Vol. 52, Special Number: pp. 17-28, November 2009 ISSN 1516-8913 Printed in Brazil

# BRAZILIAN ARCHIVES OF BIOLOGY AND TECHNOLOGY

#### AN INTERNATIONAL JOURNAL

# Characterization of Corn Landraces Planted Grown in the Campos Gerais Region (Paraná, Brazil) for Industrial Utilization

Alessandra Teixeira Barbosa Pinto<sup>1</sup>, Joyce Pereira<sup>2</sup>, Tatiana Roselena de Oliveira<sup>1</sup>, Rosilene Aparecida Prestes<sup>1</sup>, Rodrigo Rodrigues Mattielo<sup>3</sup> and Ivo Mottin Demiate<sup>1\*</sup>

<sup>1</sup>Universidade Estadual de Ponta Grossa; Avenida Carlos Cavalcanti, 4748; 84030-900; Ponta Grossa – PR – Brasil. <sup>2</sup> Curso de Engenharia de Alimentos, <sup>3</sup> Pós-Graduação em Agronomia.

#### **ABSTRACT**

This work has the objective of characterizing twenty corn landraces grown in the Campos Gerais region (Paraná State) in relation to its chemical composition (moisture, ash, protein, ether extract, dietary fiber and starch) and physical properties (weight of 1000 grains, real density, flotation index, granulometry and color). In addition, also the lab scale processing of the kernels from the varieties was carried out for producing starch; starch purity was evaluated by measuring its protein contamination. Amylose contents and viscoamylograph profile were also evaluated. The results showed that the evaluated landraces have differences in chemical composition as well as in pericarp/endosperm/germ proportions and consequently it should have different industrial applications and interest for plant breeding.

Key words: maize landraces, chemical composition, starch, physical properties, rapid viscoanalyzer

## INTRODUCTION

Corn (*Zea mays*, L.) is one of the main cultivated cereals all around the world. Its economic importance is due to many different ways of utilization from animal feeding to high technology industry (Cruz *et al.*, 2006). Although corn kernels have not an expressive importance in direct human diet, several corn products are relevant for people of low income regions.

The main parts of corn kernel are the endosperm, pericarp and the germen. Each fraction presents distinct chemical composition and the quality is dependent from the genetic material as well as from the environment conditions.

The quality of corn for food and feed is different and genetic seems to be the best way of achieving the desired characteristics. The fast evolution of biotechnology made it possible to produce many kinds of hybrids with improved yield and other interesting characteristics (Trindade, 2006). The new biotechnology approach has many advantages in comparison to traditional technologies being able to introduce characteristics that reduce risks in agriculture. As many plant breeding programs are in development, each year several new genetic materials are marketed including dozens of corn hybrids. In Brazil there are many kinds of climatic, soil and environmental conditions due to its continental extension and important differences

\_

Author for correspondence: demiate@yahoo.com

occur among the different geographical regions, including technical, economical, cultural and social aspects (Schmildt; Krause & Cruz, 2006). In this way, it is essential that plant breeders be very aware to select the correct materials for the distinct national conditions.

In this corn breeding process the landraces are of great interest (Miranda *et al.*, 2007) for identifying the needs of the farmers and exploiting their genetic variability. The landraces have potential in contributing with desired characteristics of cultivated plants including resistance to diseases and agricultural pests, improving crops and food safety (Fowler & Hodgkin, 2004, Birol; Villalba & Smale, 2007).

Several evidences show the importance of rescuing landraces for knowing their variability in terms of chemical composition of their edible parts and other economical interesting characteristics for industrial utilization as well as for breeding programs.

## **MATERIALS AND METHODS**

#### Material

Representative samples of 20 corn landraces (Table 1) were grown in randomized blocks with four repetitions. The experiment was conducted at the School Farm of Universidade Estadual de Ponta Grossa, in Ponta Grossa, Paraná, The sowing was made in November/2006 and the harvest in March/2007. The sowing fertilization was made with 300 kg ha<sup>-1</sup> of the 5-25-25 (NPK) formulation. A complimentary fertilization was made 45 days after germination with urea (100 kg ha<sup>-1</sup> of nitrogen). The corn kernels were milled in a rotor mill (Tecnal model TE-633, Piracicaba, SP, Brazil) adjusted for fine milling and the granulometry of the milled product was evaluated with a Bertel (Caieiras, SP, Brazil) system of vibrating sieves.

**Table 1** – Identification, common denomination, local of origin, city/state where the landraces were collected or bought.

Number	Races	Local of origin	City/State
1	Nutricional nº 61	Colônia Cachoeira	Rio Azul - PR
2	Milho Grande nº 52	Roça Velha	Itaiacoca – P. Grossa - PR
3	Milho Palha Roxa nº 72	Colônia Cachoeira	Rio Azul - PR
4	Milho Paiol nº 49	Colônia Barra Bonita	Prudentópolis - PR
5	Milho Branco nº 57	Rio Azul de Cima	Rio Azul - PR
6	Asteca nº60	Colônia Cachoeira	Rio Azul - PR
7	Caiano nº 63	Rio Vinagre	Rio Azul - PR
8	Roxo Índio I nº 60	Pelotas	Pelotas - RS
9	Milho Branco para Palha nº 69	Colônia Cachoeira	Rio Azul - PR
10	Milho Pérola nº 65	Salto Braço do Potingá	Rio Azul - PR
11	Carioca nº 68	Pinhalzinho	Rio Azul - PR
12	Milho Astequinha Sabugo Fino nº 70	Marumbi dos Ribeiros	Rio Azul - PR
13	Milho Vintenv nº 56	Porto Soares	Rio Azul - PR
14	Milho Palha Roxa n° 64	Invernada	Rio Azul - PR
15	Milho Encantilado nº 59	Rio Vinagre	Rio Azul - PR
16	Milho Branco nº 62	Lajeado	Rio Azul - PR
17	Milho Amarelo Antigo nº 71	Porto Soares	Rio Azul - PR
18	Crioulo Oito Carreiras nº 41	FEPAGRO	RS
19	Crioulo Pururuca n º 38	FEPAGRO	RS
20	Caiano Rajado nº 16	Pelotas	Pelotas - RS

# **Chemical composition**

The chemical characterization of corn kernels included the following analyses: moisture content (AOAC, 2000, 925.10), protein (AOAC, 2000, 920.87), ash (AOAC, 2000, 923.03), lipids (IAL, 1985), starch (Demiate et al., 2001) and dietary fiber (AOAC, 2000, 985.29).

