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# A Comprehensive Review on Therapeutic Potential of *Macrotylomauniflorum* Extract and Their Nano-Based Formulations

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**Abstract**
**Background:**WHO is recommending and enhancing the use of plant-derived medicines because they are easily available and affordable. Herbs contain different types of biological ingredients or secondary metabolites for their growth and development and these biological ingredients can be used by humans to treat various diseases and disorders.
**Main body of abstract:** Macrotylomauniflorum, frequently called horse gram in English and kulthi in Hindi, is a commonly grown plant in Asia, Australia, and Africa. It is primarily found on the soil edges of arid places in the Western Ghats region. In traditional medical cultures, it was used to cure many ailments, including bronchitis, hemorrhoids, asthma, tumors, and inflammation. The plant's seeds are widely consumed as a green vegetable in many parts of India. Because shrub extracts contain a comprehensive range of phytoconstituents, comprising tannin, flavonoids, alkaloids, triterpenoid, saponin, carbohydrates, phenolics, amino acids, and mucilage, it is believed that the seeds are rich in proteins and have medicinal qualities.Nanotechnology has opened up new possibilities for enhancing M. uniflorum extract's potency, availability, and durability, all of which will benefit the treatment of medical conditions. To tackle more chronic diseases such as asthma, cancer, and diabetes, it is necessary to introduce nanoparticles like NDDS into the standard medical system.
**Short conclusion:** The polyphenolic components of M. uniflorum function as a reducer in synthesizing nanocomposites, which are the intended phytoconstituents that led to the selection of this particular plant. A range of nanoformulations, such as zinc oxide, gold, and silver nanoparticles, have been created using M. uniflorum extract and exhibit tremendous therapeutic promise for treatingseveral illnesses.

**Abbreviations**
MU: Macrotylomauniflorum
Ag-NPs: Silver nanoparticles
ZnO-NPs: Zinc oxide nanoparticles
Au-NPs: Gold nanoparticles
HPLC: High-Performance Liquid Chromatography
DPPH: 2, 2-diphenyl-1-picrylhydrazyl
MTT: 3- (4,5-dimethylthiazol-2-yl)-2,5-diphenyl-2H-tetrazolium bromide
FTIR: Fourier-transform infrared spectroscope

## 1. Introduction

The World Health Organization (WHO) states that approximately 80% of the African and Asian populationsuse plant medicine as primary health care.[1] The demand for herbal medicine is increasing day by day due to itsminimal side effects onthe body. WHO is recommending and enhancing the use of plant-derived medicines because they are easily available and affordable.[2] Herbs contain different types of biological ingredients or secondary metabolite[3,4]for their growth and development and these biological ingredients can be used by humans to treat various diseases and disorders.

**Keywords:** Macrotylomauniflorum, Nanoparticles, Antioxidant, Antidiabetic, Anticancer, Electron microscope, Spectroscopy, Bioactive Phytoconstituents.

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These bioactive ingredients may be alkaloids, terpenes, tannins, glycosides, steroids, flavonoids, and phenolic compounds.[5] Today with the advanced knowledge of traditional medicinal herbs various medicines have been derived such as aspirin from *Filipendula,* morphine from *Papaver somniferum,* and ephedrine from *Ephedra* species.[6]According to Ayurvedagrain legumes have different types of biologically active ingredients and may be employed to cure various diseases in humans which is already mentioned in modern books like Charak Samhita and Shushruta Samhita.[7]Grain legumes also known as pulses, are traditional foods found in many countries of the world and consist of a variety of minerals and phytonutrients, as well as substantial quantities of protein and dietary fiber, making them healthier than fat.[8]The legume plant *M.uniflorum*, often known as horse gram, kulthi, and other names in different regions, is most frequently found on the soil margins of dry areas in the Western Ghats region. It is a member of the Fabaceae family and is widely cultivated in the states of Orissa, Karnataka,Andhra Pradesh, M. P., Tamil Nadu,Bihar, and in the hills of Uttaranchal in India. It is also cultivated in Sri Lanka, Australia, and Burma.[9] It has sub-erect or twining annual stems with 2.5–5 cm trifoliate leaves, white flowers, and prolonged straight pubescent pods having curving beaks. The seed of this plant is 3–4 mm wide and 6–8 mm long. The seeds are colored pale to dark reddish brown, orange-brown, or completely black. They have a trapezoidal, oblong, or moderately spherical form with mottled testa that are photo- and temperature-sensitive. It is primarily planted as a catch crop, maturing in four to six months, especially in the late summer or after a condition when it starts to rain.[10] *M. uniflorum* seeds, which are the fifth fastest-growing pulse in India, have a higher concentration of micro- and macrominerals, including secondary metabolites, and a lower concentration of anti-nutritional compounds.[11]The plant seeds are frequently eaten as green vegetables in various areas of India and are thought to be rich in proteins and have therapeutic properties. As per the study of Lavudi*et al.*[12], plant seeds have bioactive substances like isoflavonesand phenolic acids that are essential for boosting the antioxidant enzymes in cells with cancer and conferring anticancer benefits. It is a less popular bean that is typically fed to horses and regarded as a top source of dietary fiber, molybdenum, calcium, iron,and other minerals.[13]According to the Charak and SushrutaSamhitas, it was employed in ancient conventional medical cultures to cure a wide range of ailments, including hemorrhoids, asthma, tumors, inflammation, bronchitis, and liver problems[14], treating piles, hiccups, abdominal lumps, and regulating or reducing excessive sweating.[15]It has numerous health advantages, including relief from diarrhea, conjunctive inflammation, and loss of weight, irregular menstruation, constipation, and eczema of the skin. However, there are some drawbacks, including allergic responses in some people and gas and bloating due to the presence of phytic acid, which can be lessened by cooking, soaking in water, and germination.[16]It has been utilized in Ayurvedic medicine to cure a variety of illnesses, such as piles, conjunctivitis, worms, and arthritis. In Andhra Pradesh, an Indian state, the seeds were traditionally used as a jaundice cure, boiling them in water.[17]The *M.uniflorum* plant and its seeds are shown in **Figure 1**.


