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Research Paper

Formulation of Bittergourd based Recipes for Diabetes and Evaluation of its Phytochemicals

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A B S T R A C T

Bitter gourd (*Momordica charantia* Linn.) belongs to the family of Cucurbitaceae and known as one of the bitterest fruits. This study was aimed to formulate the bittergourd recipes by reducing its bitter taste, to evaluate sensory characteristics of those formulated recipes and to estimate the phytochemicals present in bittergourd recipes. Three varieties of bitter gourd recipes were developed in the current study. The sensory evaluation was done with a five-point hedonic scale. The selected recipes were subjected to phytochemical analysis. The overall acceptability score was significantly higher at 0.01% for bittergourd thuvaiyal followed by bittergourd raita and bittergourd juice. Though recipes namely thuvaiyal and raita were found to have higher acceptability their nutraceuticals levels were less in comparison to the raw bittergourd. Hence, there is necessity to explore the applicability of these plant resources which are rich in phytochemicals may have been beneficial effects of health.

***Key words*:** *Momordica charantia*, Phytochemicals, Total phenols, Flavonoids, Alkaloids, Saponins

A functional food can be a natural food, a food to which a component has been added, a food from which a component has been removed, a food where one or more components has been modified, a food in which the bioavailability has been modified or any combination of these [1]. Some plant functional foods are whole grains, fortified foods, drinks, some dietary enhancements, organic products, vegetables and their items for example green tea, broccoli, grape juice, cabbage, tomatoes, watermelon and psyllium [2]. Bitter gourd (*Momordica charantia*) is an important market vegetable in Southern and Eastern Asia and is widely spread throughout most of tropical Africa [3]. The important bitter gourd growing states are Maharashtra, Gujarat, Rajasthan, Punjab, Tamil Nadu, Kerala, Karnataka, Andhra Pradesh, West Bengal, Odisha, Assam Uttar Pradesh and Bihar [4]. Bittergourd also referred to as bitter melon, balsam pear, bitter apple and bitter African or wild cucumber. Fruits and leaves of most wild Momordica species are consumed as vegetables and have a similar bitter taste and almost identical medical uses. It has been used as a traditional antidiabetic remedy in eastern countries for many years [5]. Bitter gourd (*Momordica charantia* Linn.) belongs to the family of Cucurbitaceae and known as one of the bitterest fruits. It actually originated in India and eventually carried to China during the 14th century [4]. It is tropical and subtropical climber. Bitter gourd received different names such as bitter gourd or bitter melon for its English name and Goya for its Japanese name.

Bitter gourd is a flowering vine in the family Cucurbitaceae. It is a slender, climbing the annual vine with long-stalked leaves and yellow, solitary male and female flowers borne in the leaf axils. Leaves: simple, usually palmately 5-7 lobed, tendrils unbranched or 2 branched. It bears simple, alternate leaves 4-12 cm across, with 3-7 deeply separated lobes. Fruit: ovoid, ellipsoid, or spindle shaped, usually ridged or warty, dehiscent irregularly as a 3 valved fleshy capsule or indehiscent [6]. The young fruit is emerald green, turning to orange-yellow when ripened. On maturity, the fruit splits into three irregular valves that curl backwards and release numerous reddish-brown or white seeds encased in scarlet arils. Seeds and pith appear as white colour in unripe fruits, and red during ripening process. Bitter melon comes in a variety of shapes and sizes [6].

Bitter gourd has good demand due to its special culinary taste and it is also considered to be a good source of dietary fibers [7]. The immature fruits of bitter gourd can be fried, deep fried, boiled, pickled, juiced, and dried to drink as tea [8]. Bitter gourd is anti-diabetic, stimulant, stomachic, laxative, blood purifier and control diabetes. It is antidotal, antipyretic tonic, appetizing and antibilious [9]. The immature fruits of bitter gourd can be prepared in many ways such as frying or cooking as curries. In addition, fruits can be dehydrated, pickled or canned. They are usually blanched or soaked in salt water before cooking to reduce the bitter taste [10]. Bitter gourd contains bitter chemicals like charantin, vicine, glycosides and karavilosides along with lectin, polypeptide-p, plant insulin etc., which are hypoglycemic in action and improve blood glucose levels by increasing glucose uptake and glycogen synthesis in the liver, muscles and fat cells [11]. This study was aimed to formulate the bittergourd recipes by reducing its bitter taste, to evaluate sensory characteristics of those formulated recipes and to estimate the phytochemicals present in bittergourd recipes.

