



## **Exploration of Phytopharmacognostic Study of *Alianthus excelsa* Roxb.(Simaroubaceae) Leaves**

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### **Authors' contributions**

*This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.*

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### **ABSTRACT**

**Aim:** The purpose of this research is to look at the pharmacognostic and phytochemical properties of *Alianthus excelsa* leaves. **Methods and Materials:** The tree *Alianthus excelsa* Roxb. belongs to the Simaroubaceae family and is native to Central and Southern India. The entire methanolic extract of *Alianthus excelsa* leaves was examined for its microscopical, physicochemical, phytochemical, isolation, characterisation, and anti-inflammatory activities. Leaf powder was tested for total ash, water soluble, acid insoluble, alcohol soluble extractive, water soluble extractive, moisture content, and fluorescence property.

**Results:** Carbohydrates, phenolics, flavonoids, alkaloids, and amino acids were found in the leaf methanolic oven dried extract. To determine the existence of phenolic content in extracts, total phenolic and total flavonoid contents were calculated. Phytoconstituents such as flavonoids and saponin glycoside were found in the leaf sections throughout the experiment, which were isolated using column chromatography and characterised using IR, NMR, and mass spectroscopy. Three flavonoids and one flavonoid

**Keywords:** *Alianthus excelsa*; physicochemical; phytochemical; column chromatography.

## 1. INTRODUCTION

During the past decade, the indigenous or traditional system has gained importance in the field of medicine. In most of the developing countries, a large number of populations depend on the traditional practitioners, who are dependent on medicinal plants to meet their primary health care needs. Although, modern medicines are available, herbal medicine retained their image for historical and cultural reasons. Since the usage of these herbal medicines has increased, issues and moto regarding their quality, safety and efficacy in industrialized and developing countries are cropped up [1]. *Ailanthus excelsa* Roxb is a deciduous tree belonging to the family Simaroubaceae and is widely distributed in Asia. Its native origin is China and it is known as 'Tree of Heaven' and used in the Indian system of medicine for variety of purposes.1 It is used in wounds, skin eruption, febrifuge, bronchitis, asthma and in conditions of diarrhoea and dysentery [2,3].

*Ailanthus excelsa* is a type of *Ailanthus*. Roxb is a huge deciduous tree that grows to be 18-25 metres tall with a straight trunk and a diameter of 60-80 cm. The bark is light grey and smooth, but on larger trees it becomes grey-brown and rough, aromatic, and slightly bitter. Leaflets 8-14 pairs, long stalked, ovate or broadly lance shaped from very unequal base, 6-10 cm long, 3-5 cm wide, often curved, long pointed, hairy gland; edges coarsely toothed and often lobed; leaves alternate, pinnately compound, large, 30-60 cm or more in length; leaflets 8-14 pairs, long stalked, ovate or broadly lance shaped from very unequal base, 6-10 cm long, 3-5 cm Pharmacological activities of *A. excelsa* extracts and isolated compounds include considerable antileukemic, [4] antifungal, antibacterial [5,6], and antimalarial [7] activity. Sitosterol, Quassinoids, Ailantic Acid, Vitexin; ailanthione, glalucarubinone, malanthine, excelsin, glaucarubol, 2-6 Dimethoxy- Benzoquinone, and Melanthin [8] were discovered to be present phytochemically. For standardisation and quality assurance, the authenticity, purity, and assay of crude drugs must be cross-checked. We analysed macroscopy, microscopy, physicochemical, and phytochemical properties of leaves in this study.

## 2. MATERIALS AND TECHNIQUES

### 2.1 Plant Material Collection and Authentication

Dr. Shimpi, Taxonomist, Head of Botany Department, G. E. Society's NSC Science College, Nashik India, certified the leaves of *Alianthus excelsa* gathered in the local region of Nashik (Maharashtra, India) in June 2017. For future reference, voucher specimen No. 160 was stored in the herbarium. Formalin (5ml) is used to preserve a portion of the leaves: For histology study, a combination of acetic acid (5ml) and 70% alcohol (90ml) was created, and the remainder was shade dried, powdered, and sieved through a 20 mesh sieve before being stored in an airtight container.

### 2.2 Macroscopic Investigation

The organoleptic evaluation method was used to conduct the macroscopic studies. Leaves were observed for their arrangement, size, shape, base, texture, margin, apex, venation pattern, colour, odour, and taste [9]. As mentioned in the quality control approach, macroscopic and microscopic characteristics were investigated. Photographs were taken using a digital camera at various magnifications.

