# Phytochemical evaluation and TLC Profile of Leaf and bark in *Nyctanthes arbor–tristis* L.

Abstract: This research work mainly deals with the collection of plants, the extraction of active compounds from the bark and leaves. The Introductory phytochemical screening of *Nyctanthes arbor–tristis* L. revealed that about 20 phytochemicals present over the 21 phytochemicals were tested in the various solvent extract (Ethanol, Petroleum ether, Chloroform), with Emodin was totally absent in all kinds of extract which shows the occurrence of 8 phytochemicals were recorded. The chloroform extract shows the presence of 11 phytochemicals were recorded. All the solvent extract exhibits the occurrence of Carbohydrates, Alkaloids, Saponins while Emodin was totally absent in all kinds of extract. The compounds like alkaloids, glycosides, saponins, steroids, flavonoids have expectorant action which is very useful in the management of upper respiratory tract inflammation. During performance of TLC, the extracted solvents used are reducing sequence of polarity there in each one of them extract many of the solvents with their itself polarity rely on the active secondary metabolites containing in the plant. Coloured materials or elements have been seen directly when straight viewed versus the stationary phase even as colourless categories were encountered by spattering the plate with specific reagent which is produced colour-full areas at the regions, which are they immersed [1].

Keywords: Phytochemicals, Medicinal uses, bioactive compounds, TLC profile.

## 1. Introduction

The World Health Organization (WHO) reported by, greater than 80% of the total world population of expanding countries are depends on the traditional medicines that isolated from plant parts for the body. NAT plant normally grows in tropical and subtropical regions of the various countries in over all the world [2]. The numerous phytochemicals are being discovered with proven biological functions. However, the consequences of taking a complete plant as medication are unknown because a single plant contains a large variety of phytochemicals. Furthermore, many plants with medicinal promise still lack comprehensive scientific research to determine their phytochemical components and pharmacological activities [3]. Recently, many researchers have explored the medicinal importance of bioactive phytochemical components of leaf, flower, fruit as well as seed of *Nyctanthes arbortristis* L. [4]. Antibacterial activity of extracts of *Nyctanthes arbortristis* L. prepared with various solvents like chloroform, petroleum ether, butanol, Water, and ethanol were evaluated by agar well diffusion method [5]. It has been attempted for various pharmacological actions such as anti-arthritic, antispasmodic, antibacterial, anti-inflammatory, immunostimulant, anti-diabetic, hepato-protective, antipyretic, anti-allergic and Central Nervous System depressant [6]. That was the prehistoric time no. of plant species were used as phytomedicine for divergent diseases [7]. Osteoarthritis is an acquaint disorder of cartilage deterioration, articular muscle puffiness, bony spurs development, diminish of joint space and intra articular sclerosis [8]. Today's, natural products are responsible with

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regards to partially sanctioned drugs that are now days available [9]. The WHO also supported the use of phytochemical plants for remedial used [10]. Just like that, phytomedicinal plants are far more fascinated in that drug revelation. The phytomedicinal importance of diverge plants sub-sits in its plant derived chemical constituents that build a well define physiological steps in human body [11]. In addition to overdose of minerals than their almost exact quantity daily dose, may generate toxicity in human body [12]. Perilous toxic Phyto metals like Al, As, Hg, Cr, Cd, and Pb can be available in phytomedicinal plants [13]. Today's, people have more alert about the uncertainty companion with the presence of dangerous metals in phytomedicinal plants and their entanglement [14]. The best way of supervision of Osteoarthritis is daily workout and managed diet in routine life. Approximately all body junctures in any way damaged by Osteoarthritis but knee junctures are highly pretentious, traced by the pelvic girdle joints [15]. Osteoarthritis of lower limbs decadence pliability of important organs and cause constraint [16]. The Global grade of Osteoarthritis detailed as the major extensive locomotor system disease in the middle of the globe [17]. It is a most prevalent speculation of joints affliction in approximately or more than 100 million people in the middle of globe having age more than 45 years [18]. and which is more or less than 15% of all locomotory system disorders by WHO centre [19]. The terpenoids presence in phytomedicinal plants were first time proclaimed [20]. It is indispensable by virtue of their correspondence with necessary compounds like vitamin A and could be an enormous medical demand [21].

