



# Development and characterization of Herbal Smoothie

Mohd Shoaib Khan, Rupanjali Dubey, Anjali Gulati, Shubham Kumar, Sonia Alam, Pragya Pandey\*  
Lovely professional university, Punjab- 144411

## ABSTRACT

A smoothie is a beverage of thicker consistency prepared by blending fresh fruits, vegetables, and dairy products usually. In this experiment, an attempt was made to develop herbal smoothie by using carrot, cucumber, strawberry and *Shankpushpi* (herb) with different compositions including 0g, 0.75g, 1g, 1.25g in the final product and optimize on the basis of sensory analysis. The sample with 1.25g concentration was finalized on the basis of equality found on the various parameters i.e. color, odour, taste and mouthfeel as 22% in both color and taste and 28% in mouthfeel and odor respectively. The raw material and final product was analysed for its proximate composition and other parameters such as acidity, pH and TSS (total soluble solids). The results showed that the moisture content and ash content of the carrot, cucumber, strawberry and *Shankpushpi* herb were calculated as 89.95%, 96.45%, 93.17%, 4.73% respectively were 3.33%, 6.3%, 1.65%, 13.1% respectively. The Titrable acidity of carrot, cucumber, strawberry, control and sample with 1.25g *Shankpushpi* herb were 0.12%, 0.12%, 0.17%, 0.17%, 0.213% respectively. The pH of carrot, cucumber, strawberry, control and sample with 1.25g *Shankpushpi* herb were 5.69, 5.05, 3.40, 3.83, and 3.92 respectively. The TSS of carrot, cucumber, strawberry, control and sample with 1.25g *Shankpushpi* herb were 4°brix, 2°brix, 6°brix, 5°brix, 18°brix respectively. Storage of control and sample was done at room temperature as well as at 3°- 5°C.

## 1. INTRODUCTION

A smoothie is a beverage consisting of thicker consistency made from raw fruits, vegetables, and dairy products like milk, yoghurt and ice cream can also be used, in which all the materials are blended together. Smoothies can also be made using other ingredients such as water, ice, fruit juice, whey powder (for people who need extra protein content), plant milk, nuts, seeds, chocolates, herbs, or nutritional supplements. Smoothies made from raw fruits and vegetables consist of more dietary fiber and so they are of thicker consistency than fruit juices. (Derbyshire E, 2017) stated that smoothies (blended beverages that typically contain multiple ingredients) are popular dietary products with the potential to assist individuals in incorporating more fruit and vegetables into their diets.

Smoothies age back to the year 1940 when it was invented by Mabel Stegner on June 23, 1940. He was working in Waring Corporation when he published a recipe book called 'Recipes to make your Waring – Go-Round' and it described 12 milk smoothies. Those with blenders at that time were the ones who made smoothies. In the late 1960's, blenders were popular in the US. Smoothies became even more popular in the 1990's and 2000's by being sold at coffee shops.

Health smoothies were first invented by Steve Kuhnau who was experimenting on his health and it worked for him. It enabled him to open a health food store named "Smoothie King", where he was selling smoothie drinks and later it has grown to a chain of other stores. More than 30 years the founder of Smoothie King has expanded to Seoul, Korea and all over US. Smoothies originate back in the 1920s and were invented by Julius Freed who had stomach health problems. He made this drink and shared it with friends who named it "Orange Julius".

O'Connor L et al. (2016) is talking about intake of smoothies, then during the early years amongst those up to the age of 4 years, intakes of smoothies, fruit juices and purees daily are less than 96 ml.

(Pinket AS et al, 2015) reviewed that smoothie data is extracted and analysed separately, intakes become lower—37 ml daily amongst 3½ to 5½ years old. Unfortunately, smoothie intake data amongst UK adults is not reported in Years 5 and 6 of the UK National Diet and Nutrition Survey (NDNS). Fruit juice intake data, however, is included for which intakes are 46 ml per day amongst UK adults aged 19 to 64 years, declining to an average of 34 ml per day amongst those aged 65 years and over.

#### Variety of Smoothies

There are many different types of smoothies which can be easily made in home with a few ingredients. Different types of smoothies are:

Herbal smoothies—herbal smoothies are the ones made with the combination of herbs and using fruits as base or use of milk, and blending them all together to provide extra nutritional value. Herbs like shankhpushpi, parsley, sage, aloe vera, basil etc, can be used. Addition of herbs in smoothie is a great way to enhance its nutritive value as more antioxidants are added to it.

Green smoothies (Vegetable –Based smoothies)- green smoothie are the one consisting of kales, cauliflowers, cabbage, spinach, carrots, broccoli in base whole milk or any nut milk. They are rich in minerals and vitamins. Incorporating antioxidants, they help in maintaining weight and blood glucose level of body.

Energy-Boosting smoothies - These smoothies contain extra energy benefits in them. Eg. oat meals have carbohydrates, nuts and seeds like almonds, pistachios, flax seeds, chia seeds, melon seeds, that assist you in enhancing your energy levels.

Tropical smoothies- Tropical fruit juices from mangoes, pineapple, coconut water, watermelon juice, sweet lychee are the favourable ones for a tropical smoothie. Dairy products are added to create a creamy texture.

Protein Packed smoothie - These are low fat drinks that assist in weight loss. Low sugar fruits, milk, whey protein, soy milk, soy powder and vegetables are recommendable to provide sustainable energy.

Fruit smoothie- Fruits are mixed together with a combination of milk or yogurt in a blender.

Liquid base - Liquids like yogurt, milk, almond milk, coconut water, water are a good choice to enhance the taste and flavour of a smoothie.