### 2.3 A Microscopic Examination

Thin pieces of leaf were prepared for microscopic examination. The thin slices were then rinsed in water and stained with safranin, haematoxylin, picric acid, and dil. Iodine solution before being mounted in glycerine for visualization and confirmation of magnifications (10X, 45X).

### 2.4 Preparation of Crude Extract

Filter paper was used to pack 1 kg of powdered leaf sample into thimbles. The sample-filled thimbles were placed in the soxhlet apparatus' cylindrical sample holder and filled with organic solvents such as ethanol and methanol separately. The organic solvents were used to extract plant samples. When organic solvents were combined with plant samples, a coloured solution resulted. Extraction was carried out until the coloured solvent became clear. The temperature of the soxhlet apparatus set up was kept at 65 oC for methanol extracts during the extraction procedure. For each sample, the extraction process lasted around 10 and 12 hours. To obtain a dry residue, the methanol

extract was distilled off under reduced pressure, then dissolved in water and partitioned with distilled water and n-butanol (1:1). The water and butanol fractions were separated, and the aqueous fraction was partitioned 2-3 times with n-butanol.

## 2.5 Fluorescence Analysis

The fluorescence study is an important parameter for crude drug standardisation in the first instance. To evaluate their fluorescence behaviour, the powder medicine of leaves and bark was treated separately with different reagents and subjected to visible and ultraviolet light [10].

## 2.6 Analysis of Physicochemical and Phytochemical Compounds

The importance of physicochemical standards in determining the quality, purity, and adulteration of a given crude medicine cannot be overstated. The foreign matter, LOD, ash, and extractive values were calculated and summarised in a table, and phytoconstituents were identified using a comprehensive preliminary phytochemical screening. It's also useful for determining a crude drug's chemical composition so that it may be properly evaluated. The approach was also used to determine the total phenolic and flavonoid content of the extracts. The total phenol and total flavonoid concentrations were reported in milligramme of gallic acid equivalents/g extract and quercetin equivalents/g extract, respectively, using the mean of three readings [11,12].

## 2.7 Isolation and Characterization of Phytoconstituents from *Alianthus excelsa* Leaf

### 2.7.1 Isolation and characterization of flavonoids

In a Soxhlet apparatus at 40°, the air-dried and powdered roots of *A. excelsa* (1.4 kg) were extracted with 70 percent methanol (MeOH; 23 l). The extract was filtered and then evaporated under decreased pressure to produce methanol extract (125 g). This extract was first chromatographed using benzene, chloroform, ethyl acetate, and MeOH on a silica gel column under gradient conditions (100:0, 80:20, 60:40, 40:60, 20:80, 0:100). The eluted fractions were collected and TLC analysis was performed. Three primary fractions (E1, E2, and E3) were

created by combining similar fractions. Fraction E2 (13.6 g) was re-chromatographed on a silica gel column with a chloroform, ethyl acetate, and MeOH combination (10:0, 8:2, 6:4, 4:6, 2:8, 0:10, gradient) eluting with chloroform, ethyl acetate, and MeOH mixture (10:0, 8:2, 6:4, 4:6, 2:8, 0:10, gradient) These fractions were purified with 100 percent MeOH on Sephadex LH20 to yield compounds 1 (17 mg, 0.0034 percent w/w), 2 (13 mg, 0.0026 percent w/w), and 3 (21 mg, 0.0042 percent w/w), respectively.

### 2.7.2 Isolation and characterization of Saponin glycoside from *Alianthus excelsa* leaf:

To obtain a dry residue, the methanol extract was distilled off under reduced pressure, then dissolved in water and partitioned with distilled water and n-butanol (1:1). The water and butanol fractions were separated, and the aqueous fraction was partitioned 2-3 times with n-butanol. The combined n-butanol fraction was then evaporated and dissolved in methanol under reduced pressure. The methanolic solution was then added drop by drop to ether as a solvent to produce crude saponins precipitates. To obtain crude saponins (10 percent w/w), the solvent was decanted off and the precipitates were dried to a consistent weight at low temperature.

Column chromatography was used to separate and isolate pure saponin glycoside from CS (crude saponin). By gradient elution with Ethyl acetate followed by methanol as a more polar solvent in increasing proportions, a total of 220 fractions were obtained via column chromatography. The saponin contained in the leaves of *Alianthus excelsa* contains both triterpenoidal and steroidal component, according to the results of chemical tests conducted on all fractions. They were collected to generate the final 18 fractions based on their TLC pattern similarity. After spraying with anisaldehyde sulphuric acid (AS) reagent, the saponin compounds tested in HPTLC plates created blue, pink, violet, violet black, and yellowish brown saponin zones. With chloroform: methanol (80:20) as a solvent system and anisaldehyde sulphuric acid reagent as a visualising agent, eluted fractions 73 to 104 revealed the presence of a single spot on TLC. The same was determined to have an R<sub>f</sub> value of 0.36.