**List 1. Taxonomic Classification:** According to APG–IV (Angiosperm Phylogeny Group IV system) 2016.

| | |
|---|---|
| Kingdom | Plantae |
| Clade I | Tracheophytes |
| Clade II | Angiosperms |
| C | |
| C | |
| C | |
| F | |
| C | |
| S | - |

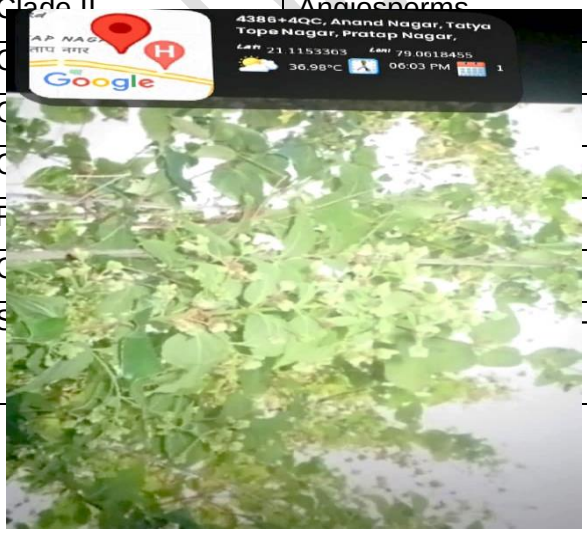


Image-1. Location for plant sample collection

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## 2.Materials and methods

**Plant material**: *Nyctanthese arbor–tristis* L. leaves and stem bark were collected from the wild forests of Wardha and Nagpur District (Latitude 21.1153363 and Longitude 79.0618455). The plants were collected and was authenticated at the Department of Botany, Bajaj College of science, Wardha. The plant material was processed to prevent the deterioration of secondary metabolites be available in the samples. The NAT plant collected materials washed and cleaned by distilled water, followed by peeling or striping leaves from the stem and desiccate at room (acceptable) temperature up to four weeks to remove moisture content. The dry sample was crushed into powder using mortar and pestle. The dry crushed sample was stored for further analysis.

### A. Preparation of plant extraction

The powder sample of plant material (50/500ml) was extracted successively with ethanol, petroleum ether and chloroform. Allow the mixture to soak for 24 to 48 hours at room temperature, shaking occasionally to ensure through mixing. After soaking, filter the mixture using Whatman's filter paper. The extract was concentrated using a rotary evaporator to recollect the solvent. The extract can be further dried to obtained solid extract by evaporating the remaining solvent completely. Stored the concentrated extract in clean, air tights container in a refrigerator at 4°C.

### B. Phytochemical analysis

The phytochemical test for various phytochemicals presents in the extract was carried out using standard methods.

### I. Carbohydrate Test

**Molisch's test**: 2ml of the extract taken with 2 drops of alcoholic solution of α-napthol was added and after mixture properly well shaken further, few drops of conc. $H_2SO_4$ was added at the edge of the sides of the test tube. The violet ring shows to indicates the sugar is present in the extract.

**II. Protein Test: Biuret test**: Up to 2ml of filtrate was taken to and 1 drop of 2% $CuSO_4$ solution was added after that 1ml of 95% $CH_3CH_2OH$ was added. Then it was continuous by excess added of KOH. The pink colour appearance which indicates that the protein is confirm.

### III. Amino acids Test:

**Ninhydrin test:** Take 0.5mg of extract with 2 drops of freshly prepared 0.2 Ninhydrin reagents were added and heated. The appearance of purple or pink colour which indicates that the presence of protein, and amino acid in the extract.

### IV. Alkaloid Test:

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**1. Mayer's test**: Take few ml of the filtrates of extract and drop of Mayers reagent ware added at the edge of the test tube. A creamy or white precipitate that means test is positive.

**2. The Wagner's test (Iodine-potassium iodine reagent)** Up to 2ml of extract with few drops of Wagner's reagent the formation of Reddish-brown precipitate which indicates that the alkaloids are present.

