**ABSTRACT**

Popular and commercially significant plant *Carica papaya* leaf has been investigated for its potential as a platelet stimulant. Papain, chymopapain, glycyl endopeptidase, and caricain are four cysteine endopeptidases that are abundantly found in latex, which is secreted by lactifers in the plant. The leaves include the glucoside carposide and the alkaloid carpaine. Alkaloids are frequent carboxylic acid salts that are organic nitrogenous bases and heterocyclic compounds found in plants. According to reports, carpine can depress the central nervous system and produce bradycardia. Blood cells called platelets, also known as thrombocytes, react to bleeding caused by blood vessel damage and take part in both innate and adaptive intravascular immune responses. They are created from totipotent marrow stem cells, and thrombopoietin controls their production. In addition to having anti-tumour and immunomodulatory actions, *Carica papaya* leaf extract has been shown to raise platelet count and hematocrit levels in dengue fever patients. Alkaloids, glycosides, tannins, saponins, and flavonoids are among the active ingredients found in leaf extract. The carpaine works by encouraging the bone marrow to create more megakaryocytes, which are in charge of producing and aggregating platelets in blood circulation. Papaya leaf juice boosts the expression of the CD110 receptor on megakaryocytes, successfully treating thrombocytopenia brought on by chemotherapy. Additionally, it keeps hematocrit levels normal and lessens platelet deterioration. Although studies on mice did not reveal any negative consequences, greater dosages may result in possible liver damage. In Sri Lanka, the extract & potential for treating dengue and promoting wound healing has been studied.

**KEYWORDS:** According to reports, carpine can depress the central nervous system and
INTRODUCTION
For thousands of years, plants have served as a significant source of medicine. Up to 80% of people, according to the World Health Organization (WHO), still largely use traditional therapies like herbs for their medications today. These plants' therapeutic efficacy is a result of a range of phytochemicals and their elemental make-up. Due to their minimal adverse effects, plant products are preferred over synthetic medications to treat a variety of ailments. As a result, the discipline of traditional medicine has recently experienced its fastest expansion. In India, the primary ingredient in ancient medical systems like Ayurveda and Siddha is medication derived from plants. Due to its effective therapeutic value, which provided the foundation for the development of newer medications to treat a variety of disorders, researchers investigated herbal compounds. The Carica papaya leaf, which is a member of the monogenetic genus caricaeae family, is one of the significant plants with a high therapeutic value.

CARICA PAPAYA
Given that its fruit is a common delicacy, Carica papaya (family Caricaceae) is one of the most well-known and commercially significant plants in the world. It is a perennial tree with a single stem that is softly forested. Its height ranges from 2 to 10 meters, and its crown of broad palmate leaves emerges from the top of the trunk. The soft, hollow, cylindrical trunk has a diameter of around 5 cm at the crown and 30 cm at the base. Despite being a native of Central America, it has been introduced to many tropical regions. The papaya plant is lactiferous because it has unique cells called lactifers, which are present in nearly all tissues and exude latex. Lactifiers secrete latex and are dispersed throughout most plant tissues. The papaya-latex is well known for being a rich source of the four cysteine endopeptidases namely papain, chymopapain, glycyl endopeptidase and caricaain. Leaves contain an alkaloid called carpaine and a glucoside named carposide.

Alkaloids
A diverse range of secondary metabolites produced by plants may not directly contribute to the growth and development of the organism. Although many of these metabolites may be produced in the lab, doing so generally involves a lot of work and produces modest yields. Although the structural complexity of some instances quickly increases the number of subdivisions, alkaloids are frequently categorized according to the kind of nitrogen-containing
structure, such as piperidine, pyrolidine, and indole. Their term, alkaloids, derives from the fact that their amine composition causes alkaline solutions in water. Numerous alkaloids biological functions frequently rely on protonation at physiological pH, which converts their amine function into a quarternary system. Based on their precursor amino acids, alkaloids are further split into categories. True alkaloids, proto alkaloids, and pseudo alkaloids are the three primary types of alkaloids according to categorization.

**Carpaine**

Papaya leaves contain alkaloids carpaine, dehydrocarpaine I and II, pseudocarpaine. Carpine has been reported to cause bradycardia and central nervous system depression. In natural product chemistry, carpaine has been classified under the group of alkaloids and it has a macrocyclic dilactone structure, a cyclic hydrocarbons that contain multiple rings and share one or more atoms. Carpine has been documented as monoclinic prisms from acetone which sublimes at 120°C under 0.05 mm pressure. It is slightly soluble in water and soluble in most organic solvents except petroleum ether.