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MATERIALS AND METHODS

Formulation of recipes and sensory evaluation

Three varieties of bitter gourd recipes were developed in the current study. They were bittergourd thuvaiyal, bittergourd raita and bittergourd juice (at different dilution levels). Recipes were standardized with the help of guidelines of FSSAI. Three variations of each recipe were carried out before they were standardized. During the process of standardization, the quantity of ingredients, cooking temperature and time, serving size, recipe yield, equipment and utensils used, recipe title and recipe category were also finalized and documented to maintain consistency during every preparation.

Sensory evaluation of bittergourd based recipes

Sensory evaluation is a scientific method used to analyze and interpret consumers acceptability perception towards a newly developed product through five senses of sight, smell, touch, taste and hearing [12]. The sensory evaluation was done with a five-point hedonic scale with a group of 30 members that constituted post graduate students and staff members of Food Service Management and Dietetics Department. The samples were evaluated by the panelists for flavor, texture, color, odor, appearance, bitterness (taste) and overall acceptability. While evaluating the samples, the sensory practices were followed according to the procedure recommended by International Food Technologist [13]. The coded samples were presented to each of the panelist. Hence, each panel member was given a serving plate with a spoon, cup and a bottle of water to rinse their mouth after each testing to avoid any bias. Instructions were given to the panelists that they have to rinse their mouth with water between tasting of each recipe to remove the taste of the previous food.

Estimation of phytochemicals in bittergourd recipes

Phytochemicals screening of bittergourd incorporated recipes namely Bitter gourd Thuvaiyal, Bitter gourd Raita and Bitter gourd Juice were performed as there is no clinical validation available claiming its nutraceutical effect. Recipes with higher acceptability score in each of the selected recipes of thuvaiyal, raita and bitter gourd juice were subjected to phytochemical analysis which included total phenolics, flavonoids, alkaloids and saponins. The detailed procedure followed for the analysis of each of the phytochemicals are listed below:

Reagents

Quercetin, Folin- Ciocalteu reagent, ascorbic acid, Aluminium chloride, Methanol, sodium carbonate and potassium acetate. The standardized bittergourd recipe namely Thuvaiyal, salad (raita) and raw bittergourd were dehydrated and pulverized following the methods of [14]. 70 g of pulverized powder and 350ml of methanol in the ratio of 1:5 was mixed and maintained for a period of 7 days at room temperature with frequent agitation.

Determination of total phenolics

The total phenolics were determined using Folin-Ciocalteu method adopted by [15]. 1 ml of extract solution was mixed with 2.5 ml of 10% Folin-Ciocalteu reagent. After 5 min, 2.0 ml of 75% sodium carbonate was subsequently added to mixture and incubated at 50^0c for 10 min with intermittent agitation. The sample was cooled and the absorbance was measured using UV spectrophotometer at 765 nm against the blank. Gallic acid was used as standard for calibration curve. The data was expressed as mg/g of gallic acid equivalents in milligrams per gram (mg GAE/g) of dry extract.

Determination of flavonoid contents

The total flavonoid content of crude methanolic extract of bittergourd and its recipes was performed by aluminum chloride colorimetric method adopted by [16]. 1 ml of the extract solution was mixed with 1 ml aluminum chloride ethanol solution and incubated for 10 minutes at room temperature; the absorbance was measured at 415 nm with UV spectrophotometer. Quercetin was used as a standard for calibration curve and the results were expressed as quercetin equivalents (µg QE/mg) of extract.

