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Phytochemical Analysis of *Simarouba glauca* Dc. and Comparison of its Bioactivity

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Authors' contributions

This work was carried out in collaboration among all authors. Authors KRP, SNH and GBJ designed the study, performed the statistical analysis, wrote the protocol and wrote the first draft of the manuscript. Authors ANP, SGW and AAD managed the analyses of the study. Author MBP managed the literature searches. All authors read and approved the final manuscript.

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ABSTRACT

Medicinal implications of plants are well known to us since the time immemorial and are well documented in the ancient literature. According to WHO report, 80% of world populations count on herbal medicines to treat themselves in case of illness. And the main reason behind this could be the side effects caused by the drugs which are recommended now a days by the general practitioners. Herbal medicines are known to produce negligible amount of side effects and that is why they are among the favorite candidates for health care. In the present study, we have attempted to rediscover the medical potential of the *Simarouba glauca. W*e have performed phytochemical analysis of *Simarouba glauca* and compared its bioactivities (antibacterial, antifungal, anti-diabetic, hemolytic, antioxidant, and antimicrobial)with synthesized zinc oxide nanoparticles.

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Keywords: Simarouba glauca; antibacterial; antifungal; anti-diabetic; antioxidant.

1. INTRODUCTION

The potential of plant to serve human beings in wellness of health and overall immunity and range of aspects has been well documented since former times [1]. According to World Health Organization, 80% of world'spopulation mainly counts on traditional medicines for health care. Plant products have been a part of phytomedicines since time immemorial [1,2]. There are about 2000 species of plants that are used for medicinal purposes and detected as having a therapeutic significance. Many of these medicinal plants, which are known to the humans since ancient time, either are on the verge of extinction or already listed as endangered. Lots of efforts are already in progress in the different corners of the world to conserve these plants and/or to enhancetheir ability to produce extra amount of medicinal compounds they are producing either by plant tissue culture techniques [3-7] or by the use of recently developed advanced techniques like Clustered Regularly Interspaced Short Palindromic Repeats with CRISPR associated protein 9(CRISPR/Cas9) [8,9]. Medicinal plants play a vital role in preventing various diseases [10] and their medicinal value lies in some bioactive constituents (like flavonoids and phenols) that produce a definite physiological action on the human body [1,11]. It has been estimated that 14 - 28% of higher plant species are used medicinally and that 74% of pharmacologically active plant derived components were discovered [12]. Out of the several hundred thousand medicinal plant species around the globe, only a small portion has been investigated so far both phytochemically and pharmacologically [12].

Plants are enriched with various phytochemicals such as vitamins, terpenoids, phenolic acids, lignins, stilbenes, tannins, flavonoids, quinones, coumarins, alkaloids, amines, betalains and other metabolites, which are rich in antioxidant activity [13]. Studies have shown that many of these antioxidant compounds possess anti-inflammatory, antiatherosclerotic, antitumor, antimutagenic, anticarcinogenic, antibacterial, and antiviral activities [14].