**Figure1**. *M.uniflorum* (A) Plant crop (B) Dried plant (C) Seed.[10,18]

Many studies on itsantioxidant capacity have been conducted recently, and the results indicate that the shrub may be able to neutralise free radicals, indicating a considerable antioxidant potential.[19-22]According to Gautam*et al.*[23], it has been used extensively since prehistoric times to treat anextensive range of situations, including stones in the kidneys[9,24], leucoderma, urinary tract issues[25], and heart illnesses.[26]

## 2. Main Text

### 2.1Pharmaceutical bioactive phytoconstituents of *Macrotylomauniflorum*

According to *Morris et al.*[27],plant seeds have high quantities of essential fatty acids, minerals, and flavonols. Employing the methods of gas chromatography, reverse-phase liquid chromatography with high performance (HPLC), and inductively linked plasma-optical emission spectral analysis, the fatty acid, flavonol, and mineral concentrations in seeds were reported.Arachidic, stearic, linoleic, oleic, gadoleic, and lignoceric acids, as well as seed quantities of myricetin, kaempferol, and quercetin, all demonstrated substantial year-end impacts. Papiya*et al.*[18] has been reported thatthe quantity of loss of drying, overall ash, and water-soluble ash were all high, although the contents of acid-insoluble and sulfated ash were found in less quantity. Additionally,Tannin, flavonoid, alkaloid, triterpenoid, steroid,saponin,carbohydrate, phenolics, glycoside, amino acid, and mucilage phytoconstituents were detected in plant extractsas representedin **Table 1**, and as per the quantitative analysis by Papiya*et al.*, the acetone extract had a high level of total polyphenols and tannins, but the methanol extract had a significant flavonoid content. TLC was used to detect ferulic acid and chlorogenic acid, whereas HPTLC was used to detect quercetin and chlorogenic acid.Suriyamoorthy*et al.*[28], utilized a phytochemical investigation to screen severalparts of the plant i.e., leaf, stem, flower, and seed extracts for phytoconstituents. Phytochemical screening was performed on extracts made from various solvents and secondary metabolic products were identified based on tests for coloration and precipitation.

**Table1.** Phytochemical Screening of *M. uniflorum*.

| S.N. | Phytoconstituents | Result |
|---|---|---|
| 1 | Alkaloids[18,28-30] | + |
| 2 | Flavonoids[18,27-30] | + |
| 3 | Saponins[18,28,30] | + |
| 4 | Glycosides[18,29] | + |
| 5 | Triterpenoids[18,28-30] | + |
| 6 | Carbohydrates[18] | + |
| 7 | Proteins[18,31,32] | + |
| 8 | Tannins[18,28-30] | + |
| 9 | Phenolic contents[18, 29, 30] | + |
| 10 | Steroids[18, 29] | + |
| 11 | Oil and Fats[27, 31] | + |
| 12 | Anthocyanins Enzyme Sources<br>Fibers, vitamins, minerals[31, 32] | + |

Suriyamoorthy*et al.*[29], utilized HPTLC fingerprint examination for the determination of phytoconstituents by using *M.uniflorum* and found that the presence of steroids, alkaloids, flavonoids, glycosides, phenols,and terpenoids is confirmed by the HPTLC fingerprinting pattern. According to Chakraborty *et al.*[30],ethanol-extracted sample seed coats had significant levels of saponin, alkaloid, flavonoid, tannin, phenol, and terpenoid, along withnatural phenols, primarily phenolic acids, flavonoids, and main antioxidant are abundant in plant seeds. It can be eaten as seeds, sprouts, or a standalone meal. It has a high protein content (22-24%), a significant amount of soluble fibers (57.2%), fat (1.1%), vitamins, and minerals (3.2%). These seeds extract exhibits strong anti-adipogenic, anti-hyperglycemic, and anti-hypercholesterolemic properties. It works to counteract oxidative stress. It can be consumed as a healthy meal and used for a variety of treatments.[31]Byassociating the retention index and mass spectral fragment structures of the identified 28 *M.uniflorum* molecules with those found in the GC-MS software library, ethanol extract underwent GC-MS analysis. Mome inositol,stigmasterol, n-hexadecanoic acid, ethyl derivatives, ethyl alpha-d-glucopyranoside, Vitamin E, linoleic acid and its esters, and 3-beta-stigmast-5-en-3-ol were the principal components discovered. The plant can be used for medicinal purposes as an antioxidant in hyperglycemia[33] and other associated illnesses since the extracts are high in linoleic acid with its esters, ethyl alpha-d-glucopyranoside, and mome inositol[34]as represented in **Table 2**.

**Table2.** Various phytoconstituents identified from *M.uniflorum* by GC-MS.[34]

| S.N. | Name | Retention time | Retention indices | Area% |
|---|---|---|---|---|
| 1 | Mome inositol | 13.027 | 1750 | 23.24 |
| 2 | stigmasterol | 26.695 | 3290 | 0.97 |
| 3 | n-hexadecanoic acid | 15.312 | 1971 | 2.76 |
| 4 | Vitamin E | 26.966 | 3154 | 2.42 |
| 5 | linoleic acid | 17.015 | 2151 | 19.79 |
| 6 | ethyl alpha-d-glucopyranoside | 12.007 | 1660 | 11.14 |

A wide range of compounds can be identified from their mass spectra using a method called mass spectrometry. Additionally,the seed extract was further subjected to GC-MS analysis by Abdullah*et al.*[35], who reported that a total of 28 substances were identified and measured. Benzene, 1-methyl-4-(1,2,2- trimethylcyclopentyl)-, (R)-, Benzene, and 1,3,6,10-Dodecatetraene, 3,7,11-trimethyl-(Z,E)- are the main bioactive compounds in seed extractas represent in **Table 3**. They found that *M. uniflorum* contained a variety of metabolites, therapeutically useful compounds, and new chemicals.