# Physical properties

The weight of 1000 kernels of each sample was made as described by Mauricio *et al.*, 2004).

The real density was evaluated as described by Mizuma, Kiyokawa & Wakai (2008), by using a pycnometer of known mass that was completely filled with xylene and weighed. A pre established

corn kernel weight was put into the pycnometer and xylene was used for completely filling the empty spaces among the kernels. The total weight was registered and used for calculations.

The flotation index was evaluated (Lojano-Alejo et al., 2007) by using a sodium nitrate solution (41 g in 100 mL of water) with a known density of 1.25 g dm<sup>-3</sup> at 23 °C. The kernels were put in this solution that was stirred. After resting for one minute, the floating kernels were counted. The hardness classification was based on a scale proposed by Salinas; Bustos & Gómez (1992), with floating index (FI) between 0 and 12 % for very hard, 13 to 37% for hard, 38 to 62% for intermediary hard, 63 to 87 % for soft and higher than 87 % for very soft kernels.

#### Starch extraction

The starch extraction was made following the method of Ji, Seetharaman & White (2004), with some adaptations. For extracting starch, 100 kernels were used, being 25 of each block. There was removal of impurities and damaged kernels. Steeping started when the kernels were put into 40 mL of a 1 % sodium metabisulphite solution at 45°C for 72 h, followed by manual pericarp and germen removal. The endosperm fraction was put into 30 mL of distilled water and finely ground. This suspension was centrifuged for 10 min at 3,200 rpm for accelerating starch separation. The pellet was recovered and mixed with excess of a 0.1 % NaOH solution, in graduated cylinders for making precipitation fast. These cylinders were cooled to around 4 °C for 24 h. The sediment was passed through a 325 mesh sieve, washing with distilled water. For neutralizing the sediment starch, addition of a 0.1 % HCl solution was made followed by distilled water washings until pH reached 6.0 - 7.0. The purified starch was then recovered by using a vacuum filtration system. The starch was dried for 24 h in a circulating oven at 45 °C until constant weight.

#### **Starch characterization**

All starch samples extracted from the corn kernels were stored in a dessicator for moisture stabilization. The analyses were made with two repetitions.

#### **Properties of the starch pastes**

The starch pastes of the 20 different starch samples were made by employing a Rapid Viscoanalyzer (RVA 4, Newport Scientific,

Warriewood, Australia) using the STD1 profile of the software Thermocline. The data shown in the results are average of two repetitions. Starch slurries at 8.0 % (14.0 % moisture basis) were analyzed, considering a final slurry weight of 28 g.

#### Statistical analysis

The results were analyzed by the ANOVA followed by the Tukey test at 95 % confidence level. These analyses were made using the ASSISTAT version 7.1 beta software (Silva, 2004). A correlation matrix was also built up in order to better discuss the results, employing the Statistica version 5.0 software, considering 95 % confidence level. The principal component analysis (PCA) was employed in order to extract information from the results, using the Pirouette version 4.11 software (Infometrix). The auto scaling processing of the data was used for producing better results.

#### RESULTS AND DISCUSSION

The raw results are presented in the Table 2. The results showed that for moisture and ash contents, there were no significant differences at 95 % of confidence. The average values of moisture ranged from 10.29 % (Milho Grande n°52) to 11.45 % (Milho Encantilado nº 59). Other published papers (Luchin; Barcaccia and Parrini, 2003; Gonçalves et al., 2003) revealed results of ash of 1.56 % and 1.27 % in corn landraces and hybrids, respectively. The ANOVA showed significant difference for the protein levels among the varieties. The variety Crioulo Oito Carreiras nº 41 had the higher protein level (12.41%), whereas the variety Caiano Rajado nº16 had the lower (10.26 %). In the literature some authors reported that the protein contents for corn landraces were between 10.47 to 12.17 % (Câmara, 2005), and in hybrids from 7.77 to 13.84 % (Jiang et al., 2007). The average value found for dietary fiber was of 13.0 % and for starch this value was of 60.54 %. Other reports stated that starch contents in corn landraces were from 67.2 % (Câmara, 2005) to more than 70.0 % (Seetharaman et al., 2001).

# **Starch extraction**

Table 3 shows the proportions of the morphological parts of corn kernel. The pericarp fraction varied from 3.73 to 7.38 %, whereas the germ fraction was between 7.05 to 12.92 %. In

other reports where commercial corn hybrids were evaluated the germ percentages were of 5.75 % (Dowd, 2003), 6.20 % (Fox *et al.*, 1992) and 6.97 % (Lopes-Filho, 1997). The *Milho Paiol* n°49 had the higher endosperm percentage (86.97 %) and

high starch extraction yield (39.87 %). On the other hand, the Asteca n°60 had the lower value (81.50 %) and one of the lowest starch extraction yield (20.22 %).