#### Benefits of smoothie

Chronic diseases are among the leading causes of death globally, and as much as 80% of these deaths are reported to be preventable with proper diet and lifestyle. Although extensive research has demonstrated that the increased consumption of fruits and vegetables offers protective health effects from many diet related non-communicable diseases, populations in both developed and developing nations consistently fall short of the recommended intake of 5 or more servings a day. By consumption of smoothie, improvement was found in waist circumference, waist-to-hip ratio and symptoms of burden linked to diet, small intestine, large intestine, and mineral needs. Despite the lack of statistically significant reductions in blood pressure, the trend toward improvements in waist circumference and waist-to-hip ratio are considered to be useful and informative of health risk. Thus, the results of this study provide preliminary support for the consumption of Green Smoothies as a possible primary prevention effort for chronic conditions.

It may also help to reduce health risks or even reverse the effects of chronic conditions. (Source: Emiko maeda, 2013)

In our capstone work, we have incorporated herb shankpushpi into smoothie and done its quality evaluation and research on its physico-chemical properties. Herbal smoothie made by us is with the combination of herb shankpushpi and using carrot, cucumber and strawberry and blending them all together to provide extra nutritional value. Addition of herbs in smoothie is a great way to enhance its nutritive value as more antioxidants are added to it.

#### Benefits of fruits and vegetables

##### Benefits of carrot

Carrot (*Daucus carota* L.) is the most important crop of Apiaceae family. Carrot is one of the vital root vegetables rich in bioactive compounds like carotenoids and dietary fibres with considerable levels of several other useful components having remarkable health-advancing properties. The colors of the carrot root flesh may be white, yellow, orange, red, purple, or very dark purple.

## 2. MATERIAL AND METHODS

### 3.1. Procurement of raw material

The raw material carrot, cucumber, strawberry free from blemishes was procured from the local market of Phagwara, Punjab (India). Carrot is a seasonal crop which remains available during the month of November to March. Cucumbers are warm season crop; they are difficult to grow during foggy, damp and summer season. Strawberry is a seasonal fruit that grows in October- November and April-May. The Herb (*Shankhpushpi*) was not available in the local market so we procured it from online site. It was a sealed packet. All the vegetables and fruit were used in fresh condition.

### 3.2. Standardization of process for production of herbal smoothie

**Smoothie was prepared according to the flowchart (Fig 1). Control sample was standardized through preliminary trials and total four treatments of herbal smoothie was developed. Their composition is presented in table no. 1.**

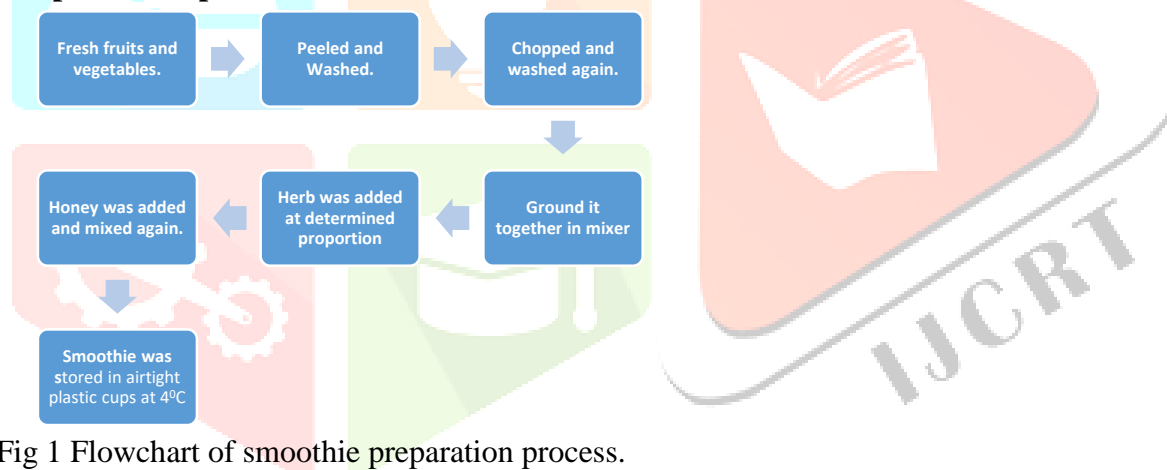


Fig 1 Flowchart of smoothie preparation process.

Table 1 Composition of smoothie

Sample No.	Carrot	Cucumber	Strawberry	Honey	Herbs
1.	30g	30g	40g	15g	0 g
2.	30g	30g	40g	15g	0.5g
3.	30g	30g	40g	15g	1g
4.	30g	30g	40g	15g	1.25g

### 3.3. Nutritional analysis

Determination of moisture, fat, ash and content was done according to standard method, AOAC, 2000. Titrable acidity was determined by Standard Method given by AOAC, 2000.

### 3.3.1. TSS & pH

Total Soluble Solid is done by 'Refractometer'. Refractometer is present in three ranges: 0–30; 30–60; 60–90. To estimate the TSS of sample, few drops of sample is put in the measuring prism and covered with daylight plate and note down the reading ( $^{\circ}$  Brix).

Determination of pH is done by using pH meter. To estimate the pH of any sample, rinse the pH electrode with distilled water and then with buffer to be used for calibration (i.e. pH- 4.01). Then dip the pH electrode into the next buffer of pH-4.01. The pH electrode should be dip in the sample and then the meter display should be locked on the buffer value, when the reading is stable, Press "Enter" to accept.

### 3.4. Sensory analysis

Sensory parameter is of great importance to both the processor and consumer. To the processor, since it attracts consumer; to the consumer since it satisfies his aesthetic and gustatory sense. Sensory quality is a combination of different sense of perception coming into play in choosing and eating a food. Appearance, which can be judged by the eye, e.g. colour, size, shape, uni- formity and absence of defects, is of first importance in food selection. Flavor embraces the sense of taste, smell and feeling. It is generally agreed that the sense of taste is limited to sweet, sour, salty, bitter and umami. Odour, a vastly complex sensation, is the most important factor in flavor.