**Table3.** Phytoconstituentswere reported from the *M.uniflorum* plant.[18,27,34-36]

| S.N. | Phytoconstituents | Parts Used |
|---|---|---|
| 1. | 1,7-Octadiyne, Benzene, 1-methyl-4-(1,2,2- trimethylcyclopentyl)-, 1-(1,5-dimethyl-4-hexenyl)- 4-methyl-, and 1,3,6,10-Dodecatetraene, 3,7,11-trimethyl-(Z,E)- [35] | Seed |
| 2. | Mome inositol, 9,12-Octadecadienoic acid, stigmasterol, n-hexadecanoic acid, ethyl derivatives, ethyl alpha-d-glucopyranoside, Octanamide, N-(2-hydroxyethyl), Vitamin E, Heneicosane, linoleic acid, 3-beta-stigmast-5-en-3-ol, and Eicosane[34] | |
| 3. | Stearic, arachidic, oleic, linoleic, gadoleic, lignoceric acids. Myricetin, quercetin, and kaempferol[27] | |
| 4. | Flavonoids, Tannins, Polyphenols[18] | |
| 5. | Flavonoids (kaempferol, daidzein, myricetin, and quercetin)[36] | |
| 6. | Phenolic compounds (ferulic acid, vanillic acid, p-cumaric acid, chlorogenic acid,3,4-dihydroxy benzoic acid, caffeic acid, syringic acid)[36] | |
| 7. | anthocyanidins (cyanidin and malvidin) andPhytosterols (stigmasterol and ß-sitosterol)[36] | |

**2.3 Reported pharmacological properties of *Macrotylomauniflorum*extract**

Recent research has demonstrated that seeds can prevent hyperlipidaemic atherosclerosis.[37] Additionally, the α-amylase inhibitor from the seeds is known to have antihyperglycemic potential[33], suggesting that extracts from plants might be beneficial in the treatment of several human infections.[27]The seed is additionally used to cure uterine calculus, bronchitis, asthma, hiccups, disorders of the brain as well as eyes, piles, inflammatory conditions, liver issues, etc., by traditional medical knowledge.[30]Recently, there are various pharmacological potentialof *M. uniflorum*has been reported which is represent in **Table 4**, including relief from diarrhea, conjunctivitis, weight loss, menstrual irregularities, constipation, and skin rashes.[38]**Figure 2** highlights the medicinal properties of *M.uniflorum* extracts that have been recently published.

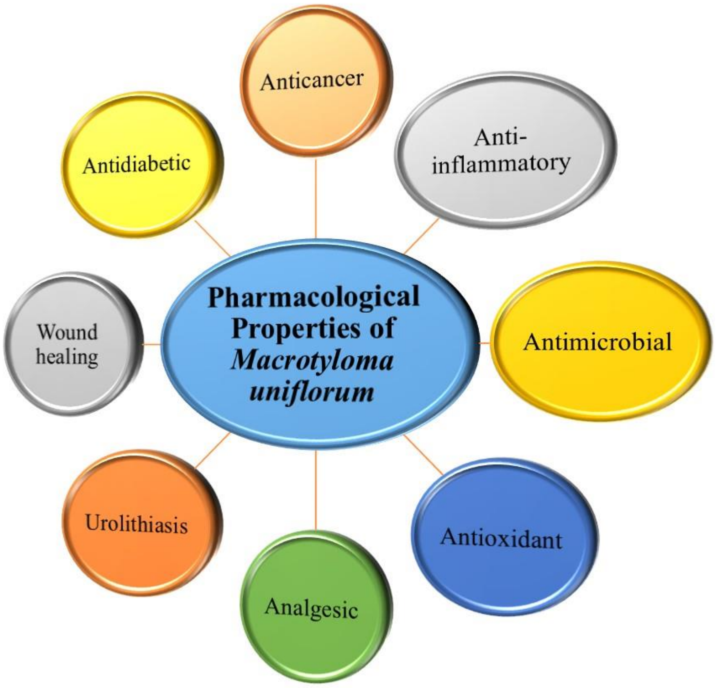


**Figure2**. Reported pharmacological effects of *M.uniflorum*Extract.[9,17,40,41,19-22,30,33,38,39]

### 2.3.1 Antioxidant Activity

Sree V. K. *et al.*[20] evaluated*the* anthelmintic and antioxidant capacitiesofseed alcoholic extracts against Pheretimaposthuma and reported that these extracts showed strong anthelmintic action. By contrasting the extracts with the reference ascorbic acid, the DPPH-freeradical-reducing capacity of the preparations was determined and the result suggests that the plant has an encouraging ability to neutralise free radicals. Additionally, in contrast to the standard, the sample with a higher concentration also demonstrated strong ferric-reducing power. As a result, both disorders caused by free radicals and parasite infections can be treated with these preparations of extracts.

Through a range of *in-vitro* methodologies, such as the DPPH experiment, total phenolic substances assay, reducing power method, and total antioxidant experiments, Ramesh *et al.*[21] assessed the antioxidant capacity of methanol-based extracts derived from two widely used legumes, *M.uniflorum,* and *V.radiata*, for their seeds and sprouts and reported the various antioxidant tests showed that the sprouts were more capable of absorbing antioxidants than the seeds.[21]

Priyanga*et al.*[22] explored the *in-vitro* antioxidant assessment of the ethanolic extract of the leaf by hydroxyl radical (OH), 2,2-diphenyl-1-picryl hydrazyl (DPPH), hydrogen peroxide ($H_2O_2$), ABTS+ cation decolorization test, nitric oxide radical (NO), metal chelating action, ferric inhibiting antioxidant power assay, and reducing power assessment employing established methods. The ethanolic stem extract revealed a better ability to reduce superoxide radicals, hydroxyl radicals, hydrogen peroxide, 2,2' azinobis-3 ethyl benzothiozoline-6 sulfonic acid, 2, 2-diphenyl-1-picryl hydrazyls radical, nitric oxide radical and metal chelating activity. Through a rise in the amount of extract, the antioxidant and reducing activities of ferric enhanced. Ascorbic acid served as the reference in each test.[22]

### 2.3.2 Antidiabetic Activity

Vasantha*et al.*[19]assess the effects of *M.uniflorum* stems and leaves with extracts made with ethanol on Alloxan-induced diabetic rat models. Utilization of plantextracts caused a considerable drop in blood sugar levels. According tothe result, the plant's extract can help you manage your blood sugar levels.