**V. Glycosides Test:**

**Borntrager's test**: Take 2ml of filtrate and add 3 ml of chloroform with well shake for few minutes. Then chloroform layer had separated after that 10% ammonium solution were added into it. Pink colour indicates that the glycosides present.

**VI. Cardiac glycosides**

**Test (Keller Killani test)**: Up to 5ml of extract mixed to 2ml of glacial acetic acid with a drop of ferric chloride sol$^{n}$. were added continuously by the add$^{n}$. of 1ml of concentrated $H_2SO_4$. The brown ring in the interface indicates that the presence of deoxy sugars of cardenoloides in the compound. The violet ring appeared below the brown ring whilst acetic acid layer a (green ring) form just side by side approach the layer in test tube.

**VII. Phenol Test**

**Gelatine test**: Up to 5ml extract, 2ml of 1% solution of gelatine containing 10% of NaCl was added. Appearance of white precipitate which indicates that phenol is present.

**VIII. Tannins Test**:

**Ferric chloride test:** The tannin present in the extract were done by taking 5ml of extract in a test tube side by side added a drop of 0.1% Ferric chloride solution. A bluish black colour / brownish green precipitation which indicates that tannin is present.

**IX. Flavonoids Test**: This Methods was used to determine the flavonoids present in the plant sample or extract [22]. 5 ml diluted ammonium sol$^{n}$. were added to a portion of the aqueous filtrate of each plant extract continuously by add$^{n}$. of concentrated H2SO4. A yellow colouration observed in sample of each extract which indicated that the flavonoids are present. After few minutes yellow colouration was disappeared at standing position.

**X. Steroids Test**:

**Salkowski test:** Up to 10 ml of chloroform were added in 1ml of each sample extract in the test tube. After that 10ml concentrated sulphuric acid was dissolved in this test tube. 2 layers were formed. The lowering layer shows yellow colour as supports with green colour fluorescence even as uppermost layer showing reddish colour. The formation of these two layers which indicates that the steroids were present.

**B. Thin layer chromatography:**

Chromatographical representations were performed on Thin Layer Chromatography Silica Gel 60F254, Aluminium Sheets of Size 6.5 cm x 5 cm (Merck, Germany). The Aqueous and Methanol Extracts of *Nyctanthes arbor–tristis* L. Were Resuspended in Respective Solvents at a Concentration of 100 mg/ml and used for TLC Analysis. The Extracts of 10 µL were

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Manually Applied to the Plate as Spot Using the Hamilton 50 µL Syringe, Positioned 1cm from the Bottom and 1.5 cm from Side of the Plate, On Each Plate with Four Applications. The Space Between Two Spot was 1.5 cm. The Spotted TLC Plates were Subjected to Development in the TLC Developing Glass Chamber Pre-saturated with Different Solvent as Mobile phase. The Developing Distance was 80mm and the developed plate was removed from the chamber and dried over the hot plate for the Evaporation of Solvents used as Mobile Phase. The TLC Plates were Transferred into the Mobile Phase Consisting of Numerous Blending of Solvent Systems of Different Polarity such as $CHCl+_{3,}\ C_6H_{14,}\ C_2H_5OH$ (4:2:4), $CHCl_{3,}\ C_6H_{14,}\ H_2O$, PE, $C_4H_8O_2$ (4:2:2:4), PE, $CHCl_{3,}\ C_6H_{14}$ (4:3:3) and permit to move on the receptive adsorbent silica gel. The consequent spots were observed under UV visible light and spraying by iodine reagent stain. The compute of the distance a compound travelled is considered as the retention factor ($R_f$), which was evaluated by using the following TLC formula: The Different Spots were Developed in each Solvent System which was Identified by means of Post-development Derivatisation with Different Spraying Agents like Iodine, Ammonia and Dragendorff's Reagents.

