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Biological Functions and Sensory Attributes of Different Skin Colored Bitter Melon (*Momordica charantia* L.) Varieties

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Abstract

Biological functions, sensory qualities, and nutritional potentials of bitter melons were examined using three different colored varieties. Several recipes have been tested for consumer acceptance. The total phenolic contents of the oven-dried and freezedried tissues from 83-89 and 85-87 mg/g are on a dry weight basis. Phenolic substances of the oven and freeze-dried materials ranged from 6.8-8.9 and 4.6-8.7 mg of chlorogenic acid equivalent (CAE)/g dry matter. Phenolic contents of SCT and flesh ranged from 4.6-8.7, 6.8-8.9 mg/CAE dry matter, respectively. The total phenolic contents of three varieties were significantly different with the highest was White followed by Light white and Dark green. Furthermore the bitter melon flesh contents considerably higher contents of polyphenolic as compared to SCT. The antioxidant activities of methanolic extracts from the bitter melon varieties dark green, white and light green ranged from 83-85, 85-88, 80-85, and 85-89% inhibition, respectively. Methanolic extracts of freeze-dried flesh from white and light green showed very high antimutagenic effects against benzo(a)pyrene with Salmonella TA98 (92-99% inhibition) TA100 (83-91% inhibition), but lower antimutagenicity activities against sodium azide that ranged from 46-54 and 17-32% inhibition, respectively. Fifty-nine participants from the University of Arkansas at Pine Bluff were recruited to take a taste testing survey evaluating the flavor, texture, and appearance of three bitter melon recipes. The experimental groups were beef stew A, and beef stew B. The flavor ratings for beef stew A and B were significantly different (p<0.1). White bitter melon (beef stew A) had the most favorable flavor rating. Texture and appearance ratings for beef stew A and B were not statistically different. The flavor is a significant factor to consider if the bitter melon is to be prepared and served to people. Future recipe studies can be done to determine the best way to cook bitter melon so that people will desire to consume it and receive all the nutritional benefits of this food item.

Keywords

Momordica charantia, Sensory Attributes. Color, Flavor, Texture, Polyphenolics, Antioxidant, Antimutagenicity

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1. Introduction

Many exotic vegetables are known for their unique nutritional and medicinal properties. Bitter Melon (Momordica charantia L.), an annual vegetable of Cucurbitaceae family, is found to be one of the critical plants of unique nutritional and medicinal qualities in the southern United States. Bitter melon is an edible fruit vegetable

commonly grown in China and India. It is a flowering vine and contains an array of novel and bioactive phytochemicals [1-4]. It has a characteristic bitter taste and has variation in bitterness depending on its color. Green bitter melon tends to be the most bitter, and bitter white melon is less bitter. When a bitter melon reaches full maturity and ripeness, it can turn orange. [1-4]

