Quality Attributes of Therapeutic Tea from Indian Herbs Sweetened with Stevia (*Stevia rebaudiana*)

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Abstract: Eating healthy food is vital for wellness and prevention of disease. Teas are aqueous extractions of crude herbs and one of the most commonly used delivery system for natural health products. Stevia (*Stevia rebaudiana*) is a sweet herb having sweetness 200 to 300 times more than sugar with zero calorific value. Therapeutically stevia is antibacterial, antifungal, anti inflammatory, antimicrobial, antiviral, antiyeast, prevents cavities, cardio tonic, diuretic, hypoglycemic, hypotensive tonic and vasodilator. Hence, sensorily acceptable therapeutic tea sweetened with stevia was formulated, optimized and assessed for various quality parameters. Therapeutic tea was optimized at 10.25% stevia leaves, 7.28% nutmeg, 32.06% arjuna bark, 5.55% licorice and 6.41% of each of ginger, cinnamon, black pepper, fennel, nagarmotha and cardamom. It is recommended that 2.34 g of tea formulation is appropriate to make 100 ml of tea infusion. Herbal tea formulations contained 8.22-9.32% protein, 18.66-18.70% ash, 42.0-45.28% carbohydrates, 72.07-82.25 mg P, 268.25-271.62 mg Ca, 80.30-83.87 mg Mg, 12.8-13.65 mg Fe, 1.82-2.60 mg Cu, 1.37-1.57 mg Zn, 3.28-3.76 mg Mn and 15.84-19.80 mg ascorbic acid per 100 g of tea mix. Bright, sparkling and clear infusion of brown colour with pleasant aroma and taste was obtained from optimized therapeutic tea formulation which would be an alternative medicine for different therapeutic purposes with minimal calories. Microbial quality of the product packed in aluminium foil bag was well up to 3 months storage at ambient temperature.

Keywords: Cinnamon (*Cinnamomum zeylanicum*), Ginger (*Zingiber officinale*), Black pepper (*Piper nigrum*), Nutmeg (*Myristica fragrans*), Licorice (*Glycyrrhiza glabra*), Nagarmotha (*Cyperus scariosus*), Cardamom (*Elletaria cardamomum*), Fennel (*Foeniculum vulgare*), Arjuna (*Terminalia arjuna*).

INTRODUCTION

Prevention of overall poor health and the many lifestyle diseases is of primary importance today. Cancers, heart diseases, hypertension, diabetes and other degenerative diseases do not appear overnight but develop gradually. Their occurrences can be greatly reduced by the choice of our food we choose today which can make a great difference in our health tomorrow. Healthy foods contain nutrients and other substances that stimulate immune system and enhance the functioning of cells, tissues and organs within cardiovascular, respiratory, nervous and other systems. Herbal tea is a non-caffeinated beverage made from the infusion of herbs, spices, or other plant material that exhibited medicinal properties. Traditional Herbal Medicine in a liquid form (extracts/ tea infusions) is more readily absorbed into the system than dried form. Teas are aqueous (water) extractions of crude herbs and one of the most commonly used delivery system for natural health products. Tea is therefore the ideal way to supply the body with enough water and at the same time high-quality active ingredients. Herbal teas are popular because of their fragrance, antioxidant properties and therapeutic applications. These are also consumed for their physical or medicinal effects, especially for their stimulant, relaxant or sedative properties.

In recent years the acceptance of low sugar food is gaining momentum due to increased health awareness. In this respect, stevia (*Stevia rebaudiana*) is an ideal plant [1]. The discovery of stevia led to a revolution in food industry as it reduces the food calorie and facilitates development of dietetic foods. Stevia is a natural sweet herb native of north-eastern Paraguay and has become very popular among Indians due to its zero calorific value and several other medicinal properties. With increasing number of diabetic patients in India it is essential to focus on a sweet herb having sweetness 200 to 300 times more than sugar. Therefore, only small amounts need to be used for sweetening purposes. The stevioside (sweetening agent) content of leaves varies from 4 to 16% [2-6]. Its primary use is as sweetener (non sucrose) to enhance the palatability of foods and drinks [7]. Stevia sweeteners are heat stable to 200 ^{0} C, acid stable and do not ferment thereby making them suitable for use in a wide range of products including baked/cooked foods and drinks [8, 9]. It is used as raw leaf or as commercially processed sweetener [10, 11]. There are more than 150 stevia species but *S. rebaudiana* is the one with significant sweetening properties [12, 13]. It is not absorbed by the intestine and is not metabolized by enzymes of gastrointestinal tract as sugar bonds in

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steviosides are beta glucosidic bond [14-17]. Therapeutically stevia is antibacterial, antifungal, anti inflammatory, antimicrobial, antiviral, antiyeast, prevents cavities, cardio tonic, diuretic, hypoglycemic, hypotensive tonic and vasodilator.