**Table 2** – Physicochemical composition and physical properties of corn landrace kernels.

RACES	MOS (%)	ASH * (%)	PRO* (%)	LIP * (%)	STA * (%)	DF * (%)	TKW * (g)	DR (g.cm <sup>3</sup> )	FI (%)	COA (%)	INT (%)	FIN (%)
Nutricional nº 61	10.82 a	1.42 a	11.04 ab	4.68 ab	66.51 a	13.58 abcd	307.60 hi	1.265 a	31.0 bcd	25.55 cde	25.61 abcd	0.93 a
Milho Grande nº 52	10.29 a	1.47 a	11.58 ab	4.60 abc	54.86 cde	13.79 abcd	398.53 abc	1.262 a	39.0 abcd	23.67 de	27.97 ab	0.68 a
Milho Palha Roxa nº72	11.17 a	1.51 a	10.32 b	3.70 cd	67.10 a	13.36 abcd	340.44 defghi	1.257 a	27.0 d	22.54 e	28.89 a	0.68 a
Milho Paiol n°49	11.08 a	1.55 a	10.69 b	4.32 abcd	65.22 ab	17.21 ab	320.36 fghi	1.248 a	52.5 abc	27.48 abcde	24.30 abcde	0.49 a
Milho Branco nº57	10.89 a	1.56 a	10.88 ab	3.73 cd	55.74 bcde	18.08 a	362.51 bcdefgh	1.225 a	60.5 a	24.51 cde	27.36 abc	0.36 a
Asteca nº 60	10.63 a	1.48 a	10.99 ab	4.98 a	59.26 abcde	13.85 abcd	317.66 ghi	1.307 a	31.5 bcd	28.44 abcd	23.76 abcde	0.68 a
Caiano nº 63	11.17 a	1.48 a	10.54 b	4.33 abcd	63.88 abcd	15.06 abc	340.52 defghi	1.287 a	28.5 cd	28.52 abcd	22.85 bcde	0.45 a
Roxo Índio I nº 60	11.03 a	1.50 a	11.04 ab	4.32 abcd	63.24 abcd	12.60 bcd	380.69 bcde	1.280 a	38.5 abcd	26.96 bcde	24.78 abcde	0.40 a
Milho Branco para Palha nº 69	10.99 a	1.43 a	11.64 ab	4.15 abcd	62.14 abcde	10.88 cd	342.40 cdefghi	1.272 a	30.0 bcd	31.67 ab	19.90 de	0.71 a
Milho Pérola nº 65	11.19 a	1.46 a	10.86 ab	4.56 abcd	58.77 abcde	14.42 abcd	303.44 i	1.320 a	28.0 d	32.49 a	19.03 e	0.49 a
Carioca nº 68	11.18 a	1.52 a	10.63 b	4.51 abcd	59.17 abcde	12.97 bcd	377.03 bcdef	1.268 a	27.5 d	28.67 abcd	23.38 abcde	0.31 a
Milho Astequinha Sabugo Fino nº 70	11.06 a	1.55 a	11.62 ab	4.58 abcd	54.00 de	16.97 ab	325.99 efghi	1.277 a	25.0 d	29.57 abc	22.09 bcde	0.35 a
Milho Vinten nº 56	10.99 a	1.39 a	11.45 ab	4.41 abcd	59.89 abcde	15.27 abc	308.91 hi	1.286 a	29.5 cd	28.20 abcd	23.61 abcde	0.46 a
Milho Palha Roxa nº 64	11.35 a	1.53 a	11.23 ab	4.58 abcd	62.36 abcde	15.25 abc	410.49 ab	1.263 a	37.5 abcd	28.64 abcd	23.49 abcde	0.39 a
Milho Encantilado nº 59	11.45 a	1.54 a	10.92 ab	4.02 bcd	59.21 abcde	13.64 abcd	386.91 bcd	1.216 a	54.0 ab	30.00 abc	21.67 cde	0.41 a
Milho Branco nº 62	10.67 a	1.48 a	10.67 b	3.63 d	64.88 abc	14.08 abcd	371.32 bcdefg	1.248 a	36.0 bcd	29.01 abcd	22.99 abcde	0.35 a
Milho Amarelo Antigo nº 71	10.82 a	1.56 a	11.15 ab	4.19 abcd	58.68 abcde	13.87 abcd	315.88 ghi	1.262 a	35.5 bcd	26.49 bcde	25.48 abcd	0.39 a
Crioulo Oito Carreiras nº 41	10.73 a	1.52 a	12.41 a	4.46 abcd	53.06 e	9.89 d	450.28 a	1.299 a	26.5 d	27.34 abcde	24.29 abcde	0.72 a
Crioulo Pururuca nº 38	11.14 a	1.47 a	10.87 ab	4.02 bcd	61.16 abcde	11.70 cd	355.48 bcdefghi	1.237 a	45.5 abcd	27.91 abcde	23.91 abcde	0.23 a
Caiano Rajado nº 16	11.38 a	1.46 a	10.26 b	3.99 bcd	63.05 abcde	11.49 cd	347.49 cdefghi	1.253 a	31.0 bcd	28.34 abcd	23.60 abcde	0.22 a
Average	11,00	1.49	11.04	4.29	60.54	13.9	353.16	1.267	35.73	27.67	24.18	0.48
Minimum	10.29	1.39	10.26	3.63	53.06	9.89	303.44	1.216	25.01	22.54	19.03	0.22
Maximun	11.45	1.56	12.41	4.98	67.1	18.08	450.28	1.32	60.5	32.49	28.89	0.93
Standard Deviation	0.28	0.05	0.51	0.36	4.02	2.08	450.28	0.026	10.1	2.46	2.36	0.93

MOS: Moisture, ASH: Ash, PRO: Protein, LIP: Lipids, DF: Dietary Fiber, STA: Starch, TKW: 1.000 kernel weight, RD: Real density, FI: Flotation Index, COA: Coarse particles (850 µm and 600 µm), INT: Intermediate particles (250 µm and 180 µm), FIN: Fine particles (125 µm and bottom).

Values with similar letters in the same column do not differ significantly (p<0,05).

**Table 3** – Fractions of the kernels and starch extraction yields.

Races	Pericarp (%)	Germ (%)	Endosperm (%)	Yield (%)	
Nutricional nº 61	5.27	10.16	84.57	31.15	
Milho Grande nº 52	6.18	11.10	82.72	33.02	
Milho Palha Roxa nº 72	6.74	7.91	85.35	24.43	
Milho Paiol nº49	5.98	7.05	86.97	39.87	
Milho Branco nº 57	5.62	10.36	84.02	27.27	
Asteca nº 60	5.57	12.92	81.50	20.22	
Caiano nº 63	5.46	9.99	84.55	35.00	
Roxo Índio I nº 60	6.35	11.74	81.91	19.05	
Milho Branco para Palha nº 69	6.53	9.39	84.07	29.75	
Milho Pérola nº 65	5.39	8.96	85.65	33.01	
Carioca nº 68	6.76	11.17	82.07	27.96	
Milho Astequinha Sabugo Fino nº 70	6.73	11.69	81.59	32.44	
Milho Vinten n° 56	5.39	10.97	83.64	25.61	
Milho Palha Roxa nº 64	5.67	10.16	84.17	35.22	
Milho Encantilado nº 59	5.33	9.93	84.74	23.56	
Milho Branco nº 62	6.20	9.38	84.43	30.21	
Milho Amarelo Antigo nº 71	3.73	9.69	86.57	31.37	
Crioulo Oito Carreiras nº 41	4.70	10.34	84.96	28.43	
Crioulo Pururuca nº 38	5.73	10.22	84.05	35.95	
Caiano Rajado nº 16	7.38	10.26	82.37	24.78	
Average	5.84	10.17	83.99	29.42	
Minimum	3.73	7.05	81.50	19.05	
Maximum	7.38	12.92	86.97	39.87	
Standard Deviation	0.82	1.31	1.57	5.40	

<sup>\*</sup> Dry weight bases

#### **Starch characterization**

In the Table 4 are shown the results of the analyses of extracted starch. The average protein content for the extracted starches was of 1.0 %, with a minimum of 0.52 % (*Milho Palha Roxa* n° 72) and a maximum of 1.42 % (*Milho Amarelo Antigo* n° 71). Ji; Seetharaman, White (2004) found protein values for corn starches extracted in laboratory in the range of 2.82 to 3.65 % and it should explain the higher extraction yields related in their report.

