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Research Article

Physicochemical and Structural Characterization of Yam (*Dioscorea rotundata*) Mucilage

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Abstract: The objective of the present study was to characterize physicochemical and structural mucilage of yam (*Dioscorea rotundata*). The extraction of the mucilage was done by the continuous bubbling method, whose basis is flotation, then proximal composition, physicochemical properties (pH, acidity, dietary fiber, starch content and color) and structural characteristics (differential scanning calorimetry, Infrared spectroscopy and thermogravimetric analysis). The proximal composition shows that the yam mucilage is a product with high protein content (27.18), minerals (18.94%) and low lipid content (1.06%), it shows physicochemical properties such as pH (6.63), acidity (0.52%) and starch content (3.99%) low average values, a high brightness percentage (L* = 82.37), a high dietary fiber content (9.73%), a glass transition of 118.29°C with a multi-stage decomposition that allows its use up to temperatures below 220°C and has as its characteristic trademark a stretching C = O present in the peptide bond R-CO-N corresponding to the amide group, all these results reflect that the yam mucilage can be used in food matrices with potentialities as a functional ingredient.

Keywords: Dietary fiber, functional ingredient, glass transition, minerals, protein

INTRODUCTION

Globally, in 2014, roots and tubers were the eighth most demanded product. Among these is the yam, which in recent years has become very important, mainly in areas of the southern hemisphere such as Africa with a participation of 96.6%, South America and Caribbean islands with 2.5% and Oceania with 0.6% (Awoyale *et al*., 2016; FAOSTAT, 2016).

The main component of yams is starch, in which studies have been centralized due to their multiple industrial uses. In the extraction of the starch from the yam, its retrieval is made difficult by the presence of components such as mucilage, which, in addition, to delaying the sedimentation process and lowering the quality of the same, have been tested for different chemical compounds for extraction, such as: ammonia solutions, pectinases, oxalic acid-sodium oxalate solution and sodium hydroxide (Daiuto *et al*., 2005; Moorthy, 1991, 2002). The extraction of starch with chemical compounds makes its use in the food industry difficult, in addition they obtain low yields of starch without recovery of mucilage; as a result, the bubbling separation process is an alternative, since yields are improved, chemical reagents are not used and percentages of mucilage are recovered from 1.66 to 8% w/w, values that are similar to that obtained by other techniques (Daiuto *et al*., 2005; Moorthy, 1991, 2002; Pérez *et al*., 2015; Tavares *et al*., 2011).

The mucilaginous material has potential as an ingredient for functional foods because it contains saponins such as diosgenin, a precursor of progesterone, which has an immunostimulating, hypolipidemic and immunological effect (Boban *et al*., 2006; Jang *et al*., 2007; Shang *et al*., 2007). At the same time, purified mucilages from tubers exhibit antioxidant properties because of their ability to attract free radicals and are important in the food industry as a food additive because of their good chemical properties as a thickener, stabilizer and emulsifier (Andrade *et al*., 2015; Behbahani *et al*., 2017; Contreras-Pacheco *et al*., 2013; Hou *et al*., 2001, 2002; Tavares *et al*., 2011; Zeng and Lai, 2014). Therefore, the objective of the present study was to characterize physicochemical and structural mucilage of yam (*Dioscorea rotundata*).

MATERIALS AND METHODS

Raw material and extraction of yam mucilage: The vegetable material used in the research corresponds to the variety of white yam (*Dioscorea rotundata*), was supplied by local producers in the city of Sincelejo

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(Colombia). For the extraction of the mucilage the bubbling method was used, whose basis is the flotation by the presence of air. Pilot-scale continuous equipment was used, operating at a water suspension ratio of yam to water 1 to 8. The mucilage was dried in a Freezone vacuum lyophilizer (Labconco, USA) for 60 h, for that purpose it was first frozen at -50°C, after drying it was macerated for later use (Tavares *et al*., 2011; Pérez *et al*., 2015).

