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Nutritional value of ten earless corn hybrids used for silage¤

Valor nutricional de diez híbridos de maíz sin mazorca utilizados para ensilaje

Valor nutritivo de dez híbridos de milho sem espiga utilizados para silagem

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Summary

Background: corn plant silage is characterized by its high nutritional value and high energy content. However, it is important to determine corn silage characteristics that affect its nutritional value, such as the cell wall constituents. **Objective:** the objective of this experiment was to evaluate the chemical-bromatological composition and apparent digestibility of 10 corn hybrids (DK265*bm3*, DK265, HS5, HS6, HTV2, HTV27, Anjou285, Mexxal, Pistachio and Buxxil). **Methods:** the hybrids were planted at INRA (Unité of Génétique Amélioration des Plantes Fourragères, Lusignan, France) in an area of 150 m^2. The experiment was conducted in triplicate. All evaluations were conducted in whole corn plants without ears. **Results:** the DK265*bm3* hybrids presented the best values for enzymatic solubility and cell wall digestibility; it was associated with reduced cell wall KL and esterified p-coumaric acid content compared with the other hybrids. The corn hybrids were evaluated before ensilage using Near Infrared Spectrometry, and a significant difference for chemical composition was found among treatments. **Conclusion:** DK265*bm3* showed superior digestibility of DM, OM, cellulose, NDF and IVDMD compared to the other hybrids.

Key words: *cattle, digestibility, hydroxycinnamic acid, lignin, ruminant.*

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Resumen

Antecedentes: el ensilaje de maíz se caracteriza por su alto contenido nutricional y energético. No obstante, la determinación de las características del ensilaje de maíz que afectan su valor nutritivo, como los constituyentes de la pared de la planta, son de suma importancia. **Objetivo:** el objetivo del presente trabajo fue evaluar la composición química y digestibilidad de 10 híbridos de maíz (DK265*bm3*, DK265, HS5, SA6, HTV2, HTV27, Anjou285, Mexxal, pistacho y Buxxil). **Métodos:** los híbridos fueron plantados en el INRA (Unité of Génétique Amélioration des Plantes Fourragères, Lusignan, France) en 150 metros cuadrados, el experimento se realizó por triplicado. Todas las evaluaciones se llevaron a cabo en plantas enteras sin mazorcas. **Resultados:** el híbrido DK265*bm3* mostró mejores valores de solubilidad y digestibilidad enzimática de la pared celular, y esto se asoció con una reducción de la pared celular y el contenido de ácido p-cumárico esterificado en comparación con otros híbridos. Los híbridos de maíz fueron evaluados antes del ensilaje con Espectrometría de Infrarrojo Cercano, y se encontraron diferencias entre los tratamientos para la composición química. **Conclusión:** el DK265*bm3* mostró mayores valores de digestibilidad de la materia seca, orgánica, celulosa, fibra detergente neutra y digestibilidad *in vitro* de la materia seca, en comparación con los otros híbridos.

Palabras clave: *ácido hidroxicinámico, digestibilidad, ganado, lignina, rumiante.*

Resumo

Antecedentes: a silagem de milho é caracterizada pelo seu alto valor nutricional e energético. No entanto, a determinação das características da silagem de milho que afetam seu valor nutricional, como os constituintes da parede vegetal são de suma importância. **Objetivo:** avaliar a composição químico-bromatológica e a digestibilidade aparente de 10 híbridos de milho (DK265bm3, DK265, HS5, HS6, HTV2, HTV27, Anjou285, Mexxal, Pistachio e Buxxil). **Métodos:** os híbridos foram plantados no INRA (Unité of Génétique Amélioration des Plantes Fourragères, Lusignan, France) em 150 m² de área; o experimento foi conduzido em triplicata. Todas as avaliações foram conduzidas nas plantas inteiras sem espigas. **Resultados:** o híbrido DK265*bm3* apresentou os melhores valores de solubilidade enzimática e digestibilidade da parede celular, e isto foi associado a redução da parede celular e do conteúdo de ácido p-coumárico esterificado comparado com os outros híbridos. Os híbridos de Milho foram avaliados antes da ensilagem usando o Espectometria de infravermelho próximo, e foi verificada a diferença entre os tratamentos para composição química. **Conclusões:** o hibrido de milho DK265*bm3* mostrou valores superiores de digestibilidade da matéria seca, matéria orgânica, celulose, fibra em detergente neutro e digestibilidade *in vitro* da matéria seca, comparado aos outros híbridos.

