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Packaging materials and storage conditions affect nutritional quality of blended guava nectar

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ABSTRACT

The present study was carried out to elucidate the influence of packaging materials and storage conditions on changes in nutritional quality of blended guava and coloured grapes nectar (50:50) during 4 months of storage. Blended juice was packaged in amber colour as well as transparent glass bottle followed by stored at low (6±1°C) and ambient temperature ((28±2 °C)). Total soluble solid (TSS), titratable acidity (TA), reducing sugar (RS) and total sugar (TS), total phenolic content (TPC), total anthocyanin (TAC), ascorbic acid (AA), antioxidant activity (AOX) and sensory evolution were recorded in freshly prepared blended juice and during the 4 month of storage. The result showed that the TSS, acidity reducing and total sugar increased whereas antioxidant, ascorbic acid, phenolic and total anthocyanin content decreased during the storage with respective of packaging material. Losses of ascorbic acid, phenolic and total anthocyanin content were higher in blended nectar which was stored at ambient temperature as compared to low temperature. The ratio of 50:50 of guava and coloured grape blends is most acceptable for the preparation of anthocyanin rich guava nectar with maximum retention of anthocyanins, flavor and other nutrient.

Keywords: Amber, Transparent, Cold storage, Anthocyanin, Antioxidants, Sensory Evolution

INTRODUCTION

Tropical fruits are gaining popularity due to their potential health benefits and exotic flavours among the tropical fruits, guava and grape have a good production potential and suitable for the production of a variety of products with good taste and flavour. Guava contains very high amount of vitamin C which helps in improving immunity and protects us from common infections and pathogens. Guava is widely cultivated all over the tropics and sub-tropical area of India. Its fruit consists of 20 per cent peel and 50 per cent pulp and remaining portion as seed core (Wrolstad *et al.,* 2006). Guava fruit normally consumed as fresh as a dessert fruit due to excellent flavour, high digestive and nutritive value, high palatability and availability. Guava is often marketed as 'super-fruits' which has a considerable nutritional importance in terms of vitamins A and C. The high content of vitamin C (ascorbic acid) in guava makes it a powerhouse in combating free radicals and oxidation that are key enemies to many degenerative diseases. The high national production, the high rate of perishability and lack of appropriate technology are some of the major factors responsible for the high amount of waste in the fruit processing sector (Queiroz, 2006). In other hand, ripe guava is highly perishable when stored at room temperature but it can be processed into various commercial products, including pulp, paste, canned slices in syrup and juice. Among these products, guava juice has become economically important in the market heat summer season. The consumption of tropical fruit juice like guava is currently growing because it is natural rich in nutrients and used as an alternative to other beverages such as soft drinks, tea and coffee (Akesowan and Choonhahirun, 2013). Coloured grapes have excellent pigment colour due to the presence of anthocyanins, phenols and betalains in their pulp. Both anthocyanins and phenols are the promising antioxidants. These antioxidants have excellent therapeutic properties and reduce the incidence of a wide range of diseases or disorders associated with free-radicals or radical oxygen species (ROS) like Parkinson's disease, hypertensive cerebra-vascular injury, cataract genesis, ocular hemorrhage, atherosclerosis, various types of cancers, gastro-intestinal disorders hypoxia, disorders related to alcoholism and ultimately ageing

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(Robertson, 2009). Fruit beverages are increasingly gaining popularity throughout the world as well as in India due to nutritive and therapeutic value over synthetic beverages, as synthetic drinks contain only water (about 88%), carbohydrates (about 12%) and provide about 48 Kcal. Thus, fruit-based beverages are far more superior than many synthetic drinks. If synthetic drinks are replaced by fruit beverages, it would be a boon to the consumers as well as to the fruit growers. Fresh fruits have limited shelf life due to the perishable nature of the fruits, they require immediate processing to avoid post-harvest losses. Therefore, it is necessary to utilize the fruits for making different products to increase their availability over an extended period and to stabilize the prices during the glut season. Hence among the beverages, nectar is one of the most important of fruit beverage, which is contains at least 20% fruit juice/pulp and 15% total soluble solids and about 0.3% acid. It is not diluted before serving (Khurdiya and Sagar, 1991). Packaging constitutes one of the most important considerations to make in the value-added chain of in the food or agro-processing industry. Packages primary functions are to contain, protect and preserve products during distribution, storage and handling. Hence, the objective of this study to elucidate the effect of packaging materials and storage condition on nutritional quality of blended guava nectar. Beverages are packaged to keep maintaining quality and freshness, to appeal to the consumers and it make convenient for storage and distribution (Roy and Singh, 1979).