A combination of herbs used for thousands of years in India for soothing and relaxing action to the body. In Ayurvedic and traditional medicine, cinnamon (*Cinnamomum zeylanicum*) is used for a wide range of conditions, including digestive disorders, diabetes, respiratory tract infection and gynaecologic disorders. As part of tea blends, cinnamon improves the taste of less tasty herbs and adds powerful antibacterial power to cold and flu remedies. Cardamom (*Elletaria cardamomum*) is considered as an excellent carminative and general digestive tonic. Cardamom stimulates appetite, soothes the mucous membrane, and relieves gas and heartburn. Indian medicine uses cardamom to treat kidney, bladder and urinary tract disorders like cystitis and nephritis.

Fennel (*Foeniculum vulgare*) seeds contain high amounts of flavonoid anti-oxidants which help to protect the body from cancers, infections, heart disease, high cholesterol, stroke, aging and other degenerative diseases. They contain numerous essential oil compounds which have digestive, carminative, and anti-flatulent properties. Fennel seeds have also been known to ease and regulate menstruation, strengthen the eyes & hair, relax the body and sharpen the memory. Fennel is an effective diuretic and relieves rheumatism, swelling and a potential drug for treatment of hypertension. Nagarmotha (*Cyperus scariosus*) or Nut grass is a pungent bitter-sweet, detoxifying herb and an important medicinal plant in Indian Systems of Medicine. It is an effective killer of intestinal worms and anti-inflammatory, analgesic, diaphoretic, diuretic, desiccant, cordial, stomachic and anti-pyretic medicine. It is widely used in Ayurveda to treat diseases of nervous system where stimulation is required. It helps in excessive thirst, cough, cold, epilepsy, dysmenorrhoea and urinary disorders

Fragrant rich nutmeg (*Myristica fragrans*) is one of the highly prized spices known since antiquity for its aromatic and curative properties. The active principles in nutmeg have many therapeutic applications in many traditional medicines as anti-fungal, anti-depressant, aphrodisiac, digestive and carminative functions. Since ancient times nutmeg and its oil were being used in Chinese and Indian traditional medicines for illnesses related to the nervous and digestive systems. Peppers (*Piper nigrum*) have been used therapeutically in dentistry as an antiseptic for tooth-decay and gum swellings. A decoction of black pepper taken twice a day helps common cold and cough. It promotes proper urination and sweating that helps get rid of harmful toxins from the body. According to Indian Herbal Medicine, Black pepper possesses anti-tumourigenic, immunostimmulatory, stomachic, carminative, anticholestrolemic and known for its strong phytochemical activities. It is a "warming" spice and helps enhance the effects of other herbs throughout the body.

Arjuna (*Terminalia arjuna*) is a medicinally valuable tree that occurs widely in india. Its bark is useful in fracture, ulcer, urethrorrhoea, leucorrhoea, diabetes, anaemia, cardiopathy, fatigue, asthma, bronchitis, tumours, internal and external haemorrhages, cirrhosis of liver and hypertension. Ginger (*Zingiber officinale*) is used in the treatment of colds for its ability to eliminate toxins and raise body heat. Ginger is also a blood thinner and inhibits platelet aggregation which may help reduce angina episodes by lowering cholesterol. It has been used successfully against seasickness, airsickness and even morning sickness in pregnant women. Licorice (*Glycyrrhiza glabra*) is useful for many ailments including asthma, athlete's foot, baldness, body odour, bursitis, canker sores, chronic fatigue, depression, colds and flu, coughs, dandruff, emphysema, gingivitis and tooth decay, gout, heartburn, HIV, viral infections, fungal infections, ulcers, liver problems, Lyme disease, menopause, psoriasis, shingles, sore throat, tendinitis, tuberculosis, ulcers, yeast infections, prostate enlargement and arthritis.

