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Research paper

Effect of Particle Size and Temperature of Soaking on Water Extraction of Kokum Rind Powder

Jidnyasa Natwar Solanki[1] and Shrikant Baslingappa Swami[2*]

[1]Department of Agricultural Process Engineering, College of Agricultural Engineering and Technology, Dr. Balasaheb Sawant Konkan Krishi Vidyapeeth, Dapoli, Dist Ratnagiri, Maharashtra, India
[2]Department of Post-Harvest Engineering, Post Graduate Institute of Post-Harvest Technology and Management, Killa-Roha, Dist: Raigad, (Dr. Balasaheb Sawant Konkan Krishi Vidyapeeth, Dapoli-Campus Roha), Maharashtra, India

*Corresponding author: swami_shrikant1975@yahoo.co.in

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ABSTRACT

Garcinia indica Choisy commonly known as Kokum belonging to the Gutifferae family. Kokum is one of the important indegenous tree spice crops grown in tropical rain forests of western ghats in Konkan, Goa, South Karnataka and Kerala. It is also flourish in ever green forest of Assam, Khasi, Jayantia hills, West Bengal and Surat district of Gujarat. Area under cultivation of Kokum is 1200 ha in South Maharashtra and total production 10200 ha tonnes with ripe fruit yield of 8.5 tonnes/ha. In the present study the effect of temperature of Soaking and time of soaking on TSS, Acidity, viscosity, anthocyanins of extract has been studied. The Kokum powder at 0.30 mm, 0.25 mm and 0.15 mm particle was taken for extraction. The Kokum powder at 0.30 mm, 0.25 mm and 0.15 mm particle sizes has been soaked in water at Kokum powder: water ratio 1:3 and 1:2. The temperature of soaking were (ambient (22°C), 40°C, 50°C, 60°C). The desirable quality of Kokum rind powder extract was based on the responses i.e. maximum TSS, maximum acidity, maximum viscosity and maximum anthocyanins. The effect of temperature of soaking and time of soaking of Kokum powder in water indicated that the optimum values i.e. the maximum values of TSS, acidity, viscosity and anthocyanins are 19.0°B, 0.47%, 14.2 cP and 7.0% respectively. These values were achieved at the 0.30 mm particle size of Kokum rind powder, the Kokum rind powder: water ratio 1:2 and 50°C soaking temperature for soaking duration 6.5 h.

Keywords: *Garcinia indica,* Kokum, Kokum rind, ratio, temperature

Garcinia indica Choisy commonly known as Kokum belonging to the Gutifferae family. *Garcinia indica* is synonymous with Garcinia Purppurea, it is known as *Bridol* in Goa, *Bhirind* or amsul in Marathi and Konkani, *Murugal* in Kannada and *Punampuli* in Malayalam (Kureel *et*. *al*., 2009). Kokum is one of the important indegenous tree spice crops grown in tropical rain forests of western ghats in Konkan, Goa, South Karnataka and Kerala. It is also flourish in ever green forest of Assam, Khasi, Jayantia hills, West Bengal and Surat district of Gujarat. Heavy concentration of Kokum tree found in Sindhudurg. The Kokum tree originated from the tropical rain forest of the Western ghats of Kerala and Malaysia. Its cultivation is confined to the coastal hilly regions of Maharashtra and Goa states. It is popularly known as "*Ratamba*". Some Kokum tree are also observed in Tamil Nadu, the Western ghats of Karnataka and Kerala, as well as parts of West Bengal, Assam and Gujarat (Kureel *et al*. 2009).

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Kokum, in its natural habit, it comes up well in a hot humid climate under partial shade. Area under cultivation of Kokum is 1200 ha in South Maharashtra and total production 10200 ha tonnes with ripe fruit yield of 8.5 tonnes/ha (Karnik *et al.* 2001). It is slow growing slender tree of moderate size having a beautiful conical shape. It bears spherical fruit with arillate seeds.

Kokum rind contains the highest concentration of anthocyanins (2.4 g/100 g of kokum fruit) (Nayak *et al*. 2010). The anthocyanins cyanidin-3-glucoside and cyanidin-3-sambubioside are the major pigment present in Kokum and is reported to occur in the ratio of 4:1 (Krishnamurthy *et al.,* 1982; Nayak *et al.* 2010). The rinds also contain two polyisoprenylated phenolics garcinol and isogarcinol (Rastogi and Mehrotra, 1990). Studies have shown that (−) hydroxycitric acid (HCA) is the major organic acid in Kokum rinds. It is responsible for the savoury taste of Kokum and is present to the extent of 4.1–4.6 and 10.3–12.7%, respectively in leaves and fruits moisture 80%, crude fibre 14.28%,tannins 2.85%, starch 1.00%, acid (hydroxy citric acid) 22.90%, ascorbic acid 0.06%, protein (N x 6.25) 1.92%, Total ash 2.57%, pectin 5.71%, crude Fat 10.00%, pigment 2.40%, carbohydrate by difference 35.00% (Sampathi and Krishnamurthy, 1982). Kokum consists of glycerids of satiric acid (55%), oleic acid (40%), palmtic acid (2.5%), hydroxyl caprice acid (10%), and linoleic acid (1.5%).

Kokum fruits contain rich amounts of anti-oxidants that combine with free radicals and avoid oxidative damage to body cells. They also support cell regeneration and repair. It is cardio tonic and useful for treatment of piles, dysentery, tumours, pains and heart complaints. Antioxidant, improves cardiovascular function, it healing property, skin care, aid digestion, it is also useful to cure cancer.

Kokum is an important culinary agent and is used as an acidulant for curries by people living in Maharashtra, costal Karnataka and Goa, India. In summer the ripe kokum rinds are ground in a pulverizer with sugar and cardamom and consumed as a cooling drink (Shenoy, 1989; Menezes, 2000; Menezes, 2002; Padhye *et al.* 2009). Addition of Kokum is supposed to enhance the taste of coconut-based curries and to remove the unpleasant smell of mackerel and sardines (Menezes, 2000; Menezes, 2002). They are also used in some vegetable dishes and to prepare chutneys and pickles (especially with prawns and cartilaginous fishes like sharks, ray fish and other cartilaginous fish) (Menezes, 2000; Menezes, 2002). The Kokum rinds are commercially used to prepare concentrate which is free from salt and sugar and can be used as an acidulant syrup and agal preparation. The concentrate could be a valuable raw material for making various fruit based liquid mixes like sarbet, solkadhi and Kokum powder. It can also be used as an important raw material for pharmaceutical industries. The local community of Goa also use the rinds to prepare wine. Dried rinds are powdered and marketed to be used as acidulant for traditional curries (Nayak *et al.* 2010).

Fruit has very short shelf-life of 2-3 days after ripening and cannot be consumed as fresh fruit hence processing is vital. Rind also has much commercial application such as colour pigment, wine, concentrate and powder etc. In the present study it is aimed to extract Kokum powder in water and to study the various properties of extracts.

