



Green Synthesis of MgO Nano Particles, Phytoconstituents, Antibacterial Activity of Stem of *Jatropha Gossypifolia* (L.)

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ABSTRACT

Jatropha gossypifolia is one of the folkloric medicinal plants belonging to the family *Euphorbiaceae*. It is used as antioxidant nutrients, blood purifier which protects against Cardio Vascular disease, Diabetes, various forms of Cancer and AIDS anticancer effects, anti – inflammatory and antimicrobial activity has been found to be chemo preventive. The present study provides information about *Jatropha gossypifolia* stem phytochemical analysis. Maceration extract method was carried out using polarity solvent Acetone (AC) were chemically screened. The main aim of the present study was to evaluate the phytochemical properties of *Jatropha gossypifolia* stem to treat various diseases. The Qualitative Phytochemical Analysis of flower revealed the presence of Alkaloids, Saponin, Tannin, Flavonoid, Terpenoids etc. Additionally, it can be identify trace elements in the materials by XRD (X-Ray Diffraction), SEM (Scanning electron microscope), FTIR (Fourier-transform infrared) spectroscopy and UV-Visible (Ultra violet-Visible) spectroscopy and EDAX (Energy Dispersive X-Ray Analysis).

Keywords: Antioxidants, *Jatropha gossypifolia*, Cardio vascular disease, Diabetes, Cancer

1. INTRODUCTION

Medicinal plants, also called medicinal herbs, have been discovered and used in traditional medicine practices since prehistoric times. Plants synthesize hundreds of chemical compounds for functions including defense against insects, fungi, diseases, and herbivorous mammals. Numerous phytochemicals with potential or established biological activity have been identified. However, since a single plant contains widely diverse phytochemicals, the effects of using a whole plant as medicine are uncertain. Further, the phytochemical content and pharmacological actions, if any, of many plants having medicinal potential remain unassisted by rigorous scientific research to define efficacy and safety. *Jatropha gossypifolia* (Euphorbiaceae), a common garden plant in tropical countries, has been used as a traditional medicine. Plants are well known as a major source of modern medicines. From ancient times, humans have utilized plants for the treatment or prevention of diseases, leading to the dawn of traditional medicine. *Jatropha gossypifolia* is one of the plants that are used in Chinese, Ayurvedic, and Thai traditional medicine for the treatment of fever, pain, and dysentery. The root, stem, leaves, fruit, seed, bark and latex of the plant are largely used for the treatment of many diseases in different parts of the world [1]. In the present study, we discussed the medicinal uses and chemical constituents of some of the species of *Jatropha*. Bark is used as a fish poison [2]. Leaves are regarded as antiparasitic, applied to scabies, rubefacient for paralysis, rheumatism; also applied to hard tumors [3]. In South Sudan, the seed as well as the fruit is used as a contraceptive [4]. List the species as a honey plant [5]. It is reported to be abortifacient, anodyne, antiseptic, cicatrizant, depurative, diuretic, emetic, hemostat, lactagogue, narcotic, purgative, rubefacient, styptic, vermifuge, and vulnerary [4], [6]. Latex is applied topically to bee and wasp stings [2] and to dress sores and ulcers and inflamed tongues [7] list it for homicide, pesticide, and raticide as well. Colombians drink the leaf decoction for venereal disease [9]. The latex was strongly inhibitory to watermelon mosaic virus [10]. Further information provided by Kirtikar and Basu [11].

2. MATERIALS AND METHODS

2.1. Plant Material

Jatropha gossypifolia (L.) leaf was collected from Bavani River in the during the month of December 2018 (Figure 1).

2.2. Preparation of extract

Fresh stem parts of *J. gossypifolia* were harvested and washed with distilled water so as to remove dust and other foreign particles. The stems were then left on a clean surface to dry well. The stem parts were air-dried under shade for 6 - 8 days. Then the dried material was grinded to fine powder using an electric grinder and stored in air tight bottles. The powdered material was used further for phytochemical screening and preparation of extracts. The voucher specimen of this plant sample related information is preserved for future reference in the Department of Chemistry, Vivekanandha College of Arts and Sciences for Women (Autonomous), Elayampalayam, Tiruchengode, Namakkal District, Tamil Nadu, India.