### 3.5. Chemical analysis

#### 3.5.1. Estimation of Beta-Carotene

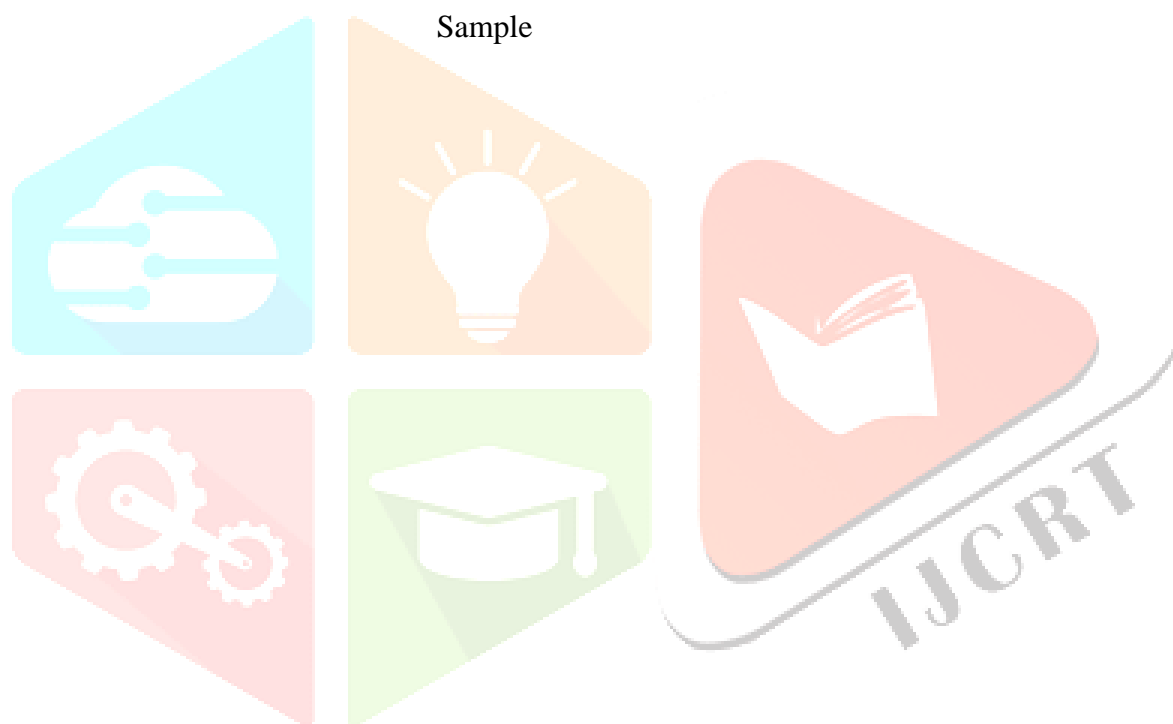
Estimation of  $\beta$ -Carotene is done by a method given by Kemmerer et al, (1943), Acetone, Petroleum Ether, Anhydrous Sodium Sulphate ( $\text{Na}_2\text{SO}_4$ ) is used as reagent. To prepare

Absorbent, mix one part of Supercel. Eluent is prepared by mixing 3% acetone in petroleum

ether. Weigh 25g of sample and dissolve in 25ml of chloroform and make up the volume to 250ml with petroleum ether. Then take 10ml of solution and dilute to 100ml with petroleum ether. Pipette 5, 10, 15, 20, 25 and 30ml of this solution in 100ml of volumetric flask. Add 3ml of acetone and dilute the mask with petroleum ether. The concentration will be 0.5, 1.0, 1.5, 2.0, 2.5, 3.0 µg/ml. Measure the color at 452nm using 3% acetone as blank using spectrophotometer.

**µg of Carotene per 100g =**

$$\frac{\text{Conc. of carotene in solution (standard curve) (}\mu\text{g/ml)} \times \text{Final vol.} \times \text{Dilution} \times 100}{\text{Weight of Sample}}$$





### 3.5.2. Estimation of L ascorbic acid

Estimation of L-ascorbic acid content is done by method given by Johnson, B.C. et al (1948). 3% Metaphosphoric acid (HPO<sub>3</sub>), Ascorbic acid Standard, Dye solution used as a reagent to estimate L-ascorbic acid Standard. Procedure for estimation of L-ascorbic acid is performed by 5ml standard ascorbic acid with addition of HPO<sub>3</sub>. Then, fill the burette with dye and titration is done after that. For semi-solid sample take 10g sample, blend with 3% HPO<sub>3</sub> and makeup the volume 100ml with HPO<sub>3</sub>. Then, filter and centrifuge and titration are performed.

Mg of Ascorbic acid per 100g or ml =

$$\frac{\text{Titre} \times \text{Dye Factor} \times \text{Volume made up} \times 100}{\text{Aliquot of extract taken for estimation} \times \text{Weight or volume of sample taken for estimation}}$$

Aliquot of extract taken for estimation X Weight or volume of sample taken for estimation

### 3.5.3. Determine the total phenolic content

Estimation of Total Phenolic content was done by method of Nabavi et al. Two hundred microlitres of extract were mixed with 2.5ml of 10% folin Cio Calteau's reagent and 2ml of 7.5% Sodium Carbonate. The reaction mixture is incubated at 45° C for 40 minutes and absorbance was measured at 700nm. Phenol is used as Standard. Standard curve is prepared.

$$\text{Total Phenolic content (T) (as gallic acid)} = \frac{C \times V}{M}$$

Where,

T = Total phenolic content in mg

C = Concentration of gallic acid

V = Volume of the extract solution in ml

M = Weight of extract in g

#### **3.5.4. Estimation of flavonoids**

The estimation of flavonoid content is determined by using a colorimeter assay developed by Bao et al., with some modification. Aliquot (0.2ml) of extract was added to 0.3ml of 5%  $\text{NaNO}_2$  and wait for 5 minutes. After that, 0.6ml of 10%  $\text{AlCl}_3$  was added followed by the addition of 2ml of 1M NaOH, then after 6minutes, add 2.1ml of distilled water. Absorbance was taken at 510nm against the reagent blank and flavonoid content was expressed as (mg rutin equivalent)

#### **3.6. Storage condition & shelf life**

For storage studies smoothie was made and packed into glass containers. These containers were previously sterilized by proper washing followed by autoclaving at  $121^\circ\text{C}$ . This process was carried out to remove any bacterial load present on the container. Freshly prepared smoothie was poured into the container leaving a minimal headspace so as to prevent bacterial contamination. These containers were placed under 2 different temperature conditions. The storage study was performed for 6 days as fruits and vegetables is highly perishable component if it is not preserved in sterilized conditions. Proximate analysis, including and microbiological analysis were done after 3 days' interval.