### 3. RESULTS AND DISCUSSION

#### 3.1 Macroscopic Feature

A tall deciduous tree with a straight trunk that can reach 30 metres in height. Leaflets 8-14 pairs, long stalked, ovate or broadly lance shaped from very unequal base, 6-10 cm long, 3-5 cm wide, often curved, long pointed, hairy gland, coarsely toothed and often lobed; leaves alternate, pinnately compound, large, 30-60 cm or more in length; leaflets 8-14 pairs, long stalked, ovate or broadly lance shaped from very unequal base, 6-10 cm long, 3-5 cm wide. Flowers many, mostly male and female on different trees, short stalked, greenish-yellow; fruit a 1-seeded samara, lance shaped, flat, pointed at ends, 5 cm long, 1 cm wide, copper, strongly veined and twisted at the base; flower clusters droop at leaf bases, shorter than leaves, much branched; flowers many, mostly male and female on different trees, short stalked, greenish-yellow.

#### 3.2 Microscopical Study

The transverse section of the lamina reveals massive, cuboidal cells of the upper epidermis, which are covered by a thin cuticle. On each epidermis, there were unicellular coverings and glandular trichomes, some of which were filled with cystolithic crystals, as well as paracytic stomata. A bi-layered, dense, radially elongated palisade lies under the upper epidermis, followed by a spongy mesophyll made up of 5-8 layers of loosely distributed parenchymatous cells. Underneath the epidermis, the midrib is made up of well-developed 6-7 layered thick wall collenchymas. Vascular bundles are bicollaterally present at three sites and are surrounded by non-lignified pericyclic fibres in continuous groupings. Above the primary vascular bundles, there are two minor secondary vascular bundles. Ground tissue is made up of polygonal parenchymatous cells that are loaded with calcium oxalate cluster and prism crystals, as well as cystolithic idioblasts (Fig. 1 and Fig. 2).

Uep: Upper epidermis, PF: Pericyclic fiber, G tri: Glandular trichome, Pal: Palisade tissue, VB: vascular bundle, Mxy: Metaxylem, Col: collenchyma, Cal oxa: Calcium oxalate crystals, C tri: covering trichome, Spa: Spongy parenchyma, Lep: Lower epidermis.

#### 3.3 Fluorescence Analysis

Each chemical has a distinct fluorescent colour. When plant materials are treated with various chemicals, they take on a variety of colours. In daylight, some plant components exhibited typical fluorescence in the visual range. Many natural materials that do not glow in daylight exhibit fluorescence when exposed to ultra violet light. When different reagents are added to non-fluorescent substances, they are converted into fluorescent derivatives or breakdown products. Fluorescence analysis was an important metric for pharmacognostic evaluation of crude pharmaceuticals, and it was used to analyse crude drugs qualitatively (Table 1).

When viewed under UV light, the crude medicine fluoresced differently at different wavelengths. This is attributable to the drug's presence of various phytochemical components [15]. Under UV light, flavones that are light yellow in aqueous conditions turn bright yellow in alkaline conditions. When phytosterols are treated with 50% H<sub>2</sub>SO<sub>4</sub>, they flash green under UV light. Under UV light, coumaric acid appears yellowish green in an alkaline state. Under short UV light, terpenoids emit a yellow green fluorescence [16]. Berberin fluoresced with a light yellow colour.

#### 3.4 Physiochemical Analysis

Both physiological and non-physiological ash are included in the total ash value. Physiological ash is created by metabolic processes in the plant, while non-physiological ash is created by contaminants in the environment. Carbonates, phosphates, nitrates, sulphates, chlorides, and silicates of various metals that have been taken up from the soil could be among them. The total ash value was particularly important in determining the purity of medications. The existence of inorganic materials was disclosed by a high percentage of total ash value, whereas the presence of insignificant amounts of siliceous matter was revealed by a very low value of acid insoluble ash. In dilute HCl, the acid insoluble ash was a portion of the total ash that was insoluble. The water soluble ash is composed of the water soluble portion of the total ash [17]. The presence of water soluble ash can be used as a crucial indicator of exhausted material (Table 2)

