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Development and Storage Studies on Ready to Use Spice-Based Paneer

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ABSTRACT

Purpose: The aim of the present study was to develop ready to use spice-based paneer, and evaluate its storage stability studies based on quality, sensory and microbial parameters.

Methods: Paneer was developed according to standard procedure followed at Mysuru Milk Dairy. Spice based paneers viz., fennel seeds, star anise and red chili paneers were prepared with addition of 2% spices. Moisture, ash, carbohydrates, protein, fat, titratable acidity and free fatty acid values in paneers were carried out as per the method of AOAC. pH was measured using microprocessor based digital pH meter. Water activity was measured by Aqualab 4TE water activity meter. Microbiological analysis was carried according to APHA method and Sensory evaluation according to Devaki and Premavalli.

Results and Discussion: The results of the present study demonstrated that moisture content of paneer ranged from 40.9-44.2%, protein 18-20.7%, fat 28-32%, ash 1.33-2%, carbohydrates as lactose 5.94-9.27%, quality parameters – pH from 5-6.2, water activity from 0.89-0.98, titrable acidity from 0.63-1.34%, free fatty acids 0.67-1.90%, overall acceptability ranged from 7.16-8.8 on 9 point hedonic scale.

Conclusion: Control paneer, star anise and red chili added paneer samples had good keeping quality till 30 days and fennel seed added paneer sample could be stored till 25 days under the refrigerated condition.

Originality/Value: The results of the study suggest that, spice based paneers had higher sensory scores compared to control paneer and it also acted as natural preservatives and antimicrobial agents.

Keywords: Fennel seeds; Star anise; Red chili; Nutrient composition; Storage stability; Spice-based paneer

INTRODUCTION

Paneer is a tempting, nutritious and delicious indigenous dairy product obtained by heat and acid coagulation of milk [1]. Paneer may be defined as the product obtained from the cow or buffalo milk or a combination of these two thereof by precipitation with sour milk, lactic acid or ascorbic acid. Paneer represents one of the soft varieties of cheese family and is used as a base material for the preparation of a large number of culinary dishes/snacks. About 5% of milk produced in India is converted into "Paneer" [2].

In comparison to other dairy products paneer has many advantages. The technology of manufacturing and handling of paneer is relatively simple. The equipment required are relatively less sophisticated and less costly. The manufacturing process of paneer is so less time, that consuming considerable amount of milk could be handled in one day. The shelf-life of paneer is reported to be only 6 days at 10°C without much deterioration in quality but the freshness of the product lost after 3 days [3].

Paneer, like other indigenous product is a highly perishable product and suffers from limited shelf life, largely because of its high moisture content [1]. The spoilage of paneer occurs mainly due to the growth of microorganisms, which bring about several physico-chemical changes leading to the development of off-flavour in the product. Over the years, various attempts have been made to increase the shelf life of paneer [4].

Fennel seed has been known since ancient times as a medicinal and aromatic spice, commonly used to flavour breads, fishes, salads and cheeses [5]. Fennel seeds were found to have a hypotensive effect [6], antimicrobial [7], antispasmodic activities [8], anti-hirsutism [9], hepato-protective [10], anti-inflammatory [11], anti-dementia [12], possess pain reliever in primary dysmenorrhoea [13], antiplatelet and antithrombotic [14], immunomodulatory [15], anticancer [16], potential in the treatment of glaucoma [17] and antioxidant [18].

Star anise (*Illicium verum*) is an evergreen small medium sized shrub from the plant family *Illiciaceae* [19]. It is used in a variety of

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dishes from beverages to deserts and savoury stews. Star anise has been utilized widely in Indian dishes, it is a principle constituent of garam masala [20]. Numerous medicinal activities shown by essential oil of star anise include: anti-microbial [21], insecticidal activity [22], anti-cancer activity [23], anti-viral, anti-inflammatory [24], anti-diarrheal activity [25], anti-fungal activity [26], and analgesic, sedative and convulsive activity [19].

Chili (*Capsicum spp*.) is an important commercial crop that is grown all over the world having superfluous nutritional and medicinal value [27,28]. Capsaicin is the main bioactive component present in chili, responsible for their incomparable pungent taste and various health benefits [29]. Bioactive amides affinin and capsaicin present in the chili have shown antimicrobial activity [30]. Other bioactive compounds like Sinapic acid, Ferulic acid, Violaxanthin etc. are present in Chili. Hot red chili peppers may be advantageous component of the diet [31].