MATERAILS AND METHOD

Procured of raw materials

The studies were conducted in the Division of Food Science & Postharvest Technology, Indian Agricultural Research Institute, New Delhi The fully mature fruits of guava *cv*. 'Allahabad Safeda' and colored grape (*cv.* 'Pusa Navrang') were procured from the Division of Fruits and Horticultural Technology, IARI, New Delhi from the main fruit orchard.Both cultivar was harvested at commercial maturity stage, followed by sorted fruits carefully were packed in CFB boxes (513 x 300 x 240 mm) and transported to division lab within 24 hr and stored at low temperature 6±2 °C with 80–90% relative humidity till further study.

2.2 Fruit juice preparation

The collected guava and coloured grapes fruit sorted to eliminate damage and diseased fruit in order to obtain sample healthy, uniform size for further processing purpose. Fruit were used for washing with tap water to remove the adhering dirt particle from the outer surface of the fruit and followed by went for the processing laboratory for extraction of pulp and juice. The flesh of the fruit was cut into halves with a stainless steel knife, the seeds were removed through a sieve, followed by juice was extracted with the help of fruit pulper (Bajaj Process Pack Limited, Noida, Uttar Pradesh). Whereas grape berries were chosen for their uniform shape, size, and color, as well as free from insects, pests, and diseases. However, harvested berries were separated from rachis. And similar unit operation was used for the extraction of grapes juice which is followed in guava. for juice extraction using a fruit pulper (500-4000 $IbIn^2$ Johnston Automation Co. New Delhi) followed by, the extracted juice was sieved to remove the seed and skin from the juice. However, in the previous studied us were standardizing the extracted juices of both the fruits were blended with defined proportion of 60:40, 50:50 and 40:60 (Guava: coloured grape). In which, the combination having 50:50 proportion was found better for the maintaining good amount of nutritional quality as compared to other blend proportions with respect of packaging material and therefore it was used for further study.

METHODOLOGY AND OBSERVATION RECORDED

During storage period of 4 months from two different packaging material (Amber colour bottle and transparent bottle). Observations on various physicochemical parameters were recorded at each 1 month of interval. Extracted juice from the both fruit taken with three replicates for further analysis. The estimation of titratable acidity (%), ascorbic acid (mg/100), total monomeric anthocyanins (mg/mL), total sugars (%) and reducing sugars (%) was carried out by Ranganna (1999).

Determination of sensory properties

The sensory assessment of the extracted juice was done with the help of a sensory panel of 10 – semi-skilled participated and rating was calculated on 9factor hedonic scale (1 = dislike extremely, 9 = like extremely), Extracted juice/ pulp evaluated through panel of ten semi-skilled inclusive of post graduate students of the division of Food Science and Postharvest Technology at ICAR IARI New Delhi (Álvarez and Barbut, 2013).

Determination of TSS, RS, TS TA and AA

A digital handheld device was used to measure the total soluble solids (TSS) of the samples. Add a drop of filtered juice on a hand refractometer at ambient temperature 20 ºC. where the prisms indicated were recorded, (range 0-50º Brix ATAGO make; logo Japan). RS were estimated by crushing weighed quantity of fruit and making known volume followed by titration against a known volume of Fehling solution (A&B), using methylene blue as indicator. The appearance of brick red precipitate was noted as end point. The results were expressed as %.A known weight of sample was taken and clarified by the addition of 2.0 mL of 45% lead acetate. Followed by lead acetate was neutralized with addition of 2.0 mL of 22% potassium oxalate solution. The clarified solution was made to a known volume with distilled water and filtered. The TS were estimated by hydrolyzing the clarified solution with 5 ml of concentrated hydrochloric acid (HCl) for 48 hat ambient temperatures. The hydrolyzed solution was neutralized with alkali (40% NaOH) and it was titrated against Fehling solution using methylene blue indicator and it was expressed in % (Ranganna, 1999). Taken of 5 ml aliquot juice to estimate the TA of the juices were titrated with 0.1N sodium hydroxide (NaOH). Phenolphthalein use as an indicator. Acidity was calculated and expressed as % (Ranganna, 1999). AA was estimated with the help of dyes (2,6-dichlorophenol-indophenol). 2 ml each of grape and guava juice were titrated with a solution of 0.1% 2, 6-dichlorophenolindophenol and sample was prepared in 3% metaphosphoric acid. AA concentration was calculated by the pink titration method. The amount of ascorbic acid expressed in mg 100 m L^{-1} (Ranganna, 1999).

