

New Food Product Development by Incorporation of *O. bracteatum* Extract to Produce Memory Enhancing Milk Toffee

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Abstract

*Product development is the life-blood of companies and societies. In the present study a new nutraceutical (memory enhancing) milk toffee was produced by incorporating of Eastern Medicine *O. bracteatum*. Among the various combination, the *O. bracteatum* mixed toffee containing 120mg/5g was found to be better for its nutritional quality and sensory evaluation. The addition of 120mg/5gm of *O. bracteatum* to produce milk toffee imparts memory enhancing impact on the children according to the daily dose of the Eastern medicine. The metallic coated polythene wrapper was used to wrap the final product, 50-micron polythene bags were used to pack and were kept for 6 months at $27 \pm 2^\circ\text{C}$ and were refrigerated at $5 \pm 2^\circ\text{C}$. The chemical changes in the final product containing *O. bracteatum* during storage have shown that the moisture content is decreased from 6.9 to 6.6 % and 6.9 to 6.8%, TSS is increase from 82.7 to 85.7°Brix and 82.7 to 85.1°Brix and titrable acidity from 0.17 to 0.15% and 0.17 to 0.16% at ambient and refrigerated condition respectively. The sensory evaluation shows that the colour was changed from 8.6 to 7.9 and 8.6 to 8.1, texture 8.7 to 7.9 and 8.7 to 8.1, flavour 8.5 to 7.9 and 8.5 to 8.0, taste 8.8 to 8.1 and 8.8 to 8.2, and overall acceptability (9-point Hedonic) was 8.65 to 8.0 and 8.65 to 7.9 at ambient temperature and refrigerated conditions respectively. The results after 6 months indicated increase in TSS, while acidity and moisture content were decreased. The decrease in sensory quality during storage period, however the final product was found to be acceptable even after 6-month storage. There are several confectionery products sold in the market and are happily consumed by the children (6 to 13 years of age) and few of them have medicinal properties, but no milk toffee with memory enhancing properties has been reported in the literature earlier. Therefore, the introduction of this first ever memory enhancing toffee in the market will improve the health of children as well as the product cycle of the industry. It also has the market competent price therefore the product is likely to gain the attraction of consumer and improves the market trends.*

Keywords: New food product development, toffee, memory enhancing, Eastern Medicine

Introduction

Eastern (Unani) medicines are a traditional way and considered as an important source of treatment of various diseases in Pakistan (Hussain, Saeed *et al.* 2006, Bodeker and Ong 2005). These herbal medicines have successfully been used to treat respiratory diseases, liver dysfunction, gastrointestinal and urinary tract infections, genital diseases, cardiovascular diseases and even for brain dysfunction (Ahmad, Rehman *et al.* 2010). *Onosma Bracteatum* (*gaozaban*) belongs to the family *Boraginaceae*. *O. bracteatum* is used as a key ingredient in many Eastern medicines and Ayurvedic forms. It contains lipids, carbohydrates, alkaloids, phenolic compound, aliphatic ketones, naphthazarines and naphthaquinones. The dried leaves and flowers of the wall of *O. bracteatum* forms a medicine, which is used as a tonic, rejuvenating, laxative, diuretic and provide a soothing effect in the internal part of organs of the digestive tract. It helps as a

spasmolytic (Kumar, Kumar *et al.* 2013). *O. bracteatum* has been proven to possess marked effect on mental alertness, behavior as well as it also possesses memory enhancing effect. Due to its memory enhancing effect a product Khamera gaozaban is generally given to the school going children for mental alertness and to augment their memory (Anser, Najam *et al.* 2014). *O. bracteatum* contains pharmacologically active substances with anti-diarrheal and antimicrobial properties (Gautam, Bithel *et al.* 2017). *O. bracteatum* appeared to have magnificent antioxidant properties and is reported to have anti-anxiety and depression reducing impact (Asif, Hayee *et al.* 2019). *O. bracteatum* is found useful in asthma, bronchitis, heart diseases, diuretic, alterative, syphilis as well as leprosy (Kirtikar and Basu 1999, Menghani, Sudhanshu *et al.* 2011).

Keeping in view the medicinal, especially memory enhancing effect of *O. bracteatum*, and reluctant behavior of children to take Eastern medicine, in the current study, a new milk toffee product was formulated and produced aiming to contain appropriate amount of *O. bracteatum* for augmenting memory of the school going children. Newly established toffee is not only capable of fighting with various diseases but it is also ready to compete in market due to its taste and cheap cost. Establishment and marketing of such toffee can be an attractive way to treat such common diseases and to enhance the memory of children. The neuro-pharmacological screening and market research may further be carried out expanding the area of study.