Figure 1. *Jatropha gossypifolia* (L.) Habitat and Stem

2.3. Extraction

Maceration process was conducted with the pellet by magnetic stirrer (a). 9g of KOH pellets was dissolve in the 50ml of water and it is slowly dropped into the above mixed solution, and precipitation is form slowly; the molar ratio of $Mg(NO_3)_2/KOH$ was 3 : 9. The temperature of the precipitation process was about 350°C. The mixed solution was stirred for 4 hours. And above content was filtered with the No.1 whatman filter paper by washing with acetone and dried it for 60 - 80°C in the hot air oven (b) for 4 hours. Then precipitated material was transfer into the silica crucible and heated at 400°C in the muffle furnace (c) for 4 hours. And the silica crucible was kept in the desiccators for cooling. The composite were well grind in fine powder particles with the help of mortar and pestle (d). The fine powder particles is washed with the ethanol, after the evaporation of solvent ethanol. The dried plant material was pulverized into fine powder using a grinder (mixer). 6g crude powder of *J. gossypifolia* (stem part) part was dissolve in 60 - 80ml of water in the 400ml beaker. The beaker was heated for 20 - 25mins in the Bunsen burner. Take 14.5g Nickel nitrate and it is dissolve in 100ml of water. From the hot extract of crude powder, take 10ml and slowly dropped into the above mixed solution. Then the temperature of

precipitation process was about 350°C. The mixed content was stirred for 4 hours. And the content was filtered with the No.1 whatman filter paper and the extract in the watch glass; the sticky greenish-brown substances were obtained and stored in desiccators for further use. Then it dried for 1 hour in room temperature. The dried substances were washed thoroughly with ethanol and grind well with the help of mortar and pestle. Some of the extracts of each solvent were used for the qualitative phytochemical screening, then identification of the various classes of phase composition of the products were characterized by an XRD (X - Ray Diffraction); SEM (Scanning Electron Microscope); FT - IR (Fourier Transform Infrared Spectroscopy); UV - Visible (Ultra Violet - Visible Spectroscopy); and additionally it is also allowed to test for antioxidant activity.

2.4. Preliminary Phytochemical Screening

Qualitative Phytochemical analysis of the crude powder of the stem collected was determined according to the standard procedures to identify the constituents as described by. Foam test for saponins, Salkowski and Liebermann-Burchard test for terpenoids and triterpenoids, Ferric chloride test (FeCl_3) test for tannins, Killer-Killani test for cardiac glycosides, Fehling's test for reducing sugars, xanthoproteic test for proteins, iodine test for starch and ammonia test for detection of flavonoids were performed to identify the constituents present in the extracts of the stem part of the plant.

2.4.1. Test for alkaloids

Extracts were dissolved individually in dilute Hydrochloric acid and filtered.

a. Dragendroff Test

Bismuth nitrate was dissolved in distilled water and acetic acid, Potassium iodide was dissolved in distilled water Equal volume of A and B mixed together the solution was taken distilled with distilled water and glacial acetic acid and used as drangendroff's reagent.

The solvent extract was adding drangendroff's reagent was mixed well the orange colour indicates the presence of alkaloids.

b. Mayer's Test

Filtrates were treated with Mayer's reagent (Potassium Mercuric Iodide). Formation of a yellow colored precipitate indicated the presence of alkaloids.

c. Wagner's Test

Filtrates were treated with Wagner's reagent (Iodine in Potassium Iodide). Formation of brown/reddish precipitate indicates the presence of alkaloids.

2.4.2. Test for phenolic compounds

Lead acetate Test

The lead acetate was dissolved in distilled water and used as lead acetate reagent. The solvent extract of *Jatropha gossypifolia* was adding with lead acetate solution was mixed well. The white colour precipitate indicates the presence of phenolic compounds.