Herbshave long been used to treat diabetes because theycontain potent antioxidants, flavonoids, polyphenols, as well as proteins. They are among the sources with the highest concentrations of antioxidants, which reduce blood sugar and have other positive health effects[42].

Gupta *et al.*[33] produced diabetes in mice withstreptozotocin, theyexamined the anti-diabetic effects of α-amylase inhibitor extracted from the seeds and reported that the α-amylase inhibitor from *M. uniflorum* has antihyperglycemic action and can be employed as a dietary therapy for type 2 diabetes mellitus.[33]

### 2.3.3 Antimicrobial Activity

P *et al.*[39] have studied the phytochemistry of the plant materials and showed that they contained carbohydrates, steroids, tannins, phenol, proteins, and amino acids but did not contain alkaloids, glycosides, flavonoids, or saponins, and demonstrated a moderate to significant antibacterial activity towards 9 human pathogens. Alcoholic extract performed more effectively than water-based extract at reducing bacterial growth when antibacterial activity towards pathogens was taken into account.[39]

The most effective source of nutritious food, phytoconstituents, and bioactive ingredients can be found in plant seeds. So, using a conventional *in-vitro* methodology, Mathaiyan*et al.*[43]assess the phytochemical profile, antibacterial and antioxidant capabilities of several preparations of plant. The water-based extract of seed included a substantial quantity of phytochemicals, including protein, flavonoids, steroids, phenols, saponins, tannins, terpenoids, and carbohydrates. Interestingly, at higher concentrations, this water-based extract exhibited outstanding antibacterial action against bacterial pathogens. The antioxidant capacity of several kinds of free radicals, including DPPH and phosphomolybdenum, was also dose-dependent. Thus, the findings suggest that regular consumption of foods derived from *M.uniflorum*seeds can lower the buildup of reactive oxygen compounds and improve cell viability, leading to maximum longevity.[43]

**2.3.4 Anticancer Activity**

*M.uniflorum*, with its distinctive traits, is well known to play a significant part in plant defense reactions. Because the impact of its extracts on cytotoxicity has not been studied, the objective of Chakraborty *et al.*[8] was to assess the extracts' impact on cancer cell lines as well as infectious pathogens using practical and analytical methods. The phytochemical as well as analytical tests (TLC and GC-MS) revealed the abundance of various beneficial substances in *M.uniflorum*. The point that plant species can be utilized as a substitute functional food with high nutritional content has been supported by moderate to high antioxidant and cytotoxic activities of *M.uniflorum* extracts towards the MG 63 cell line.[8]

Employing the Trypan Blue Exclusion experiment to assess cytotoxicity and the DPPH radical scavenger analysis to assess antioxidant activity by Chakraborty *et al.*[30], and demonstrated that the seed coating sample that was extracted with ethanol encompassed large amounts of phytochemicals such as flavonoid, phenolic, tannin, alkaloid, saponin, and terpenes and exhibited cytotoxic and anti-oxidant capabilities towards the B16F10 and B16BL6 cell strains. Additionally, initial scavenging properties and trypan blue exclusion analysis on the B16F10 and B16BL6 cell lines revealed that it hassignificant anticancer action accompanied by these compounds.[30]

Rao *et al.*[40]investigated the existence of several phytochemicals as well as the antibacterial, antioxidant, and antitumor properties of extracts that were obtained from various *M. uniflorum* sections and reported that the susceptibility of Gram-negative bacteria was higher than that of Gram-positive bacteria. Steroids, tannins, alkaloids, flavonoids, saponins, and phenols were discovered to be present in extracts that were tested for a variety of significant phytochemicals. In terms of antioxidant activity measured using several techniques, including DPPH, FRAP, DMPD, and hydrogen peroxide (HP) radical scavenger seed coat extract outperforms all other extracts with 73, 76, 74, and 70%, respectively, at a concentration of 100 mg/ml. The seed coat extract has the greatest anticancer potency of any substance tested by the SRB Assessment towards the human renal adenocarcinoma cell line 786-O, with 66.2% viable cells.[40]

**2.3.5 Hepatoprotective and Antiobesity Activity**

With the use of paracetamol to Wistar albino rats orally for ten days, hepatotoxicity was induced by Ramasamy*et al.*[17]In comparison to the standard medicine silymarin, the oral administration of both ethanol-based extract as well as water-based extract considerably improved biochemical indicators in the hepatotoxicated rats. The ethanolic extract's hepatoprotective properties offer therapeutically superior efficacy, which was further substantiated by histological analysis. Concerning the hepatoprotective action, the phytochemical analysis identifies an abundance of phytoconstituents including phenolic compounds, flavonoids, and alkaloids.[17]

By investigating its impact on dietary intake, gaining weight, human serum lipid levels, SGOT, SGPT, and fat mass in rats, Kumar *et al.*[44], evaluated the anti-hypercholesterolemic impact of *M. uniflorum* extract. A considerable drop in total cholesterol, triglycerides, SGPT, SGOT, VLDL, and LDL, as well as a substantial rise in high-density lipoprotein or HDL, were the results of consuming water and ethanol extracts of the test plant for five weeks. When compared to those administered with a water-based extract, ethanol extract-treated groups considerably increased their faeces' elimination of cholesterol levels. The total body weight in the groups receiving the extract of water was substantially lower than that in the groups gettingwith ethanol extract. They suggest that the plantextracts exhibit powerful anti-hypercholesterolemia and anti-obesity effects, suggesting that they may be beneficial as functional components.[44]

Vadivelu*et al.*[45] tested both the leaves and seeds of *M.uniflorum* over antiobesity characteristics using an *in-vitro* pancreatic lipase reduction assay and molecular docking experiments, and found that a mixture of the leaves along with seeds reduced obesity in animals provided a high-fat diet.[45]

