



Comparative Analysis of Phytochemicals and Antioxidant Potential of Ethanol Leaf Extracts of *Psidium guajava* and *Syzygium jambos*

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Abstract

Background: Plant-based drugs for various human ailments are becoming very important in the current domain of therapeutics. **Aim:** *Psidium guajava* and *Syzygium jambos* are two such plant species known for their medicinal properties in traditional systems of medicine like Ayurveda. **Methods:** Phytochemical analysis including GCMS, and antioxidant studies (DPPH) was carried out for both plant extracts. **Results:** Comparative phytochemical analyses of ethanol extracts of both these plants have shown the existence of bioactive components like tannins, polyphenols, alkaloids, flavonoids and terpenoids. These phytochemicals were quantified and the ethanol extracts were subjected to GCMS analysis which showed the presence of cis- β -farnesene, cis-calamenene, copaene, humulene, caryophyllene, phytol, neophytadiene, n-hexadecanoic acid etc, many of which possess diverse properties like antimicrobial, antibiofilm, antioxidant and anti-inflammatory. DPPH and reducing power assays revealed the excellent radical scavenging activity of the extracts. **Conclusion:** Among the two plants under the current study, *S. jambos* extract showed better results when compared to *P. guajava* concerning the antioxidant potential and the quantity of flavonoids, alkaloids, polyphenols and tannins present in the plant samples.

Keywords: Antioxidants, DPPH, GC-MS, *Psidium guajava*, Phytochemicals, *Syzygium jambos*

1. Introduction

Plants have always been considered a crucial part of curing several ailments because of the wide variety of phytochemicals present in them. There are so many plants that are continuously being explored for their benefits to mankind and among those plants, *P. guajava* is one. *P. guajava* is commonly called guava and is one of the most popular fruits seen in tropical and sub-tropical areas including India. It is a member of the family *Myrtaceae*, class *Magnoliopsida*, and phylum *Magnoliophyta* and is loaded with numerous beneficial components¹. Traditionally, the plant's fruits and leaves have been used to treat conditions like gastric trouble, flatulence, diarrhoea, wounds and ulcers. Since it is very economical, it is also called the poor man's apple in tropical areas². Although guava does not belong to the

citrus family of fruits, it is loaded with more vitamin C (80mg in 100g of fruit) than citrus fruits³. Thus, reflecting the antioxidant properties of the plant. Apart from the good taste, the fruit also helps in good bowel movement because of the required percentage of dietary fibre present in them and hence helps in preventing constipation. In Japan, guava leaves tea is used due to the phenolic components present in them and is believed to regulate blood sugar levels⁴. The practice of using the extract of guava leaves to cure asthma attacks, bronchitis and dysentery is prevalent in many parts of the world even today. There is also evidence showing the susceptibility of clinical isolates like *Pseudomonas aeruginosa*, *Staphylococcus* spp. and *Proteus* spp. to *P. guajava*⁵. The magnitude of bioactivity of different parts of *P. guajava* is tremendous and therefore the applications can be unlimited.

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Another plant from the family of *Myrtaceae* is *S. jambos* which is comparatively less explored. It is a plant extensively seen in Central America and South Asian countries like Nepal, the Philippines, Indonesia, and Malaysia. However, the Indian sub-continent has adopted this plant and has multiple vernacular names by which it is called such as *Panineer champa*, rose apple, Malabar plum, water apple etc⁶. The common name of *S. jambos* in Brazil is *jambolão* and there is a practice of using the leaf infusion to treat diabetes⁷. This plant has been utilized, like guava, by traditional doctors in Kerala, India to treat conditions like ulcers, wounds, and dermatopathy⁸. Some studies report the various health benefits of *S. jambos* such as antiviral, antimicrobial, anti-inflammatory, anti-cancer, antidermatophytic, analgesic and hepatoprotective activities⁹. The present study has been focused on examining the various phytochemicals found in the ethanol-based leaf extracts of *P. guajava* and *S. jambos* and the extent to which a few of the main classes of phytochemicals are present. The particular compounds found in the extract were also revealed using GC-MS analysis. To highlight their antioxidant properties, both the reducing power assay and the DPPH assay were carried out.

2. Materials and Methods

2.1 Preparing Plant Samples

For the current study, leaves of *P. guajava* and *S. jambos* (*Panineer champa*) were collected from Thrissur district in Kerala, India and were dried in the shade for fourteen days. Further, they were ground into a fine powder and stored for later use in a zip-lock bag.

2.2 Extraction

5g of each plant powder was taken and extraction was done using a Soxhlet apparatus using 150ml ethanol as the solvent. Extraction was carried out for six hours subjected to 40% heat. This extract was used for further analysis.

2.3 Qualitative Analysis of Phytochemicals

Various tests were done to screen the extract for the presence of specific phytochemicals. The results were tabulated once the tests were performed as per standard procedures¹⁰.

2.4 Quantitative Examination of Phytochemicals

2.4.1 Total Polyphenol Content

Singleton method with a few modifications was followed to estimate *P. guajava* and *S. jambos* ethanol leaf extracts' total polyphenol content. 0.5 millilitres of each extract were placed in a test tube, and two millilitres of 7.5% Na₂CO₃ and 2.5 millilitres of 10% Folin-Ciocalteu reagent were added. With the aid of a shaker, the tubes were vigorously shaken. After the combined solution was left undisturbed for 30 minutes, the absorbance at 765 nm was measured. The total amount of polyphenols was calculated as milligrams of gallic acid equivalent per gram of dry matter extract using the standard gallic acid curve¹¹.