Palavras chave: *ácido hidroxicinâmico, digestibilidade, gado, lignina, ruminante.*

Introduction

Corn plant silage has high nutritional value and energy content. However, it is important to determine corn silage characteristics that affect its nutritional value, such as cell wall constituents (Oba and Allen, 1999, 2000; Ballard *et al.*, 2001; Barrière *et al.*, 2001; Thomas *et al.*, 2001; Rodrigues *et al.*, 2002; Ferreira *et al.*, 2005). New corn varieties and hybrids with superior nutritional profile for animal feed are currently in the market (Oliveira *et al.*, 2011).

The lignification level of the cell wall constitutes a limiting factor in forage digestibility (Baucher *et al.*, 1998; Boudet, 2000). Factors besides lignin also influence digestibility. The arrangement between lignin and its precursors and the other components of the cell wall can be responsible for many of the limitations observed in forage digestibility (Jung, 1989, 1996; Morrison *et al.*, 1998; Deschamps, 1999; Barrière and Emile, 2000). Some histological studies have demonstrated that lignin-containing material is poorly degraded by rumen microorganisms (Akin, 1998).

Composition and contents of lignin in genetically improved plants have been studied. Mutants of brown midrib corn (*bm*) differ from regular corn in that they present low lignin levels, reduced levels of esterified *p*-coumaric acid and syringin units in the lignin, resulting in improved cell wall digestibility (Jung, 1996). A study by Méchin (2000) investigating different corn lineages showed improved cell wall digestibility for the *bm3* lineage, which was associated with small lignin content.

The *bm3* hybrids present low cell wall lignin levels and are therefore of interest for a comparative study among different genotypes. According to Barrière and Argillier (1998), *bm* plants have little commercial value, although they present higher digestibility when compared with other plants, which is essentially due to its lower forage production. Given the possibility of obtaining lineages of equivalent and/or superior quality to *bm3*, researchers believe it might be possible to obtain hybrids from eared corn lineages with the same quality as *bm3* lineage hybrid, but superior DM production.

The objective of this study was to evaluate the chemical-bromatological characteristics and the *in vitro* digestibility of earless corn genotypes.

Materials and methods

Nine regular eared corn genotypes (DK265, HS5, HS6, HTV2, HTV27, Anjou 285, Mexxal, Pistachio and Boxxil), and a *bm* (DK265 bm3) were planted at INRA (Unité de Gènètique et d'Amélioration des Plantes Fourragères, Lusignan, France) using three repetitions and an average plot size of 150 m^2. A 0.75 m spacing between lines was adopted for a density of 95,000 plants/ha. Seeding was performed in May, 2002, and all of the hybrids were harvested in September, 2002, 152 days after planting.

Ten plants per plot were harvested at random to determine the hybrid's chemical composition. After this assessment, the samples were composed by plot, and all ears were removed. The plants without ears were pre-milled, identified, and dried in a heater at 60 ºC for 72 hours and then milled using a 1 mm diameter sieve for subsequent chemical analyses.