Based on the above information the present study was undertaken with the following objective. To study the extraction of Kokum rind powder in water at varied temperatures and particle sizes.

MATERIALS AND METHODS

Materials

The Kokum powder from dry rinds of *Garcinia Indica* have been used from NAIP laboratory of Department of Agricultural Process Engineering, CAET, Dapoli.

1. Moisture content

The initial moisture content of the Kokum rind was determined by using hot air oven method. The 50-60 g kokum rind was kept in the moisture boxes. The moisture boxes were kept in hot air oven at 105˚C±1

for 24 h. The final weight of Kokum rind after 24 h was recorded. The moisture content of the Kokum rind was determined by following formula (Chakraverty, 1994).

$$\text{Moisture Content (db)\%} = \frac{W_m}{W_d} \times 100$$

Where, W_m = weight of moisture, g

W_d = weight of bone dry material, g

Soaking of Kokum rind powder

50g of dried Kokum rind with initial moisture content 7 %db was ground in the pulverize (Make: M/S SEW, Kudal) to a particle size≤ 0.3mm, ≤0.25mm and ≤0.15mm respectively at varied time intervals 5, 9 and 12 minutes respectively. The Kokum rind powder so obtained were soaked in water i.e.1:3 and 1:2 (Kokum powder: water) at varied temperatures i.e. ambient (22±2); 40, 50 and 60°C respectively. The temperature of soaking was achieved by keeping the beakers in the waterbath (Make: Unix96 Biotechnolab, Mumbai) by adjusting the knob to 40, 50 and 60°C respectively with an accuracy of ±**1**°C. The Kokum extract in water was recorded at varied time intervals i.e. 1, 2, 3, 4, 5, 6, 7 and 8h respectively. The experiment was replicated four times.

The treatment details are given in Table 1.

1. Measurement of Total Soluble Solids (TSS) of Kokum Extracts at different temperatures

The TSS of all the Kokum extract was measured by using digital refractometer (Make: Atago, Japan). The prism of the refractometer was cleaned with the help of distilled water and tissue paper. The distilled water was used as the TSS value of distilled water is zero and is known. This was used as standard for calibration. Kokum extract samples Kokum rind powder: water ratios 1:3 (at varied particle sizes i.e. 0.300 mm, 0.250 mm and 0.150 mm) and at the varied temperature 22, 40, 50 and 60°C for varied time intervals i.e. 1, 2, 3, 4, 5, 6, 7, 8h respectively was taken for the study. Extract sample was then taken into the beakers of 50 ml capacity. The sample was stirred with the help of spoon just to ensure a proper mixing and to have homogeneity. The temperature readings were taken using thermometer in ˚C. Then 0.3-0.5 ml sample of fruit product was placed on the prism with the help of plastic spoon. Plastic spoon was used so as to avoid the damage to the screen and the prism. The equipment gives its TSS reading within 5 s. Readings were taken at different temperature ranging from 22˚C to 60˚C. Four replications were taken at each temperature. The average value

Table 1: Treatment details for the Kokum extraction in water

Sl. No.	Kokum rind powder: Water	Particle size of kokum powder	Temperature of extraction (°C)	Time interval (h)
1	1:3	0.30 mm	22	1, 2, 3, 4, 5, 6, 7, 8
2			40	1, 2, 3, 4, 5, 6, 7, 8
3			50	1, 2, 3, 4, 5, 6, 7, 8
4			60	1, 2, 3, 4, 5, 6, 7, 8
5	1:3	0.25 mm	22	1, 2, 3, 4, 5, 6, 7, 8
6			40	1, 2, 3, 4, 5, 6, 7, 8
7			50	1, 2, 3, 4, 5, 6, 7, 8
8			60	1, 2, 3, 4, 5, 6, 7, 8
9	1:3	0.15 mm	22	1, 2, 3, 4, 5, 6, 7, 8
10			40	1, 2, 3, 4, 5, 6, 7, 8
11			50	1, 2, 3, 4, 5, 6, 7, 8
12			60	1, 2, 3, 4, 5, 6, 7, 8

Various properties i.e. Total soluble solids, Acidity, Viscosity and anthocyanins of the extract have been determined.

was then calculated from four readings for each temperature. Same procedure was followed for remaining all the products. The graph of TSS versus temperature (°C) and time of extraction (h) was then plotted and results were obtained.

2. Acidity of Kokum extract

Acidity was calculated by using titration method (Ranganna, 1986). 1g of Kokum extract was taken for the study. 20 ml distilled water was added to it. Pipette out 1 ml of this sample in conical flask and 100 ml distilled water was added to it. 2-3 drops of phenolphthalein indicator was added to it. The solution was titrated with 0.1 N NaOH. End point is feint pink colour. Acidity was calculated by using equation:

$$\text{Total Acid, \%} = \frac{\text{B.R.} \times \text{Normality} \times \text{Vol. madeup} \times \text{equivalent wt. of acid} \times 100}{\text{vol. of sample taken for estimation} \times \text{wt. of sample} \times 1000}$$

3. Viscosity measurements of Kokum Extracts at different temperatures

Kokum extract of 100 ml was taken for the viscosity determination. Viscosity measurements were carried out using Brookfield viscometer model (DV-II+ Pro). The measurements were carried out at temperatures 22 ±1°C, 40°C, 50°C and 60°C for the extracts at different time intervals i.e. 1, 2, 3, 4, 5, 6, 7 and 8h respectively. Spindle no. 63 was used for the viscosity determination in the present research work at 150 rpm rotational speed. Four replications were performed. 100 ml of extract sample were taken in beaker and viscosity was measured at 150 rpm rotational speed the readings were recorded after its stabilization.

The Brookfield viscometer was leveled on the platform and the spindle no 63 was attached to the viscometer by screwing it onto the lower shaft. Kokum extract at various time intervals and at various temperature of extraction were filled in a 100 ml beaker. The spindle was dipped in the sample up to mark on it at the center of the beaker. The rheological data was recorded at the speeds mentioned above by pressing the enter key on the equipment. Viscosity was displayed in centipoise (cP). Four replications for each sample at each temperature were taken. Similar procedure was followed for extract for different particle size and at various temperatures and Kokum rind powder: water ratios.

4. Anthocyanin

Anthocyanin of the Kokum extract was determined by spectrophotometric method (Ranganna, 1986). In this method, Anthocyanin was extracted with ethanolic HCL (85:15). 1 g Kokum extract was taken and mixed with 10 ml of ethanolic HCL and was kept overnight in refrigerator at 4°C. Next day after making volume with ethanolic HCL, mixture was filtered. Absorbance of this filtered solution was recorded at 535 nm against blank solution. The absorbance was reported as anthocyanin content of dried Kokum rind.

