Amazonian Journal of Plant Research *©2017 Universidade Federal do Pará*
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http://www.ajpr.online - Amaz. Jour. of Plant Resear. 2(2):175-182. 2018

Amazonian Journal of Plant Research

Original Paper

Effect of UV-C radiation on quality and enzymatic browning in processed cassava

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Received: 13 May, 2018. Accepted: 27 August, 2018
First published on the web August, 2018
Doi: 10.26545/ajpr.2018.b00021x

Abstract

The objective of this work was to evaluate the treatment with ultraviolet-C (UVC) radiation on quality and enzymatic dimming in minimally processed cassava kept under refrigeration. The cassava pieces were treated with 3.0 kJ m^{-2} on both sides and stored for nine days at 10 ºC and evaluated for physicochemical, sensory and enzymatic analysis. During the storage period, no significant effect was observed between the treated and non-UV-C treated samples on the loss of fresh mass, firmness, soluble solids content, titratable acidity and pH, showing that in this dose the treatment with UV-C radiation does not alter the physical and sensory properties of processed cassava. However, exposure to UV rays significantly reduced the index of physiological deterioration and the activity of the polyphenoloxidase and peroxidase enzymes throughout the storage time with reflection in the preservation of the staining of the samples when compared to the control. Because it does not alter physicochemical properties and considerably inhibits darkening during storage, treatment with UV-C radiation is a promising technology for the conservation of minimally processed cassava.

Key-words: *Manihot esculenta* Crantz, Oxidase Enzymes, Postharvest

Introduction

Cassava (*Manihot esculenta* Crantz) is present in more than 100 countries, especially in developing countries, because of its rusticity and the ability to produce high amounts of starch in climate and soil conditions in which other crops would not survive, in food security (Vieira et al., 2011; Freire et al., 2014).

In Brazil, cassava is among the country's main crops, most of which are cultivated by small and medium-sized producers (Mezette, Blumer and Veasey, 2013). The North region represents 33.5% of the national production, followed by the Northeast region with 25.6%, with the State of Pará being the largest Brazilian producer, with an estimated harvest of 4.21 million tons in 2017 (CONAB, 2017). However, it is observed that the commercialization of these roots in natura presents little consumer acceptability due to the difficulty of peeling and little practicality, besides the limited useful life due to the rapid physiological deterioration (Ramos et al., 2013; Rinaldi, Vieira and Fialho, 2015).

The minimum processing, in this context, arises to respond to a new consumption trend and have been increasingly accepted in world markets (Santos and Oliveira, 2012), allowing the production of a differentiated and innovative product, adding value to the raw material (Almeida et al., 2010). In addition, the production of cassava is a very important factor in the production of cassava.

However, minimal processing acts as a stress inducer in plant tissue, because tissue injury, inherent to processing, changes the physiology of products

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(Chitarra and Chitarra, 2005), in cassava, for example, the rapid physiological deterioration is marked by the performance of enzymes that in contact with oxygen promote both the appearance of fine dark blue vascular streaks causing the product to darken as to the action on the carbohydrates leading to the softening of the pulp (Saravanan et al., 2016).

Thus, it is fundamental to adopt post-harvest conservation technologies that minimize these deleterious effects with a reflection on the preservation of quality and the maintenance of the life of minimally processed cassava, such as ultraviolet C.

It has been used in recent years in post-harvest because it is highly effective in the elimination of microorganisms, because it does not form toxic residual compounds on the surface of the product, the low cost of application, the non-production of odor and the inactivation of enzymes such as PPO, involved in the process of darkening of plant tissues (Manzocco et al., 2011; Ribeiro, Canada and Alvarenga, 2012; Urban et al., 2016).

However, studies reporting the application of UV-C radiation on table cassava physiology are still scarce. The objective of this study was to evaluate the effect of UV-C radiation on the quality and control of enzymatic browning in minimally processed table cassava roots during refrigerated storage.

Material and Methods

Plant material

Causavara table cassava roots of white color were harvested at the age of nine months in the municipality of Monte Alegre, Pará. The material was packed in plastic baskets and transported to the Technology Laboratory of the State Technological Education School of Pará - EETEPA.