Determination of the physicochemical properties of the yam mucilage: The proximal composition of the yam mucilage was developed according to the AOAC methodology (2012), determining the moisture content (AOAC 925.10), ash (AOAC 923.03), fat (AOAC 920.85), crude protein (AOAC 920.87), crude fiber (AOAC 920.86) and total carbohydrates by difference, according to Eq. (1) (Morillas-Ruiz and Delgado-Alarcón, 2002):

$$\%\text{total carbohydrates} = 100-(\%\text{moisture}+\%\text{protein}+\%\text{fat}+\%\text{ash}+\%\text{fiber}) \quad (1)$$

For the physicochemical characterization of the yam mucilage, the following analyses were carried out: pH: by method 981.12 (AOAC, 2012), titratable acidity: by method 942.15 (AOAC, 2012) expressed in allantoic acid, dietary fiber: insoluble by method 991.42 and soluble according to method 993.19 (AOAC, 2012), starch: was analyzed by enzymatic hydrolysis determining the concentration of reducing sugars using the DNS method, the standard curve was performed with glucose to determine the starch concentration by interpolation (Miller, 1959).

Color: The instrumental measurement of lyophilized yam mucilage color was carried out with a Colorimeter Minolta CR400 (Minolta, USA) according to the methodology proposed by Fernández-Vazquez *et al.* (2013) and the results were expressed in accordance with the CIELAB system with reference to illuminant D65 and a visual angle of 10°. The color measurements were expressed in terms of lightness L* (L* = 0 for black and L* = 100 for white) and the chromaticity parameters a* (-a* = greenness and +a* = redness) and b*(-b* = blueness and +b* = yellowness). From these parameters, chroma (C_{ab}*) and hue (h_{ab}), were calculated according to Eq. (2) and (3), respectively. The colorimeter was calibrated with a standard white plate (L* = 94.8, a* = -0.78 and b* = 1.43) before each series of measurements:

$$C_{ab}^{*} = \sqrt{(a^{*})^{2} + (b^{*})^{2}} \quad (2)$$

$$h_{ab}^{*} = \arctan \frac{b^{*}}{a^{*}} \quad (3)$$

Euclidean distance between two points in the three-dimensional space defined by L*, a* and b* were used for calculating color differences, ΔE_{ab}* Eq. (4):

$$\Delta E_{ab}^{*} = \sqrt{(\Delta L^{*})^{2} + (\Delta a^{*})^{2} + (\Delta b^{*})^{2}} \quad (4)$$

Determination of the structural properties of yam mucilage: The following analyzes were applied to the yam mucilage for its structural characterization.

Differential Scanning Calorimetry (DSC): Thermal properties of yam mucilage were determined by using differential scanning calorimetry DSC Q2000 (TA Instruments, USA). Approximately 8.3 mg of mucilage was used, initiating the process at a temperature of 25°C, which was placed in a hermetically sealed capsule and analyzed in a temperature range of -90 to 200°C with a heating and cooling ramp of 5°C min^{-1} and a modulation of 60s, according to the methodology described by Zhang *et al.* (1997) modified.

Fourier-Transform Infrared (FTIR) spectroscopy: The IR spectra of the yam mucilage was determined using an infrared spectrometer (FTIR) (Digilab Excalibur FTS 3000 Series, United States) with DTGS detector in the spectral range of 470 to 4000 cm^{-1} at a resolution of 4 cm^{-1}. Samples were analyzed by transmission into 7 mm diameter KBr tablets and were prepared by a mucilage mixture of yam: KBr in a ratio of 1:5 (Razavi *et al*., 2016).

Thermogravimetric Analysis (TGA): 10.320 mg of sample were introduced into an alumina crucible and subjected to heating at 30 to 1000°C with a linear heating rate of 10°C min^{-1}. The study was performed under a Nitrogen atmosphere at a flow rate of approximately 50 mL*min^{-1}. At the end of the test, the residue was discarded. The equipment was initially calibrated with a sample of Indio*Aluminum^{-1} (Martin *et al*., 2017).