Through the use of male albino Wistar rats as their model and a high-fat diet alongside RNA extraction and semiquantitative RT-PCR analysis, Bharathi*et al.*[46] examined the impact of a medicinal plant formulation referred to *M.uniflorum* (used an equal amount of seeds and leaves powdered form) on obesity-associated oxidative stress-triggered hepatic damage. The results suggest that the plant formulation has antioxidant as well as anti-inflammatory potential, which helps the body fend off the harmful effects of obesity-induced high-fat diet. They also showed that a high-fat diet elevates liver enzymes and depletes antioxidants. They offer convincing evidence that using plant-based formulations can help reduce oxidative stress linked to obesity.[46]

**2.3.6 Wound Healing Capacity**

Appropriate treatments are necessary for exposed burn wounds to promote faster healing and guard against infection. Using plant extracts as a means of treating burns and wounds has been a common practice for many years and is an essential part of therapeutic care.[47]

Muthukumar*et al.*[41] employedrat experimental wounds,which were treated with a sheet-like wound dressing material containing *M.uniflorum* extract (MPE), fish scale collagen (FSC), and physiologically clotted fibrin (PCF) and reported that by inhibiting the expression of cyclooxygenase-2 (COX-2) and induced nitric oxide synthase, MPE was found to expedite the healing process as well as minimize inflammation. The healing of wounds has been accelerated by the biocomposite sheet's increased production of collagen and downregulation of matrix metalloproteinases(MMPs).Increasing the rate at which wounds heal was made possible by the use of plant extract.[41]

Once the permeable collagen sponges (CS) were created from fish scales, preparations of *M.uniflorum* (CSPE) and mupirocin (CSM) were added individually to the sponges to provide them with antibacterial action. According to Muthukumar*et al.*[47] the CSPE's resistance to collagenase enzyme and compressive durability were improved by the addition of plant extract. By using fibroblast and keratinocyte cell lines to test the sponges' biological compatibility *in-vitro*, the results showed that the sponges were biologically compatible. A preliminary in-vivo study using small animals could examine CS, CSM, and CSPE as burn/wound therapy materials in light of the findings.[47]

The antioxidative, antiurolithiatic, and wound-healing properties of plantseeds were assessed by Faujdar*et al.*[48], and discovered that the range of preserved cell viability with varying doses of plant extracts was 77.4%–90.74%. Plant extracts EC50 and EC90 produced wound closure rates of 85.66% and 91.09%, respectively. They suggest that these herbal extracts are beneficial in reducing the risk variables associated with urolithiasis.[48]

### 2.3.7 Anti-inflammatory and Analgesic Activity

Ashraf *et al.,*[25]has been reported that the seeds are diaphoretic, anthelmintic, emmenagogue, and diuretic. Additionally, it helps with bronchitis, urinary stones, and asthma. *M.uniflorum* seed dosages of 200 and 400 mg/kg were examined. All of the outcomes were meaningful activities.[25]

Vasishta*et al.*[38]conducted a preliminary phytochemical study on an ethanolic extract to determine its free radical scavenging activity as well as anti-inflammatory activities. The polyphenolic chemicals such as tannins and flavonoids in the ethanolic extract have the most promise. The extract of the plant had anti-inflammatory and antioxidant effects *in vitro* mediated scavenger, chelating, and diminishing potential.[38]

### 2.3.8 Urolithiasis and Kidney Stones

The kidney stones that are generated by dietary minerals in the urine are also referred to as calculi and crystal aggregations. The complex development of lithiasis is caused by several physicochemical processes, including supersaturation, the nucleation process, progress, accumulation, as well as retention inside the kidneys.[49]

For verification of their anti-lithiatic ability,Pal Sharma *et al.*[49]utilized an *in-vitro* modelwhich have been taken from Unnati*et al.*[50], for *M.uniflorum* seeds and cystone as a standard compound for dissolving kidney calculi and crystals of calcium oxalate. The aqueous fraction dissolved kidney stones and calcium oxalate more effectively than the other fraction. The results of the investigation into the minerals that create stones will give a clear picture of how to manage the danger of stone formation. Primary evidence supporting potent anti-lithiatic properties has been provided by this research.

Alongside their antioxidant as well as amino acid profiles, seeds of this plantthat were gathered from several Himalayan locations of Himachal Pradesh, India, have been investigated by Gautam*et al.*[23], and reported that when compared to alcohol and hydroalcohol, the water-based extract of Sundernagar and Rampur seeds had the best capacity for inhibition. The aqueous extract of Sundernagar seeds exhibited the greatest amount of inhibition in both the artificial urine assay and nucleation, correspondingly. Compared to high proteins germinated seeds, the Tris-buffer (pH 8.0) extract of dry seeds exhibited greater inhibition containing little protein. The secondary metabolites of horse gram protein may play a role in the suppression of CaOx crystals, rather than only the protein itself.[23]

Its antilithiatic action and nutritional profile have received a lot of attention, but no effort has been made to identify the nutrient and mechanism underlying the activity in the decoction of its seeds. Based onthe analysis of Gautam*et al.*[51],itisthe most effective treatment for kidney stones issues and malnutrition.India uses a lot of plantseeds because of their urolithiatic and diuretic properties.