The samples were analyzed to determine the level of crude protein (CP), soluble carbohydrates (according to AOAC, 1984), neutral detergent fiber (NDF), acid detergent fiber (ADF), acid detergent lignin (ADL; according to Goering and van Soest, 1970) and lignin Klason (LK; according to Effland, 1977). Lignification level (ADL and LK) was expressed as a percentage of NDF. Ferulic acid and *p*-coumaric acid contents were determined after treating NDF with soda (according to Morrisson *et al.,* 1993). This method uses a combination of two alkaline treatments in distinct dosages. Esterified *p*-coumaric acid was determined by incubation of NDF samples in soft alkaline hydrolysis (NaOH, 2N) at room temperature for 20 hours with mechanical shaking. Total ferulic acid was determined by incubation of NDF samples in a severe alkaline hydrolysis (NaOH, 4N) at 170 ºC for 2 hours. Subsequently, samples from both the soft and severe hydrolysis received the same treatment. An internal pattern (p-anisic acid) was added before the first centrifugation (380 x g for 10 min). The liquid portion of the samples was adjusted to pH 2 using 3N hydrochloric acid for the samples from the soft hydrolysis and 6N hydrochloric acid for the samples from the severe hydrolysis. All samples were placed in a refrigerator at 4 ºC. Samples were then centrifuged again to eliminate the insoluble polymeric components (hemicelluloses and the polysaccharide-lignin complexes) that formed after acidification. The liquid portion was then removed with the introduction of ethyl acetate, and sodium sulfate (Na_2SO_4) was added to eliminate the water content in the material. The solvent was then vaporized under reduced pressure at 45 ºC. The residue containing hydroxycinnamic acid was then recovered by adding 2.2 mL methanol before high-performance liquid chromatography (HPLC) analysis, as described by Chabbert *et al.* (1994).

Dry matter *in vitro* digestibility (IVDMD) was determined through enzymatic solubility according to Ronsin (1990). This method involves a sequence of digestive enzymatic attacks. For the first attack, pepsin in acid was used and a secondary attack was achieved with the use of a mixture of celluloses and hemicelluloses. The difference between initial sample DM quantity and residual DM quantity after the enzymatic treatment constitutes the digested DM quantity.

Cell wall digestibility (INDSGP) was also estimated by a mathematical equation obtained from the dosage of the different chemical constituents of the forage based on Near Infrared Spectrometry (NIRS). From these dosages, digestibility was determined by subtracting from the IVDMD the quantities of amide, soluble carbohydrates and proteins in the samples (INDSGP = 100*[IVDMD–amide–soluble carbohydrate–proteins]/[100–amide–soluble carbohydrate–proteins]) (Argillier *et al.*, 1996).

Besides INDSGP, cell wall digestibility (DNDF) was also evaluated through an indirect method according to Argellier *et al.* (1998), who presented the hypothesis that the non-NDF fraction is completely digested and that the equation (IVDNDF = 100*(IVDMD–(100–NDF)/NDF) is based on the level of NDF and IVDMD.

It is important to emphasize that these two indirect methods (INDSGP and IVDNDF) for determining DNDF present flaws. For example, not all proteins and amides are digested in the rumen and intestines.

The experimental approach used for these evaluations was to split the plot blocks with ten treatments and three repetitions according to the following mathematical model:

$$Y_{ij} = \mu + B_j + e_{ij}$$

where:

Y_{ijkl} = observation of the genetic variety and the repetition j

μ = general constant

H_i = effect of the hybrid i; i = 1, 2, 3, 4, 5, 6, 7, 8, 9 and 10

B_j = effect of the block k; k = 1, 2 e 3

e_{ij} = random mistakes associated with each observation Y_{ij}.

The data were submitted a Duncan test yielding 5% probability when the test was significant. Variables NDF, ADF, ADL, LK, esterified *p*-coumaric acid, and total ferulic acid were evaluated for each genotype (Dk265, Anjou285, Pistache, Buxxil, Mexxal, HTV2, HTV27, HS5 and HS6) in the regression equation with the objective of better explaining the results concerning IVDMD, INDSGP and, mainly, IVDNDF.

