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Comparison of Pasting and Gel Stabilities of Waxy and Normal Starches from Potato, Maize, and Rice with Those of a Novel Waxy Cassava Starch under Thermal, Chemical, and Mechanical Stress

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Functional properties of normal and waxy starches from maize, rice, potato, and cassava as well as the modified waxy maize starch COLFLO 67 were compared. The main objective of this study is to position the recently discovered spontaneous mutation for amylose-free cassava starch in relation to the other starches with well-known characteristics. Paste clarity, wavelength of maximum absorption ($\lambda_{\max}$), pasting properties, swelling power, solubility, and dispersed volume fraction measurements and gel stability (acid and alkaline resistance, shear, refrigeration, and freeze/thaw stability) were evaluated in the different types and sources of starch included in this study. $\lambda_{\max}$ in the waxy cassava starch was reduced considerably in comparison with that of normal cassava starch (535 vs 592 nm). RVA peak viscosity of waxy cassava starch was larger than in normal cassava starch (1119 vs 937 cP) and assumed a position intermediate between the waxy potato and maize starches. Acid, alkaline, and shear stability of waxy cassava starch were similar to normal cassava except for alkaline pH, at which it showed a low effect. Gels from normal root and tuber starches after refrigeration and freeze/thaw had lower syneresis than cereal starches. Gels from waxy starches (except for potato) did not present any syneresis after 5 weeks of storage at 4 °C. Waxy cassava starch was the only one not showing any syneresis after 5 weeks of storage at −20 °C. Natural waxy cassava starch is, therefore, a promising ingredient to formulate refrigerated or frozen food.

KEYWORDS: Syneresis; gel stability; functional properties; waxy cassava starch; storage

INTRODUCTION

The markets for industrial starches are expanding, and current demand is satisfied mostly by four crops: potato, maize, wheat, and cassava or tapioca (*Manihot esculenta* Crantz) (*1*, *2*). Cassava is particularly important as a source of starch in tropical and subtropical regions of the world. About 84.5% of the dry root weight of cassava is starch (*3*). Compared with other root and tuber tropical crops, cassava starch and its biosynthesis have been well studied (*1*−*8*). The starch granules are generally round (oval), with a flat surface on one side (truncated). The size of individual granules ranges from 5 to about 40 μm, with reported averages varying from 5.4 to 17.2 μm (*5*, *6*). However, recently a mutation with considerably smaller starch granule size has been reported (*9*). Starches from cassava and potato share some similarities: they produce relatively bland pastes, with higher viscosity, better clarity, and lower retrogradation rates than starches from cereals. They also possess lower levels of proteins and lipids (*1*, *2*). On the other hand, the functional properties of potato starch are influenced by the presence of phosphate monoester groups in its amylopectin and their large granule size (*5*).

There is a widespread variation of cassava starch biochemical and functional properties reported in the literature (*5*, *6*, *10*). In a description of analyses made in starches from more than 4000 genotypes recently published, average amylose content was found to be 20.7% (*3*). An amylose-free (or waxy) natural mutation has also been reported (*11*) as well as the application of genetic transformation for the development of cassava with a waxy starch (*12*).

Many comparisons of the physicochemical and functional properties of starches from different crops have been published (*1*, *2*, *13*−*19*). Normal cassava and potato starches have high swelling power and dispersed volume fraction compared with starches from other tropical root and tuber crops (*19*). Comparisons between normal and transgenic waxy cassava starch demonstrated that the latter increased the clarity and stability of gels and increased the granule melting temperature by almost 2 °C, without affecting particle size or chain length distributions or phosphorus contents (*12*).

Despite the diversity offered by native starches from different crops, they cannot satisfy the different demands by different industries [adhesives, agrochemicals, cosmetics, detergents, food,

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medical oil and gas, paper and board, pharmaceutical, plastics, purification, beverages, textile (*1*, *2*)], and chemical or physical modifications in vitro are required. However, these modifications are often environmentally unfriendly and incur additional costs, and there is a growing preference by consumers for natural, unmodified products. Therefore, there are increasing interest and growing opportunities (conventional breeding and genetic transformation) in achieving some of these modifications in planta (*1*, *2*). The recent discovery of two starch mutations in cassava (*9*, *11*) is relevant because it widens the applications and competitiveness of cassava starch, because they offer the advantages of modifications in planta, and because this crop is a key source of starch for tropical regions of the world. There are ongoing efforts to develop commercial cassava varieties with waxy starch. Field selections will be conducted in Thailand by mid 2010.

The aim of the present study was to compare physicochemical and functional properties of normal and waxy starches from different crops, taking advantage of the recent availability of a natural mutation for waxy cassava starch in genotype AM206-5 (*11*).

