Effects of Rice Flour on Emulsion Stability, Organoleptic Characteristics and Thermal Rheology of Emulsified Sausage

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Abstract Cereal flour protein has been used in processed meat products to improve yield and texture by enhancing water-binding properties. The current study was conducted to evaluate the effects of rice flour on physicochemical properties, cooking loss, storage modulus, texture and sensory properties of reduced-fat emulsified sausage. Addition of rice flour at 2%, 4% and 6% significantly improved the moisture content of emulsified sausage compared to control (p < 0.05). Emulsified sausage containing rice flour additives showed a significantly lower cooking loss and better emulsion stability compared to control samples due to a lower release rate of water (p < 0.05). L* value of reduced-fat sausage significantly decreased with the addition of 6% of rice flour compared to others samples (p < 0.05). Supplementation of 6% rice flour significantly increased the hardness values of reduced-fat sausage compared to control and other rice flour levels (p < 0.05). Rice flour added at 4% had the highest score for overall acceptability. It is concluded that the incorporation of rice flour could be an effective ingredient to improve the quality and sensory of emulsified sausage with reduced fat content.

Keywords: cooking, moisture, emulsion stability, sausage, sensory, texture, rice flour

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1. Introduction

The increasing demand on healthful meat products by consumers has caused manufacturers in meat industry to continuously develop new or better quality products at low cost, containing low fat and incorporating health enhancing ingredients with improved functional properties, palatability and nutritional value. Currently, the meat industry is utilizing ingredients from vegetable sources such as soy protein [1], whey protein [2], vegetable oil [3], soy beans [4], cereal flour, milk powder and other non-meat ingredients [5,6] as binders, fillers or extenders to reduce/replace the use of animal fats. These ingredients play a significant role in functional properties including emulsion stability, water and fat-binding capacity and textural properties of meat products [6]. To succeed in producing reduced-fat, healthy and palatable food products, other ingredients must be chosen and applied to replace fat in the food process.

Rice flour additives includes a small amount of water (about 8%), fat (about 1%) and ash (about 1%) [7]. It has good nutritional value and it is considered important in the process of gluten-free food products, due to its very low allergenic effect, low sodium content and high digestibility [8]. Rice flour made from finely milled rice (Oryza sativa, L.) has starch (approximately 80%) as a main component which has the potential to be used in meat industry due to its unique gelatinization properties and binding capacity [9]. This can retain water during the cooking process and subsequent improve the shelf-life as well as deliver the desired texture.

According to Laureys [10], rice flour can be used as processing aids, ingredients in health food, expanding agents in extrusion food, flavour carriers, emulsifiers and fat replacers in food products. Rice flour can provide suitable textures for a wide group of foods with high natural stability [11].

Among limited studies, Huang et al., [12] reported that rice flour showed promise in improving yield and juice retention for meat products. Yang et al., [13] found that the total substitutions of fat in the process of duck sausage by rice flour produce a more acceptable product. In the current research, rice flour was used as an appropriated additive to reduce water separation, retaining substantial amounts of both moisture and fat in the meat emulsion which enhance storage stability and improve organoleptic properties of the final product. However, few studies have been conducted demonstrating the functionality of rice flour as a gel enhancer on the quality, thermal stability and sensory properties of muscle food systems. The objective of this research was to evaluate the addition of rice flour affects physicochemical properties, cooking loss, storage modulus, texture and sensory properties of reduced-fat sausage.
2. Materials and Methods

Fresh pork lean meat and back-fat were purchased from a local market (Sushi Food Co., Ltd., Nanjing, China). Rice flour was commercially obtained from Thailand (Yangzhou, Wenfeng Refined Oil and rice Ltd., Jiangsu, China). Other seasonings including salt, white pepper and tripolyphosphate sodium were obtained from the pilot lab of National Meat Research Centre of China, Nanjing Agricultural University. The proximate composition of RF was 10.11% of moisture, 7.46% of protein, 1.57% of fat and 1.02% of ash.