The apparent amylose contents of corn landrace starches did not differ at 95 % confidence level and the average value was of 17.66 %, in a range from 13.14 % (*Milho Paiol* n° 49) to 21.51 % (*Milho Astequinha Sabugo Fino* n° 70). In the case of total amylose, there was statistical difference among the starches. The average amylose content was of 25.59 %, in a range of 19.10 % (*Roxo Índio* I n° 60) to 33.86 % (*Milho Palha Roxa* n° 72).

**Table 4** – Hot paste viscosity, Peak viscosity, gelatinization temperature, final viscosity, breakdown and setback.

Races	HPV (cP)	PV (cP)	P onset(°C)	FV (cP)	Breakdown (cP)	Setback (cP)
Nutricional nº 61	1248 a	1818 ab	78.43 bcde	1897 abc	794 bcdef	873 bcd
Milho Grande n° 52	1036 bcde	1795 ab	77.63 efg	1836 abc	808bcdef	849 bcd
Milho Palha Roxa n°72	991 bcde	1804 ab	77.55 fg	1793 abc	867 abcd	857 bcd
Milho Paiol n°49	1008 bcde	1550 e	78.45 bcde	1578 bc	715efgh	743 cd
Milho Branco nº 57	904 de	1398 fg	78.50 bcd	1593 bc	568 i	763 cd
Asteca nº 60	1108 abc	1808 ab	80.85 a	1940 a	759 defg	891abc
Caiano nº 63	968 bcde	1726 bc	78.40 bcdef	1714 abc	813 bcdef	801 bcd
Roxo Índio I nº 60	957 cde	1808 ab	78.43 bcde	1985 a	900 abc	1078 a
Milho Branco para Palha nº 69	939 cde	1317 g	81.33 a	1581 bc	425 j	689 d
Milho Pérola nº 65	875 e	1785 ab	78.53 bc	1802 abc	964 a	981 ab
Carioca nº 68	1005 bcde	1750 ь	78.83 ь	1799 abc	800 bcdef	849 bcd
Milho Astequinha Sabugo Fino nº 70	1044 bcde	1764 ь	78.43 bcde	1902 abc	764 cdefg	903 abc
Milho Vinten nº 56	1066 bcd	1790 ab	78.45 bcde	1907 ab	782 bcdef	899 abc
Milho Palha Roxa nº 64	982 bcde	1700 bcd	78.48 bcde	1719 abc	831 abcde	800 bcd
Milho Encantilado nº 59	1015 bcde	1571 de	77.65 defg	1753 abc	606 hi	788 cd
Milho Branco nº 62	970 bcde	1495  ef	78.08 bcdef	1754 abc	630 ghi	840 bcd
Milho Amarelo Antigo nº 71	1061bcd	1908 a	75.95 i	1755 abc	909 ab	756 cd
Crioulo Oito Carreiras nº 41	997 bcde	1783 ab	$76.83~\mathrm{gh}$	1844 abc	841 abcde	902 abc
Crioulo Pururuca nº 38	920 de	1705 bcd	$76.05 \; \mathrm{hi}$	1572 c	834 abcde	701 d
Caiano Rajado nº 16	1129 ab	1608 cde	77.75 cdef	1700 abc	691 fghi	783 cd
Average	1011	1693.98	78.22	1771.13	764.93	837.08
Minimun	875	1317	75.95	1571.5	425	689
Maximun	1247.5	1908	81.33	1985.5	963.5	1077.5
Standard Deviation	85.24	155.04	1.27	124.55	128.99	93.29

 $P_{\text{onset}}\text{: gelatinization temperature; HPV: hot paste viscosity, PV: peak viscosity, FV: final viscosity}$ 

Values are average of duplicate runs

Values with similar letters in the same column do not differ significantly (p<0,05).

The viscoamylographic pattern is presented in Table 4 with the main points of the analysis (viscosity peak, hot paste viscosity, final viscosity, breakdown, setback and gelatinization temperature). The profile is typical for most of the starch samples, the final viscosity is high due to retrogradation and the paste is relatively stable to stirred cooking, when comparing with cassava

starch (Schoch & Maywald, 1968; Cereda *et al.*, 2001), for example.

For some samples there were lower values of final viscosities when compared to viscosity peaks. The samples that had this viscoamylographic pattern were *Milho Palha Roxa* n° 72, *Caiano* n° 63, *Milho Amarelo Antigo* n° 71 and *Crioulo Pururuca* n° 38. For the *Amarelo Antigo* n° 71 and *Crioulo Pururuca* n° 38 varieties, this pattern should be

associated with the low levels of total amylose (23.85 and 21.91 %, respectively) that reduce final viscosity (Salgo & Juhász, 2008; Blazek, 2008). The *Milho Palha Roxa* n° 72 and *Caiano* n° 63 varieties had high lipid complexed amylose levels (13.18 and 10.14 %, respectively), what have direct influence on viscosity. As written by Nelles *et al.*, (2000), a small amount of lipids or phospholipids in starch affects the paste properties. Starch has free fatty acids and phospholipids in amounts directly related with amylose, distributed asymmetrically in the granules.

# **Correlation analysis**

In the Table 5 the correlations among results are shown. The protein content had a negative correlation with starch content (r= -0.66, at 95 % confidence level), in the same way as reported by Chander et al. (2008). Fox et al. (1992) also found this correlation for corn hybrids (r = -0.63; p<0.01). The hardness, as estimated by the flotation index, did not have correlation with the protein content, although some authors found positive correlation (Narváez-González et al., 2006), attributed to strong association between starch granules and protein with absence of empty intergranular spaces in the endosperm (Gibbon; Wang & Larkins, 2003, Gibbon & Larkins, 2005). Pereira et al. (2008) reported that corn kernels with hard or dent endosperms have abundance of organized protein bodies what explain the compact of these endosperms.

A positive correlation was observed between lipid content and kernel real density (r = 0.67, at 95 % of confidence). Although not expected this could be explained by a study for wheat of Scott (1951) cited by Posner (1985). The author reported that wheat germen had a real density of 1.34 g cm<sup>-3</sup>, very close to that of the whole grain, of 1.36 g cm<sup>-3</sup>. In the germ, beyond lipids, there are important levels of protein and sugars, which explain the data reported by Posner (1985). Furthermore the studied corn landraces presented high percentage of germ from 7.0 to 12.0 %. As expected the germ percentage and lipid content were positively correlated (r= 0.51, 95 % of confidence).

