EXTRACTION OF COLOR FROM INDIGENOUS PLANTS AND TO STUDY THE EFFECT OF THESE EXTRACT ON RTS (READY TO SERVE) BEVERAGE

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Abstract: Food color is one of the classes of food additives added to food products for improvement of sensory quality. It is not only the sensory quality improved rather it supplies the nutrients in many cases of food pigments. The *Marua* has Rosmarinic acid as an antioxidant and the beverage was found to be acceptable for 2-4 days after its addition. The color intensity after incorporating into the beverage was good but shows a declination after a short period of time. The color of papaya beverage after adding *Marua* extract was much better than curry leaves extract. The appeal of banana beverage increased a lot after addition of *Marua* extract & it seems much more appetizing than before. While the incorporation of the curry leaves extract changes color to a great extent that looks attractive but the flavor imparts by the extract is not so acceptable. The addition imparts aromatic smell and slightly bitterness to the RTS which was not liked by the judges. In an average shelf life in an incubator at about 37^0C it lasts for about a 2-4 days. The essential oils from *Marua* also contain d-limonene that has anti carcinogenic properties. It also contains vitamin B_6 and magnesium the vitamin B_6 prevents the formation of harmful compounds in the body such as homocysteine and magnesium makes the cardiac muscles and blood vessels healthy so that blood flows without any interruption.

Keywords: Extraction, Effect, Indigenous plants

INTRODUCTION

Food color is one of the classes of food additives added to food products for improvement of sensory quality. It is not only the sensory quality improved rather it supplies the nutrients in many cases of food pigments. Many fruits and vegetables contain naturally occurring food colors like Chlorophyll, Carotene, Lycopene, Anthocyanins etc. all these pigments occur in plastids for example chlorophyll occurs in the Chloroplast. Here, we study the colors from *Ocimum basilicum* and *Murraya koenigii*.

The word basil comes from the Greek (basileus), meaning "king", as it is believed to have grown above the spot where St. Constantine and Helen discovered the Holy Cross. *Ocimum basilicum* (common basil) is the members of the family Lamiaceae. In India it is commonly known by the name Marua in northern region. The Oxford English Dictionary quotes speculations that basil may have been used in "some royal unguent, bath, or medicine". Basil is most commonly recommended to be used fresh; in cooked recipes it is generally added at the last moment, as cooking quickly destroys the flavor. The fresh herb can be kept for a short time in plastic bags in the refrigerator, or for a longer period in the freezer, after being blanched quickly in boiling water.

Ideal colour properties

- It should be non toxic & non carcinogenic.
- It has solubility in both fats as well as in water for its wide usage.
- It should be stable to light, pH change, baking & boiling Temp.
- It must be approved and confirm to government specification.

Ocimum basilicum (Marua)

Murraya koenigii (Curry patta leaves)

Objectives of color extraction

- To extract colors from various indigenous plants
- To standardized the level of colors to be incorporated in RTS beverage.

- To study the difference in concentration of color by different extraction technique
- Improved appearance, flavor and enjoyment of food.
- Reduces cost and improved quality.
- Improved stability and reduced wastage in storage.
- Should be chemically stable in the foods in which they.

MATERIAL AND METHOD

Chlorophyll and carotenoid extraction by arnon method

The Arnon, a botanist in 1949 developed a method of chlorophyll & carotenoids extraction from plants. This method is even used to calculate the extraction of chlorophyll A and chlorophyll B.

Procedure

100-500 mg of plant material was homogenized with 10-15 ml of 80% acetone in a pastel & mortar until tissue become colorless. A pinch of $CaCo_3$ was also added to avoid the destruction of chlorophyll and other pigments. Centrifuge the liquid at 2000 r.p.m. for about 10-15 minutes. Filter it. Now make the volume to 10 ml. with acetone. Absorbance was recorded at 480, 510 & 663 nm Using spectrophotometer.

$$\text{Total carotenoids} = \frac{7.6\ A_{480} - 14.9\ A_{510} \times V}{\alpha \times 1000 \times w}$$

Where,
A = absorbance.
α = light path in cm.
v = volume in m.
w = fresh weight in grams.
Quantity = mg/gm on fresh as well as dry wt.

$$\text{Chlorophyll a (mg/gm fresh wt)} = \frac{12.3\ A_{663} - 0.06\ A_{645} \times V}{\alpha \times 1000 \times w}$$

$$\text{Chlorophyll b (mg/gm fresh wt)} = \frac{19.3\ A_{645} - 3.6\ A_{663} \times V}{\alpha \times 1000 \times w}$$

Paper chromatography by tswett in 1906

Theory

In this technique the sample to be analyzed is applied in the form of a spot near one edge of a Whattman filter paper. The Cellulose of paper acts as the stationary phase. An appropriate system, which functions as a mobile phase, is then allowed to flow over the sample spot. On coming in contact with the mobile phase, various components of the sample get partitioned between the stationary and mobile phase. Those constituent having higher affinity for the stationary phase move slowly as compared to the component have higher affinity for mobile phase.