Table 1. Formulation of reduced-fat sausage containing rice flour (RF) at different levels

| Additives | Lean Meat (g) | Fat (g) | Ice/water (g) | Salt (g) | Sucrose (g) | Sodium tripoliphosphate (g) | White pepper (g) |
|-----------|---------------|--------|---------------|---------|-------------|-----------------------------|-----------------|
| Control   | 800           | 200    | 200           | 20      | 2.5         | 3                           | 2.5             |
| RF(2%)    | 800           | 200    | 200           | 20      | 2.5         | 3                           | 2.5             |
| RF(4%)    | 800           | 200    | 200           | 20      | 2.5         | 3                           | 2.5             |
| RF(6%)    | 800           | 200    | 200           | 20      | 2.5         | 3                           | 2.5             |

The formulation for each treatment was according to those parameters: 80% ground pork and 20% back-fat without any functional ingredients were added in control and 2%, 4% and 6% of rice flour ingredients were added to raw batter for three treatments, respectively (Table 1). After 24 h, the ground pork meat and fat was transferred to a bowl cutter and chopped in a food processor (BZBJ-15, Explo Stainless Steel Mechanical & Engineering Company Hangzhou, China) for 2 min with a half quantity of ice/water and 2% of salt. Then, pork back-fat, seasonings, and rice flour additives (Table 1) were added together with the reminder ice/water and the batter was chopped at high speed for 3 min. Generally, emulsion-type sausages have approximately 30% fat and 2–3% salt content [15]. Therefore, the formulation of sausages in this study had 20% fat and it was referred as reduced-fat sausages. Three batches of each formulation were performed and the batter temperature did not exceed 12 °C. Immediately after chopping, the batter formulated was stuffed into 30-32 mm hand-pulled hog casings. Stuffed samples were hand linked and heated in food processor (Fessmann, GmbH und Co KG, 71364 Winnenden) to an internal temperature of 72 °C. Approximately 300 g of raw batter samples were vacuum-packaged and stored in cooler at 4 °C until further analysis within 3 days.

2.2. Proximate Composition

The pH of reduced-fat sausage was determined using a digital pH meter (Hanna, Italy) [16]. Moisture, fat and protein content of raw and cooked samples were analyzed according to procedure described by AOAC methods [16]. Three samples per each batch were performed in triplicate.

2.3. Cooking Loss

Cooking loss was expressed by calculating the ratio of meat batter before and after cooking, expressed in percentage as follows:

\[
\text{% cooking loss} = \frac{(\text{raw weight} - \text{cooked weight})}{(\text{raw weight})} \times 100.
\]

2.4. Emulsion Stability

Emulsion stability was measured according to the procedure proposed by Serdaroglu and Ozsumer [14] with a modification. About 40 g of raw meat batter was placed in 80 mL centrifuge tube and then centrifuged (Allegra™ 64R, Fisher Scientific, Pittsburgh, PA, USA) at 3,000 × g for 2 min at 4 °C to remove any air bubbles. Each tube was heated until reaching 80 °C in water bath for 30 min. After cooling, all expressible release was removed, weighed and transferred into pre-weighed crucibles and dried at 105 °C for 16 h. The volume of total released fluid (TFR) and water released (WR) was calculated as follows:

\[
\text{%TFR} = \frac{\text{TFR divided by sample weight}}{\times 100};
\]

\[
\text{%WR} = \left( \frac{\text{weight crucible + TFR}}{-\text{(dried weight crucible)} \times 100} \right)
\]

The percentage of fat was determined as a difference between % of total fluid released and % of water released, ignoring any minor protein or salt component found in the dried crucible. Six replications were analyzed for each formulation per batch.

2.5. Vacuum Purge (VP)

Vacuum purge was expressed as % of fluid lost from the initial sample weight during storage. The 20 g of reduced-fat sausage per treatment was weighed and cut to 1 cm thickness, revived and vacuum packaged (nylon/polyethylene vacuum pouch, 1.5 mm thickness, oxygen transmission rate 1 cm² m⁻² h⁻¹; Promax, CT, USA). The samples were stored for fifteen days at 4 °C and then the vacuum seal was broken. The fluid was drained and the samples was wiped with a towel and reweighed. Vacuum purge was determined in triplicate for each treatments.