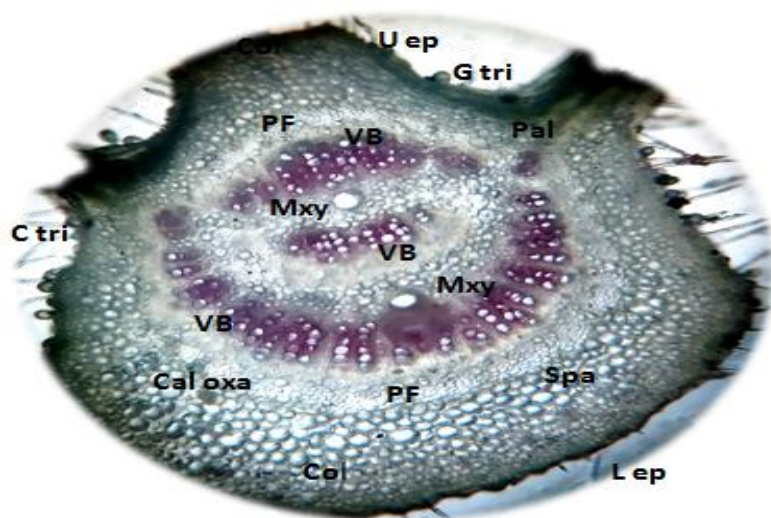
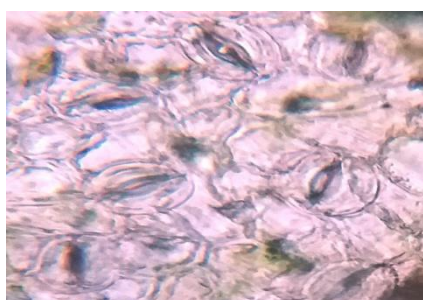
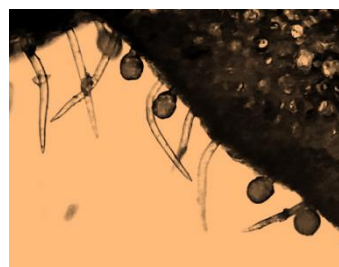


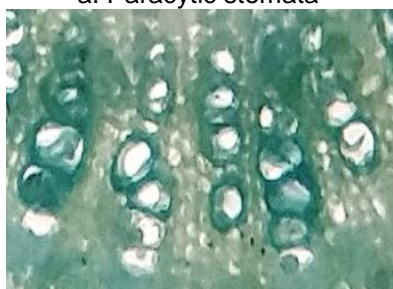
Fig. 1. Transverse section of leaf of *A. excelsa* leaf



a. Paracytic stomata



b. Unicellular covering & glandular stomata



c. Xylem vessels along with phloem



d. Spongy mesophyll

Fig. 2. Magnified view of at 45 X

Table 1. Fluorescence analysis of *Alianthus excelsa* powder

Reagents	Visible light	UV light (365nm)
Plant+1N NaOH Aqueous	Yellow	Bright yellow
Plant+1N NaOH Alcoholic	Light yellow	Bright yellow
Plant+1N Hcl	Light green	Dark green
Plant+H <sub>2</sub> SO <sub>4</sub> (1:1)	Light yellow	Green
Plant+HNO <sub>3</sub> (1:1)	Dark yellow	Yellow green
Plant+Ammonia	Yellow green	Yellow green
Plant+Iodine	Light green	Brown
Plant+5%FeCl <sub>3</sub>	Light green	Brown
Plant+Acetic acid	Light green	Green

**Table 2. Physicochemical evaluation of *Alianthus excelsa* leaves**

Sr. No.	Parameters	Percentage (w/w)
1.	Total ash	9.23
2.	Acid-insoluble ash	3.17
3.	Water soluble ash	4.93
4.	Moisture content	5.6
5.	Extractive values	
	i) Alcohol soluble	10.09
	ii) Water soluble	11.12
	iii) Chloroform soluble	2.9
	iv) Petroleum ether	1.2

**Table 3. Phytochemical analysis of *Alianthus excelsa* extract**

Sr. No.	Chemical constituents	Aqueous extract	Methanol extract	Chloroform extract	Petroleum ether extract
1.	Test for carbohydrates				
	a) Molisch test	+++	+++	---	---
	b) Benedict's test	+++	+++	---	---
	c) Fehling's test	+++	+++	---	---
2.	Test for Alkaloids				
	a) Mayer's test	+++	+++	---	---
	b) Dragandroff's test	+++	+++	---	---
	c) Wagner's test	+++	+++	---	---
	d) Hager's test	+++	---	---	---
3.	Test for Glycoside				
	a) Modified borntrager's test	+++	+++	+++	+++
	b) Legal's test	+++	+++	+++	+++
4.	Test foe saponin				
	a) Froth test	+++	+++	---	---
	b) Foam test	+++	+++	---	---
5.	Test for Phytosterol				
	Salkowski test	---	+++	---	+++
	b) Libermann burched's test	---	+++	---	+++
6.	Test for Phenols				
	a) Ferric chloride test	---	+++	---	---
7.	Test for tannin				
	a) Gelatin test	---	+++	---	---
8.	Test for flavonoids				
	a) Alkaline test	---	++	---	---
	b) Lead acetate test	---	++	---	---
9.	Protein & Amino acids				
	a) Xanthoprotein test	+++	+++	---	---
	b) Ninhydrin test	+++	+++	---	---