Anthocyanin was calculated by using equation:

$$\text{Total Optical Density} = \frac{\text{OD} \times \text{vol. made up} \times 100}{\text{wt. of sample}}$$

$$\text{Total Anthocyanin (mg)/(100g)} = \frac{\text{otal OD} \times 100}{98.2}$$

RESULTS AND DISCUSSION

Effect of Temperature of soaking and time of soaking of Kokum rind powder in water for Kokum rind powder: water (1:3) for 0.300 mm particle size of Kokum rind powder

1. TSS (T_{ss})

Fig. 1 shows the effect of soaking temperature and time of soaking of Kokum rind powder in water on the TSS of the extract at 0.300 mm particle size. The TSS for the extract was 7.47°B to 18.57°B. As the temperature of soaking increases from 22°C to 60°C the TSS of extract increases. As the soaking time increases the TSS of the extract increases. The effect of temperature (°C) of soaking and time of soaking

(h) on TSS of the extract at 1:3 (Kokum rind powder: water) for particle size 0.300 mm is represented by Equation.

$$T_{SS} = 6.520 - 0.33T - 0.013t - 0.0117T^2 + 0.002t^2 + 0.023Tt$$

$$R^2 = 0.991$$

Where, T- Temperature (°C); t - time (h)

From above equation, it was also observed that the combined effect of temperature of extraction and time of extraction have positive effect on TSS of the extract and it increases the TSS of extract. Table 2 shows the ANOVA for the TSS of Kokum extract. It was observed that effect of soaking temperature and soaking time is highly significant at F = 0.4729

Table 2: ANOVA for TSS of Kokum rind powder: water (1:3) at particle size 0.300 mm at different temperature

Responses	Degree of Freedom	Sum of Squares	Mean Square	Significance *F
Regression	7	56.23631	8.03375	
Residual	24	407.7072	16.9878	0.4729
Total	31	463.9435		

*$p \leq 0.05$.

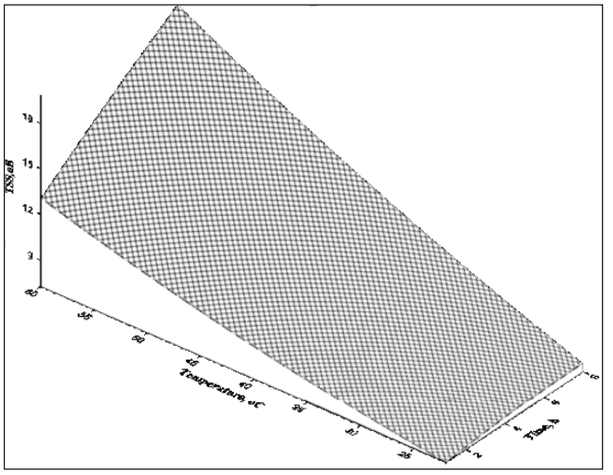

Fig. 1: Surface plot of effect of temperature of soaking and soaking time of Kokum rind powder: water on TSS at 1:3 level of 0.300 mm particle size

2. Acidity (A_c)

Fig. 2. shows effect of soaking temperature and time of soaking of Kokum rind powder in water on the Acidity of the extract at 0.300 mm particle size. The Acidity of the extract was 0.37% to 0.57%. As the temperature increases from 22°C to 45°C the acidity of extract increases gradually up to 45°C followed by decreasing trend up to 60°C. As the soaking time of the Kokum rind powder increases Acidity of the extract decreases. The effect of temperature (°C) of soaking and time of soaking (h) on acidity of the extract at 1:3 (Kokum rind powder: water) for particle size 0.300 mm is represented by equation.

$$A_c = 0.277 - 0.016T - 0.017t - 0.001T^2 - 0.001t^2 - 0.001Tt$$

$$R^2 = 0.978$$

Where, T- Temperature (°C); t - time (h)

From above equation, it was also observed that the combined effect of temperature of extraction and time of extraction have negative effect on acidity of the extract. Table 3 shows the ANOVA for the acidity of Kokum rind powder extract. It was observed that effect of soaking temperature and soaking time is highly significant at F = 3.2186.

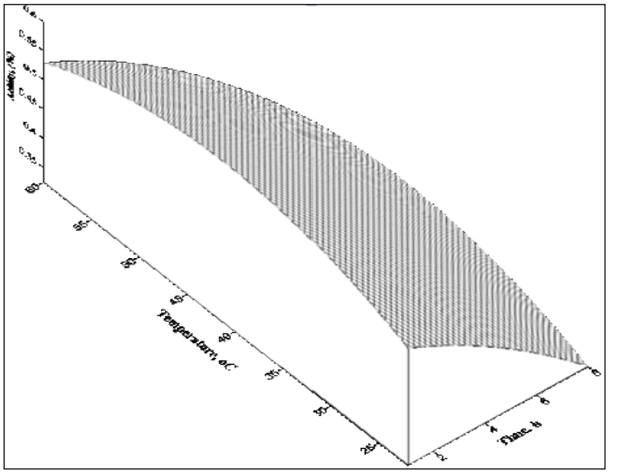

Fig. 2: Surface plot of effect of temperature of soaking and time of soaking Kokum rind powder: water on Acidity at different temperature at 1:3 level of 0.300 mm particle size

Table 3: ANOVA for Acidity of Kokum rind powder: water (1:3) at particle size 0.300 mm at different temperature

Responses	Degree of Freedom	Sum of Squares	Mean Square	Significance *F
Regression	7	0.16345	0.02335	
Residual	24	0.17410	0.007255	3.2186
Total	31	0.33755		

*$p \leq 0.05$.

3. Viscosity (V_s)

Fig. 3 shows the effect of temperature of soaking and soaking time of Kokum rind powder in water on the viscosity of the extract at 0.300 mm particle size. The viscosity for extract was 3.3 cP to 16.57 cP. As the temperature of soaking increases from 22°C to 60°C the viscosity of the extract increases. The soaking time of the Kokum rind powder in water increases the viscosity of the extract decreases. The effect of temperature (°C) of soaking and time of soaking (h) on viscosity of the extract at 1:3 (Kokum rind powder: water) for particle size 0.300 mm is represented by equation.

$$V_s = 5.872 - 1.779T + 0.046t - 0.061T^2 - 0.001t^2 + 0.046Tt$$

$$R^2 = 0.990$$

Where, T - Temperature (°C); t - time (h)

From above equation, it was also observed that the combined effect of temperature of extraction and time of extraction have positive effect on viscosity of the extract the viscosity of the extract increased. Table 3 shows the ANOVA for the viscosity of Kokum rind powder extract. It was observed that effect of soaking temperature and soaking time is highly significant at F = 1.2217.