2.4.2 Total Content of Flavonoids

In each test tube, 0.5 ml of the ethanol leaf extracts of *P. guajava* and *S. jambos* were combined with 1.5 ml of methanol. Subsequently, 0.1 ml of AlCl₃ was added to 2.8 ml of distilled water and 0.1 ml of 1M potassium acetate. After thoroughly mixing the solution, the test tube was allowed to sit at room temperature for half an hour. The absorbance was measured with a spectrophotometer at 415 nm. Using the standard quercetin curve, the total flavonoid content was calculated and represented as milligrams of Quercetin Equivalent (QE) per gram of dry matter extract¹².

2.4.3 Tannin Content

50 ml of distilled water was added to a conical flask containing 0.5 g of each plant sample, and the mixture was stirred for an hour. Using a volumetric flask, the filtrate was made up to 50 ml after the solution was filtered. 5 ml of this filtrate and 2 ml of 0.1M FeCl₃ made in 0.1M HCl and 2 ml of 0.008M K₄Fe(CN)₆·3H₂O were added to a tube. Following a 10 minute incubation period, the absorbance was measured at 395 nm using a spectrophotometer¹³. A tannic acid standard curve was used to calculate total tannin content, which was then reported as µg tannic acid equivalent per gram of dry matter extract.

2.4.4 Alkaloid Content

3g of the dried plant powder of *P. guajava* and *S. jambos* were taken in an Erlenmeyer flask. The flask was filled

with 20 ml of 10% acetic acid prepared in ethanol and covered right away. For four hours, the solution was not disturbed. Following the incubation period, the solution was filtered, and a dropwise addition of concentrated ammonium hydroxide was made to the filtrate until no more precipitate formed. Thereafter, the solution was allowed to stand such that the precipitate could settle down. After collecting the precipitate, it was filtered after cleaning with 10% ammonium hydroxide. To find the number of alkaloids in each gram of the sample powder, the residue was dried and weighed¹⁴.

2.5 Antioxidant Activity (DPPH Assay)

The commonly used 2,2-Diphenyl-1-picryl hydrazyl (DPPH) assay was employed to evaluate the extracts' antioxidant capacity. To make up the volume to 2 ml, a range of extract volumes (50 μ l, 100 μ l, 150 μ l, and 200 μ l) were added to 1 ml of 0.2 mM DPPH solution and pure methanol solution. For sixty minutes, the tubes were incubated in the dark. At 517 nm, the absorbance was measured using ascorbic acid as the reference and methanol as the blank solution¹⁵. The percentage antioxidant activity was calculated using the formula $[(Ac-As)/Ac] \times 100$, where Ac is the absorbance of the control at 517 nm and As is the absorbance of the sample at 517 nm (Control was 0.2mM DPPH)¹⁶.

2.6 Reducing Power Assay

Another frequently followed technique to assess the antioxidant activity of plant extracts is the reducing power assay. A range of dilutions (0.2, 0.4, 0.6, 0.8 and 1 ml) of the ethanol leaf extracts of *P. guajava* and *S. jambos* were taken in clean test tubes and 2.5 ml of phosphate buffer (pH 6.6) was added. Then, freshly prepared 1% potassium ferricyanide was added into each of the tubes to incubate them at 50°C for 20 minutes. Post the incubation phase, the tubes were centrifuged for 10 minutes at 6000 rpm. Subsequently, 2.5 ml of 10% trichloroacetic acid was added, and 2.5 ml of the supernatant was carefully pipetted out into new tubes without disturbing the pellet. An equal amount of distilled water was added to the tubes and 0.5 ml of 0.1% ferric chloride was added. Upon the addition of 0.1% FeCl₃, the yellow solution immediately changed its colour to green. Next, at 700 nm, the absorbance was measured using ascorbic acid as a reference¹⁵.

2.7 GC-MS Analysis of *P. guajava* and *S. jambos*

Analysis of various compounds present in the ethanol extract of guava leaves was performed using a GC-MS (Shimadzu model no: QP2010 SE). It is a single quadrupole mass spectrometer. The ethanol extract obtained after Soxhlet extraction could not be injected into the machine directly and therefore required pre-treatment. Hexane was used as the solvent for extraction, and it was vortexed to mix the compounds thoroughly into the solvent¹⁷. The compound analysis for *S. jambos* was done using a Perkin Elmer Clarus 80 GC-MS. Helium was used as a carrier gas during the separation process, with a steady flow rate of 2 millilitres per minute. During the chromatographic run, 220°C was the injector temperature. One microliter of sample was put into the device, and the oven's temperature was set at this level: 50°C was held for two minutes, then 150°C was reached at a rate of 15°C min⁻¹, and finally 250°C was reached at a rate of 30°C min⁻¹, which was held for eight minutes. The conditions for the mass detector were: 230°C for the ion source, 250°C for the inlet line, 70eV for the ionization mode electron impact, 0.2 seconds for the scan period, and 0.1 seconds for the scan interval. The component fragments from 40 to 600 Da spectrums were compared to the database of known component spectra kept in the GC-MS NIST (2014) library¹⁸. The peaks of all the prominent compounds obtained in the chromatograph with their bioactive properties are discussed in the results.