Determination of TAC, TPC and AOX activity

The TAC from the juice samples were analyzed by using pH differential method and expressed as mg of cyanidin-3-glucoside equivalent per kilogram of fresh weight. Folin–Ciocalteu reagent method was used to determine the TPC of fruit juice as mg of gallic acid equivalent/100 g (Shahbaz *et al.,* 2014). Total AOX activity of both fruit guava and grape was evaluated using DPPH (2, 2-diphinyl-1-picryl-hydrazyl-hydrate) free radical method as described by Sethi *et al. (*2020).

STATISTICAL ANALYSIS

Based on the design, data from the experiment were analyzed using two-way ANOVA (Analysis of variance) and LSD (Panse and Sukhatme, 1954). Analysis of the data was conducted using SAS (Statistical Analysis System, US) with three replications at significant effect (p < 0.05). However, this is available at ICAR-IASRI, New Delhi-110012.

RESULTS AND DISCUSSION

Effect on TSS

As regarding the quality parameters, TSS content in blended nectar was significantly (p≤0.05) affected by packaging material and storage condition, which was shown in (Table 1**)**, with majority of a significant increasing trend was observed in TSS content of guava and grape blended nectar in respect of packaging material, storage temperature as well as storage period. However, the maximum and minimum total soluble solid content increased was observed by 7.40% and 6.18% at ambient and low temperature condition in amber colored bottle respectively. Whereas in case of transparent bottle, the maximum and minimum value of TSS was observed by 4.02 % and 3.93 % at ambient and low temperature condition during the four month of storage. Overall point of view, the highest and lowest value of TSS was found by 3.78% and 3.13 % in transparent and amber glass bottle respectively. The TSS content increased in nectar during storage, it might be due to the conversion of left-over polysaccharides into soluble sugars. Similar findings were also reported by Deka *et al.*

(2004), who observed that total soluble solids showed an increasing trend throughout the storage period. In other hand, these results are in good agreement with the findings of Sharma and Singh (2005), who was reported that the TSS of lime juice increased with an increase in storage period up to 90 days.

Table 1: Influence of packaging material on total soluble solids (°Brix) of blended nectar of guava and grape (50:50) during storage period

Packaging material	Storage period(month)				
	0	1	2	3	4
AMBRT	15.50[f]	15.51[abcdef]	15.61[abcdef]	15.90[abcd]	16.20[a]
AMBLT	15.50[f]	15.26[ef]	15.50[bcdef]	15.80[abcd]	16.00[abc]
TBRT	15.50[f]	15.40[cdef]	15.50[bcdef]	15.53[abcdef]	16.07[ab]
TBLT	15.50[f]	15.30[def]	15.40[cdef]	15.47[abcdef]	15.60[abcdef]

Effect on titratable acidity (TA)

As shown in Table 2, that there was a significant increase in TA content of guava and grape blended nectar during storage. Nevertheless, amber colored bottles and low temperature significantly preserved the loss in TA. The increase in acidity content was lesser in amber colored bottles (0.37%) than transparent bottles (0.38%). Similarly, the increasing in acidity content was lower by 10.0% at low temperature [LT] (6±1 °C) than at ambient temperature [AT] (28±2°C) by 13.88% during the four months of storage. Overall, higher TA was recorded in nectar packed in amber colored bottles (10%) at low temperature compared to in transparent bottles.