MATERIALS AND METHODS

Raw Material. *Cassava Starch Isolation*. Freshly cut pieces of cassava roots were suspended in tap water and crushed in a 4 L capacity Waring commercial blender (New Hartford, CT). The slurry was filtered through a 100 μm sieve. The starch was allowed to settle and the supernatant decanted off. Solids were washed with distilled water twice and centrifuged at 8000 rpm for 10 min (*20*). The sample was then dried in an oven with fan-forced ventilation at 40 °C for 2 days (Thelco Oven model 28, Precision Scientific Subsidiary of GCA Corp., Chicago, IL). Starches from four nonwaxy genotypes were included in this study. Mutant 7 is a starch from a genotype that has frequently shown distinctive properties. The other three genotypes have been released for commercial production and are adapted to contrasting environments in Colombia. CM 523-7 is a variety adapted to the acid soil savannas. MPER 183 is adapted to the midaltitude valley environment. MTAI 8 was released in Thailand as Rayong 60 and is adapted to subhumid conditions. The study also includes the waxy cassava starch from clone AM 206-5 (*11*). This clone was obtained from a self-pollination that allowed the expression of the recessive trait responsible for the production of amylose-free starch. It is a natural mutation similar to those observed in other crops. The starch of this clone completely lacks amylose, based on DSC analysis. All cassava starches were obtained following the same extraction protocol on root samples from plants grown at the CIAT Experimental Farm in Palmira, Colombia, which is approximately 1000 m above sea level (masl) and harvested at 10 months after plating.

Commercial Starches. Normal and waxy commercial starches were used in this study. Maize (*Zea mays*) starches were from Roquette, Lestrem, France, and potato (*Solanum tuberosum*) starches from Avebe, The Netherlands. Normal rice (*Oryza sativa*) was from Bangkok Starch Industrial Co., Ltd., Thailand, and waxy rice was from Remy Industries, Belgium. In addition, the modified maize starch COLFLO 67 from National Starch (France) was also included in the study. This starch was obtained by reticulation/stabilization of native waxy maize starch.

Starch Characterization. *Moisture Content*. This parameter was determined on 10 g of starch samples (iw) which were then placed at 100 °C for 24 h and weighed again (fw). Moisture expressed as percentage was determined as (fw/iw) × 100. Three different quantifications per starch sample were made, and mean values were then calculated.

Amylose Content. This parameter was measured following standard colorimetric procedures (*21*). Starch granules were first dispersed with ethanol and then gelatinized with 1 N sodium hydroxide. An aliquot was then acidified and treated with a 2% iodine solution, which produces blue-black stain coloration. The color intensity, which is related to amylose content, was then measured with a spectrophotometer (620 nm wavelength) and compared with standard curves (using amylose concentrations ranging from 0 to 30%). Curves were obtained using purified amylose extracted from potato tubers and amylopectin extracted from cassava roots [variety AM 206-5 (*11*)]. Three different quantifications per starch sample were made, and mean values were then calculated.

Paste Clarity. The methodology suggested by Craig et al. (*22*) was used. A 1% dry base (db) aqueous dispersion of starch was boiled at 97 °C (1000 masl) with thorough shaking every 5 min for 30 min. Transmittance was measured after the starch had cooled to room temperature at 650 nm. Two different quantifications per starch sample were made, and mean values were then calculated.

Wavelength of Maximum Absorption (λ_{max}). The formation of iodine complexes with amylose, amylopectin, and their mixture was determined after solubilization in 1 M KOH for 3 days at 4 °C under stirring. The solution was diluted to 0.1 M KOH at a final polymer concentration of 1 g L^{-1}. One milliliter of 0.1 N HCl, 3 mL of distilled water, and 0.2 mL of iodine solution (2% KI, 0.2% I_2) were added to 1 mL of glucan solution in 0.1 N KOH. The absorption spectrum was recorded with a μQUANT spectrophotometer (BioTek Instruments, Winooski, VT) between 450 and 700 nm (*19*).

Pasting Properties. Hot starch dispersion viscosity profiles were obtained with a Rapid Visco Analyzer model RVA-4 series (Newport Scientific, Australia). Starch (1.25 g db) was dispersed in distilled water (near 23 cm^3) to 5% suspension. Starch concentration is critical for RVA results. The concentration used was adequate for comparing different starches and fell within the range of concentrations frequently reported the literature. Viscosity was recorded using the following temperature profile: hold at 50 °C for 1 min, heat from 50 to 90 °C at 6 °C min^{-1}, hold at 90 °C for 5 min, and then cool to 50 at 6 °C min^{-1}. The gel was then maintained for 2 min at 50 °C with continuous stirring at 160 rpm. Four parameters were measured: pasting temperature (PT), peak viscosity (PV), hot paste viscosity at the end of the plateau at 90 °C (HPV), and cool paste viscosity (CPV) at 50 °C (19.33 min analysis). With them, three additional parameters were calculated: breakdown (BD), estimated as PV − HPV; setback (SB), estimated as CPV − PV; and consistency (CS), estimated as CPV − HPV.