##### **3.6.1. Titrable acidity**

Determination of Titrable acidity is done by Standard Method given by AOAC, 2000. Dilute the aliquot of sample with distilled water and then titrate with 0.1 N NaOH solutions by using few drops of 1% phenolphthalein solution as an indicator and then note down the ml of NaOH used and calculate the result as percent anhydrous citric acid or other acid.

$$\text{Titration acidity(\%)} = \frac{[\text{ml of NaOH used}] \times [0.1 \text{ N NaOH}] \times [\text{Milliequivalent Factor}]}{\text{Gram of Sample}} \times 100$$

### Gram of Sample

#### 3.6.2. pH & TSS

Total Soluble Solid is done by 'Refractometer'. Refractometer is present in three ranges: 0– 30; 30–60; 60–90. To estimate the TSS of sample, few drops of sample is put in the measuring prism and covered with daylight plate and note down the reading (° Brix).

Determination of pH is done by using pH meter. To estimate the pH of any sample, rinse the pH electrode with distilled water and then with buffer to be used for calibration (i.e. pH- 4.01). Then dip the pH electrode into the next buffer of pH-4.01. The pH electrode should be dip in the sample and then the meter display should be locked on the buffer value, when the reading is stable, Press "Enter" to accept



## 4. RESULTS AND DISCUSSION

### 4.1. Proximate analysis of raw material

#### Moisture content

**Carrot-** The moisture content of native and modified carrots samples is shown in table that varies from 90.7-89.95%. The moisture content of the carrots 9.31% whereas with the moisture content of carrot samples are shown in table that varies from 90.7 – 89.95%. The moisture content of the native carrot was 90.7% whereas with the continuation of the test it gradually decreased to 89.95%.

The mean of these samples was calculated to be 89.97% as shown in



Figure 4-7 Carrot sample

**Cucumber-** As shown in (table no. 5) the readings are partially same which varies from 96.44 – 96.6%. The data shows that cucumber has higher moisture content as compared to carrots. Technically, the mean as calculated is 96.45% as shown in table. The reason for this may be due to carrots contain high amount of dietary fibers, increasing its moisture level. Not only are they high in water content, they also contain important nutrients that play a part in hydration like magnesium and potassium.



**Figure 4-8 Fresh cucumber**

**Strawberry-** The table below shows the data of moisture content of strawberry which varies from 93.24 – 93.5%. The numbers below show clearly there is no decrease of moisture content as we continued to test, whereas the mean data shows 93.30%. As strawberry are rich source of vitamin C, and antioxidants, the water trapping ability of strawberry were noted.

**Herbs-** The table below shows the moisture content which varies from 3.6 – 4.73%. The mean as calculated is 4.73%. Herb, which we used was in powdered form where as other mentioned raw materials were used fresh from farm.

These were no moisture loss traces as the herb we used was hermetically sealed which endures its low water gain capacity



Figure 4-9 Moisture analyzed herb sample

Table 4-2 Moisture content of Raw material

Sr. No.	Sample	Moisture content %
1.	Cucumber	96.453%
2.	Carrots	89.97%
3.	Strawberry	93.30%
4.	Herbs	4.73%



## Ash content of raw material

### *Carrot-*

Accurately weigh carrot sample and put in to a previously dried and weighed crucible. The crucible with sample was heated gently on flame for complete charring and then it was heated in a muffle furnace at  $550 \pm 10^\circ\text{C}$  for 4-5 hours, until ash was formed. Then cooled in desiccators and weighed. The percent ash content was calculated. As you can observe in fig. 10 & 11 before and after effect of ashing carrot.



Figure 4-10 Raw carrot sample



Figure 4-11 Ash content of carrot

### *Cucumber-*



Ash is composed of minerals and because they are rudiments, they will always exist in the exact form as they are. The crucible was weighed before keeping it in the muffle furnace at  $550 \pm 10^\circ\text{C}$  for 4-5 hours, until ash was formed. Then the sample was kept in the desiccator for cooling and then the weight of taken and the ash content was calculated.

#### ***Strawberry-***

The amount of ash present in the strawberry flour ranged from 1.47%-2%. We can observe that the strawberry has less ash content than the cucumber; this may have been due to the quantities of seeds present in the residues. The same procedure was followed to calculate the mineral content.

#### ***Herb-***

Content of mineral significantly depends on macro and microelements. In the case of herbs, the degree of increase of minerals in leaves which are eaten directly or subjected to drying methods is of significant meaning. The analyzed herbal plants differ in macronutrient content due to differences in species. The ash content in herb was in range of 13% - 13.5%. Similar procedure was followed to analyze the ash content.

Table 4-3Ash content of Raw material

Sr. no.	Sample	Ash content%
1.	Carrot	3.33
2.	Cucumber	6.3
3.	Strawberry	1.65
4.	Herb	13.1

## **Titration acidity of raw material**

### ***Carrot-***

As per our testing results we have three samples of carrot. The sample 1 shows reading, 0.12% of titration acidity. Sample 2 shows value of 0.11% and sample 3 shows value of 0.12%. Titration acidity increased with days of storage. Total titration acidity of the samples increased with days of storage, with the sample stored at ambient temperature recording higher range of total titration acidity. Increase in titration acidity of the carrot juice may possibly be due to reactions between the carrot juice components and chemicals at low temperature.