RESULTS AND DISCUSSION

Proximal composition of yam mucilage: Table 1 shows the chemical composition of lyophilized yam mucilage. It has a lower carbohydrate content (47.19%) than that obtained for other lyophilized mucilages of yams, 65% (Contado *et al*., 2009; Tavares *et al*., 2011). This is due to the efficiency of the mucilage starch separation process, by the continuous bubbling method. The carbohydrate content present in the mucilage is polymers that are composed mainly of a complex network of carbohydrates that give it part of its characteristics. It is worth noting that the high protein

Table 1: Chemical composition of yam mucilage

Component	Content (%, d.b.)
Moisture	3.29±0.41
Protein	27.18±1.46
Crude fiber	1.06±0.13
Fat	2.34±0.00
Ash	18.94±0.02
Carbohydrates	47.19

Table 2: Physicochemical properties of yam mucilage

Properties	Value
pH	6.63±0.16
Titratable acidity (% allantoic acid)	0.52±0.07
Starch (%, d.b.)	3.99±0.05
Total dietary fiber (TDF) (%, d.b.)	9.73±0.36
Insoluble dietary fiber (IDF) (%, d.b.)	4.53±0.08
Soluble dietary fiber (SDF) (%, d.b.)	5.2±0.43
SDF/IDF	1.15

Table 3: Color parameters of the yam mucilage

Parameters	Value
L*	82.37±0.40
a*	0.91±0.02
b*	15.71±1.24
h^*_{ab}	86.69±0.22
C^*_{ab}	15.41±1.82
ΔE*	19.01±1.16

L* = lightness; a* = chromaticity parameter (green-red); b* = chromaticity parameter (blue-yellow); h^*_{ab} = hue; C^*_{ab} = chroma; ΔE* = color differences

content (27.18%), which is beneficial because they may contribute to the emulsifying properties of the mucilage, being non-polar molecules and which are covalently bound to carbohydrates (McClements and Gumus, 2016).

On the other hand, the high content of ash and, therefore, of minerals, gives the mucilage importance as a possible source of these micronutrients (Montero Quintero *et al*., 2015). High amounts of minerals such as calcium, iron and phosphorus have been reported in species of the *Dioscoreagenus spp.* (González, 2012). As well as micronutrients such as potassium, magnesium, sulfur, copper, manganese and zinc for yam mucilages (Tavares *et al*., 2011) and leaves of gooseberry (Martin*et al*., 2017).

Physicochemical properties of yam mucilage: The physicochemical properties of lyophilized yam mucilage are shown in Table 2. These values indicate that the mucilage has a pH close to neutrality, similar values have been reported for nopal mucilage, 5.5-6 (Contreras-Padilla *et al*., 2016), Chinese yam (*Dioscorea opposita Thunb*) 6.96 (Ma *et al*., 2017) and lyophilized yam mucilage, 6.3 (Tavares *et al*., 2011). Also, it presents a small proportion of free acids reflected in titratable acidity (0.52), an important feature because its repercussions on sensorial characteristics would be few when incorporated into food matrices. At the same time, a mucilage with low starch content (3.99%) was obtained as compared to other mucilages of yams, which are around 50%, (Contado *et al*., 2009; Tavares *et al*., 2011), this may be due to the difference of the extraction methods used. It should be noted that the bubbling method is more efficient to separate the mucilage from the starch because it allows a better fixation of carbohydrates and soluble proteins in the foam released in the process (Pérez *et al*., 2015).

The mucilage of yam had a TDF content of 9.73%, which positions it as a source of fiber (Sánchez, 2016). Its IDF content is similar to that reported for mucilages obtained from different nopal varieties: *Opuntia tomentosa* (4.03%), *Opuntia hyptiacantha* (5.5%), *Opuntia ficus-indica* (5.43%). The values of SDF are lower (5.2%) than those reported for the different varieties of nopal, 51.79-67.51% (Rodríguez-Gonzalez *et al*., 2014), which may be due to the characterization of the nopal mucilage the study reports the carbohydrate as SDF. However, fiber contents were higher than those of *Ziziphus mauritiana Lam* mucilage, which presented values of 4.03% IDF and 2.82% SDF (Thanatcha and Pranee, 2011). High TDF values in food products are beginning to gain a lot of attention worldwide because of their importance in human health, mainly attributed positive effects on colon cancer, absorption of bile acids, cholesterol and blood glucose, therefore, studies have been emphasized in search of new economic sources of TDF (Aboulfazli *et al*., 2015; Matos and Chambilla, 2010; Sangeethapriya and Siddhuraju, 2014; Soukoulis *et al*., 2014).

