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Ultrasound Assisted Extraction and Microwave Assisted Extraction of Carotenoids from Melon Shells

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Abstract—Cantaloupes (muskmelon and watermelon) contain biologically active molecules such as carotenoids which are natural pigments used as food colorants and afford health benefits. ß-carotene is the major source of carotenoids present in muskmelon and watermelon shell. Carotenoids were extracted using Microwave assisted extraction (MAE) and Ultrasound assisted extraction (UAE) utilising organic lipophilic solvents such as acetone, methanol, and hexane. Extraction conditions feed-solvent ratio, microwave power, ultrasound frequency, temperature and particle size were varied and optimized. It was found that the yield of carotenoids was higher using UAE than MAE, and muskmelon had the highest yield of carotenoids when was ethanol used as a solvent for 0.5 mm particle size.

Keywords**—**Carotenoids, extraction, muskmelon shell, watermelon shell.

I. Introduction

OVER the past few years, there has been a surge interest in fat-soluble compounds such as Vitamin-A & E, coenzyme Q_{10} and carotenoids due to their beneficial effects on human health. Vitamin-A is essential for normal cell growth, cell differentiation, immunological functions and vision [1], [2]. WHO reported that vitamin-A deficiency affects about 190 million preschool-aged children and 19 million pregnant women mostly in Africa and South-East Asia. Carotenoids are extensively studied for the prevention of cancers and cardiovascular diseases. They are also important for anti-oxidant activity, intercellular communication and immune system activity [3]. Besides their biological properties carotenoids are utilized as natural anti-oxidant for the formulation of functional foods. Lycopene and ß-carotene are authorized natural pigments that can be used in the drying of various kinds of the food products.

Carotenoids are natural pigments which are metabolized by plants, algae and bacteria, which are responsible for the yellow, orange and red colors in various fruits and vegetables. The interest was focused on waste by-products from food industry such as seeds, peels, stems and leaves which are the cheapest source of natural anti-oxidants (carotenoids) and they can later be used in pharmaceutical, cosmetics and food industries [4]. Carotenoids can be classified into two groups on the basis of functional groups; xanthophylls containing oxygen as a functional group and carotenes which contains only parent hydrocarbon chain without any functional group.

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Soxhlet extraction (SE) is the most widely used unit operation for the recovery of bioactive compounds from a broad range of plant origin matrices. Though the yield of carotenoids obtained by SE is generally high, it is not preferred due to possible degradation of thermo-sensitive compounds such as carotenoids at high temperature and long extraction time. UAE and MAE can also be used to separate hydrophilic flavonoids and hydrophobic carotenoids.

UAE is a simpler and more efficient alternative to conventional solvent extraction, and occasionally, to modern extraction techniques. The efficiency of ultrasound-assisted as an alternative to conventional solvent extraction is mainly attributed to acoustic cavitation. Acoustic cavitation is caused by the interaction between the ultrasonic waves, the liquid, and the dissolved gas and can be generated in liquids in the frequency range of 20 kHz to >1 MHz, above critical power levels. MAE is a relatively new extraction technique, which utilizes microwave energy to heat the solvent and the sample to increase the mass transfer rate of the solutes from the sample matrix into the solvent.

MAE has a number of advantages like shorter extraction time, less solvent, higher extraction rate, over traditional method of extraction of compounds from various sources, especially natural products. Operational parameters temperature, sample size, moisture content, particle size, solvent type, power and frequency, sonication/microwave time and liquid to solid ratio are crucial to achieve high extraction efficiency [5], [6].

In the present study, muskmelon shell and watermelon shell were used as a potential source of carotenoids. Extraction of carotenoids from muskmelon shell and watermelon shell were carried out by Ultrasound Assisted Extraction (UAE) and MAE. Hexane and ethanol were used as a solvent. The main goal was to determine the influence of solvent, F-S ratio, temperature and extraction time on extraction of carotenoids to optimize the optimum extraction parameters.

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II. Materials and Methods

A. Materials

The muskmelon and watermelon samples were collected from the local juice shops. These shells were washed with distilled water to remove sand and dust particles from outer surface. They were dried in sunlight for 3 days and then dried in hot air oven for 30 min at 50 °C. Then, they were grinded into fine particles. The ground particles are separated based on size by using sieves. The mesh number was 16(0.6 mm), 25(0.5 mm). After sieving, the particles are labeled and stored in air tight containers.