In the laboratory, the roots were manually peeled and submitted to minimal processing. The cutting was carried out longitudinally in pieces with an average size of 7.0 cm in length and 2.0 cm in diameter. After the processing, the root pieces were sanitized in 5 mg chlorinated solution L^{-1}, centrifuged and allowed to dry at room temperature (25 °C) on paper towels.

After drying, the pieces of cassava were divided into two lots and submitted to treatment with UV-C radiation at the concentration of 3.0 kJ m^{-2} determined in preliminary tests. The lot that was not exposed to radiation consisted of the control treatment.

Treatment with UV-C radiation

For treatment with UV-C radiation the cassava pieces were arranged without overlapping in a translucent glass pan inside a stainless steel casing and irradiated with the aid of three low pressure mercury lamps located at a distance of 30 cm above and below the surface of the samples. The UV-C lamps were heated for 30 minutes prior to irradiation to ensure reliable results. The intensity of UV-C radiation (254 nm) was monitored using a radiometer (Lutron Electronic Co., Ltda., Taiwan).

Then, the samples of both treatments were conditioned in styrofoam trays of expanded polystyrene (EPS), coated with 14 micron PVC plastic film and conditioned in refrigerator at 10 ± 2 °C and 87% RH for nine days.

Physical-chemical and sensory analyzes

It was evaluated: loss of fresh mass, determined using a semi-analytical balance, with an accuracy of 0.01 g, calculating by difference the weight loss between the initial day and the one in each evaluation period, the results being expressed as a percentage of fresh mass (%).

The firmness of the samples was determined using a TA-XT2 texturometer, by compressing the samples at a speed of 2 mm s^{-1} with an aluminum cylinder of 100 mm in diameter and the firmness was determined by the maximum force during compression in Newton (N).

For the determination of the color was used "Minolt" colorimeter, model CR-300, with reading of the coordinate L * with the results expressed in luminosity (L *).

Soluble solids content was measured by refractometry using 10 g of macerated pulp and homogenized in a refractometer Quimis digital refractometer according to AOAC recommendations (2012) and results expressed in degrees Brix (° Brix).

The titratable acidity content expressed as % citric acid per 100 g of pulp was determined by titration of known mass of homogenized pulp and diluted with distilled water with a standard solution of 0.1 M sodium hydroxide, to 1% phenolphthalein, following the AOAC recommendation (2012).

The pH was obtained by direct reading of the crushed pulp in digital pH meter (TECNAL, model MPA 2010), duly calibrated in buffer solution 4.0 and 7.0 according to AOAC (2012).

Post-harvest physiological deterioration (DFP) through visual analysis of the product and assigning notes on a hedonic scale of 4 points, where: 0 = no sign of dimming, 1 = surface with up to 25% of dimming, 2 = surface area between 26 and 50% dimming, 3 = surface with up to 75% dimming, 4 = surface with more than 75% dimming (Andrade et al., 2016).

Pxtraction and activity of polyphenoloxidase (PPO) and peroxidase (POD)

Polyphenoloxidase (PPO) and peroxidase (POD) activity extraction and assay were performed according to the methodology described by Coelho et al. (2017). For this purpose, 0.50 g of the homogenized tissue surface was used in 1.2 mL of 0.2 M sodium phosphate buffer (pH 6.0) in ice-cold pistil at 4 °C for two minutes followed by centrifugation at 10.000 xg for 21 minutes at 4 °C, the collected supernatant was used as an enzyme extract. For the PPO activity assay, 100 µL of the enzyme extract was added with 1.6 mL of 0.2 M phosphate buffer (pH 6.0) and 1.2 mL of 0.2 M catechol. After 10 minutes in the bath dry at 20 °C, the readings were taken every 2 minutes at 10 second intervals in a spectrophotometer at the absorbance of 420 nm. The PPO activity was calculated as a function of the variation of the absorbance (dA) $.min^{-1}.mg^{-1}$ of protein. The determination of POD activity was performed according to the methodology proposed by Simões et al. (2015). The reaction mixture was composed of 100 µL of the enzyme extract, 1 mL of 0.2 M phosphate buffer (pH 6.0), 100 µL of guaiacol (0.5%) and 100 µL of hydrogen peroxide (0.08%) previously kept in a water bath at 20 °C for 10 minutes, after which the spectrophotometer readings were performed at 25 °C at 470 nm absorbance. The activity of the POD was calculated as a function of the variation of the absorbance (dA) $.min^{-1}.mg^{-1}$ of protein.