Phytochemicals are chemicals, which are naturally present in plants. Recently phyto chemicals have become more popular due to their countless medicinal uses. Phytochemicals play a vital role against number of diseases such as asthma, arthritis, cancer etc. unlike pharmaceutical chemicals these phytochemicals do not have any side effects. Phytochemicals are derived from different parts of plant such as leaves, flowers, roots, pulps, seeds, etc. [15]. Phytochemicals are also known as "Man friendly medicines" as they cure diseases without causing any harm to human beings [15]. To produce synthetic molecular analogs, a large proportion of plant based compounds are used as lead molecules in drug discovery which implies that phytochemicals play a crucial role in diversity oriented synthesis of natural product like pharma compounds [16]. It is a rain fed waste land evergreen edible oil tree. *Simarouba glauca* belongs to the family Simaroubaceae. It contains various groups of chemical components [17]. Quassinoids is the main active group of chemicals in *Simarouba glauca* which belongs to triterpine family [16]. Pharmacological properties of *Simarouba glauca* are haemostatic, antihelmenthic, antiparasitic, antidysentric, antipyretic and anticancerous. *Simarouba glauca* is a native species to Florida in the United States, southern Florida, South America, and the Lesser Antilles [18]. *Simarouba glauca* is indigenous to the rainforest and other tropical areas in Mexico, Cuba, Haita and Central America. Its cultivation depends on rainfall distribution, water holding capacity of the soil and sub-soil moisture. *Simarouba glauca* is found in several cultivated parts of India and was introduced in India during 1960s by plant genetic resources in the research station at Amravati in Maharashtra and University of agriculture science Banglore in 1986 [19]. *Simarouba glauca* is suited for temperature range of 10 to 50°C. It can grow at elevations from sea level to 1,000 m (3,300 ft). It grows 40 to 50 ft (12 to 15 m) tall and has a span of 25 to 30 ft (7.6 to 9.1 m) [18]. It has bright green leaves, small white flowers and small red fruits [16]. Leaves of *Simarouba glauca* are bluish oily green and smooth at apex and are pinnately compound. It has 3-21 leaflets. Simarouba leaf decoction when taken in limited amounts can raise the immunity, reduce appetite loss and increase quality of life in cancer patients [18]. *Simarouba glauca* has very good anti-bacterial, anti-tumorous properties so it is very effective in reducing the size of tumors and secondary infections in cancer patients and in curing cancer of first/second stages, whereas in later stages it can considerably increase the quality of life [18].Nanotechnology involves the use of materials having nanoscale dimensions in

the range of 1–100 nm [20]. Nanomaterials are particles having nanoscale dimension, and nanoparticles are very small sized particles with enhanced catalytic reactivity, thermal conductivity, non-linear optical performance and chemical steadiness owing to its large surface area to volume ratio [21,22].

The nanomaterials can be synthesized by different methods including chemical, physical, irradiation and biological methods. The development of new chemical or physical methods has resulted in environmental contaminations, since the chemical procedures involved in the synthesis of nanomaterials generate a large amount of hazardous byproducts [23,24]. Hence the green synthesis methods for the nanoparticle synthesis are being used now a day's which are clean, safe, eco-friendly and environmentally nontoxic. Moreover, in this method there is no need to use high pressure, energy, temperature and toxic chemicals [24,25,26]. Nanoparticles possess a wide array of application in the different fields' viz., medicine, electronics, and therapeutics and as diagnostic agents [24]. Nano-particles have started being considered as nano-antibiotic because of their antimicrobial activity [22]. They also have potential application in the field of medicine like drug discovery, biological activities such as anti-microbial, antioxidant etc. and diagnosis of diseases [20].

The Zinc oxide nanoparticles have found fabulous applications in bimolecular, diagnostics and microelectronics [24], electromagnetic coupled sensors [20]. The Zinc oxide nano particles have been used to remove arsenic and sulphur from water even through bulk zinc oxide cannot absorb arsenic. It is because nanoparticles have much larger surface areas than bulk particles [27]. They have fascinating properties and extensive applications. Bio-inspired synthesis of the zinc oxide nanoparticles has been achieved using environmentally and eco-friendly accepted systems.

In this research work, we have performed phytochemical analysis of *Simarouba glauca* and compared its bioactivities with synthesized zinc oxide nanoparticles.

2. MATERIALS AND METHODS

2.1 Collection and Preparation of Plant Material

Simarouba glauca leaves were collected from local areas of Aurangabad, MS, India. After collection of leaves they are cleanedwith distil waterto remove all the impurities and unwanted materials. Dried to remove all the moisture content present in leaves so that it can be stored for further analysis. Complete drying of leaves is followed by grinding to convert them in powder form for further use.

2.2 Preparation of Plant Extract

Methanol extract: For the methanol extract, 200 ml of methanol and 20 gm of dry powder placed in soxhlet extractor. Extraction was carried out for 3hrs and 9 cycles were performed. Dark green colour extract was obtained.