Results

The effect of genotype was significant ($p<0.05$), as determined by the Duncan test, for all the chemical and digestibility characteristics evaluated among the corn hybrids (Table 1). Average values for crude protein (CP) found in this experiment varied from 5.08% to 7.14%.

Soluble carbohydrate content varied from 3.40% to 8.23% among the evaluated hybrids (Table 1). However, it was verified that the highest values of NDF (71.48%) and ADF (42.09%) were observed in reduced soluble carbohydrate content genotypes. Through correlation analysis results (Table 2), it was verified that NDF and ADF contents were negatively correlated ($r = -0.84$ and -0.77) in regard to soluble sugar content.

The DK265*bm3* hybrid presented the smallest NDF ($P<0.05$) and ADF (33.29%) values and the highest values ($P<0.05$) for DM digestibility (58.84%) and DNDF (36.56%) (Table 1). However, this hybrid did not differ ($P>0.05$) from the DK265 hybrid in terms of NDF content (65.82%), and it presented smaller ($P<0.05$) LK (10.96%) and ADL (3.28%) values than the DK265 hybrid (14.16% and 5.89%, respectively). In this study, the LK and ADL contents also presented a negative correlation to IVDMD, INDSGP, and IVDNDF.

As shown in table 1, the differences ($P<0.05$) in hydroxycinnamic acid content were verified among the hybrids. The esterified coumaric acid content varied from 6.46 mg/g to 15.97 mg/g of NDF and the total ferulic acid content varied from 6.61 mg/g to 7.62 mg/g of NDF. Esterified *p*-coumaric acid was negatively associated with IVDMD ($r = -0.66$), INDSGP ($r = -0.62$) and

IVDNDF (r = -0.42) contents. The Mexxal, HTV2, and HTV27 genotypes presented a low total ferulic acid content associated with reduced digestibility (Table 1). Conversely, the DK265*bm3* and DK265 genotypes presented high total ferulic acid content associated with good digestibility.

Table 1. Chemical composition of corn hybrid plants without ears (average values).

Hybrids	Items										
	%CP[1]	%SC[2]	%NDF[3]	%ADF[4]	%LK[5]	%ADL[6]	EsterCu[7]	TotalFe[8]	%IVDMD[9]	%IVDNDF[10]	%INDSGP[11]
Dk265	5.95^{abc}	8.23^{a}	65.82^{de}	35.52^{f}	14.16^{b}	5.89^{de}	13.03^{c}	7.49^{b}	49.09^{b}	22.68^{bc}	40.68^{b}
Anjou285	5.10^{c}	5.03^{bc}	71.41^{b}	39.13^{bc}	14.29^{ab}	5.84^{e}	13.40^{c}	7.43^{b}	44.67^{cd}	22.53^{bc}	38.43^{bcd}
Dk265*bm3*	7.14^{a}	7.26^{ab}	64.88^{e}	33.29^{g}	10.96^{c}	3.28^{f}	6.46^{e}	7.70^{a}	58.84^{a}	36.56^{a}	51.91^{a}
Pistachio	5.40^{bc}	5.65^{abc}	69.91^{bc}	38.17^{cd}	14.67^{ab}	6.36^{cde}	13.29^{c}	7.39^{bc}	46.45^{bc}	23.41^{b}	39.79^{bc}
Buxxil	6.07^{abc}	7.23^{ab}	67.69^{cd}	36.34^{ef}	14.70^{ab}	6.25^{cde}	10.96^{d}	7.29^{c}	46.17^{bc}	20.51^{bc}	37.93^{cde}
Mexxal	5.08^{c}	3.40^{c}	74.55^{a}	42.09^{a}	15.73^{a}	7.20^{a}	14.65^{b}	6.61^{f}	41.58^{e}	21.63^{bc}	36.16^{de}
HTV2	5.89^{abc}	6.01^{abc}	69.09^{bc}	37.93^{cde}	14.22^{ab}	6.58^{bc}	12.98^{c}	7.11^{d}	45.78^{c}	21.49^{bc}	38.43^{bcd}
HTV27	5.92^{abc}	4.20^{c}	71.48^{b}	40.44^{b}	15.31^{ab}	7.06^{ab}	15.97^{a}	6.98^{e}	42.04^{de}	18.92^{c}	35.49^{e}
HS5	6.17^{abc}	5.73^{abc}	68.42^{c}	37.19^{def}	15.06^{ab}	6.27^{cde}	12.84^{c}	7.62^{a}	47.32^{bc}	23.00^{b}	40.20^{bc}
HS6	6.77^{ab}	4.30^{abc}	70.11^{bc}	38.94^{bcd}	15.30^{ab}	6.44^{cd}	13.62^{c}	7.26^{c}	44.70^{cd}	21.12^{bc}	37.81^{cde}
CV[12]	13.37	27.28	2.05	2.50	5.48	5.48	4.26	3.11	3.38	8.40	3.40
SD[13]	0.920	1.993	2.988	2.611	1.430	1.430	2.508	0.368	4.868	5.002	4.619
	*Effects										
Hybrids	0.09	0.025	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Repetition	SN	SN	SN	SN	SN	SN	SN	SN	SN	SN	SN