Nutrition-related health problems such as hypertension,

* Corresponding author E-mail address: islams@uapb.edu (S. Islam) diabetes, aging, obesity, arthritis, cardiovascular disease, are predominant between deprived country and urban inhabitants. particularly minorities in the delta region [5-6]. Human beings are naturally adapted to eating foods which have a balanced ratio of triglycerides and phospholipids. Based on the overall balance of triglyceride/phospholipid ratio, foods from plant sources have less adverse effects on human health than foods from animal sources [7]. When these balanced foods are consumed, peroxidation problems of polyunsaturated fats and lipids, associated with many of the human sicknesses, are remarkably minimized [8] In addition to the commonly used natural vegetable and herbs in the US, many exotic herbs and vegetables are known for their exceptional nutritious properties [9]. During the past decade or two, Americans have understood that they could switch their own health problems and reduce the risks of many chronic illnesses by changing certain dietary behaviors [10]. Certain vegetables comprise a profusion of polyphenolics, isoflavones, anthocyanins, amino acids, minerals, vitamins and other antioxidants that are related with protection from cancer, aging, cardiovascular diseases, diabetes, hypertension [8, 11-16]. The most popular and useful group of vegetables belong to the family Cucurbitaceae [17]. The green warty fruits of this plant are used as a vegetable, rich in vitamin A, vitamin C, and iron [18]. Fruits vary in size (1.0"-9.8" long and 1.0-5.9" wide), shape (oval, round, oblong, club, etc.), fruit color (dark green to creamy white), maturity (45-80 days), fruit per plant (6-85) and yield per plant (0.8-12) Ibs). The fruits, leaves, and roots of bitter melon are conventionally thought to have medicinal value in reducing blood sugar levels for diabetic patients [19-20]. The various therapeutic possessions of bitter melon are well known in eastern Asia [21]. Sofowora [22] reported several uses of M. charantia traditional medicines Africa. Pharmacologically, the hypoglycemic properties of the plant organs were established by Lotlikar et al. [23]. The polypeptides from the seeds and fruits of M. charantia were considered as antidiabetic agents [24-28]. Cucurbitacines isolated from several species of the family Cucurbitaceae showed antitumor effects [29-31]. Lin et al. [32] isolated two lectins from seeds of M. charantia, momordin and agglutinin, the momordin inhibiting protein synthesis by Ehrlich Ascites cells. Licastro et al. [33] announced that two proteins isolated from the seeds of *M. charantia* inhibited protein synthesis and subsequent DNA fusion in normal and leukemic human outlying blood lymphocytes. Spreafico et al. [34] reported a protein inhibitor from M. charantia showing immunomodulatory activity in mice. Although bitter melon is used by the ethnic people from Asia, Africa, and Europe, American consumers have not widely developed a taste and appreciation for it. Therefore the functional compounds contained in these foods and their therapeutic effects are needed to study and clinically proven thoroughly. Research is required to identify suitable genotypes of this vegetable for their production potential and nutritional and/or nutraceutical values. In these ongoing studies, we have determined that bitter melon can be successfully produced in Arkansas and the southern states. In sum, bitter melon is a nutritious food item that needs to prepare and eat for the benefit of the human body.

2. Materials and Methods

2.1. Sample Preparation

Three varieties of bitter melon (Light green, Dark green and White) were tested in replicated field trials on Calloway silt loam soil at the UAPB Agricultural Research Center in Pine Bluff. Experiments were conducted in randomized complete block designs with four replications. Plants were grown on upright trellises with 10 ft. between rows and 12 ft. within rows. Two four-week-old seedlings were transplanted in each hole. N, P, and K fertilizers were applied pre-plant using 200 lbs per acre of 13-13-13. Irrigation was provided by a drip irrigation system. Weeds were controlled by covering the beds with black plastic. Roundup (Glyphosate) was used in controlling weeds around the beds. Melons were harvested twice a week starting in mid-June and ending mid-September. Fruit samples were stored in sealed plastic bags in ice chests for phytochemical analysis. The bitter melons were washed with deionized water, drained at ambient temperature and cut in half lengthwise. The seeds were removed, and flesh was thinly sliced and collected separately from the seeds. The flesh was divided into two halves. One half was freeze-dried, and the other half was oven dried. Dried flesh was then ground to flour and passed through a 60-mesh U.S.A. standard testing sieve (W. S. Tyler, Incorporated, Mentor, OH).

2.2. Determination of Phenolics

Total phenolics of the flesh and SCT (Seed coat tissue), the of bitter melons, were determined by Folin-Ciocalteu method [35]. One hundred milligrams of each sample was weighed into a screw-cap test tube and vortexed with 10 mL of methanol. The dispersion was heated in a water bath of 65°C for 2h and allowed to cool at room temperature. To 1 mL of the clear solution in a screw-cap test tube, 1.0 deionized water was added. The tubes were vortexed and allowed to stand for 2h at room temperature. Absorption of the solution at 726 nm was measured using a spectrophotometer. The total phenolic content was expressed as chlorogenic acid equivalents in mg/g dry material.

2.3. Antioxidant Activity

Antioxidant activity was carried out by oxidizing linoleic acid methyl ester (MeLo) in the presence of phenolic extracts

as antioxidants [36]. Two mg of the extracts were dissolved in 10 mL of methanol. Five hundred microliters of the extracts solution were added into 0.2 g of MeLo (500 ppm extract in MeLo), and the methanol was evaporated under a stream of nitrogen at ambient temperature. Five hundred microliters of methanol were added into 0.2 g of MeLo for blank as a reference. Oxidation of MeLo in the present of the extract was carried out in at 40°C for 72h. Two mg of sample aliquots were taken at the starting point (zero time) and after 72h of oxidation (at 40°C) and dissolved in 10 mL of 2,2,4trimethylpentane (isooctane). The conjugated absorption of the aliquots was read using spectrophotometer (Shimadzu Model UV-1601, Kyoto, Japan) at a wavelength of 234 nm. The antioxidant activities were expressed as percentage inhibition of conjugated diene hydroperoxides formation of MeLo after 72h of oxidation comparing with blank from MeLo as a reference antioxidant as follows: % inhibition= [(AB(72h) - AB(0h))- (AE(72h) -AE(0h)/ (AB(72h) - AB(0h))] x 100; where A= absorbance, E= extract, and B= blank.