Ethyl acetate extract: 200 ml of ethyl acetate and 20 gm of dry powder were placed in soxhlet extractor. Extraction was carried out for 3 hrs and 9 cycles were performed. Dark green color extract was obtained. Methanol and ethyl acetate extract were evaporated using rotary evaporator and powder obtained was collected for further analysis. The powder was then kept in sterile bottles, under refrigerated conditions, until further use.

Preliminary phytochemical analysis: The extracts were subjected to preliminary phytochemical testing to detect for the presence of different chemicals or compounds. *S. glauca* leaves extract were screened for the presence of alkaloids, flavonoids, carbohydrates, glycosides, tannins, terpenoids, saponins and steroids.

Test for alkaloids: Mayer's test: In 1 ml of extract add 1 ml of Mayer's reagent. Appearance of creamy white precipitation indicates the presence of alkaloids.

Test for flavonoids: In 5 mg of extract powdered 10 ml of sodium hydroxide. Yellow fluorescence indicates the presence of flavonoids.

Test for terpenoids: Salkowski's test: In 5 ml of extract add 2 ml of chloroform and 3 ml of concentrated sulphuric acid. Reddish brown colour indicates the presence of Terpenoids.

Test for steroids: In 5 mg of extract add 2 ml of acetic acid unhydride and 2 ml of sulphuric acid. Green colour indicates the presence of steroids.

Test for saponins: 1 mg of extract was mixed with 5 ml of distilled water and shaken well. Appearance of creamy white miss bubbles indicates the presence of saponins.

Test for tanins: 5 mg of extract was mixed with 3 ml of distilled water and heated on a water bath. The mixture was filtered and add 5 mg of ferric chloride to the filtrate. Dark green colour indicates the presence of tanins.

Test for carbohydrates and glycosides: Benedict's test: In 2 ml of extract add 2 ml Benedict's reagent, heat in boiling water bath for 5 min, appearance of precipitation indicates the presence of glycosides.

2.3 Qualitative Analysis of Phyto-chemicals by Thin Layer Chromato-graphy

In the TLC analysis, 1 cm from the edge of the thin layer plate for a straight line across the plate. Spot the extract in the line with the capillary tube to dry the spot and place the plate in a beaker with a mobile phase. Layer the beaker when the solvent reaches more than 1 cm from the top of the plate. Remove the beaker from the solvent line before the Rf values have been calculated.

2.4 Synthesis of Zinc Oxide Nanoparticles

For the synthesis of zinc oxide nanoparticles, prepare 1 mM of Zinc acetate in 50 ml of sterile distilled water. 20 ml of 0.1 M sodium hydroxide and 25 ml plant leaf extract was then addedto the 1 mM Zinc acetate solution. Thissolution was then kept incubated on magnetic stirrer for 3 hrs. Appearance of yellow color indicates the presence of Zinc oxide nanoparticles. This mixture was then centrifuged to separate supernatant and pellete. Supernatantwas discarded and pellete was dried in the hot air oven. Dried powder was then preserved in an airtight bottle for further studies. Characterization of zinc oxide nanoparticles was carried out by using UV visible spectrophotometry.

2.5 Antibacterial Activity

In vitro antibacterial activities were examined for methanol, ethyl acetate extract and zinc oxide nanoparticles. Antibacterial activities of plant leaf extracts and zinc oxide nanoparticles against three pathogenic bacteria (*E. coli, S. auerus, and Bacillus subtilis*) were investigated by the well diffusion method. The petriplateswere poured with approximately 25 ml autoclaved nutrient agar media each. Agar plates were inoculated with three different bacterial colonies and spread with sterile swab. Four wells were punched on each plate using a sterile cork borer of 8 mm diameter. 100 µL of each extract was pipetted into respective wells. Standard leaf extract was used as a control.The Petri plates were incubated overnight at 37°C and the anti-bacterial activity was measured after 18 hrs of incubationby measuring the zone of inhibition.

