



A review of analysis of different phytochemicals present in *Moringa oleifera*, their extraction and separation methods along with nutritional and medicinal properties of *Moringa oleifera*.

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ABSTRACT: Many people today experience damaging consequences on their skin as a result of synthetic chemical-based therapies. According to research, the *Moringa oleifera* extract possesses a number of antibacterial characteristics and also functions as an anti-inflammatory and antioxidant agent. It provides a wide range of nutritional and therapeutic benefits, including the ability to lower blood sugar, treat inflammation, soothe sore throats, and more. *Moringa oleifera* includes a variety of phytochemicals, which are found in various areas of the plant and have varying antibacterial and therapeutic activities. Techniques include maceration, sublimation, percolation, distillation, soxhlet, and decoction processes can be used to extract the phytochemical contents. Thin Layer Chromatography, High-Performance Liquid Chromatography, Mass Spectrometry, and the Fractionation Method can all be used for separation and purification.

Keywords: *Moringa oleifera*; therapeutic; antibacterial; phytochemicals; extraction; separation; purification.

1. INTRODUCTION:

The drumstick or horseradish tree, *Moringa oleifera*, is a small to medium-sized evergreen or deciduous tree (Özcan, 2018). According to Bentham and Hooker's system of classification, the taxonomic hierarchy of *Moringa oleifera* is as follows:

KINGDOM: Plantae

SUBKINGDOM: Phanerogams

DIVISION: Angiospermae

CLASS: Dicotyledonae

SUBCLASS: Polypetalae

SERIES: Disciflorae

ORDER: Sapindales

FAMILY: Moringaceae

GENUS: *Moringa*SPECIES: *oleifera*

Figure 1- The *Moringa oleifera* tree.

2. **HABITAT:**

Although it can also be found in tropical America and Africa, the *Moringa oleifera* tree is most frequently seen in tropical Asia. India, Ethiopia, the Philippines, and Sudan are all major producers of *Moringa oleifera*. It flourishes in tropical Asia, Latin America, the Caribbean, Florida, and the Pacific Islands as well as West, East, and South Africa (Ojewumi et. al., 2022; Abdulkadir et. al., 2018; Razis et. al. 2014; Price, 2007). The species *Moringa oleifera*, which is native to South Asia and grows in the foothills of the Himalayas, is the most economically useful species. It is widely planted throughout the tropics. Tropical, subtropical, and semiarid regions are where the *Moringa* tree is most commonly grown (Razis et. al. 2014). Its favourable pH range is between neutral and slightly acidic, well-drained, sandy or loamy soil (Gopalakrishnan et. al, 2016; Kashyap et. al., 2022). As a heliophyte or plant that prefers the sun and heat, *Moringa oleifera* can withstand some cold but not very low temperatures (Tamang et. al., 2021; Raja et. al., 2016; Palada and Chang, 2003). According to estimates, India produces the most *Moringa*, making up around 80% of the world's supply. Apart from India, commercial *Moringa* production has been established in South Africa, Ghana, Mozambique, Kenya, and Zambia (Raja et. al., 2016).

3. PHYTOCHEMICALS PRESENT IN *Moringa oleifera*:

Phytochemicals are substances found in plants that are produced spontaneously. As secondary metabolites, phytochemicals are produced by plants. The stem, seeds, leaves, fruits, and flowers are popular places to find them. Alkaloids, saponins, tannins, steroids, phenolic acids, glucosinolates, flavonoids, terpenes, carotenoids, quercetin, caffeoylquinic acids, kaempferol, beta-sitosterol, and other phytochemical components are all found in *Moringa oleifera* (Imohiosen et. al., 2014; Anwar et. al., 2006; Kasolo et.al., 2010).