3. Results

3.1 Qualitative Analysis of Phytochemicals

The extracts of *P. guajava* and *S. jambos* were prepared using the powdered plant materials (Figures 1 and 2). Plants produce several compounds that are bioactive and are predominantly used for their protection. Some of the most common compounds present in plants are saponins, tannins, alkaloids, carotenoids, flavonoids, polyphenols and specific polysaccharides. All these compounds contribute towards a plant's antimicrobial, anti-inflammatory, anticancer, antioxidant, antibiofilm properties and much more¹⁹. A number of these compounds were subjected to qualitative analysis in the current investigation using ethanol extracts of both *P. guajava* and *S. jambos* (Table 1).

3.2 Measurement and Evaluation of Phytochemicals

3.3 Total Polyphenol Content

From the standard gallic acid curve (Figure 3), the concentration of polyphenols in the ethanol leaf extracts of *P. guajava* and *S. jambos* in the current study was found to be 0.883 mg/ml and 0.875 mg/ml respectively. Consequently, 88.3 mg and 87.5 mg of gallic acid equivalent per gram of dry extract of the samples, respectively, represent the overall content of polyphenols.

3.4 Total Flavonoids Content

In the current study, the aluminium chloride method was followed to find the total quantity of flavonoids

in the sample and from the standard quercetin curve, the concentration of flavonoids in the ethanol extract of *P. guajava* leaves was discovered to be 0.626 mg/ml. Therefore, the final concentration of flavonoids can be reported as 62.6mg quercetin equivalent per gram of dry extract. On the other hand, the concentration of flavonoids in ethanol extract of *S. jambos* leaves was found to be 0.8206 mg/ml and hence, from the standard quercetin curve (Figure 4), it can be reported as 82.06mg quercetin equivalent per gram of dry extract.

3.5 Tannin Content

From the standard curve (Figure 5), the concentration of tannins was found to be 1.389µg/ml for guava leaves. The



Shade dried leaves of *Psidium guajava*



Powdered leaves



Soxhlet extraction of *Psidium guajava* leaves using ethanol

Figure 1. Sample preparation of *Psidium guajava* leaves for extraction.



Shade dried leaves of *Syzygium jambos*



Powdered leaves



Soxhlet extraction of *Syzygium jambos* leaves using ethanol

Figure 2. Sample preparation of *S. jambos* leaves for extraction.

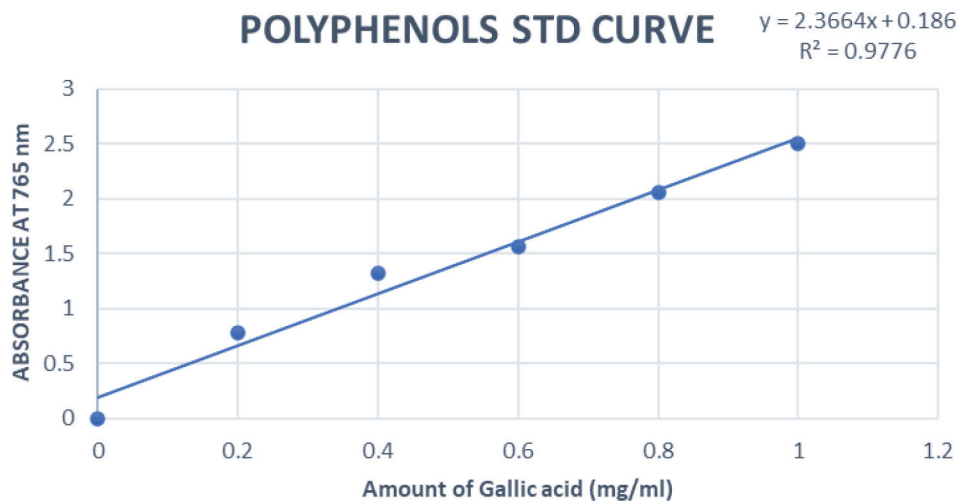
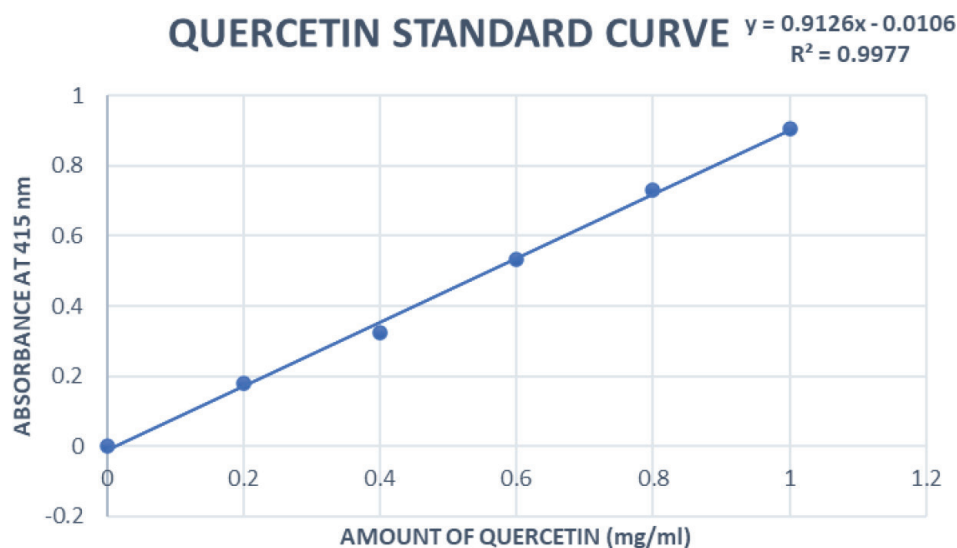
Table 1. Qualitative analysis of phytochemicals in ethanol leaf extracts of *P. guava* and *S. jambos*