2.4.3. Test for flavonoids

a. Shinodo's Test

The plant extract in alcoholic solutions, few pieces of magnesium turnings were added with the few drops of concentrated hydrochloric acid was added and gently heated. Development of red or orange colour indicates the presence of flavonoids.

b. Ferric Chloride Test

Extracts were treated with 3-4 drops of ferric chloride solution. Formation of bluish black colour indicates the presence of flavonoids.

c. Sodium hydroxide Test

Sodium hydroxide was dissolved in distilled water and then the solution was added with test solution. Development of yellow orange colour solution denotes the presence of flavonoids.

2.4.4. Test for glycosides

a. Killer killani's Test

Test solution was mixed with acetic acid, concentrated sulphuric acid and ferric chloride. Development of brown colour denotes the presence of glycosides.

b. Legal Test The plant extracts few drops of pyridine and sodium nitroprusside was added and gently mixed. Development of red to pink colour solution indicates the presence of glycosides.

2.4.5. Test for terpenoids

The plant extracts was mixed with chloroform and sulphuric acid. Formation of red colour aqueous solution at the top and green fluorescence at bottom layer indicates the presence of terpenoids.

2.4.6. Test for Tannins

The extracts was mixed with ferric chloride is dissolved in distilled water. A development of dark green colour indicates the presence of tannins.

2.4.7. Test for Saponins

a. The plant extracts was mixed with sodium carbonate dissolved in distilled water. The formation of honey comb like frothing indicates the presence of saponins. **b. Foam Test:** 0.5 gm of extract was shaken with 2ml of water. If foam produced persists for ten minutes it indicates the presence of saponins.

3. RESULTS AND DISCUSSION

Plants are the great importance to the health of individuals and communities from time immemorial. Plant kingdom provides a tremendous reservoir of various phytochemicals with potential therapeutic properties. The major phytochemicals of interest are alkaloids, tannins, flavonoids, phenolic compounds, steroidal saponins (saponins); however, other diverse groups of naturally occurring phytochemicals such as unsaturated sterols, triterpenoids, and essential oils are also present. These phytochemicals play important role in herbivore deterrence due to astringency or they may act as phytoalexins, killing bacteria that the plant recognizes as a threat. Plant extracts are used to treat numerous human diseases and have prominent effect on the animal system, important therapeutic properties and antimicrobial activities against various pathogens. Plant can function as sources of anti-cancer agents. Percent extractive value of various fractions of partially purified hot extracts is depicted. Maximum percent extractive value was observed for aqueous fraction (Table 1).

All the tested phytochemicals were found to be present in dried plant material (stem) as shown in (Table 2). The presence of flavonoids and tannins in the stem is likely to be responsible for the free radical scavenging activity. Flavonoids and tannins are phenolic compounds and plant phenolics are a major group of compounds that act as primary antioxidants or free radical scavengers. These findings give credence to the traditional medicinal application of the stem as remedies for sores, rash and bewitchment, internal and external wounds and infections. Flavonoids have been referred to as nature's biological response modifiers because of strong experimental evidence of their ability to modify the body's reaction to allergies, virus and carcinogens. They show anti-allergic, anti-inflammatory,

antimicrobial and anti-cancer activity. Cardiac are used in the treatment of congestive heart failure and cardiac arrhythmia. They are also, used to strengthen a weakened heart and allow it to function more efficiently. Steroids anti-inflammatory effects. Glycosides, flavonoids, tannins have hypoglycemic activities. Saponins possess hypocholesterolemic and anti-diabetic properties. The terpenoids have also been shown to decrease blood sugar level in animal studies. Steroids and triterpenoids showed the analgesic properties. The steroids and saponins are responsible for central nervous system activities.