Table 4. Absorbance observed in estimation of total phenolic content

Sr. No.	Conc. (µg/ml)	Absorbance (nm)			Mean ± SEM
		I	II	III	
<b>Standard (Gallic acid)</b>					
1	20	0.0750	0.0744	0.0748	0.0746 ± 0.0002
2	40	0.1536	0.1532	0.1553	0.1534 ± 0.0002
3	60	0.2481	0.2484	0.2492	0.2486 ± 0.0001
4	80	0.3172	0.3170	0.3164	0.3168 ± 0.0002
5	100	0.3685	0.3790	0.3780	0.3787 ± 0.0001
<b>Samples</b>					
Methanolic extract	400	0.31740	0.31570	0.31876	0.3172 ± 0.0008

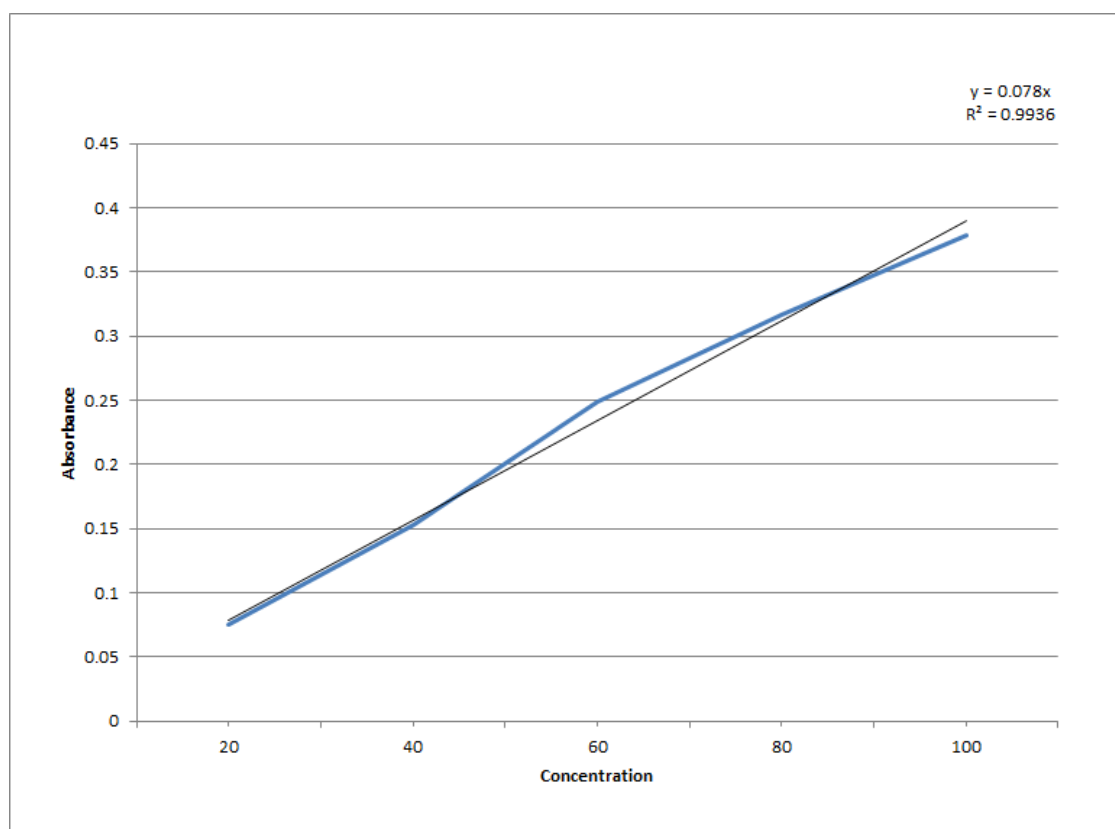


Fig. 3. Showing calibration curve of Gallic acid

Table 5. Absorbance observed in estimation of total flavonoids content

Sr. No.	Conc. (µg/ml)	Absorbance (nm)			Mean ± SEM
		I	II	III	
<b>Standard (Rutin)</b>					
1	20	0.1945	0.1953	0.1949	0.195 ± 0.0002
2	40	0.3888	0.3893	0.3891	0.389 ± 0.0001
3	60	0.5825	0.5826	0.5831	0.583 ± 0.0002
4	80	0.7781	0.7786	0.7788	0.779 ± 0.0002
5	100	0.9327	0.9329	0.9321	0.933 ± 0.0002
<b>Samples</b>					
MeoH extract	400	0.9392	0.9393	0.9392	0.931 ± 0.0002