The drying characteristics were studied in hot air oven at a temperature of 50 °C, to determine the moisture content of muskmelon and watermelon shell. Initially 10 grams of fresh raw material was taken in the petri plate. The initial and the final weight of petri plate was noted. The sample weight was noted for every 30 minutes until attaining a constant weight.

B. Extraction of Carotenoids Using MAE

1 gram of 0.5 mm musk melon shell samples were placed in microwave oven with known volume of extracting solvent for various microwave powers of 200 W, 300 W, 400 W and 500 W. The extraction yield was calculated. Equilibrium time and feed-solvent ratio were determined by varying the microwave power, temperature and were optimized for the highest yield of β-carotene. The same procedure was repeated for water melon samples.

C. Extraction of Carotenoids Using UAE

1 gram of sample was sonicated for 1 to 10 min using 20 mL of extracting solvent in an ultrasonic bath at frequency of 40 kHz, temperature of 40 ºC and 50 ºC. The effect of concentration on carotenoids with ultrasonic duration was determined. Equilibrium time and feed-solvent ratio were determined by varying the ultrasonic strength, temperature and optimized for the highest yield of β-carotene. The same procedure was repeated for water melon samples.

D. Analysis of Carotenoids

After extraction, the extracted solvent was analyzed using UV-Vis Spectrophotometer and absorbance values were noted for each feed-sample ratio. The ratio of absorbance (455nm/483nm) was maintained between 1.14 and 1.18. The concentration of β-carotene present in the extracted solvent was determined using absorbance value by following formula.

$$\beta\text{-carotene } q = (A \times S \times V) / (E \times W)$$

where, A = Absorbance in 455 nm, S = volume of solvent in mL, V = volume of extractant taken in cuvette, E = Coefficient of absorbance, W = weight of sample taken for extraction.

III. Results and Discussion

The extraction of carotenoids from muskmelon shell and watermelon shell using MAE and UAE were studied. Extraction conditions varied were feed-solvent ratio (1:10 to 1:40), microwave power (100 W, 150 W, 200 W and 300 W), ultrasound frequency (40 kHz), temperature (30, 40, 50, and 60 °C), solvent (hexane) and particle size (0.5 mm).

In MAE, the effect of concentration on carotenoids with duration of microwaved extraction was determined.

A. Equilibrium Time

Equilibrium extraction time was studied for musk melon, and water melon shells were shown in the figures below,

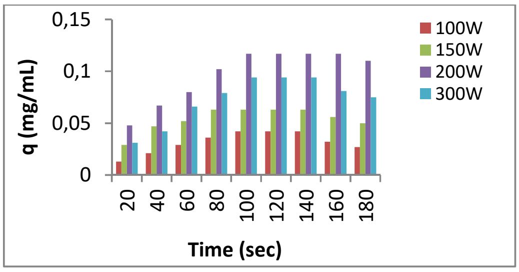

Fig. 1 Equilibrium time for muskmelon varying microwave power

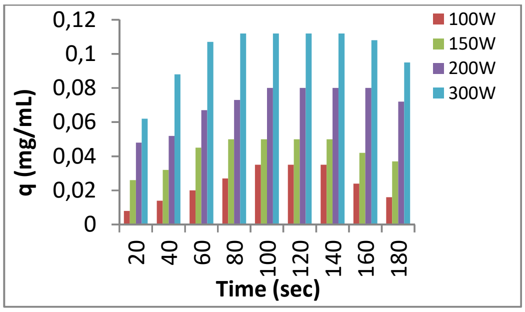

Fig. 2 Equilibrium time for watermelon varying microwave power

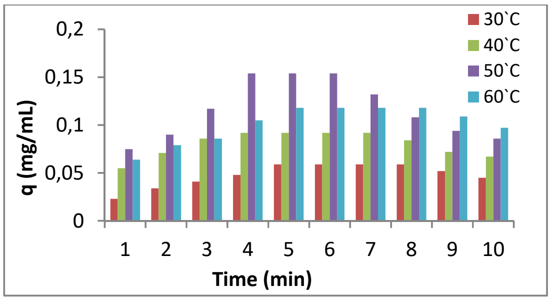

Fig. 3 Equilibrium time for muskmelon by UAE

It was studied from Figs. 1 and 2 that the optimum conditions obtained for extraction of carotenoids from muskmelon shell were 200 W, 0.5 mm and 120 s and for watermelon the optimum conditions are 300 W, 0.5 mm, and 120 s. The microwave irradiation for more duration with lesser microwave strength yielded better recoveries than lesser time with higher strength. It may be due to the reason that in open vessel microwave extraction, when higher microwave strength was applies, solvent gets evaporated rapidly. But, when lesser microwave strength is applied for more time, there is sufficient time for solvent to dissolve target analyte within it, resulting in enhanced recoveries of target analyte. So, more irradiation time with lesser strength was preferred.