Methodology

Chemicals and Glassware

Analytical grade chemicals were used for analysis. All glassware was pre-rinsed with 10% HCl followed by de-ionized water. The ingredients of milk toffee including cane sugar (local), corn syrup (local), milk powder (Nastle Nido), hydrogenated fat (local) and salt (National Foods) were purchased from super market.

Collection of *O. bracteatum*

O. bracteatum was purchased from an herbal store located at Nazimabad, Karachi. Identification of the plant was done by the Botany Department of the University.

Preparation extracts

The dirt and other possible impurities were removed by washing the sample with distilled de-ionized water. Drying of *O. bracteatum* was carried out at room temperature for fifteen days. The dried plant was grinded into powder and was kept in sterile plastic bags before taken for further processing.

Plant Extract

Extract was obtained by cold water extraction. *O. bracteatum* powder was carefully weighed (60g) and soaked in cold distilled water (300mL). Electric rotator at 200 rpm was used to shuck the soaked *O. bracteatum* for 24 hours. The suspension was then filtered through Whatman filter paper. The suspension was then centrifuged for 7 minutes at 4400rpm three times. The supernatant was collected and diluted to 1000mL to obtain 8mg/mL *O. bracteatum* (Zeb, Sajid *et al.* 2015).

Packaging materials

Metallic coated Polyethylene and butter paper were used to wrap the toffees and 50-micron Polyethylene bags were purchased from market.

Preparation of toffee

Four types of toffees were prepared using 0, 80, 120 and 160mg/mL of *O. bracteatum* and constant amount of other ingredients such as sucrose, corn syrup, cream, milk powder, butter, salt and water. The extract was mixed with other ingredients into stainless steel container. The mixture was then heated to obtain 80°Brix TSS content. The mixture was then mixed with a little amount of salt (dissolved in water) and heated to obtain 82–83°Brix TSS. The heated mass was spread on 1.5 to 2.5 cm stainless steel cubes molds (smear with fat). The heated mass was cooled for three hours to solidify.

Organoleptic Evaluation

Standard 9-point hedonic scale procedure was used to carry out the sensory (organoleptic) evaluation of the finally produced toffees (Amerine, Pangborn *et al.* 1965). The mean score of each quality parameter such as overall acceptability, colour, texture, flavor and taste was recorded which was analyzed by 10 trained/and semi trained panelist.

Packaging Material and Storage

The metallic coated polythene wrapper was used to wrap the final product, 50-micron polyethylene bags were used to pack and were kept for 6 months at $27 \pm 2^\circ\text{C}$ and at refrigerated temperature $5 \pm 2^\circ\text{C}$.

Physical and Chemical Analysis

Physical tests of the final product such as moisture, water activity, pH, ash, colour and chemical analysis such as sucrose, reducing sugar and fat contents, titrable acidity, peroxide value, crude protein, crude fiber and carbohydrates were carried out according to the standard methods of AOAC (Helrich 1990). The texture was analyzed by texture analyzer, and the calorific value was estimated by Bomb Calorimeter.

Microbial Analysis

Standard Plate Count (SPC) method was used to record microbial counts. Petri dishes with Potato Dextrose Agar (PDA) as the cultivation medium were kept for incubation at $37 \pm 5^\circ\text{C}$ for 2 days. The colonies were counted by colony counter.

Result and Discussion

Table 1: Standardization of Toffee Recipe

Parameters	Control	Sample1	Sample2	Sample3
<i>O. bracteatum</i> extract (mg/5g) toffee	0	80	120	160
Organoleptic acceptability score*	8.50	8.20	8.65	8.15
Remarks	Control	Not Selected	Selected	Not Selected

* Nine point hedonic scale:

Keeping other ingredients constant 80, 120, and 160mg *O. bracteatum* were incorporated in 5g of memory enhancing milk toffee to establish three different compositions. The sensory analysts recommended the sample with 120 mg/mL

O. bracteatum. As compare to the control sample, both had almost the same taste, texture and mouth feel but were differ in colour, the sample with *O. bracteatum* was darker in appearance. It can be revealed from the sensory results that the acceptability of milk toffee with *O. bracteatum* was found to be above average.