Averages with different letters in the same column differ through the Duncan test ($p<0.05$). [1]Crude protein; [2]Soluble carbohydrates; [3]Neutral detergent fiber; [4]Acid detergent fiber; [5]Lignin klason; [6]Acid detergent lignin; [7]Esterified *p*-coumaric acid; [8]Total ferulic acid; [9]Dry matter *in vitro* digestibility; [10]Neutral detergent fiber *in vitro* digestibility; [11]Digestibility by mathematical equation (INDCGP = 100*(IVDMD-amid-soluble carbohydrate-proteins)/(100-amid-soluble carbohydrate-proteins); [12]Coefficient of variation; [13]Standard deviation.

The total ferulic acid content was negatively correlated with NDF content (r = -0.68), ADF (r = -0.72), ADL (r = -0.80), LK (r = -0.63), and positively correlated with DM digestibility (r = 0.70) (Table 2).

It was confirmed that the best multiple regression equation for IVDMD was achieved using the variables NDF, ADF, ADL, LK, esterified *p*-coumaric acid, and total ferulic acid (IVDMD = 30.36054 + 0.85393*NDF – 1.51373*ADF + 0.73632*ADL – 0.62914*LK – 0.00485*Cester + 0.34744*Ftotal; Residual Standard Mistake (RSM) = 1.16; r^2 = 85%). However, the multiple regression equation using two variables that better explained the results concerning IVDMD was IVDMD = 89.27539-0.45282-*NDF-2.74102*ADL; RSM = 1.28; r^2 = 79%.

According to the multiple regression equation analysis for INDSGP the best equation selected used NDF, ADF, ADL, LK, esterified *p*-coumaric acid, and total ferulic acid (INDSGP = 30.36054 + 0.85393*NDF-1.51373*ADF + 0.73632*ADL – 0.62914*LK –0.00485*Cester + 1.34744*Ftotal; RSM = 1.40; r^2 = 66%). However, the multiple regression equation using two variables that better explained the results concerning INDSGP was INDSGP = 54.54912 – 2.55033*ADL – 0.45705*LK; RSM = 1.50; r^2 = 54%.

As with IVDMD and INDSGP, the values concerning IVDNDF were better explained using multiple regression analysis with the same variables (IVDNDF = -14.90636 + 1.57937*NDF – 1.96071*ADF + 0.41507*ADL – 0.81950*LK + 0.12213*Cester + 0.91468*Ftotal; RSM = 1.68; R^2 = 55%). However, the best equation to explain IVDNDF used two variables, ADF and NDF, as shown by the following equation: DNDF = -1.23464 – 1.98305*ADF + 1.41937NDF; RSM = 1.61; R^2 = 51%.