Table 3 shows the results of the color parameters of lyophilized yam mucilage. Which have a color with high percentage of brightness and tend to be clear (L* = 82.37), similar results were found in nopal mucilage (*Opuntia Ficus*) with values between 73-83 (Contreras-Padilla *et al*., 2016), badari mucilage (*Ziziphus mauritiana Lam*) 81.84 and commercial stabilizers such as guar gum (84.14) and xanthan gum (90.76) (Thanatcha and Pranee, 2011), but superior to chia (*Salvia hispanica L.*) mucilage with L* = 6.22 (De la Paz Salgado-Cruz *et al*., 2013). The gloss similarity of lyophilized yam mucilage with guar gum and xanthan gum allows its use as a food additive without affecting sensory properties in food matrices (Thanatcha and Pranee, 2011).

The yam mucilage presents a deviation from the achromatic point corresponding to the red color (a* = 0.91) lower than that found in chia mucilage (a* = 1.77). Therefore, the latter tends more to red color (De la Paz Salgado-Cruz *et al*., 2013), but slightly higher than guar gum (-0.75) and xanthan gum (-0.72) (Thanatcha and Pranee, 2011). On the other hand, the yam mucilage has a deviation towards yellow (b* = 15.71), similar to that of *Ziziphus mauritiana Lam* (17.58), guar gum (12.56) and xanthan gum (9.95), (Thanatcha and Pranee, 2011), but higher than that of chia mucilage with b* = 6.07 (De la Paz Salgado-Cruz *et al*., 2013). In addition, it has a low hue (h* = 86.69) near 90° whereby it tends to yellow and has a low color saturation (C* = 15.41) which tends to gray. The values of the parameter deviation of the achromatic point, saturation and hue of the yam mucilage may be due to the content of impurities as natural pigments (chlorophyll and tannins) present in the plants (Pérez *et al*., 2008) and temperatures of lyophilization drying used in the mucilages (Campos *et al*., 2016).

The color variation of the lyophilized yam mucilage (ΔE*) was 19.01 units concerning to the standard, which is higher than the minimum difference

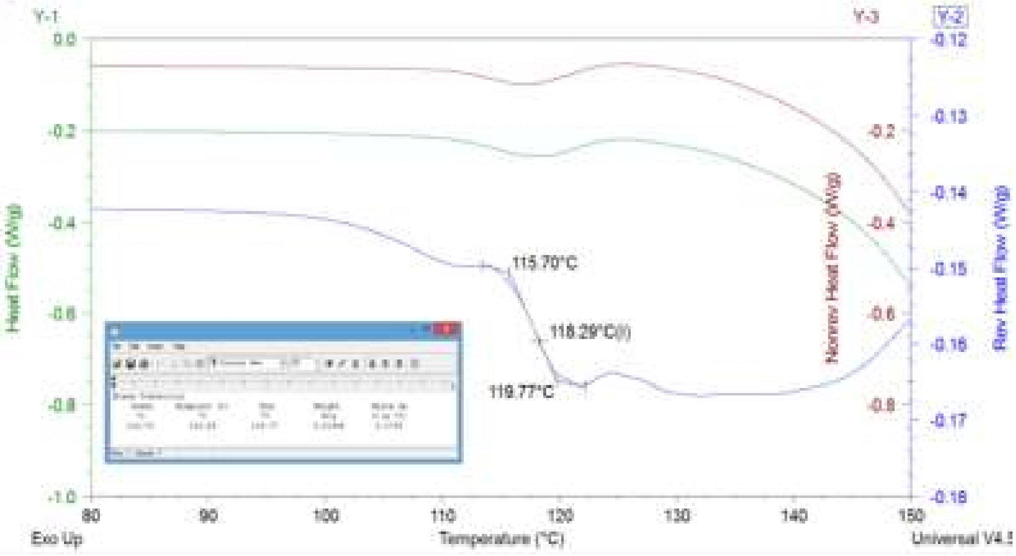

Fig. 1: Differential scanning calorimetry of the mucilage of lyophilized yam

that the average observer can perceive (5 or 6), which may affect the color of the product in which this mucilage is applied. It should be noted that the sensory attributes are the first to be appreciated and the consumers relate it to the quality of the product (Mathias and Ah, 2014; Wrolstad and Smith, 2010), so it is necessary to make measurements of the color parameters of the food matrix with different proportions of the mucilage to determine its possible effect on the visual perception of the consumer.