Table 2: Sensory score of fresh toffees

Parameters	Control	Sample
Colour	8.4±0.02	8.6±0.01
Texture	8.4±0.04	8.7±0.03
Flavour	8.5±0.01	8.5±0.02
Taste	8.7±0.03	8.8±0.03
Organoleptic acceptability score*	8.50±0.03	8.65±0.03

* Nine point hedonic scale:

The sensory score of fresh toffee has been summarized in Table 2. The colour of final product was observed to be 8.4 and 8.6 for control and sample prepared with mixing *O. bracteatum*. This might be due to bronze colour formation by *O. bracteatum*. The score for texture for control and sample prepared with mixing *O. bracteatum* was 8.4 to 8.7. Flavour score for control and sample prepared with mixing *O. bracteatum* was found to be similar i.e. 8.5. The taste scores for control and sample prepared with mixing *O. bracteatum* was observed as 8.7 and 8.8. Both of the products control and sample prepared with mixing *O. bracteatum*, were found to be acceptable. The composition and better sensory scores of both of the products might have made both of the product acceptable.

Table 3: Physical Analysis of the Fresh Toffees

Parameters	Controlled sample	Sample with <i>O. bracteatum</i>
Water Activity (A_w)	0.5	0.58
pH	6.8±0.01	6.8±0.01
Texture Analysis (deformation N)	4.39±0.32	4.35±0.41
Colour	Light bronze	Bronze
Calories (kcal/5g)	143.6±1.2	147.14k±1.9

The physical analysis of fresh controlled and the sample containing *O. bracteatum* have been recorded in Table 3. Water activity was recorded as 0.56 and 0.58 A_w for control and sample prepared with mixing *O. bracteatum* respectively. The basic reason for migration of moisture in both of the products is the difference in water activity. Rapid migration of moisture is due to increase in water activity in both of the products. The wrapper of the milk toffee protects the milk toffee from moist air absorption.

The results of pH analysis have provided an evident result that since the pH is neutral; it is suitable according to the product.

Hardness is one of the variables that are used to determine the texture profile of the food. Milk toffee in this study is made from milk and fat processes without ageing. The hardness in newton is found 4.35N and 4.39N for control and sample prepared with mixing *O. bracteatum* respectively. The texture of milk toffee is happened to be affected by the additional moisture content, the interaction between milk protein and fat-soluble flavonoids. Moisture and fat content governs the texture i.e. runny, soft, chewy etc. The colour of controlled milk

toffee was light bronze, the addition of aqueous extract of *O. bracteatum* to the milk toffee cause the colour to one tone darker. Colour is one of the important qualities that attract consumers visually (Hutching 2003). The colour of milk toffee is also influenced by the thermal process and flavonoid content of *O. bracteatum*. It has been reported in literature that solution containing flavonoid will appear darker i.e. red, orange or yellow when thermally processed. The food product containing sugar appears to be darker when getting caramelized (Pricina and Karlina 2013).

The calorific values indicate that the milk toffee contains only 6.7% of the calories a child needs in a day. Therefore, it can be easily utilized with the confidence of not gaining weight.

Table 4: Chemical Analysis of the Fresh Toffees

Parameters	Controlled sample	Sample with <i>O. bracteatum</i>
Moisture Content(%)	6.5±0.04	6.9±0.05
Ash(%)	0.77±0.03	0.82±0.02
Reducing Sugar%	37±0.25	36.4±0.62
TSS (°Brix)	83.5± 1.00	82.7± 1.2
Fat Content%	23.6± 0.94	23.8± 0.85
Titration Acidity%	0.18± 0.05	0.17± 0.02
Peroxide Value	< the Detection Limit	< the Detection Limit
Crude Protein%	1.23 ± 0.05	1.35 ± 0.03
Crude Fiber%	0.24 ± 0.01	0.60 ± 0.01
Carbohydrate%	52.76 ± 1.05	55.65 ± 0.94
Microbial Analysis		
Standard Plate Count	< the Detection Limit	< the Detection Limit

The results of chemical analysis of the fresh final product are summarized in Table 4. Food quality can be maintained by maintaining less moisture content in the food commodities; that will reduce the deterioration of food by decreasing microbial growth. In the current study the moisture content is found to be lesser which ascertains the quality of good product.

The ash content in the controlled sample is found to be less than the ash content in the sample with *O. bracteatum*. It is mainly because *O. bracteatum* extract is obtained from a plant source which could be the reason of higher value in the later product. The ash content in both of the samples is found to be in acceptable limits, which make the product with *O. bracteatum* acceptable.