Table 1: Extraction of different partially purified fractions of *J.gossypifolia* Stem Extract

S. No	Solvent	Percent extractive value
1.	Petroleum ether Fraction (PEF)	4.9
2.	Benzene Fraction (BEF)	1.72
3.	Chloroform fraction (CHF)	2.37
4.	Acetone fraction (ACF)	1.4
5.	Ethanol fraction (ETF)	4.5
6.	Methanol fraction (MEF)	3.37
7.	Aqueous fraction (AQF)	6.65

Table 2. Phytochemical Screening of *J.gossypifolia* Stem

S. No	Test	Presence /Absence
1.	Alkaloids (Drangendroff's Test)	+
2.	Phenolic compounds (PbCH ₃ COO Test)	+
3.	Flavonoids (Shinoda's Test)	+
4.	Glycosides (Killer Kiliani's Test)	+
5.	Terpenoids (Salkowski Test)	+
6.	Tannins (FeCl ₃ Test)	+
7.	Saponins (Frothing Test)	+

3.1. X-Ray Diffraction (XRD)

X-Ray Diffraction is a powerful, non-destructive technique for characterizing crystalline materials. It provides information on structures, phases, preferred crystal orientation and other structural parameters such as average grain size, crystallinity and crystal defects. The characteristic x-ray diffraction pattern generated in a typical XRD analysis provides a unique "fingerprint" of the crystals present in the sample. When properly interpreted, by comparison with standard reference patterns, this fingerprint

allows identification of the crystalline form. The particle size of the prepared samples were determined

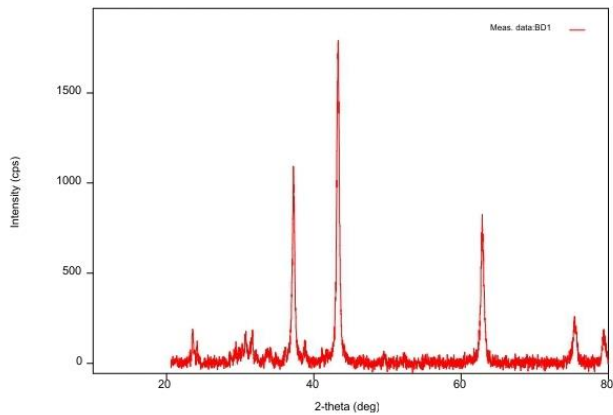


Figure 2. XRD spectrum of MgO Nano Particles

by using Scherrer's equation as follows $D \approx 0.9\lambda/\beta\cos\theta$ where D is the crystal size, λ is the wavelength of X-ray, θ is the Bragg's angle in radians and B is the full width at half maximum of the peak in radians (Figure 1).

3.2. Fourier Transform Infrared Spectroscopy (FT-IR)

The interaction of surface biological group with the metal can be studied by the valuable analytical tool FT-IR. The sample is characterized by FT-IR within the wavelength range of 800-20000 cm^{-1} . Infrared spectroscopy is an important technique inorganic chemistry. It is an easy way to identify the presence of certain functional group in a molecule. This technique was applied to determine the group that was present in the surface of nanoparticles. The FT-IR spectrum of NiO nanoparticles exhibit peaks at 1570 cm^{-1} , 1370 cm^{-1} , 1100 cm^{-1} , 942 cm^{-1} , 884 cm^{-1} , 844 cm^{-1} and 803 cm^{-1} . A peak at 1570 cm^{-1} corresponds to N-O stretching (Figure 3). A vibration at 1370 cm^{-1} corresponds to C-H bending methyl. A peak at 1570 cm^{-1} corresponds N-O stretching. A peak at 1100 cm^{-1} corresponds to C-O stretching of Carbonyl. The bond appeared at 803 cm^{-1} could be assigned for the C-H bending vibration.

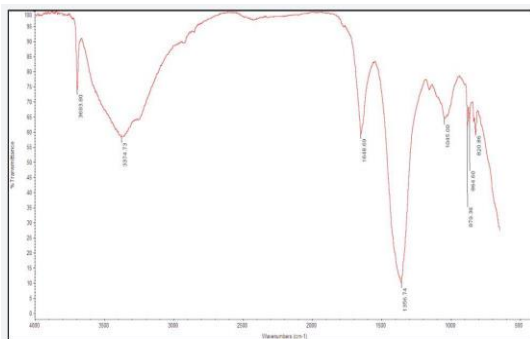


Figure 3. FT-IR spectrum A: MgO Nano Particles using *Jatropha gossypifolia* stem extract, B: Nanoparticles.