Experimental design and statistical analysis

The experimental design was completely randomized in a 2x6 factorial arrangement, that is, two treatments (with UV-C radiation and without UV-C radiation) and six storage times (0, 1, 3, 5, 7, 9 days) , with three replicates and the experimental plot composed by trays of 250 g.

The results were submitted to analysis of variance followed by a comparison of means by the Tukey test at 5% of significance with the aid of statistical software SAS (Statistical Analysis System, versão 9.3).

Results and discussion

The loss of fresh mass in the manioc pieces did not vary significantly with respect to treatment with UV-C radiation (Figure 1), observing up to 2.3% over the nine days of evaluation.

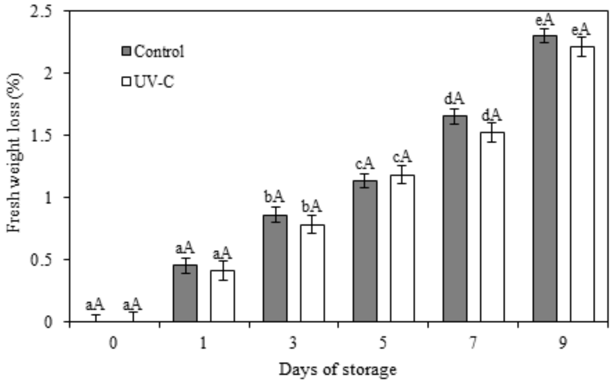

Fig. 1. resh mass loss (%) in roots of minimally processed cassava treated with UV-C radiation and stored for nine days under refrigeration (10 ± 2 °C).

Although the effect of UV-C radiation was not observed, it was observed that in both treatments there was a low percentage (2.3%) in the loss of fresh, lower mass, for example, as verified by Andrade et al. (2016) that obtained percentages ranging from 4 to 6% in table maniocs of cultivars Causavara and Manteiguinha stored at 10 °C for 10 days. These authors concluded that the Causavara cultivar showed the lowest percentages in the mass loss during the storage period, so it is possible that a characteristic of this cultivar is a higher post-harvest resistance to moisture loss. The firmness of the samples remained virtually unchanged over seven days of refrigerated storage, observing a slight decrease on the last day of evaluation in both treatments (Figure 2). This maintenance of firmness is associated with the refrigerated condition and the coating with PVC plastic film which restricts the gas exchanges, and consequently the loss of moisture from the interior of the package to the external environment. According to Junqueira et al. (2010), the firmness of the plant tissue is composed of numerous factors, some of which are environmental, others due to processing and post-harvest conservation, however, different cultivars vary greatly in their rate of deterioration of firmness due to loss of moisture. The latter effect corroborates with that observed in the analysis of fresh mass loss suggesting that the cultivar Causavara used in this work presents resistance to the minimum processing through the smaller loss of fresh mass with reflection in more turgid, firm samples.

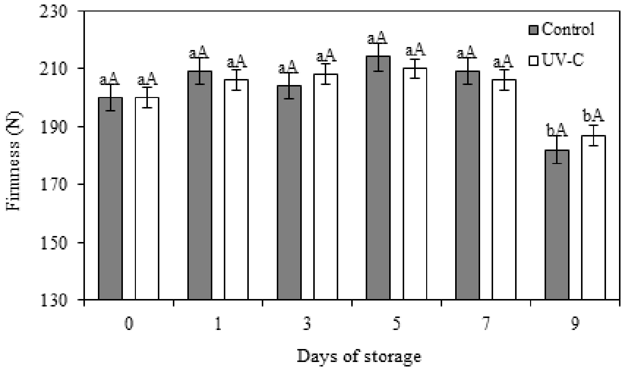

Fig. 2. Firmness (N) in roots of minimally processed cassava treated with UV-C radiation and stored for nine days under refrigeration (10 ± 2 °C).

Soluble solids content (SS) in °Brix is used as a quality indicator, as it may suggest the taste of food as "sweet". Figure 3 shows the mean values of SS in untreated and UV-C treated cassava pieces over nine days of storage.