An expected negative correlation was found between flotation index and real density (r= -0.74, at 95 % of confidence); this fact was already reported by Salinas; Bustos & Gómez (1992), that found r= -0.94, at 95 % of confidence.

The granulometric profile evaluated by the percentage of coarse, intermediary and fine

particles produced by corn kernels milling, revealed a negative correlation between coarse and intermediary particles (r= -0.93, at 95 % of confidence). The hardness classification of the grains allows understanding of this correlation. The hard endosperm kernels had higher percentage of coarse particles (retained in the 850 µm and 600 µm sieves) (*Milho Pérola* n° 65) and the kernels classified as intermediary that had particles between 250 µm and 180 µm of diameter (*Milho Grande* n° 52). In this way, we expected to have a significant correlation between hardness and the particle size distribution, but this fact did not happen.

In the processing of corn grains for starch extraction, the correlations confirmed the expected results. There were negative correlations between germ and endosperm percentages and endosperm and pericarp percentages (r=-0.85 and r=-0.55, respectively, at 95 % of confidence).

Starch extraction yield had a positive correlation with endosperm percentage (r= 0.50, at 95 % of confidence) and negative with germ percentage (r= -0.49, at 95 % of confidence).

The apparent viscosity profile (viscoamylography) of the starches was influenced by some of the studied parameters. The flotation index correlated negatively with peak viscosity (r= -0.48, at 95 % of confidence) and also with the final viscosity (r= -0.51, at 95 % of confidence). Corn varieties with low flotation index, classified in this work as hard corn (Salinas, Bustos & Gómez, 1992), presented higher average values of final viscosity (1799 cP), breakdown (772 cP) and peak viscosity (1719 cP). Seetharaman et al. (2001) when comparing starch paste properties of starches from corn with different hardness concluded that only very soft (floury) endosperms produced pastes with distinct characteristics. The starches from endosperms had higher gelatinization temperatures and lower values of peak viscosity, final viscosity, breakdown and setback. Due to this report, it would be expected that hard endosperms presented higher values for peak and final viscosity, breakdown and setback. The real density results confirmed this expectation as there was a positive correlation with viscosity peak (r= 0.50), final viscosity (r= 0.56), breakdown (r= 0.48) and setback (r= 0.60), always at 95 % of confidence level. Almeida-Domingues; Suhendro & Rooney

(1997) reported that hard endosperm particles need more time for hydration and starch gelatinization for resulting in viscous pastes.

In relation to complexed amylose, negative correlations were found with final viscosity (r=-0.45) and setback (r=-0.46). This is discussed in other papers (Ribeiro & Seravalli, 2004) that mention that lipids affect the setback as they form complexes with helicoidal amylose and make difficult water absorption in the granules. Jane *et al.* (1999) reported that the reduction in the viscosity peak is due to lipids and phospholipids complexation with amylose.

Wickramasinghe *et al.* (2009) studying starches from tuberous crops observed the influence of the granular swelling on peak viscosity and breakdown. These authors correlated positively breakdown with granular swelling and viscosity peak and stated that the bigger granules swollen more intensively. During stirred cooking, on the other hand, apparent viscosity dropped drastically. The corn starches from landraces of our study also presented a positive correlation between viscosity peak and breakdown (r=0.90), similar to the correlation found by Hou *et al.* (2008) (r=0.89; P<0.01).

Other points of the viscoamylograms were correlated as peak and final viscosities (r= 0.70, at 95 % of confidence). This was expected as these points are interdependent.

The viscosity peak and the setback were positively correlated (r= 0.56, at 95 % of confidence) due to amylose liberation during the starch granules swelling. Also the setback and the breakdown and the breakdown and the final viscosity were positively correlated (r= 0.55 and r= 0.49, respectively). Breakdown is an indicative of granular disruption related with amylose liberation. High concentrations of free amylose become available in the polymer solution and reassociate increasing the final viscosity (setback).

The gelatinization temperature and the breakdown presented a negative correlation (r= -0.49, at 95 % of confidence), what could be due to the energetic demand for complete swelling of the granules. As the gelatinization temperature rises the granular swelling becomes difficult and limited and then the breakdown is lower. It is important to clarify that in this work we used the STD1 RVA profile and analysis is very short, taking only 13 min to be completed.