Determination of alkaloids

The sample extract (1mg) was dissolved in dimethyl sulphoxide (DMSO), added 1ml of 2 N HCl and filtered. This solution was transferred to a separating funnel, 5 ml of bromo-cresol green solution and 5 ml of phosphate buffer were added. The mixture was shaken with 1, 2, 3 and 4 ml chloroform by vigorous shaking and collected solution in a 10-ml volumetric flask and diluted to the volume with chloroform. A set of reference standard solutions of atropine (20, 40, 60, 80 and 100 µg/ml) were prepared in the same manner as described earlier. The absorbance for test and standard solutions were determined against the reagent blank at 470 nm with an UV/Visible spectrophotometer. The total alkaloid content was expressed as mg of AE/g of extract [17].

Determination of saponins

Total saponin determination was done using anisaldehyde reagent. Sample solution was prepared in water. Weighed 10 mg of diosgenin, dissolved in 16 mL of methanol, and add 4 mL of distilled water. Standard solutions of diosegenin (20, 40, 60, 80 and 100 µg/ml) were prepared 80% aqueous methanol. Mixed thoroughly and start pipetting immediately. For total saponins estimation 500µl of sample, 500µl of 0.5% anisealdehyde reagent, were mixed and kept aside for 10 min. Later, 2 ml of 50% sulphuric acid reagent was added and tubes were mixed. Tubes were then kept in water bath with constant temperature of 60º. After 10 min tubes were cooled and absorbance was taken at 435 nm. Same method was followed for standard solution. The amount of saponins was calculated as saponin equivalent from the calibration curve of standard [17].

Statistical analysis

Data were analysed using IBM SPSS 20 software (IBM corp., Armonk. NY, USA). Quantitative data were presented as Mean ± standard deviation. One-way ANOVA (analysis of variation) and post hoc least significant difference test was done to identify the significant difference in the sensory characteristics of bittergourd recipes at $p \leq 0.05$.

RESULTS AND DISCUSSION

Standardization of bittergourd recipes

Each portion of three recipes namely bittergourd thuvaiyal, bittergourd raita and bittergourd juice were prepared thrice. Also, each item in the recipe was standardized in triplicates to assess their consistency in taste, appearance, portion size and yield and presented in (Table 4). The standardized procedure was then documented for quantity of ingredients as indicated in (Table 1-3).

Table 1 Recipe of Bittergourd Thuvaiyal

Ingredients	Quantity
Bittergourd	25g
Bengal gram dhal	2g
Red gram dhal	2g
Red chilli	1g
Coconut	10g
Tamarind	2g
Asafoetida	1g
Curry leaves	1g
Urad dhal	1g
Oil	3g
Salt	2g

Preparation method

- Roast bengal gram, red gram dhal and red chilli in medium flame with constant stirring.
- Roast grated coconut till it turns slightly brown.
- Cut bittergourd into pieces then add water and squeeze it and roast them till it becomes golden brown.
- Add salt and tamarind and grind all the ingredients together to coarse paste.
- Heat oil in a pan and add mustard seeds, urad dhal, curry leaves. Pour it on the top of the thuvaiyal.
- It can be served with rice, dosai, idly etc.

Table 2 Recipe of Bittergourd Raita

Ingredients	Quantity
Bitter gourd	10g
Ash gourd	5g
Cucumber	5g
Cashewnut	2g
Cumin seeds	1g
Small onion	2g
Green chilli	1g
Curd	50ml
Coriander leaves	1g
Salt	1g
Oil	2g
Coconut	2g

Preparation method

- Wash the bittergourd and slit it lengthwise. Remove seeds and cook till it tenders.
- Wash the ashgourd and remove seeds and skin. Chop and cook the gourd till it tenders.
- Grind grated coconut, green chilli, cashewnut, cumin seed into smooth paste.
- Add cooked bittergourd, ashgourd, cucumber and curd and mix well.
- Heat oil in a small pan and add cumin seeds. Pour it on the top.
- It can be served with variety rice, chappathi etc.