Table 4: ANOVA for viscosity of Kokum rind powder: water (1:3) at particle size 0.300 mm at different temperature

Responses	Degree of Freedom	Sum of Squares	Mean Square	Significance *F
Regression	7	101.1942	14.4563	
Residual	24	283.989	11.8328	1.2217
Total	31	385.1832		

*$p \leq 0.05$.

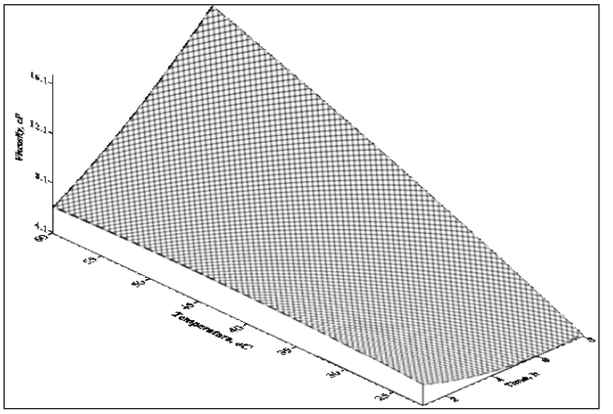

Fig. 3: Surface plot of effect of temperature of soaking and time of soaking of Kokum rind powder in water on Viscosity, cP at 1:3 level of 0.300 mm particle size

4. Anthocyanin % (A_n)

Fig. 4. shows the effect of Temperature of soaking and time of soaking of Kokum rind powder in water on the Anthocyanins of the extract at 0.300 mm particle size. The anthocyanin of extract was 0.92% to 5.30%. As the temperature of soaking increases from 22°C to 60°C the anthocyanins of the extract increases. Also on the soaking time of the Kokum rind powder in water increases, the anthocyanins of the extract increases. The effect of temperature (°C) of soaking and time of soaking (h) on of the extract at 1:3 (Kokum rind powder: water) for particle size 0.300 mm is represented by equation.

$$A_n = 2.848 - 0.272T - 0.112t - 0.044T^2 - 0.002t^2 - 0.001Tt$$

$$R^2 = 0.988$$

From above equation, it was also observed that the combined effect of temperature of extraction and time of extraction have negative effect on anthocyanins of the extract and it decreases anthocyanins of extract. Table 5 shows the ANOVA for the Anthocyanins of Kokum rind powder extract. It was observed that effect of soaking temperature and soaking time is highly significant at F = 0.3067

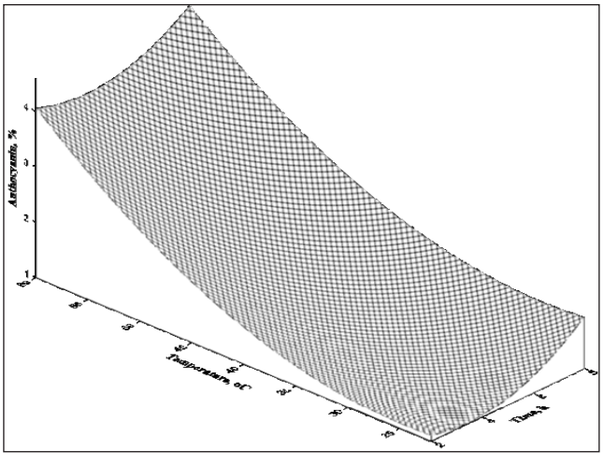

Fig. 4: Surface plot of effect of Temperature of soaking time of Kokum rind powder in water on Anthocyanins, % at 1:3 level of 0.300 mm particle size

Table 5: ANOVA for Anthocyanins of Kokum rind powder in water (1:3) at particle size 0.300 mm at different temperature

Responses	Degree of Freedom	Sum of Squares	Mean Square	Significance *F
Regression	3	1.5159	0.5053	
Residual	12	197674	1.6472	0.3067
Total	15	21.2834		

*$p \leq 0.05$.

5. Superimposed contour plots for the extracts

Desirable qualities of Kokum rind powder extract are maximum TSS, Maximum Acidity, Maximum Viscosity and maximum Anthocyanins. Fig. 5 shows the superimposed contour plots for effect of soaking temperature and soaking time of Kokum rind powder in water on TSS, Acidity, Viscosity, Anthocyanins.

It was observed that the optimum zone of Temperature of soaking and time of soaking of Kokum rind extract occurred at Temperature of soaking 35 (ºC) to 48 (ºC), Time of soaking, 5.2 (h) to 6.8 (h). At this optimum zone the TSS is 12.75 ºB to 15 ºB; Acidity is 0.41 % to 0.48 %; Viscosity is 10 cP to 12 cP and Anthocyanins are 1.8 % to 2.95 %.

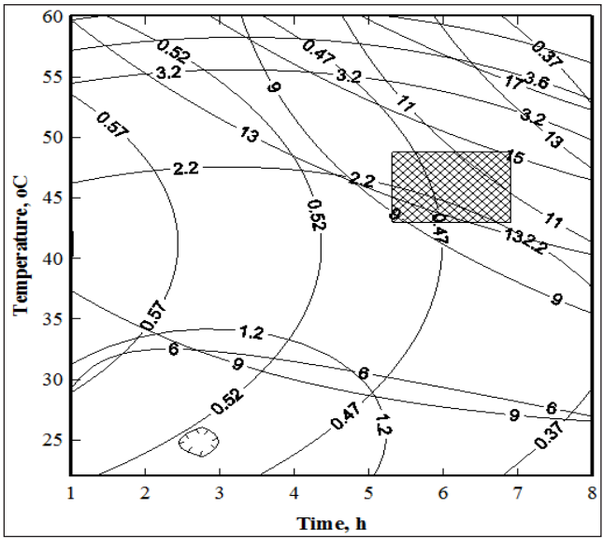

Fig. 5: Superimposed contour plots showing the optimum zone of Temperature of soaking and time of soaking Kokum rind powder in water at 1:3 level of 0.300 mm particle size

Effect of Temperature of soaking and time of soaking of Kokum rind powder in water for Kokum rind: water (1:3) for 0.250 mm particle size of Kokum rind powder.

1. TSS (T_{ss})

Fig. 6 shows the effect of soaking temperature and time of soaking of Kokum rind powder in water on the TSS of the extract at 0.250 mm particle size. The TSS for extract was 12.02°B to 15.57°B. As the temperature of soaking increase from 22°C to 45°C, The TSS of extract decreases followed by increasing trend with increase in temperature from 45°C to 60°C. As the soaking time of the Kokum rind powder increases, the TSS of the extract decreases. The effect of temperature (°C) of soaking and time of soaking (h) on TSS of the extract at 1:3 (Kokum rind powder: water) for particles size 0.250 mm is represented by equation.

$$T_{ss} = 14.422 - 0.2202T - 0.123t - 0.009T^2 + 0.001t^2 + 0.0011Tt$$

$$R^2 = 0.999$$

From above equation, it was also observed that the combined effect of temperature of extraction and time

of extraction have positive effect on TSS of the extract and it increases the TSS of extract. Table 6 shows the ANOVA for the TSS of Kokum rind powder extract. It was observed that effect of soaking temperature and soaking time is highly significant at F = 0.9187.