Table 2: Influence of packaging material on titratable acidity (%) of blended nectar of guava and grape (50:50) during storage period

Packaging material	Storage period(month)				
	0	1	2	3	4
AMBRT	0.36[d]	0.37[cd]	0.37[cd]	0.38[bcd]	0.38[bcd]
AMBLT	0.36[d]	0.36[d]	0.37[bcd]	0.39[abc]	0.41[ab]
TBRT	0.36[d]	0.36[d]	0.37[cd]	0.37[cd]	0.38[bcd]
TBLT	036[d]	0.37[cd]	0.37[cd]	0.38[bcd]	0.40[ab]

The increase in acidity of nectar during storage might be due to formation of organic acids by ascorbic acid degradation as well as progressive decrease in the pectin content. It is also might be due to the formation of acids from sugar. The results are also in conformity with the findings of (Bal *et al.,* 2014) who reported a significant increase in acidity of guava nectar during storage. in other hand Similar finding has also been reported by (Pandey and Singh, 1999), (Choudhary and Dikshit, 2006) in guava beverages.

Effect on reducing sugar (RS)

There was a significant increased RS of guava and grape blended nectar during storage condition. The RS content show increasing trend with the increasing of storage period (Table 3). The increase in RS content was more in amber colour bottle (5.68 %) than transparent bottle (5.60 %), similarly the increasing RS content was at low temperature (6.60%) than room temperature (7.9%). In general, no significant difference in storage condition was recorded in blended nectar for four months of storage (Table 3). The increase in RS during storage may be attributed to gradual inversion of non-reducing sugars to reducing sugar by the hydrolysis process. These results are in close conformity with the findings of (Brekke *et al.,* 1976), who studied that the decrease in sucrose content in nectar was correlated with an increase in storage temperature. Tiwari and co-worker (2000) reported that an increase in RS content during storage of the RTS beverages prepared from guava: papaya (70:30) blends. Roy and Singh Roy (1979) reported significant increase in

RS ranged from (27.09 to 33.75%) during storage in bale squash and nectar. Similarly, Kalra *et al.* (1991) reported that in mango nectar.

Table 3: Influence of packaging material on reducing sugar (%) content in blended nectar of guava and grape (50:50) during storage period

Packaging material	Storage period (month)				
	0	1	2	3	4
AMBRT	5.30^{d}	5.50^{bcd}	5.66^{abcd}	5.89^{abc}	6.12^{a}
AMBLT	5.30^{d}	5.50^{cd}	5.60^{bcd}	5.76^{abcd}	6.10^{a}
TBRT	5.30^{d}	5.50^{cd}	5.60^{bcd}	5.70^{abcd}	6.06^{a}
TBLT	5.30^{d}	5.40^{d}	5.50^{cd}	5.66^{abcd}	6.00^{ab}

Effect on total sugar (TS)

There was a significant increase in TS of guava and grape blended nectar during four month of storage period (Table 4). The increase in TS content was higher in amber colour bottle 6.35%), than transparent bottle (14.67%). Similarly, the increase in TS content was lower at LT (15.32%) than at RT (17.19%). A significant increase TS of blended nectar was observed during storage. The increase in reducing sugar as well as TS corresponded to the increase in TSS (total soluble solids) and ultimate decrease in non-reducing sugar in both the beverages during storage period was observed. The variation in different fractions of sugar might be due to hydrolysis of polysaccharides like starch, pectin and inversion of non-reducing sugar into reducing sugar, as increase in reducing sugar was co-related with the decrease in non- reducing sugar. The increased level of TS was probably due to conversion of starch and pectin into simple sugars. The present findings are in alien with the report of Tripathi *et al. (1992)*, who reported that there was a continuous increase in the level of TS (11.2 % to 18.6 %) in pineapple-guava blended RTS, they were also reported that total sugars and reducing sugar increased, with the increase in storage period. Choudhary, (2004) reported that there was an increasing trend of total and reducing sugar in guava nectar and RTS with an increasing period of storage under ambient conditions.