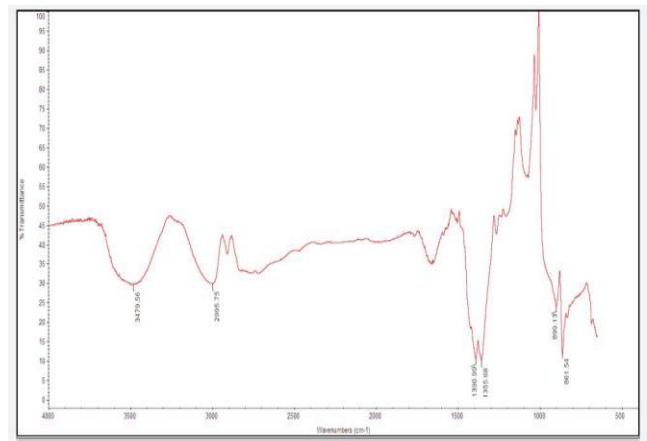


Figure.4. XRD spectrum of MgO nanoparticles using *Jatropha gossypifolia* Stem extract

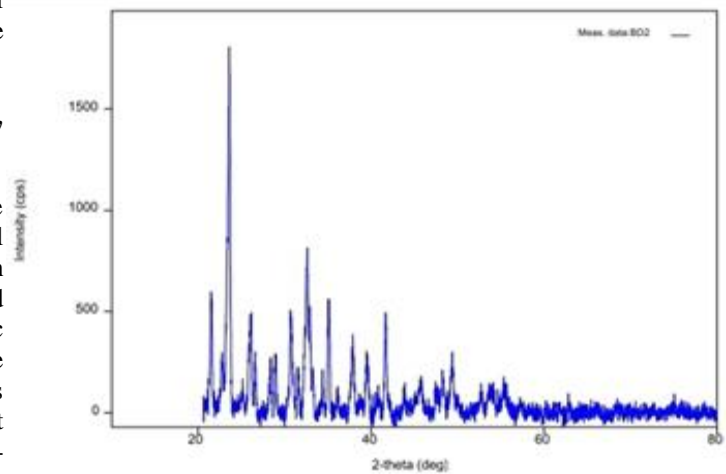


Figure 5. Phytoconstituents Screening Test

antibacterial Activity of Stem of *Jatropha Gossypifolia* (L.)

3.3. Assays for Antibacterial Activity

Bacterial strains of *Staphylococcus aureus*, *Bacillus subtilis*, *Bacillus cereus* and *Klebisella* were procured from Department of Microbiology, Vivekanandha College of Arts and Sciences for Women (Autonomous), Tiruchengode. Disc diffusion method [15] was used to assess the effect of plant extracts on bacteria. The plant extract were added at concentration of 2.5, 5, and 10 % (dissolved in respective solvents) to the aseptic medium and agar plates were prepared. All the tested microorganisms were grown previously in the same agar plates individually. Disc without plant used as control. The plates were left for aerobic incubation at 30°C for 16 hrs and growth of the organisms was observed in treatments. Presence (+) or absence (-) of antibacterial activity and the size (diameter) of the inhibition zones was also need in respective of solvent.

Antibacterial activity of MgO Nanoparticles prepared by using *Jatropha Gossypifolia* (stem) extract. Anti-Oxidant activity of MgO nanoparticles were carried out to test the bacterial efficiency of the nanoparticles. It has demonstrated that highly reactive metal oxide nanoparticles exhibit excellent biocidal action against gram positive and gram negative bacteria. The toxicity is depends on both the exposure and size of metal nanoparticle. The test was carried out using different concentrations of the nanoparticles by disc diffusion method. MgO nanoparticles exhibit higher zone of inhibition against the bacterial pathogens such as *Staphylococcus aureus*, *Bacillus subtilis*, *Bacillus cereus* and *Klebisella*.