2.4. Antimutagenic Activity

The antimutagenic activity of the methanolic extract from freeze-dried bitter melon flesh and seed from varieties India white and China green was determined by the method of Ames et al. [37]. The histidine requiring stains of *Salmonella typhimurium* TA98 and TA 100 were used for this test. Benzo(a)pyrene and sodium azide was used as mutagens for mutagenic and antimutagenic analyses. The percentage inhibition of mutagenesis was calculated using the following equation: inhibition% = [1-(number of revertants in the presence of fraction/number of revertants in the absence of fraction)] X 100.

2.5. Sensory Evaluations

Fifty-nine participants were recruited to take a taste testing survey evaluating the flavor, texture, and appearance of three bitter melon recipes: beef stew A, beef stew B, and salad. The distributions for gender were 54% female, 37% male, and 9% unspecified. The average age of a respondent was 46 with ages ranging from 7 to 78 years of age. The race distribution was 46% Black, 19% White, 14% Asian, and 22% unspecified. Educational levels were distributed such that 17% were High School educated or under, 51% were trained above High School level, and 32% had general educational standards. Beef stew A contained Dark green (15 cups) bitter melon, and beef stew B carried white bitter melon (15 cups). The salad included bottle gourd and was a compliment the meal, not a primary experimental category. The ingredients for the three recipes are shown below (Table 1).

Table 1. Bitter melon recipe with Beef Stew A and B ingredients.

Beef Stew Recipe (A and B) *	
1 1/4 cup	All-purpose flour
10 pounds	Beef stew meat
1 1/4 cup	Cooking oil
5 cups	Onions, chopped
1 1/4 cup	Instant beef bouillon granules
$10 \operatorname{cups} (2\operatorname{qt} + 2 \operatorname{cups})$	Water
20 cloves	Garlic, minced
5 teaspoons	Dried basil, crushed
5 teaspoons	Dried thyme, crushed
22 ½ cups	Potatoes, peeled and cubed
20 cups	Carrots, sliced
10 cups	Celery, sliced
15 cups	Bitter melon * (A=green; B=white)

*The only difference between beef stew A and B is that the former has green bitter melon and the latter has white bitter melon.

The flavor, texture, and appearance for each recipe were rated on a scale of 1 to 5. A rating of 1 meant that the recipe was "liked very much," and a rating of 5 meant that the recipe was "disliked very much." A rating of 3 meant that the recipe was "neither like or disliked." Participants filled out paper surveys after tasting each sample. The ratings were given on the categories of flavor, texture, and appearance.

2.6. Statistics

All values are reported as means of three determinations. A Split-plot complete randomized design was conducted using JMP 5 software package (SAS 2002), and Tukey HSD procedure was performed for the significance of differences at the 5% level. Results and Discussion

3. Results and Discussion

3.1. Nutritive Value of Bitter Melon

The bitter melon fruit has a higher content of vitamin C, potassium, calcium, magnesium, and dietary fiber as compared to some other commercial vegetables [18] (Table 1). It also has niacin, thiamin, riboflavin, vitamin A, protein, organic acids, and other nutrients. Physical separation of bitter melons resulted in flesh and seed fractions. Bittermelon has traditionally has been used as a medicinal food in many countries [1-3] and demanded to have therapeutic consequences due to its content of biologically active compounds [3]. Several medicinal belongings of bitter melon have been reported by different investigators namely antioxidant, anti-diabetic, anti-ulcerogenic, anti-tumor, anti-lipolytic, analgesic, antiviral, analgesic, abortifacient, hypoglycemic, immunomodulatory, etc. [1-4, 11, 39-40]. Several authors have also established the comparative law toxicity of all parts of the bitter melon plant when consumed orally [1, 3]. Therefore, bitter melon and its products are employed by natural health practitioners for treating various human conditions.