Detection

The detection of colored compounds takes place under UV light.

Extraction with various alcoholic solvents

Here various alcoholic solvents are used such as Methanol, Ethanol, Hexane, and Propan-2-ol, Chloroform etc. 10 gm leaves dipped in the each solvent and then allow for shaking for about 12 to 24 hrs. Discard the colorless leaves, filter the solution and examine the absorbance at visible region with the help of spectrophotometer. Now allow these solvents to evaporate under the shade. Note down the weight of extract. The thick concentrate is the plant extract which is used further for the estimation of shelf stability, aroma and flavor development in the beverages.

Extraction & partial purification of chlorophyll from plant material using dioxane

A convenient laboratory method for partial purification of chlorophylls using dioxane is studied by Keiji Iriyama, Nagao Ogura they define that Chlorophyll in crude extracts with 90%-methanol or 80%-acetone was mixed with dioxane and then precipitated by drop-wise addition of water. Chlorophyll is precipitated in spherocrystalline form, leaving the bulk of carotenoids in solution. This is an efficient method for preparing chlorophyll from various plant materials, including higher plants and algae as well as photosynthetic bacteria (bacterio-chlorophyll).

Sensory evaluation

Beverage sample were offered to 5 panelists and sensory evaluation was conducted based on 9 point hedonic scale. The result of evaluation is as follows.

Data in hedonic scale	Score
Like extremely	9
Like very much	8
Like moderately	7
Like slightly	6
Neither like nor dislike	5
Dislike slightly	4
Dislike moderately	3
Dislike very much	2
Dislike extremely	1

RESULT AND DISCUSSION

The *basil* contains Rosmarinic acid which works as an antioxidant. Data on % recoveries from leaves of *basil* and *Murraya* during color extraction by different solvents has given below.

Marua sample A (Ethanol)

Extract obtained I	= 0.5230 gm.
Extract obtained II	= 0.4010 gm.
Extract obtained III	= 0.5249 gm.
Mean	= 0.4829 gm

Sample B

Extract obtained I	= 0.7200 gm.
Extract obtained II	= 0.7516 gm.
Extract obtained III	= 0.6882 gm.
Mean	= 0.7199 gm

Sample A (Methanol)

Extract obtained I	= 0.6080 gm.
Extract obtained II	= 0.3022 gm.
Extract obtained III	= 0.6456 gm.
Mean	= 0.5186 gm

Sample B

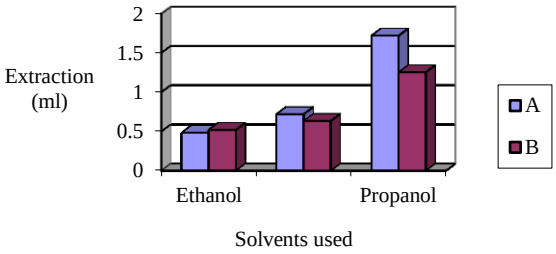
Extract obtained I	= 0.4050 gm.
Extract obtained II	= 0.4949 gm.
Extract obtained III	= 1.0024 gm.
Mean	= 0.6341 gm

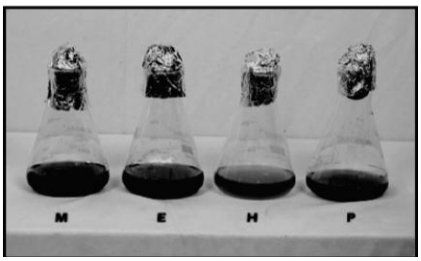
Sample A (Propan-2-ol)

Extract obtained I	= 1.8690 gm.
Extract obtained II	= 1.8640 gm.
Extract obtained III	= 1.6640 gm.
Mean	= 1.7990 gm

Sample B

Extract obtained I	= 1.3311 gm.
Extract obtained II	= 1.3244 gm.
Extract obtained III	= 1.1077 gm.
Mean	= 1.2544 gm





From these solvent extraction techniques the maximum extract is obtained by the use of propan-2-ol.

Curry Patta Leaves (*Murraya koenigii*) ethanol

Sample A

Extract obtained I	= 0.3002 gm.
Extract obtained II	= 0.3332 gm.
Extract obtained III	= 0.3066 gm.
Mean	= 0.3133 gm

Sample B

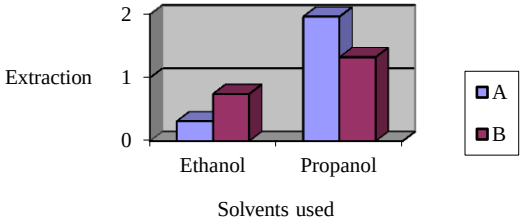
Extract obtained I	= 0.7131 gm.
Extract obtained II	= 0.7327 gm.
Extract obtained III	= 0.7872 gm.
Mean	= 0.7443 gm.