Paneer contains numerous nutritional components and varying in composition, it includes moisture, fat, protein, carbohydrates as lactose and minerals. Storage stability depends on many factors such as the quality of raw materials, nutritional composition and condition of paneer making, storage and transport. Keeping quality of paneer varies depending upon the ingredients used, storage temperature and refrigeration condition. Therefore in the present study, an attempt was made to improve the storage period of paneer by adding natural preservatives and antimicrobial agents viz., spices such as fennel, star anise and red chilli flakes, which has proved beneficial in increasing the shelf life of the products and also these spices will be beneficial in improving the taste and enhancing the flavour of paneer which will be bland in taste. The developed paneer was evaluated for nutritional, quality and microbial parameters and the storage period was established in the present study.

MATERIALS AND METHODS

Materials

Collection of milk and other ingredients: Fresh cow's milk, other ingredients such as fennel, star anise, red chili flakes and lemon (source for citric acid) were procured from the local market of Mysuru.

Chemicals: All chemicals and media used in the study were of analytical grade and were purchased from S D fine chemicals limited, Mumbai -30 and nice chemicals (P) Ltd., Kerala, India.

Packaging and Storage of control and different spice-based paneer: Prepared control paneer and different spice-based paneer were packed in low density polyethylene pouches (LDPE), stored at refrigerated condition (4°C) for shelf life evaluation.

Methods

Preparation of control paneer: Paneer was prepared as per the procedure led down by Bhattahacharya et al. [3]. Milk standardized to 5% fat and 8.5% SNF was heated upto 85°C (the desired temperature for coagulation). The milk was heated and then cooled to 70°C to 75°C. Freshly prepared coagulation was heated to 70°C prior to addition in order to maintain the coagulation temperature (70°C), then 2% citric acid solution, the coagulate was added to milk with constant and gentle stirring till curd formation then it was allowed to settle in whey for about 5 minutes. Curd was separated out from the whey by pouring the content over muslin cloth, stretched over an empty vessel. The separated curd was filled into hoops lined with muslin cloth and for about 20-25 minutes. The pressed Paneer was removed from the hoop and transferred to chilled water (4 to 6°C for 2 hours) and then placed on wooden plank to allow loose water for about 10-15 minutes. Then cut the paneer into cubes.

Preparation paneer by the addition of spices

a. Preparation of fennel paneer

Milk was standardized to 5% fat and 8.5% SNF

Heated up to 85°C and then cooled to 70 to 75°C

Crushed fennel (2%) was added to milk

Paneer was prepared according to the procedure followed to develop control paneer.

b. Preparation of star anise paneer

Milk was standardized to 5% fat and 8.5% SNF

Heated up to 85°C and then cooled to 70 to 75°C

Coarsely powdered star anise (2%) was added to milk

Paneer was prepared according to the procedure followed to develop control paneer.

c. Preparation of red chili flakes paneer

Milk was standardized to 5% fat and 8.5% SNF

Heated upto 85°C and then cooled to 70 to 75°C

Red chili flakes (2%) was added to milk

Paneer was prepared according to the procedure followed to develop control paneer.

Organoleptic evaluation

The developed control paneer and spice-based paneers were evaluated for sensory characteristics such as appearance, colour, texture, flavour, taste and overall acceptability, and were served to semi trained panelists for organoleptic evaluation on a 9-point hedonic scale, with score 9 as excellent and score 1 as disliking. Sensory evaluation was carried out by 30 semi trained panel members [32].

Analysis

Moisture content, ash content and free fatty acid (FFA) values in control and spice-based paneer were carried out as per the method of AOAC [33]. Carbohydrates, protein and fat contents were carried out as per the method described by AOAC [34]. Titratable acidity was carried out as per the method of AOCS [35]. pH was measured using microprocessor based digital pH meter (Cyber Scan, Model PH 1500, Eutech Instruments, India). Water activity was measured by Aqualab 4TE water activity meter. Microbiological analysis was carried according to APHA [36] and data were transformed into logarithms of the number of colony forming units (CFU/ml).