### ***Cucumber-***

We tested titration acidity for cucumber also three samples, sample 1 shows reading of 0.12%, sample 2 showed value of 0.12% and 0.12% for sample 3 also. Decline in Titration Acidity is an important outcome during ripening, as it renders the fruit less acidic and sour. Since organic acids, such as malic or citric acid, are primary substrates for respiration, a reduction in acidity is expected in respiring fruits. As compared to the carrot results, cucumber showed no difference in the results, as it may be due to the factor that cucumber was fresh and equally ripened.

### ***Strawberry-***

The results of strawberry came out to be, sample 1 showed value of 0.28%, sample 2 showed value of 0.12% and sample 3 results was 0.12%. The titration acidity (TA) of strawberry was significantly affected by harvesting the fruits at different stages of maturity. The acidity in fruit is an important factor in determining fruit maturity and quality. The acidity of strawberry fruit, generally, increases to upmost in mature fruit before declining more quickly in the later stages of ripening. The drop in acidity is attributed to increasing consumption of organic acids in the

process of respiration consequential decreased acidity as the fruit advances from one stage of development to another.



Figure 4-12 acidity analysis

This study was conducted to increase the nutritional value of smoothie by incorporating *Shankhpushi* herb. With increasing health awareness people are becoming more inclined towards functional food products. Therefore an attempts has been made to optimize the ratio of herbs which respect to their sensory profile. Further, the product was analyzed for its proximate value, physico-chemical value, sensory attributes and storage qualities of the same. The Fig 2 and 3 pictorial representation of the samples.



Fig 2 smoothie samples



**Fig 3 control and herbal (1.25g) smoothie**

#### 4.2. Sensory analysis

The results of sensory analysis are represented in fig 3. The sensory analysis revealed that sample 1 showed 36% taste and 31% of mouth feel and the sensory results were acceptable but not very much liked. The liking percentage increased to 42% in sample 2 and taste was acceptable. It was observed that sample 2 has less than half percentage of color and appearance which is 11% and it contains 0.5g of herbs. Sample 4 showed highly acceptable percentage for all sensory characteristics. Therefore sample 4 was selected for further analysis. In sample 4, 1.25g of herb was added which was interestingly acceptable and encouraged profoundly. It has been noticed that all the criteria were purely taken into consideration and sample 4 was the product which as compared to other three samples was highly accepted. So we can say that sample 4 was our perfect product which, as the data says, has 28% of colour and appearances, 28% of odour, 22% of taste and lastly, 22% of mouth feel, which is 100% acceptable.



Fig 4 Sensory Profile of developed Smoothie

#### 4.3 Physicochemical properties of developed herbal smoothie-

Titration acidity of controlled as results showed were, sample 1 was 0.128%, sample 2 was 0.142% and sample 3 was 0.192% controlled sample just contain carrot, strawberry and cucumber. Sample titration acidity showed results like, sample 1 showed 0.214%, sample 2 showed 0.192% and sample 3 showed 0.230% sample contain carrot, strawberry and cucumber plus herb too.

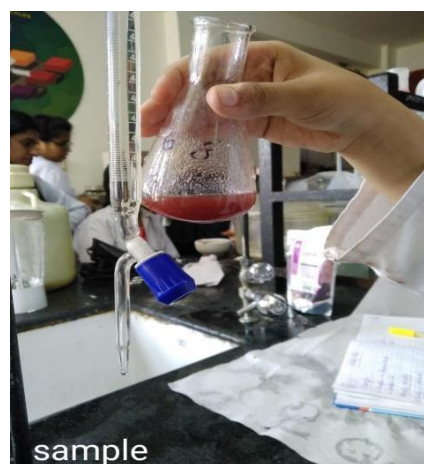
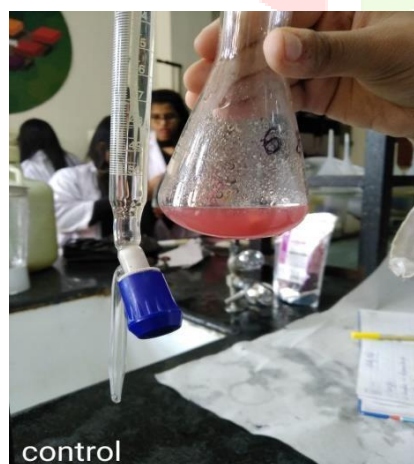


Figure 4-13 Acidity analysis of control

Figure 4-14 acidity of herbal smoothie

**Table 4-4 Acidity content of Raw and Herbal Smoothie**

Sr. no.	Sample	Titration acidity
1.	Carrot	0.12
2.	Cucumber	0.12
3.	Strawberry	0.17
4.	Controlled	0.17
5.	Sample	0.213

#### pH & TSS

In pH as we calculate with the help of pH meter, the results we got are, for carrot pH is 5.69, cucumber is 5.05, strawberry is 3.40, for control pH is 3.83 and for sample is 3.92. The relatively high pH of carrot juice (pH ~ 6) makes it unique among popular commercial juices, such as orange or apple juices, which have pH values below 4.5. The low-acid nature of carrot juice makes it more susceptible to spoilage and pathogenic organisms, which can be countered by acidification. This issue is of particular interest in fresh carrot juice products that require refrigeration. In addition, carrot juice can be subjected to lower pH conditions when it is mixed with fruit juices in juice blend products. For strawberry the right pH media needed to provide the best conditions for most agricultural plants to absorb the nutrients efficiently. For hydroponic cucumbers (5.5–6.0) and nitrification (7.5–9.0) requires reconciliation to improve systems integration. Controlled showed optimum pH value as it was the mixture of all the ingredients. The sample pH showed higher value due to presence of herb.