Research carried out by the scientific research institute of Heath & Nutrition, RNGTs, Ukraine, suggested the long term use of Stevia products causes no harm and has beneficial effects for obese or diabetic laboratory animals [18]. Smirnova [19] reported that stevioside is a harmless and nontoxic natural sweetener with high sweetness, good solubility and low energy value. Barathi [20] discussed the uses of stevioside, in bakery, soft drinks, beverage sector, and its household and medicinal uses. Herbal tea based on dried Stevia leaves has been developed by the Russian company Lis-V [18]. Stevia have also been tried in quite a variety of herbal teas [21]. Several low calorie drinks were prepared by Maia *et al*. [22] using different sweeteners including stevia. Looking to the diversified benefits and therapeutic value of all these

herbs as well as to promote physical and mental health, a therapeutic herbal tea was optimized and assessed for various quality parameters.

MATERIALS AND METHODS

Dried Stevia leaves were obtained from department of Plant Breeding and Genetics, College of Agriculture, Indore, India. Ginger rhizome, arjuna bark, cinnamon bark, black pepper seeds, nutmeg fruits, Licorice root, nagarmotha root, cardamom fruit, fennel seeds were obtained from Medicinal and Herbal Garden of the University. To make a therapeutic tea infusion, the herbal tea formulations were added in freshly boiled water, covered and kept for 7 min, strained and served. In order to know the maximum acceptable level of herbs in herbal infusions, the infusions were put up before a panel of 20 experts to evaluate various quality attributes like appearance, colour, taste, flavour, brightness, strength, texture by using 9-point hedonic scale [23]. Finally four formulations i.e. F_1, F_2, F_3 and F_4 were developed using 6.41, 6.85, 7.28, 7.71% nutmeg and 6.41, 5.98, 5.55, 5.12 % licorice respectively. The amount of stevia (10.25%), arjuna bark (32.06%) and ginger, cinnamon, black pepper, fennel, nagarmotha, cardamom are same (6.41%) for all formulations.

The herbal tea formulations were analyzed for moisture, ash, fat and protein [24], carbohydrate by acid hydrolysis process of Hassid and Abraham [25], ascorbic acid by method of Ranganna [26] and Titrable acidity by AOAC [27]. Total Soluble Solid (TSS) was measured by using hand refractometer of $0\text{-}32^0$ Brix range. Ca and Mg by varsenate titration method of Black [28], S content by method of Bardslay and Lancaster [29] and P content by vanadomolybdate method of Koenig and Johnson [30] were determined. The micronutrients Fe, Cu, Zn and Mn were determined by atomic absorption spectroscopy (Yarian Techtron Model aa-120 Australia).

The best combination of herbal tea formulation found during acceptability studies was selected for storage studies. The samples were packed in LDPE (50 gauge), HDPE (100 gauge) and aluminium foil bags and stored for 3 months at ambient temperature ($28 \pm 2\ ^0$C, RH 80 ± 5%). The samples were analyzed periodically for total plate count [31]. All the experiments were carried out in triplicate. Data were analyzed statistically using analysis of variance (ANOVA) and differences among the means were determined for significance at $p<0.05$ using Duncan's multiple range test [32].

RESULTS AND DISCUSSION

Preliminary experiments were conducted to find out the optimum level of each herb for the preparation of herbal infusions. In the primary sensory evaluation test, the panellists judged the infusions prepared from formulations containing 10-12% dried stevia leaves, 6-8% nutmeg, 30-35% arjuna bark, 4-8% licorice, 5-7% each of ginger, cinnamon, black pepper, fennel, nagarmotha and cardamom. They suggested that incorporation of higher amount of stevia in tea infusions adversely affected the taste and aroma of the final product as well as addition of enhanced level of nutmeg and licorice may impart the good flavor to tea infusion .Addition of ground arjuna bark created the problem during preparation of herbal infusion by developing turbidity and therefore adversely affected the texture of tea infusion. The panel members suggested that the small pieces of arjuna bark along with all other herbs should be used for acceptable colour and flavour of tea infusion. The panellists also suggested that the herbal tea must be infused for 7 min to prepare good quality infusion.