Vitamins/Protein/Fat Component Minerals (mg) Edible portion (%) 84.0 Ca 22.0 Vitamin A (mg) 0.04 93.8 260.0 Thiamin (mg) 0.05 Water (%) K Energy (KJ) 20.0 Mg 16.0 Riboflavin (mg) 0.03 0.9 Carbohydrate (g) 0.20 Fe Niacin (mg) 0.40 Dietary fiber (g) 3.30 Na 3.0 Vitamin C (mg) 50.00 Zn Organic acids (g) 0.11 0.1 Protein (g) 0.90 0.60 Fat (g) 0.10 Ash (g)

Table 2. The nutritive configuration of per 100g comestible serving of Bitter Melon (Momordica charantia) Fruits [18].

3.2. Phenolic Contents

Total phenolic content of oven and freeze-dried bitter melon tissues from four varieties are given in Table 5. Overall, phenolic content in oven-dried samples was significantly higher than the freeze-dried samples. Phenolic substances of the oven and freeze-dried materials ranged from 6.8-8.9 and 4.6-8.7 mg of chlorogenic acid equivalent (CAE)/g dry matter, correspondingly. Phenolic contents of SCT and flesh ranged from 4.6-8.7, 6.8-8.9 mg/CAE dry matter,

respectively. The total phenolic contents of three varieties were significantly different with the highest was White followed by Light white and Dark green. Furthermore the bitter melon flesh contents considerably higher contents of polyphenolic as compared to SCT. In most plants, total phenolic compounds have been determined to be the main antioxidant compounds [11-16]. This study found that bitter melon fruits contain higher amounts of phenolic compounds as compared to some other plants [11, 13, 15].

Table 3. Total phenolic contents of bitter melon tissues (mg/g dry matter).

Bitter melon	Oven dried	Oven dried		Freeze dried	
Varieties	Flesh	SCT	Flesh	SCT	
Light Green	$7.9 \pm 0.06c$	7.1 ±0.12b	7.4 ±0.06d	$4.6 \pm 0.10d$	
White	$8.9 \pm 0.12b$	8.7 ±0.36a	$8.7 \pm 0.04c$	$7.9 \pm 0.09a$	
Dark Green	6.9 ± 0.06 d	6.8 ± 0.07 b	$6.8 \pm 0.14b$	$5.7 \pm 0.25c$	

^{*}Values are means \pm SD of three determination, Mean with different letters in the same column are significantly different (p<0.05).

3.3. Antioxidant Activities of Bitter Melon Extract

There was no significant difference in the antioxidant activities (% inhibition) of bitter melons among varieties and drying methods (oven and freeze-dried). The antioxidant activities of Indian green, Indian white, China green, and China white ranged from 79-88, 79-87, 80-86, and 79-87% inhibition, respectively. The antioxidant activities of the oven-dried samples and the freeze-dried samples were 79-88 and 79-86% inhibition, respectively. The antioxidant activities of the methanolic extracts of flesh and SCT were not significantly different, while they were considerably higher than that of

seeds. Shu-Jing and Lean-Teik [38] reported that bitter melon extracts possess potent antioxidant and free radical scavenging activities. These antioxidant activities could have contributed, at least partly, to the therapeutic benefits of the specific traditional claims of wild BM. The property useful in determining the non-enzymatic antioxidant values of fruits is the capacity of the extracts to inhibit the free radical. This reduction of the free radical diminishes cellular damage by allowing the neutralization of reactive oxygen species present in the extracts. Our results are however in agreement with other investigators [1-4], indicating the bitter melon is a high antioxidant comprising foods.

Table 4. Antioxidant activities (% inhibition) of different bitter melon varieties.

Varieties Oven-dried Flesh	Oven-dried	Oven-dried		Freeze Dried	
	SCT	Flesh	SCT		
Light Green	85 ±3.8a	89 ±0.4a	86 ±1.6a	87 ±0.8a	
White	$86 \pm 2.7a$	88 ±1.1ab	85 ±2.0a	86 ±0.5a	
Dark Green	$84 \pm 2.0a$	83 ±1.4c	85 ±0.8a	85 ±1.2a	

^{*}Values are means \pm SD of three determination, ** SCT= Seed coat tissue; Mean with different letters in the same column are significantly different (p<0.05).