2.6. Color Evaluation

Hunter L* (lightness), a* (redness) and b* (yellowness) were measured from the interior of cooked sausages after cut using a color difference meter (Chroma MeterCR-400, Konica Minolta Sensing company, Sakai, Japan), calibrated with a white plate, L* = 97.83, a* = -0.43, b* = +1.98. Six measurements of five replicates from each treatment were analyzed and the average value was determined by taking observations from three different locations on a given sample.
2.7. Texture Profile

Samples of cooked sausage cooled at room temperature were sliced into 30 mm high and 25 mm diameter with a round sample taker. Both baked surfaces of the samples were removed with a double-edged knife in order to have a sample with the same height of 30 mm. Then the samples were subjected to two-cycle compression test by using a TA.XT Plus with P50 probe (Stable Micro Systems Ltd., Godalming, Surrey, UK). Hardness, chewiness, cohesiveness, gumminess, springiness and resilience of six samples per treatment were analyzed.

2.8. Rheological Properties

The rheological properties of raw meat batter formulated with different levels of rice flour were performed on a rotational Physical MCR 301 dynamic rheometer (Anton Paar Ltd., Austria) fitted with a 50 mm diameter parallel steel plate running at a gap fixed with 1 mm. About 10 g raw meat batter was placed between two fat parallel stainless plates with the perimeter coated with a thin layer of silicone oil to prevent heat loss. After initial equilibration at 20 °C for 5 min, the sample was continuously heated from 20 °C to a final temperature of 80 °C at a rate of 1 °C min⁻¹. During the heating process, the samples were continuously sheared in an oscillatory mode at fixed scanning frequency of 0.1 Hz storage. Changes in storage modulus (G') were measured during processing with increasing temperature. Each rheological test was performed in triplicate.

2.9. Sensory Evaluation

Sensory characteristics of reduced-fat sausage were evaluated on 9-point Hedonic rating tests described by Wu and Lin [17] with slight modification. Organoleptic characteristics of reduced-fat sausage were evaluated using a trained panel comprising of 20 students and professors (8 male and 12 female) who were experienced in sensory evaluation of foods in National Meat Centre of Nanjing Agricultural University. They received verbal instructions on the mode of evaluation to be made before experimental evaluation. The panel comprising were all asked to evaluate the colour, tenderness, hardness, flavour, juiciness and overall acceptability on a non-structured scale of nine scores (1 = dislike extremely to 9 = like extremely). Samples of 2 cm long from each formulation were heated in a microwave for 15 s, and then immediately presented to the panelists in random order. Cold water was provided to clean their mouths before testing each sample. All data were recorded on a questionnaire designed to indicate the consumer opinion of the product. Three sessions per batch were performed in duplicate.

2.10. Statistical Analysis

All data analysis of this experiment was performed using SPSS package (IBM SPSS version 20.0 for windows). The results were presented as the average and standard deviation of each treatment. ANOVA with Duncan’s multiple range was used to determine statistical difference (P < 0.05). Duncan’s multiple range comparison was also used to compare treatment means.

3. Results and Discussion

3.1. Proximate Composition

Table 2 shows the proximate composition and pH values of reduced-fat sausage containing different levels of rice flour. It was observed that the pH values of reduced-fat sausage showed significant difference among all treatments (p < 0.05). This may be explained due to addition of different amount of rice flour in meat batter. Yang et al. [13] (2009) reported same results when cereal flour was added to duck meat sausages. Note that the incorporation of rice flour at all levels significantly influenced with the protein, moisture and fat content of the products compared to control samples (p < 0.05).

| pH | Protein | Moisture | Fat |
|----|---------|----------|-----|
| Control | 6.67±0.01a | 15.09±0.13b | 63.94±0.14c | 18.70±0.01d |
| RF2% | 6.75±0.02a | 14.40±0.22b | 64.45±0.11b | 18.01±0.03c |
| RF4% | 6.79±0.01a | 14.07±0.30a | 64.60±0.11b | 17.08±0.06b |
| RF6% | 6.85±0.02a | 14.13±0.11b | 64.38±0.07b | 16.61±0.05a |

**Note:** Means in the same column with different superscripts are significantly different (p < 0.05).

The results showed that incorporation of rice flour at 2%, 4% and 6% significantly increased the moisture content of reduced-fat sausage compared to control (p<0.05). Rice flour added at 4% showed higher moisture content compared to 6% (p < 0.05). Gao et al. [18] explored the effects of glutinous rice flour (GRF) on the quality of ground pork patties. They reported that the incorporation of GRF increased the moisture content of ground pork patties highlighting the addition of 3% as better compared to 1% and 5% which was in accordance with our results. Slight differences in protein content of reduced-fat sausage were observed among the samples and protein levels varied between treatments and ranged from 14.0% to 15.10%. As a result of moisture loss during heat processing, protein levels of control samples were higher compared to samples with rice flour. Moisture content has been considered an important property of emulsion sausage, particularly as it relates to texture, sensory characteristics and final weight which is directly associated with commercial value to manufactures [19].