- i. **Alkaloids**: Basic heterocyclic nitrogenous chemicals with physiological activity are known as alkaloids. Alkaloids are frequently helpful as nutritional supplements and are well-known as anaesthetics, cardioprotective, and anti-inflammatory agents (Imohiosen et. al., 2014).
- ii. **Saponins**: Natural substances found in plants called saponins get their name from their ability to create stable, soap-like foam in aqueous solutions. A carbohydrate moiety is joined to a triterpenoid or steroid in saponins. As a hypolipidemic drug, saponins can be used to treat dyslipidemia by lowering levels of density lipoprotein and cholesterol. Additionally, it lowers cancer risks and blood cholesterol levels (Imohiosen et. al., 2014; Chumark et. al., 2008).
- iii. **Tannins**: Tannins are a class of biomolecules that are either phenolic or polyphenolic that are found in plants and act as a deterrent to herbivores. Tannins have astringent properties, making them effective in the treatment of skin eruptions, tonsillitis, and pharyngitis. Additionally, they function as antibacterial, anti-inflammatory, antioxidant, and anticancer agents (Imohiosen et. al., 2014).
- iv. **Steroids**: Biologically active substances with 17 carbon atoms organised in four rings in a particular chemical configuration are known as steroids. They are artificial lipids or substances that can bind to fat. Due to their anti-inflammatory properties, steroids are used to treat allergies (Imohiosen et. al., 2014).
- v. **Phenolic acids**: When combined with a phenolic ring, phenolic acids, also known as phenol carboxylic acids, are aromatic acid compounds. Phenolics are crucial to plant development, particularly in the formation of lignin and pigments. They aid plants' structural integrity as well. Since phenolic acids are easily absorbed by the intestinal walls, they function as antioxidants and prevent human gut cells from being harmed by free radical oxidation processes (Jimenez et. al., 2017).
- vi. **Glucosinolates**: While glucosinolates are classified as secondary metabolites of plants that contain glycosides containing sulphur and nitrogen that are responsible for the plant's pungent characteristics, glucosinolates themselves are not pungent. In addition to acting as antioxidants and anti-inflammatory substances, glucosinolates also have antibacterial capabilities (Bohinc et. al., 2012; Saini et. al., 2016).
- vii. **Flavonoids**: Phytochemicals found in plants include flavonoids. They consist of a diverse range of compounds with phenolic structures. There is widespread knowledge of the anti-inflammatory, anti-cancer, cardioprotective, and antibacterial effects of flavonoids (Imohiosen et. al., 2014; Saini et. al., 2016).
- viii. **β -sitosterol**: A class of phytosterols known as β -sitosterol possesses anti-inflammatory effects (Liao et. al., 2018).
- ix. **Quercetin**: Polyphenols can be found in quercetin, a type of flavonoid. The benefits to the heart are numerous. Furthermore, it has anti-inflammatory and antihistamine action (Ali et. al., 2015; Ferreira et. al., 2008).
- x. **Caffeoylquinic acids**: The term "caffeoylquinic acids" refers to plant metabolites that contain quinic acid and have been acylated with one or more caffeoyl groups. It is primarily an antioxidant and has anti-inflammatory qualities (Hamed et. al., 2020; Magana et. al., 2021).
- xi. **Kaempferol**: A type of flavonoid known as kaempferol is made up of several polyphenol biomolecules and may be categorised according to its phenol ring structure. Kaempferol helps with heart health, has anti-cancer benefits, and has anti-inflammatory qualities (Alam et. al., 2020; Ali et. al., 2015; Ferreira et. al., 2008).
- xii. **Terpenes**: Terpenes are naturally occurring plant substances, or phytoconstituents, that are primarily in charge of giving plants their distinctive scent. It can act as an antioxidant, an anti-inflammatory, and an antibacterial agent (Emmanuel et. al., 2014).
- xiii. **Glycosides**: A molecule known as a glycoside is one in which sugar is joined to a functional group, often a non-carbohydrate component. The non-sugar group is known as aglycone, and the sugar group is known as glycone. It is crucial to the defensive mechanisms against damage brought on by herbivores or pathogens (Akinyeye et. al., 2014; Imohiosen et. al., 2014).
- xiv. **Eugenol**: A phenolic group is found in naturally occurring aromatic compounds like eugenol. It helps to relieve stomach and toothaches. In addition, it has many antimicrobial characteristics and functions as an antioxidant (Akinyeye et. al., 2014).

4. EXTRACTION METHODS:

Various extraction techniques exist, such as the maceration method, the soxhlet extraction method, the decoction method, the distillation method, the sublimation method and the percolation method.

Solvents can be either polar or non-polar and come in a variety of varieties that are utilised for phytochemical extraction. To create a solvent system, you can utilise one or more solvents either separately or in combination. Methanol, ethanol, acetone, ethyl acetate, water, and other polar solvents are regularly utilised, although non-polar solvents like chloroform, ether, and hexane are more frequently used. (Akinyeye et. al., 2013 -2014; Oluduro, 2012; Tshabalala et. al., 2019; Rajamanickam and Sudha, 2013; Sravanthi and Rao, 2014; García-Beltr'an et. al., 2020; Bichi et. al., 2012; Imohiosen et. al., 2014; Hossain et. al., 2020; Busani et. al., 2011; Rahman et. al., 2009; Kumar et. al., 2011; Abdallah, 2015; Walter et. al., 2010; Abalaka et. al., 2012; Arora and Onsare, 2013; Pal et. al., 1994; Prasajak et. al., 2020).