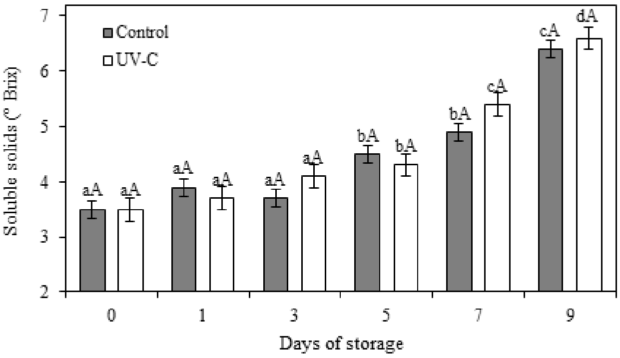

Fig. 3. Soluble solids content (°Brix) in roots of minimally processed cassava treated with UV-C radiation and stored for nine days under refrigeration (10 ± 2 °C).

It is observed that between the initial day and after nine days of storage the SS content increased significantly from 3,5 °Brix to more than 6,5 °Brix, respectively (Figure 3). These values are similar to those observed by Rinaldi, Vieira and Fialho (2015), with three cultivars of minimally processed table manioc whose average values ranged from 3.4 to 6.4 °Brix. This increase is mainly due to metabolic reactions such as carbohydrate degradation or the loss of fresh mass resulting in the accumulation of sugars in the pulp of the samples during storage.

Regarding the treatments, no significant effect of UV-C radiation on the accumulation of sugars was observed, observing similar mean values throughout the storage period (Figure 3).

The content of titratable acidity was influenced only by storage time, not differing in relation to treatment with UV-C radiation (Figure 4).

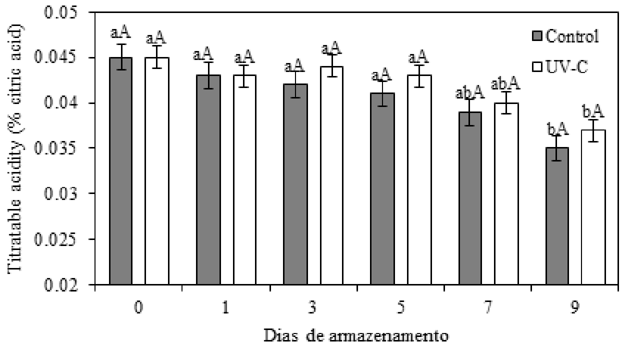

Fig. 4. Titratable acidity (% citric acid) in roots of minimally processed cassava treated with UV-C radiation and stored for nine days under refrigeration (10 ± 2 °C).

Mean values of acidity showed a tendency to decrease with storage time, with the highest value being 0.045% citric acid at day zero and the lowest value of 0.035% citric acid at nine days of storage (Figure 4). This reduction in titratable acidity during refrigerated storage is common in plant products due to the use of the acids of its composition as an energy substrate for respiratory metabolism. In the case of mangoes that were minimally processed and treated with antioxidants, the titratable acidity content progressively decreased during post-harvest storage (Andrade et al., 2016; Rinaldi et al., 2017), corroborating the findings of the present study. pH is a very important factor in the conservation of food, since it limits the development of microorganisms that promote the loss of sensorial quality of the product. According to Figure 5, mean pH values increased with storage time, with no significant difference between samples treated with or without UV-C radiation. At the beginning of storage (day zero) until the fifth day of evaluation the pH values maintained stability ranging from 5.89 to 5.93, respectively. From the seventh day onwards, there is a more expressive increase with an average value higher than 6.0 on the last day of evaluation (Figure 5). In summary, treatment with UV-C radiation (Figures 3, 4 and 5) did not alter the taste-related chemical properties of the samples. This result seems to be a useful observation, since it confirms the softness of UV-C technology as a post-harvest treatment. Results were similar to those observed in other minimally processed products such as apple (Gómez et al., 2010), pineapple (Pan and Zu, 2012), tomato (Pataro et al., 2015), watermelon (Gómez et al., 2015) and pitaya (Nimitkeatkai and Kulthip, 2016), for example, where exposure to treatment with UV-C radiation did not compromise quality over the storage period.