Structural properties of yam mucilage: In Fig. 1, the mean value at inflection reflecting a glass transition (Tg) of lyophilized yolk mucilage at 118.29°C and a specific heat of 0.1733 J/g *°C is shown. These values manifest the change of structure of the polymer material in the endothermic process. The Tg of lyophilized yam mucilage is superior to that of mucilage of other species such as *jaracatia*, 95°C (Heidemann *et al*., 2014), yam, 83.34°C (Tavares *et al*., 2011), chía, 48.79°C (Velázquez-Guitierrez *et al*., 2015), Tamarind seeds (*Tamarindus indica L.*) 57.93°C (Alpizar-Reyes *et al*., 2017) and Barbary fig (*Opuntia ficus-indica*) 45°C (León-Martinez *et al*., 2010), indicating the amorphous nature of lyophilized yam mucilage, whereby it presents a polymer chain more difficult to disintegrate with the thermal processes. The importance of knowing the values of heat capacity and the amorphous transition of polymers is that they are necessary to obtain qualitative and quantitative information about the physical and chemical changes that involve endothermic processes (heat absorption), exothermic (heat loss) or changes in heat capacity (Alpizar-Reyes *et al*., 2017; Magon *et al*., 2013; Tavares *et al*., 2011).

The FTIR spectra of mucilage and native yam starch are shown in Fig. 2, where absorbance (%) was plotted with respect to wavelength and characteristic peaks of mucilage are observed at 3317 cm^{-1} and starch at 3418 cm^{-1}, which correspond to the -OH groups of alcohols, phenol, water and carboxylic acids involved in the hydrogen bonds of the molecules. It should be noted that at 2930 cm^{-1} there is a C-H stretching of the alkyl group (Ahuja *et al*., 2011; Behbahani *et al*., 2017; Heidemann *et al*., 2014; Tavares *et al*., 2011) and bands occur in the region between 800 and 1200 cm^{-1}, which could be dominated by superimposed annular vibrations with stretching vibrations of the C-OH and CO groups of the alcohol and the vibration of the COC glycosidic bond (Ahuja *et al*., 2011; Li *et al*., 2016; Razavi *et al*., 2016).

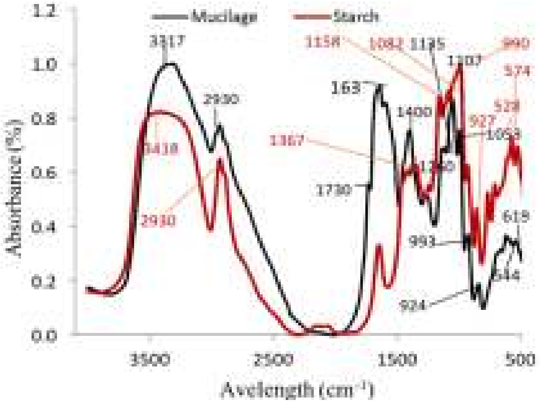

Fig. 2: Infrared spectra of lyophilized yam mucilage and native starch of yam

The most significant difference between infrared spectra of starch and yam mucilage corresponds to the peak at 1638 cm^{-1}, which is far more intense in the mucilage spectrum. This peak corresponds to the C=O stretching present in the R-CO-N peptide bond corresponding to the amide group (Faccio *et al*., 2015;