From different studies, phytochemicals were extracted and gave the best results in ethanol, methanol, chloroform and hexane in comparison to other solvents (Akinyeye et. al., 2013 -2014; Oluduro, 2012; Tshabalala et. al., 2019; Rajamanickam and Sudha, 2013; Sravanthi and Rao, 2014; García-Beltrán et. al., 2020; Bichi et. al., 2012; Imohiosen et. al., 2014; Hossain et. al., 2020; Busani et. al., 2011; Rahman et. al., 2009; Kumar et. al., 2011; Abdallah, 2015; Walter et. al., 2010; Abalaka et. al., 2012; Arora and Onsare, 2013; Pal et. al., 1994; Prasajak et. al., 2020).

Sr. No.	Extraction Methods	Ideal for phytochemicals	Results	Citation
1.	Maceration	Thermolabile phytochemicals including alkaloids, flavonoids, tannins and saponins.	The mixture needs more purification after extraction as it is not in its purest form.	(Lezoul et. al., 2020; Balamurugan et. al., 2019)
2.	Soxhlet	Alkaloids, flavonoids, tannins, steroids, polyphenols and glycosides.	Increases efficiency of solubility of the phytochemicals and thus, no filtration is required, which gives more accurate results.	(Gopalsatheeskumar, 2018; Nieto et. al., 2008)
3.	Decoction	Heat stable phytochemicals including phenolic acids, carotenes and glucosinolates.	The mixture obtained needs to be filtered in order to get heat stable phytochemicals.	(Lezoul et. al., 2020; Balamurugan et. al., 2019)
4.	Sublimation	Suitable for phytochemicals ideal at low temperature such as terpenes.	Removes excess impurities from the mixture giving the phytochemicals in pure form.	(Šamec et. al., 2022)
5.	Distillation	Suitable for oil-based phytochemicals including eugenol and caffeoylquinic acids.	The mixture is separated using filtration, giving organic solvent-free phytochemicals.	(Magaña et. al., 2021; Ulanowska and Olas, 2021)
6.	Percolation	Ideal for phenolic acids, flavonoids and saponins.	The mixture obtained is in pure form, thus, no further purification is required.	(Balamurugan et. al., 2019; Azwanida, 2015)

Table 1- Comparative Analysis of different extraction methods along with results.