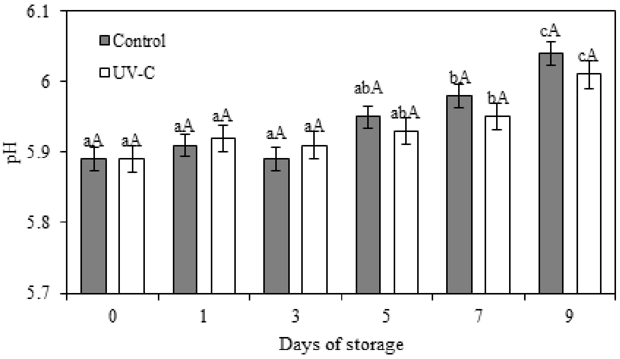

Fig. 5. pH in roots of minimally processed table manioc treated with UV-C radiation and stored for nine days under refrigeration (10 ± 2 °C).

The mean values for the post-harvest physiological deterioration index (CPD) observed in cassava pieces are shown in Figure 6.

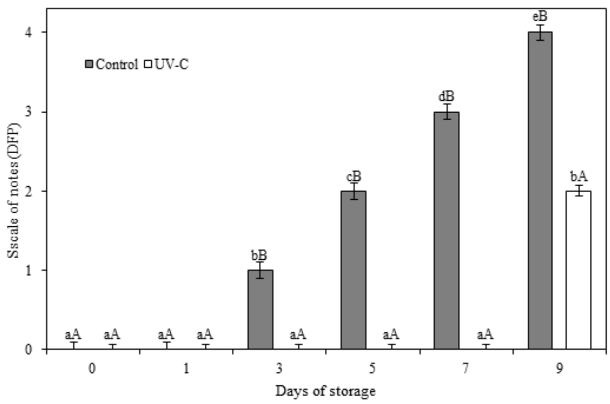

Fig. 6. Post-harvest physiological deterioration (DFP) in roots of minimally processed cassava treated with UV-C radiation and stored for nine days under refrigeration (10 ± 2 °C).

In general, the DFP began to manifest on the third day, specifically on the samples not treated with radiation when they were characterized with note 1 (surface with up to 25% darkening) and with note 4 (surface with more than 75% darkened) end of nine days. At seven days of storage, irradiated samples showed no symptoms of PFD, with a slight darkening at the end of nine days when they were characterized with note 2 (surface between 26 and 50% darkened) (Figure 6).

Inhibition of darkening induced by UV-C treatment over seven days is superior to the four days of inhibition observed in table manioc pieces treated with anti-curing substances (RAMOS et al., 2013). On the other hand, Andrade et al. (2016) evaluating two table cassava cultivars processed and treated with antioxidants did not observe darkening of the tissues during 10 days of storage at 10 °C.

DFP analysis corroborates that observed in polyphenoloxidase activity (PPO) (Figure 7).

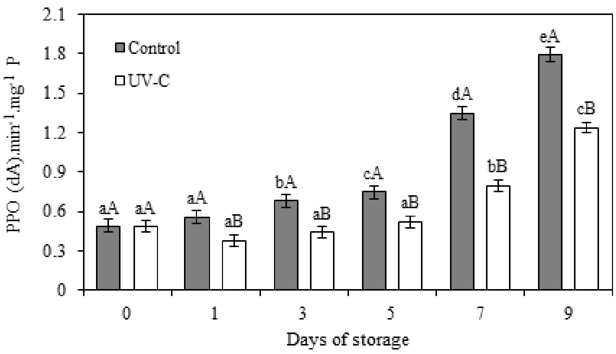

Fig. 7. Specific activity of polyphenoloxidase (PPO) in roots of minimally processed cassava treated with UV-C radiation and stored for nine days under refrigeration (10 ± 2 °C).

In the samples treated with UV-C radiation the specific activity of the PPO remained stable for seven days observing a slight increase at nine days of storage. On the other hand, in samples not exposed to UV-C treatment PPO activity showed a remarkable increase during the entire storage period, especially after the fifth day of analysis (Figure 7).

This beneficial effect of UV-C on PPO activity has already been mentioned in products submitted to minimum processing such as pineapple (Pan and Zu, 2012) and potato roots (Rocha et al., 2015). In these studies, the authors suggest that the hormonal effect of UV-C radiation stimulates the synthesis of bioactive compounds that act as antioxidant agents in the enzymatic inhibition of PPO.