2.6 Antifungal Activity

In vitro antifungal activities were examined for methanol, ethyl acetate extract and zinc oxide nanoparticles. Antifungal activities of plant leaf extracts and zinc oxide nanoparticles against pathogenic fungus (*Asperillusflavus)* were investigated by the well diffusion method. All plates were autoclaved with PDA agar media of approx. 25 ml. Aspergillus flavus colonies were inoculated with agar plates and sterile swab spread. Each plate was hit with four wells using a sterile 8 mm diameter cork borer. The pipetting into respective wells was 100 µL of each extract. Standard leaf extract was used as a control. The Petri plates were incubated overnight at 37°C and the antifungal activity was measured after 48 hrs of incubationby measuring the zone of inhibition.

2.7 Haemolytic Assay

The haemolytic activity of the extract was determined using agar diffusion technique on blood agar plate. Blood agar was prepared and well measuring 8 mm were made on the agar using cork borer. The wells were filled with 100 µL of different concentration of plant extracts solution and zinc oxide nanoparticles solution. Standard leaf extract was used as a control. The plates were then incubated at 37ºC for 24 hrs and the haemolytic activity was measured after 24 hrs of incubationby measuring the zone of inhibition.

2.8 Antioxidant Activity

Scavenging of H_2O_2 radicals: The ability of the extracts to scavenge H_2O_2 was determined according to the reported method (reference). A solution of H_2O_2 (40 mM) was prepared in phosphate buffer (pH 7.4). Extract of selected concentrations in distilled water were added to H_2O_2 solution (0.6 mL, 40 mM). The absorbance of H_2O_2 at 230 nm was determined after 10 minutes against a blank solution containing phosphate buffer without H_2O_2. Ascorbic acid was used as a standard antioxidant. The %

of H_2O_2 scavenging of both extracts and standard compounds were calculated. % scavenged $[H_2O_2]$ = [(A−A)/A] x 100 where A was the absorbance of the control and A was the absorbance in the presence of the sample of extract (rewrite this formula correctly again).

DPPH (1,1-Diphenyl -2-picryl hydarzil) free radical scavenging activity: 0.1 mM solution of DPPH in ethanol was prepared. 500 µg of sample was added in 1 ml of solution. The mixture was shaken vigorously and allowed to stand room temperature for 30 min. Then the absorbance was measured at 517 nm by using UV visible spectrophotometer. Lower absorbance of reaction mixture indicates higher free radical scavenging activity. The % DPPH scavenging effect was calculated using following equation.The capability to scavenge the DPPH radical was calculated using the following equation: DPPH scavenging effect (%) = [(A0 – A1)/A0) × 100], where A0 is the absorbance of the control reaction, and A1 is the absorbance of presence of all of the extract samples and standard.

Antidiabetic activity: The antidiabetic activity of the extract was determined using starch agar plates. Starch agar plates were prepared by using 0.1% starch and 1% nutrient agar. After that 5wells measuring 8 mm were made on the agar plates using cork borer. The wells were filled with given concentrationsof phosphate buffer and amylase enzyme and sample as indicated below.

1st well – 40 ul amylase + 60ul phosphate buffer = 100 ul

2nd well – 100 ul buffer

3rd well – 40 ul amylase + 40 ul phosphate buffer + 20 ul sample = 100 ul

4th well – 40 ul amylase + 20 ul phosphate buffer + 40 ul sample = 100 ul

5th well – 40 ul amylase + 60 ul sample = 100 ul

3. RESULTS AND DISCUSSION

3.1 Preliminary Phytochemical Analysis

Phytochemical screening is a preliminary step in the characterization of plant products. Preliminary screening for presence or absence of a particular class of compound is qualitative in nature and form a robust base for the quantitative estimation of bioactive components. Phytochemicals were screened for Alkaloids, Carbohydrates, Glycosides, Coumarins, Proteins & Amino acids, Phenolic compounds, Tannins, Flavonoids, Terpenoids.