3.4. Antimutagenicity of Bitter Melon Extract

All the bitter melon varieties studied showed higher antimutagenic effects against benzo(a)pyrene with *Salmonella* TA98 (92-100% inhibition) and *Salmonella* TA100 (79-86% inhibition) but lower antimutagenic effects

against sodium azide (data not shown). A similar finding was reported by several authors [39-40]. Several authors [39-40] discussed chemopreventive agents as blocking agents that prevent carcinogens from reaching or reacting with critical target sites, and as suppressing agents that inhibit the evolution of the neoplastic process in cells that otherwise

would become malignant. The bitter melon extract contains both agents [40]. The indication from epidemiologic, scientific and laboratory studies proposes that cancer may be caused by multifaceted interfaces between genetic and environmental influences. The risks of colorectal cancer are likely to result from exposure to carcinogens associated with daily food intake because adulteration of carcinogenic ingredients in food is not always preventable. Cancer deterrence may be carried out, though, by a decrease in the number of actions which convey about exposure to colon carcinogens. This theoretical model leads to the concept that the incidence of cancer could be abridged or prevented by the management of inhibitory compounds, either as separate drugs or as naturally happening elements of the day-to-day food. Although the exact mechanism of the chemopreventive effects of bitter melon is not yet known, these findings suggest that bitter melon is a possible chemopreventive agent against carcinogenesis.

 Table 5. Antimutagenic activities (% inhibition) of different bitter melon

 varieties

Bitter Melon extracts	TA98	TA100
Light Green	98.3	91.1
Dark Green	91.7	83.5
White	98.4	88.7

^{*}Values are mean of duplicate determination; Concentration of extract: 500 µg/plate; benzo(a)pyrene and sodium azide (10 µg/plate).

3.5. Sensory Evaluation and Quality Criteria

The quality criteria of bitter melon are a fresh appearance with uniform coloration and firm without excessive seed development. The bitter melons are harvested, selected for size and uniformity of fruit surface characteristics and commonly packed in carton or wood boxes containing 5, 10, or 20 kg of fruit. It is a chilling sensitive vegetable and maybe air-cooled to 10°C-12°C [37-38]. Bitter melons are intermediate in perishability. The common postharvest defects are seed development, softening and ripening with the internal /external color change. The recommended temperature and relative humidity for postharvest handling of bitter melon are 10-12.5°C and 85-90%, respectively, with an estimated shelf-life 7 to 14 days [41-42]. In this present study with a sample of 59, the average flavor ratings between Beef stew A and B were statistically different (p<0.1) (Figure 1). No significant differences were found in texture or appearance ratings between beef stew A and B (Figure 1). These results indicate that bitter white melon (beef stew B) has a more pleasant flavor than green bitter melon (beef stew B). The flavor difference can be attributed to bitterness difference between the white and green varieties, the latter being the most bitter. The bitter melon juice was assessed for

sensory characteristics by a panel of 10 partial-trained judges, using a 9 point hedonic scale scheme for diverse parameters like color and appearance, flavor, taste and overall suitability. The mean values of 10 partial-trained judges were measured for assessing the quality and specified that the color and appearance score of fresh beverage was 7.3 while flavor score was 7.7, taste score was 7.5, and overall acceptability score was 7.6 [43]. The appearance was influenced by the TSS levels. However, the addition of citric acid contributed towards better flavor, and the taste was much attributed to the appropriate sugar-acid blend in the product [43].

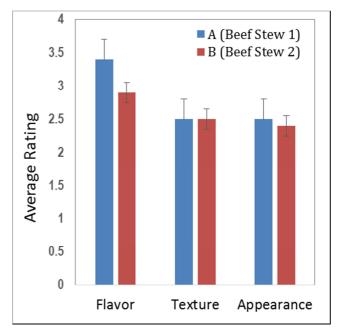


Figure 1. Average Flavor, Texture, and Appearance Ratings for Beef Stew A and Beef Stew B. *Flavor Ratings for Beef Stew A and B are significantly different. p < 0.1. N=59.

4. Conclusions

Evaluating bitter melon on the basis of flavor, texture, and appearance is essential, especially if agricultural researchers desire to market this highly nutritious food to the general public. This study has shown that flavor is a factor in alluring the consumer to eat food. In addition, bitterness tends to be a deterrent for some people, and the marketing of bitter white melon may be the best option for this population. More studies like this should be continued with different bitter melon recipes. Furthermore, Bitter melon is an excellent source of phenolic compounds, antioxidant, and antimutagen. This can find application in food products and dietary supplements. The phenolic extracts showed high inhibition effect to Prevent lipid oxidation. These natural plant phenolics can be an excellent antioxidant which may be applied in many food systems to maintain the food quality.

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