Microbiological Quality of Water of Green Coconut With Different Coatings

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Abstract. *The objective of this work was to evaluate the effect of different biodegradable coatings on the microbiological quality of coconut water 'Anão Verde' variety stored for 40 days. The presence of coliforms at 45 ºC and 35 ºC, mesophilic aerobics, filamentous fungi and yeasts in the water was lower in fruit coated with chitosan and chitosan + gelatin. The contamination was higher in the water of fruits coated with gelatin and in the control fruits (no coating). Throughout storage time, only three genera of fungi were identified in coconut water: Penicillium, Fusarium and Paelomyces. The genus Penicillium was found more frequently in relation to Fusarium and the latest in relation to Paelomyces. During the storage, the waters of the fruits coated with gelatin, gelatin + carboxymethylcellulose and the control fruits presented higher frequency of contamination with these fungi, while the waters of fruit coated with chitosan and chitosan + gelatin had a lower frequency. The fruit coated with chitosan and chitosan + gelatin better preserved the microbiological quality of the coconut water for natural consumption in the own fruit.*

Keywords. *Cocos nucifera*, storage, fungi, bacteria, chitosan, gelatin

Introducão

Coconut water is a refreshing and tasty drink, very popular in Brazil and other tropical countries. It consists mainly of sugars and minerals, nitrogenous substances and vitamins, wich characterizes a product of easy contamination and rapid deterioration in contact with the environment. The chemistry sanitization of the coconut surface is limited due to the husk irregularities and to the calyx and stalk structures that facilitate microbial attachment. The application of biodegradable coatings has been focused to control the development of microorganisms and to increase the shelf life of products. Several authors mention the antimicrobial activity of chitosan to inhibit the growth of microorganisms, among them, Fusarium sp (Ravi Kumar, 2000).

The objetive of this study was to evaluate the effect of different biodegradable coatings on the microbiological quality of coconut water during storage for 40 days.

Material & Methods

Coconut (*Cocos nucifera* L.), 'Anão Verde' variety with eight months of age was used. After cleaning, sorting and sanitizing, the fruits were separated into portions to applied the coatings (Table 1) through immersion for 1 minute and then they were dried at room temperature.

Table 1. Treatments and respective formulations used to coat the coconuts

Treatment	Formulations
1	Chitosan 1.5 % + glycerol (20% v/p chitosan) + PEG-400 (10% v/p chitosan)
2	Carboxymethilcellulose (CMC) 3% + gelatin 5%
3	Chitosan 1.5% + gelatin 5%
4	CMC 3% + stearic acid (15% p/p CMC) + PEG-400 (10% v/p CMC)
5	Gelatin 5% + glycerol (15% v/p gelatin) + PEG-400 (10% v/p gelatin)
6	Control (pure water)

After that the fruits were stored at 12 ± 2 ºC, 80% RH for 40 days. The experimental unit was composed of two fruits.

The following microbiological analysis of water were done every 10 days: total coliforms (35 ºC) and fecal coliforms (45 ºC); counting of mesophilic aerobics, filamentous fungi and yeasts; identification of filamentous fungi. The analysis followed the methodology proposed by Vanderzant & Splittstoesser (1992), modified by Smith (1995), and Downes & Ito (2001). The analysis of filamentous fungi was based on methodology described by Pitt & Hocking (1997) and Samson et al. (2004) for isolation and identification, respectively. A completely randomized design with split plot in time was used and the interaction between the factors time and coatings was investigated. The results were submitted to analysis of variance and the Scott Knott 5% average test.

Results e Discussion

A significant interaction between the coatings and the storage time in all microbiological evaluations was observed, except for filamentous fungi and yeasts that showed isolated effect of coatings and storage time (Table 1).

Table 1. Anlysis of Variance

Variable	Sources of variation		
	Coatings	Time	Coatings x time
Coliforms 35 °C	0.00 **	0.00 **	0.00 **
Coliforms 45 °C	0.00 **	0.00 **	0.00 **
Mesophilic Aerobics	0.02 *	0.00 **	0.02 *
Filamentous fungi and yeasts	0.01 **	0.01 **	0.61

*: F significative (p=0.02); **: F significative (p<0.01)

At 40 days of storage, there was no presence of coliform at 45 °C in water for fruit coated with chitosan, gelatin + CMC and chitosan + gelatin (Table 2).