As regards peroxidase activity (POD), there was a trend similar to that observed for PPO (Figure 8), that is, the exposure of cassava samples to UV-C radiation was able to reduce significantly the activity of this enzyme, especially until the seventh day of storage when the staining of the samples was similar to that observed on the initial day.

In comparison, the POD activity in the samples not treated with radiation (Figure 8), is significantly higher after three days, remaining constant until the end of the storage period.

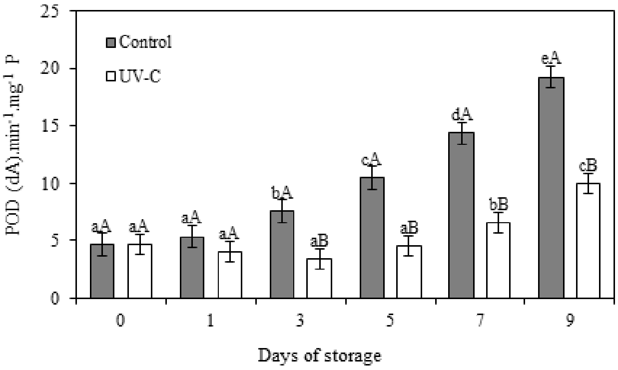

Fig. 8. Specific activity of peroxidase (POD) in roots of minimally processed cassava treated with UV-C radiation and stored for nine days under refrigeration (10 ± 2 °C).

In general, the performance of these enzymes occurs when fruits and vegetables are mechanically injured during harvesting, handling, packaging, transportation and storage procedures or when they are processed in the industry (Chitarra and Chitarra, 2005). For most vegetables, darkening of tissues leads to reduced quality due to altered sensory properties and decreased nutritional properties (Parkin, 2010).

Regarding the staining of the samples (Figure 9), it was observed that there was a reduction in the luminosity value (L *) of both treatments, indicating the gradual darkening with the storage time.

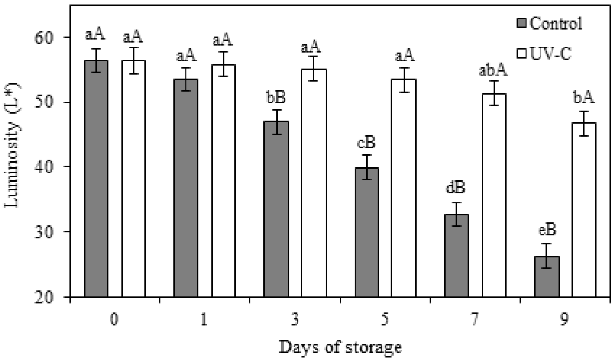

Fig. 9. Luminosity (L *) in roots of minimally processed cassava treated with UV-C radiation and stored for nine days under refrigeration (10 ± 2 °C).

In general, the lower L * values were observed in the non-irradiated samples, whose mean values increased from 56,4 on the initial day to less than 20.0 at the end of the storage period, corroborating with the observed darkening tendency. This large change in tissue staining is specifically due to injury caused by minimal processing, thereby inducing decomposition of the PPO and POD enzymes and their substrates, as observed in Figures 7 and 8, respectively.

In the irradiated samples, the loss of luminosity during the nine days was only 10.0 L °, that is, the color of the cassava was practically unchanged in relation to the initial day of storage, except for the slight yellowing presented on their surfaces. In this sense, the preservation of coloration is closely related to the lower activity of the PPO and POD enzymes, since these affect significantly the visual characteristic of the vegetal products during the storage.

Conclusions

The application of UV-C radiation does not alter the physical-chemical properties and presents as a technology capable of inhibiting the enzymatic darkening in roots of minimally processed cassava for up to seven days in refrigerated condition.

Conflict of interest: All authors declare no conflict of interest.

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Cite this article as:

Sanches, A.G.; Silveira, A.G.; Oliveira, A.R.G. de; Cordeiro, C.A.M. 2018. Effect of UV-C radiation on quality and enzymatic browning in processed cassava. **Amaz. Jour. of Plant Resear** 2(2): 175-182.

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