Patel *et al.*[9] investigated the impact of seed aqueous extract (AEMU) on rat urolithiasis caused by ethylene glycol, and suggested that AEMU had substantial efficacy in urolithiasis, which may be related to its diuretic medication, suppressive effects on calcium oxalate crystal growth, and capacity to raise inhibitory levels while lowering promoter concentrations.[9]

**Table4.A brief review of therapeutic potential of different parts of the *M. uniflorum*has been tabulated based on evaluation techniques, target receptor site, enzymatic action, and possible pharmacological activity.**

| S. No. | Plant Parts | Method of Evaluation (*In-vivo*/*In-vitro*) | Targeted receptor/Enzymatic activity/Cell lines | Pharmacological activity |
|---|---|---|---|---|
| 1. | Plant extract | *In-vivo* Anthelmintic activity (Earthworm), and *In-vitro*DPPH assay, Ferric reducing power assay | 2, 2-diphenyl-1-picrylhydrazyl, Potassium ferricyanide radical | Anthelmintic and Antioxidant activity[20] |
| 2. | Seed extract | *In-vitro*DPPH assay | 2, 2-diphenyl-1-picrylhydrazyl | Antioxidant activity[21] |
| 3. | Leaves extract | *In-vitro*DPPH, OH, $H_2O_2$, ABTS+, NO radical, and ferric reducing power assay | 2, 2-diphenyl-1-picrylhydrazyl, Hydroxy, Hydrogen peroxide, Nitric oxide, and Potassium ferricyanide radical | Antioxidant activity[22] |
| 4. | Stem and leaves extract | *In-vivo*antidiabetic activity in alloxan-induced rats | Alloxan-induced rats | Antidiabetic activity[19] |
| 5. | Seed extract | *In-vitro*α-amylase inhibition assay | α-amylase | Antidiabetic activity[33] |
| 6. | Seeds extract | *In-vitro*agar well diffusion methods | Bacterial Pathogens including, Klebsiella pneumonia, Pseudomonas argentinensis,Escherichia coli,etc. | Antibacterial activity[39] |
| 7. | Seed extract | *In-vitro*agar well diifusion methods, and *In-vitro*DPPH, Phosphomolybdenum assay | Bacterial Pathogens including, Escherichia coli, Klebsiella pneumonia, S. aureus, etc. | Antibacterial and Antioxidant activity[43] |
| 8. | Plant Extract | *In-vitro*agar well diffusion methods, In-vitro DPPH and *In-vitro*human osteosarcoma cell (MG 63) | 2, 2-diphenyl-1-picrylhydrazyl,Shigellasp, Salmonella sp. Etc, MG-63 cell lines | Antibacterial, Anticancer and Antioxidant activity[8] |
| 9 | Seed extract | *In-vitro*DPPH, In vitro trypam blue exclusion method against B16F10 and B16BL6 cell lines | 2, 2-diphenyl-1-picrylhydrazyl, B16F10 and B16BL6 cell lines | Cytotoxic and Antioxidant activity[30] |
| 10. | Seed coat extract | *In-vitro*DPPH, DMPD, FRAP, and Hydrogen peroxide assay, *In-vitro*SRB assay | 2, 2-diphenyl-1-picrylhydrazyl, $H_2O_2$, Potassium ferricyanide radical, renal adenocarcinoma cell line 786-O | Antioxidant and Anticancer activity[40] |
| 11. | Seed extract | *In-vivo*Hepatoprotective potential | Paracetamol induced hepatotoxicity in rats | Hepatoprotective activity[17] |
| 12. | Plant extract | *In-vivo*antihypercholesterolemic activity on Sprague-Dawley rats | Examined the human serum lipid levels, SGOT, SGPT, and fat mass in rats | Antihypercholesterolemic activity[44] |
| 13. | Leaves and Stem extract | *In-vitro*pancreatic lipase reduction assay and *In-vivo*antiobesity assay in hight fat diet rat | Pancreatic lipase enzyme | Antiobesity activity[45] |
| 14. | Seed extract | *In-vitro*DPPH, ABTS+, *In-vitro*calcium oxalate monohydrate(COM) crystal inhibition assay and *In-vivo*scratch assay | 2, 2-diphenyl-1-picrylhydrazyl, calcium oxalate monohydrate(COM) crystal | Antioxidant, Antiurolithiatic and wound healing [48] |
| 15. | Seed extract | *In-vivo*analgesic, anti-inflammatory, diuretic activity by using Swiss albino mice | acetic acid induced writhing test, rat paw edema test, hot plate test | Analgesic, Diuretic and Anti-inflammatory activity[25] |
| 16. | Plant | *In-vitro*protease inhibition | Protease enzyme, 2, 2- | Antioxidant and Anti- |

| | | | | |
|---|---|---|---|---|
| | extract | assay and *In-vitro* DPPH assay | diphenyl-1-picrylhydrazyl | inflammatory activity[38] |
| 17. | Seed extract | *In-vitro*calcium oxalate crystal inhibition assay | Calcium oxalate crystal | Antiurolithiatic[23,49] |
| 18. | Seed extract | *In-vivo*antiurolithiatic procedure | Urolithiasis induced by ethylene glycol in rat | Antiurolithiatic[9] |

**3.Reported nanoformulationsynthesized fromM.*uniflorum*extracts**

The *M.uniflorum* extract has been shown to produce a variety of pharmacological effects.[8,17,19,20,25,39,49] By utilizing metallic nanoparticles combined with its extracts, the pharmacological activity can be increased.[52,53]Their distinctive size and large loading capacity seem to allow them to transport medications at high concentrations to disease locations[54].For example, Leonard *et al.,*[55]Jagtap*et al.,*[56]Jayaseelan*et al.,*[57] He *et al.,*[58] and Saxena*et al.,*[59]formulated biocompatible metallic nanoparticles capped by plant extracts, improved their stability and pharmacological activity.Implementing *Citrulluscolocynthis* leaf extract, Satyavani*et al.*[60] created silver nanoparticles for healing burns and open wound infections.As per the phytochemical screening, the presence of polyphenol constituents in *Macrotylomauniflorum* serves as a reducing agent for metal ionsto produce metallic nanoparticles, which are the desiredconstituents for the choice of this plant.[61]*M.uniflorum* extracts have been used to make a extensive variety of nanoparticles, comprisingcomposite sheet[47,62], zinc oxide[63], gold[64], and silver nanoparticles[12,61,65,66]as representedin **Table 5**. There are various metallic nanoparticles have been reported recently from plant extracts with different dimensions, forms, structures, and morphologies.[63,67–69]