**Table 5** – Correlation among the results of characterization of the grains and of the starches of 20 corn landraces.

```
MOS
            -1
 ASH
          ns
                 1
 PRO
         -0.45
                ns
 LIP
                      ns
          ns
                ns
 STA
          ns
                ns
                     -0.66
                            ns
  DF
                ns
                      ns
                            ns
          ns
                                  ns
 TKW
          ns
                ns
                      ns
                            ns
                                  ns
  RD
          ns
                ns
                      ns
                           0.67
                                  ns
                                       ns
                                             ns
  FI
          ns
                ns
                      ns
                            ns
                                  ns
                                        ns
                                             ns
                                                  -0.74
 COL
                ns
                      ns
                            ns
                                  ns
                                        ns
                                             ns
                                                   ns
                                                         ns
          ns
 COA
         0.46
                ns
                      ns
                            ns
                                  ns
                                        ns
                                             ns
                                                   ns
                                                         ns
 INT
          ns
                ns
                      ns
                            ns
                                  ns
                                       ns
                                             ns
                                                   ns
                                                         ns
                                                                    -0.93
 FIN
          ns
                ns
                      ns
                            ns
                                  ns
                                       ns
                                             ns
                                                   ns
                                                        ns
                                                              ns
                                                                    ns
 PER
          ns
                ns
                      ns
                            ns
                                  ns
                                        ns
                                             ns
                                                   ns
                                                         ns
                                                              ns
                                                                    ns
                                                                          ns
                           0.51
                                 -0.46
 GER
          ns
                ns
                      ns
                                       ns
                                             ns
                                                   ns
                                                         ns
                                                               ns
                                                                    ns
                                                                          ns
 END
          ns
                ns
                      ns
                            ns
                                  ns
                                        ns
                                             ns
                                                   ns
                                                         ns
                                                               ns
                                                                    ns
                                                                          ns
                                                                                ns
 YIE
          ns
                ns
                      ns
                            ns
                                  ns
                                       ns
                                             ns
                                                   ns
                                                        ns
                                                              ns
                                                                    ns
                                                                          ns
                                                                                ns
                                                                                      ns
                                                                                           -0.49
PRO STA
          ns
                ns
                      ns
                            ns
                                  ns
                                       ns
                                             ns
                                                   ns
                                                        ns
                                                              ns
                                                                    ns
                                                                          ns
                                                                                ns
                                                                                      ns
                                                                                            ns
                                                                                                 ns
                                                                                                        ns
                                                        -0.52
AMY AP
          ns
                ns
                      ns
                            ns
                                  ns
                                       ns
                                             ns
                                                   ns
                                                              ns
                                                                    ns
                                                                          ns
                                                                                ns
                                                                                      ns
                                                                                            ns
                                                                                                 ns
                                                                                                       ns
AMY_TO
                                                                                                                  0.59
                                  ns
                                                                                           -0.48
                                                                                                 ns
          ns
                ns
                      ns
                            ns
                                       ns
                                             ns
                                                   ns
                                                        ns
                                                              ns
                                                                    ns
                                                                          ns
                                                                                ns
                                                                                      ns
                                                                                                       ns
                                                                                                             ns
                                                                                                 0.51
                                                                                                                        0.83
AMIY_LIP
                                                                                           -0.65
          ns
                ns
                      ns
                            ns
                                  ns
                                       ns
                                             ns
                                                   ns
                                                        ns
                                                              ns
                                                                    ns
                                                                          ns
                                                                                ns
                                                                                      ns
                                                                                                       ns
                                                                                                             ns
                                                                                                                   ns
                                                                                           0.48
AMYLO
                                                                                                                        -1.00
          ns
                ns
                      ns
                            ns
                                  ns
                                       ns
                                             ns
                                                   ns
                                                        ns
                                                              ns
                                                                    ns
                                                                          ns
                                                                                ns
                                                                                      ns
                                                                                                 ns
                                                                                                       ns
                                                                                                             ns
 Ponset
          ns
                ns
                      ns
                            ns
                                  ns
                                       ns
                                             ns
                                                   ns
                                                         ns
                                                               ns
                                                                    ns
                                                                          ns
                                                                                ns
                                                                                      ns
                                                                                            ns
                                                                                                 ns
                                                                                                       ns
                                                                                                             ns
                                                                                                                   ns
                                                                                                                         ns
                                                                                                                               ns
                           0.54
                                                  0.50
                                                        -0.48
                                                             -0.52
                                                                               0.50
                                 ns
                                                                    ns
          ns
                ns
                      ns
                                       ns
                                             ns
                                                                          ns
                                                                                      ns
                                                                                            ns
                                                                                                 ns
                                                                                                       ns
                                                                                                             ns
                                                                                                                   ns
                                                                                                                         ns
                                                                                                                              ns
 HPV
          ns
                ns
                      ns
                            ns
                                  ns
                                       ns
                                             ns
                                                   ns
                                                              ns
                                                                    ns
                                                                          ns
                                                                                      ns
                                                                                            ns
                                                                                                             ns
                                                                                                                   ns
                                                                                                                         ns
                                                                                                                                          ns
                                                        -0.51
                                                                               0.50
                                                                    ns
          ns
                ns
                      ns
                                 ns
                                       ns
                                             ns
                                                              ns
                                                                          ns
                                                                                      ns
                                                                                                             ns
                                                                                                                   ns
                                                                                                                                          ns
                                                  0.48
                                       ns
                                             ns
                                                                          ns
                ns
                      ns
                           ns
                                 ns
                                                        ns
                                                                                ns
                                                                                            ns
                                                              -0.46
                                                                               0.46
```

MOS: Moisture; ASH: Ash; PROT: Protein; LIP: Lipids; STA: Starch; DF: Dietary Fiber; TKW: 1.000 kernel weight; RD: Real density; FI: Flotation Index; COL: Color; COA: Coarse particles; INT: Intermediate particles; FIN: Fines particles; PER: Pericarp; GER: Germ; END: Endospern; YIE: Yield; PRO\_STA: Protein Starch; AMY\_AP: Apparent amylase; AMY\_TO: Total amylase; AMY\_LIP: Amylose-lipids complex; AMYLOP: Amylopectin; Ponset: Gelatinization temperature; PV: Viscosity pasting; HPV: Hot paste viscosity; FV: Final viscosity; BD: Breakdown and ST: Setback.

# **Principal component analysis (PCA)**

The PCA was used to explore the variability of samples considering the selected physicochemical and physical aspects of the grains and of their starches. For an initial analysis, all the variables were considered to check the degree of explained variance. By using ten factors the explained variance reached 92.29 %. The two first PC were responsible for explaining 36.06 % (PC1 = 22.42 % and PC2 = 13.66 %) of total variance of the original data. For improving the variance explanation of the model, some variables were excluded: moisture, thousand kernels weight, color, total amylose, fine particles percentage and setback. With this next data the PCA with ten factors explained 96.22 % of the total variance. The two first PCs explained 41.52 % (PC1 = 25.58% and PC2 = 16.84 %) of total variance of the original data. The samples were grouped in the same manner as in the first PCA, with all variables.

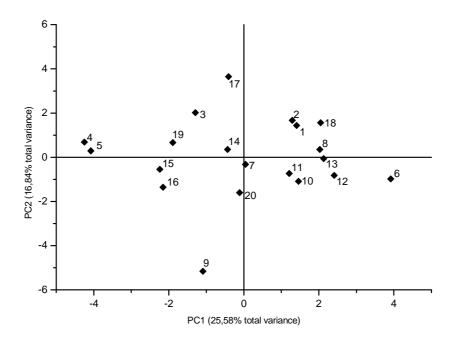
The positive PC1, in the right side (Fig 1), is associated with the percentage of germ, lipids, real density, protein, intermediary size particles, breakdown, peak and hot paste viscosities and gelatinization temperature. In the left side of the PC1 are the variables starch, ash, dietary fiber, percentage of endosperm and of pericarp, starch yield and flotation index. This PC should be considered a dimension of endosperm density and hardness as the samples presenting higher real densities and lower flotation indexes are located in the positive side of the component. On the other hand, the less dense kernels presenting higher flotation indexes are located in the negative side of the PC1.