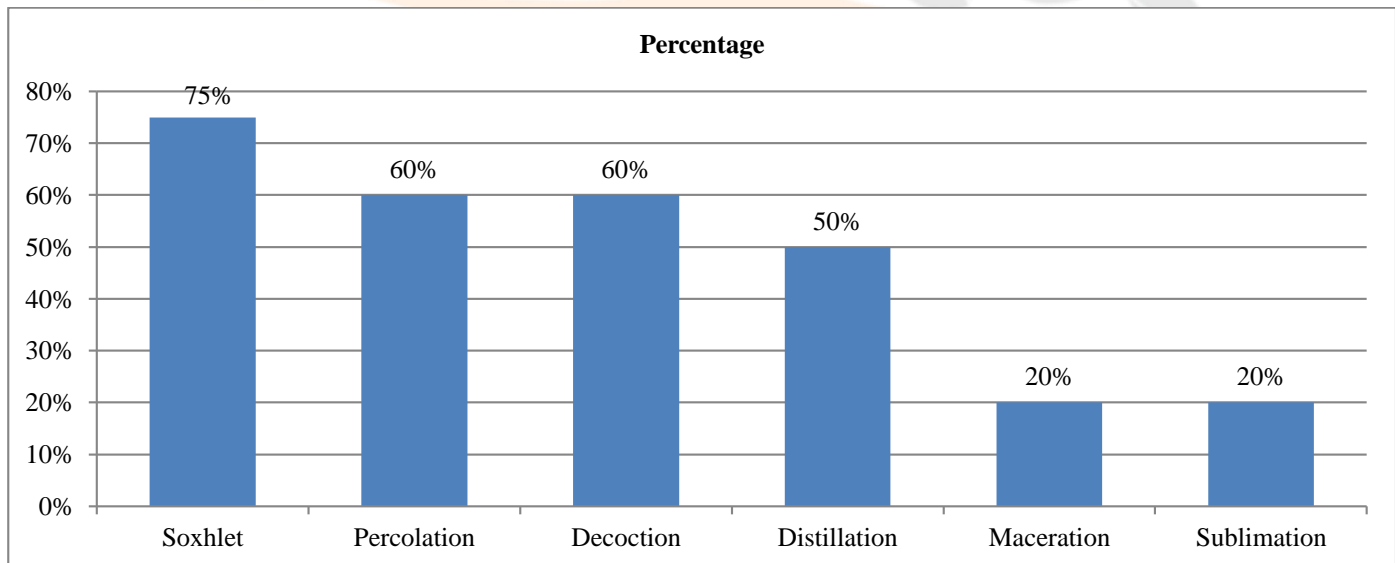


Chart 1- Analysis of different extraction methods efficiency using a bar chart (Azwanida, 2015; Lezoul et. al., 2020; Balamurugan et. al., 2019; Gopalsatheeskumar, 2018; Nieto et. al., 2008; Šamec et. al., 2022; Magaña et. al., 2021; Ulanowska and Olas, 2021; Abubakar and Haque, 2020; Gronbach et. al., 2020; Chemat and Boutekedjiret, 2015).

5. SEPARATION METHODS:

There are several different separation methods, such as High-Performance Liquid Chromatography (HPLC), Fractionation, Mass Spectroscopy and Thin Layer Chromatography (TLC).

Sr. No.	Separation Methods	Ideal for Phytochemicals	Results	Citation
1.	Thin Layer Chromatography	Saponins, alkaloids, flavonoids, steroids, carotenoids and tannins.	Phytochemicals were detected after spraying the required visualizing agent but confirmation is needed by other separation and purification methods. TLC is not solely reliable.	(Abubakar and Haque, 2020; Sonam et. al., 2017)
2.	High Performance Liquid Chromatography	Phenolic acids, flavonoids and alkaloids.	Phytochemicals were separated. HPLC provides both qualitative and quantitative results in a single assay. It has more efficiency when coupled with other techniques such as MS and TLC.	(Abubakar and Haque, 2020; Silva et. al., 2017)
3.	Mass Spectroscopy	Kaempferol, caffeoylquinic acids, quercetin, polyphenols, glucosinolates, anthocyanins and alkaloids.	Determination of phytochemicals using mass spectroscopy was more efficient method. It gave more accurate results when coupled with gas chromatography (GC-MS), HPLC (HPLC-MS), etc.	(Abubakar and Haque, 2020; Banu and Cathrine, 2015)
4.	Fractionation	Carotenoids, terpenoids, anthraquinones, glycosides, phenolic acids, eugenol and tannins.	Phytochemicals fractions were easily and efficiently separated. The fractionation technique was most accurate.	(Abubakar and Haque, 2020; Hamzaoui et. al., 2013)

Table 2- Comparative Analysis of different separation methods along with results.