Reducing sugars are derived by hydrolysis of disaccharides. In this study, major sources of reducing sugars are lactose and sucrose in milk toffee. However, glucose, galactose, and fructose may also be produced during thermal hydrolysis. Further free amino acids of milk protein and reducing sugars underwent with Millard reaction resulting in light brown colour; caramelized appearance and sweet-bitter taste. The TSS content of product with *O. bracteatum* (82.7°Brix) was found to be less than of that control sample (83.5 °Brix).

The fat content is found to be within the range of 24% in both controlled and the sample with *O. bracteatum*. It is reported to be in the limit which is imparting texture to the toffee and also playing a part in the mouth feel of the product. The soft texture of milk toffee is due to emulsification of milk fat; therefore, the fat

release is prevented during the thermal process. Milk proteins such as casein cover the fats droplets. Milk proteins are also utilized as functional compounds therefore, they can interact with fat-soluble materials and reduce the loss of their functionality during high temperature and oxidation process (Tavares and Noreña 2019).

The titerable acidity (TA) were found to be 0.18% in the controlled sample and 0.17% in the final product (Table 4). The milk protein positively affects the value of titerable acidity.

Peroxide value is found to be less than the detection limit. This indicates that the product will stay stable for a longer time, and will not get rancid or lose its characteristics.

Plate count value was found to be less than the detection limit. Confections are generally resistant to bacterial growth, but pathogens (if present) survival in the product for a long time is possible (Lund, Baird-Parker *et al.* 2000).

The per day frequency of any medicine is dose and number of times per day the medicine is taken is used to calculate the total daily dose. In the preparation of milk toffee with the incorporation of *O. bracteatum*, the daily dosage was considered as a prime requirement. The daily dose of *O. bracteatum* is 450mg per day for children ageing from 6 to 13. The product contains 120mg per 5g of the milk toffee sample. It can be therefore recommended to consume 3-4milk toffees a day which is adequate to be used by the children for attaining the maximum efficacy of the Eastern Medicine.

The constituents of the product produced in the current study are natural, no artificial colour and flavour is added to prepare the milk toffee. The addition of corn syrup to the composition of product has positive effects on Texture, transparency, gloss and storage stability. Starch improves the water retention that is very important for better shelf life otherwise growth of moulds and some microbes will occur. To ensure the best shelf life of around 6 months, toffee was stored at room temperature and less humid environment.

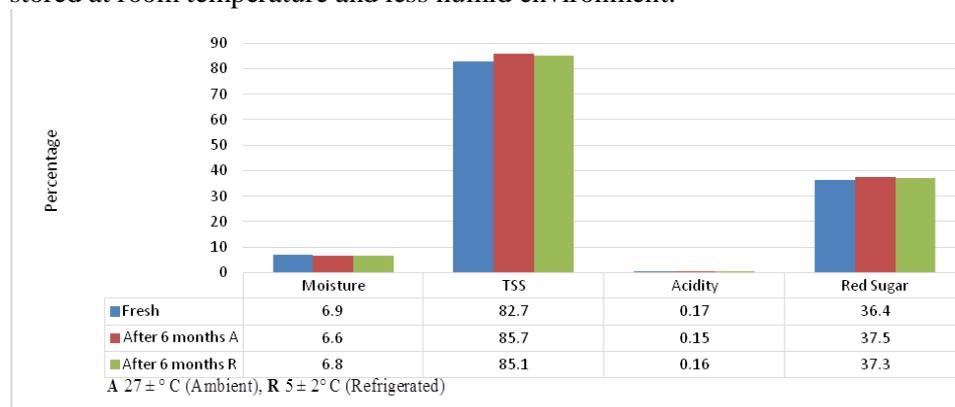
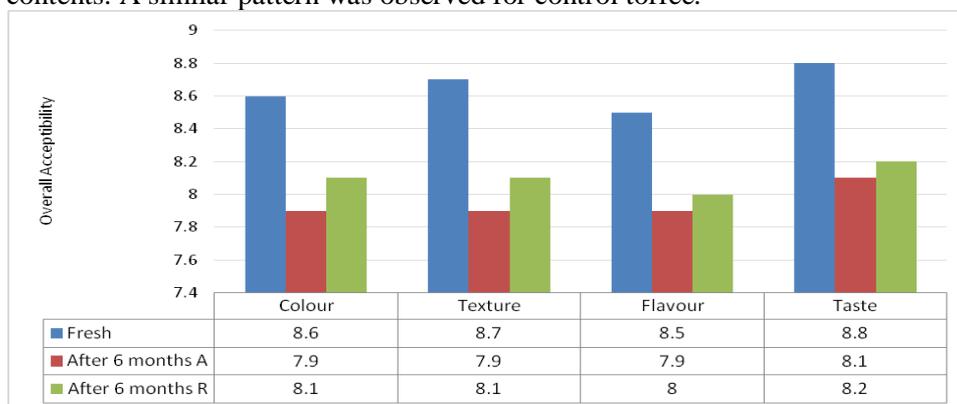