Table 2. Most probable number (MPN) of propagules of coliforms at 45 ºC and coliforms at 35 °C and counting of mesophilic aerobic bacteria, filamentous fungi and yeasts in coconut water using different coatings for 40 days of storage at 12 ° C

Coatings	Days	Coli 45 ºC (MPN mL^{-1})	Coli 35 ºC (MPN mL^{-1})	Mesophilic Aerobics (CFU mL^{-1})	Filamentous and Yeasts (CFU mL^{-1})
Chitosan	0	ND	ND	$2.0x10^1$	1
Gelatin + CMC	0	ND	5	$5.1x10^1$	3
Chitosan + gelatin	0	ND	ND	$3.9 x10^1$	2
CMC	0	ND	4	$5.4 x10^1$	5
Gelatin	0	ND	ND	$1.32 x10^2$	7
Control	0	ND	ND	$1.02 x10^2$	$2.4 x 10^1$
Chitosan	10	ND	ND	$2.8 x10^1$	5
Gelatin + CMC	10	ND	ND	$1.81 x10^2$	6
Chitosan + gelatin	10	ND	ND	$6.9 x 10^1$	5
CMC	10	ND	ND	$3.30 x10^2$	$3.9 x 10^1$
Gelatin	10	ND	ND	$1.71 x10^2$	$1.88 x 10^2$
Control	10	ND	ND	$7.66 x10^2$	$6.52 x 10^2$
Chitosan	20	ND	ND	$1.2 x 10^1$	6
Gelatin + CMC	20	ND	ND	$3.3 x 10^1$	$1.0 x 10^1$
Chitosan + gelatin	20	ND	ND	$1.1 x 10^1$	8
CMC	20	ND	ND	$4.5 x 10^1$	$1.1 x 10^1$
Gelatin	20	ND	ND	$2.5 x 10^1$	$5.4 x 10^1$
Control	20	ND	ND	$1.86 x10^2$	$8.5 x 10^1$
Chitosan	30	ND	ND	$3.53 x10^2$	$4.8 x 10^1$
Gelatin + CMC	30	ND	9	$4.71 x10^2$	$2.83 x10^2$
Chitosan + gelatin	30	ND	ND	$3.69 x10^2$	$6.8 x 10^1$
CMC	30	4	7	$4.10 x10^2$	$6.5 x 10^1$
Gelatin	30	4	11	$4.80 x10^2$	$2.09 x10^2$
Control	30	9	26	$8.92 x10^2$	$3.67 x10^2$
Chitosan	40	ND	ND	$3.93 x10^2$	$5.3 x 10^1$
Gelatin + CMC	40	ND	11	$1.053 x 10^3$	$1.02 x10^2$
Chitosan + gelatin	40	ND	16	$7.03 x10^2$	$9.2 x 10^1$
CMC	40	ND	7	$2.040 x 10^3$	$4.80 x10^2$
Gelatin	40	11	15	$1.487 x 10^3$	$3.37 x10^2$
Control	40	4	9	$1.1767 x 10^4$	$8.48 x10^2$

At 30 days of storage coliforms at 35 ºC were not observed in the treatments with chitosan and chitosan + gelatin and also in all the treatment with chitosan at 40 days of storage(Table 2).

These data indicate that chitosan coatings can be effective to reduce the infestation of coliforms during storage and can help to keep away these microorganisms in the water of fruit stored until 40 days.

The number (CFU) of mesophilic aerobic bacteria, filamentous fungi and yeasts (Table 2) increased during the storage. The lower number of CFU of aerobic mesophilics was observed in the water from the fruit treated with chitosan and chitosan + gelatin throughout the storage period.

During storage only three genera of fungi in coconut water were identified in this order of frequency: Penicillium, Fusarium and Paelomyces. Fungi were not identified at the beginning of storage. At 10 days the first contaminations arised and increased until the end of storage. During storage, the water from fruits treated with chitosan and chitosan + gelatin had lower rates of contamination. The highest frequency of fungi (Penicillium and Fusarium) on water from fruits coated with gelatin suggests that gelatin facilitates the infestation of these fungi. Gelatin is an animal protein containing the essential amino acids, except tryptophan, which may have favored the faster growth of fungi. The lower frequency of fungi on water from coconuts treated with chitosan and chitosan + gelatin shows the effect of antimicrobial properties of chitosan on growth of Penicillium and Fusarium.

Conclusions

Coconut coated with chitosan and chitosan + gelatin better preserved the microbiological quality of coconut water for natural consumption in the own fruit, indicating the antimicrobial propertie of chitosan.

The main fungi identified in the coconut water during storage were *Penicillium*, *Fusarium* and *Paelomyces*, in that order of frequency.

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