Swelling Power, Solubility, and Dispersed Volume Fraction Measurements. Swelling power (SW) and solubility patterns (SO) (*23*) were determined using 1% db (w/w) starch dispersions (0.28 g db dispersed in 27.72 g of distilled water) at 60, 75, and 90 °C. The low concentration used in the study was chosen to obtain an optimal separation between the pellet and supernatant phases after centrifugation. Paste was prepared in RVA starting at 35 °C for 1 min, increasing temperatures at a 6 °C min^{-1} rate. Three different and independent analyses were made, holding final temperatures at 60, 75, or 90 °C for 2.5 min. Stirring was at 960 rpm for the first minute and then maintained at 160 rpm during the entire analysis. The paste was immediately transferred to a 50 cm^3 centrifuge tube. The supernatant and sediment after centrifugation for 10 min at 6000*g* at 25 °C were collected and weighed (Wsu and Wse, respectively) and then dried at 100 °C for 24 and 48 h, respectively, and weighed (Dsu and Dse, respectively). The values thus obtained were used to calculate three parameters: concentration of soluble material in the supernatant (solubility), swelling power, and volume fraction of the dispersed phase (Φ) as follows:

$$\text{solubility (\% db)} = 100 \times \text{Dsu}/0.28$$

$$\text{swelling power}(g_{\text{water}}/g_{\text{starch}}) = (\text{Wse} - \text{Dse})/\text{Dse}$$

$$(\Phi) = [27.91 - (\text{Wsu} - \text{Dsu})]/27.91$$

Factor 27.91 is calculated as total volume (cm^3) of the paste. Starch specific density is 1.5 g cm^{-3}.

$$27.91 = 27.72 + (0.28/1.5)\text{cm}^3$$

Gel Stability. *Acid and Alkaline Resistance*. The starch sample (5%, w/v, db) was dispersed in 25 mL of distilled water, in 25 mL of 0.1 M potassium chloride and sodium borate solution (pH 9), or in 25 mL of 0.2 M citrate−phosphate buffer (pH 3). An RVA was used with the same profiles mentioned above for the analysis of pasting properties. Cool paste viscosity (CPV) at 50 °C (19.33 min of analysis) for the different pH values was reported as CPV_{pH3}, CPV_{water}, and CPV_{pH9}. The acidity effect was defined as the ratio between CPV_{pH3} and CPV_{water} and the basic effect as the ratio between CPV_{pH9} and CPV_{water} (*14*, *24*). These formulas indicate that pH-resistant gels have values around 1. Susceptible gels, on the other hand, will result in values relatively distant from 1 (either close to 0 or above 2).

Table 1. Physicochemical Properties of Different Types of Starch (Normal, Waxy, or Modified) of Different Crops[a]

starch type	amylose content (%)	paste clarity (%)	λ_{max}
Normal Starches			
maize	19.9 (±0.4)	11 (±2.7)	590
potato	27.7 (±0.5)	88 (±0.8)	591
rice	9.7 (±0.6)	10 (±1.2)	570
CM 523-7 (cassava)	19.8 (±1.3)	50 (±3.5)	593
MPER 183 (cassava)	19.5 (±1.8)	51 (±3.8)	590
MTAI 8 (cassava)	16.5 (±0.6)	47 (±0.8)	592
Waxy Starches			
maize	0.0	42 (±1.1)	529
potato	7.7 (±0.8)	92 (±1.4)	550
rice	0.0	13 (±0.6)	531
cassava (AM 206-5)	0.0	61 (±0.7)	535
Other Starches			
COLFLO 67	0.0	5 (±0.1)	na
Mutant 7 (cassava)	21.3 (±1.1)	47 (±4.1)	592

[a] Standard deviations are presented within parentheses. na, not available.

Shear Stability. The shear effect was measured using the following ratio: CPV_{shear}/CPV (*14*). CPV is the same parameter described above under Pasting Properties. CPV_{shear} was recorded using the same temperature profile, but there was a change in stirring: 2 min after the maximum temperature (90 °C) had been reached, stirring was increased to 960 rpm for 3 min. Then the stirring and temperature profile continued as in the standard pasting properties analyses.

Refrigeration and Freeze/Thaw Stability. Paste was prepared in RVA (5% db, w/w, containing 0.1% sodium azide) from 35 °C, increasing temperatures at a 6 °C min^{-1} rate to 93 °C (the boiling temperature of water at Cali, Colombia, is 97 °C) and maintained during 2 min. Protocol followed common procedures (*12*, *14*, *18*) with slight modifications (pastes were prepared using the RVA, and the starch concentration was 5% rather than 4%).

Syneresis after Refrigeration. Ten centrifuge tubes were filled with approximately 6 g of gel (WG) and stored at 4 °C. The study lasted for 5 weeks. Every 7 days, two tubes were taken out of the refrigerator and held at room temperature during 1 h. Tubes were centrifuged at 5000 rpm for 10 min, and supernatant was separated and weighed (WS). Syneresis was calculated as WS/WG × 100.

Syneresis after Freeze/Thaw. Ten centrifuge tubes were filled with approximately 6 g of gel (WG) and stored at −20 °C. The study lasted for 5 weeks. Every 7 days, two tubes were taken out of the freezer and held during 1.5 h in a water bath (30 °C). Samples were then centrifuged (5000 rpm for 10 min), and then supernatant was separated and weighed (WS). Syneresis was calculated as WS/WG × 100.

All analyses were replicated. In the cases of moisture and amylose content, paste clarity, and swelling power, three independent replications were used. For wavelength of maximum absorption, pasting properties and gel stability test results are based on two replications.