Phytochemicals	<i>Psidium guajava</i>	<i>Syzygium jambos</i>
Alkaloids	+	+
Flavonoids	+	+
Glycosides	+	+
Phenol	+	+
Saponins	-	-
Sterols	+	+
Tannins	+	+
Terpenoids	+	+

sample was diluted six times as the colour was extremely dark. Therefore, 8.334 μ g of tannins was present in 0.5g of the plant powder and it can be further reported as 0.016mg tannic acid equivalent per gram of dry matter extract. Similarly, for *S. jambos*, the concentration of tannins was found to be 1.805 μ g/ml. The sample was diluted thrice and thus, 5.415 μ g of tannins was present in 0.5g of the plant powder. Hence, the result could be represented as .018mg tannic acid equivalent per gram of dry matter extract.

3.6 Alkaloid Content

In this study, the precipitate obtained was filtered after washing with ammonium hydroxide solution. The filter

**Figure 3.** Calibration curve for polyphenols using gallic acid.**Figure 4.** Calibration curve for flavonoids using quercetin.

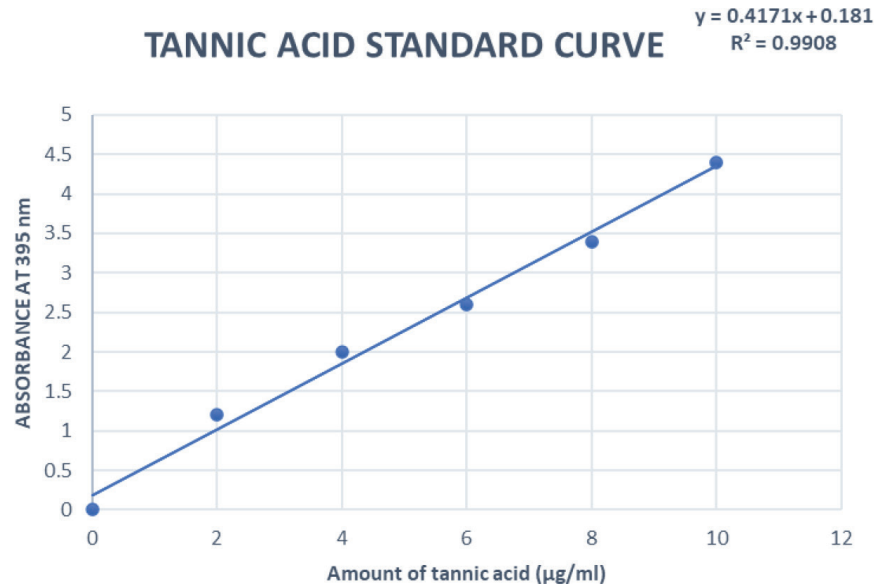


Figure 5. Calibration curve for tannins using tannic acid.

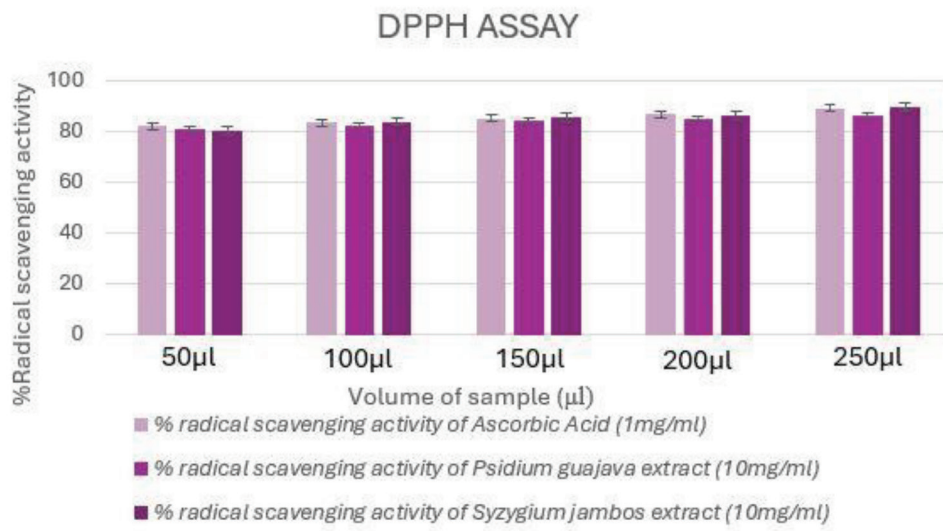


Figure 6. Comparison of the percentage antioxidant activity between standard ascorbic acid and ethanol leaf extract of *Psidium guajava* and *S. jambos*.

paper weighed 1.06g before filtration but after filtration and drying, it weighed 1.52g. Subtracting the values, the quantity of alkaloids present in 3g of the plant powder was deduced to be 0.46g or 460mg. Therefore, it can be concluded that 1g of guava leaves powder contains 153.33mg of alkaloids. Likewise, after filtration and drying the precipitate obtained from *S. jambos* weighed 0.575g or 575mg. Thus, 1g of *S. jambos* leaves powder contains 191.66mg of alkaloids.