Table 6: ANOVA for TSS of Kokum rind powder in water (1:3) at particle size 0.250 mm at different temperature

Responses	Degree of Freedom	Sum of Squares	Mean Square	Significance *F
Regression	7	5.23871	0.74838	
Residual	24	19.5494	0.81455	0.9187
Total	31	24.7881		

*$p \leq 0.05$.

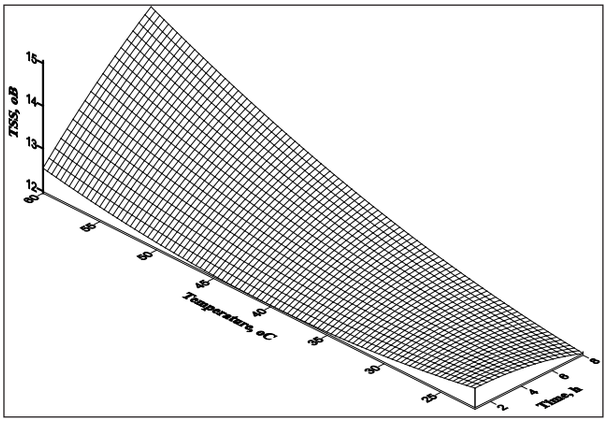

Fig. 6: Surface plot of effect of Temperature of soaking and time of soaking of Kokum rind powder in water on TSS at 1:3 level of 0.250 mm particle size

2. Acidity (A_c)

Fig. 7 shows effect of temperature of soaking and time of soaking of Kokum rind powder in water on Acidity of the extract at for 0.250 mm particle size. The Acidity for the extract was 0.17% to 0.67%. As the temperature of soaking increases from 22°C to 50°C the acidity of extract increases, however further increase in temperature decreases acidity of extract. It can be seen from the graph that as the soaking time of the Kokum rind powder in water increases the acidity of extract decreases. The effect of temperature (°C) of soaking and time of soaking (h) on acidity of extract at 1:3 (Kokum rind powder: water) for particle size 0.250 mm is represented by equation.

$$A_c = -0.206 + 0.034T + 0.041t - 0.010T^2 - 0.001t^2 + 0.001Tt$$

$$R^2 = 0.755$$

Where, T- Temperature (°C); t - time (h).

From above equation, it was also observed that the combined effect of temperature of extraction and time of extraction have positive effect on TSS of the extract and it increases acidity of extract. Table 7 shows the ANOVA for the acidity of Kokum rind powder extract. It was observed that effect of soaking temperature and soaking time is highly significant at F = 0.6887.

Table 7: ANOVA for acidity of Kokum rind powder in water (1:3) at particle size 0.250 mm at different temperature

Responses	Degree of Freedom	Sum of Squares	Mean Square	Significance *F
Regression	6	8.038	1.340	
Residual	26	2.604	0.100	0.6887
Total	32	10.641		

*$p \leq 0.05$.

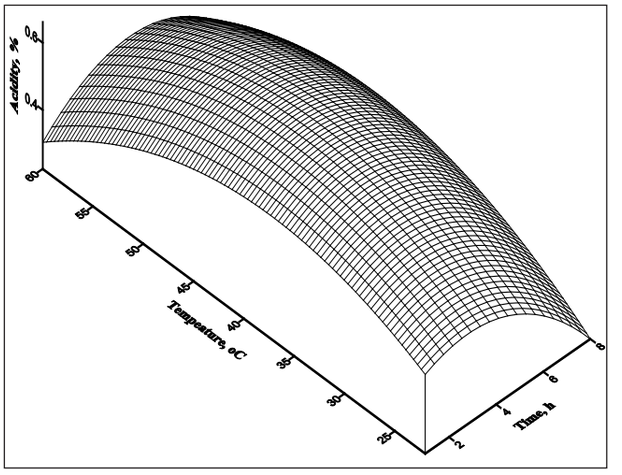

Fig. 7: Surface plot of effect of temperature of soaking and time of soaking Kokum rind powder in water on Acidity at 1:3 level of 0.250 mm particle size

3. Viscosity (V_s)

Fig. 8 shows effect of soaking temperature and time of soaking of Kokum rind powder in water on viscosity of the extract at 0.250 mm particle size. The viscosity for the extract was 1.06 cP to 13.72 cP. As the temperature of soaking increases from 22°C to 60°C the viscosity of extract decreases. The time of soaking of the Kokum rind powder in water increases the viscosity of extract decreases initially up to 4h and then the viscosity of the extract increases. The effect of temperature (°C) of soaking and time of soaking (h) on viscosity of extract at 1:3 (Kokum rind powder: water) for particle size 0.250 mm is represented by equation.

$$V_s = 8.456 - 1.959T - 0.250t + 0.141T^2 - 0.002t^2 + 0.043Tt$$

$$R^2 = 0.968$$

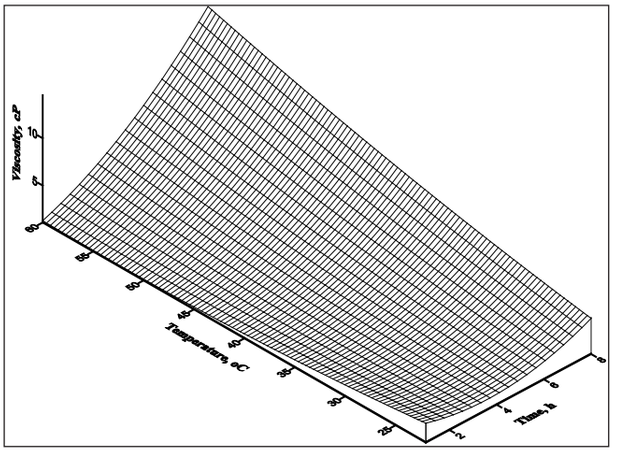

Fig. 8: Surface plot of effect of temperature of soaking and time of soaking of Kokum rind powder in water on Viscosity at 1:3 level at 0.250 mm particle size

From above equation, it was also observed that the combined effect of temperature of extraction and time of extraction have positive effect on viscosity of the extract and it increases viscosity of extract. Table 7 shows the ANOVA for the viscosity of Kokum rind powder extract. It was observed that effect of soaking temperature and soaking time is highly significant at F = 5.3408.

Table 7: ANOVA for viscosity of Kokum rind powder in water (1:3) at particle size 0.250 mm at different temperature

Responses	Degree of Freedom	Sum of Squares	Mean Square	Significance *F
Regression	7	251.2624	35.8946	
Residual	24	161.2989	6.7207	5.3408
Total	31	412.5613		

*$p \leq 0.05$.