RESULTS

The physicochemical properties of the different starches included in this study are presented in **Table 1**. Paste clarity in normal starches ranged from 10% (rice) to 88% (potato). Paste clarity of normal cassava was intermediate, around 50%. Waxy starches from each crop showed higher paste clarity than their respective normal starches. The change was particularly noticeable in the case of maize and much smaller in the cases of potato and rice. The modified starch COLFLO 67 had very low paste clarity (about 5%). As expected, amylose content in waxy starches (including COLFLO 67) was negligible except for potato, which showed values around 8% (this can be explained by the branched amylopectin molecules of potato starch (*25*)).

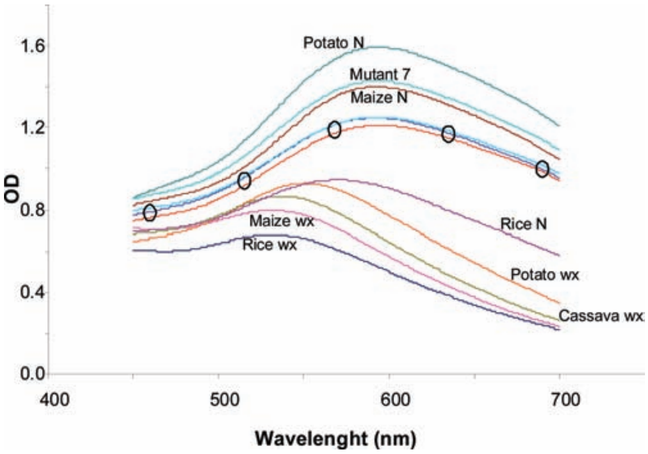

Figure 1. Absorption spectra of different starches analyzed in this study. The rings group together the three commercial cassava clones.

Table 2. Effect of pH and Shear on Gels from Different Types of Starch (Normal, Waxy, or Modified) of Different Crops[a]

starch type	pH acid effect	pH alkaline effect	shear effect
Normal Starches			
maize	1.23 (±0.06)	4.12 (±0.41)	1.13 (±0.02)
potato	0.58 (±0.03)	1.53 (±0.02)	0.52 (±0.02)
rice	0.72 (±0.05)	3.64 (±0.26)	0.94 (±0.04)
CM 523-7	0.39 (±0.05)	3.22 (±0.35)	0.59 (±0.03)
MPER 183	0.43 (±0.10)	2.77 (±0.29)	0.67 (±0.03)
MTAI 8	0.53 (±0.01)	3.73 (±0.06)	0.55 (±0.07)
Waxy Starches			
maize	0.45 (±0.01)	2.32 (±0.27)	0.35 (±0.03)
potato	0.33 (±0.02)	2.26 (±0.10)	0.65 (±0.05)
rice	0.69 (±0.02)	2.66 (±0.24)	0.52 (±0.04)
cassava	0.50 (±0.04)	2.77 (±0.19)	0.55 (±0.05)
Other Starches			
COLFLO 67	1.05 (±0.03)	1.33 (±0.05)	1.09 (±0.12)
Mutant 7	0.36 (±0.01)	3.15 (±0.04)	0.60 (±0.03)

[a] Standard deviations are presented within parentheses.

The normal rice starch analyzed in our study had a relatively low level of amylose (around 10%). λ_{max} values were clearly higher in the gels from normal starches compared with waxy starches from different crops. **Figure 1** illustrates the absorbance spectra of gels from different starches in the presence of iodine. The color of waxy gels was brown; starch containing amylose developed blue gels. The intensity of the color was related to amylose content; the highest absorbance spectra are for the starches with highest amylose content.

Effects of pH and shearing in gels from the starches analyzed in this study are presented in **Table 2**. Acid conditions in normal starches showed a minimum effect on maize starch, increasing the viscosity of its gels (1.23), followed by rice (0.72), potato (0.58), and then the different cassava samples (from 0.39 in CM 523-7 to 0.53 in MTAI 8). Mutant 7 had the lowest resistance among nonwaxy cassava samples (0.36). Differences in the performances of gels from waxy and normal starches from cassava or rice were not large (**Table 2**). On the other hand, waxy maize starch had a considerably lower resistance to acid medium than normal maize starch (0.45 vs 1.23). In the case of potato the effect on acid pH resistance was also lower in the waxy starch compared with normal starch, but the difference was not as marked as in the case of maize.