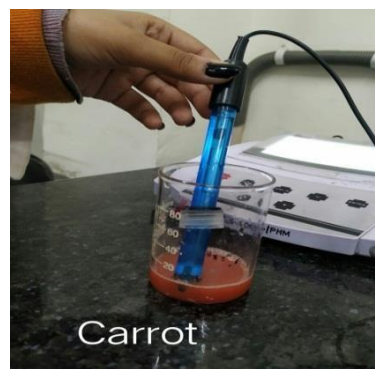
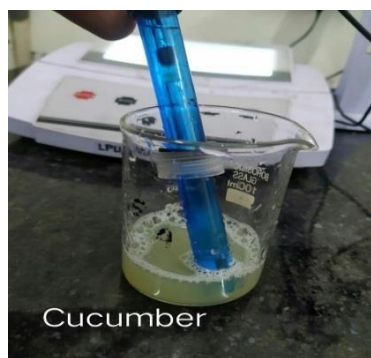


Figure 4-15 pH analysis of Cucumber, carrot and strawberry

Table 4-5 pH analysis of control and herbal smoothie

Sr. no.	Sample	Ph
1.	Carrot	5.69
2.	Cucumber	5.05
3.	Strawberry	3.40
4.	Control	3.83
5.	Sample	3.92

For Total Soluble Solids, we used refractometer with 0-30° Brix range. °Brix is the unit of refractometer. For carrots the results came as 1°Bx. For cucumber the results came as 2°Bx, for strawberry it showed 6°Bx and for both controlled and sample the results showed 5°Bx and 18°Bx respectively. As carrot and cucumber both contain abundant water content the TSS came out to be low, whereas for sample the TSS came out to be high because it is the mixture of all the fruits and vegetable we used and herb too. The samples were freshly collected from the farm and all the proximate analysis were done on the same day itself.



Table 4-6 TSS of control and herbal smoothie

Sr. no.	Sample	TSS (°Brix)
1.	Carrot	4
2.	Cucumber	2
3.	Strawberry	6
4.	Control	5
5.	Sample	18

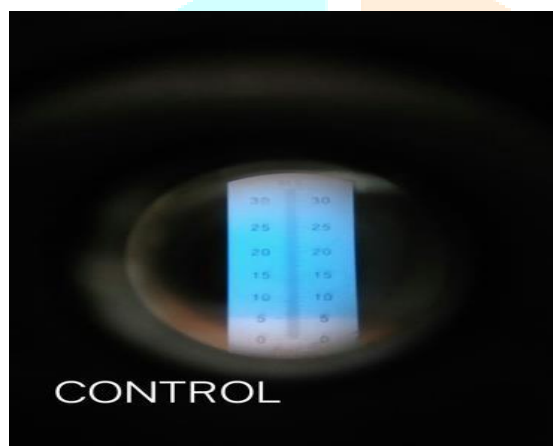


Figure 4-16 TSS of herbal smoothie

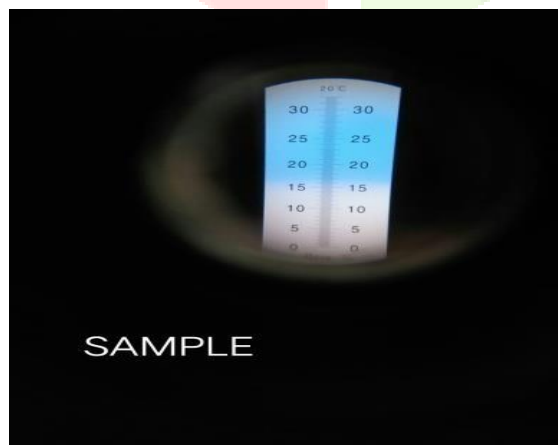


Figure 4-17 TSS of Control sample



#### 4.2.1. Fat estimation

##### *Sample-*

As we performed fat estimation test, the samples showed results, sample 1 showed estimated value of 0.336 g/100g, sample 2 showed value of 0.343 g/100g and sample 0.354 g/100g. Tests conducted by D. Saranyambiga, Dr. Rita Narayanan and Dr. V.S. Vadivoo the values of fat estimation were, 0.30 g/100g for control and 0.34 g/100g for sample. Other lipids, although present in relatively small quantities, play crucial roles as enzyme cofactors, electron carriers, light-absorbing pigments, hydrophobic anchors for proteins, Chaperones to aid membrane protein folding, emulsifying agents in the digestive tract, hormones, and intracellular messengers Nelson and Cox, 2004.

##### *Controlled-*

Similarly, performed fat estimation on our controlled sample, the results were, sample 1 showed value of 0.183 g/100g, sample 2, 0.173 g/100g and sample 3 showed values of 0.179 g/100g. Fahy et al. (2005- 2009) developed a classification system for lipids as follows: lipids are hydrophobic or amphipathic small molecules that may originate entirely or in part by carb-anion-based condensations of thioesters and/or isoprene units. Tests done by Arcos, J.A., García the value came to be 0.7 g/100g.

Table 4-7 Fat content of control and herbal sample

Sr. no.	Samples	Fat content (g/100g)
1.	Sample (with herb)	0.344
2.	Controlled (without herb)	0.178

### 4.3. Chemical analysis

#### 4.3.1. Estimation of Beta-carotene

##### *Sample-*

As we conducted test of  $\beta$ -carotene on our sample, it showed the value ranging from 370.821-369.366  $\mu\text{g}/100\text{g}$ . Similarly, as others we took three different samples, so sample 2 showed value of 369.366  $\mu\text{g}/100\text{g}$ . But as the data tells, the sample 2 and sample 3 both showed the same value. According to the study performed by Magdalena Buniowska and Eva Arrigoni they conducted test of  $\beta$ -carotene under heat treatment, a range of three identified carotenoids included  $\beta$ -cryptoxanthin (0.10–0.21 mg/100 g),  $\alpha$ -carotene (1.66–1.98 mg/100 g) and  $\beta$ -carotene (2.24–2.74 mg/100 g) for spinach and carrot smoothie. As their studies uses heat treatment the values may likely put a difference. The high temperature promotes the isomerization of double bonds, which results in brightening of the resulting colour in the fruits and vegetables.