Statistical analysis

The data obtained for all the parameters and effect of storage on them was statistically analyzed through student t-test to see the critical difference at 5% level of significance using CPCS1 software.

RESULTS AND DISCUSSION

Nutritional composition of control and different spice-based paneer

Control and all the different spice-based paneer were evaluated for nutrient composition. Control paneer had moisture content of 41.2%, protein 18%, fat 32%, ash 1.66% and carbohydrates 7.14%. Fennel seed paneer had moisture 40.9%, protein 20.5%, fat 28%, ash 1.33% and carbohydrates 9.27%. Star anise paneer had moisture 41.4%, protein 20.7%, fat 28%, ash 2% and carbohydrates 7.9%. Red chili flakes paneer had moisture 44.2%, protein 19.2%, fat 29%, ash 1.66% and carbohydrates 5.94%. Developed red chili flakes paneer showed lesser in carbohydrates, higher in moisture. This is because of the addition of blanched red chili flakes paste increased the moisture content. According to Shivakumar et al., [37] control paneer had moisture 50.74%, protein 22.8%, fat 28.88% and ash 1.60%**.** When compared with the literature, the results which obtained from the present study showed that there was significant decrease in moisture in control, fennel paneer and star anise paneer. Protein content was higher in all the spice-based paneer and there was reduction in fat content when compared with the control (Table 1).

Storage studies of control and spice-based paneer

Shelf life of any product shows its ability for being stored for a definite period of time without any deteriorating effects on its quality parameters. Storage life of developed products indirectly shows the market life of the product. Now a day there has been increasing trend of the consumers about foods free from chemical preservatives because of their possible toxic effect in human beings. The consumers are also demanding foods with long shelf life and absence of risk of causing food borne diseases. There is an increasing demand for foods containing natural ingredients. The consumers demand has forced the food industry to exploit potential of natural alternatives for synthetic antimicrobial compounds.

The spices offer a promising alternative in food safety. The spices have been well known for their medicinal, preservative and antioxidant properties [38]. In addition to imparting flavour, certain spices prolong the shelf life of foods due to their bacteriostatic or bactericidal activity and some prevent rancidity in food by their antioxidant activity [39].

The turmeric was suitable for incorporation in paneer and also effective in extending the shelf life of paneer. The addition of turmeric in paneer at the rate of greater than 0.6% significantly decreased the sensory score of paneer. Addition of turmeric at the rate of 0.6% extended the shelf life of paneer up to 12 days on storage at 7±1°C [40].

In the present study, four different types of paneer viz., Control, fennel, star anise and red chili flakes paneers were developed and were evaluated periodically for quality parameters, sensory parameters and microbial parameters. pH, Water activity, titrable acidity and free fatty acids, were considered as quality parameters, sensory evaluation for overall acceptability and microbiological studies were studied in the stored samples. Acceptability scores are helped to establish shelf life of paneer samples.

Changes in quality parameters

The results on the changes in quality parameters during storage of control and different spice-based paneer are given in Table 2. As shown in the table there was decrease in pH from 6 to 5 in control paneer during first month of storage. In fennel, star anise and red chili flakes paneer, pH was decreased from 6 to 5.2, 6.1 to 5.6 and 6.2 to 5.8 respectively. Bhattacharya et al., [3] observed similar decreasing trend in pH in citric acid and cultured whey treated samples of vacuum packaged skim milk paneer and paneer prepared from standardized buffalo milk (pH from 6.60 to 5.80) respectively during storage under room refrigeration temperature (7°C).

There were decreasing trends in water activity from 0.98 to 0.90 in control paneer during first month of storage. In fennel, star anise and red chili flakes paneer, water activity was decreased from 0.96 to 0.89, 0.98 to 0.90 and 0.97 to 0.90 respectively. Das et al., [41] observed similar decreasing trend in water activity from 0.99 to 0.92 when stored at 4°C±1°C.

As shown in Table 2**,** Titrable acidity in control paneer increased from 0.63 to 0.90, in fennel, star anise and red chili flakes paneer, titrable acidity were increased from 0.72 to 1.34, 0.63 to 0.99 and 0.72% to 1.08% of lactic acid respectively. Khatkar et al., [42] estimated the titrable acidity and were found to be 0.16 to 0.43 in 5 days in LDPE when stored at 8±1°C. The increase in titratable acidity of control and spice-based paneer samples is a natural process. The increase in titrable acidity of paneer during storage was also reported previously by various research workers [3].