**3.1 Silver/Gold nanoparticles (Ag-NPs, Au-NPs)**

Lavudi*et al.*[12] employed the ovarian carcinoma cell line PA-1, and plant extracts to synthesize Ag-NPs and examined the effectiveness of these particles in slowing tumor growth. In a DPPH study, they observed that 76.08% of them produced at 500 µg/ml of concentration, indicating their activity. The MTT assay demonstrates the effectiveness of MU-Ag-NPs in lowering cell viability. Regarding the findings that were obtained, they conclude that MU-Ag-NPs have a remarkable ability to prevent the proliferation of ovarian cancerous cells *in-vitro* by generating damage to DNA and apoptosis.[12]

By comparing the biological actions of its biosynthesized Ag-NPs with plant flour and its prepared extracts, Sudha*et al.*[65]provided a low-cost source of mineral substances with antidiabetic activity through the use of *in-vitro* α-amylase inhibition and *in-vitro*struvite crystals inhibition assays. According to their findings, Ag-NPs with typical particle sizes of 30 to 60 nm exhibit superior anti-diabetic activity in a dose-dependent manner when compared to the other two extract samples (water extract and ethanol extract). When plant extracts and nanoparticles were compared, it was shown that the MU-Ag-NPs had a greater degree of inhibition of struvite crystal than the plant extracts. As a result, it may show that the seed flour extracts including their Ag-NPs were an exciting natural herbal remedy that had potential mineral content, antidiabetic, and anti-urolithiatic actions that may be employed as a valuable ingredient in dietary compositions.[65]

Vidhu *et al.*[61]and Aromal*et al.*[64], synthesized silver (Ag-NPs) as well as gold nanoparticles (Au-NPs) by employingseeds and dried parts of plant *M.uniflorum*, respectively. UV-visible, TEM, X-ray diffraction and FTIR analyses were used for evaluationof nanoparticles.The silver nanoparticleshad a cubic structure with a face-centered according to the XRD examination. TEM scans and UV spectra showed well-distributed silver nanoparticles with an anisotropic shape and a size of around 12 to 17 nm. FTIR spectra represent the different kinds of functional groups that exist in the bio-molecule that covers the nanoparticles.[61,64]

Dhanalakshmi*et al.*[66]manufactured silver nanoparticles with the help of aqueous 0.1 mM solution of $AgNO_3$. Ag-NPs in a solution of water have an absorbance peak at about 450 nm caused by a surface plasmon resonance within the ultraviolet-visible spectrum. The existence of multiple functional groups responsible for encapsulating the silver nanoparticles was validated by FTIR examination, and the dimension of the nanoparticles were verified by SEM examination, which reveals that the particles belonged in the 30 to 50 nm diameter region.[66]

**3.2 Zinc Oxide nanoparticles (ZnO-NPs)**

Sali*et al.*[63] synthesized metal oxide nanoparticles (ZnO and Ag-ZnO) by using *M.uniflorum* leaves extracts with the help of Zinc sulfate, and structural validation of nanoparticles was accomplished by TEM, SEM, UV-Vis, FTIR, and Zeta potential methods.On cultures of different bacterial species, including B. subtilis, streptococci, and E. coli, the minimum inhibitory concentration of synthesized nanoparticles was determined by the 96-well plate method. An ELISA plate reader was used to record the absorbance at 620 nm.They reported thatnanoparticleshavebetter results for antibacterial activity by damaging the DNA, and cell membranes due to reactive oxygen species (ROS), with MIC values of 62.5 µg/ml. Compared to ZnO nanoparticles, Ag-ZnO nanoparticles demonstrated a stronger antibacterial activity.[63]

ZnO nanoparticles were produced from plant seeds by Sudha*et al.,*[70] who then showed dose-dependent antibacterial efficacy towardsKlebsiella, Staphylococcus aureus, Pseudomonas aeruginosa,and Escherichia coli. The investigators also evaluated the dose-dependent effects of antioxidant and antidiabetic agents on α-amylase and DPPH, respectively. By employing MCF-7 cell lines in the MTT experiment, they proposed that the ZnO nanoparticles were useful in reducing cell viability. ZnO-NPs can attach to CT-DNA through a partial intercalation binding process, according to the interaction between ZnO-NPs and CT-DNA.[70]

**3.3The composite sheets**

Muthukumar*et al.*[47], utilized*M.uniflorum*extracts along with fish scale and mupirocin to produce a composite sheet that was used to cure burns and wounds. By employing the modified agar well diffusion method, they concluded that the formulation had a considerable antibacterial action. Biological compatibility was also tested by using fibroblast and keratinocyte cell lines, resultssuggest that the sheets were biologically compatible. According to the outcomes, it might be explored as burn/wound treatment materials.[47]Muthukumar*et al.*[62] made a composite sheetby using *M.uniflorum* extracts, fish scale collagen (FSC),and physiologically clotted fibrin (PCF)to provide antibacterial characteristics. The biocompatibility of synthesized biomaterial wasdetermined by developing NIH 3T3 fibroblasts and keratinocyte cell line (HaCaT) and reported that the FSC: PCF: MPE biocomposite sheets were biologically compatible, andcould be used as a burn/wound healing material.[62]

**Table5.A brief review of nanoparticles synthesized from different parts of the *M. uniflorum*has been tabulated based on standardization and evaluation techniques, target receptor site, enzymatic action, and possible pharmacological activity.**