**Retention factor ($R_f$) values was determined by following formula:**

$$\mathbf{R}_f = \frac{\textit{Distance travelled by solute (cm)}}{\textit{Distance travelled by solvent (cm)}}$$

**Result and Discussion:**

**The following are the results obtained.**

**Table I Presence (+Ve) or Absence (–Ve) of Primary and Secondary metabolites with Different solvents in the Extract of *Nyctanthes arbor –tristis* L.**

| Sr. No. | Name of sample | Ethanol | Petroleum Ether | Chloroform |
|---|---|---|---|---|
| I | Carbohydrate | +ve | +ve | +ve |
| II | Amino acid | –ve | +ve | –ve |
| III | Protein | –ve | +ve | –ve |
| IV | Steroids | –ve | +ve | –ve |
| V | Alkaloids | +ve | +ve | +ve |
| VI | Flavonoids | –ve | +ve | –ve |
| VII | Glycosides | –ve | –ve | +ve |
| VIII | Cardiac glycosides | +ve | –ve | +ve |
| IX | Tannins | –ve | +ve | +ve |
| X | Phenols | –ve | +ve | +ve |

The Successful evaluation of botanical phytocompounds from plant sample was largely dependent on the type of solvent were used in the extraction procedure. Today's phytochemical study on the plant of Nyctanthes arbor-tristis using different solvent containing extracts betray the alkaloids, glycosides, flavonoids, phenols, saponins, steroids, amino acids, tannins, terpenoids, quinones, anthraquinones and coumarin was present. Next to ethanol and showed the presence of rich variety of secondary metabolites. The results revealed that *Nyctanthes arbor–tristis* L. leaves and Bark as a rich source of bioactive compounds. These findings suggested that *Nyctanthes arbor–tristis* L. leaves and Bark have a potential source of natural, antioxidant which have great importance as therapeutic agent for many chronic diseases. In addition, the Ethyl Acetate, Petroleum Ether, Chloroform as well as ethanol extracts for *Nyctanthes arbor–tristis* L. leaves and Bark contain a higher content of bioactive compounds, which will be used for future research on this plant.

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**Table II: Phytochemical evaluation and TLC Profile Perform by using chromatographic Rf values of different solvent extracts of leaves and Bark in *Nyctanthes arbor–tristis* L.**

| Extract | Solvent System | Normal slide Number of spots | $R_f$ values | UV light Number of spots | $R_f$ values | Iodine and Dragendorff" s Reagent, Ammonia Number of spots | $R_f$ values |
|---|---|---|---|---|---|---|---|
| **NATCE** | $CHCL_3$, $C_6H_{14}$, $C_2H_5OH$ (4:2:4) | 1 green, 1 yellow | 0.231, 0.136 | 2 light green, 1 dark green, 1 yellow green | 0.316, 0.366, 0.533, 0.716 | 1 light green, 1 dark yellow, 1 brown, 1dark green | 0.316, 0.312, 0.716, 0.366 |
| **NATWE** | | 1 yellow | 0.312 | 1 dark yellow, 1 brown | 0.483, 0.716 | 1 light green, 1 light yellow | 0.316, 0.76 |
| **NATPEE** | | 2 green, 2 yellow | 0.423, 0.321, 0.521, 0.211 | 2 dark green, 3 dark yellow, 1 light yellow | 0.266, 0.35, 0.483, 0.65, 0.716, 0.75 | 1 dark green, 2 brown, 1 light yellow, 1 dark yellow | 0.312, 0.713, 0.716, 0.76, 0.483 |
| **NATEAE** | | 1 brown, 2 yellow | 0.412, 0.421, 0.321, 0.134 | 2 dark yellow, 2 dark green, 1 brown | 0.216, 0.516, 0.55, 0.683, 0.716 | 1 dark green, 1 light green, 1 dark yellow | 0.316, 0.366, 0.438 |
| **NATEE** | | 1 brown, 1 yellow | 0.416, 0.612 | 1 dark green, 1 dark yellow, 1 light yellow, 1 brown | 0.2, 0.35, 0.566, 0.783 | 1 light green, 1 dark green, 1 orange and brown | 0.345, 0.232, 0.266 |
| **NATCE** | $CHCL_3$, $C_6H_{14}$, $H_2O$, PE, $C_4H_8O_2$ (4:2:2:4) | 1 green, 1 yellow, 1 brown | 0.352, 0.132, 0.326 | 2 dark yellow, 1 dark yellow, 2 brown | 0.15, 0.266, 0.4, 0.566, 0.7 | 1 dark blue green, 1 dark yellow, 1 light green, 1 light blue | 0.766, 0.483, 0.283, 0.416 |
| **NATWE** | | 1 brown | 0.423 | 1 dark yellow, 1 light yellow, 1 light green | 0.283, 0.483, 0.766 | 1 dark blue, 1 light green, 1 dark yellow | 0.25, 0.7, 0.483 |
| **NATPEE** | | 2 yellow, 1 brown, 1 green | 0.521, 0.412, 0.324, 0.231 | 2 dark green, 1 light green, 1 dark green, 1 brown | 0.266, 0.416, 0.616, 0.8, 0.816 | 1 dark green, 1 dark yellow, 1 light yellow, 1 light green | 0.083, 0.116, 0.683, 0.516 |
| **NATEAE** | | 2 yellow, 1 brown, 2 green | 0.116, 0.45, 0.321, 0.126 | 2 dark green, 1 dark green, 1 light green, 2 brown | 0.083, 0.25, 0.383, 0.55, 0.633, 0.683 | 1 blue, 1 dark yellow, 1 light yellow, 1 light green | 0.683, 0.283, 0.15, 0.65 |
| **NATEE** | | 1 green, 1 yellow, 1 brown | 0.241, 0.512, 0.116 | 1 dark green, 1 light yellow, 2 light green, 1 brown | 0.283, 0.483, 0.6, 0.7 | 1 dark green, 1 brown, 1 dark yellow | 0.433, 0.733 |

