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Estimation of Antioxidant, Antimicrobial and Antifungal Activity of Nelumbo nucifera

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ABSTRACT

Background: *This study's goal was to conduct preliminary phytochemical screening, physicochemical screening, determine the total quantity of flavonoids and phenols, and antioxidant activity (by DPPH, PRA) antimicrobial activity (by disk diffusion method, MIC) and antifungal activity (by Inoculum, MFC).*

Material and methods: *Physicochemical and phytochemical screening of Nelumbo nucifera, testing of TPC, TFC in extract. Antioxidant activity was determined using the DPPH method and power reducing assay. Antibacterial effect was observed determined utilizing the disk diffusion procedure and MIC, and Antifungal activity was determined using of inoculum method and MFC.*

Results: *TPC extract was determined at the level of 25.12 ± 3.58 mg/g, while TFC extract was determined at the level of 34.23 ± 4.84 mg/g. The DPPH activity test shows IC50 34.05342 ±, reducing power assay was determine at the level of 2.02 ± 0.05, the disk diffusion method shows zone of inhibition was 12.5mm and 12.6mm and MIC was shows in 100mg/ml and the inoculum shows zone of inhibition was 11mm and 10.7 mm, and MFC was shows in 100mg/ml.*

Conclusion: *It was assessed that Nelumbo nucifera's methanolic extract has an antioxidant, antimicrobial and antifungal characteristics.*

Keywords *– Green price, green advertisement, green perceived value, environmental knowledge, green purchase intention*

INTRODUCTION

China and India have used the perennial water plant Nelumbo nucifera Gaertn (Nymphaeaceae) as a medicinal herb. For more than 400 years, it has been recorde Substance that inhibits the oxidation of other molecules.d in China's most famous medical text. Traditional medicine makes use of the plant's leaves, seeds, blooms, and rhizome, among other parts. Sore throat, chest pain, spermatorrhea, vitiligo, variola, enteritis, respiratory infection, vomiting blood, nosebleed, coughing up blood, blood in urine, metrorrhagia, bleeding between menstrual periods, high levels of fats in the blood, high body temperature, a severe gastrointestinal infection, liver disease, and excessive thirst belongs to the ailments that ancient medicine has found to benefit from the use of different plant parts. Following traditional claims of N. nucifera's medical properties, researchers have

worked hard to scientifically validate its usage in treating a variety of ailments.[1] Ayurveda and Chinese medicine are two traditional medical systems that use various sections of the lotus plant, from the base to the tip. Substance that inhibits the oxidation of other molecules, anti-swelling, immune-modulating, fever-reducing, germ-fighting, virus-fighting, fungus-fighting, water pills, anti-amnestic, blood clot preventing, antiarrhythmic, medicine for controlling decreasing cholesterol levels in diabetic individuals, managing blood sugar concentrations, anti-weight gain, age-defying, anti-artery-clogging, anti-scarring, tranquilizer, neuroprotective, recollection-enhancing, contraceptive, protects the liver, protective of the skin, heart-protective, and cancer-fighting characteristics are just a few of the biological and pharmacological activities that have been found in the plant as a whole, along with in its raw extracts, parts and elements. The lotus plant's profusion of concerning bioactive properties phytochemicals, comprising of polyphenolic compounds, flavonoids, compounds found in plants that are known for their health benefits, and phenolic acids, another type of beneficial compound, exist in nature, active compounds such as alkaloids and terpenoids, muscle-enhancing drugs, long-chain carboxylic acids, and sugar compounds, has been associated with its remarkable capacity to support health and mitigate illness.[2]

MATERILAS AND METHODS

All chemicals used were of high quality for analysis Methanol, Ethanol, DPPH (1, 1-Diphenyl-2-picryl-hydrazyl), Concentrated sulphuric acid (H2SO4), Butylated hydroxyl toluene (BHT), Phosphate buffer (pH6.6), Potassium ferricyanide (K3Fe (CN) 6), Potassium ferricyanide, Trichloro-acetic acid, Ascorbic acid, Tannic acid , Sodium carbonate, Distilled water, Aluminum chloride, Quercetin standard solution, Sodium nitrite, Sodium chloride, Streptomycin Standard, Gentamycin Standard, Mueller-Hinton agar, Fluconazole Standard, Sabouraud dextrose agar, Folin-Ciocalteu reagent, Ferric chloride (FeCL3), Dragendorff's reagent, Million's reagents, Chloroform, Ninhydrin, Concentrated hydrochloric acid (HCl).

Collection of Plant Material and Preparation of Plant Extract

We gathered Nelumbo nucifera seeds from the local grocery shop of Banmore, Morena M.P. (India) during the month of March, 2024. The plant matter was crushed into tiny fragments, subsequently allowed to atmosphere dry for a duration of 2 days. Once dried, the samples were finely grind into powder and kept in desiccators until the removal process. Utilizing absolute methanol, the extraction was performed in a Soxhlet apparatus for a duration of 10 hours. The solvent was then removed through evaporation Utilizing a rotary evaporator machine, and the resulting crude samples were kept in desiccators.