Table 2. Genotypic correlation coefficients for chemical composition and digestibility of nine corn hybrids*.

	CP[1]	SC[2]	NDF[3]	ADF[4]	ADL[5]	LK[6]	IVDMD[7]	IVDNDF[8]	INDSGP[9]	EsterCu[10]	TotalFe[11]
CP	1	-0.11	-0.26	-0.28	-0.22	0.17	0.18	-0.04	0.05	-0.07	0.08
		0.5721	0.1745	0.1538	0.2650	0.3909	0.3625	0.8170	0.7990	0.6973	0.6591
SC		1	-0.84	-0.77	-0.61	-0.80	0.72	0.11	0.42	-0.45	0.58
			0.0001	0.0001	0.0007	0.0001	0.0001	0.5564	0.0265	0.0182	0.0014
NDF			1	0.93	0.73	0.75	-0.82	-0.07	-0.55	0.53	-0.68
				0.0001	0.0001	0.0001	0.0001	0.7170	0.0029	0.0042	0.0001
ADF				1	0.86	0.78	-0.91	-0.32	-0.72	0.70	-0.72
					0.0001	0.0001	0.0001	0.0945	0.0001	0.0001	0.0001
ADL					1	0.77	-0.83	-0.45	-0.72	0.63	-0.80
						0.0001	0.0001	0.0160	0.0001	0.0003	0.0001
LK						1	-0.77	-0.32	-0.63	0.57	-0.63
							0.0001	0.0949	0.0004	0.0018	0.0003
IVDMD							1	0.62	0.90	-0.66	0.70
								0.0005	0.0001	0.0002	0.0001
IVDNDF								1	0.83	-0.42	0.28
									0.0001	0.0284	0.1490
INDSGP									1	-0.62	0.59
										0.0005	0.0011
EsterCu										1	-0.42
											0.0258
TotalFe											1

[1]Crude protein; [2]Soluble carbohydrates; [3]Neutral detergent fiber; [4]Acid detergent fiber; [5]Acid detergent lignin; [6] Lignin klason; [7]*In vitro* digestibility dry matter; [8]*In vitro* digestibility neutral detergent fiber; [9]Digestibility by mathematical equation (INDCGP = 100*[IVDMD-amid-soluble carbohydrate-proteins]/[100-amid-soluble carbohydrate-proteins]); [10]Esterified *p*-coumaric acid; [11]Total ferulic acid.
* Dk265, Anjou285, Pistachio, Buxxil, Mexxal, HTV2, HTV27, HS5 and HS6.

Discussion

The *bm3* hybrid presented the highest CP content, which is similar to some eared hybrids. These results were lower than the values found by Oba and Allen (1999) who studied two corn hybrids (9.7% and 9.5%), and by Rodrigues *et al.* (2002) who evaluated the Agroceres 5011 hybrid (9.43%). However, the values found by those authors were obtained from entire plants in silage form, whereas earless plants were used in the present work. Zeoula *et al.* (2003), working with the stems and sheaths of five corn hybrids at different phases of maturity (25% to 40% MS) obtained CP values between 3.58% to 3.86%. However, Ferreira *et al.* (2005) reported no significant differences among five earless corn genotypes, with values ranging from 7.2% to 9.4%. Corn plants are characterized by their high-energy content (mainly amide) and low CP content, which can be influenced by agronomic factors (insufficient and/or poorly implemented fertilization) and genetic factors. Most recent hybrids have low CP content as showed by Caetano *et al.* (2011), who tested C805 hybrid and found low CP values (2.91%).

The negative correlation between NDF and ADF contents in regard to soluble sugar content can be explained by the higher metabolic activity in some plants, leading to decreased soluble nitrate, protein, and carbohydrates, and an increase in cell wall components (Van Soest, 1994). The genetic characteristics of each plant must also be considered, as they might lead to an increased or decreased cell wall proportion. Similarly, the results found between DK265*bm3* and DK265 hybrids led us to assume that even if the NDF content had a negative correlation with INDSGP content (Table 2), this did not explain the differences in digestibility values; therefore, other factors were involved, such as lignin content and possible connections to low molecular weight molecules.