Table 3. Pasting Characteristics from Different Types of Starch (Normal, Waxy, or Modified) of Different Crops (5% Suspensions)[a]

starch type	PT (°C)	PV (cP)	BD (cP)	SB (cP)	CS (cP)
			Native Starches		
maize	89.0 (±0.85)	176 (±4)	−30 (±4)	−15 (±3)	−45 (±6)
potato	65.2 (±0.06)	2550 (±15)	1204 (±29)	−1082 (±2)	108 (±5)
rice	84.0 (±0.40)	343 (±7)	22 (±2)	12 (±0)	34 (±2)
CM 523-7	63.3 (±0.12)	1006 (±14)	500 (±22)	−364 (±8)	137 (±14)
MPER 183	64.8 (±0.12)	979 (±12)	482 (±15)	−267 (±10)	215 (±8)
MTAI 8	63.7 (±0.00)	876 (±13)	455 (±0)	−338 (±4)	117 (±4)
			Waxy Starches		
maize	70.9 (±0.00)	973 (±22)	307 (±25)	−289 (±4)	31 (±2)
potato	65.9 (±0.12)	2491 (±49)	1287 (±30)	−1268 (±35)	13 (±3)
rice	67.0 (±0.71)	498 (±16)	39 (±1)	15 (±6)	54 (±4)
cassava	67.4 (±0.00)	1119 (±11)	631 (±8)	−595 (±12)	37 (±4)
			Other Starches		
COLFLO 67	69.0 (±0.12)	na[b]	na	na	na
Mutant 7	68.0 (±0.17)	887 (±20)	324 (±4)	−157 (±16)	167 (±12)

[a] Standard deviations are presented within parentheses. [b] Not available.

The modified COLFLO 67 had better stability than standard waxy or normal maize starches with a value very close to 1.

Alkaline pH effect increased viscosity and was also highest in the case of normal maize starch (4.12) and lowest for potato (1.53). Normal cassava starches assumed intermediate values ranging from 2.77 (MPER 183) to 3.73 (MTAI 8). Waxy cassava did not show much difference compared with normal cassava starches for the effects of alkaline pH, with a slightly better resistance. The alkaline pH resistance of waxy maize gels was considerably better (2.32) than that of the normal maize gels (4.12). The effect of alkaline pH on waxy potato starch, on the other hand, increased from 1.53 to 2.26. COLFLO 67 had the lowest effect of alkaline pH (1.33) of all the starches analyzed (**Table 2**).

Shear effect values among normal starches were lowest in maize and rice (1.13 and 0.94, respectively), highest in potato (0.52), and intermediate for cassava (ranging from 0.55 in MTAI 8 to 0.67 in MPER 183, **Table 2**). Waxy cassava and potato starches did not show much difference in shear effect compared with their normal counterparts. In the case of waxy maize, on the other hand, there was a drastic resistance reduction (from 1.13 to 0.35). The shear effect of COLFLO 67 (1.09) was similar to that of normal maize.

Pasting characteristics of the different starches analyzed are presented in **Table 3**, as well as in **Figures 2** and **3**. The lowest PT for normal starches was found in root and tuber starches. Cassava clones CM 523-7 and MTAI8 had the lowest values (63.3 and 63.7 °C, respectively), with slightly higher values in potato (65.2 °C) and the other cassava starches (64.8 °C in MPER 183 and 68.0 °C in Mutant 7). Cereals had the highest PT: 84.0 °C for rice and 89.0 °C for maize. PT in the case of waxy potato was about the same as for normal potato (65.9 vs 65.2 °C). Waxy cassava showed a slightly higher value (≈2 °C) compared with the normal versions (except for Mutant 7, which showed the highest PT among cassava starches). Waxy maize and COLFLO 67, on the other hand, had a drastic reduction in PT (from 89.0 to around 70 °C, **Table 3**).

As expected, PV of normal starches was highest in potato (2550 cP), lowest in maize (176 cP) and rice (343 cP), and intermediate in the different cassava starches (ranging from 876 to 1006 cP). The absence of amylose in the starch increased PV in cassava (to 1119 cP) and especially in maize (to 973 cP). Waxy rice did not show a very large increase in PV (498 cP compared with 343 cP).

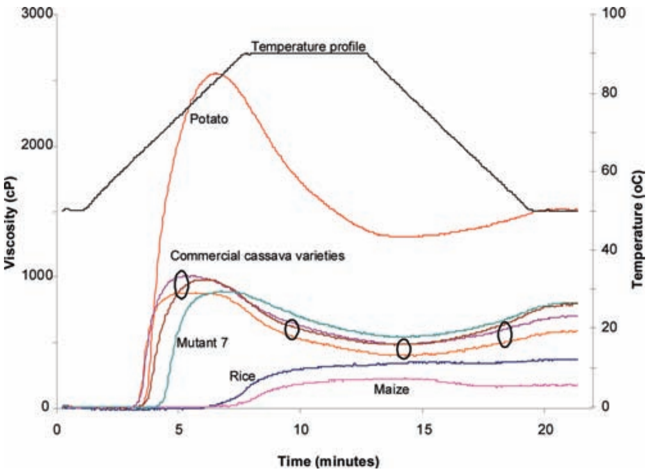

Figure 2. RVA amylograms (5% suspensions) of normal starches from different crops. Amylograms going through the rings in the plot belong to the three commercial cassava clones included in the study (CM523-7, MTAI 8, and MPER 183).