Sample A (Propan-2-ol)

Extract obtained I	= 1.9441 gm.
Extract obtained II	= 2.0083 gm.
Extract obtained III	= 1.9444 gm.
Mean	= 1.9656 gm.

Sample B

Extract obtained I	= 1.3128 gm.
Extract obtained II	= 1.3272 gm.
Extract obtained III	= 1.3398 gm.
Mean	= 1.3266 gm.



Absorbance study of Marua leaves extract by using spectrophotometer

Solvent used	Wave length	% absorbance
Ethanol		2.098
	480 nm	1.619
		2.293

solvent used	wavelength	% absorbance
Methanol		2.041
	480 nm	1.683
		2.325

Propan-2-ol		2.080
	480 nm	1.749
		2.317

Methanol		1.354
	645 nm	1.665
		1.473

Ethanol		1.370
	645 nm	1.764
		1.444

Propan-2-ol		2.567
	645 nm	2.035
		2.400

Absorbance study of curry leaves extract by using spectrophotometer

Ethanol		2.567
	480 nm	2.035
		2.400

Hexane		2.224
	480 nm	1.810
		1.983

Propan-2-ol		2.449
	480 nm	2.723
		2.767

All these extracts are measured and weighed and then incorporated into the papaya and banana juice. The fresh papaya juice when stored at 4^{o}c had excellent visual appearance after 7 to 10 days of storage, however after 7 days faint of greenish color starts with decreased acceptability of juice. These products after 14 days storage were found to have unacceptable flavor quality. Maximum permitted level of colors is expressed as mg/kg and refer to mg of color per kg (liter) of food that is ready to eat. For many colors the term" Quantum Satis" is used for the permitted level. However, usage should be in accordance with good manufacturing practices. The results of sensory evaluation were carried out at the departmental level. The color of the product accepted at the excellent level while the flavor of RTS was liked by the judges at moderate level. The *Ocimum basilicum* extract were liked most in flavor as well as in color and *Murraya koenigii* extract was discard due to bitter flavor formation.

DISCUSSION AND CONCLUSION

The extraction of color was done by using different alcoholic solvents such Ethanol, Methanol, Propan-2-ol and Hexane etc. but maximum recovery was found from Propan-2-ol which was determined by the absorbance study using spectrophotometer.

However the *Marua* has Rosmarinic acid as an antioxidant and the beverage was found to be acceptable for 2-4 days. The color of papaya beverage after adding *Marua* extract was much better than curry leaves extract. The appeal of banana beverage increased a lot after addition of *Marua* extract & it seems much more appetizing than before. Although, the shelf life of beverage was not studied but the decrease in acceptability seen initially after some time due to chemical changes that takes place in the beverage due to improper preservation techniques. While the incorporation of the curry leaves extract changes

color to a great extent that looks attractive but the flavor imparts by the extract is not so acceptable. The addition imparts aromatic smell and slightly bitterness to the RTS which was not liked makes it shelf stable. In an average shelf life in an incubator at about 37^0C it lasts for about a 2-4 days. The essential oils from *Marua* also contain d-limonene that has anti carcinogenic properties. It also contains vitamin B_6 and magnesium the vitamin B_6 prevents the formation of harmful compounds in the body such as homocysteine and magnesium makes the cardiac muscles and blood vessels healthy so that blood flows without any interruption. Thus we say that *Marua* addition into the RTS makes them nutritionally fit and also add taste to our palate.

Future recommendations

Changing lifestyles have resulted in a greater demand for food that will stay fresh and safe for a long period. As the population is growing, more food is needed and hence the use of additives requirements to provide a total safe food supply will be more in demand. According to my own opinion if the *Marua* flavored juice is prepared in a commercial scale by examining it's all properties prior its addition into the juices, meats and the bakery products, make them highly stable and deprive the Indian population from various deficiency diseases. *Ocimum basilicum* is a cosmopolitan species found everywhere in the Indian climate growing as a wild variety thus it is easily accessible and beneficial for the low class and middle class peoples that suffer from various malnutrition problems. The China is a thickly populated country which consumes the *Marua* as soup. In Indonesia and Thailand *Ocimum basilicum* consumes in salad as well as mix in the vinegar for maintain its preserving action. Marua works as a medicine in various diseases. Chewing of *Marua* leaves makes our gum strong and relieves from toothache problems such as foul smell. Thus its recommendation into the toothpaste should be mandatory. At last from the properties studied during this research reveals us the importance of *Ocimum basilicum* that add taste to our palate and makes our lives healthy. A. Micel and A. Moncada have studied that the expansion of the medicinal and aromatic plant sector requires new studies to rationalize their cultivation. Due to its toxicity, the use of methyl bromide as a sterilizing agent will be banned in the near future. Basil is very susceptible to soil disease and therefore, alternate options for its cultivation, such as hydroponics methods, need to be considered. One of these hydroponics methods is the floating system which is the easiest and least expensive way to produce leafy Vegetables when soil cultivation is no longer feasible. The experiment was carried out in a greenhouse from February to April 2002.

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