##### *Controlled-*

Our controlled sample, in which we did not add any herb, the results were as, sample 1 showed the value of 365.251  $\mu\text{g}/100\text{g}$ . As we took other readings, the results of sample 2 and sample 3 came as, 366.380  $\mu\text{g}/100\text{g}$  and 365.183  $\mu\text{g}/100\text{g}$  respectively. Carotenoids in which molecules do not show the presence of the  $\beta$ -ion ring do not exhibit pro-vitamin activity. According to the study performed by Kok Wei Tan<sup>1</sup>, Brigitte A. Graf contained on average 21.02 mg  $\beta$ -carotene ( $\pm 4.38$  mg, ranging from 16.07 mg to 26.32 mg) in their avocado smoothie. The materials used by us showed only minimum amount of  $\beta$ -carotene, which we calculated in micrograms.

Table 4-8 Beta-carotene of control and herbal sample

Sr. no.	Samples	$\beta$ -carotene ( $\mu\text{g}/100\text{g}$ )
1.	Sample ( with herb)	369.836

2.	Controlled ( without herb)	365.61
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#### 4.3.2. Estimation of L-ascorbic

##### *Sample-*

As of our sample we performed L-ascorbic test, the results were, sample showed value of 5.836 mg/100g. Similarly, we took two more readings, sample 1 showed the value of 5.812 mg/100g and sample 3 showed the value of 5.861 mg/100g. This research work shows that carrots and cucumbers contain an appreciable amount of antioxidant properties, vitamins, minerals and macronutrients which are required for the proper functioning of the body. Tests performed by Taiwo Ayodele Aderinola and Kemi Elizabeth Abaire shows value of  $14.48 \pm 0.32$  mg/100 mg ,  $21.07 \pm 0.26$  mg/100 mg and  $24.48 \pm 1.11$  mg/100 mg respectively basically of smoothie made up of lemon, kale and celery and parsley herb. With results it has been clearly shown that the ascorbic acid present in lemon is more than carrot and cucumber.

##### *Controlled-*

In controlled as we did not add any herb, the results were, sample 15.946 mg/100g, as of sample 2 the results were, 5.796 mg/100g and sample 3 the results estimated to be 5.782 mg/100g. Experiments done by Canan Ece TAMER and Fatma Zehra YEKELER the results were 1.10708 mg/100g and 0.53526 mg/100g as they used orange extract, milk, and ascorbic acid concentrate. Ascorbic acid determination results obtained by cyclic voltammetry were compared with those obtained by the volumetric method with dichlorophenol indophenol. The addition of herbal extracts allows the production of cold beverages of high nutritional value, which are preferred by consumers for their sensory properties. It also helps to serve these products for consumption across all seasons.

Table 4-9 L-ascorbic acid of control and herbal smoothie

Sr. no.	Samples	L-ascorbic acid (mg/100g)
1.	Sample (with herb)	5.83
2.	Controlled (without herb)	5.842

#### 4.3.3. Total phenolic content

##### Sample-

Smoothie was also tested for total phenolic content and the results were calculated to be, sample 1 shows reading of 1278.3 mg/L, sample 2 showed the result, 1285.76 mg/L and sample 3, 1285.78 mg/L. according to the study performed by Sushant Aryal, Manoj Kumar Baniya the estimated value for their vegetable smoothie, the results were,  $292.65 \pm 0.42$  mg/L and  $287.73 \pm 0.16$  mg/L. As fruits have higher antioxidants, the results vary. Phenolic compounds are important plant constituents with redox properties responsible for antioxidant activity.

##### Controlled-

The results for controlled sample was, sample 1 showed value 1269.81 mg/L. sample 2 showed value estimated to be, 1268.33 mg/L and lastly, sample 3 showed value of 1279.33 mg/L. More research done by Keskin-Šašić, Tahirović, their result for total phenolic content of fruit smoothie was estimated to be, 1086.60 mg/L. The reason behind this decrease in total phenolic content was, it was non-centrifuged, and proteins residing in solutions of non-centrifuged samples increased the antioxidant capacity of those fruits.

Table 4-10 Total Phenol content of control and herbal smoothie

Sr. no.	Sample	Total Phenolic content (mg/L)
1.	Sample (with herb)	1283.7
2.	Controlled (without herb)	1272.49



**Figure 4-18 TPC estimation of sample**

#### **4.3.4. Estimation of flavonoids**

##### **Carrot-**

For flavonoids, sample 1 estimated value ranges from 3.7-4.4 (mg/g). Sample 2 showed value of 3.1 mg/g and sample 3 showed the value of 4.4 mg/g. Carrots are also a good source of disease-fighting flavonoids that provide antioxidants that neutralize free radicals in our bodies. The value ranges from 3.7 mg/g but it can be seen that it increases to 4.4 mg/g. As carrots were grinded for smoothie preparation, the minor losses on flavonoids can affect the values.

##### **Cucumber-**

The flavonoids tests we conducted in *Cucumis sativus*, the results were, sample 1 showed value of 2.14 mg/g, sample 2 values came to be 2.73 mg/g and sample 3 values was 1.56 mg/g. As cucumber constitutes more amounts of carotenoids, flavonoids and phenolic content, these parameters may differ due to the species also. According to Saxena, et al., flavonoids have been reported to exert multiple biological property including antimicrobial, antioxidant, cytotoxicity, anti-inflammatory, as well as antitumor activity.

##### **Strawberry-**

As compared to carrots and cucumber, strawberries contain high amount of flavonoids content.

Sample 1 showed value of 7.04 mg/g, sample 2 showed value of 7.54 mg/g and sample 3 have

decreasing value of 6.53 mg/g. Flavonoids in strawberry include the flavonols quercetin and kaempferol Wang and Zheng, 2001. The flavonoids content value may affect by the grinding. Therefore, if found to be of value in improving antioxidant capacity or the balance of specific phenolic compounds, accessions from these progenitor species could be readily incorporated into a strawberry breeding program.