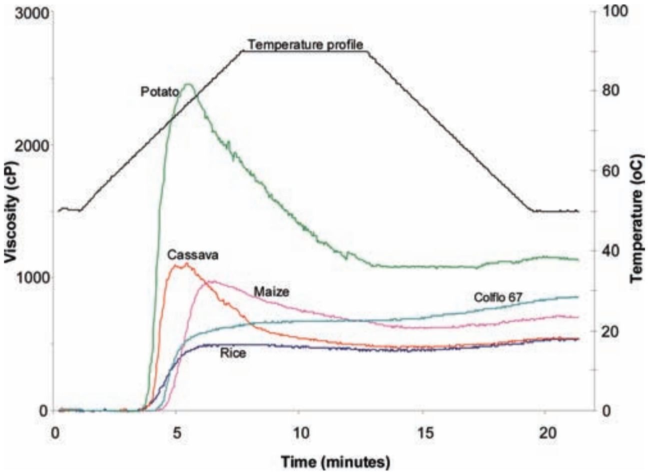

Figure 3. RVA amylograms (5% suspensions) of waxy starches from different crops. COLFLO 67 is a modified commercial waxy maize starch.

COLFLO 67 had a peculiar amylogram with a high heat and shear stability (**Figure 3**) continuously increasing viscosity. Therefore, it was not possible to determine PV, BD, SB, or CS values. A longer analysis may have shown better changes in viscosity. The RVA profile used, however, is the standard in many laboratories and facilitates comparisons between starches.

Table 3 also presents the results of BD, which was negative in maize (−30 cP), intermediate in normal cassava starches (ranging from 324 cP in Mutant 7 to 500 cP in CM 523-7), and highest in potato (1204 cP). BD in waxy maize starch was much higher than in its normal counterpart (307 vs −30 cP) and slightly higher in waxy versus normal cassava and potato starches. Potato showed the highest drop in viscosity (SB) among normal starches (−1082 cP), followed by cassava (values ranging from −157 to −364 cP) and then by maize (SB value of −15 cP). Mutant 7 showed a distinctive performance among nonwaxy cassava starches, with a small SB magnitude (−157 cP). The absence of amylose accentuated the SB value of different starches: for maize from −15 to −289 cP; for cassava from an average of −323 cP (excluding Mutant 7) to −595 cP; and in potato from −1082 to −1268 cP. Normal and waxy rice starches had positive SB values and were similar to each other.

The last column in **Table 3** summarizes the values for CS. Cereal starches had lower values. Normal maize starch had actually a negative value (−45 cP), whereas rice starch had a small positive one (34 cP). Normal starches from cassava and potato had values more or less similar with those from several cassava cultivars slightly higher than that of potato. Amylose-free starches had opposite effects on the CS of maize (which increased from −45 to 31 cP) and rice (increasing from 34 to 54 cP), compared with that of cassava and potato (which were reduced to 37 and 13 cP, respectively, from values > 100 cP).

The reliability of the information provided in **Table 3** is adequate, based on the standard deviation values provided. **Figure 2** helps in visualizing the main trends in viscosities of different normal starches: a very high PV for potato starch, low values for cereal starches, and intermediate performance for the cassava starches. The delayed gelatinization of Mutant 7 is also apparent. **Figure 3** illustrates how the recently discovered waxy starch mutation would offer a performance intermediate between waxy maize and waxy potato starches, with regard to PV.

Figure 4, panels **a** and **b**, present the values for SO and SW, respectively, estimated at three different temperatures. **Figure 4c** illustrates the Φ values for the starches analyzed. The coefficients of variations (*26*) for SO and SW were ≤6 (data not presented) except for SO at 60 °C (which for most cases was still too low for starch to start the absorption of water). This information is, therefore, considered to be reliable. Normal cereal starches (rice and maize) showed low SO at 60 and 75 °C but the highest SO at 90 °C. As expected, SO of waxy starches was generally lower than in their normal counterparts. However, in the case of waxy cassava SO increased, particularly at 90 °C. COLFLO 67 had a very low SO, which did not increase with temperature.

Normal maize starch had the lowest SW values regardless of the temperature (**Figure 4b**). At 60 °C normal potato starch had swelling values intermediate between those of maize and cassava. Differences in SW were generally not large. At 75 and 90 °C, however, SW were much higher in the waxy version of the starches of each crop, particularly in the cases of cassava and potato. SW for COLFLO 67 was always higher than for normal maize, particularly at 75 °C.

Φ for waxy potato starch at 90 °C reached the value of 1. There was no separation between pellet and supernatant after centrifugation of the gel (**Figure 4c**). To achieve separation it would have been necessary to reduce the concentration of the starch below 1%. This result suggests that SW of waxy potato at 90 °C could be even higher than the value presented in **Figure 4b**.