Free fatty acids values of all control and spice-based paneer got increased. Khatkar et al., [42] resulted that there was increase in free fatty acids in paneer samples from 0.175 to 0.541 in 5 days in LDPE when stored at 8±1°C. The increase in free fatty acids was mostly due to lipolysis action.

Changes in sensory parameters

One of the most important criteria for evaluation of foods is their acceptability which is based on the sensory attributes. In this study, the products were served to a semi-trained group of panelists with 30 panel members. Samples were randomly drawn for each experimental block, coded and served to the panelists. Initially control and spice-based paneer cubes- paneer butter masala and rated for their organoleptic characteristics in the terms of appearance, color, texture and body, flavor, taste and

Table 1: Nutritional composition of control and different spice-based paneer (n=3).

Samples	Moisture (%)	Protein (%)	Fat (%)	Ash (%)	CHO (%)*
Control	41.2 ± 0.03	18.0 ± 0.02	32 ± 0.07	1.66 ± 0.03	7.14 ± 0.23
Fennel	40.9 ± 0.12	20.5 ± 0.21	28 ± 0.02	1.33 ± 0.15	9.27 ± 0.14
Star anise	41.4 ± 0.15	20.7 ± 0.11	28 ± 0.04	2.00 ± 0.13	7.90 ± 0.16
Red chili flakes	44.2 ± 0.06	19.2 ± 0.18	29 ± 0.09	1.66 ± 0.10	5.94 ± 0.18

*CHO – Carbohydrate

overall acceptability on the 9 point hedonic scale. As shown in Table 3, the control paneer butter masala was rated as 8.1 for appearance and texture, and 8.0 for colour, flavor, taste and overall acceptability (OAA). Fennel paneer butter masala had score of 8.6 for appearance, 8.47 for colour, 8.52 for texture, 8.74 for flavor, 8.75 for taste and 8.6 for OAA. Star anise paneer butter masala had score of 8.9 for appearance, 8.54 for colour and texture, 8.7 for flavour, 8.66 for taste and 8.76 for overall acceptability. Red chili flakes paneer butter masala had score of 8.8 for appearance, taste and overall acceptability, 8.78 for texture, 8.94 for colour and 8.81 for flavor. All the four products showed good sensory scores and were highly acceptable. Among all spice-based paneer butter masala, red chili flakes paneer had highest scores for all sensory parameters. Initially acceptability scores of control and different spice based paneer butter masala viz., Fennel, Star anise and red chili flakes was 8, 8.6, 8.76 and 8.8 on 9 point hedonic scale and decreased slowly with the storage period and fennel paneer got spoiled in 25th day of storage.

Changes in microbial parameters

The microbiological quality of paneer depends upon the condition of pre and post manufacturing, handling, packaging and storage of the product. Ratiba et al., [43] studied effect of cardamom, thyme and clove powder on the composition and quality of white cheese made from goat's milk. The authors found that the total viable count (TVC) of control significantly increased throughout the storage period while, count of different treatments decreased upto 15 days. It then slightly increased till the end of storage period. Cardamom, thyme and clove concentration of 0.20, 0.15 and

Table 2: Quality parameters of control and spice-based paneer (n=3).