On the basis of above findings, finally 4 formulations were developed with different proportions of nutmeg and licorice. The optimum levels of all herbs were finalized by further sensory analysis of tea infusions (Table **1**). The formulation F_3 was the best with regards to all sensory parameters. Its herbal infusion exhibited brown colour with strong flavour and panellists designated it's as strong tea. These results are also supported by Tanaka *et al.* [33] as they reported that hot water infusion of stevia loaded with dark brown colour particles containing pigment of the leaves and soluble polysaccharides. All the tea infusions were smooth in texture and the scores for brightness (8.00) showed no perceivable variation between the formulations. The sensory scores for appearance, taste & flavour of tea infusions showed a steady increase (7.40-8.00) with formulations which may be due to increase in nutmeg and decrease in licorice concentration in the formulations as nutmegs provide aromatic oils such as myristicin, eugenol, elemicin and safrole.

Apart from that, good taste of therapeutic tea was also due to the presence of glycyrrhizin, an active principle of licorice, 50 times more sweetener than sucrose which provide a sweet after taste as stated by Huxley [34]. Good taste and aroma of tea may also be due to the incorporation of cardamom which has a strong unique taste with aromatic fragrance due to its volatile oil accompanied by cineol, terpinene, limonene,

Table 1: Sensory Quality Attributes of Different Therapeutic Tea Infusions

	F_1	F_2	F_3	F_4	SEm	CD at 5%
Sensory Quality (n=20)						
Appearance	7.60	7.70	8.00	7.90	0.29	NS
Brightness	8.00	8.00	8.00	8.00	0.058	NS
Taste	7.40	7.70	8.20	8.00	0.29	NS
Flavor	7.60	7.70	8.10	8.00	0.58	NS
Colour	LB	OB	B	B	-	-
Strength	MT	MT	ST	ST	-	-
Texture	SC	SC	SC	SC	-	-

B-Brown, OB-Orangish Brown, LB-Light Brown, MT-Mild Tea, ST-Strong Tea, SC-Smooth & Clear, F_1-F_4 - As in text.

sabinene and terpineol [35]. The combination of characteristic odour and flavour of ginger volatile oil containing a mixture of zingerone, shogaols and gingerols, sweet flavor from cinnamaldehyde present in cinnamon and cyperine content of nagarmotha essential oil also enhanced the overall flavour of tea infusion. Arjuna bark also gave a triterpene arjungenin, triterpene glucosides I, II and III [36].

The best formulation (F_3) optimized at 10.25% stevia leaves, 7.28% nutmeg, 32.06% arjuna bark, 5.55% licorice and 6.41% of each of ginger, cinnamon, black pepper, fennel, nagarmotha and cardamom. It is recommended that 2.34 g of tea formulation is appropriate to make 100 ml of tea infusion. Savita *et al.* [37] also standardized the quantity of stevia for tea as small as 0.2 to 1.0 g which is in accordance with the present findings. Herbal saffo tea using safflower petals as a major ingredient in combination with other Indian herbs standardized by Satwadhar [38]. Variety of herbal teas were also developed using ginger, licorice, cinnamon, stevia and other herbs [21]. Several formulation of low calorie drinks based on acerola were prepared by Maia *et al.* [22] using different sweeteners including stevia. Lisitsin and Kovalev [18], Smirnova [19], Megeji *et al.* [39] considered stevia to be harmless, natural high potency sweetener with zero calories, nontoxic and suggested the long term use of stevia products whereas Kostina [40] inferred potential use of steviosoide as a sweetener in dairy beverages.

Protein, crude fibre, ash content of therapeutic tea mix increased with formulations (Table **2**). This may be due to gradual increase in the amount of nutmeg in formulations consisting of good amount of all these nutrients. All macro and micro nutrients found to be in increasing order with formulations due to blending of nutmeg with various herbs at various levels. Ozcan and Akbulut [41] analyzed the mineral contents of 31 medicinal and aromatic plants collected from the south region of Turkey and reported the highest values of Ca and P in *F.vulgare*. Savita *et al*. [37] investigated the mineral composition of dry stevia leaves and stated stevia as a mineral loaded ingredient required to protect the body, regulate and maintain the various metabolic process.