Qualitative phytochemical investigation of extract of *Simarouba glauca* leaves showed the presence of saponins, tannins, flavonoids, glycosides, alkaloids, steroids, terpenoids. Methanol and ethyl acetate extract of *Simarouba glauca* leaves contain maximum number of phytoconstituents. The results of qualitative phytochemical analysis areshowed in the Tables 1, 2, 3.

Table 1. Preliminary phytochemical analysis of methanolic and ethyl acetate extracts

Sr No.	Phytochemicals	Methanol extract	Ethyl acetate extract
1	Alkaloids	+	+
2	Glycosides	+	+
3	Flavonoids	+	+
4	Terpenoids	+	+
5	Steroids	+	+
6	Saponins	-	+
7	Tannins	+	+

3.2 Qualitative Determination of Bioactive Compounds by Thin Layer Chromatography

Methanol and Ethyl acetate extract obtained from *S. glauca* leaves was used to perform thin layer chromatography in order to establish the purity and composition of materials. The purified components of Methanolic and Ethyl acetate extract of S. glauca leaves were identified by iodine test. For different components different bands were observed under ultraviolet light (Figs. 1, 2).

3.3 Synthesis of Zinc Oxide Nanoparticles

In visual observation colour of zinc acetate solution was changed to yellow after addition of *simarouba* leaf extract and yellowish precipitate indicates the formation of zinc oxide nanoparticles. For further confirmation UV visible spectroscopy was carried out.

UV-visible spectroscopy is usually conducted to confirm the synthesis of zinc oxide nanoparticles. The absorption spectrum was recorded for the

sample in the range of 200 – 400 nm. The spectrum showed the absorbance peak at 310 nm corresponding to the characteristic band of zinc oxide nanoparticles.

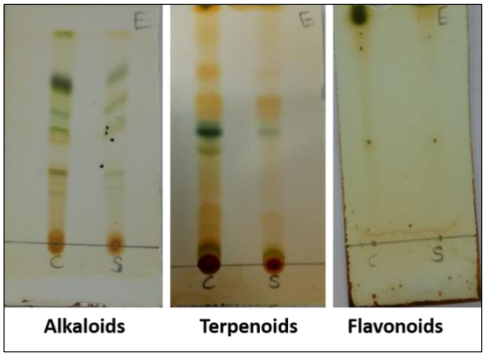

Fig. 1. TLC for ethyl acetate, showing the separation of alkaloids, Teroenoids and flavonoids

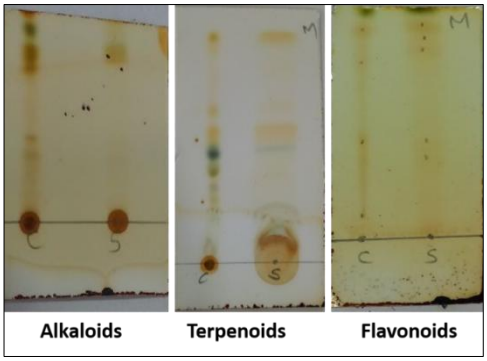

Fig. 2. TLC for methanol, showing the separation of alkaloids, teroenoids and flavonoids

3.4 Antibacterial Activity

The results of antimicrobial activity of methanol, ethyl acetate extracts and zinc oxide nanoparticles of *E. coli, S. auerus, and Bacillus subtilis* are given in Table 4. From the results it was found that zinc oxide nanoparticles have widerinhibitory zone as compared tomethanol and ethyl acetate extracts. Ethyl acetate extract have the least inhibitory zone. These results clearly indicate that zinc oxide nanoparticles have enhancedantibacterial activity as compared to methanol and ethyl acetate extracts(Tables. 3, 4) (Figs. 3, 4).

3.5 Antifungal Activity

The antifungal assay showed that *Simarouba glauca* has antifungal property against the tested fungi *Aspergillus Niger*. In this analysis, zinc oxide nanoparticles were found to be more effective as compared to methanolic and ethyl acetate extracts against the growth of fungi. The results of antifungal activity of methanolic, ethyl acetate extracts and zinc oxide nanoparticles of *Aspergillus flavus*is given in Table 5.