4. Anthocyanin % (A_n)

Fig. 9 shows effect of soaking temperature and time of soaking of Kokum rind powder in water on anthocyanins of the extract at for 0.250 mm particle size. The anthocyanin for the extract was 1.39% to 3.55%. As the temperature of soaking increases from 22°C to 60°C the anthocyanins of extract increases. The soaking time of the Kokum rind powder in water increases the anthocyanins of extract increases. The effect of temperature (°C) of soaking and time of soaking (h) on anthocyanins of extract at 1:3 (Kokum rind powder: water) for particle size 0.250 mm is represented by equation.

$$A_n = 0.656 - 0.162T - 0.004 - 0.004T^2 + 0.001t^2 + 0.001Tt$$

$$R^2 = 0.994$$

Where, T – temperature; t - time

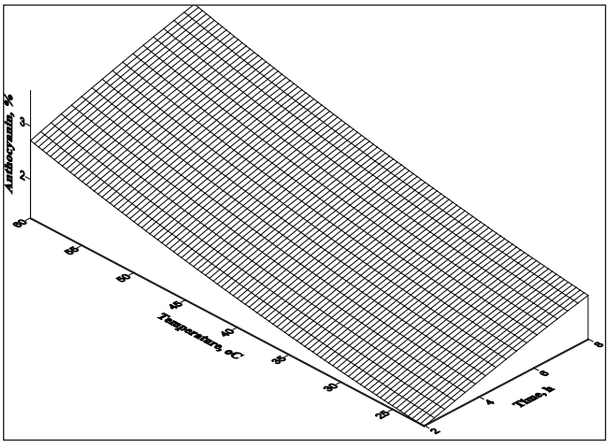

Fig. 9: Surface plot of effect of temperature of soaking and time of soaking of Kokum rind powder in water on Anthocyanins at 1:3 level of 0.250 mm particle size

From above equation, it was also observed that the combined effect of temperature of extraction and time of extraction have positive effect on Anthocyanins of the extract Table 8 shows the ANOVA for the anthocyanins of Kokum rind powder extract. It was observed that effect of soaking temperature and soaking time is highly significant at F = 1.2223.

Table 8: ANOVA for anthocyanins of Kokum rind powder in water (1:3) at particle size 0.250 mm at different temperature

Responses	Degree of Freedom	Sum of Squares	Mean Square	Significance *F
Regression	3	1.7131	0.5710	
Residual	12	5.6061	0.4671	1.2223
Total	15	7.3193		

*$p \leq 0.05$.

5. Superimposed contour plots for the extracts

Desirable qualities of Kokum rind powder extract are maximum TSS, Acidity, Viscosity, Anthocyanins. Fig 10 shows the superimposed contour plots for effect of soaking temperature of Kokum rind powder in water on TSS, Acidity, Viscosity, Anthocyanins.

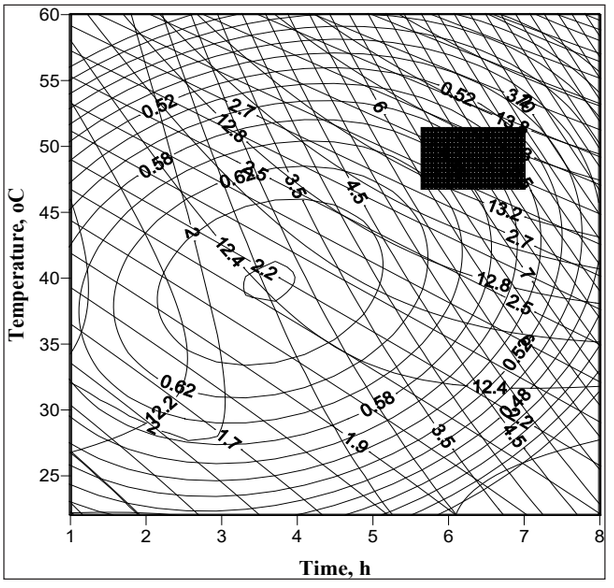

Fig. 10: Superimposed contour plots showing the optimum zone of Temperature of soaking and time of soaking Kokum rind powder in water at 1:3 level of 0.250 mm particle size

It was observed that the maximum value of TSS 13°B to 13.8°B, acidity 0.51% to 0.59 %, Viscosity 5.81 cP to 9.62 cP, anthocyanins 2.6% to 3.25% was observed in optimum zone where the temperature of soaking 46^0C to 53°C and time of soaking 5.6 h to 7.1 h. The optimum zone of maximum TSS, acidity, viscosity, anthocyanins shown in Fig 10.

Effect of Temperature of soaking and time of soaking of Kokum rind powder in water for Kokum rind powder: water (1:3) for 0.150 mm particle size of Kokum rind powder.

1. TSS (T_{ss})

Fig. 11 shows the effect of temperature of soaking and time of soaking of Kokum rind powder in water on the TSS of the extract at 0.150 mm particle size. The TSS of extract was 11.55°B to 18.12°B. As the temperature of soaking increase from 22°C to 60°C, the TSS of extract increases. The time of soaking of the Kokum rind powder increases, the TSS of the extract increases. The effect of temperature (°C) of soaking and time of soaking (h) on TSS of the extract at 1:3 (Kokum rind powder: water) for particles size 0.150 mm is represented by equation.

$$T_{ss} = 15.913 - 0.039T - 0.041t - 0.004T^2 - 0.001t^2 + 0.008Tt$$

$R^2 = 1.0$

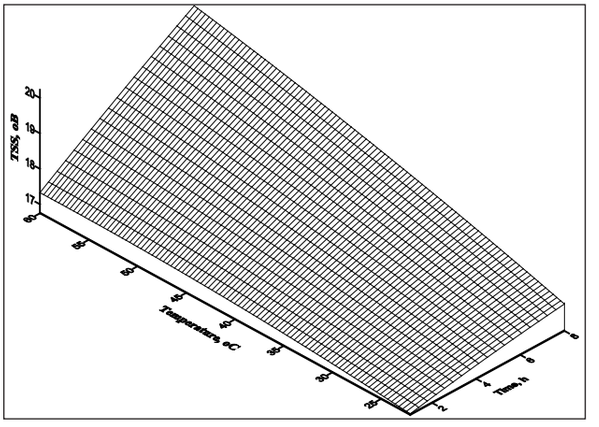

Fig. 11: Surface plot of effect of Temperature of soaking and time of soaking of Kokum rind powder in water on TSS at 1:3 level of 0.150 mm particle size

From above equation, it was also observed that the combined effect of temperature of extraction and time of extraction have positive effect on TSS of the extract and it increases the TSS of extract.

Table 9 shows the ANOVA for the TSS of Kokum rind powder extract. It was observed that effect of soaking temperature and soaking time is highly significant at F = 2.5729.