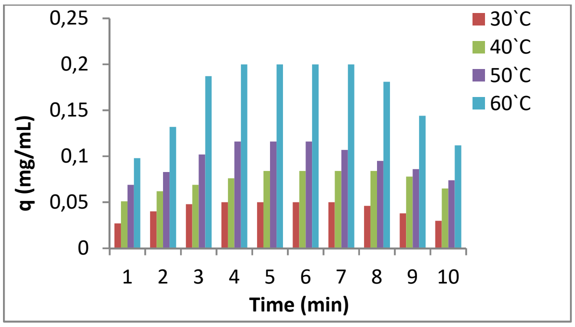

Fig. 4 Equilibrium time for watermelon by UAE

B. Feed-Solvent Ratio

The feed-solvent ratio was determined for high concentration of carotenoids extraction by varying with microwave power.

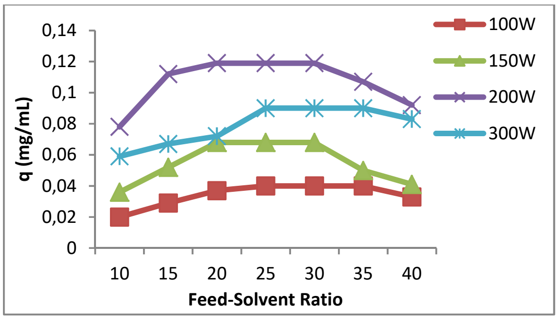

Fig. 5 F-S ratio for muskmelon varying microwave power

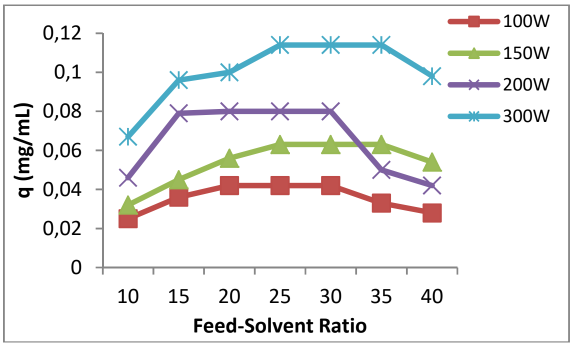

Fig. 6 F-S ratio for watermelon varying microwave power

From the above graphs, it was studied that the optimum feed-solvent ratio as 1:25 and particle size 0.5 mm was adopted for extraction of carotenoids in muskmelon and watermelon shell. It was observed that the small particle size gives the higher concentration of carotenoids; because the smaller particle has the higher surface area. Higher surface area provides more contact between feed and solvent so extraction yield increases. The concentrations of carotenoids are high in muskmelon shell than in watermelon shell [7].