3.6 Haemolytic Assay

The zone of haemolysis was directly proportional to concentration of the extract. From the results it was found that zinc oxide nanoparticles (Table. 6) have enhancedzone of haemolysisas compared tomethanol and ethyl acetate extracts (Fig. 5).

3.7 Antioxidant Activity

Antioxidant acitivity of the zinc oxide nanoparticles, methanolic and ethyl acetate extracts from the plant leaf *SSimarouba glauca was* carried out with hydroxyl radical scavenging assay and DPPH scavenging activity.

3.8 Antidiabetic Activity

Amylase inhibition detection was carried out using starch agar plates technique. The amylase activity inhibition zone appeared around the wells having zinc oxide nanoparticles, methanolic and ethyl acetate extract (Table. 7) (Fig. 6).

A large number of biologically active plant constituents have been found in the Simarouba glauca extract in the phytochemical analysis. The result showed variations in the number of phytochemicals obtained from both extracts. More than 124 metabolites were detected in the ethyl acetate extract compared to both solvent extracts. This may be due to the high solubility of the active compounds of Simarouba glauca with ethyl acetate by extraction compared to the petroleum ether solvent.

In addition to these results, it is also documented that different light intensities have various effects on the efficiency of the metabolites produced by the plants as seen in case of *Glechoma longituba* [28]. Zang et al. (2015) explained, chloroplasts produces different metabolites, and chloroplasts development mainly depends on the different light intensities [29,30,31]. Similar kind of effects can also be possible in case of *Simarouba glauca* and appropriate light intensity management might be feasible to obtain higher yields of secondary metabolites in it, which might

be helpful in improving its either antibacterial, antifungal, anti-diabetic, hemolytic, antioxidant, and antimicrobial. Moreover,*S. glauca*and similar kind of medicinal plants' specific antimicrobial activity isnowwell established, so one can also make the efforts to do the molecular studies, trace back the proteins and genes mainly responsible for these effects and clone them with easy, cheapand quick methods of cloning [32] for further studies by considering its commercialization in mind. Producing these antimicrobial components in bacteria is easy and quick and additionally we do not have to destroy plants for this, which will be environment friendly and is the absolute need of the current time.

Table 2. Ethyl acetate

Parameters	Stationary phase	Mobile phase	No. of spots	R_f value
Alkaloids	Silica gel	Benzene : ethanol(45:5)	10	0.04, 0.22, 0.27, 0.31, 0.36, 0.47, 0.54, 0.61, 0.75, 1
Glycosides	Silica gel	Ethyl acetate : butanol : water(20:20:15)	5	0.16, 0.30, 0.66, 0.76, 0.95
Flavonoids	Silica gel	Ethyl acetate : formic acid : glacial acetic acid : water (20:2:2:5)	4	0.42, 0.81, 0.87, 0.93
Terpenoids	Silica gel	Toluene : ethyl acetate	11	0.65, 0.09, 0.13, 0.21, 0.34, 0.44, 0.50, 0.59, 0.63, 0.73, 0.98

Table 3. Methanol

Parameters	Stationary phase	Mobile phase	No. of spots	R_f value
Alkaloids	Silica gel	Benzene : ethanol(45:5)	7	0.29, 0.37, 0.72, 0.95, 0.78, 0.81, 0.89
Glycosides	Silica gel	Ethyl acetate : butanol : water (20:20:15)	2	0.35, 0.91
Flavonoids	Silica gel	Ethyl acetate : formic acid : glacial acetic acid : water (20:2:2:5)	5	0.10, 0.40, 0.75, 0.89, 0.95
Terpenoids	Silica gel	Toluene : ethyl acetate (9:1)	7	0.27, 0.33, 0.41, 0.49, 0.56, 0.66, 1