| S. No. | Nanoformulation | Plant Parts | Characterization Techniques | Reaction Time | Required Temp. & pH | Size of Nanoparticle | Shape of Nanoparticle | Targeted Receptor/Enzymatic activity/ Cell lines | Validation Methods | Pharmacological Activity |
|---|---|---|---|---|---|---|---|---|---|---|
| 1. | Silver Nanoparticles (Ag-NPs) | Plant Extract | Zeta Potential, X-ray diffraction Pattern, Transmission Electron Microscopy, Scanning Electron Microscopy, Fourier Transform Infra-Red spectroscopy, Ultraviolet-Visible spectroscopy, etc. | 20 min | 70 – 80℃ | 50nm – 100 nm | Spherical and oval | 2, 2-diphenyl-1-picrylhydrazyl, PA-1 Ovarian cancer cell lines | *In-vitro* DPPH Assay, *In-vitro* MTT Assay | Better Antioxidant and Anticancer activity[12] |
| | | Seed | | 5 hours | Room Temp. | 30nm – 60 nm | Spherical and Hexagonal | α-amylase and struvite crystals | *In-vitro* α-amylase inhibition assay and *in-vitro*struvite crystals inhibition assay | Nanoparticles have greater Antidiabetic and Antiurolithiatic activity compared to water and ethanol extract[65] |
| | | Seed | | 20 – 60 min | Room Temp & pH – 6, 7, 8, 9, 10 | 12 nm | Face centered cubic structure | - | Spectral AnalyticalTechnique | Characterization/ Validation[61] |
| | | Seed | | 1 hour | - | 30nm – 50nm | Spherical | - | | Characterization/ Validation[66] |

| | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| 2. | Gold Nanoparticles (Au-NPs) | Plant Extract | | 2 - 20 min | 373 K & pH – 4, 5, 6 7, 8 | 14nm – 17nm | Spherical | - | | Characterization/ Validation[64] |
| 3. | Composite Sheet: FSC:PCF:MPE | Plant Extract | | 48 hours | - | - | Fibrous and porous sur-face which | NIH 3T3 fibroblasts and human keratinocyte cell line (HaCaT) | Modified agar well diffusion method, *In-vitro* MTT Assay | Better Antibacterial activity and Biologically compatible[62] |
| | Composite Sheet: CS:CSM:CSPE | Plant Extract | | 48 hours | - | The pore size in sheet 30 - 70µm | Collagen sponges hold tri-helical structure | Fibroblast and keratinocyte cell lines | *In-vitro* MTT Assay | More wound Healing and Antibacterial activity[47] |
| 4. | Zinc Oxide Nanoparticles (ZnO-NPs), Ag-ZnO nanoparticles | Leaves | | 20 min | 70°C | 120.16 nm and 91.17 nm | Polycrystalline in nature and face-centered cubic structure | By damaging the DNA, cell membranes due to reactive oxygen species (ROS) | 96-well plate with ELISA plate reader | Greater Antibacterial activity compared to ZnO nanoparticles[63] |
| | | Seed | | 60 min | 80°C | 30nm – 60nm | Spherical | 2, 2-diphenyl-1-picrylhydrazyl, α-amylase, MCF-7 cell lines | *In-vitro* DPPH Assay, *In-vitro* MTT Assay, *In-vitro* α-amylase inhibition assay | Antioxidant, Antidiabetic, and Anticancer activity[70] |

**4. Conclusion**

The detailed literature review of *M. uniflorum* reveals the existence of significant pharmaceutical bioactive phytoconstituents, such as tannins, flavonoids, fiber, essential fatty acids,alkaloids, and phenolic acid, which are reported to manage various health problems, including cancer, diabetes, inflammation, asthma, and kidney stones.The majority of the research is focused on managing and treating urolithiasis. Sudha et al., Patel *et al.,* and Gautam*et al*. have described a substantial potential to treat urolithiasis as well as break kidney stones due to its crystal-dissolvingproperties. Due to the presence of polyphenolic and flavonoids constituents Lavudi*et al*. derived antioxidant and anticancer properties from *M. uniflorum*. Through the development of innovative medication delivery techniques, the plant extracts can be utilized more efficiently and optimally by mixing them into modern dosage forms. The synthesis of metallic nanoparticles has been the subject of current research, which has been thoroughly reviewed in this study along with a summary of its biological purposes. Nanotechnology provided new opportunities for improving the effectiveness, distribution, and durability of *M. uniflorum* extract, which will have a positive effect on the management of health issues. Therefore, to combat more chronic diseases like diabetes, cancer, and asthma, nanoparticles must be incorporated into the regular medical system as NDDS. These methods involve inclosing plant extracts in nanoparticles with stirring procedures utilized by various researchers. A thorough understanding of the molecular mechanisms behind the synthesis of plant-mediated nanoparticles is crucial to making this technique economically competitive with more traditional methods.Even though a molecular mechanism has been suggested in numerous publications, nearly all of them are merely plausible theories lacking strong experimental backing. Since the hypothesis differs with different phytoconstituents present in plants as well, a more thorough analysis is needed to understand the true molecular mechanism of a given plant system, and it will continue to be of great help.Recently, there are different nanoparticles such as silver, zinc, gold, and composite sheets have been developed from plant extract and exhibit significant efficacy against a range of health issues such as bacterial infection, urolithiasis, diabetes, ovarian cancer, etc.These nanoparticles can also be studied for a variety of other medical conditions, including depression, anxiety, lung cancer, inflammation, Parkinson's, Alzheimer's, and diseases related to the kidneys and liver.For example, Muthukumar*et al*.synthesized a composite sheet from *M. uniflorum*, fish scale, and mupirocin (containing a short chain of fatty acid linked to monic acid by an ester linkage) to treat burns and wounds by using *in-vitro* and *in-vivo*methodologies, and they reported that the sheets are biocompatible and may be employed for the management of other skin problems such as eczema, fungal infections, acne, etc. Nanoparticles can target multiple receptor sites, including α-amylase, α-glucosidase, and DPP-4, to aid in the control of diabetes. While Gupta *et al*. and Sudha*et al*. have mainly concentrated on α-amylase, diabetes can also be treated by targeting other sites. Similarly, Lavudi*et al*.synthesized nanoparticles for the treatment of cancer, but they only studied ovarian cancer cell lines. Since lung, prostate, and breast cancer are the three main cancers that cause mortality in humans, these nanoparticles could also be tested on these cancer cell lines. Further research is necessary to investigate the pharmacological potential of synthesized nanoparticles derived from different extracts of *M. uniflorum*.

**Declarations**

**Ethics approval and consent to participate**

Not applicable.

**Consent for publication**

Not applicable.

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Not applicable

**Competing interests**

The authors declare that they have no competing interests

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**Plant authentication**

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