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|---|---|---|---|---|---|---|---|
| **NATCE** | PE, $CHCL_3$, $C_6H_{14}$ (4:3:3) | 1 yellow, 1 brown | 0.321, 0.514 | 1 light green, 1 dark yellow, 1 brown | 0.116, 0.516, 0.683 | 1 blue, 1 light green, 1 dark green, 2 dark yellow | 0.516, 0.866, 0.733, 0.65, 0.35 |
| **NATWE** | | 1 brown | 0.341 | 1 dark green, 1 dark yellow | 0.283, 0.483 | 1 brown, 1 dark green, 1 dark yellow | 0.55, 0.65, 0.383 |
| **NATPEE** | | 1 yellow, 1 green | 0.534, 0.162 | 1 light green, 1 dark green, 2 dark yellow | 0.15, 0.35, 0.483, 0.65 | 1 light blue, 1 light green, 1 dark green, 2 dark yellow, 1 brown | 0.25, 0.266, 0.283, 0.6, 0.7, 0.15 |
| **NATEAE** | | 2 yellow, 1 green, 1 brown | 0.465, 0.112, 0.116, 0.341 | 2 dark green, 1 light green, 1 dark yellow, 2 brown | 0.266, 0.35, 0.433, 0.516, 0.733, 0.866 | 1 dark green, 1 brown, 2 dark yellow | 0.283, 0.416, 0.8, 0.633 |
| **NATEE** | | 1 green, 1 yellow, 1 brown | 0.172, 0.342, 0.521 | 2 dark yellow, 1 dark green, 1 light green, 1 brown | 0.25, 0.35, 0.383, 0.483, 0.65 | 1 brown, 1 dark yellow, 1 dark green | 0.483, 0.616, 0.55 |

The Introductory phytochemical screening of *Nyctanthes arbor–tristis* L. revealed that about 20 phytochemicals present over the 21 phytochemicals were tested in the various solvent extract (Ethanol, Petroleum ether, Chloroform), with Emodins was totally absent in all kinds of extract which shows the occurrence of 8 phytochemicals which includes Carbohydrate, Amino acid, Protein, Steroid, Alkaloid, Saponin, Tannin, Terpenoids, Triterpenes, Fatty acids, Resins, Quinones. The chloroform extract shows the presence of 11 phytochemicals which includes, carbohydrate, Alkaloid, Glycoside, Cardiac glycoside, Saponins, Tannin, Fatty acid, Coumarins, Phenol, Quinone, Resin. All the solvent extract exhibits the occurrence of Carbohydrates, Alkaloids, Saponins while Emodins was totally absent in all kinds of extract (Table-I). TLC is used in separating various phytochemicals based on their polarity and interaction with the stationary or the standing phase and the moving or mobile phase. Different classes of compounds in *Nyctanthes arbor–tristis* L. such as Alkaloid, Flavonoids, Tannins, Saponins, Terpenoids and Phenolic compounds which exhibit different $R_f$ values. The $R_f$ values are used to characterize the different phytochemical compounds which are present in the extracts. The phytochemical compounds will be obtained by different Rf values, due to their polarity. The polar compound will have strong interaction with stationary phase on TLC and travel shorter path. Meanwhile, the non-polar compounds have weaker interaction with stationary phase and travel longer path. The plates below show the developed chromatogram which resulted from the various solvent extracts and the $R_f$ values of the constituents separated (table-II).