The PC2 was characterized by the particle size distribution, staying in one side that of coarse and in the other of intermediary granulometry. The grains that after milling produced higher amounts of coarse particles are located in the negative side of PC2 and the samples with higher amounts of intermediary size particles are in the positive side. As a consequence, these two variables allow a separation among grains with different hardness, as the varieties with proportionally more coarse

particles have their endosperms classified as hard. In this way, the interaction of flotation index, real density, coarse and intermediary particles were responsible for the separation of the varieties in four groups (Fig 1) identified with numbers from 1 to 4, in decreasing order of endosperm hardness. The group 1 is classified as low-intermediary hardness, as the samples presented higher flotation indexes, lower densities and higher percentage of intermediary particles. The group 4, on the other hand, includes grains with higher densities, lower flotation indexes and higher percentage of coarse particles, being classified as hard.

Another grouping showed in Fig 1 includes the grains that presented higher percentage of germ and lipids in the positive side of PC1. This fact explains the correlation between real density and lipid content (r = 0.67 at 95 % of confidence).

Another PCA analysis was carried out considering only the main physicochemical results (protein, dietary fiber, lipids and starch contents) and the morphological fractions (endosperm, pericarp and germ percentages) of the corn landraces. In the Fig 2 the results clearly show that in the endosperm fraction is located the great part of starch, in the pericarp the fiber fraction and in the germ is rich in lipids and protein. This analysis allows the identification of two main groups in the corn landraces studied (Fig 3). The first group includes samples that have more endosperm and starch and are located at the negative side of the PC1 (varieties marked as 3 and 4). The second group (varieties marked as 2, 6, 12 and 18) is characterized by kernels with higher percentage of germ and consequently with more protein and lipids. The variety identified as 16 stayed between the two groups due to its intermediary chemical composition. The varieties with the higher levels of dietary fiber are those located in the center of the graph. The variety coded as 20 had higher percentage of pericarp and due to that is isolated in the upper part of the graph. The other samples, 8, 11 and 13 are in between the groups separated by dietary fiber, lipids, protein and germ presenting intermediary values for these parameters.



**Figure 1** – Grouping of corn landraces according to their physicochemical and physical properties and their starches.

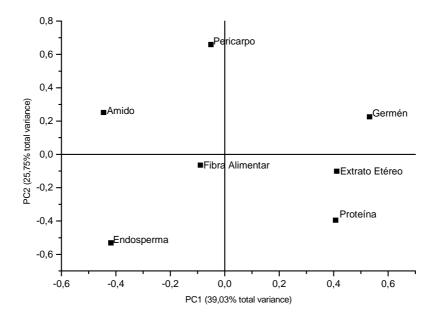
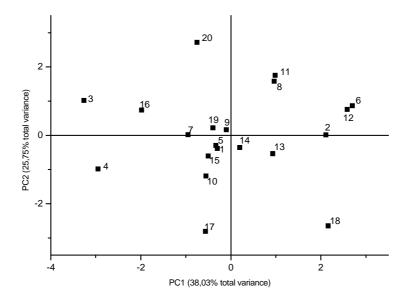


Figure 2 - Effect of the selected variables in the PCA analysis (loadings).



**Figure 3 -** Grouping of corn landraces by selected physicochemical characteristics and morphological fractions.

#### CONCLUSION

The corn landraces presented relatively high germ percentage and consequently high lipid contents. The physical properties of corn grains like flotation index, real density and particle size distribution are important parameters for classifying endosperm hardness that was correlated with physicochemical characteristics. Grains with hard endosperms tend to present higher real density, lower flotation index and higher percentage of coarse particles after milling. The opposite pattern was met for intermediary hard kernels.

# **AKNOWLEDGMENTS**

The financial resources for the conduction of this study were supported by the project BioAgroPar financed by FINEP, SETI/PR, and Fundação Araucária/PR; and by CNPq/Brazil.The authors thank to the *Coordenação de Aperfeiçoamento de Pessoal de Nível Superior* (CAPES) for scholarships.

#### RESUMO

Esse trabalho teve o objetivo de caracterizar vinte variedades de milho crioulo cultivadas na região dos Campos Gerais (Estado do Paraná) em relação a sua composição química (umidade, cinzas, proteína, extrato etéreo, fibra alimentar e amido) e propriedades físicas (peso de 1000 grãos, densidade real, índice de flotação, granulometria e cor) Além disso, foi feito o processamento dos grãos em escala de laboratório para a extração do amido, sendo mensurado o teor de proteína. Foram avaliados os conteúdos de amilose e o perfil viscoamilográfico. Os resultados mostraram que os milhos apresentaram diferentes composições químicas e proporções pericarpo/endosperma/ gérmen e consequentemente podem ter diferentes aplicações industriais e interesse ao melhoramento de plantas.

# **REFERENCES**

Almeida-Dominguez, H. D.; Suhendro, E. L; Rooney, L. W. (1997), Factors Affecting Rapid Visco Analyser Curves for the Determination of Maize Kernel Hardness. *Journal of Cereal Science*, **25**, 93-102.

- AOAC (2000), Official methods of analysis of AOAC international. 17<sup>th</sup>.ed. Washington.
- Birol, E.; Villalba, E. R.; Smale, M. (2007), Farmer Preferences for *Milpa* Diversity and Genetically Modified Maize in Mexico. Washington: International Food Policy Research Institute, pp. 44.
- Blazek, J. (2008), Role of amylose in structure-function relationship in starches from Australian wheat varieties. Doctor of Philosophy, Thesis, University of Sydney, Sidney.
- Câmara, R. J. (2005), Cultivares crioulas de milho (Zea mays, L.) em sistema de produção orgânico Desempenho agronômico das plantas e composição química das sementes. Mestrado em Agronomia Dissertação, Universidade do Oeste do Paraná, Marechal Cândido Rondon, Paraná.
- Cereda, M. C. et al. (2001), Propriedades gerais do amido. ed. Fundação Cargill, CAMPINAS, pp. 50-60.
- cruz, j. c.; konzen, e. a.; pereira filho, i. a.; marriel, i. e.; cruz. i.; duarte, j. o.; oliveira. m. f.; alvarenga, r. c. (2006), Produção de Milho Orgânico da Agricultura Familiar. *In-Ministério da Agricultura e Abastecimento*. Circular Técnica, pp. 1-17.
- Demiate, I. M.; Konkel, F. E.; Pedroso, R. A. (2001), Enzymatic determination of starch in *doce de leite* using dialysis. *Ciência e Tecnologia de Alimentos*, 21, 339-342.
- Dowd, M. K. (2003), Improvements to laboratory-scale maize wet-milling procedures. *Industrial Crops and Products*, **18**, 67-76.
- Fowler, C.; Hodgkin, T. (2004), Plant genetic resources for food and agriculture: Assessing global availability. *Annual Review of Environmental Resources*, **29**, 143-179.
- Fox, S. R.; Johnson, L. A.; Hurburgh, C.R.; Dorsey-Redding C.; Bailey T. B. (1992), Relation of grain proximate composition of physical properties to wetmilling characteristics of maize. *Cereal Chemistry*, **69**, 191-197.
- Gibbon, B. C.; Larkins, B. A. (2005), Molecular genetic approaches to developing quality protein maize. *Trends in Genetics*, **21**, 227-233.
- Gibbon, B. C.; Wang, X.; Larkins, B. A. (2003), Altered starch structure is associated with endosperm modification in Quality Protein Maize. Proceedings of the National Academy of Science of United States of America, 100, 15329–15334.
- Gonçalves, R. A.; Santos, J. P.; Tomé, P. H. F.; Pereira, R. G. F. A.; Ascheri, J. L. R.; Abreu, C. M. P. (2003), Rendimento e composição química de cultivares de milho em moagem a seco e produção de grits. *Ciência e Agrotecnologia*, **27**, 643-650.
- Hou, H. X.; Dong, H. Z.; Lui, C. F.; Zhang H. (2008), Relationships Between Some Physicochemical Properties of Starches from Maize Cultivars Grown in East China. *Starch/Stärke*, **60**, 305–314.