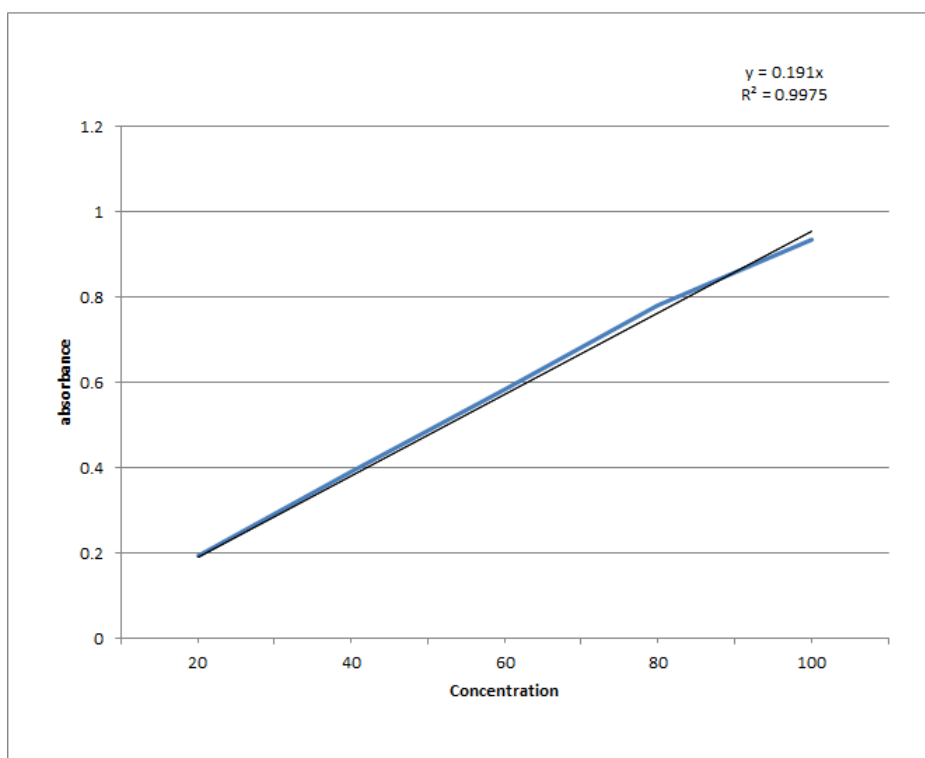


Fig. 4. Showing calibration curve of rutin

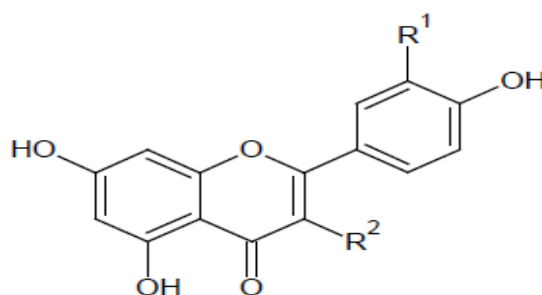


Fig. 5. Structure of flavonoids isolated from *Alianthus excels*

### 3.5 Phytochemical Analysis

Analysis of phytochemicals Because most of the plant's chemical elements dissolved in water and methanol, the percentage yield was much boosted compared to other solvents (Table 3).

### 3.6 Estimation of Total Phenolic Content

The total phenolic compound concentration was estimated using the gallic acid calibration curve equation. Folin-Ciocalteu reagent was used to determine the total phenolic content of the extract. The total phenolic content of *A. excelsa*

leaves methanolic extract was found to be 1.01 percent (Table 4 and figure 3).

### 3.7 Estimation of Total Flavonoid Content

The absorbance of the standard and test samples were mentioned in the table above. On the basis of calibration, the total concentration of flavonoids was estimated. The total flavonoids content of *A. excelsa* methanolic extract was measured using an aluminium chloride colorimetric technique and a rutin standard curve at varied doses, yielding 1.22 percent (Table 5 and Fig. 4).