Figure 1: Changes in Chemical Properties of Final Product after 6 month

The chemical constituents of toffee prepared after incorporating *O. bracteatum* after 6 months were analyzed and recorded as Fig 1. The moisture loss of toffee was recorded between 6.9 to 6.6 % and from 6.9 to 6.8 % at ambient temperature

and refrigerated condition respectively. The temperature difference might be a reason of such decrease in moisture content.

The values of TSS of toffee prepared from *O. bracteatum* were observed to be increased from 82.7 to 85.7°Brix at ambient temperature and from 82.7 to 85.1°Brix at refrigerated storage conditions, respectively. The decreased moisture content might be the reason of the increase in TSS in 6-month period. Maximum decrease in acidity value was observed from 0.17 to 0.15 % at ambient conditions and for refrigerated conditions from 0.17 to 0.16 %. Increase in the reducing sugars content was observed from at ambient temperature, and from 36.4 to 37.3% at refrigerated condition after 6-month. Spontaneous hydrolysis of non-reducing sugars might be the main reason behind the increase in reducing sugar contents. A similar pattern was observed for control toffee.



A 27 ± ° C (Ambient), **R** 5 ± 2° C (Refrigerated)

Figure 2: Comparison of Sensory quality of Control & final product after 6 month storage

The sensory properties of toffee prepared incorporating *O. bracteatum* during storage are recorded as Fig 2. A decrease in sensory score of final product was observed from 8.65 to 8.0 during 6-month of storage at ambient temperature, whereas sensory score is found to be decreased from 8.65 to 7.9 for the toffees stored in refrigerated condition. The colour became darker in ambient temperature (8.6 to 7.9) than the refrigerated temperature (8.6 to 8.1). The high temperature at ambient condition might affect the colour and appearance. The colour of milk toffee is influenced by the thermal process and flavonoid content of *O. bracteatum*. A decreasing pattern i.e 8.7 to 7.9 and 8.7 to 8.1 in the texture score of *o. bracteatum* mixed toffee at ambient and refrigerated condition has been observed. A decreasing pattern for flavor score during storage was observed. At ambient temperature, a higher decrease was observed (8.5 to 7.9) than refrigerated storage (8.5 to 8.0). The high temperature at ambient condition might affect the flavor more. The taste score was found to be decreasing from 8.8 to 8.1 and 8.8 to 8.2 at the end of 6 months at ambient temperature and refrigerated condition respectively. The high temperature at ambient condition might affect the taste more.

The acceptability of the product by the panel members after 6 months storage confirms that the minimum changes which might have occurred due to microbes

were within the safe limit for human consumption. The cost of toffee was calculated as per existing prices at the time of the study.

Conclusion

In the present study a new memory enhancing milk toffee was produced by incorporating of Eastern Medicine *O. bracteatum*. Among the various combination, the *O. bracteatum* mixed toffee containing 120mg/5g was found better than other combinations in respect to organoleptic properties and nutritional quality. The addition of 120mg/5gm of *O. bracteatum* to produce milk toffee imparts memory enhancing impact on the children according to the daily dose of the Eastern medicine. The toffees prepared were wrapped in metallic coated polythene wrapper, packed in 50-micron polythene bags and stored at ambient ($27 \pm 2^\circ\text{C}$) as well as refrigerated ($5 \pm 2^\circ\text{C}$) condition for 6 months. The storage studies of toffees packed in 50-micron polyethylene bags indicated that the TSS, reducing and total sugars increased with the advancement of storage period, while moisture and acidity content decreased. The rate of reactions was relatively higher at ambient temperature than refrigerated temperature. Though the sensory quality of toffees also decreased at faster rate during storage period at ambient condition than the refrigerated condition yet the toffees were found to be acceptable even after 6 months at both the conditions. There are several confectionery products sold in the market and are happily consumed by the children (6 to 13 years of age) and few of them have medicinal properties, but no milk toffee with memory enhancing properties has been reported in the literature earlier. Therefore, the introduction of this first ever memory enhancing toffee in the market will improve the health of children as well as the product cycle of the industry. It also has the market competent price therefore the product is likely to gain the attraction of consumer and improves the market trends.

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