- IAL (1985) Normas Analíticas do Instituto Adolfo Lutz: Métodos Químicos e Físicos para Análise de Alimentos. São Paulo: 3ª ed, pp.302-30.
- Ji, Y.; Seetharaman, Y. J. K., White, P. J.(2004), Optimizing a small-scale corn starch extraction method for use in the laboratory. *Cereal Chemistry*, **81**, p. 55-58.
- Jiang, H. Y.; Zhu, Y. J.; Wei, L. M.; Dai, J. R.; Song, T. M.; Yan, Y. L. Chen, S. J. (2007), Analysis of protein, starch and oil content of single intact kernels by near infrared reflectance spectroscopy (NIRS) in maize (Zea mays L.). *Plant Breeding*. 126, 492-497.
- Lojano-Alejo, N.; Carrillo, G. V.; Pixley, K..; Palacios-Rojas, N.(2007), Physical properties and carotenoid content of maize kernels and its nixtamalized snacks. *Innovative Food Science and Emerging Technologies*, **8**, 385-389.
- Lopes-Filho, J. F. (1997), Moagem úmida do milho para produção de amidos e subprodutos. *Boletim SBCTA*, **31**, 42-47.
- Lucchin, M; Barcaccia, G; Parrini, P. (2003a), Characterization of a flint maize (Zea mays L. convar. Mays) Italian landrace: I. Morphophenological and agronomic traits. *Genetic Resources* and Crop Evolution, 50, 315-327.
- Mauricio, R. A. S.; Figueroa, J. D. C.; Taba, S.; Reyes, M. L. V.; Rincón, F. S.; Mendoza, A. G. (2004), Characterization of maize accessions by grain and tortilla quality traits. *Rev. Fitotec. Mexico*, 27, 213-222.
- Miranda, G. V.; Souza, L. V. De; Santos, I. C. Dos; Mendes, F. F. (2007), Resgate de variedade crioulas de milho na região de Viçosa-MG. *Revista Brasileira Agroecologia*, **2**, 1145-1148.
- Mizuma, T; Kiyokawa, Y.; Wakai, Y. (2008), Water Absorption Characteristics and Structural Properties of Rice for Sake Breweering. *Journal of Bioscience and Bioengineering*, **106**, 258–262.
- Narvaéz-González, E. D.; Figueroa-Cárdenas, J De D.; Taba, S.; Tostado, E. C.; Peniche, R. A. M.; Sánchez, F. R. (2006), Relationships between the microstructure, physical features and chemical composition of different maize accessions from Latin América. *Cereal Chemistry*, **83**, 595-604.
- Nelles, E. M.; Dewar, J.; Bason, M. L.; Taylor, J. R. N. (2000), Maize Starch Biphasic Pasting Curves. *Journal of Cereal Science*, **31**, 287-294.
- Pereira, R. C.; Davide, L. C.; Pedrozo, C. A.; Carneiros, N. P.; Souza, L. R. P.; Paiva, E. (2008), Relationship between structural and biochemical characteristics and texture of corn grains. *Genetics of Molecular Research*, **7**, 498-508.
- Philippeau, C.; Monredon, F. Le D. De; Michalet-Doreau, B. (1999), Relationship between ruminal starch degradation and the physical characteristics of corn grain. *Journal of Animal Science*, 77, 238-243.

- Posner, E. S. (1985), The technology of wheat germ separation in flour Mills. *Association of Operative Millers*. 4577-4592.
- Ribeiro, E. P.; Seravalli, E. A. G. (2004), Química dos Alimentos. 1. ed. São Paulo: Edgard Blücher, pp. 184.
- Salgo, A.; Juhasz, R. (2008), Pasting Behavior of Amylose, Amylopectin and Their Mixtures as Determined by RVA Curves and First Derivatives. *Stärke*, **60**, 70-78.
- Salinas, M. Y.; Bustos, F.; Gomez J. H. (1992), Comparación de métodos para medir la dureza del maíz (Zea mays L.). *Archivos Latinoamericanos de Nutrición*, **42**, 59–63.
- Schmildt, E. R; Krause, W; Cruz, C. D. (2006), Melhoria da eficiência dos experimentos de indicação de cultivares de milho. *Ciência e Agrotecnologia*. **30**, 72-80.
- Schoch, T. J.; Maywald, E. C. (1968), Preparation and properties of various legume starches. *Cereal Chemistry*, **45**, 564-573.

- Seetharaman, K.; Tziotis, A.; Borras, F.; White, P. J.; Ferrer, M.; Robutti, J. (2001), Thermal and functional characterization of starch from Argentinean corn. *Cereal Chemistry*, **8**, 379-386..
- SILVA, F. de A. E. (2004), Assistat versão 7.1 beta. Departamento de Engenharia agrícola do CCT-UFGC, Campina Grande PB.
- Trindade, C. C. (2006), Sementes crioulas e transgênicos. Uma reflexão sobre sua relação com as comunidades tradicionais. Trabalho apresentado no XV Congresso Nacional do Conpedi, 15-18 Nov, Manaus, Amazonas.
- Wickramasinghe, H. A. M.; Takigawa, S.; Endo, C. M.; Yamauchi, H.; Noda, T.(2009), Comparative analysis of starch properties of different root and tuber crops of Sri Lanka. *Food Chemistry*, **112**, 98–103