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Effect of different solvents on recovery of total monomeric anthocyanin from mangosteen peel (*Garcinia mangostana* L.)

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Abstract

The yield of Total monomeric anthocyanins (TMA) were analysed for optimisation of extraction condition by varying solvents in fresh and dry peel of mangosteen. Solvents include Hot water, Ethanol and Methanol were used as test solvents. They were later acidified with 0.1% HCl, 1% citric acid and 1% acetic acid to identify the increase in yield of anthocyanins. Results evidently showed that extraction variable was affecting the yield of Total monomeric anthocyanin. The treatment showed significant results when processed statistically. The maximum yield of anthocyanin obtained when extracted with test solvent methanol followed by ethanol. Among them solvents acidified with 0.1% HCl showed the maximum yield followed by acidified citric acid solvent. Considering the negative impact of methanol and HCl on human, anthocyanin extracted with ethanol acidified with citric acid were found to be the optimal solvent for food uses.

Keywords: Mangosteen, anthocyanins, solvents, acidified solvents

Introduction

Mangosteen (*Garcinia mangostana* L.) called as the “Queen of fruits” is one of tropical fruits which has sweet tangy taste. It is widely grown in South East Asia and in the tropical areas^[1]. They have edible white flesh inside which are fibrous and inedible rind which are purple in colour. The peel of mangosteen is rich in anthocyanin and their use is recently been identified as drug entrants^[2]. Various compounds on the rind includes flavonoids epicatechin, anthocyanins and xanthone derivatives are responsible for the beneficial action. They found to have ease for laxative menstruation, thrush medication, fever, drug for treating dysentery^[8]. The purple colour of the fruits is due to the presence of anthocyanin which tend to act as greater antioxidant. Anthocyanin prevents the oxidative stress and block the free radicals produced^[11].

Numerous studies revealed that anthocyanin is a potential natural colourant in the food processing industry and their contribution is known for anticancer, antimicrobial and anti-inflammatory purposes^[11]. Various solvents are used for extraction of anthocyanins such as aqueous, ethanol, methanol, acetone, chloroform etc^[17]. Later the acidified solvent extraction were introduced to improve the yield. Study says that acidification were done with citric acid, acetic acid, Hydrochloric acid, formic acid etc. Nowadays many modern extraction techniques were assisted for extraction which includes ultrasound, High pressure, Microwave etc^{[4][19]}. The purpose of acidification is to denature the membranes of the cell wall where the anthocyanin are maximally located. Anthocyanin is very sensitive to pH and degrade at higher temperature^[14]. Hence the main objective of this paper is to study the optimal extraction condition for the recovery of crude anthocyanin content from the mangosteen peel.

Materials

Mangosteen fruits were purchased from Local market, Chennai. Fruits were carefully selected without any blemishes and yellow latex. They were washed completely to remove dirt's and fruits were stored in the freezer. The thin purple skin on the rind alone was peeled off, shade dried for 10-14 hrs until moisture content reaches less than 10% and taken for analysis.

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Methods

Effect of solvent on extraction of TMA

Extraction was done as per the modified procedure of Dam, 2017^[6]. Sample with solvent ratio 1:10 were used with extraction time for 1hr at room temperature. Firstly, Aqueous, Ethanol (50% v/v) and Methanol (50% v/v) were used for the extraction of anthocyanin from fresh and dry peel.

Effect of acidified solvents on extraction of TMA

Extraction done using acidifying solvents was done as per the modified procedure of Dam, 2017^[6]. Samples were acidified with 0.1% HCl, 1% citric acid and 1% acetic acid to compare the yield between the fresh and the dry peel of mangosteen.

Estimation of total monomeric anthocyanin content

Anthocyanin was estimated by pH differential method. Monomeric anthocyanin undergoes reversible structural change from oxonium form at pH 1 to colourless hemiketal form at pH 4.5. Hence the difference in the absorbance is equal to the concentration of monomeric anthocyanin in the sample and expressed as mg cyanidin-3-glucoside/100g of sample. Reagents 0.025M KCl (pH 1) and 0.4M sodium acetate (pH 4.5) are the two buffer solutions which are used for the determination of TMA. Samples were diluted in 1:10 ratio and absorbance were measured at 510nm and 700nm using UV spectrophotometer^[13].

$$\text{Anthocyanin pigment (mg/L)} = \frac{A \times MW \times DF \times 1000}{\epsilon \times l}$$

Where,

A = (A_{520nm} - A_{700nm}) pH_{1.0} - (A_{520nm} - A_{700nm}) pH_{4.5}

MW = 449.2 g/mol for cyanidin-3-glucoside (molecular weight)

DF = Dilution factor

ε = 26,900 (molar absorptivity of cyanidin-3-glucoside)

l = cell path length in (1cm)

1000 = factor for conversion from g to mg.

Statistical analysis

All experiments were conducted in six trials and an analysis of variance was performed using VETSTAT software. The results were expressed as mean ± SE and least significant difference at P < 0.05 was calculated using Duncan's multiple range test to determine the effect of optimised solvent on time, temperature and pH on anthocyanin content with significant differences in results.