Table 4. Antibacterial activity and zone of inhibition for respective sample

Sr. no.	Extracts	Conc. In mg/ml	Diameter of zone of inhibition in cm		
			E. coli	***S. auerus***	***B. subtilis***
1	Methanol	1	1.1	1.1	1.1
		2	1.2	1.1	1.2
		3	1.2	1.2	1.3
		4	1.2	1.2	1.3
2	Ethyl acetate	1	1.1	1.1	1.1
		2	1.1	1.1	1.1
		3	1.1	1.1	1.1
		4	1.1	1.1	1.1
3	Zinc oxide nanoparticles	1	1.2	1.1	1.3
		2	1.3	1.3	1.4
		3	1.4	1.5	1.5
		4	1.5	1.5	1.5

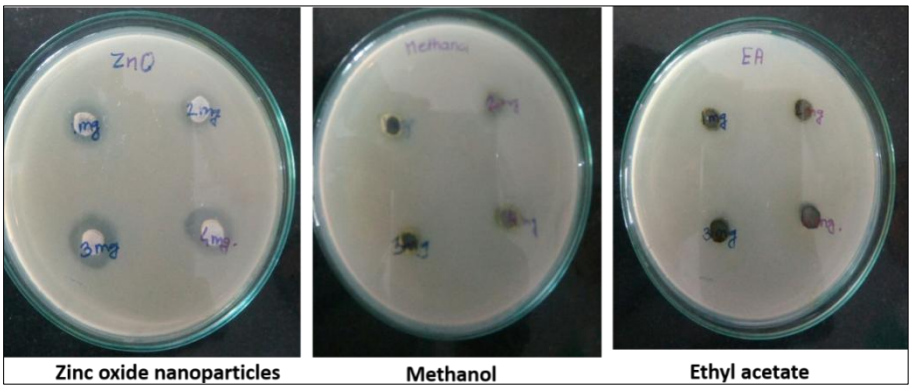

Fig. 3. Antibacterial activity, in against from left zinc oxide nanoparticals, methanol, ethyl acetate

Table 5. Antifungal activity and zone of inhibition for respective sample

Sr. no.	Extracts	Conc. In mg/ml	Diameter of zone of inhibition in cm
			Aspergillus flavus
1	Methanol	1	1.1
		2	1.2
		3	1.2
2	Ethyl acetate	1	1.1
		2	1.1
		3	1.1
3	Zinc oxide nanoparticles	1	1.2
		2	1.3
		3	1.5

