Preliminary phytochemical screening and thin layer chromatography of selected extract of Moringa oleifera leaf

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Abstract
Moringa oleifera commonly known as Drumstrick, grows in different countries of the tropics and subtropics regions. It is a vibrant affordable source of phytochemicals and has high nutritive and medicinal value. Every part of plant contains beneficial nutritive components. It is rich source of calcium, magnesium, Zinc, potassium, iron, and copper.

Materials and Methods: The present study, aims to conduct preliminary phytochemical analysis by biochemical test and to analyze thin layer chromatography (TLC) of different polarity solvent extracts.

Results: Qualitative phytochemical analysis of Moringa leaves shows the presence of flavonoid, polyphenol, terpenoids, glycosides, steroid, alkaloids, saponin and tannins in Methanol, Ethanol, Petroleum ether solvent of plant extract. TLC analysis was carried out using different polarity extracts solvents reveal the presences of different colour compounds.

Conclusions: Thus The results obtained in the present study indicates that Moringa oleifera plants extracts contain various phytochemical which are medicinal importance.

Keywords: Moringa oleifera, Bioactive compound, Retention factor, Thin layer chromatography, Soxhlet extraction

Introduction
Herbal drugs are used since ancient time to cure diseases due to presence of phytochemical compound. Phytochemicals are naturally present in the different parts of medicinal plant and have potential to kill or static growth of infectious pathogen. Secondary metabolites are the byproducts of primary metabolism and are synthesized in large variety which include alkaloids, steroids, flavonoids, terpenoids, glycoside, saponia, tannis, phenolic compounds etc. [1]. Therefore basic phytochemical investigation, identification and isolation of bioactive compounds provides evidence for scientific data of traditional use of medicinal plant [2].

Moringa oleifera is a small, fast-growing evergreen or deciduous tree that usually grows up to 10 to 12m in its height, open crown of drooping fragile branches, feathery foliage of trip innate leaves and thick corky, whitish bark [3]. Moringa oleifera Lam is used as a highly nutritive vegetable in many countries. Its young leaves, flowers, seeds and tender pods are commonly consumed and they are having some medicinal properties. Different parts of this plant are being employed for the treatment of various ailments in the indigenous system of medicine.(4-6) It possesses antitumor, antipyretic, analgesic [7] antiepileptic, anti-inflammatory, antilucre, antispasmodic, diuretic, antihypertensive, cholesterol lowering, antioxidant, antiobiotic, renal [5, 8] and hepatoprotective activities [9]. Moringa oleifera has traditionally been used in the treatment of malaria, parasitic diseases, skin diseases, hypertension and diabetes. Three phenol derivatives, quercetin showed antibacterial activity, Niazinin-A and Stigmasterol also exhibit antibacterial activity. In vitro studies also showed that flavonoids isolated from M.oleifera exhibit considerable antimicrobial activity. Its leaves has low calorific value and used in the diet of obese [11]. The Moringa plant provides a rich and rare combination of zeatin, quercetin, Kaempferom and many other phytochemicals. (Sharma)

Therefore, the aims and objectives of this research is to determine the phytochemical constituents of the leaf extracts of Moringa oleifera as well as separation, isolation and identification of phytochemical having bioactivity.
Material and Method
Experimental plant
*Moringa oleifera* leaves were collected botanical garden of A.N. College, Patna, Bihar, India

Preparation of plant extract
Leaves were collected, thoroughly washed in tap water, shade dried, powered by grinder. 50 g of powered weigh and extracted using methanol, ethanol, petroleum ether solvents through soxhlet extraction. The extract was dried at low room temperature under pressure in a rotary vacuum evaporator [9]. The extracts were collected as fine powered and then subjected to qualitative phytochemical screening and TLC analysis studies [14]. The dried extract was properly stored in the refrigerator at 4 °C for further investigation and experimentation.

Phytochemical screening
Experiments were conducted for the screening and identification of eight phytoconstituent present in the *Moringa oleifera* leaf and study were carried out in Methanol, Ethanol, Petroleum ether extracts by using the standard procedure described by [15, 18].

Thin layer chromatographic studies
Each solvent extract was placed to silica gel plates for thin layer chromatography (TLC) analysis. Plate were marked and glass capillaries were used to place the sample. The TLC plates were placed in camber containing Chloroform: Methanol: H2O (7:3:1) solvent (mobile phase) and leave for 20 min to develop bands and was visualized under UV rays [20]. After the solvent running plates are dried and sprayed iodine reagents to detect the bands on the TLC plates. The movement of the separated compound was expressed by its retention factor (Rf), values were calculated by formula. Rf = Distance traveled by the solvent front / Distance traveled by the solvent.