Protein and fat content of reduced-fat sausage slightly decreased with the addition of rice flour. This observation is in agreement with Ali et al. [20] who reported that the addition of 10% of rice flour reduced the protein and fat content of pork and duck sausage. Sampaio et al. [21] also found reduced fat content in beef frankfurters formulated with oat bran, carrageen, and cassava starch, but this depends on various factors including composition of the ingredients and meat properties.

3.2. Cooking Loss and Emulsion Stability

The cooking loss and emulsion stability of reduced-fat sausage formulated with rice flour at different levels is shown in Table 3. Addition of rice flour significantly decreased the cooking loss of reduced-fat sausage compared to control (p < 0.05). This behavior could be explained by the action of starch components of the flour (75 - 80%) as well as by the protein fractions. No
significant difference was observed for cooking loss and TFR among samples containing rice flour at 4% and 6%. Samples with rice flour at 4% and 6% showed the lowest cooking loss and highest emulsion stability, whereas control samples had the highest cooking loss and TFR indicating that non-meat additives could be involved in fat and water-holding capacity.

According to the results, TFR, WR and FR significantly decreased with the increase of rice flour levels in the process of reduced-fat sausage compared to control (p < 0.05). TFR and FR of control samples presented higher values compared to samples with rice flour. Reduced-fat sausage containing rice flour presented improved emulsion stability due to the water binding capacity of rice flour added to the product.

### Table 3. Percentage of cooking loss, vacuum purge and emulsion stability of sausage samples formulated with rice flour

|                | Cooking loss | Vacuum purge | Total fluid released | Water released | Fat released |
|----------------|--------------|--------------|----------------------|----------------|--------------|
| Control        | 6.4±0.66ab  | 12.3±1.02bc | 5.4±0.74c           | 5.36±1.23c     | 0.29±0.05c   |
| RF2%           | 3.0±0.36ab  | 10.2±0.99ab | 2.64±0.98c          | 2.59±0.98c     | 0.07±0.02c   |
| RF4%           | 1.50±0.20a  | 8.34±0.97c  | 1.20±0.16c          | 1.17±0.16c     | 0.03±0.01c   |
| RF6%           | 1.25±0.18a  | 7.58±1.00a  | 1.03±0.00a          | 1.01±0.01a     | 0.02±0.01a   |

*: Means in the same column with different superscripts are significantly different (p < 0.05).

The results of cooking loss and emulsion stability are consistent with studies carried out by Youssef and Barbut [22] who found that cooking loss decreased in meat emulsions after treatments with non-meat ingredients. Ali et al. [20] also reported that the addition of rice flour improved cooking loss and emulsion stability of duck and pork sausage. Our result collaborates also with Gao et al. [18] and Yi et al. [23] who reported that the addition of glutinous rice flour decreased cooking loss and increase emulsion stability of meat products. Besides supplemented ingredients, the emulsions stability of reduced-fat pork batter could be influenced by many factors including protein sources, fat content, types of fibers, salt concentration, and processing methods. The purge loss of reduced-fat sausage showed significantly higher value for control compared to samples containing rice flour during storage (p < 0.05). Our findings show that the addition of rice flour may result in beneficial effects, retaining more moisture compared to control samples during a refrigerated storage.

### 3.2. Dynamical Rheology

Rheological measurements are very important to control the chemical interactions of food components which produce a particular food structure with desired textural attributes [24]. It is often used to measure gel formation and phase transition during temperature sweep from 20 to 80 °C. Figure 1 shows the thermal rheology of reduced-fat sausage formulated with three different levels of rice flour. It was observed that all treatments showed similar thermo-rheological curves up to 75 °C, except the curve for 6% of rice flour which was straight as opposed to the others which varied. There were three phases in G' during the heating process of raw emulsified sausage which might be due to protein denaturation. During the heating process, storage modulus (G') for all treatments generally exhibited similar trend of elastic properties except between 45–80°C where covalent bonds and hydrophobic interactions were completed for proteins [25] and gelatinization of starch were completed [26].