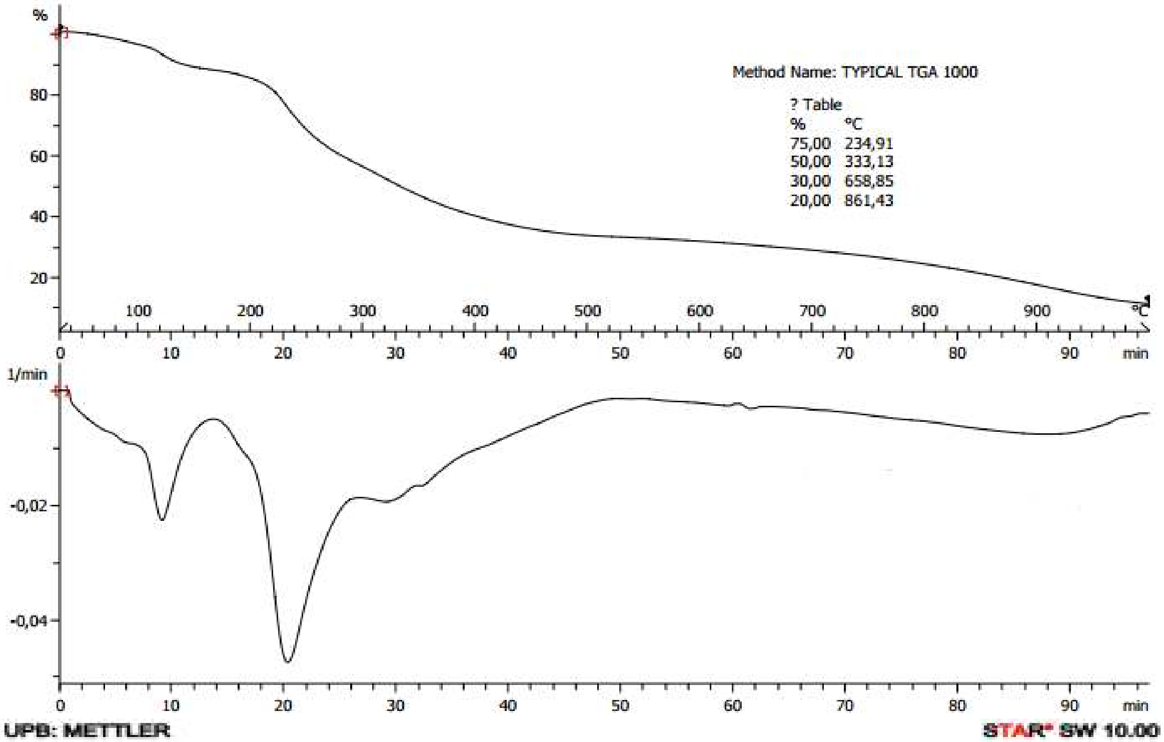

Fig. 3: Thermogravimetric curve of the lyophilized yam mucilage (top) and derivative curve (bottom)

Larkin, 2011) and is related to the high protein content (27.18%) found in yam mucilage (Table 1). Similar peak (1637 cm^{-1}) reports Ma *et al*. (2017) for Chinese yam (*Dioscorea opposita Thunb*). The importance of FTIR spectrometry is because it is widely used to characterize the molecular and material structure of natural polymers and often this characterization results in the identification of functional groups and modes of their binding to the polysaccharide backbone (Singh and Bothara, 2014).

The thermogravimetric and derivative curve of lyophilized yam mucilage indicate a multi-stage decomposition (Fig. 3). The first mass loss of about 5% occurred between 0 and 120°C, which is due to the loss of volatile compounds, mainly corresponding to the evaporation of the water contained in the polymer (first peak of the derivative curve). Then in the area between 220-280°C there is a mass loss of 25%, due to the decomposition of low molecular weight peptides and carbohydrates (second derivative curve peak). The final mass loss is attributed to the degradation of the high molecular weight polysaccharides, such as proteins and lipids, among other organic compounds (330-500°C) and after to the complete decomposition of the material, leaving the organic matter (minerals). These results are close to those reported for yam mucilage (Tavares *et al*., 2011), nopal (Han *et al*., 2016), *Pereskia aculeata* leaves (Martin *et al*., 2017) and *jaracatiá* (Faccio *et al*., 2015), reporting decomposition in different stages, highlighting the three regions mentioned. Therefore, concerning to thermal stability, the use of lyophilized yam mucilage is not possible at temperatures more than of 220°C, because processes occurring above this temperature may cause distortion and degradation of the polymer, causing it to lose its applicability and functional properties (Granados, 2015). However, in the food industry the additives are subjected to temperatures below 220°C; For example, in the Ultra High Temperature (UHT) process of the dairy industry, a temperature of 135°C is used.

CONCLUSION

The lyophilized mucilage of yam *(Dioscorea rotundata*) presents high protein content (27.18%), ashes (18.94%) and fibers (9.73%), which potentiates it as a functional ingredient. Also, its acidity, pH and color similar to other hydrocolloids used in the industry, would facilitate the inclusion in different food matrices. Finally, it has a higher glass transition temperature than other hydrocolloids, such as gums and starch and has the highest percentage of decomposition at temperatures more than 220°C, which allows its use in different products and processing conditions.

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CONFLICT OF INTEREST

The authors of this study do not have conflict of interest to report.

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