Table 9: ANOVA for TSS of Kokum rind powder in water (1:3) at particle size 0.150 mm at different temperature

Responses	Degree of Freedom	Sum of Squares	Mean Square	Significance *F
Regression	7	13.4486	1.92123	
Residual	24	17.9209	0.7467	2.5729
Total	31	31.3695		

*$p \leq 0.05$.

2. Acidity (A_c)

Fig. 12 shows effect of temperature of soaking and time of soaking of Kokum rind powder in water on Acidity of the extract for 0.150 mm particle size. The Acidity for the extract was 0.21% to 0.64%. As the temperature of soaking increases from 22°C to 60°C the acidity of extract increases. The soaking time of the Kokum rind powder in water increases the acidity of extract decreases. The effect of temperature (°C) of soaking and time of soaking (h) on acidity of extract at 1:3 (Kokum rind powder: water) for particle size 0.150 mm is represented by equation.

$$A_c = 0.958 + 0.004T - 0.020t - 0.002T^2 + 0.001t^2 - 0.001Tt$$

$$R^2 = 0.983$$

From above equation, it was also observed that the combined effect of temperature of extraction and time of extraction have negative effect on acidity of the extract and it decreases acidity of extract. Table 12 shows the ANOVA for the acidity of Kokum rind powder extract. It was observed that effect of soaking temperature and soaking time is highly significant at F = 10.8496.

Table 12: ANOVA for Acidity of Kokum rind powder in water (1:3) at particle size 0.150 mm at different temperature

Responses	Degree of Freedom	Sum of Squares	Mean Square	Significance *F
Regression	7	0.82166	0.11738	
Residual	24	0.25965	0.0108	10.8496
Total	31	1.08132		

*$p \leq 0.05$.

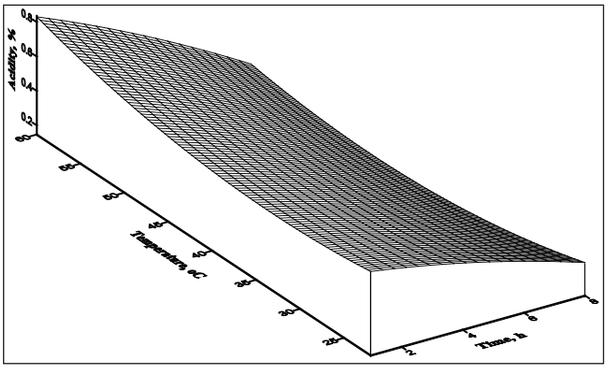

Fig. 12: Surface plot of effect of temperature of soaking and time of soaking of Kokum rind powder in water on Acidity at 1:3 level of 0.150 mm particle size

3. Viscosity (V_s)

Fig.13 shows effect of soaking temperature and time of soaking of Kokum rind powder in water on viscosity of the extract for 0.150 mm particle size. The viscosity for the extract was 2.13 cP to 8.76 cP. As the temperature of soaking increases from 22°C to 45°C the viscosity of extract increases further increase in temperature from 45°C to 60°C, the viscosity of extract decreases. The soaking time of the Kokum rind powder in water increases the viscosity of extract increases. The effect of temperature (°C) of soaking and time of soaking (h) on viscosity (cP) of extract at 1:3 (Kokum rind powder: water) for particle size 0.150 mm is represented by equation.

$$V_s = -11.193 - 0.093T - 0.808t + 0.027T^2 - 0.010t^2 + 0.020\ Tt$$

$$R^2 = 0.970$$

From above equation, it was also observed that the combined effect of temperature of extraction and

time of extraction have positive effect on viscosity of the extract and it increases viscosity of extract. Table 13 shows the ANOVA for the viscosity of Kokum rind powder extract. It was observed that effect of soaking temperature and soaking time is highly significant at F = 4.6735.

Table 13: ANOVA for viscosity of Kokum rind powder in water (1:3) at particle size 0.250 mm at different temperature

Responses	Degree of Freedom	Sum of Squares	Mean Square	Significance *F
Regression	7	247.258	35.3226	
Residual	24	181.39	7.5579	4.6735
Total	31	428.648		

*$p \leq 0.05$.

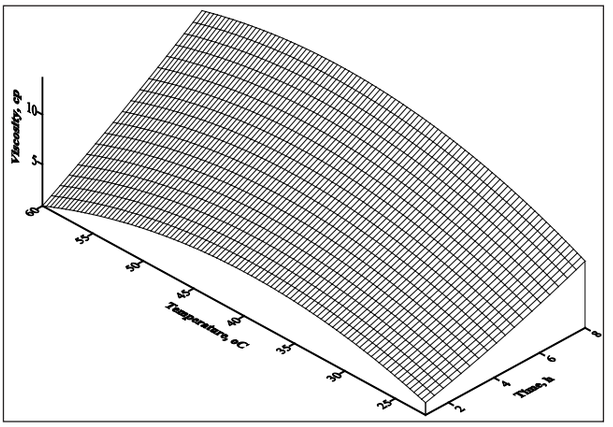

Fig. 13: Surface plot of effect of temperature of soaking of Kokum rind powder in water on Viscosity at 1:3 level at 0.150 mm particle size

4. Anthocyanin % (A_n)

Fig. 14 shows effect of temperature of soaking of Kokum rind powder in water on anthocyanins of the extract for 0.150 mm particle size. The anthocyanins for the extract was 1% to 5.6%. As the temperature of soaking increases from 22°C to 38°C the anthocyanins of extract decreases, however further increase in temperature from 38°C upto 60°C increases anthocyanins of extract. The time of soaking the Kokum rind powder in water increases the anthocyanins of extract increases. The effect of temperature (°C) of soaking and time of soaking (h) on anthocyanins of extract at 1:3 (Kokum rind powder: water) for particle size 0.150 mm is represented by equation.

$$A_n = 3.235 - 0.202T - 0.135t - 0.026T^2 + 0.002t^2 + 0.007Tt$$

$$R^2 = 0.986$$

From above equation, it was also observed that the combined effect of temperature of extraction and time of extraction have positive effect on Anthocyanins of the extract. It was increases anthocyanins of extract. Table 14 shows the ANOVA for the viscosity of Kokum rind powder extract. It was observed that effect of soaking temperature and soaking time is highly significant at F = 2.7301.

Table 14: ANOVA for anthocyanins of Kokum rind powder in water (1:3) at particle size 0.150 mm at different temperature

Responses	Degree of Freedom	Sum of Squares	Mean Square	Significance *F
Regression	3	9.2237	3.0746	
Residual	12	13.5139	1.1261	2.7301
Total	15	22.7377		

*$p \leq 0.05$.