Table 3 Recipe of Bittergourd juice

Ingredients	Quantity
Bitter gourd	30g
Water	80ml

Preparation method

- Wash and cut bittergourd into pieces.
- Grind the bittergourd without adding water.
- Add 20 ml of bittergourd extract and make it into 100ml by water

Table 4 Standardisation of bittergourd recipes

Recipe	Cooking time (mins)	Serving portion	Recipe yield (g/ml)		
			Variation-1	Variation-2	Variation -3
Bittergourd thuvaiyal	30	1	83	83	83
Bittergourd raita	20	1	82	82	83
Bittergourd juice	10	1	100	100	100

The (Table 4) presents standardization of bittergourd recipes. The bittergourd recipes were done in three variations. After preparation the yield of the thuvaiyal was 83g and it took 30 mins for the preparation, bittergourd raita in variation I and II yield were 82g, whereas variation III yield was 83g and each portion took 20 mins for preparation. All the three variations of bittergourd juice were prepared for 100ml. To prepare each portion it took 10 minutes.

Sensory evaluation of bittergourd recipes

The bittergourd recipes were evaluated for its sensory characteristics and the data are presented in (Table 5)

Table 5 Sensory evaluation of all Bittergourd Recipes

Components	Bittergourd thuvaiyal	Bittergourd raita	Bittergourd juice
Colour	4.26 ± 0.86	4.13 ± 0.77	4.06 ± 0.86
Appearance	4.13 ± 0.77	4.46 ± 0.73	$4.13 \pm 0.86^{ab**}$
Flavour	3.73 ± 0.82	$4.40 \pm 0.96^{a**}$	$2.93 \pm 0.58^{ab**}$
Texture	3.80 ± 0.80	$4.20 \pm 0.76^{a*}$	$3.56 \pm 0.89^{ab**}$
Taste	4.56 ± 0.97	$4.23 \pm 1.00^{a*}$	$2.76 \pm 1.13^{ab**}$
Consistency	4.96 ± 0.94	4.23 ± 0.81	$3.90 \pm 0.88^{ab**}$
Bitterness	3.53 ± 1.10	3.76 ± 1.00	$3.03 \pm 0.71^{ab**}$
Overall acceptability	4.17 ± 0.89	$3.45 \pm 0.65^{a**}$	$3.01 \pm 0.92^{ab**}$

Values are Mean ± SD of bittergourd thuvaiyal, raita and juice. ANOVA test were used for comparison.
'a' denotes significant difference at 5% level between bittergourd thuvaiyal and bittergourd raita, bittergourd juice,
'b' denotes significant difference at 5% level between bittergourd raita and bittergourd juice.
'*' shows p value <0.005 and '**' shows p value <0.001

The bittergourd juice was prepared in five different dilutions as 1:9 (10 ml of bittergourd juice and 90 ml of water), 1:4 (20 ml of bittergourd juice and 80 ml of water), 3:7 (30 ml of bittergourd juice and 70 ml of water), 2:3 (40 ml of bittergourd juice and 60 ml of water) and 1:1 (50 ml of bittergourd juice and 50 ml of water). Among that the dilution

made with one portion of bittergourd juice and four portion of water was scored the best and it was selected for standardization and further analysis of phytochemicals.

The results of (Table 5), the overall acceptability score was significantly higher at 0.01 per cent for bittergourd thuvaiyal followed by bittergourd raita and bittergourd juice. Among the five points hedonic scale food attributes, the appearance, flavour and texture of bittergourd raita scored higher than that of the other bitter gourd recipes. In contrast the score of food attributes such as colour, taste and consistency was found to be higher for bitter gourd thuvaiyal. Unavoidably, the bitterness of bitter gourd juice was found to be higher in comparison to other two recipes.

Phytochemicals estimation of bittergourd and bittergourd based recipes

Four major phytochemicals like total phenols, total flavonoids, total alkaloids and total saponins were identified and quantified from raw bittergourd and in bittergourd recipes. Among these phytochemicals, total phenols, total flavonoids and total saponins were estimated using aqueous extract while total alkaloids were estimated from nonaqueous extract.