Result and Discussion

Phytochemical screening
The present study of phytochemical screening of *Moringa oleifera* leaves reveals the presence of active phytoconstituents. The active phytochemical compounds such as alkaloid, flavonoid, steroid, terpenoid, saponins, glycosides, polyphenols and tannins are major compound to be investigated and the results are presented in table 1. In these screening process alkaloids, saponins, polyphenol, tannins, glycosides, flavanoids and terpenoids shows different types of results in different solvents extracts. Among these phytochemical screening, Alkaloids, steroid, glycosides, polyphenol and tannin were present in all three solvent. Flavanoid is found absent in Methanol solvent whereas terpenoids and saponin were absent in petroleum ether solvent. All the eight active compound are found present in ethanol solvent. The phytochemical such as alkaloid, Flavonoid, saponin are good agent of Anti-oxidant and act as anti bacterial, antiviral, anti tumor. The result of qualitative analysis of phytochemical presented in *Moringa oleifera* leaf depicted in respective table shows that leaves are rich in phytochemical that have antioxidant activities and several medicinal value [13].

Thin layer chromatographic studies
TLC analysis of selected solvent (Methanol, Ethanol, Petroleum ether) extract of *Moringa oleifera* leaf was carried out to separate and isolate bioactive compound present in extract. The result of present study indicated in Table 2. And TLC plate run picture is shown in fig 1. Solvent used for Thin layer chromatographic studies of *Moringa oleifera* leaf is Chloroform: Methanol: H2O (7:3:1) for all three extract. TLC analysis of the methanol extract, reveals the presence of 5 spots with Rf values 0.26, 0.31, 0.51, 0.73, 0.91. Solvent run was 9.2 cm. TLC of ethanolic extract, reveals the presence of five compounds having Rf value are 0.35, 0.56, 0.77 and 0.96 respectively. Solvent run of ethanolic extract was 9.0 cm. TLC of petroleum ether extract reveals the presence of five compound having Rf value 0.25, 0.33, 0.58, 0.72 and 0.97 with different colour spectrum. Solvent run for petroleum ether was 9.0cm. Swathi S. (2016) studies Phytochemical screening and TLC studies of *Moringa oleifera* extract: their antibacterial and antioxidant activities indicated the different colour band of leaf extract produce in different solvent and shows similar result of TLC.

<table>
<thead>
<tr>
<th>S. No.</th>
<th>Phyto-Chemicals</th>
<th>Methanol</th>
<th>Ethanol</th>
<th>Petroleum Ether</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Alkaloids</td>
<td>+</td>
<td>-</td>
<td>+</td>
</tr>
<tr>
<td>2</td>
<td>Flavonoids</td>
<td>-</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>3</td>
<td>Glycosides</td>
<td>+</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>4</td>
<td>Steroids</td>
<td>+</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>5</td>
<td>Polyphenol</td>
<td>+</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td>6</td>
<td>Terpenoids</td>
<td>+</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td>7</td>
<td>Saponins</td>
<td>+</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td>8</td>
<td>Tannin</td>
<td>+</td>
<td>+</td>
<td>+</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Extract</th>
<th>Solvent taken</th>
<th>Solvent Run</th>
<th>Peaks Obtained(cm)</th>
<th>Peak Colour</th>
<th>Rf Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Methanol</td>
<td>Chloroform: Methanol: H2O (7:3:1)</td>
<td>9, 2</td>
<td>2.4, 2.9, 4.7, 6.8, 8.4</td>
<td>Light Brown, Brown, Green, Yellow, Light Blue</td>
<td>0.26, 0.31, 0.51, 0.73, 0.91</td>
</tr>
<tr>
<td>Ethanol</td>
<td>Chloroform: Methanol: H2O (7:3:1)</td>
<td>9</td>
<td>2.6, 3.2, 5.1, 7.0, 8.7</td>
<td>Light Brown, Brown, Green, Yellow, Light Blue</td>
<td>0.28, 0.35, 0.56, 0.77, 0.96</td>
</tr>
<tr>
<td>Petroleum ether</td>
<td>Chloroform: Methanol: H2O (7:3:1)</td>
<td>9</td>
<td>2.3, 3.0, 5.3, 6.5, 8.8</td>
<td>Light Brown, Brown, Green, Yellow, Light Blue</td>
<td>0.25, 0.33, 0.58, 0.72, 0.97</td>
</tr>
</tbody>
</table>
Fig 1: Photo of TLC plate

References