**Properties of carpaine compounds**

<table>
<thead>
<tr>
<th>Name</th>
<th>Carpaine</th>
</tr>
</thead>
<tbody>
<tr>
<td>Chemical Name</td>
<td>12, 25-Diaza-13, 26-dimethyl-2, 15-Dioxatricyclo [22.2.2.2] triacontane-3,16-dione</td>
</tr>
<tr>
<td>Formula</td>
<td>C28H50N2O4</td>
</tr>
<tr>
<td>Molecular Mass</td>
<td>478.70 g/mol</td>
</tr>
<tr>
<td>Melting Point</td>
<td>121 °C</td>
</tr>
<tr>
<td>Solubility</td>
<td>30 mg/ml in water at 80 °C, less than 0.5 mg/ml at 25 °C</td>
</tr>
<tr>
<td>pH in aqueous medium</td>
<td>4.6</td>
</tr>
<tr>
<td>Crystal Structure</td>
<td>The molecule is flexible and the changes in the conformations are brought about by the hydrogen bonding of the Protonated-N atoms with two Br atoms and the water molecule</td>
</tr>
<tr>
<td>Chemical structure</td>
<td><img src="image" alt="Chemical structure" /></td>
</tr>
</tbody>
</table>
PLATELETS
When a blood artery is injured and begins to bleed, platelets, also known as thrombocytes, work with coagulation factors to clump together and create a blood clot. Megakaryocytes from the bone marrow or lung produce platelets, which are cytoplasmic pieces that travel through the bloodstream without having cell nuclei. Only mammals have platelets; in other vertebrates (such as birds and amphibians), thrombocytes circulate as whole mononuclear cells. Platelets play a key role in hemostasis, the process of halting bleeding at the site of ruptured endothelium.

First, adhesion happens when platelets stick to things outside the injured endothelium. Second, they undergo a shape change, activate receptors, and release chemical messengers.

Thirdly, they aggregate to form connections with one another via receptor bridges. The coagulation cascade is activated in conjunction with the formation of this platelet clog (primary hemostasis), resulting in fibrin deposition and linkage (secondary hemostasis).

Structure
Structurally the platelet can be divided into four zones, from peripheral to innermost.

- Glycoproteins needed for platelet adhesion, activation, and aggregation are abundant in the peripheral zone. For instance, GPVI, GPIIb/IIIa, and GPIb/IX/V.
- Microtubules and microfilaments are abundant in the sol-gel zone, which helps the platelets keep their discoid form.
- Platelet granules are abundant in the organelle zone. Factor V, factor VIII, fibrinogen, fibronectin, platelet-derived growth factor, and chemotactic substances are among the clotting mediators found in alpha granules. Delta granules, also known as dense bodies, contain the platelet-activating mediators ADP, calcium, and serotonin.
- The manufacture of thromboxane A2 takes place in the membraneous zone, which is made up of membranes produced from megakaryocyte smooth endoplasmic reticulum and arranged into a dense tubular system. To facilitate the release of thromboxane A2, this thick tubular structure is joined to the surface platelet membrane.

Shape
Inactive platelets in circulation are biconvex discoid (lens-shaped) formations with a maximum diameter of 2-3 m. Cell membrane projections cover the surface of activated platelets.
Development

- The totipotent marrow stem cells are the source of platelets.
- Thrombopoietin, a hormone generated in the kidneys and liver, controls the generation of megakaryocytes and platelets.
- Between 1,000 and 3,000 platelets are produced by each megakaryocyte during the course of its existence.
- Reserve platelets are kept in the spleen and released when needed by splenic contraction brought on by the sympathetic nervous system. A healthy adult produces 10 platelets each day on average.
- Circulating platelets have a life expectancy of 8 to 9 days on average. A Bcl-xL timer is part of the internal apoptotic regulatory system, which regulates how long individual platelets live.
- In the spleen and liver, phagocytosis breaks down old platelets.

REVIEW OF LITREATURE

The literature review encompasses systematic information on whether *Carica papaya* leaf extract increases platelets. It was found that very little information was available on the plants of this species.


10. Vuong QV, Hirun S, Roach PD, Bowyer MC, Phillips PA, Scarlett CJ. Effect of extraction conditions on total phenolic compounds and antioxidant activities of Carica papaya leaf aqueous extracts. Journal of Herbal Medicine, 2013 Sep 1; 3(3): 104-11.

OBJECTIVES OF THE STUDY

- The present study was aimed to determine Carica papaya used to increase the platelet count and study its mechanism of action.
- The carpaine is extracted from papaya leaves using both hot and cold extraction methods.
- The present investigation aims to evaluate the pharmacognostical studies such as morphology and microscopy of leaf. The physico-chemical parameters such as loss on drying, total ash content, and sulphated ash were determined. Phytochemical parameters of C. papaya plant were analysed in accordance with Ayurvedic Pharmacopeia of India.

