

COMPARISON BETWEEN HIGH ISOSTATIC PRESSURE WITH HEATING AND CONTROLLED MATURATION ON STRUCTURAL MODIFICATION OF CASHEW TREE GUM (*Anacardium occidentale* L.)

ABSTRACT

Polysaccharides are frequently modified by technological processes. The high isostatic pressure (HIP) and controlled maturation (CM) are physical methods, where pressure and temperature are the main mechanisms of alteration, respectively. This work evaluated the effect of HIP with heating (HIPH) and CM on chemical and physical aspects of cashew tree gum (CG). The exudate was modified by HIP at 600 MPa and 60 °C for 6 or 12 h, and CM at 110 °C for 24 or 48 h. The CG was isolated and analyzed on monosaccharide composition; linkage analysis; MW; flow behavior index; and, differential scattering calorimetry. Both processes increased the MW and reduced the apparent viscosity of the gum. HIPH decreased the consistency index in 70% and the controlled maturation in 10-30%, but it did not promote variation on flow behavior index (n), while, HIPH approximated the n value to a Newtonian fluid. Both processes reduced the melting enthalpy. Thus, both HIPH and controlled maturation produced physical and chemical modifications of CG.

INTRODUCTION

Cashew tree gum (CG) is a branched acid heteropolysaccharide obtained from the crude exudate of cashew tree (*Anacardium occidentale* L.)¹. This biopolymer emerges as a new profitable product to the cashew tree culture, which has cashew nuts and peduncle (cashew) as commercial products, especially in the off-season. The applications attributed to CG are vast and can be used in the manufacture of medications, cosmetics, and food².

CG has a branched structure whose molar mass distribution reveals two majority peaks (28000 and 67000 g/mol), and one peak greater than 500000 g/mol that was attributed to the protein-polysaccharide complex³. In terms of centesimal composition, CG is predominately carbohydrate (97.7-98.3%)¹. The monosaccharide composition is mainly composed of galactose (73.9%), glucose (12.9%), and arabinose⁴. Besides that, CG contains approximately 1% protein which enables its use as an emulsifier¹.

To expand its functionality, polysaccharides undergo diverse modification processes⁵. Some physical methods used are: high isostatic pressure (HIP)⁶, controlled maturation (CM)⁷, ultrasound⁸, and dynamic high pressure^{4,9}. HIP emerged as a non-thermal pasteurization or sterilization process for food production. The benefits of using pressure rather than heat for microbiological safety are related to reduced sensory and nutritional modification of foods¹⁰. This pressure is transmitted uniformly throughout the food¹¹. HIP equipment allows pressure, temperature, and time control. Although the method has emerged as a non-thermal processing, it is often used for mild heating (50-70 °C) to optimize results¹². CM is a patented process that uses high temperatures for a certain period under controlled conditions. The process is applied to a crude plant exudate and has as objective the polymerization of the heteropolysaccharide chain, mainly the increase of the protein-polysaccharide fraction, responsible for the exudate's emulsifying property. So far, this process has only been used in plant exudates of the genus *Acacia*⁷, and in corn fiber gum¹³. In gum arabic, CM intensifies the union of the protein fraction

with the arabinose and galactose monosaccharides, forming the arabinogalactan-protein complex, a substance that gives emulsifying capacity to the gum⁷.

As with gum arabic, CG also comes from a plant exudate and contains the monosaccharides arabinose and galactose, in addition to protein fraction, which boosted the investigation of CM in CG. Furthermore, the application of pressure favors the occurrence of chemical and biochemical reactions, therefore, the use of HIP combined with heating (HIPH) can also be an interesting option in the formation of the protein-polysaccharide complex.

OBJECTIVE

This work evaluated the effect of HIPH and CM on the chemical and physical characteristics of CG.

