2 population.

An assessment of the effectiveness of selection for protein content was made by comparing the results of this study with those of Cianzio and Fehr (1982), in which no selection was practiced between backcrosses. Both studies used Pando as the donor parent and high-yielding lines with average protein as the recurrent parents. The mean of BC_2F_2-derived lines was 12 g kg^{-1} higher in protein than the recurrent parent in their study, whereas the means for the three populations in this study were 18 to 28 g kg^{-1} higher than the recurrent parents. The line with the highest protein was 26 g kg^{-1} higher than the recurrent parent in their study, whereas the lines with the highest protein were 49 to 71 g kg^{-1} higher than their recurrent parent in this study. This comparison indicated that selection between backcross generations was effective in maintaining a distinctly higher level of protein in a population.

The advantage of selection for high protein between backcross generations is particularly important for obtaining a relatively high frequency of lines with sufficiently higher protein than the recurrent parent, from which high-yielding lines can be identified. If it is assumed that a backcross-derived line should have at least 30 g kg^{-1} more protein than the recurrent parent, none of the BC_2F_2-derived lines in the population developed from Pando without selection by Cianzio and Fehr (1982) would meet this criterion. In this study, 18% of the BC_2F_2-derived lines in the Weber population, 43% in the A76-202015 population, and 40% in the A76-304020 population were at least 30 g kg^{-1} higher in protein than their recurrent parent. Of the lines that were not significantly different in yield from the recurrent parent, six in the Weber population, three in the A76-202015 population, and four in the A76-

304020 population were at least 30 g kg^{-1} higher in protein than the recurrent parent.

The average protein content of the A76-304020 population was considerably higher than that of the other two populations (Table 1). This is due to the higher protein content of the recurrent parent of that population, compared with the protein content of the other two recurrent parents. To obtain a higher protein level among BC_2F_2-derived lines, it is advisable to consider the protein content of high- yielding cultivars in selecting a recurrent parent.

REFERENCES

Cianzio, S.R., and W.R. Fehr. 1982. Genetic variability for soybean seed composition in crosses between high- and low-protein parents. J. Agric. Univ. P.R. 66(2):123–129.

Hartwig, E.E., and K. Hinson. 1972. Association between chemical composition of seed and seed yield of soybeans. Crop Sci. 12:829–830.

In Vitro Digestibility of Dry Matter and Cell Wall Constituents of Smooth Bromegrass Forage[1]

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ABSTRACT

The availability of forage cell walls to ruminants is dependent on the concentration and digestibility of cell wall constituents. The objectives of this study were to quantify genotypic variation for in vitro digestibility of individual cell wall constituents and relate this variation to variation in concentration of cell wall constituents of smooth bromegrass (*Bromus inermis* Leyss.). Eighteen clones were grown at Arlington, WI, and sampled in 1982 and 1984. Forage samples were harvested to simulate a normal first cutting. Samples were analyzed for cell wall constituents of dried forage and forage that had been previously digested by rumen microorganisms. In vitro digestibility (IVD) of cell wall constituents was computed from differences obtained between undigested and digested forage samples. Genotypic variation was significant for IVD of neutral detergent and acid detergent fiber (NDF and ADF, respectively), cellulose, and hemicellulose, but not for acid detergent lignin (ADL). In vitro digestibilities of dry matter, NDF, ADF, cellulose, and hemicellulose were mutually correlated, with $r \geq 0.87$. Concentration and IVD of cell wall constituents generally were not correlated, with $-0.52 \leq r \leq 0.22$. Selection for divergent IVD of dry matter (IVDMD) resulted in similar changes to IVD of NDF, ADF, cellulose, and hemicellulose; differences between high and low IVDMD clone groups ranged from 37 to 42 g kg^{-1} for IVD of cell wall constituents. The additional information provided by determining IVD of cell wall constituents makes these selection criteria potentially valuable. Because of their close interrelationships and similar correlated selection responses, IVD of NDF, which is the easiest and least expensive to determine, would satisfactorily represent the other IVD variables in smooth bromegrass.

Additional index words: Bromus inermis Leyss., Cellulose, Hemicellulose, Fiber, Lignin, Repeatability.

DIGESTION of forage plant cell walls by ruminants is influenced by cell wall structure and composition. The development of grass genotypes with improved energy availability for ruminants will likely require changes in the relative concentrations and/or digestibilities of specific cell wall constituents. These changes, in turn, will depend on the identification of meaningful and repeatable selection criteria.

Genetic variation exists in smooth bromegrass (*Bromus inermis* Leyss.) for concentrations of cellulose, hemicellulose, and lignin (Ehlke and Casler, 1985; Reich and Casler, 1985), as determined by the detergent system of forage analysis (Goering and Van Soest, 1970). Lignin is the most important chemical factor limiting in vitro dry matter digestibility (IVDMD) of smooth bromegrass forage (Casler, 1986; Ehlke et al., 1986). Cellulose concentration is also an important factor limiting the genetic potential for increased IVDMD of smooth bromegrass, whereas hemicellulose concentration seems to have little effect on IVDMD (Casler, 1986). Conversely, because hemicellulose and lignin are covalently linked and there is no evidence for cellulose-lignin covalent bonding (Morrison, 1974, 1980; Selvendran, 1983), biochemical evidence suggests that hemicellulose may be a greater limiting factor than cellulose to energy utilization by ruminants.

Changes in cell wall structure and composition with advancing maturity, as they affect energy utilization by ruminants, are becoming increasingly well-understood. Lignin, cellulose, and hemicellulose concentrations, expressed as proportions of total dry matter, increase with advancing maturity. Reductions in the availability of structural carbohydrates to rumen microorganisms with advancing maturity are primarily attributed to increased lignification of the cell wall (Morrison, 1974). However, degradation of delignified hemicellulose by purified rumen hemicellulases increases with advancing maturity, primarily due to a reduction in the proportion of *L*-arabinose side branches (Brice and Morrison, 1982). The genetic relationships of cell wall structure and composition to energy availability are not well understood. Limited results in smooth bromegrass have led to a suggestion of cell wall ideotypes with reduced lignin and cellulose concentration and either reduced or unchanged hemicellulose concentration (Casler, 1986). A thorough investigation of these proposed ideotypes will require knowledge of the relative availability of all cell wall constituents for degradation by rumen microorganisms.

The objectives of this study were to quantify genotypic variation for in vitro digestibility of cellulose,

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