Table 4: Influence of packaging material on total sugar (%) content in blended nectar of guava and grape (50:50) during storage period

Packaging material	Storage period (month) Total Sugar				
	0	1	2	3	4
AMBRT	10.70^{h}	11.70^{defg}	11.89^{cde}	11.95^{bcd}	12.45^{a}
AMBLT	10.70^{h}	11.47^{efg}	11.68^{defg}	11.83^{def}	12.34^{ab}
TBRT	10.70^{h}	11.47^{efg}	11.60^{defg}	11.79^{defg}	12.54^{a}
TBLT	10.70^{h}	11.40^{g}	11.45^{fg}	11.72^{defg}	12.27^{abc}

Effect on ascorbic acid (AA)

The AA content showed a declining trend with the increase in the four month of storage period. The reduction in AA content was lesser in amber colour bottle (3.05%) than transparent bottle (6.08%). Similarly, the reduction of AA (AA) content was lower at Low temperature [LT] (6.12%) than at room temperature [RT] (8.99%). In overall point of view, the minimum reduction in AA content was recorded in nectar stored at LT in amber colored bottles by (7.56 %) and maximum in transparent bottles at RT by (13.78%). A significant reduction in AA content of guava and grape blended nectar was observed during storage (Fig 1). The decreasing trend of AA content with increase in the storage period was found in blended guava and grape nectar and this decrease in ascorbic acid content might be due to oxidation of Vitamin-C by trapped oxygen in glass bottles, which resulted in formation of dehydro-ascorbic acid and also due to the effect of processing, storage time and exposure to light. Similar results were also noted by Mall and Tandon, (2007). Rani and Babu, (2015) who was also reported that the decrease in AA and antioxidant activity during storage of 90 days.

Effect on total phenols (TPC)

There was a significant reduction in TPC of guava and grape blended nectar during storage. The TPC showed declining trend with the increase in storage period. The reduction in TPC was lesser in amber coloured bottles (7.18%) as compare to in transparent bottles (9.27%). Similarly, the reduction in TPC content was lower at LT (7.28%) than at RT (9.16%). In overall point of view, the minimum reduction of TPC in content was recorded in nectar stored at LT in amber coloured bottles (6.33%) and maximum reduction in transparent bottles at RT by (10.29%). TPC of the anthocyanin rich guava nectar decreased with increase in storage period at both the storage condition of AT and LT significantly. So alien our finding, Shahbaz *et al.* (2014) reported that the TPC of pomegranate juice showed a slight decrease in both rooms as well low temperature. In other hand some contrast study investigated that, an increase in the concentration of sinapic acid for 4 months; after which, there was a decrease, a trend similar to the one observed in the present study (i.e., initial increase followed by decrease). Other studies have also reported an increase in the concentration of TPC compounds, particularly p-coumaric acid and ferulic acids in carrot juice during the storage period (Martínez-Flores *et al.,* 2015).

Effect on total antioxidant activity (AOX)

The AOX content showed declining trend with the increase in storage period. The reduction AOX in was lesser in amber colored bottles (9.44%) than transparent bottles (15.03%) Similarly, the reduction in antioxidant content was lower at LT (12.13%) than at AT (12.34%). Overall, the minimum reduction in antioxidant content was recorded in nectar stored at LT in amber colored bottles (14.5 %) and maximum in transparent bottles at AT by (25.83%). Similar this finding, Shahbaz *et al.* (2014) was also reported that the antioxidant content of pomegranate juice showed a slight decrease in both rooms as well low temperature showed by DPPH and ABTS assays. In other hand, similar to our study, antioxidant value losses were less than 30% during storage, and it might be due to polymeric compounds formed during storage which compensated for the loss of AOX due to monomeric anthocyanin degradation (Wilson, 1980).

Effect on total anthocyanin content (TAC)

A significant reduction in TAC of guava and grape blended nectar during the four months of storage. The anthocyanin content showed declining trend with the increase in storage period. The reduction in TAC content was lesser in amber coloured bottles (9.06 %) than transparent bottles (27.07%). Similarly, the reduction in TAC content was lower at LT (18.26%) than at RT (29.4%), In all minimum reduction in content was recorded in nectar stored at LT in amber coloured bottles (15.02 %) and highest in transparent bottles at AT (32.64%). As per our result supported by (Bobbio and Bobbio, 1989) who was reported that, the interplay of anthocyanin and ascorbic acid causes both compounds to degrade, resulting in pigment loss and this also occurs in the presence of amino acids, phenols, and sugars. As a result, anthocyanin and AA degrade simultaneously in blended fruit juice (Table 5).