#### Herb-

As the results shows, sample 1 value estimated to be, 0.78 mg/g, sample 2 values was 0.74 mg/g and sample 3 showed values of 0.80 mg/g. The values do not differ in this case because the herbs are in dried form, so no extra degradation can take place. The flavonoids content in herb is less as compared to other fruits and vegetables, because many of the reason they are dehydrated and most of these phenols get degraded, only some part of it is left.

Table 4-11 Total flavonoid content of raw material

Sr. no.	Samples	Flavonoids (mg/g)
1.	Carrot	3.73
2.	Cucumber	2.14
3.	Strawberry	7.04
4.	Herb	0.77

#### 4.4. Storage & Shelf life

For storage studies smoothie was made and packed into glass containers. These containers were previously sterilized by proper washing followed by autoclaving at 121°C. This process was carried out to remove any bacterial load present on the container. Freshly prepared smoothie was poured into the container leaving a minimal headspace so as to prevent bacterial contamination.

These containers were placed under 2 different temperature conditions. The storage study was performed for 6 days as fruits and vegetables is highly perishable component if it is not preserved in sterilized conditions. Proximate analysis, including and microbiological analysis were done after 3 days' interval.

4 samples-

2 sample: Room temperature (control & sample) as shown in fig. 21.

2 sample:  $3^{\circ} - 5^{\circ}\text{C}$  (control & sample) as shown in fig. 22.



Figure 4-19 Stored sample at room temperature



Figure 4-20 Stored sample at refrigeration temp

Titration acidity-

We have observed the Titrable acidity of control before keeping it for the shelf life the titrable acidity of our control sample is 0.17% but we prepared the fresh sample again before keeping it for the shelf to observe the acidity before & after shelf life. Therefore, before keeping it for the shelf life the titrable acidity was about 0.302% but after the shelf life period of 5 days the acidity of the control increased due to the spoilage of the smoothie.

We have observed the Titrable acidity of control before keeping it for the shelf life the titrable acidity of our sample is 0.213% but we prepared the fresh sample again before keeping it for the shelf to observe the acidity before & after shelf life. Therefore, before keeping it for the shelf life the titrable acidity was about 0.379% but after the shelf life period of 5 days the acidity of the control increased due to the spoilage of the smoothie.

pH-

We have observed the pH of control before keeping it for the shelf life the pH of our control sample is 3.83 but we prepared the fresh sample again before keeping it for the shelf to observe the pH before & after shelf life. Therefore, before keeping it for the shelf life the pH was about 3.83 but after the shelf life period of 5 days the pH of the control decreased due to the spoilage of the smoothie. The pH after the shelf life was 2.1.

S.No.	Before storage	After Storage
1	3.83	2.1

We have observed the pH of control before keeping it for the shelf life the pH of our control sample is 3.83 but we prepared the fresh sample again before keeping it for the shelf to observe the pH before & after shelf life. Therefore, before keeping it for the shelf life the pH was about 3.92 but after the shelf life period of 5 days the pH of the control decreased due to the spoilage of the smoothie. The pH after the shelf life was 1.84 this is because of herbs present in it.



S. No.	Before storage	After Storage
1	3.92	1.84

Total soluble solid-

We have observed the TSS of control before keeping it for the shelf life the TSS of our control sample is 5<sup>0</sup> Brix but we prepared the fresh sample again before keeping if for the shelf to observe the pH before & after shelf life. Therefore, before keeping it for the shelf life the TSS was about 5<sup>0</sup> Brix but after the shelf life period of 5 days the TSS of the control increased due to the spoilage of the smoothie. The brix after the shelf life was 10<sup>0</sup> Brix. As you can observe the after the spoilage the increment of the brix was about 5<sup>0</sup>.

S. No.	Before storage	After storage
1	5 <sup>0</sup> Brix	10 <sup>0</sup> Brix

We have observed the TSS of control before keeping it for the shelf life the TSS of our control sample is 18<sup>0</sup> Brix the brix is high in the sample due to herbs present in it, as we prepared the fresh sample again before keeping if for the shelf to observe the pH before & after shelf life. Therefore, before keeping it for the shelf life the TSS was about 15<sup>0</sup> Brix but after the shelf life period on 5 days the TSS of the control increased due to the spoilage of the smoothie. The brix after the shelf life was 25<sup>0</sup> Brix. As you can observe the after the spoilage the increment of the brix was about 10<sup>0</sup>.

S. No	Before storage	After storage
1	15 <sup>0</sup> Brix	25 <sup>0</sup> Brix

### 3. CONCLUSION

Smoothies are popular dietary products with the potential to assist individuals in incorporating more fruit and vegetables into their diets. As the title suggests we developed a herbal smoothie with the herb Shankpushpi with different compositions including 0g, 0.75g, 1g, 1.25g in the final product and optimize on the basis of sensory analysis. The control was in the ratio of 30:30:40 in which 30g carrot, 30g cucumber and 40g of strawberry are added. The sample with 1.25g concentration of herb was finalized on the basis of equality found on the various parameters. With the addition of the herb Shankpushpi, it served enormous benefits including the improvement in the function of nervous system, boosting memory, helps in the treatment of disorders/syndromes such as hypertension, hypotension, anxiety neurosis, stresses etc. whereas with the addition of carrot, cucumber and strawberry in the smoothie, the anti-oxidative, antacid, anti-inflammatory and anti-carcinogenic properties increased significantly. Carrot being a good source of beta carotene, fiber, vitamin K1, potassium, and antioxidants, helps in lowering cholesterol levels and improved eye health. Cucumber being rich in vitamin C, vitamin K, magnesium helps in promoting hydration, prevents ulcerative colitis, and lowers blood sugar level. While strawberry being a sodium-free, fat-free, cholesterol-free, low-calorie food, is rich in vitamin C, fiber, manganese, potassium, and antioxidant (polyphenols), which helps in increasing HDL (good) cholesterol, lowering your blood pressure, and guard against cancer. So with the introduction of herbal smoothie into the regular diet, it may have a greater impact on the health quality of life for individuals.

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