PLANT PROFILE

Biological Source: It consists of dried leaves of Carica papaya

Family: Caricaceae

Synonyms of Carica papaya Linn.

Indian and International synonyms of Carica papaya Linn. and different species of Carica papaya Linn. are described.

<table>
<thead>
<tr>
<th>COUNTRY</th>
<th>NAMES</th>
</tr>
</thead>
<tbody>
<tr>
<td>India</td>
<td>Papita</td>
</tr>
<tr>
<td>Holland</td>
<td>Tree melon</td>
</tr>
<tr>
<td>France</td>
<td>Papaya</td>
</tr>
<tr>
<td>Australia</td>
<td>Paw Paw</td>
</tr>
</tbody>
</table>

Brazil | Mamo
---|---
UK | Papa, Paw Paw

**TAXONOMICAL CLASSIFICATION**

- Domain: Flowering plant
- Kingdom: Plantae
- Sub Kingdom: Tracheobionta
- Class: Magnoliopsida
- Super division: Spermatophyta
- Phylum: Steptophyta
- Order: Brassicales
- Family: Caricaceae
- Genus: Carica
- Botanical Name: *Carica papaya* Linn

**GEOGRAPHICAL SOURCE**

Papaya (*Carica papaya* L.) is a tropical plant mainly produced by India, Brazil, Mexico, and Nigeria.

**MORPHOLOGY OF LEAVES**

- Color - Green.
- Odour - Characteristics.
- Taste - Very bitter taste.
- Shape - Broad, flat, and deeply, palmately lobed Size 50-70 cm diameter and 18-90 cm in length.

**ETHNOMEDICINAL USE**

Used in the treatment of
- Symptoms related to dengue fever
- Inflammation
- Improving blood sugar control
- Supporting skin
- Hair health
- Preventing cancer.
MATERIALS AND METHODS

COLLECTION OF SAMPLES
Crisp leaves (plant verification example number p 06010717) of the Carica papaya were collected fresh in July, washed in water, cleaned with a suitable material to remove dust, shade-dried at room temperature, and ground using a blender processor.

Extraction
Cold water extraction
5 L of ultra-pure water and 1 kg of pulverized leaf samples were combined, and the extract was filtered before being put into a plastic bag. The extract was kept in a freezer at 20°C. The frozen sample was then moved into a freeze-dry vessel and lyophilized using a freeze drier device to produce powder. The powder was then gathered, weighed, and saved for later use in a labeled bottle at 20°C.

Hot Water Extraction
5 L of ultra-pure water and 1 kg of pulverized leaf samples were combined, and the mixture was heated to 40 °C for 3 hours before being allowed to cool to room temperature and being filtered through cloth. The extract was placed in a plastic bag and kept in a freezer set at 20°C. The frozen sample was then moved into a freeze-dry vessel and lyophilized using a freeze drier device to produce powder. The powder extracts were then gathered, weighted, preserved in a labelled bottle at 20°C for future use, and then used.

Extraction of carpaine
Papaya leaves have been collected at a farm in India, Negeri, Sembilan, Malaysia. We utilized both the young (green) and the aged (yellow) leaves. Young leaf stalks were also cut apart and utilized. Following a thorough cleaning under running water, the samples were rinsed in distilled water. The leaves were divided into pieces measuring about 7 cm long and 9.0 cm wide. Using a low speed electrical blender (Panasonic, Malaysia), half of the samples were mixed with distilled water (ratio of 300 g sample to 200 mL distilled water), while the other half was left unblended. The samples of both kinds (blended and unblended) were stored overnight at -12 °C in a deep freezer.

The dried materials were crushed into powder after freeze drying in preparation for further chemical analysis. Since freeze drying successfully preserves the majority of the bioactive compounds during drying, it was chosen as the approach. Alkaloid in a warm water bath at
60°C for a one hour. To separate the ethanol extract from the plant remains, the mixture was decanted and filtered. Using a rotary-evaporator, the resultant extract was subsequently concentrated. Alkaloids were extracted using acid-base extraction (ABE), and ethanol was recovered for further usage. It was acidified with tartaric acid (3%) to improve its solubility in water, and then it was filtered through the kieselghur layer using a Buchner funnel. After that, the ammonia solution was used to basify the acidic filtrate until the pH rose. freeze-dried papaya leaf and stem powders were steeped in 95% ethanol.