Table 5: Influence of packaging material on total anthocyanin (CGE/kg of blended juice) content in blended nectar of guava and grape (50:50) during storage period

Packaging material	Storage period (month)				
	0	1	2	3	4
AMBRT	38.60[a]	36.60[abc]	34.20[abcdefg]	32.50[fgh]	28.50[ji]
AMBLT	38.60[a]	37.20[ab]	36.80[abc]	35.20[bcdef]	32.80e[fgh]
TBRT	38.60[a]	35.50[bcde]	33.60[defg]	30.40[hi]	26.00[j]
TBLT	38.60[a]	36.20[abcd]	34.40[abcdefg]	32.40[gh]	30.30[hi]

Effect on sensory evaluation of blended nectar

It is clear from the Table 6 that overall acceptability for nectar stored in Amber coloured bottles was better as compared to transparent bottles. This is due to higher retention of colour and flavour in amber coloured bottles. Although the nectar stored at RT remained acceptable till the end of storage period of four months (OA=

7.2), the colour faded much (score = 6.8). The highest acceptability was recorded for nectar stored in amber coloured bottles at LT which the panelists rated high (OA = 7.8) even after four months of storage. The organoleptic scores were higher up to 30 days of storage there after decreased with increase in storage period. storage. However, the acceptability rate decreased due to colour range and the product was slightly acceptable which might be due to conversion of vitamin C and polyphenol into di or poly carbonyl compounds. Similar findings were observed by Tiwari (2000), in guava and papaya blends. The other possible reasons could be the loss of volatile aromatic substances responsible for flavour and taste which decreased acceptability in storage at ambient condition. The present findings are in accordance with the view of (Jain and Asati, 2004) who reported a decrease in overall acceptability of guava with storage period.

Table 6: Influence of packaging material on sensory evaluation (Overall acceptability) content in blended nectar of guava and grape (50:50) during storage period

Storage temperature	Storage period (months)	Amber colour bottles				Transparent bottles			
		Colour	Flavour	Taste	Overall	Colour	Flavour	Taste	Overall Acceptability
RT	0	8.2^{a}	8.4^{a}	8.5^{a}	8.4^{a}	8.0^{ab}	8.4^{a}	8.5^{a}	8.4^{a}
	1	7.9^{ab}	8.3^{ab}	8.2^{ab}	8.1^{ab}	7.8^{ab}	8.0^{abcd}	7.9^{abc}	8.0^{abc}
	2	7.8^{ab}	7.5^{bcde}	8.0^{ab}	7.9^{abc}	7.5^{abc}	7.8^{abcde}	7.5^{bc}	7.7^{abc}
	3	7.6^{abc}	7.4^{cde}	7.6^{bc}	7.6^{ab}c	7.4^{abc}	7.3^{edf}	7.2^{c}	7.5^{bc}
	4	7.3^{bc}	7.1^{ef}	7.2^{c}	7.5^{bc}	6.9^{c}	6.5^{f}	6.3^{d}	7.2^{c}
LT	0	8.2^{a}	8.4^{a}	8.5^{a}	8.4^{a}	8.0^{ab}	8.4^{a}	8.5^{a}	8.4^{a}
	1	7.8^{ab}	8.2^{abc}	8.3^{a}	8.2^{abc}	7.9^{ab}	8.3^{ab}	8.3^{ab}	8.2^{ab}
	2	7.7^{abc}	8.1^{abcd}	8.2^{ab}	8.1^{ab}	8.0^{ab}	8.2^{abc}	8.2^{ab}	8.0^{abc}
	3	7.6^{bc}	7.9^{abcde}	8.1^{ab}	8.0^{abc}	7.8^{ab}	8.1^{abcd}	7.9^{abc}	7.8^{abc}
	4	7.5^{abc}	7.8^{abcd}e	7.9^{abc}	7.8^{abc}	7.7^{abc}	8.0^{abcd}	7.8^{abc}	7.6^{abc}

In conclusion, the blend ratio (50:50) of guava and coloured grapes retained higher physico-chemical attributes during the 4 months at low temperature (6±1 °C). Additionally, amber coloured bottle showed better results over the transparent bottles with respect to sensory and nutritional qualities of the nectar irrespective of storage conditions. Hence, these beverages could be stored safely up to four months in amber colored bottles at low temperature without losing the quality such as AOX, anthocyanin, phenolics and sensorial properties.

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