Table 6 Phytochemicals present in selected bittergourd recipes

Recipe	Total Phenols (mg GAE/100g)	Total Flavonoids (mg QE/100g)	Total Alkaloids (mg AE/100g)	Total Saponins (mg/100g)
Raw bittergourd	350	38	172	63
Bittergourd thuvaiyal	303	33	154	58
Bittergourd raita	268	28	142	45

The present data in (Table 6, Fig 1) clearly shows that raw bittergourd was found to have high composition of selected phytochemicals followed by that bittergourd thuvaiyal and bittergourd raita. Raw bittergourd was found to be rich source of nutraceutical with less overall acceptability score (Table 5). The bittergourd juice was not evaluated for its nutraceutical property as it was only the water diluted fraction (1:4) of raw bittergourd. From the sensory evaluation prospective, though recipes namely thuvaiyal and raita were found to have higher acceptability their nutraceuticals levels were less in comparison to the raw bitter gourd.

There are few evidential reports by [18], [19] of sensory evaluation by hedonic scaling respectively by nine point and five-point scale for bitter gourd juice. No other reports to date are available for the other bitter gourd recipes of bitter gourd thuvaiyal and raita. According to reports by [20], the acceptability of the bitter gourd was less among the public mainly due to their bitterness sensation. Hence, to increase the consumption of bitter gourd, alternative recipes of bitter gourd juice and raita can be advocated among the public with high overall acceptability score. Reports are available for the presence of phytochemicals / nutraceuticals both in dry bitter gourd powder [21], [22], [23]. In contrast, the quantification of phytochemicals or nutraceuticals in the bitter gourd recipe is so far not available. Hence, in this study quantification of phytochemicals were done to increase the acceptance of the other bitter gourd recipes stating their health benefits nutraceutical effect.

Bitter gourd

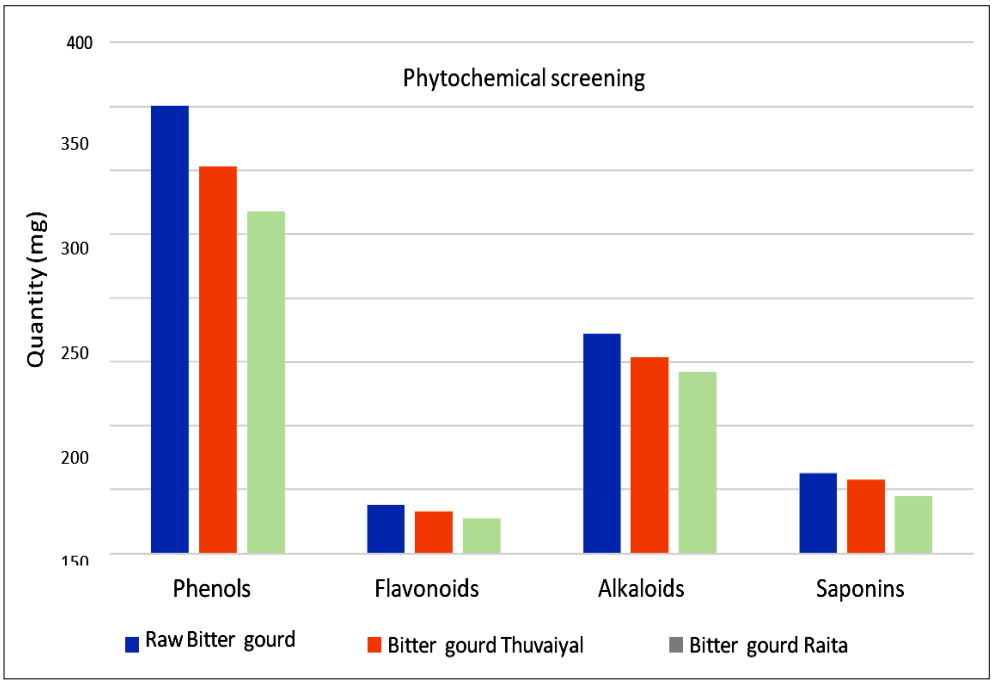

Fig 1 Comparison of Phytochemical content in selected bitter gourd recipes

CONCLUSIONS

The plant-based bio-active compounds have the effective dosage response with minimal side effects, when compared to the synthetic compounds. The medicinal properties of bittergourd may be due to the presence of the active biochemicals and phytochemicals. The study showed that the plants are a source of significant natural therapeutic agents and may be beneficial in diabetes and other health problems. Hence, there is necessity to explore the applicability of these plant resources which are rich in phytochemicals may have been beneficial effects of health.

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