3.7 Antioxidant Activity (DPPH Assay)

The radical scavenging property (Figure 6) for all the dilutions of the standard and extracts was estimated by substituting the values in the prescribed formula and is represented in Supplementary Table 1. The highest concentrated sample showed the best percentage of radical scavenging activity for the ethanol extract of guava leaves (86.24%) and ethanol extract of *S. jambos* leaves (89.43%).

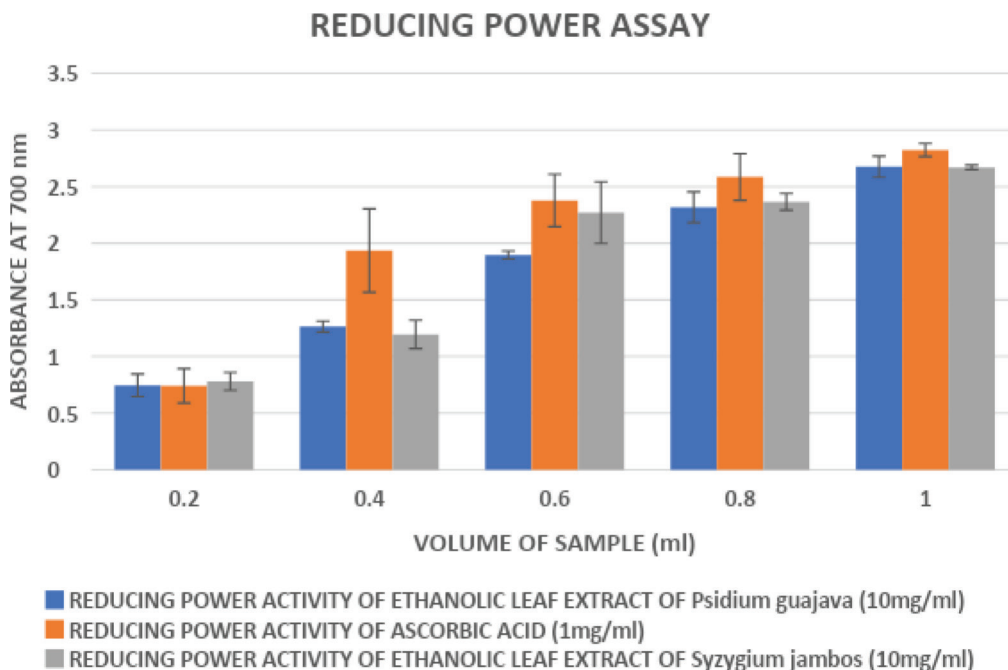


Figure 7. Comparison between reducing the power of ethanol extract of *P. guajava* leaves standard ascorbic acid and ethanol leaf extract of *S. jambos*.

3.8 Reducing Power Assay

The reducing power of the extracts and reference ascorbic acid was tested for a range of dilutions, and it was observed that as the concentration increased, the final colour obtained was a darker shade of green. The dilutions taken were 0.2 ml, 0.4 ml, 0.6 ml, 0.8 ml and 1 ml of the extract and the absorbance readings recorded at 700 nm were 0.74, 1.26, 1.89, 2.31 and 2.67 respectively for *P. guajava*. Likewise, for *S. jambos*, the values recorded were 0.78, 1.19, 2.27, 2.36 and 2.67 respectively (Figure 7). The experiment was done in triplicates and the mean values against standard deviation are shown in Supplementary Table 2.

3.9 Phytochemical Constituents of Ethanol Extract of *P. guajava* Leaves Detected by GC-MS

About nine prominent peaks were observed when a GC-MS analysis was done for the ethanol extract of *S. jambos* (Figure 8). Phytol, neophytadiene, phenol-3-penta decyl, and 1,8,11,14-heptadecatetraene, (Z, Z, Z) were some of the components detected in the extract with immense biological activities and are listed in Table 2. On the other hand, a total of 30 components (Figure 9) were identified in the leaves of *P. guajava*

extracted with ethanol and are listed in Table 3 along with their biological activities. Some of the most important components present are caryophyllene, hexacosane, hexatriacontane, tetratetracontane, tetracosane, heneicosane, 1,6,10-dodecatrien-3-ol,3,7,11, trimethyl- and β -bisabolene.

4. Discussion

The leaves of *P. guajava* extracted in ethanol are known to possess beneficial phytochemicals in abundance. The phytochemicals present in the leaves were qualitatively and quantitatively analysed in the present study. The antioxidant action of different plants is mostly attributed to the phenolic compounds present in them. The ethanol leaf extracts of both *P. guajava* and *S. jambos* showed tremendous antioxidant properties and can be deduced that the polyphenols play a major role in stabilizing the antioxidants. The Folin-Ciocalteu method revolves around the migration of electrons from phenolic components to phosphomolybdic/phosphotungstic acid in an alkaline medium to form a blue-coloured complex. In an earlier study, the concentration of total phenolic compounds was compared between aqueous, methanol and ethanol