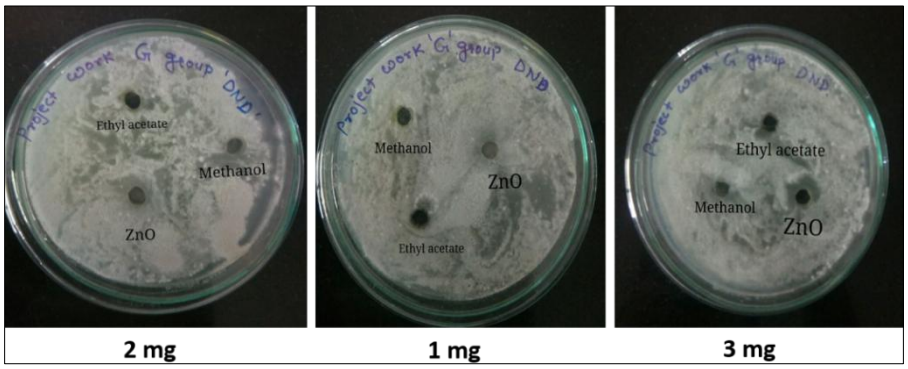

Fig. 4. Antifungal activity at different concentration level

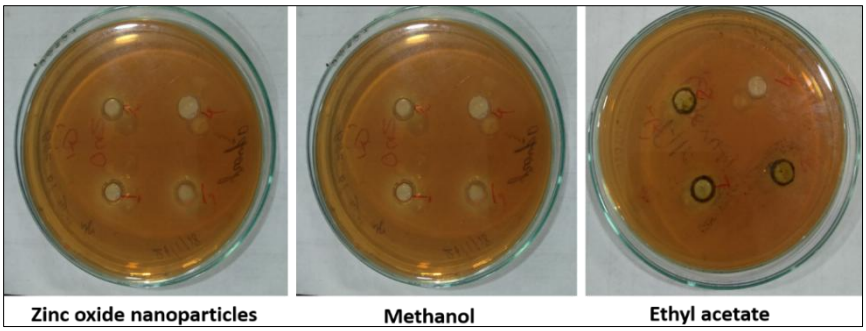

Fig. 5. Haemolytic activity in against from left zinc oxide nanoparticals, methanol, ethyl acetate

Table 6. Zones of haemolysis for respective sample

Extracts	Zones of haemolysis in mm		
	Conc. of extracts in mg/ml		
	1 mg/ml	2 mg/ml	3 mg/ml
Methanol	1.2	1.3	1.4
Ethyl acetate	1.2	1.2	1.4
Zinc oxide nanoparticles	1.3	1.3	1.4

Table 7. Zones of antidiabetic activity in against from left zinc oxide nanoparticals, methanol, and ethyl acetate

Extracts	Zones in cm				
	Well no.				
	1	2	3	4	5
Methanol	1.2	-	1.1	1	-
Ethyl acetate	1.5	-	1.2	1.2	-
Zinc oxide nanoparticles	1.2	-	1.2	1.1	-

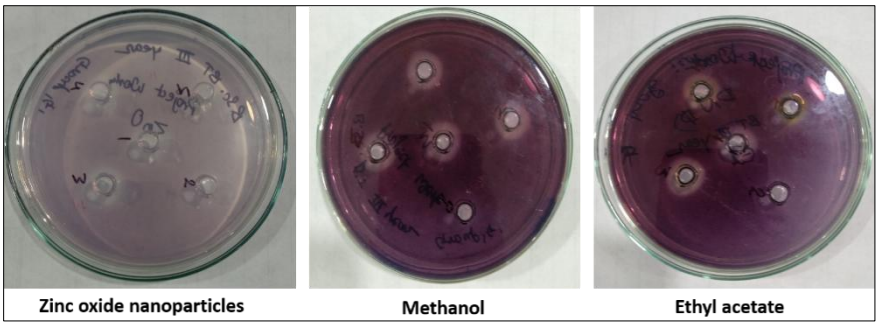

Fig. 6. Antidiabetic activity in against from left zinc oxide nanoparticals, methanol, and ethyl acetate

The present study revealed that the *S. glauca* leaf posess antimicrobial activity, which is big plus of this study. Currently the world is facing the problem of antimicrobial resistance and pathogenic bacteria are developing resistance to the commercially available antibiotics. Therefore, it is an absolute necessity to keep looking for new antimicrobial solutions. And who know this *Simarouba glauca*extract might be a solution to nullify the effect of antibiotic resistance in bacteria and could become a most favored medicine in the near future.

4. CONCLUSION

Based on our findings we conclude that *Simarouba glauca* plant leaves have significant amount of phytochemical compounds. Zinc oxide nanoparticles of *Simarouba glauca* have shown enhanced *in vitro* antioxidant, antibacterial activity, haemolytic activity, Antifungal activity compared to methanolic and ethyl acetate extracts of *Simarouba glauca.* The present study revealed that the *S. glauca* leaf possessantimicrobial activity, which is big plus of this study. Currently the world is facing the problem of antimicrobial resistance and pathogenic bacteria are developing resistance to the commercially available antibiotics. Therefore, it is an absolute necessity to keep looking for new antimicrobial solutions. And who know this *Simarouba glauca*extract might be a solution to nullify the effect of antibiotic resistance in bacteria and could become a most favored medicine in the near future.

CONSENT

It is not applicable.

ETHICAL APPROVAL

It is not applicable.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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