METHODOLOGY

The crude cashew tree exudate was donated by EMBRAPA Tropical Agroindustry (Brazil). Initially, the raw exudate particles from cashew tree were placed in a polyethylene terephthalate bottle with a lid and stirred manually to reduce their size. Then, the exudate was sieved through 3.5-mesh (5.66 mm) and 10-mesh (2 mm) sieves. The material used for the modification processes was the retentate from the 10-mesh sieve, and the permeate from the 3.5-mesh sieve, covering the size range from 2 to 5.66 mm. Subsequently, the exudate was dried in an oven with forced air circulation at 60 °C for 23 h (time in which the samples, $n = 30$, presented constant mass) (data not shown). The loss of water in the samples was around $2.65 \pm 0.08\%$.

To the HIPH process, the raw exudate samples were packed in flexible plastic packages (LDPE-Nylon-LDPE, 16 μm thick - TecMaq, Brazil) under vacuum and later processed in HIP equipment at 60 °C, 600 MPa for 6 h and 12 h. After that, the CG was isolated. The CM process was selected based on a patent described by Al-Assaf et al.⁷. Samples of cashew tree exudate were placed in aluminum trays and put in an oven at 110 °C for 24 h and 48 h. After that, the CG was isolated. The isolation process of CG, determination of MW, rheology behavior, monosaccharide composition, glycosyl-linkage analysis, uronic acid sugars and the calorimetric analysis of the modified and control samples were realized according to the methodology described by Porto et al.⁴. The similarities and differences between the results were subjected to analysis of variance (ANOVA) and Tukey's test using the Statistica 7 software (Statsoft, USA).

RESULTS AND DISCUSSION

Table 1 shows the MW results of CG: a) without processing (control); b) processed by HIPH for 6 and 12 h; and, c) with CM for 24 and 48 h.

All chromatograms exhibited two main peaks and traces of larger and smaller peaks. There was an increase in the size of the CG fractions when it was processed by both HIPH and CM ($p < 0.05$), with no difference between the processes ($p > 0.05$), represented by peak 1. There was a decrease ($p < 0.05$) in the molecular weight of CG's peak 2 when it was processed by HIPH, and a higher reduction when the gum was processed by CM ($p < 0.05$). Both processes promoted an increase in the concentration of the 1 \rightarrow 2-linked of arabinose and a reduction in the glucose terminal linkage. However, it was the CM process that

promoted most changes in the structure of the gum, as it reduced the concentration of the terminal and 1→3,6-linked of galactose and increased the number of 1→6-linked.

Table 1.: Molecular weight (Da) and monosaccharides linkage (mol%) of CG processed by HIPH at 100 and 600 MPa for 6 and 12 h and by CM at 110 °C for 24 and 48 h.