The indispensable class of were clearly identified on chromatogram. The different coloured spots like dark green colour indicated that the presence of tannin, dark yellow colour signify the occurrence of flavonoids, brown colour signify the occurrence of alkaloids, light yellow green colour signify the occurrence of steroids, blue green colour signify the occurrence of phytosterols (Image-2), this clearly indicates that the most necessary secondary metabolites or phytochemicals are found in the bark and leaves extract in *Nyctanthes arbor–tristis* L. The compounds like alkaloids, glycosides, saponins, steroids, flavonoids Saponins, a special class of glycosides (Image-3 and 4), have expectorant action which is very useful in the management of upper respiratory tract inflammation. The extracted solvents used are reducing sequence of polarity there in each one of them extract many of the solvents with

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their itself polarity rely on the active secondary metabolites containing in the plant. Coloured materials or elements have been seen directly when straight viewed versus the stationary phase even as colourless categories were encountered by spattering the plate with specific reagent which is produced colour-full areas at the regions, which are they immersed [23]. The following spraying systems were used. The Alkaloid present in the sample were detected by spraying the freshly prepared Dragendorff" s reagent, Ammonia, and Iodine on TLC plates.

A positive reaction in the chromatogram (orange and brown) was confirmatory evidence that the alkaloid was present in the extract [24]. The existence of flavonoid was detected by the emergence of colour in the plate a positive reaction was emergence of yellow colour spot by exposure of ammonia [25]. On that basis experimental Pretend the Alkaloid, Tannins and Saponins contents are responsible for its antibacterial activity [26]. The occurrence of Phenolic group in the plants keep safe them from Microbial attack, Insect and Herbivores damages [27]. The number of active chemical compounds also contain other functional characteristic features like Anti-inflammatory, Antimutagenic, Hypocholestemic and Antiplatelet Aggregation Properties [28]. Such phytochemical necessary compounds carried out their specific activity by Combining with Protein, Lipids or any other Components of the Bacterial Cell Membrane which are associated to many indispensable physiological functions by there, disrupting the characters as well as functional behaviour of the cell membrane [29]. Cardiac Glycosides is also Occurs to useful in treatment of Heart failure and Supraventricular Arrhythmias [30].

The necessary class of Phytoconstituents known as Cardiac Glycosides have very important role in Medicine because their proper actions on Heart and used in Cardiac Insufficiency [31]. It is a particular action helps in the treatment of Congestive Heart Failure [32]. Moreover, Glycosides, Flavonoids and Tannins have Hypoglycaemic Activities [33]. Saponins is a best class of compound which is an Active Constituents with a marked Hormonal Activity, support in the Absorption of Nutrients [34]. The phytochemicals have unique properties of Precipitating and Coagulating Red Blood Cells, Binding Cholesterol, Formation of Foams in Aqueous Solutions and Hemolytic Activity [35]. Steroids filled important Roles at Different Stages of Mammalian Development Comprising Prenatal Development, Growth, Reproduction, Sexual and Social Behaviour [36]. Steroids had been Reported to have Antibacterial Properties [37].