The obtained values indicate that LK and ADL contents could explain the variation in digestibility values: 59% and 68% for IVDMD; 39% and 51% for INDSGP; and 10% and 17% for DNDF content, respectively. Mechin (2000) observed that 28% of DNDF variation between corn lineages could be explained by LK content and 25% of the variation could be explained by NDF content. This negative effect of esterified coumaric acid on digestibility could be explained by the positive correlation with LK (r = 0.57) and ADL (r = 0.63). According to Argillier *at al*. (1996), *p*-coumaric acid is predominantly related to lignin, and its influence over polysaccharide degradation is probably related to direct and indirect negative effects of lignin. It can be inferred from these results that total ferulic acid content alone is insufficient to explain the results in terms of digestibility; therefore, other factors are involved, such as the proportion of ester types and ether bonds, which were not evaluated in the present work. This evidence has been confirmed by several authors (Jung, 1989; Marvin *et al.*, 1995; Morrison *et al.*, 1998; Deschamps, 1999; Barrière and Emile, 2000; Ferreira *et al.*, 2005) who suggested that digestibility variations can be better explained by variations in lignin and total hydroxycinnamic acid content, specifically by their proportions of ester and ether bonds.

Argiller *et al.* (1996) studied the existing variations in six corn hybrids and demonstrated 8% difference in IVDNDF content between MBS847 x Co125 (36.4%) and F2 x F113 (44.4%) hybrids, which presented similar lignin contents (21.3% and 21.4%, respectively). The authors also confirmed differences in characteristics of hydroxycinnamic acid and monomeric lignin composition, but the MBS847 x Co125 hybrid presented higher *p*-coumaric acid concentration and smaller proportion of ether and ester ferulic acids compared with the F2 x F113 hybrid. Mechin *et al.* (2000) did not observe differences among lignin contents in the W117, F2, and F251 corn hybrid lineages. However, the observed differences between these lineages were related to the digestibility of cell wall contents ranging from 10% to 12%. The authors maintained the theory that differences in digestibility can be better explained by the kind of bonds between lignin and hemicelluloses.

The correlation analyses (Table 2) demonstrated that NDF explained 30% of the INDSGP results, whereas the total ferulic acid content explained only a fraction of the digestibility values. Therefore, it was observed that NDF and total ferulic acid, when individually evaluated, did not present the same confidence level as when evaluated as a whole to explain INDSGP values of the studied hybrids. Mechin (2000), working with different corn lineages, observed that only 28% of DNDF results could be explained by LK content, and 58% of the results could be better explained through the grouping of esterified *p*-coumaric acid and the relationship between syringyl and guaiacyl (S:G) units. In the present work, however, chemical analyses were not performed in terms of the monomeric composition of lignin or in terms of the types of bonds between ferulic acid and hemicelluloses, which can present negative effects on IVDNDF (Argillier *et al.*, 1996; Boudet, 2000).

The mutant genotype DK265*bm3* presented the best results concerning the chemical evaluations studied, and, due to these characteristics, it is necessary to explain the existing variations between regular/normal hybrids. The HTV27 genotype presented low digestibility of the NDF fraction, which was associated with small differences, compared with the other studied genotypes, in the contents of NDF, ADF, LK, and ADL. It is supposed that other factors can interfere with digestibility, such as the types of ester and ether connections, and possible differences in the monomeric composition of lignin.

The multiple equations presented in this study are a good resource to explain IVDMD results for assessing cell wall using NIRS (INDSGP) and, mainly, for evaluating NDF *in vitro* digestibility. DK265*bm3* showed superior values compared with the other hybrids for digestibility of DM, OM, cellulose, NDF, and IVDMD.

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