Quality parameters	Period in days	Control paneer	Fennel paneer	Star anise paneer	Red chili flakes paneer
pH	0	6.0 ± 0.01	6.0 ± 0.17	6.1 ± 0.11	6.2 ± 0.07
	10	5.7 ± 0.04^{c}	5.9 ± 0.15^{b}	5.9 ± 0.12^{c}	6.0 ± 0.09^{c}
	20	5.4 ± 0.12^{c}	5.6 ± 0.11^{c}	5.7 ± 0.11^{a}	5.9 ± 0.12^{b}
	30	5.0 ± 0.11^{b}	5.2 ± 0.12^{a}	5.6 ± 0.14^{c}	5.8 ± 0.10^{a}
Water activity	0	0.98 ± 0.19	0.96 ± 0.16	0.98 ± 0.12	0.97 ± 0.04
	10	0.94 ± 0.02^{a}	0.94 ± 0.19^{a}	0.96 ± 0.19^{b}	0.95 ± 0.06^{a}
	20	0.92 ± 0.18^{b}	0.92 ± 0.11^{a}	0.93 ± 0.13^{a}	0.93 ± 0.11^{b}
	30	0.90 ± 0.36^{a}	0.89 ± 0.12^{c}	0.90 ± 0.15^{c}	0.90 ± 0.01^{a}
Titrable acidity (% Lactic acid)	0	0.63 ± 0.13	0.72 ± 0.13	0.63 ± 0.11	0.72 ± 0.12
	10	0.63 ± 0.18^{a}	0.99 ± 0.12^{a}	0.72 ± 0.17^{b}	0.90 ± 0.06^{a}
	20	0.72 ± 0.21^{b}	1.17 ± 0.14^{a}	0.90 ± 0.16^{c}	0.90 ± 0.05^{a}
	30	0.90 ± 0.12^{c}	1.34 ± 0.15^{a}	0.99 ± 0.17^{a}	1.08 ± 0.06^{b}
Free fatty acid (%/g)	0	0.78 ± 0.23	0.67 ± 0.12	0.78 ± 0.14	0.73 ± 0.03
	10	0.90 ± 0.16^{b}	0.67 ± 0.21^{a}	0.78 ± 0.06^{c}	0.84 ± 0.18^{a}
	20	1.77 ± 0.18^{a}	1.07 ± 0.17^{c}	1.00 ± 0.17^{a}	1.17 ± 0.06^{a}
	30	1.90 ± 0.16^{b}	1.87 ± 0.11^{a}	1.32 ± 0.07^{c}	1.84 ± 0.02^{a}

***Note:** Values with different superscripts are significant difference with initial period at the level, a: p<0.0001, b: p<0.001, c: p<0.01, d: p<0.05

Table 3: Sensory scores of control and flavored Paneer butter masala (n=30).

S. No	Paneer	Period In days	Appearance	Colour	Texture	Flavor	Taste	Overall acceptability
01	Control paneer	0	8.10 ± 0.18	8.00 ± 0.19	8.10 ± 0.18	8.00 ± 0.22	8.00 ± 0.12	8.00 ± 0.10
		10	7.67 ± 0.02^{c}	7.65 ± 0.16^{a}	7.76 ± 0.14^{c}	7.79 ± 0.11^{a}	7.40 ± 0.11^{a}	7.62 ± 0.09^{c}
		20	7.42 ± 0.07^{c}	7.35 ± 0.13^{a}	7.35 ± 0.07^{c}	7.55 ± 0.12^{a}	7.19 ± 0.02^{c}	7.36 ± 0.23^{a}
		30	7.22 ± 0.06^{c}	7.11 ± 0.12^{c}	7.14 ± 0.09^{c}	7.20 ± 0.11^{c}	7.05 ± 0.01^{c}	7.16 ± 0.12^{a}
02	Fennel	0	8.60 ± 0.08	8.47 ± 0.04	8.52 ± 0.01	8.74 ± 0.19	8.75 ± 0.06	8.60 ± 0.04
		10	8.47 ± 0.19^{c}	8.32 ± 0.15^{c}	8.42 ± 0.06^{a}	8.58 ± 0.12^{c}	8.63 ± 0.09^{b}	8.43 ± 0.05^{c}
		20	8.37 ± 0.08^{c}	8.25 ± 0.07^{b}	8.27 ± 0.11^{b}	8.35 ± 0.19^{b}	8.35 ± 0.14^{a}	8.30 ± 0.07^{a}
03	Star anise	0	8.90 ± 0.05	8.54 ± 0.13	8.54 ± 0.13	8.70 ± 0.03	8.66 ± 0.09	8.76 ± 0.09
		10	8.62 ± 0.07^{b}	8.35 ± 0.19^{b}	8.30 ± 0.16^{a}	8.50 ± 0.02^{c}	8.20 ± 0.26^{a}	8.55 ± 0.34^{c}
		20	8.37 ± 0.04^{c}	8.15 ± 0.09^{a}	8.12 ± 0.02^{c}	8.33 ± 0.13^{b}	8.18 ± 0.18^{b}	8.42 ± 0.21^{b}
		30	8.25 ± 0.19^{c}	8.08 ± 0.07^{b}	8.02 ± 0.16^{b}	8.22 ± 0.10^{c}	8.02 ± 0.07^{a}	8.07 ± 0.12^{b}
04	Red chili flakes	0	8.80 ± 0.12	8.94 ± 0.11	8.78 ± 0.06	8.81 ± 0.18	8.80 ± 0.12	8.80 ± 0.01
		10	8.49 ± 0.15^{c}	8.78 ± 0.09^{a}	8.55 ± 0.06^{c}	8.34 ± 0.03^{a}	8.59 ± 0.06^{a}	8.54 ± 0.17^{b}
		20	8.15 ± 0.13^{b}	8.47 ± 0.29^{b}	8.35 ± 0.02^{b}	8.20 ± 0,08^{c}	8.25 ± 0.01^{b}	8.38 ± 0.08^{a}
		30	8.05 ± 0.16^{a}	8.03 ± 0.17^{c}	8.24 ± 0.05^{a}	8.10 ± 0.01^{b}	8.09 ± 0.05^{a}	8.14 ± 0.18^{c}