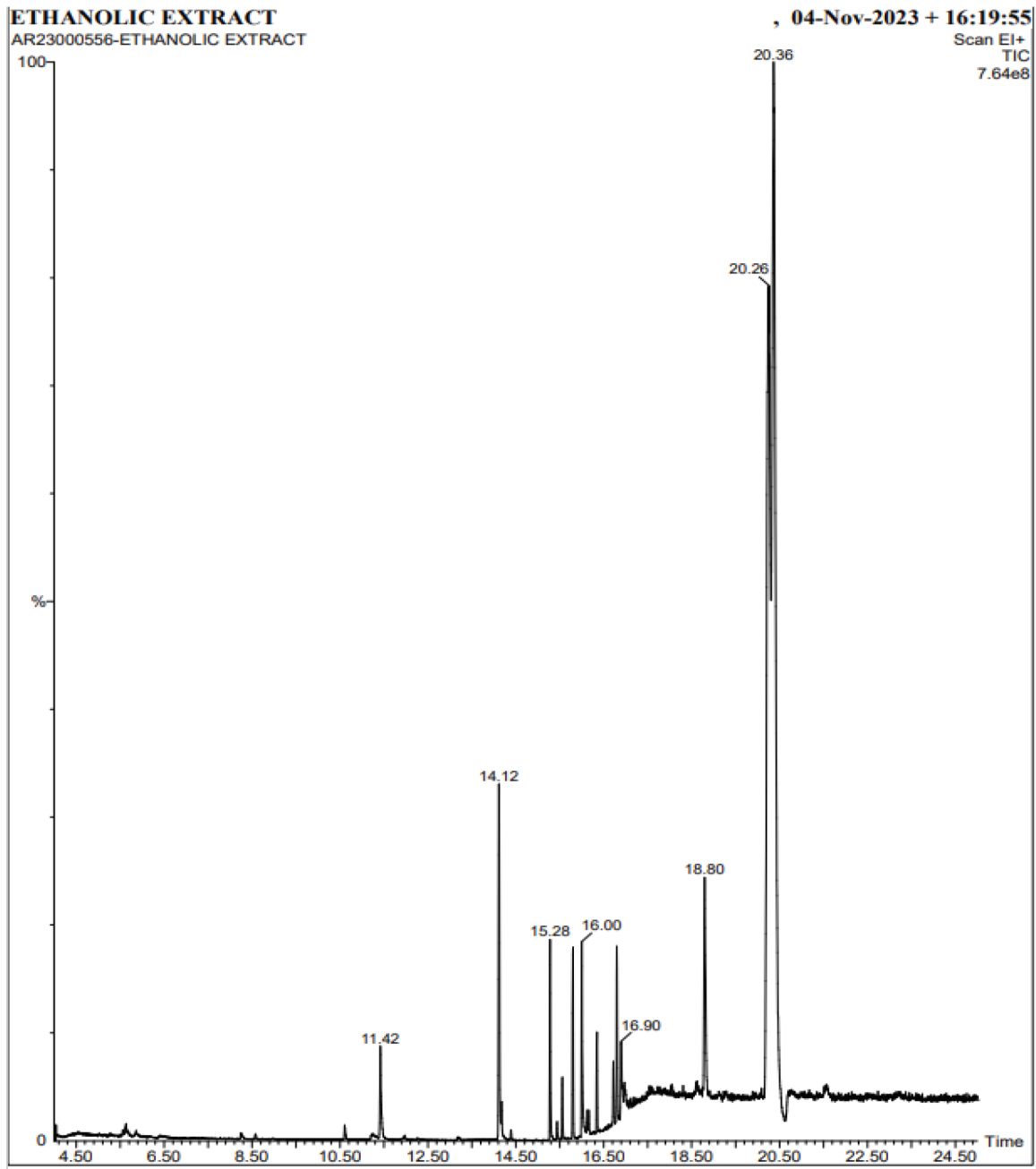


Figure 8. Chromatogram obtained from the GC-MS analysis of the ethanol leaf extract of *S. jambos*.

extracts of guava leaves and was concluded that the aqueous extraction had the maximum amount of total phenol compounds followed by pure ethanol extract and pure methanol extract. The concentration of polyphenols in the ethanol extract was deduced to be 110mg caffeic acid equivalent per gram of dry extract which is close to the values found in this investigation⁵³. A study was also conducted in the recent past to check the impact of altitude and solvents on the concentration

of phytochemicals in guava leaves. From the study, it was concluded that guava trees growing in higher altitudes and ethanol extract of guava leaves showed the highest concentration of polyphenols (331.84 mg GAE/g)⁵⁴. It has been also proven that guava leaves when extracted with 50% and 70% ethanol showed 574.5 mg GAE/g and 426 mg GAE/g total phenolic compounds respectively. Other related studies reiterate the fact that more polyphenols are observed as the

Table 2. Compounds in the ethanol leaf extract of *S. jambos* identified by GC-MS analysis and their biological properties

Name of the compound	Biological Activity	Reference
Dodecyl acrylate	Antibacterial	20
Neophytadiene	Antimicrobial and anti-inflammatory	21
Hexadecenoic acid, methyl ester	Antimicrobial activity against MDR strains	22
n-hexadecanoic acid	Anti-inflammatory activity	23
Phytol	Antioxidant, apoptosis-inducing, anti-inflammatory, antimicrobial.	24
Phenol-3-pentadactyl	Oxygen scavenging and anti-inflammatory.	25
1,8,11,14-heptadecatetraene, (Z,Z,Z)	anti-inflammatory, antiulcer, antitumor, antiparasitic, analgesic, antiviral, antibacterial, insect deterrent and antifungal.	24,25