4. Conclusion

The development of novel compounds with phytochemical activity is an urgent need. In this work, we used easily available, inexpensive starting materials and procedure is easy to carry out in any laboratory. Use of toxic reagents is avoided. Here we have reported eco-friendly synthesis of MgO Nanoparticles using *Jatropha gossypifolia* stem extract. It has been used as reducing agent and also a capping agent in the MgO Nanoparticle synthesis. This process was completely undertaken through green synthesis route. The freshly prepared stem extract was added to 14.5g Magnesium nitrate and the reaction takes place at room temperature. The synthesized Nanoparticles were of rod shaped and the particles sizes were 25-35nm. In this study, green synthesis methods were pollutant free and eco-

friendly. The traditional medicine of *Jatropha gossypifolia* extract with Magnesium nitrate results in the formation of Magnesium oxide Nanoparticles. Magnesium oxide Nanoparticles characterized by UV-VIS, FT-IR and XRD. Further, these synthesized MgO Nanoparticles from *Jatropha gossypifolia* shows antibacterial activity on both gram positive and gram negative bacteria which could be excellent antibacterial agent to treat diseases caused by bacteria.

Conflicts of Interest

There are no conflicts of interest.

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Compound	Name of microorganisms employed			
	Gram Positive Bacteria (+) (mm) (Zone of inhibition (mm))			Gram Negative Bacteria (-) (Zone of inhibition (mm))
	<i>Bacillus subtilis</i>	<i>Bacillus cereus</i>	<i>Staphylococcus aureus</i>	<i>Klebsiella species</i>
Nanoparticles	4.1	3.6	6.2	3.5
Streptomycin (Antibiotics)	5.9	4.9	5.2	5.3

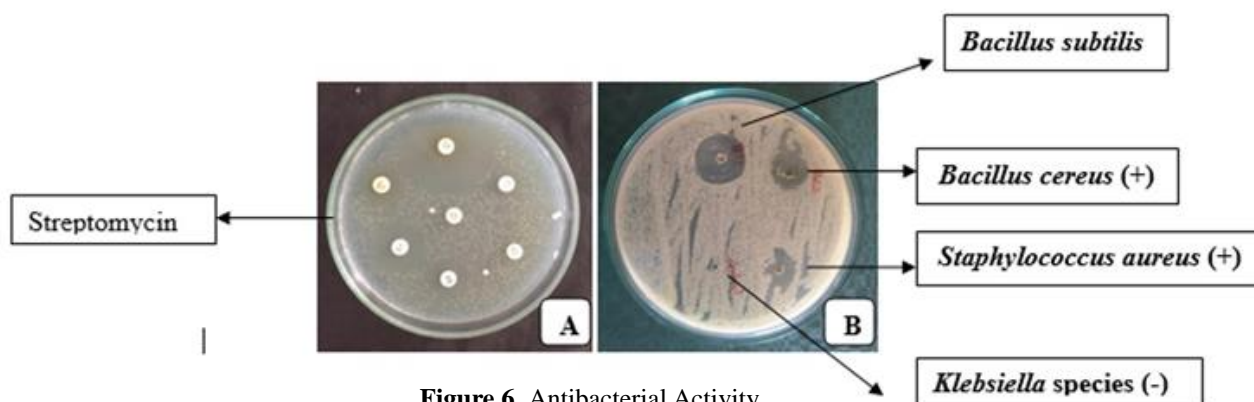


Figure.6. Antibacterial Activity

Table 1. Antibacterial activity of Chemical name using *Streptomycin* as a standard drug - zone of inhibition

Compound Name	Name of microorganisms employed			
	Gram Positive Bacteria (+) (Zone of inhibition (mm))			Gram Negative Bacteria (-) (Zone of inhibition (mm))
	<i>Bacillus subtilis</i>	<i>Bacillus cereus</i>	<i>Staphylococcus aureus</i>	<i>Klebsiella species</i>
Nanoparticles	4.3	4.6	5.2	4.5
Ciprofloxacin	5.9	3.9	4.2	4.3

Table 2. Antibacterial activity of Chemical name using *Ciprofloxacin* as a standard drug – zone of inhibition

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