* OAA- Overall Acceptability

***Note:** Values with different superscripts are significant difference with initial period at the level, a: p<0.0001, b: p<0.001, c: p<0.01, d: p<0.05

Table 4: Microbial analysis of developed Paneer (n=3).

Serial number	Samples	Period in month	Standard plate count (CFU/gm)	Coliforms (CFU/gm)	Yeast & Moulds (CFU/gm)
1	Control	0	28×10^3	Nil	24×10^1
		1	75×10^3	Nil	52×10^1
2	Fennel	0	19×10^3	Nil	5×10^1
		1	115×10^3	1×10^1	39×10^1
3	Star anise	0	12×10^2	Nil	3×10^1
		1	51×10^2	Nil	8×10^1
4	Red chili	0	14×10^3	Nil	Nil
		1	40×10^3	Nil	6×10^1

0.20% respectively had the highest effect on TVC especially 0.20% cardamom.

The developed paneer samples were evaluated for its microbial qualities and are represented in Table 4. Singh et al., [44] studied microbial quality of turmeric incorporated paneer for standard plate count (SPC) and yeast & moulds. The rate of change in standard plate count of samples containing turmeric is slower than the samples without turmeric. David [45] examined peanut paneer for microbiological studies such as standard plate count and coliform test. He reported that coliform count was completely absent in the prepared peanut paneer samples. The microbiological analysis carried out in the present study, clearly shows the sterilization of the products and the microbial readings were within the limits of FSSAI. SPC had shown increasing trend with extended storage however on storage at refrigerated temperature, SPC was present in control and all spice-based paneer samples was less in quantity according to FSSAI specification.

Yeast and moulds were present in all spice-based paneer samples. coliform was nil in control paneer, star anise and red chili flakes paneer were stable upto one month at refrigerated temperature (4°C). Fennel paneer got spoiled on 25th days due to microbial growth (mould growth) on the surface of the paneer samples. Fennel paneer samples had less keeping quality under the refrigeration condition. Control paneer, star anise and red chili flakes paneer samples were not spoiled till 30th days under refrigeration condition (4°C), so these paneer samples had good keeping quality more than 30 days under the refrigeration storage condition. Star anise and red chili paneer samples rich in antioxidants and red chili and star anise were not allowed for the microbial growth on the surface of the paneer. The microbiological analysis showed the sterilized condition of the paneer, coliform was nil, which was reflecting the safety of the product.

CONCLUSION

It is concluded from the study that addition of spices such as fennel seeds, star anise and red chili improved the organoleptic parameters of Paneer cubes. Protein was increased and fat was decreased in all spice-based paneer samples when compared to control paneer. Among the spice-based paneer studied, red chili-based paneer was found to be the best spice-based paneer due to its storage stability and overall acceptability scores. Quality parameters such as titrable acidity and free fatty acids got increased; water activity and pH got decreased in all control and spice-based paneer. The paneer samples were stable for 30 days at refrigerated condition. Addition of spices not only improved the flavor and taste of the paneer samples, it also acted as natural preservatives and antimicrobial agents.

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