### 3.8 Isolation and Characterization of Phytoconstituents from *Alianthus excelsa* Leaf

#### 3.8.1 Isolation and characterization of flavonoids

Fraction E2 (13.6 g) was re-chromatographed on a silica gel column with a chloroform, ethyl acetate, and MeOH combination (10:0, 8:2, 6:4, 4:6, 2:8, 0:10, gradient) eluting with chloroform, ethyl acetate, and MeOH mixture (10:0, 8:2, 6:4, 4:6, 2:8, 0:10, gradient) These fractions were purified with 100 percent MeOH over Sephadex LH20, yielding compounds 1 (17 mg, 0.0034 percent w/w), 2 (13 mg, 0.0026 percent w/w), and 3 (21 mg, 0.0042 percent w/w). Elemental analysis, UV, IR, <sup>1</sup>HNMR, <sup>13</sup>CNMR, and MS data, as well as comparisons with existing data, were used to determine the structures of these isolated compounds.

Apigenin: R<sup>1</sup>=H, R<sup>2</sup>=H; Leuteolin: R<sup>1</sup>=OH, R<sup>2</sup>=H; Quercetin: R<sup>1</sup>=OH, R<sup>2</sup>=OH.

Compound (1): elements percent for: (C 50.7, H 33.03, N<0.5) C<sub>15</sub>H<sub>10</sub>O<sub>5</sub> (molecular weight: 270); MS/MS: m/z 220 (M-H); UV<sub>max</sub> (MeOH) nm: 271.2, 347; NaOAc nm: 274.2, 326.8, 392.7; NaOAc+H<sub>3</sub>BO<sub>3</sub> nm: 267.8, 342.2; AlCl<sub>3</sub> nm: 275.6, 303.4, 344.4, 383; AlCl<sub>3</sub>+ HCl nm: 276.6, 302, 342.6; NaOH nm: 276.2, 326, 393.5; IR: 3274 cm<sup>-1</sup>: OH, 1647 cm<sup>-1</sup>: C=O, 1607, 1549 and 1499 cm<sup>-1</sup>: aromatic ring; <sup>1</sup>H NMR (DMSO-d<sub>6</sub>, 400 MHz): δ 12.3, 10.4, 10.6 (1H, s, 5, 7, 4'-OH), 7.7 (2H, d, J 7.2 Hz, H-2'/H-6'), 6.8 (2H, d, J 7.2 Hz, H-3'/H-5'), 6.70 (1H, s, H-3), 6.47 (1H, d, J 2.1 Hz, H-8), 6.5 (1H, J 2.1 Hz, H-6); <sup>13</sup>C NMR (100 MHz, DMSO-d<sub>6</sub>): δ 184.6 (s, C-4), 165.2 (s, C-2), 164.6 (s, C-7), 163.5 (s, C-5), 162.3 (s, C-4'), 156.8 (s, C-9), 127.7 (d, C-2'/C-6'), 123.2 (s, C-1'), 117.8 (d, C-3'/C-5'), 104.6 (s, C-10), 103.7(d, C-3), 97.7 (s, C-6), 94.8 (d, C-8).

Compound (2): elements percentage for: (C 48.09, H 32.76, N<0.5) C<sub>15</sub>H<sub>10</sub>O<sub>6</sub> (mol. wt. 286); MS/MS: m/z 285 (M-H); UV<sub>max</sub> (MeOH) nm: 255.4, 350.2; NaOAc nm: 266.2, 405; NaOAc+H<sub>3</sub>BO<sub>3</sub> nm: 257.9, 374.2; AlCl<sub>3</sub> nm: 273.6, 418.6; AlCl<sub>3</sub>+HCl nm: 265.7, 273.6, 298, 296, 360.6; NaOH nm: 272.2, 406.6; IR: 3117 cm<sup>-1</sup>: -OH, 1661 cm<sup>-1</sup>: C=O, 1616, 1508 and 1448 cm<sup>-1</sup>: aromatic ring; <sup>1</sup>H NMR (DMSO-d<sub>6</sub>, 400 MHz): δ 11.98 (1H, s, 5-OH), 7.49 (1H, dd, J

7.7 Hz, H-6'), 7.38 (1H, d, J 2.2 Hz, H-2'), 6.98(1H, d, J 7.8 Hz, H-5'), 6.72 (1H, s, H-3), 6.40 (1H, d, J 2.2 Hz, H-8), 6.26 (1H, d, J 2.2 Hz, H-6). <sup>13</sup>C NMR (100 MHz, DMSO d<sub>6</sub>): δ 183.4 (C-4), 166.1 (C-2), 162.4 (C-7), 160.8(C-5), 157.5 (C-9), 147.5 (C-4'), 144.5 (C-3'), 122.4 (C-1'), 119.9 (C-6'), 115.7 (C-5'), 111.4 (C-2'), 103.5 (C-10), 102.3 (C-3), 97.9 (C-6), 92.9 (C-8).