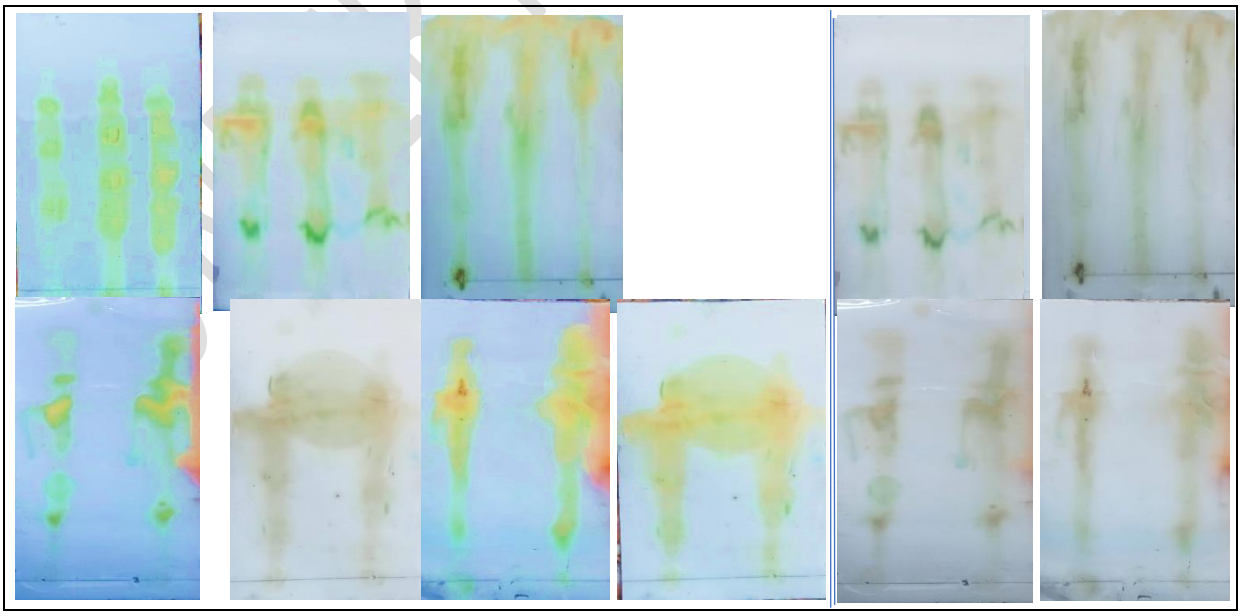

Image 2: TLC profile of *Nyctanthes arbor–tristis* L. Different solvent systems examined under UV light to find phytochemicals found in the extracts. (A) $CHCL_3$, $C_6H_{14}$, $C_2H_5OH$ (4:2:4) B) $CHCL_3$, $C_6H_{14}$, $H_2O$, PE, $C_4H_8O_2$ (4:2:2:4) (C) PE, $CHCL_3$, $C_6H_{14}$ (4:3:3) NATCE: *Nyctanthes arbor–tristis* L. Chloroform Extract, NATWE: *Nyctanthes arbor–tristis* L. Water Extract, NATPEE: *Nyctanthes arbor–tristis* L. Petroleum Ether Extract, NATEAE: *Nyctanthes arbor–tristis* L. Ethyl Acetate Extract, NATEE: *Nyctanthes arbor–tristis* L. Ethanol Extract.

Image 3: TLC profile of *Nyctanthes arbor–tristis* L. Different solvent systems shows visualised spots to find phytochemicals found in the extracts. (A) $CHCL_3$, $C_6H_{14}$, $C_2H_5OH$ (4:2:4) B) $CHCL_3$, $C_6H_{14}$, $H_2O$, PE, $C_4H_8O_2$ (4:2:2:4) (C) PE, $CHCL_3$, $C_6H_{14}$ (4:3:3) NATCE: *Nyctanthes arbor–tristis* L. Chloroform Extract, NATWE: *Nyctanthes arbor–tristis* L. Water Extract, NATPEE: *Nyctanthes arbor–tristis* L. Petroleum Ether Extract, NATEAE: *Nyctanthes arbor–tristis* L. Ethyl Acetate Extract, NATEE: *Nyctanthes arbor–tristis* L. Ethanol Extract.