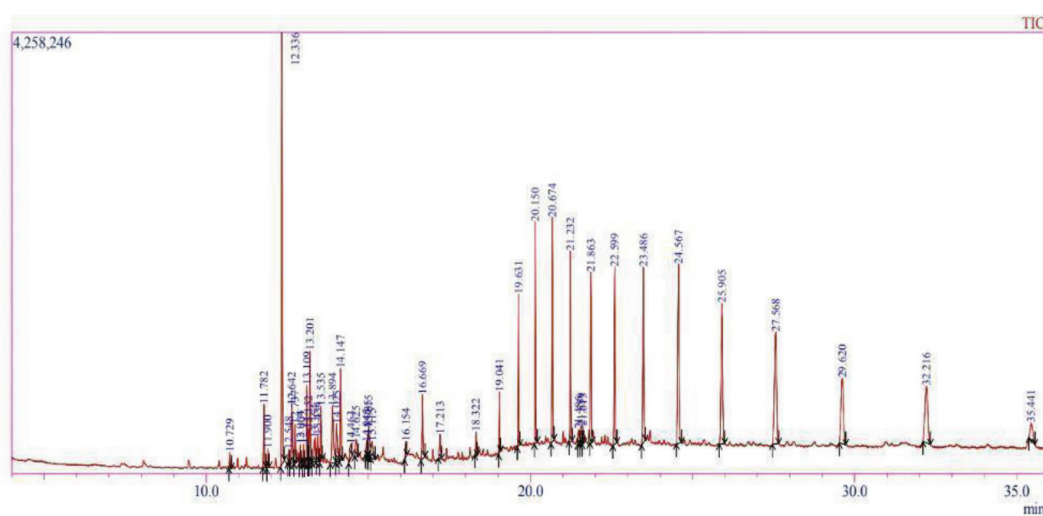


Figure 9. Chromatogram obtained from the GC-MS analysis of the ethanol leaf extract of *P. guajava*.

concentration of water in the extract increases⁴². Since 100% ethanol was used for extraction in the present study, the values obtained for the total polyphenols can be relied upon. In a study conducted by Gabriela Bonfanti *et al.*, the total phenolic content in an aqueous extract of *S. jambos* leaves was found to be 108.2 mg GAE/g which is slightly higher than the concentration obtained in the current study⁷. Flavonoids are sometimes referred to as vitamin P and it has been proven that the colour of the plants is imparted by this group of phytochemicals. In an earlier study by Rica Hartati *et al.*, the total flavonoid content was compared between ethanol, ethyl acetate and hexane extracts of guava leaves and the highest concentration of flavonoid was observed in the hexane extract (9.68g QE/100 g or 96mg QE/g) which was followed by ethanol and ethyl acetate extracts⁵⁶. The flavonoids quantified in

the present study were less than 96 mg QE/g and can be correlated with the earlier literature available. The concentration of flavonoids obtained in the ethanol extract of *S. jambos* leaves was higher than in guava (82.06mg QE/g) and is comparable to the results obtained by Gabriela Bonfanti *et al.*, (85.55mg QE/g)⁷. Tannins are molecules that form strong complexes with proteins. They are of two types: hydrolysable tannins and condensed tannins. In the current study, the amount of tannin overall in the guava leaf ethanol extract was quantified using tannic acid as the standard. In a study conducted recently, a comparison was drawn between the concentration of tannins present in aqueous, ethanol and methanol extracts of guava leaves and it was concluded that the aqueous extract showed the highest concentration of tannins which was followed by methanol and ethanol extracts. The ethanol

Table 3. Compounds in the ethanol leaf extract of *P. guajava* identified by GC-MS analysis and their biological properties