	Molecular weight (Da)				
	Control	HIPH		Controlled maturation	
		6 h	12 h	24 h	48 h
Peak 2	4.34 x 10 ⁴ A	4.38 x 10 ⁴ B	4.38 x 10 ⁴ B	4.38 x 10 ⁴ B	4.37 x 10 ⁴ AB
Peak 3	2.08 x 10 ⁴ A	2.07 x 10 ⁴ A	2.07 x 10 ⁴ A	2.05 x 10 ⁴ A	2.05 x 10 ⁴ A
	Monosaccharides linkage (mol%)				
	Control	HIPH		Controlled maturation	
		6 h	12 h	24 h	48 h
Terminal-L-arabinofuranosyl→	2.74 ± 0.53 ^A	2.34 ± 0.19 ^A	1.95 ± 0.07 ^A	2.34 ± 0.37 ^A	2.81 ± 0.38 ^A
1→2-L-arabinofuranosyl	0.39 ± 0.06 ^A	0.96 ± 0.18 ^B	1.08 ± 0.10 ^B	0.82 ± 0.14 ^B	0.73 ± 0.14 ^{AB}
1→3-L-arabinofuranosil	1.12 ± 0.05 ^A	0.95 ± 0.15 ^{AB}	1.23 ± 0.10 ^A	1.09 ± 0.12 ^A	0.71 ± 0.08 ^B
Terminal-D- glucopyranosyl→	12.93 ± 0.96 ^A	5.74 ± 0.68 ^{BC}	6.67 ± 0.07 ^B	11.21 ± 1.24 ^A	4.00 ± 0.81 ^C
1→3-D-glucopyranosyl	ND*	7.20 ^{**}	6.26 ^{**}	1.72 ± 0.17	8.92 ^{**}
Terminal-D- galactopyranosyl→	15.53 ± 0.12 ^A	15.60 ± 1.37 ^A	15.27 ± 0.46 ^A	12.06 ± 0.93 ^B	11.58 ± 0.47 ^B
1→3-D-galactopyranosyl	6.50 ± 0.74 ^A	.68 ± 0.93 ^A	7.84 ± 1.03 ^A	6.16 ± 1.22 ^A	6.34 ± 1.22 ^A
1→4-D-galactopyranosyl	9.04 ± 0.87 ^{ABC}	10.44 ± 1.04 ^{BC}	10.74 ± 0.96 ^C	8.38 ± 0.28 ^{AB}	8.23 ± 0.54 ^A
1→6-D-galactopyranosyl	20.63 ± 2.77 ^A	18.75 ± 1.21 ^A	16.94 ± 1.20 ^A	29.55 ± 2.46 ^B	31.43 ± 1.02 ^B
1→3,6-D-galactopyranosyl	22.21 ± 0.96 ^A	22.44 ± 1.39 ^A	23.12 ± 0.72 ^A	17.75 ± 0.26 ^B	16.50 ± 1.08 ^B
2,3,4→6-deoxi- mannoypyranosyl	2.31 ± 0.34	2.31 ^{**}	2.31 ^{**}	ND*	ND*

ND = non-detected.

Figure 1 shows the flow curves obtained for unprocessed CG (control) and processed CG treated with HIPH at 600 MPa and 60 °C for 6 and 12 h, and treated with CM at 110 °C for 24 and 48 h at 10% concentration (w/v) and shear rates ranging from 0.1 to 300 s⁻¹. It can be observed that the shear stresses in the processed samples were lower than in the control sample, considering the same shear rate. The tension reduction probably occurs because the gum undergoes changes in its conformation⁵.

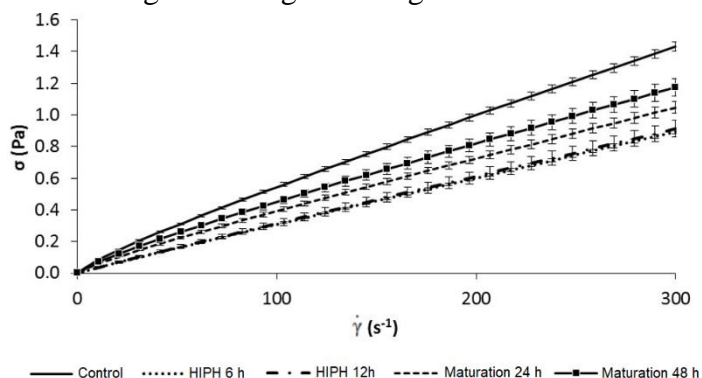


Figure 1.: Flow behavior (25 °C) of CG solutions (10% w/v): unprocessed (Control) and processed by HIPH at 600 MPa and 60 °C for 6 h (HIPH 6 h) and 12 h (HIPH 12 h) and processed by CM at 110 °C for 24 h (Maturation 24 h) and 48 h (Maturation 48 h). σ and $\dot{\gamma}$ are shear stress and shear rate, respectively.

Table 2 presents the results of the indexes of consistency (k) and flow behavior (n) of CG (10% w/v) control, processed by HIPH at 600 MPa (60 °C) for 6 (HIPH 6 h) and 12 h

(HIPH 12 h), and processed by CM at 110 °C for 24 (Maturation 24 h) and 48 h (Maturation 48 h).