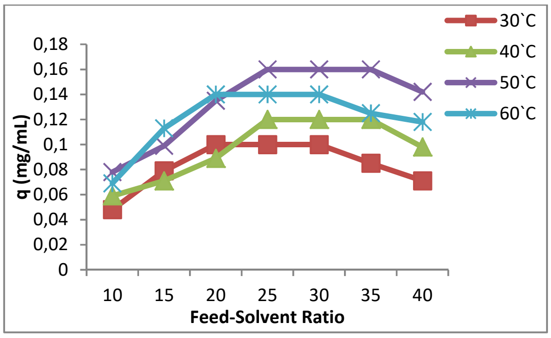

Fig. 7 F-S ratio for muskmelon using UAE

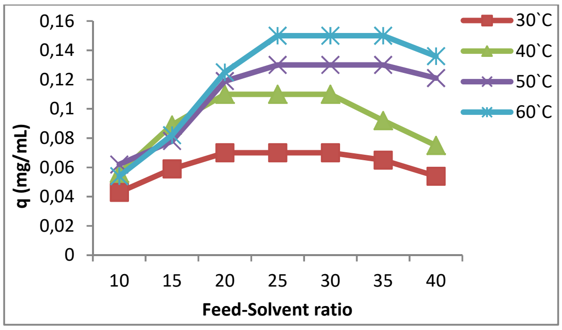

Fig. 8 F-S ratio for watermelon using UAE

The extraction time was very low in UAE compared to other extraction methods. This is due to the cavitation effects, which caused the intensification of faster mass transfer and thus increased interaction between the solvent and plant material. The collapse of cavitation bubbles near plant material surface produces micro-jets, causing tissue disruption and a good penetration of the solvent into the plant material. The above figure shows that the rate of carotenoid extraction was high, during the first 5 min and then decreased considerably thereafter. A reason for this was due to the large difference between the initial carotenoid concentration of the extraction solvent and its solubility. Another reason for this, initially high rate could be that carotenoids located in the outer region of the plant powder were more readily accessible than those in the inner part in which the plant tissues were intact. The extraction from the outer part was attributed to external mass transfer, which in this case was convective since fluid motion was provided because of ultrasonic cavitation. Later, carotenoids from the inner part of the plant powder must diffuse through the pores of the plant material, resulting in a much slower extraction rate.

C. Extraction Temperature

The extraction temperature was determined for high concentration of carotenoids extraction by varying the microwave power.

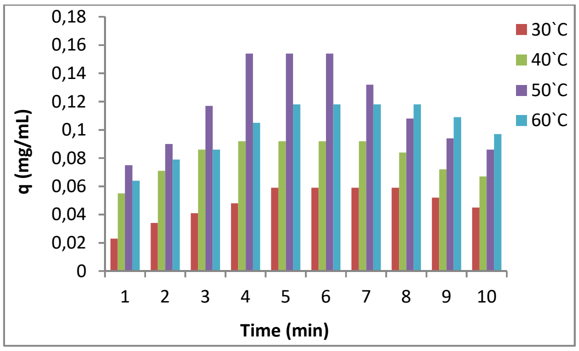

Fig. 9 Extraction yield for muskmelon based on time

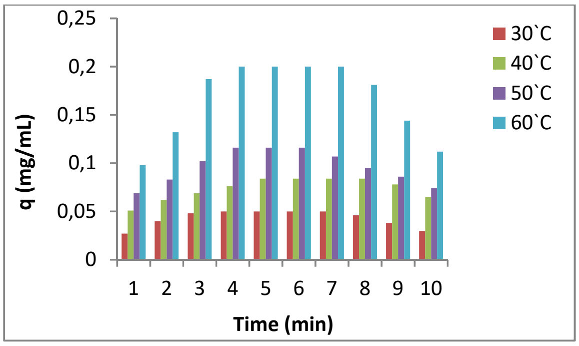

Fig. 10 Extraction yield for watermelon based on time

The extraction efficiency was found to increase with increasing temperature at certain level due to increased solubility of carotenoids in extraction solvent. From all the extraction techniques reviewed, the increase of temperature affects positively the mass transfer process and consequently the extraction yield of carotenoids. In addition, at higher temperature, the liquid viscosity and density decreased, resulting in increased mass transfer. Furthermore, because of decreased solvent viscosity, cavitation bubbles within the fluid occurred more easily as the cohesive force.

IV. Conclusion

The extraction of carotenoids from muskmelon shell and watermelon shell using MAE and UAE were studied. Extraction conditions are feed-solvent ratio (1:10 to 1:40), microwave power (100 W, 150 W, 200 W and 300 W), ultrasound frequency (40 kHz), temperature (30, 40, 50, and 60 °C), solvent (Hexane and Ethanol) and particle size (0.6 mm and 0.5 mm). In MAE, the optimum conditions obtained for extraction of carotenoids from muskmelon shell were 400 W, 0.5 mm and 120 s and for watermelon the optimum conditions are 500 W, 0.5 mm, and 120 s. In UAE, the optimum conditions for extraction of carotenoids from muskmelon shell were 40 kHz, 60 °C, and 5 min and for watermelon the optimum conditions were 40 kHz, 70 °C, and 5min. The extracted carotenoids were analyzed using UV-Visible Spectrophotometry and High performance liquid chromatography (HPLC). In MAE, the yield of carotenoids was found to be 590.85 µg/g for muskmelon and 474.72 µg/g for watermelon. In UAE, the yield of carotenoids was found to be 775.25 µg/g for muskmelon and 639.54 µg/g for watermelon. The study concluded that, UAE gives the higher yield of carotenoids than MAE and also muskmelon had the highest yield of carotenoids when ethanol used as a solvent for 0.5 mm particle size. The extraction temperature, microwave power, and F-S ratio are found to be most important parameters affecting the extraction efficiency of carotenoids from muskmelon and watermelon shell.

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