Name of Compounds	Biological Activity	References
Hexacosane	Antimicrobial, antibacterial effects against <i>Staphylococcus aureus</i> and <i>Streptococcus pneumoniae</i>	27
Tetratetracontane	Antioxidant and cytoprotective activities	28
Hexatriacontane	Anti-inflammatory, analgesic activity, radical scavenger and antioxidant activity	29
Tetracosane	Cytotoxic effect against colon cancer cells and estrogen-dependent breast cancer cells, antidiarrheal, cardiotoxic, laxative, anthelmintic and removes fatigue, anti-inflammatory, used in peptic ulcer treatment	30
Nonadecane,2-methyl-	Antibacterial	31
Bis(2-ethylhexyl) phthalate	Apoptosis inhibitor, anti-microbial, cytotoxic activity	32
5,8,11,14- Eicosatetraenoic acid, methyl ester	Antioxidant, anti-inflammatory	33
Heneicosane	Antimicrobial activity against <i>Streptococcus pneumoniae</i>	34
Nonadecane	Antimicrobial, antioxidant, anticancer, anti HIV	31
1-Octadecyne	Antimicrobial	35
Caryophyllene	Antibacterial activity against <i>S. aureus</i> , antifungal activity, strong antioxidant effects, and anti-proliferative effects against cancer cells, demonstrated potent inhibition against clonogenicity, migration, invasion and spheroid formation in colon cancer cells.	36
Tridecane	Anti-inflammatory, anti-fungal anti-bacterial against gram-positive bacteria	27
Copaene	Antioxidant, antimicrobial and cytotoxic	37
Tetradecane	Antibacterial diuretic and anti-tuberculosis and antifungal	38
Caryophyllene	Antimicrobial local anaesthetic, anticarcinogenic and anti-inflammatory activities.	39
cis-beta-Farnesene	Sedative, anti-inflammatory, antifungal and antibacterial	40
Alpha-cubebene	Anti-bacterial, antifungal antioxidant and anti-inflammatory	
Humulene	Anti-inflammatory, anti-fungal, anti-bacterial, anti-cancer	39
Naphthalene-1,2,4a,5,6,8a-hexahydro-4,7-din	Antifungal, antioxidant and antibacterial	42
Cis-alpha-Bisabolene	Antimicrobial and antioxidant	43
1H-Cyclopenta[1,3] cyclopropa[1,2] benzene	Antioxidant, Insectifuge, anticancer, antimicrobial, antigenotoxic anti-inflammatory, nematocidal, anthelmintic, antihistaminic, antieczemic	44
Beta-bisabolene	It demonstrates particular cytotoxicity against cancerous cells	45
Cis - calamenene	Dendritic cells that were treated with calamenene showed increased activating capacity of T cells of the immune system. As a result, this can be used in immunotherapy treatments	46
1,6,10-Dodecatrien-3-ol,3,7,11-trimethyl	Antibacterial and antifungal	47
Caryophyllene oxide	Inhibits PI3K/Akt/mTOR signalling pathway is significantly inhibited by it, demonstrating its; hence antitumor.	48
Cubenol	Feeding habit inhibitor of plant pests like <i>H. obscurus</i> ; alternative to chemical pesticides	49
Hexadecane	Anti-inflammatory and anti-tumour activity	50
Epoxy-alpha-terphenyl acetate	It acts as a competitive inhibitor of a hepatic enzyme that is used in clinical drug metabolism	51
Octadecane,3-ethyl-5-(2-ethylbutyl)-	Antimicrobial effects	52

extract showed 95.35 mg TAE/g which is much higher when compared to the result obtained in the current study⁵⁷. Alkaloids more often act as the defence system in plants where they ward off insects and predators. These nitrogen-rich compounds are extensively used in the medical domain as anaesthetics, cardioprotective agents, and anti-inflammatory agents⁵⁸. A study done by Oncho *et al.*, using the same protocol that has been used in this study showed an alkaloid concentration of 96.67 mg/g and 84.33 mg/g of powdered guava leaves from two different districts in Ethiopia. These values are relatively lower than the concentration of alkaloids obtained in the current study. The variations could be because of the drastic difference in the climatic conditions between the Gursum and Babile districts of Ethiopia and Kerala in India¹⁴. Among *P. guajava* and *S. jambos*, the latter showed relatively higher concentrations of alkaloids.

Many antioxidants present in plants are non-enzymatic and are either expressed constitutively or when challenged with biotic or abiotic stress. Catalase, superoxide dismutase, glutathione reductase, and glutathione peroxidase are examples of enzymatic antioxidants in plants, whereas low-molecular-weight compounds such as ascorbic acid, flavonoids, phenolic acids, and glutathione comprise the majority of non-enzymatic antioxidants. This antioxidant activity in plants is a reflection of their genetic make-up where they fundamentally produce myriad phytochemicals to perform their physiological functions and in response to environmental stress conditions. Hence, several *in vitro* studies are being done to validate the antioxidant potential of plants⁵⁹. To assess or estimate the antioxidant activity of any substance, the DPPH assay is extensively used. 2,2-diphenyl-1-picrylhydrazyl is a stable free radical that is quenched by phenolic substances in plant extracts. The purple-coloured reagent gets converted into a light-yellow colour when a hydrogen ion is received from the phenolic components, thereby making the reagent 2,2-diphenyl-1-picryl hydrazine⁶⁰. In one of the earlier studies, the percentage of free radical scavenging activity of guava leaves was found to be 95.784% which was slightly more than the result obtained in the current study⁶¹. Another very common assay performed to check the antioxidant potential of plants is the reducing power assay. The solution's colour shifts from yellow to various green and

blue hues due to the antioxidants breaking down the ferricyanide molecules into ferrocyanide. Therefore, the measurement of Perl's Prussian blue complex at 700 nm gives the concentration of Fe^{2+} ions⁵⁹. Comparing both the ethanol extracts, the antioxidant potential was found to be slightly more in *S. jambos*. Most of the components detected in this study were found to have excellent bioactivity and have been discussed in detail in Tables 2 and 3.

5. Conclusion

Psidium guajava and *Syzygium jambos* are plants from the *Myrtaceae* family and have been in use since ancient times for food and medicinal purposes. These plants are loaded with compounds that have high medicinal value and therefore one of the reasons for which it is cultivated in abundance in most places. A few classes of phytochemicals were quantified in this study which were comparable with the previous studies conducted. Considering the amount of toxins or free radicals that accumulate in our system daily, there is a vast scope to formulate the ethanol extract of *P. guajava* leaves and *S. jambos* leaves into jellies, ointments, cosmetic products etc. Compounds with anticancer properties could be studied in great detail to incorporate them in the cancer treatment regime. Thus, a synergistic approach of combining both extracts could be experimented with for better results and applications in the field of medicine and cosmetics.

6. Acknowledgements

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