Table 2.: Indexes of flow consistency (k) and flow behavior (n) of CG (10% w/v) control (unprocessed), processed by HIPH at 600 MPa and 60 °C for 6 (HIPH 6 h) and 12 h (HIPH 12 h), and processed by CM at 110 °C for 24 h (Maturation 24 h) and 48 h (Maturation 48 h).

Rheological parameters	Modification processes				
	Control	HIPH 6 h	HIPH 12 h	Maturation 24 h	Maturation 48 h
k	0.010 ±	0.003 ±	0.003 ±	0.007 ±	0.009 ±
	0.000 ^A	0.000 ^B	0.000 ^B	0.000 ^C	0.000 ^D
n	0.873 ±	0.966 ±	0.968 ±	0.885 ±	0.860 ±
	0.001 ^{AC}	0.013 ^B	0.007 ^B	0.002 ^C	0.003 ^A
R²	0.999	0.999	0.999	0.999	0.999

*Same letters on the same line indicate that the samples are not significantly different at 5% significance level between the different processes.

According to the results of Table 2, processing by HIPH reduced k in 70% ($p \leq 0.05$), without difference among the processes ($p > 0.05$). While the CM processing reduced the k value by 30% (Maturation 24 h) and 10% (Maturation 48 h) ($p \leq 0.05$). For n, the CM process did not promote differences when compared to the control sample ($p > 0.05$); the HIPH process produced a CG solution with a behavior closer to that of a Newtonian fluid ($n = 1$) ($p \leq 0.05$), with no difference between the processes.

According to Table 3, the enthalpy of desolvation did not vary ($p > 0.05$) for any of the control and processed samples.

Table 3.: Enthalpies of desolvation, melting, and complete decomposition of the control CG (without processing), processed by HIPH 6 h at 60 °C, and CM at 110 °C for 24 and 48 h.

Samples	Enthalpy (J/g)		
	Desolvation	Melting	Complete decomposition
Control	136.31 ± 7.55 ^A	2.84 ± 0.24 ^A	12.08 ± 1.45 ^A
HIPH 6 h	132.09 ± 12.07 ^A	2.30 ± 0.23 ^B	20.51 ± 2.66 ^B
Maturation 24 h	130.32 ± 10.82 ^A	0.31 ± 0.06 ^C	13.01 ± 2.39 ^A
Maturation 48 h	135.24 ± 4.54 ^A	0.24 ± 0.04 ^C	5.88 ± 0.65 ^C

*Same letters on the same column indicate that the samples are not significantly different at 5% significance level between the different processes.

For melting enthalpy, there was a 19% reduction when the sample was processed by HIPH 6 h compared to the control sample. About CM processes, there were reductions in the melting temperature of 89 and 92% for 24 and 48 h, respectively, in relation to the control sample. However, the most distinct behavior occurred in the enthalpy of complete decomposition. While the HIPH promoted an increase in 70%, the CM 48 h reduced in 51% when compared to the control sample. This result demonstrates that the HIPH

preserves the molecule more than the CM process, due to a greater need for energy for the degradation.

CONCLUSION

Physical processing by HIPH and CM promoted chemical and physical changes in the CG. Both processes increased the concentration of the 1→2-linked of arabinose; reduced the terminal linkage of glucose; promoted a slight increase in the MW; and reduced its apparent viscosity and melting enthalpy. Moreover, the CM reduced the concentration of terminal linkages and 1→3.6-linked of galactose; increased the number of 1→6-linked; and reduced the enthalpy of complete decomposition of CG; while the HIPH increased the gum's enthalpy of complete decomposition. The increase in the MW of the CG and the reduction in the consistency of its solutions, promoted by the modifications by HIPH and CM, can be positive for the gum's emulsifying and encapsulating properties as they promote greater steric repulsion (greater MW) and increase the diffusivity of the gum to the oil-water interface.

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