PHYSIOCHEMICAL EVALUATION STUDIES
Physicochemical parameters were determined as per guidelines of WHO. Loss on drying, total ash value, Sulphated ash were determined.

DETERMINATION OF LOSS ON DRYING (LOD)
Weigh about 1.0gm of powdered drug into a weighed china dish. Dry in the hot air oven at 105°C, until two consecutive weighing do not differ by more than 0.5mg. Cool in a desiccator. The weight after drying was noted, loss on drying was calculated, the percentage was expressed as %w/w with reference to air dried sample, and the weight after drying was weighed.

DETERMINATION OF TOTAL ASH VALUE
Weight accurately about 1 g of sample in a China dish. Heat the China dish carefully over a small flame to char the material Ignite in a muffle furnace at 500 ± 25°C for one hour. Grey ash is obtained. Cool the China dish in the desiccators. If wetting shows ash to be carbon- free, remove the dish from desiccators. Weigh the China dish and repeat the operations for two successive weighing. Record the lowest mass and finally calculate the result.

DETERMINATION OF SULPHATED ASH
Weight accurately about 1- 2g of sample in a china dish. Add the sulfuric acid and heat the china dish carefully over a small flame until white fumes no longer evolve to remove any excess sulfuric acid in the sample. Ignite in a muffle furnace at 800 ± 25°C for one hour. Grey ash is obtained. Cool the China dish in the desiccators. If wetting shows as, remove the dish from desiccators. Weigh the China dish and repeat the operations for two successive weighing. Record the lowest mass and finally calculate the result.
Chemical test

Detection of Alkaloids

Mayer’s test
Filtrates were treated with Mayer’s reagent (potassium mercuric iodide). The formation of a yellow-colored precipitate indicates the presence of alkaloids.

Detection of Flavonoids

Alkaline reagent test
Extracts were treated with a few drops of sodium hydroxide solution. The formation of intense yellow color, which becomes colorless on the addition of dilute acid, indicates the presence of flavonoids.

Detection of Saponins

Foam test
0.5 g of the extract was shaken with 2 ml of water. If foam produced persists for 10 minutes it indicates the presence of saponins.

Detection of Tannin

Gelatin test
To the extract, a 1% gelatin solution containing sodium chloride was added. The formation of a white precipitate indicates the presence of tannins.

Detection of glycosides

Killer killiani test
Glycoside dissolves in a mixture of 1% ferric sulphate solution in (5%) of glacial acetic acid. Add one or two drops of concentrated sulphuric acid. The blue colour grows due to the presence of deoxy sugar.

Test for Carbohydrates

Fehling’s Test
Take 2 ml of given sample solution in a clean test tube. Add 2 ml of Fehling’s solution A and Fehling’s solution B to it. Keep the solution in a boiling water bath for about 10 minutes. If there is the formation of red precipitate then the presence of carbohydrate is confirmed.
**Test for Protein**

**Biuret test**
An alkaline solution of protein is treated with a drop of aqueous copper sulfate and a bluish violet color is obtained. Note: Formation of violet coloration confirms the presence of Proteins.

**Detection of steroids**

**Liebermann-Burchard test**
When chloroform solution of steroid is treated with acetic anhydride and concentrated sulphuric acid, green colour is formed.

**MECHANISM ACTION OF CARPAINE**

![Diagram of mechanism action of Carpaine](image-url)
Mechanism Action of carpaine
Numerous studies have shown that *C. papaya* leaf enhances platelet synthesis by altering gene expression. Platelet-activating factor receptor (PTAFR) and arachidonate 12-lipoxygenase (ALOX-12) genes are two that are affected by the action of *C. papaya* leaf extract. According to the flow chart, increased expression of particular genes encourages the bone marrow to make more megakaryocytes. These megakaryocytes are the stem cells in charge of producing platelets, and when they mature, they separate into tiny particles known as platelets, which boosts platelet production and aggregation in the bloodstream. Recent experimental research has demonstrated that the carpaine found in *C. papaya* leaves is what causes this mode of action.

Juice from *C. papaya* leaves aids in boosting the megakaryocytes production of CD110 receptors, which are useful in preventing chemotherapy induced thrombocytopenia. By minimizing the breakdown of platelets, *C. papaya* leaf can help treat thrombocytopenia. The functioning and reproducing components of viruses, known as proteases, are found in the leaves of the *Carica papaya* plant and can be bound by flavonoids to stop viral reproduction. As seen in Figure, this procedure reduces platelet oxidation and maintains normal hematocrit levels. Additionally, the leaf extract has antioxidant and free radical scavenging qualities that hinder oxidation and stop hemolysis and bleeding. These extracts also increase the synthesis of platelets by boosting ALOX-12 and PTFAR activity by 15 and 13 and 14 times, respectively.