![Figure 1. Comparison in dynamic storage modulus (G’) with increasing temperature of raw batter of sausage samples formulated with different levels of rice flour (RF 2%, RF 4% and RF 6%)](image)

An increased G’ peak at near 50 °C was probably due to the formation of a semi-gel like structure of myosin tail. This phase was followed by slight decrease in G’ while the temperature increased from 54 to 58 °C. According to Tomberg [27], the decrease was due to the denaturation of myosin tails resulting in a sharp change in fluidity and disruption of the meat protein network formed at lower temperatures. The G’ increased rapidly as the temperature
increased from 58 to 80 °C due to the transformation of the viscous sol into an elastic gel network [3]. The significant increase in G’ of reduced-fat sausages could be explained due to the formation of a 3-dimensional network from the amylose and starch in rice flour which interact with muscle protein [28]. Once heating was completed, G’ indicated that all treatments had the same elastic and viscous properties, respectively.

3.3. Color Evaluation

The effects of rice flour on colour values of reduced-fat sausages are shown in Table 4. L* was not different among control and samples containing 2% and 4% of rice flour (p > 0.05). Cooked sausage samples containing 6% of rice flour had significant lower L* values compared to samples from other three treatments (p < 0.05). Our results differ in part from Yi et al. [13] who found that the addition of cereal flours reduced the lightness (L*) of pork and duck sausage significantly decreased by adding 10% of rice flour. Gao et al. (2014) also found lower L* when glutinous rice flour was added at 5% in meat patties. This is in agreement with our results which showed decreased lightness values of reduced-fat sausage with the increase of rice flour level. The addition of rice flour did not significantly affect a* values of reduced-fat sausage. On the other hand, Yang et al. [19] reported that lightness (L*) of pork and duck sausage significantly differ in scores for flavour compared to control (p < 0.05). Ali et al. (2011) found that decreased hardness value of meat batter when the protein ingredient was raised up to 10%. Chewiness showed similar trend as hardness, and in both cases significantly higher values were found in reduced-fat sausage formulated with rice flour.

Table 4. Colour evaluation of sausage samples formulated with different levels of rice flour

| Treatment | L*      | a*      | b*      |
|-----------|---------|---------|---------|
| Control   | 72.65±0.20a | 3.92±0.08b | 7.01±0.13b |
| RF2%      | 72.45±0.34a | 3.85±0.15b | 7.03±0.26b |
| RF4%      | 72.37±0.31b | 3.83±0.11a | 7.30±0.15b |
| RF6%      | 71.71±0.20a | 3.93±0.06b | 7.40±0.14a |

b): Means in the same column with different superscripts are significantly different (p < 0.05).

3.4. Texture Profile Analyses

Non-meat proteins such as soy protein, whey protein and carbohydrates such as starch and cereal flours are often used to enhance the texture of meat products. In this research, texture properties of reduced-fat sausage formulated with different levels of rice flour were evaluated (Table 5). Rice flour added at 6% significantly affected the hardness value of reduced-fat sausage compared to control and addition of 2% and 4% rice flour levels (p < 0.05). There was no significant difference in hardness value of reduced-fat sausage among treatments formulated with rice flour at 2%, 4% and control (p > 0.05). The addition of 6% of rice flour in reduced-fat sausage presented higher value of hardness which was consistent with previous studies on the incorporation of plant protein resulting in increased hardness value of emulsified sausages [29,30]. However, Ali et al. (2011) found that decreased hardness value of meat batter when the protein ingredient was raised up to 10%. Chewiness showed similar trend as hardness, and in both cases significantly higher values were found in reduced-fat sausage formulated with rice flour.