Compound (3): elements percent for (C 46.17, H 31.15, N<0.5) C<sub>15</sub>H<sub>10</sub>O<sub>7</sub> (mol. wt. 302); MS/MS: m/z 301 (M-H); UV<sub>max</sub> (MeOH) nm: 257.4, 374.6; NaOAc nm: 276.6, 332.8, 398.9; NaOAc+H<sub>3</sub>BO<sub>3</sub> nm: 274.6, 388; AlCl<sub>3</sub> nm: 276.8, 364, 436.8; AlCl<sub>3</sub>+HCl nm: 236.8, 374, 429S; NaOH nm: 328.5, 429.8; IR: 3278 cm<sup>-1</sup>: -OH, 1662 cm<sup>-1</sup>: C=O, 1612, 1529 and 1450 cm<sup>-1</sup>: aromatic ring; <sup>1</sup>HNMR (DMSO-d<sub>6</sub>, 400 MHz): δ 12.8 (1H, s, 5-OH), 9.3 (1H, s, 4'-OH), 7.78 (1H, d, J 2.1 Hz, H-2'), 7.52 (1H, dd, J 7.4 Hz, H-6'), 6.8 (1H, d, J 7.4 Hz, H-5'), 6.39 (1H, d, J 2.1 Hz, H-8), 6.15 (1H, d, J 2.1 Hz, H-6); <sup>13</sup>CNMR (100 MHz, DMSO-d<sub>6</sub>): d 177.3 (C-4), 163.8 (C-7), 161.8 (C-5), 157.2 (C-9), 146.8(C-3'), 147.6 (C-2), 144.9 (C-4'), 136.8 (C-3), 125.7 (C-1'), 120.9 (C-6'), 114.8 (C-5'), 114.0 (C-2'), 102.0 (C-10), 97.1 (C-6), 92.4(C-8).

#### 3.8.2 Isolation and characterization of Saponin glycoside from *Alianthus excelsa* leaf

The isolated fraction CS-73(Compound 4) was subjected to UV analysis on Shimadzu 2010 instrument & absorption maxima (λ max.) of isolated compounds was found to be 349.5nm which resembles to λ max value of homoanular diene keto steroids. The melting point for the isolated compound CS-73 was determined in open capillaries in an electrothermal melting apparatus & was found to be 205°C respectively.

The results of chemical tests (libermann-Burchard, salkowski, Whitby and trichloroacetic acid test) performed for the isolated compound showed positive results indicating presence of steroidal saponin (Mohammad Ali).

White amorphous powder. m.p.: 272-274° C. <sup>1</sup>H-NMR (500 MHz, CDCl<sub>3</sub>): 0.74, 0.76, 0.90, 0.92, 0.94, 0.97 (each 3H, s, CH<sub>3</sub> x6), 1.12 (3H, s, H-27), 2.78 (1H, dd, J= 3.6, 13.2 Hz, H-18),

3.28 (1H, dd, J= 11.2, 4.4 Hz, H-3), 5.19 (1H, t, J=3.5 Hz, H-12). <sup>13</sup>C-NMR (125 MHz, Pyridined5): δ C (from C-1 to C-30) 36.2, 29.4, 76.4, 38.6, 54.6, 19.6, 34.6, 38.6, 46.4, 38.6, 24.8, 125.4, 143.6, 41.6, 27.6, 22.6, 45.7, 41.0, 46.4, 31.5, 34.6, 32.8, 27.8, 15.8, 15.3, 16.2, 24.6, 181.4, 34.3, 24.7.

#### 4. CONCLUSION

Quality, purity, and sample identification all require standardisation. Preliminary phytochemical experiments revealed that the methanolic extract of *Alianthus excelsa* has more phytoconstituents than the chloroform, petroleum, and aqueous extracts. Because of the presence of multiple functional groups, the drug powder had a variety of fluorescence characteristics. TLC may be useful for identifying and authenticating crude drugs. The goal of this project is to develop certain pharmacognostical criteria. The papers mentioned above provide information on *Alianthus excelsa*'s identification, chemical components, and physical characteristics. These researches aid in the identification and verification of plant material. To achieve reproducibility, proper identification and quality inspection of the starting components are required.

#### CONSENT

It is not applicable.

#### ETHICAL APPROVAL

It is not applicable.

#### COMPETING INTERESTS

Authors have declared that no competing interests exist.

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