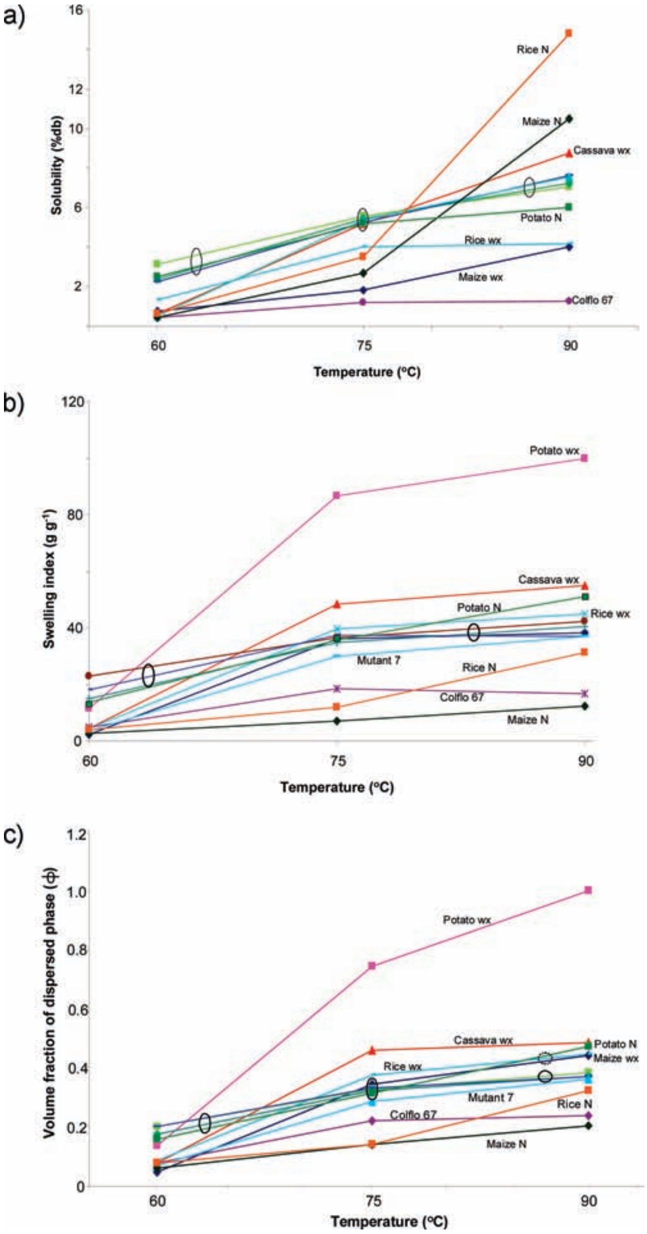

Figure 4. (**a**) Solubility at 60, 75, and 90 °C of the different types of starches analyzed in this study. The circles group the performance of the four nonwaxy cassava starches. (**b**) Swelling power at 60, 75, and 90 °C of the different types of starches analyzed in this study. (**c**) Volume fraction of the dispersed phase (Φ) at 60, 75, and 90 °C of the different types of starches analyzed in this study. The circles group nonwaxy cassava starches (except Mutant 7), nonwaxy potato, and waxy maize starches.

Syneresis in gels maintained in refrigerated conditions for up to 5 weeks is illustrated in **Figure 5**. The highest syneresis values were observed in gels from normal maize and potato starches and COLFLO 67. Cassava Mutant 7 showed the highest syneresis for a nonwaxy cassava starch, followed by MTAI 8 (but with considerably lower values). Gel from waxy potato had much lower syneresis than its normal counterpart, but it was still measurable. Gels from waxy maize, rice, and cassava as well as normal rice and cassava starches (CM 523-7 and MPER 183) had negligible levels of syneresis, showing a very stable behavior under storage at 4 °C.

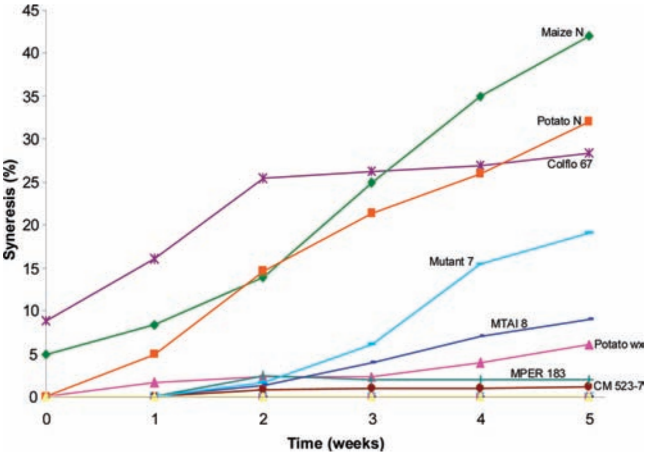

Figure 5. Refrigeration stability for up to 5 weeks of gels from different types (normal, waxy, or modified) of starches from different crops (maize, potato, rice, and cassava).

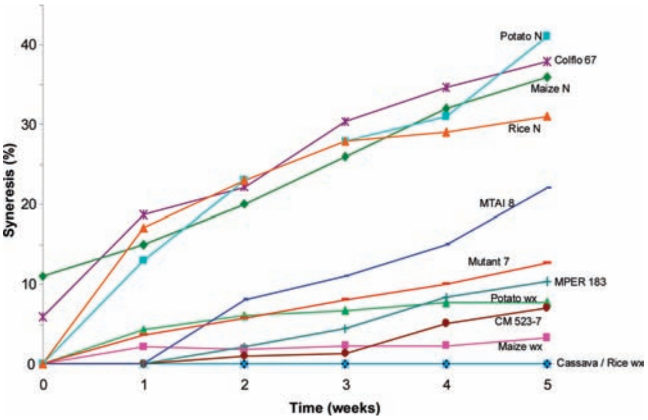

Figure 6. Freeze/thaw stability for up to 5 weeks of gels from different types (normal, waxy, or modified) of starches from different crops (maize, potato, rice, and cassava).