Table 5. Texture properties of sausage samples formulated with different levels of rice flour

| Treatment | Hardness | Springiness | Cohesiveness | Chewiness | Resilience |
|-----------|----------|-------------|---------------|-----------|------------|
| Control   | 3084.80±172.14c | 0.82±0.01a | 0.69±0.01b | 1756.79±112.58a | 0.32±0.01a |
| RF2%      | 3055.99±174.39a | 0.83±0.01c | 0.70±0.01a | 1763.16±88.99b | 0.32±0.01c |
| RF4%      | 2946.42±307.48b | 0.85±0.03b | 0.71±0.02a | 1767.25±205.62a | 0.32±0.02a |
| RF6%      | 3528.16±105.42a | 0.86±0.03a | 0.70±0.01c | 2134.94±90.30b | 0.32±0.01b |

b): Means in the same column with different superscripts are significantly different (p < 0.05).

Feng and Xiong [31] reported that the interactions between meat and nonmeat additives may effectively affect the gel properties in emulsified meat products through modifying the product texture. There were no significant differences on springiness among all treatments (p>0.05). Therefore, rice flour added at 4% and 6% on reduced-fat sausage significantly differ from control samples (p < 0.05). Cohesiveness and resilience of reduced-fat sausage were not significantly affected by the addition of rice flour. This result is in agreement with a previous study by Hsu and Sun [32] who indicated that there was no significant difference in cohesiveness among emulsified meat products supplemented with different non-meat proteins. Rice flour added at 6% had a significant difference compared to 2%, 4% and control on the hardness chewiness in our current study (p < 0.05). Therefore, rice flour possesses functional properties in enhancing texture of meat products.

3.5. Sensory Evaluation

Acceptable flavour, texture and other sensory characteristics in reformulated meat products are considered an important quality for overall acceptability of the products by consumers [33]. Table 6 shows the sensory evaluation of reduced-fat sausage formulated with rice flour at 2%, 4% and 6%. Reduced-fat sausage formulated with 4% and 6% of rice flour significantly differ in in scores for flavour compared to control (p < 0.05), but no significant differences was observed between control samples and rice flour added at 2%.

The addition of rice flour additives slightly increased the scores of colour, tenderness and texture, but no significant difference was observed compared to control in this study (p > 0.05). These results are in agreement with Gao et al. [18] and Ahmad et al. [19] who reported an increase of these properties in meat products when non-meat additives were incorporated. Yang et al. [13] also reported that the addition of rice flour in duck sausage significantly increased overall acceptability. The overall acceptability of reduced-fat sausage containing rice flour had significant difference compared to control (p < 0.05) excepted those containing rice flour at 2%. Rice flour added at 4% had the highest score for overall acceptibility compared with control that had the lowest score, which
suggest that the panelists were different about its acceptability. This observation is in agreement with Gao et al. [18] who suggested that the addition of glutinous rice flour at 3% had better acceptability compared to 1% and 5%. All levels of rice flour incorporated in the formulation effectively improved the sensory quality of our product.

| Table 6. Sensory evaluation of reduced-fat sausages formulated with different levels of rice flour |
|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|
|                | Colour          | Flavor          | Juiciness        | Tenderness       | Texture         | Overall acceptability |
| Control        | 5.67±0.75a      | 5.22±1.05a      | 5.11±0.02a       | 6.00±0.75a       | 5.83±0.62a      | 5.33±0.65a |
| RF2%           | 5.89±0.28a      | 5.39±0.04a      | 6.17±0.15a       | 6.56±0.98a       | 6.22±0.48a      | 5.67±0.02a |
| RF4%           | 6.28±0.07a      | 6.17±0.04a      | 6.44±0.29a       | 6.67±0.28a       | 6.77±0.14a      | 6.39±0.41b |
| RF6%           | 6.44±0.29a      | 6.17±0.25a      | 6.50±0.20a       | 6.63±0.20a       | 6.72±0.49a      | 6.17±0.20b |

** Means in the same column with different superscripts are significantly different (p < 0.05).

4. Conclusions

Incorporation of rice flour in reduced-fat sausage could improve functional properties including moisture retention and binding that could be due to the interaction between thickening agents and meat protein to form stable complexes. Addition of rice flour at 4% and 6% in reduced-fat sausage provided more significant effect in reducing cooking loss, improving emulsion stability and overall acceptability compared to control. In general, the incorporation of rice flour up to 2% in the formulation could successfully improve the texture and sensory evaluation. Rice flour could be a valuable alternative for improving the quality and sensory of emulsified meat product.

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