<table>
<thead>
<tr>
<th>Bioactive Component</th>
<th>Mechanism of Action</th>
</tr>
</thead>
<tbody>
<tr>
<td>Carpaine</td>
<td>An alkaloid Stimulates ALOX-12 and PTFAR genes to produce megakaryocytes that mature and release more platelets. Stimulates CD110, L.e., a receptor on megakaryocytes results in high platelet production.</td>
</tr>
</tbody>
</table>

RESULTS AND DISCUSSION
MACROSCOPIC EVALUATION
The leaves of the *Carica papaya* are green, smell distinctively, and taste quite bitter. Additionally, they have a broad, flat, and strongly palmately lobed shape and are between 50 and 70 cm in diameter and between 18 and 90 cm in length.

PHYSIOCHEMICAL EVALUATION
For the purpose of establishing standards for crude pharmaceuticals, the quantitative
assessment of various pharmacognostic characteristics is helpful. A crucial factor in identifying medication adulteration or inappropriate management is the physical continual inspection of the pharmaceuticals. The drug's moisture content isn't excessive, thus it could prevent the growth of bacteria, fungus, or yeast. The estimation of the ash value and acid-insoluble ash value is crucial in the assessment of crude medicines. The assessment of drug purity, or the presence or absence of foreign inorganic materials such as metallic salts and/or silica, depends heavily on the total ash. The outcomes of the physicochemical parameter study of crude papaya powder are displayed in table below.

<table>
<thead>
<tr>
<th>S.NO</th>
<th>PARAMETER</th>
<th>VALUE (% w/w)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>LOSS ON DRYING</td>
<td>7.28</td>
</tr>
<tr>
<td>2.</td>
<td>TOTAL ASH VALUE</td>
<td>11.2</td>
</tr>
<tr>
<td>3.</td>
<td>SULPHATED ASH VALUE</td>
<td>9.2</td>
</tr>
</tbody>
</table>

PRELIMINARY PHYTOCHEMICALS STUDIES

Initial phytochemical analysis revealed the presence of alkaloids, flavonoids, carbohydrates, proteins, tannins and saponins.

<table>
<thead>
<tr>
<th>S.NO</th>
<th>TEST</th>
<th>Presence /Absence</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>Alkaloids</td>
<td>+</td>
</tr>
<tr>
<td>2.</td>
<td>Flavonoids</td>
<td>+</td>
</tr>
<tr>
<td>3.</td>
<td>Tannins</td>
<td>+</td>
</tr>
<tr>
<td>4.</td>
<td>Glycosides</td>
<td>-</td>
</tr>
<tr>
<td>5.</td>
<td>Steroids</td>
<td>-</td>
</tr>
<tr>
<td>6.</td>
<td>Carbohydrates</td>
<td>+</td>
</tr>
<tr>
<td>7.</td>
<td>Proteins</td>
<td>+</td>
</tr>
<tr>
<td>8.</td>
<td>Saponins</td>
<td>+</td>
</tr>
</tbody>
</table>

RESULT: PRESENT (+), ABSENT (-)

CONCLUSION

*Carica papaya* is prized for its therapeutic properties. As a natural medicament for the treatment of infectious disorders, this is helpful in alternative systems of medicine. Phytochemicals found in the roots, stems, leaves, and fruits of *Carica papaya* L. are a highly rich source of this plant's nutrients. Different portions of plants can be used to extract phytochemicals including, alkaloids, tannins, flavonoids, glycosides, phenolic compounds.

The purpose of the current study was to examine the powdered drug validity as well as the physiochemical and phytochemical makeup of the *Carica papaya* plant. The final results were 7.28% loss on drying, total ash 11.2% and 9.2% Sulphated Ash. The study on
phytochemicals found a variety of alkaloids, flavonoids, carbohydrates, proteins, tannins, and other substances saponins and glycosides.

*Carica papaya* leaf extract improves platelet synthesis by altering gene expression, affecting PTA FR and ALOX-12 genes. This leads to increased megakaryocyte production, boosting platelet production and aggregation in the bloodstream. Carpaine found in *C. papaya* leaves is responsible for this action. The leaf extract also reduces platelet breakdown, inhibits viral reproduction, and has antioxidant and free radical scavenging properties. It also increases platelet synthesis by boosting ALOX-12 and PTFAR activity.

**REFERENCE**


17. O. Oche, A. Rosemary, O. John, E. Chidi, S.M. Rebecca, U.A. Vincent, Chemical constituents and nutrient composition of *Carica papaya* and Vernonia amygdalina leaf