Results

Table 1: Effect of solvents on extraction of TMA

Solvents	Fresh peel (FP)	Dry peel (DP)	t - test value
Hot water	57.49±0.53 ^a	44.84±0.09 ^a	23.26**
Ethanol	73.25±0.10 ^b	57.53±0.67 ^b	23.05**
Methanol	75.92±0.08 ^c	61.20±0.74 ^c	19.64**
F value	1.8388	3.3621	

Table 2: Effect of acidifying solvents on extraction of TMA

Solvents	Fresh peel (FP)	Dry peel (DP)	t - test value
T1	73.14±0.23 ^c	65.13±0.30 ^c	20.66**
T2	92.85±0.39 ^f	82.35±0.10 ^f	26.03**
T3	104.80±0.84 ⁱ	88.58±0.03 ⁱ	19.19**
T4	69.16±0.85 ^b	58.20±0.14 ^b	12.65**
T5	89.37±0.49 ^e	80.74±0.09 ^e	17.28**
T6	99.14±0.17 ^h	86.58±0.03 ^h	69.76**
T7	64.40±0.36 ^a	55.13±0.26 ^a	20.68**
T8	85.47±0.42 ^d	78.99±0.11 ^d	14.86**
T9	96.77±0.09 ^g	82.60±0.10 ^g	101.80**
F value	1.4394	0.4606	

- T1 – Acidifying water HCl
 T2 – Acidifying Ethanol HCl
 T3 – Acidifying Methanol HCl
 T4 – Acidifying water Citric acid
 T5 – Acidifying Ethanol Citric acid
 T6 – Acidifying Methanol Citric acid
 T7 – Acidifying water Acetic acid
 T8 – Acidifying Ethanol Acetic acid
 T9 – Acidifying Methanol Acetic acid

Table 1 shows the respective mean ± SE values of the effect of Solvents (Hot water, Ethanol and Methanol) and Table 2 shows the respective mean ± SE values of the effect of Acidifying Solvents: Hot water, Ethanol, Methanol acidified with 0.1% HCl, 1% citric acid and 1% acetic acid on Total Anthocyanin content (mg CGE/100g) in Fresh peel (FP) and Dried peel (DP - shade drying).

Average of Six Trials, # Total Monomeric Anthocyanin content.

Small case superscripts represent significant differences between treatments.

** represents Statistically Highly significant (P≤0.01)

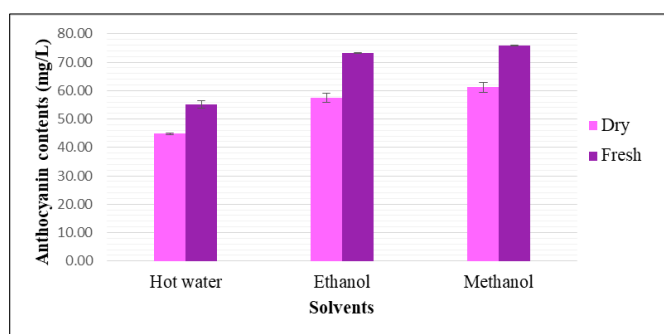


Fig 1: Effect of Extraction solvents on yield of TMA in Fresh and Dry peel of Mangosteen

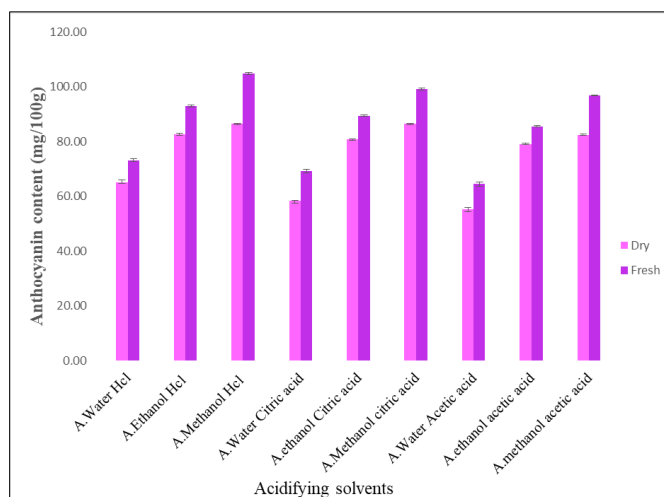


Fig 2: Effect of Acidified solvents (A.) on yield of TMA in Fresh and Dry peel of Mangosteen

Discussion

Effects of extraction solvents on yield of TMA in fresh and dry peel of mangosteen

Anthocyanin in the crude extracts extracted using three different test solvents had the yield ranged from 44.84±0.23 to 104.80±0.43 mg cyanidin /100g. The results were comparable with the author Chaovanalikit (2012)^[2] who reported that anthocyanin content was 179.49mg cyanidin/100g when cryogenically milled outer pericarp of

mangosteen was extracted with acetone as test solvent. From Fig.1 and 2, It is observed that mangosteen peel extracted with test solvent methanol and Ethanol acidified with 0.1% HCl has given more yield of anthocyanins followed by methanol and Ethanol acidified with 1% citric acid as test solvents has given higher yield with the mean and standard values of 89.37 ± 0.24 mg/100g cyanidin equivalent. However for food use, Ethanol is identified as a better and safe solvent^[5]. Similar results were reported by author Dam (2017) who stated Ethanol acidified with 1.5% HCl as optimised solvent for anthocyanin extraction^[6]. Between Fresh and Dry peel of mangosteen, extract from fresh peel has given more yield with the mean and standard values of 92.85 ± 0.44 mg cyanidin/100g than the shade dried peel with the mean and standard values of 82.60 ± 0.25 mg cyanidin /100g. It is perceived that acidification of test solvent increased the yield of crude anthocyanin.

Conclusion

Results showed that mangosteen is credible source of anthocyanin which possibly exhibit good antioxidant activity when employed in food matrix. The optimised extraction condition was found to be good with ethanol acidified with 1% citric acid. The optimised solvent may be recommended for food and drug application whereas extraction with methanol and acidified solvent (HCl) can be used for production of synthetic dyes for fabrics, paints and printing.

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