Syneresis, evaluated as a parameter for freeze/thaw stability, is illustrated in **Figure 6**. The largest values were observed in normal potato, maize, and rice starches and COLFLO 67, with magnitudes increasing consistently through time. There was an interesting variation in different nonwaxy starches from cassava with relatively high syneresis values for MTAI 8, followed by Mutant 7, MPER 183, and CM 523-7. Waxy potato and maize starches had significantly lower syneresis values compared with their normal counterparts and had a performance similar to those of several normal cassava starches. Waxy cassava and rice starches showed very interesting performances with no syneresis measurable up to 5 weeks under freezing conditions.

DISCUSSION

The main contribution of this study is to locate the performance of the recently discovered spontaneous mutation of amylose-free cassava starch (*11*) and to compare it with those of well-known normal and waxy starches.

Paste clarity of normal cassava was intermediate between those of the cereals (maize and rice) and potato (**Table 1**). Paste clarity was low in COLFLO 67, higher in normal, and highest in waxy maize. Paste clarities of waxy starches were higher than their respective normal counterparts, as reported in the literature (*2*, *27*−*29*). The low λ_{max} value for normal rice was also reported by Tetchi et al. (*19*). Acid pH resistance was lower in waxy than in normal maize. There seems to be no major change in the shear effect of waxy cassava starch compared with its normal counterpart (**Table 2**).

The general shape of the amylogram of the amylose-free cassava mutant would fall between those of waxy maize and waxy or normal potato starches (**Figures 2** and **3**). In the case of cassava, PV tends to increase but not as much as the increase observed between normal and waxy maize starch. In a comparison between transgenic waxy cassava and wild type cassava starch (*12*) it was found that the latter had a higher PV. We have found the opposite in our study. PV in normal maize and rice was lower than in the respective versions, as reported in the literature (*2*, *18*, *29*, *30*).

Waxy and normal potato starches did not show much difference in PT. However, in the case of maize, PT was lower in waxy than in normal starch. In the case of cassava, there was a slight increase in PT of waxy compared with the nonwaxy cassava samples. PT of Mutant 7 was the highest of the studied root and tuber starches. SB was smaller in waxy starches compared with their normal counterparts, as already reported in the literature (*29*).

SO values at 60 °C were very low, in agreement with the information from **Table 3** regarding pasting temperatures > 60 °C. In general, SO values in waxy starches increased compared with their normal counterparts as the final temperature was increased (**Figure 4a**). Waxy cassava starch had lower SO at 60 °C but higher SO at 90 °C, compared with the normal cassava starches. Trends in the case of potato starch could not be clearly established because of difficulties in separating the different phases at 90 °C. SO was measured under the shearing conditions of the RVA, which offers the advantage of a uniform paste but may result in slight overestimations.

According to the literature, swelling in normal maize increases with temperature but the rate of change was much higher in waxy maize (*2*, *18*). Similarly, swelling of waxy potato starch increased much more rapidly with higher temperatures than normal potato starch (*17*). We found similar trends in our study. Normal cassava starch showed the highest SW at 90 °C. It had values similar to those of normal potato at 75 °C, lower values at 90 °C, and higher values at 60 °C. In this study, SW values of normal cassava agree with those reported elsewhere (*31*).

Syneresis in normal cassava was low, but it was much lower in waxy cassava (**Figures 5** and **6**). This is particularly relevant in the case of freeze/thaw analyses in which waxy cassava and rice were the only starches with total absence of syneresis until the end of the experiment. This finding agrees with results on transgenic waxy cassava starch already reported (*12*) and highlights an advantage of using waxy cassava in comparison with other waxy and nonwaxy starches. It has been reported that even after three freeze/thaw cycles the only starch showing no syneresis was the amylose-free transgenic cassava starch (*12*). The low syneresis value observed for normal rice in **Figure 5** may be due to the low amylose content of the particular rice starch analyzed in this study.

Pasting properties of the waxy cassava starch reported in this study have slight differences with those in the original paper (*11*). This in part is due to differences in the sample arising from different environmental conditions in which AM 206-5 was grown.

As stated by BeMiller (*32*), the potential for new commercial starches can be even greater when biological and chemical modifications are combined. BeMiller also stated that after thousands of studies, starch remains a beautifully mysterious substance. Our study provides insight into several distinctive properties of the recently discovered spontaneous mutation of an amylose-free cassava starch. This, in turn, will define the applications in which it could potentially offer advantages over normal cassava starch or as an alternative to amylose-free starches from other crops.

ABBREVIATIONS USED

PT, pasting temperature; PV, peak viscosity; HPV, hot paste viscosity; CPV, cool paste viscosity; BD, breakdown; SB, setback; CS, consistency; SW, swelling power; SO, solubility patterns.

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