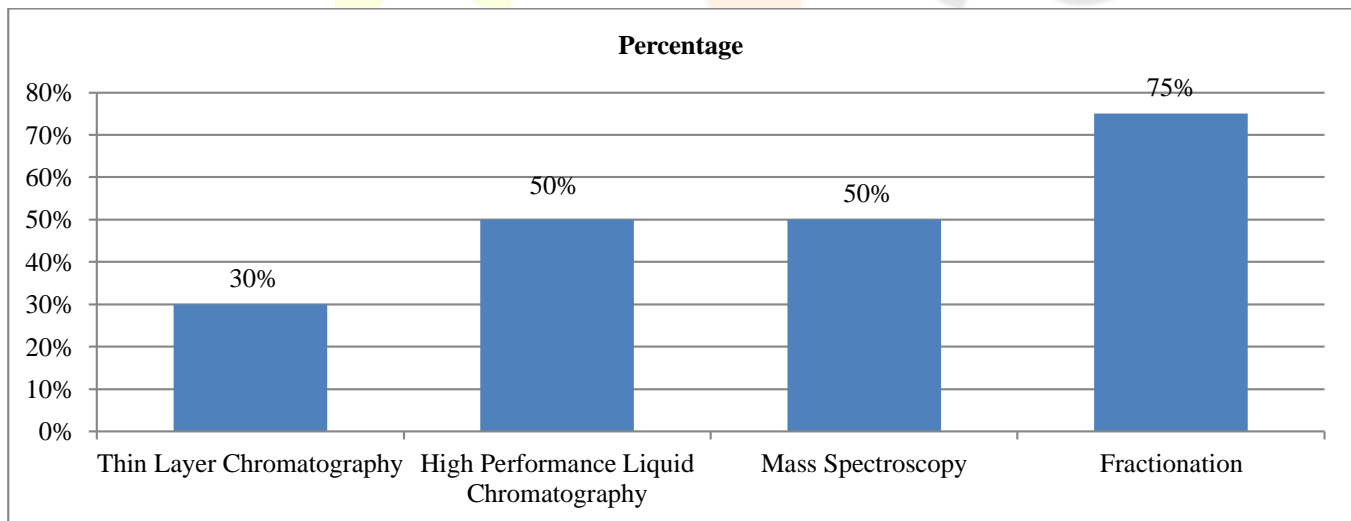


Chart 2- Analysis of different separation methods efficiency using a bar chart (Abubakar and Haque, 2020; Sonam et. al., 2017; Silva et. al., 2017; Banu and Cathrine, 2015; Hamzaoui et. al., 2013).

6. NUTRITIONAL AND MEDICINAL PROPERTIES:

For its nutritional benefits as a source of proteins and important amino acids like cysteine, methionine, tryptophan, and lysine, *Moringa oleifera* is used as a vegetable, herbal tea, and processed meals. In addition to being nutrient-dense, *Moringa oleifera* offers a number of medical qualities, such as the seeds ability to shield the liver from toxins, oxidation, and injury. Heart issues, eye conditions, and dyspepsia are all treated using *Moringa oleifera* root. The stem contains anti-tubercular qualities, cures ulcers, and acts as a painkiller for earaches. The gum portion is used to treat dysentery, fill cavities in teeth, and ease migraines, among other conditions. The *Moringa oleifera* flower reduces liver lipids and treats inflammation. The benefits of the leaves include relief from bronchitis, sore throat, and

blood sugar regulation (Anwar et. al., 2006). Additionally, *Moringa oleifera* contains a wide range of chemicals with antibacterial, antifungal, antioxidant, anti-inflammatory activities (Chumark et. al., 2008; Charoensin, 2014; Oduro et. al., 2008).

Sr. No.	Parts of <i>Moringa oleifera</i>	Nutritional and Medicinal Properties	Citation
1.	Root	It reduces constipation, kidney pain, cures eye conditions, benefit the circulatory system, treats dyspepsia and acts as a stimulant in paralytic problems. It tends to have anti-inflammatory, antiepileptic and diuretic properties.	(Mishra et. al., 2011)
2.	Leaves	It cures sore throat, bronchitis, eyes and ear infections, control blood glucose levels and reduces glandular swelling. It possesses certain cardioprotective, hepatoprotective and antiulcer properties.	(Charoensin, 2014)
3.	Bark	It cures eye diseases, prevents enlargement of spleen, works as a painkiller from the ear aches, heals ulcers. It serves to have various anti-tubular properties.	(Maurya and Singh, 2014)
4.	Gum	It tends to heal dysentery, fills cavities in teeth and eases migraine. It is also used to treat syphilis and rheumatism.	(Anwar et. al., 2006)
5.	Flower	It reduces liver lipids, reduces cholesterol cures muscle diseases and hysteria. It serves to hypocholesterolemic and antiarthritic properties.	(Anwar et. al., 2006; Patil et. al., 2022)
6.	Pods	The pods possess anti-inflammatory, cures diarrhoea, liver illness and relief from joint pain.	(Shahat et. al., 2022)
7.	Seeds	It has several diuretic and cardioprotective properties. Apart from this, it decreases the liver lipid profile.	(Patil et. al., 2022)

Table 3- A table representing the nutritional and medicinal properties of different parts of *Moringa oleifera*.

7. CONCLUSION:

Natural sources deliver positive effects devoid of any negative side effects. The natural source, *Moringa oleifera*, has been emphasised in order to concentrate on natural treatments for various forms of skin problems. The extraction of various phytochemicals utilising polar and non-polar solvents by using diverse extraction procedures, such as maceration, decoction, soxhlet, percolation, distillation, and sublimation, can be summed up by reviewing many reviews and research publications. Using the Soxhlet extraction method, the highest yield and most trustworthy results were attained. Utilising techniques like thin layer chromatography, high-performance liquid chromatography, mass spectrometry, and fractionation, separation and purification were accomplished. More accurate than other methods was the fractionation approach.

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