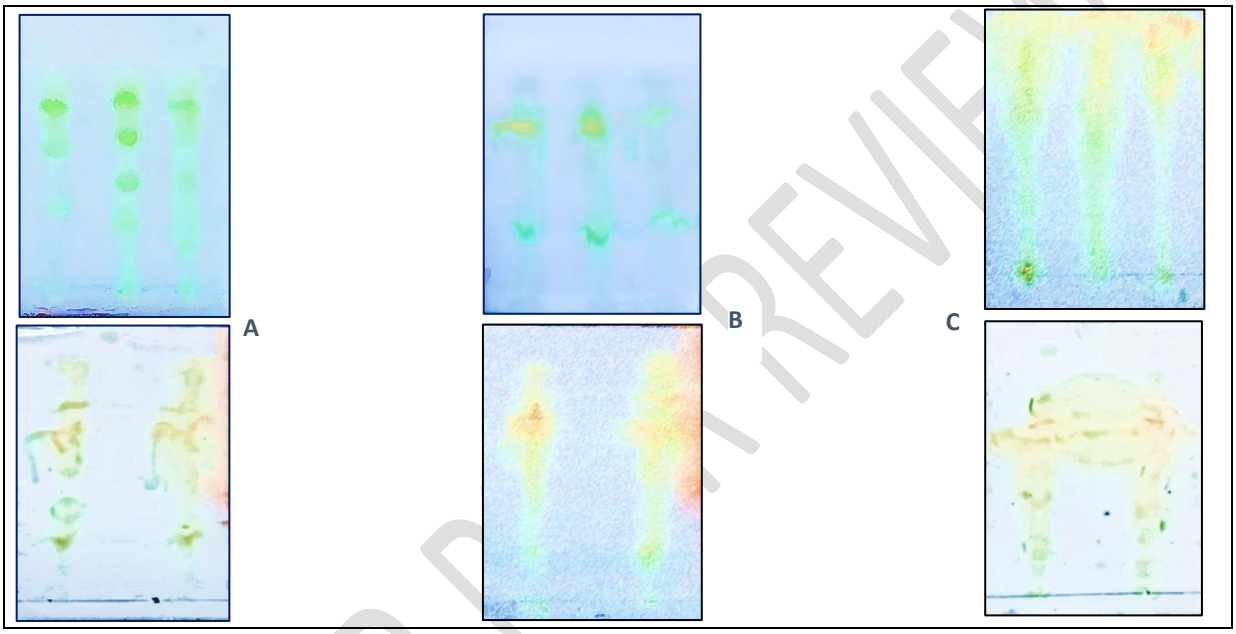


Image 4: TLC profile of *Nyctanthes arbor–tristis* L. Different solvent systems examined by spraying Iodine reagent to find phytochemicals present in the extracts. (A) $CHCL_3$, $C_6H_{14}$, $C_2H_5OH$ (4:2:4) B) $CHCL_3$, $C_6H_{14}$, $H_2O$, PE, $C_4H_8O_2$ (4:2:2:4) (C) PE, $CHCL_3$, $C_6H_{14}$ (4:3:3) NATCE: *Nyctanthes arbor–tristis* L. Chloroform Extract, NATWE: *Nyctanthes arbor–tristis* L. Water Extract, NATPEE: *Nyctanthes arbor–tristis* L. Petroleum Ether Extract, NATEAE: *Nyctanthes arbor–tristis* L. Ethyl Acetate Extract, NATEE: *Nyctanthes arbor–tristis* L. Ethanol Extract.

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**Conclusion:**

This study of Phytochemical screening of Petroleum ether and chloroform extracts of *Nyctanthes arbortristis* L. revealed more concentrated phytochemicals in Petroleum ether and chloroform extracts when compared to that of ethanol extracts of the plant during which was formed during various phytochemical tests by using standard protocol. When the actual demonstration was done of different phytochemicals in different solvents by using Thin Layer Chromatography which revealed that every phytochemical showed different absorption rate in different solvent to formed different $R_f$ values and based on the Rf values calculations was found that the phytochemicals is present in plants which was shown above. It has been

analysed and conclude that the plant containing phytochemicals present in different concentrations in plants based on their rate of absorption on TLC plates by using different reagents to forms a cleared coloured spots at different solvent system concentration. This Study Confirmed that the use of *Nyctanthes arbortristis* L. Plant Material Supply for the pharmaceutical industry and demonstrated a suitable control of many biochemical and physiological activities like anti-bacterial, anti-arthritic, anti-malarial as well as anti-cancerous.

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