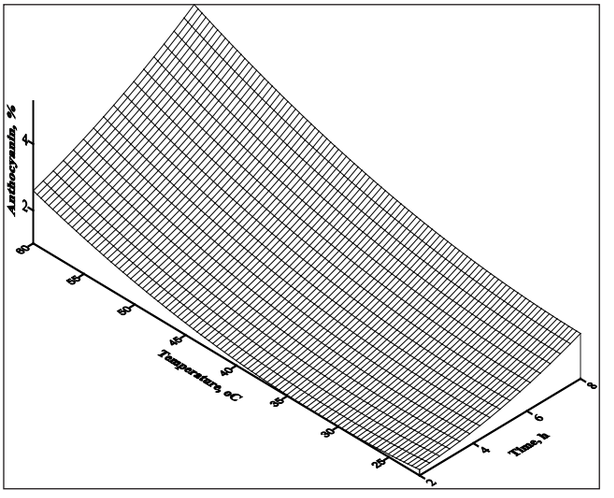

Fig. 14: Surface plot of effect of temperature of soaking and time of soaking of Kokum rind powder in water on Anthocyanins at 1:3 level of 0.150 mm particle size

5. Superimposed contour plots for the extracts

Desirable qualities of Kokum rind powder extract are maximum TSS, Acidity, Viscosity, Anthocyanins. Fig 15 shows the superimposed contour plots for effect of soaking temperature of Kokum rind powder in water on TSS, Acidity, Viscosity, Anthocyanins.

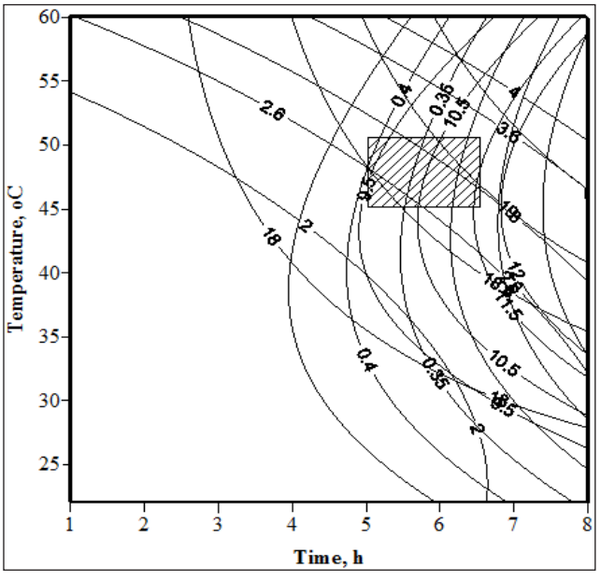

Fig. 15: Superimposed contour plots showing the optimum zone of Temperature of soaking and time of soaking kokum rind powder in water at 1:3 level of 0.150 mm particle size

It was observed that the maximum value of TSS 18.8°B to 19.57°B, acidity 0.37% to 0.42%, Viscosity 9.4 cP to 11 cP and anthocyanins 2.95% to 3.3% was observed in optimum zone where the Soaking temperature 45°C to 50.1°C and Soaking time 5 h to 6.5 h. The optimum zone of maximum TSS, acidity, viscosity, anthocyanins shown in Fig. 15.

Optimization of process conditions

Desirable qualities of Kokum rind powder extract are maximum TSS, Acidity, Viscosity and Anthocyanins. The Kokum rind powder extract were prepared at different Kokum rind powder: water ratios i.e. 1:3 and 1:2 by using particle sizes 0.30 mm, 0.25 mm and 0.15 mm respectively. The optimum zones for the TSS, Acidity, Viscosity and anthocyanins for Kokum rind powder: water i.e. 1:3 at particle size 0.30 mm, 0.25mm, 0.15mm respectively.

The optimum zones for TSS, Acidity, Viscosity, Anthocyanins for Kokum rind powder: water at 1:2 at particle size 0.30 mm, 0.25 mm, 0.15 mm respectively. The optimum values were taken and presented in Table 15, it can be seen that the maximum value of TSS, Acidity, Viscosity, anthocyanins was observed at Kokum rind powder: water ratio (1:2) at particle size 0.30 mm which is observed at 50°C extraction temperature and 6.5h. of extraction time. The optimum values for TSS 19°B, Acidity 0.47%, Viscosity 14.2 cP and anthocyanins 7% observed at the above mentioned process condition.

The Particle Size, Temperature of soaking and soaking Time have significant effect on TSS, Acidity, Viscosity, anthocyanins of Kokum rind powder extract. The optimum zone of Temperature of soaking and time of soaking of Kokum rind extract for particle size

Table 15: Optimum zone values for different particle size of Kokum rind powder soaked with water at different Soaking temperature

	Kokum rind powder: water (1:3)			**Kokum rind powder: water (1:2)**		
	0.300 mm	**0.250 mm**	**0.150 mm**	**0.300 mm**	**0.250 mm**	**0.150 mm**
Optimum Temp., °C	48	53	50.1	50	53	50
Optimum time, h	7.1	6.8	6.5	6.5	6.5	6.8
TSS	13.8	15	19.57	**19**	19.48	19
Acidity	0.48	0.59	0.42	**0.47**	0.44	0.38
Viscosity	9.62	12	11	**14.2**	10.2	10.6
Anthocyanins	2.95	3.25	3.3	**7**	6.75	6.75

0.300mm at 1:3 (Kokum rind powder: water) level, occurred at Temperature of soaking 35 (°C) to 48 (ºC) and Time of soaking, 5.2 (h) to 6.8 (h). At this optimum zone the TSS is 12.75 °B to 15 °B; Acidity is 0.41 % to 0.48 %; Viscosity is 10 cP to 12 cP and Anthocyanins are 1.8 % to 2.95 %. For particle size 0.250mm at 1:3 level, TSS 13°B to 13.8°B, acidity 0.51% to 0.59 %, Viscosity 5.81 cP to 9.62 cP, anthocyanins 2.6% to 3.25% was observed in optimum zone where the temperature of soaking 46°C to 53°C and time of soaking 5.6 h to 7.1 h. The maximum value of TSS 18.8°B to 19.57°B, acidity 0.37% to 0.42%, Viscosity 9.4 cP to 11 cP and anthocyanins 2.95% to 3.3% was observed in optimum zone where the Soaking temperature 45°C to 50.1°C and Soaking time 5 h to 6.5 h for particle size 0.150 mm at 1:3 level.

CONCLUSION

Following conclusions were drawn from the present investigation are as follows:

1. Effect of soaking time and soaking temperature was observed to be maximum for Kokum rind powder: water at 1:2 for particle size 0.300 mm.
2. Optimum Soaking time 5 h to 6.5 h and Soaking temperature was 45°C to 50.1°C. It was observed that the maximum value of TSS 18.8°B to 19.57°B, acidity 0.37% to 0.42%, Viscosity 9.4 cP to 11 cP, anthocyanins 2.95% to 3.3% was observed in optimum zone.

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