Fennel seeds, black peppercorns, arjuna bark and nutmeg are also concentrated with minerals like copper, iron, calcium, potassium, manganese, selenium, zinc, and magnesium. Potassium is an important component of cell and body fluids that helps control heart rate and blood pressure. Manganese and copper are used by the body as co-factors for the antioxidant enzyme, superoxide dismutase. Iron is essential for cellular respiration, red blood cell production and as a co-factor for cytochrome oxidases enzymes. Nutmeg and black peppercorns are also rich in many vital B-complex vitamins including vitamin C, folic acid, riboflavin, niacin, vitamin A and many flavonoid anti-oxidants like beta-carotene and cryptoxanthin that are essential for optimum health. These compounds help the body removes harmful free radicals and helps protect from cancers and diseases. It has also been found that piperine present in black pepper can increase absorption of selenium, B-complex vitamins, beta-carotene, as well as other nutrients from the food.Cinnamon is also an excellent source of manganese and a very good source of dietary fibre, calcium and iron. Both calcium and fibres can bind to bile salts and help remove them from the body. By removing bile, fibres help to prevent the damage that certain bile salts can cause to colon cells, thereby reducing the risk of colon cancer. In addition, when bile is removed by fibres, the body must break down cholesterol in order to make new bile. This

Table 2: Chemical Parameters of Different Therapeutic Tea Formulations

Chemical parameters (n=3)	F_1	F_2	F_3	F_4	SEm	CD at 5%
Moisture %	9.57	9.78	9.68	9.97	0.58	NS
Protein %	8.22	8.45	8.99	9.32	0.58	NS
Fat %	5.40	6.04	6.60	7.00	0.58	NS
Carbohydrate %	45.28	45.04	43.04	42.00	0.58	1.88
Ash %	18.66	18.68	18.69	18.70	0.58	NS
Crude fiber %	11.00	11.44	11.80	12.10	0.58	NS
P mg/100g	72.07	75.67	79.56	82.25	0.58	1.88
Ca mg/100g	268	269	270	271	2.93	NS
Mg mg/100g	80.30	81.31	82.89	83.87	2.93	NS
S mg/100g	167	175	189	200	28.87	NS
Zn mg/100g	1.37	1.48	1.49	1.57	2.87	NS
Cu mg/100g	1.82	2.00	2.25	2.60	0.58	NS
Fe mg/100g	12.8	12.99	13.25	13.65	0.58	NS
Mn mg/100g	3.28	3.45	3.62	3.76	0.58	NS
TSS ^{0}Brix	1.02	0.9	0.4	0.8	-	-
Titrable acidity %	0.005	0.007	0.007	0.008	-	-
Ascorbic acid mg/100g	15.84	17.16	18.48	19.80	-	-

NS-Not significant, F_1-F_4 - As in text.

process can help to lower high cholesterol levels, which can be helpful in preventing atherosclerosis and heart disease [42].

TSS 0.4 to 1.02 ^{0}Brix, titrable acidity 0.005 to 0.008% and ascorbic acid 15.84 to 19.80 mg/100g was observed in all the formulations .The concentration of different herbs did not affect these parameters appreciably. This may be due to lower amount of tea blend used for the preparation of infusions. Maia *et al.* [22] also observed that no statistically significant variation were found for pH, Brix, acidity ratio and reducing and non reducing sugar contents of low caloric drinks prepared from various sweeteners. The microbial count of all formulations increased with increase in storage period in all packaging materials (Table **3**). Uptake of moisture from atmosphere having high humidity also adversely affected the microbial quality of therapeutic tea formulation. Least microbial population (31 x 10^5 cfu/g) was noted in the formulation packed in aluminium foil bags on 90th day of storage. The reason may be due to low water vapour transmission rate of aluminium foil. Cinnamon is also useful as a food preservative to inhibit the growth of common food-borne bacteria such as Salmonella and E coli. The antimicrobial properties of cinnamon are due to eugenol present in essential oil.

CONCLUSION

Tea is an effective vehicle for administering the medicinal components of plant .An acceptable infusion of brown colour with pleasant aroma and taste was obtained from optimized formulation containing 10.25% stevia leaves, 7.28% nutmeg, 32.06% arjuna bark, 5.55% licorice and 6.41% of each of ginger, cinnamon, black pepper, fennel, nagarmotha and cardamom.

Table 3: Microbial Count (10^5cfu/g) of Therapeutic Tea Formulation (F_3) During Storage

Storage Days	Packaging Materials		
	LDPE	HDPE	Aluminium Foil
0 day	30	30	30
45 day	41	32	30
90 day	53	40	31

Aluminium foil bags are more suitable for packaging herbal tea formulations at ambient temperature. There is a need for creating awareness among the people about the nutritional and therapeutic values of stevia along with other herbs. Regular consumption of this tea combination would be therapeutic and relieve ailments including several illnesses.

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