Percentage Yield:

The weight of the residual extract was measured and percent yield. The calculation was done using the following equation.

$$\text{Percentage yield } \% = \frac{W1}{W2} \times 100$$

Where: W1 amount of dry extract in grams post extraction

W2 = The overall amount of powder in grams used for extraction

Physicochemical screening of plant material

Loss on Drying:

To begin, weigh an empty china dish. Next, weigh the 4 grams of powdered medication to find out how much moisture is present into the sample drug. After that, put the china dish in the hot air oven and preheat it to 105 °C so that the powder's moisture evaporates. And after an hour, take the china dish out of the desiccator with the use of tongs. After cooling to room temperature once more, weigh the china dish and repeat the operation three times or more to ensure a constant weight.[3, 4]

$$\text{Loss on drying } \% = \frac{W2 - W3}{W2 - W1} \times 100$$

Where,

(W1) = Weight of china dish when empty

(W2) = Combined weight of the china dish and the sample prior to drying

(W3) = Total weight of china plate and dried sample combined

Ash value

Total ash Value:
I accurately weighed 4gm of powdered drug into a silica crucible that had been tarred. I incinerated it sample was heated in a muffle furnace at 450°C until all carbon was removed. After cooling the crucible, I weighed it againI computed the proportion of ash compared to the material that has been dried in air.[3, 4]

$$\text{Percentage of Total ash} = \frac{\text{Ash weight}}{\text{Weight of sample}} \times 100$$

Acid Insoluble Ash:
The entire 25ml of 2N HCl was mixed with ash and boiled for a period of time on a water bath, then filtered through ash-free filtering paper. Afterwards, the silica crucible, which had been weighed beforehand, was used to hold the filter paper. It was then burned heating to 450°C in a muffle furnace and maintaining that temperature and all carbon was removed. After cooling, The crucible was a container weighed, and the rate of acid insoluble The amount of ash was determined.[3, 4]

$$\text{Percentage acid insoluble in ash} = \frac{\text{Weight of ash that is not dissolved in acid}}{\text{Sample's mass}} \times 100$$

Initial mass of silica crucible =

After weight of silica crucible + sample =

Extractive Value

Ethanol Soluble Extractive Value:
Approximately 4 grams of coarse powdered drug that had been dried in the air was measured. The substance was soaked in 100millilitres of 90% ethanol within a sealed container for a full day, with regular shuddering for the initial 6 hours. Next, went away from settle for a duration of 18 hours. Afterwards, it was quickly strained while taking measures to avoid losing the liquid used for dissolving substances. 25 ml was measured of the filtered liquid dried on a bottom that is flat tarred plate, heated at a temperature of 105 degrees Celsius, and then measured after evaporation. The proportion of ethanol soluble extractive values were being dissolved determined.[3, 4]

$$\text{Alcohol soluble extractive percentage} = \frac{\text{Residue weight}}{\text{Mass of drug}} \times 100$$

Value of Extractives that Dissolve in Water:
Approximately 4 grams dried out drug in powder form mixed by 100 milliliters of water treated with chloroform sealed flask and left to macerate for a full day. Frequently shuddering during the initial 6 hours. And then left to sit for a period of 18 hours. After that, it occurred quickly strained while making sure not to lose the solvent. 25 milliliters of the filtered liquid became dried in a dish with a flat bottom at a temperature of 105 degrees Celsius until it reached dryness, then measured. The amount of value obtained from a water soluble extract determined as a percentage.[3, 4] The calculation was done by using formula given below.

$$\text{Proportion of water soluble extractive content} = \frac{(W2 - W1) \times Vs}{W \times VF} \times 100$$

Where,

W1 = The weight of a china dish when it is empty

W2 = China dish weight and dried sample

W = Weight of drug

Vs = Solvent taken

VF = Pipette out ¼ of filtrate and transfer to china dish

Phytochemical Screening of Plant Extraction

The extract was analyzed qualitatively for its phytochemical properties. Carbohydrates, proteins, flavonoids, alkaloids, glycosides, amino acids, saponins, steroids, and tannins are present.[5, 6]

Test for Carbohydrates:

Molisch's Test: A few drips of alpha-nephthol are added in ethanol to Two to three milliliters of filtrate, shake, and then include concentrated solution. Pour out sulfuric acid from the edges of the test pipe. A purple circle is created where two liquids meet.[5, 6]

Test for Protein:

Million's Test: Combine 3 milliliters of extract with 5 milliliters of Million's reagents. Bran powder. Heating causes a red precipitate to form, which can either remain solid or dissolve to create a red solution.[5, 6]

Test for Amino Acids:

Ninhydrin Test: 3ml of the filtrate mixed Add 3 droplets of 5% ninhydrin solution and then warm in the water bath for 10 minutes with boiling water. A color that is either purple or bluish has emerged.[5, 6]

Test for Steroids:

Salkowski Reaction: Combine 2 milliliters of extract with 2 milliliters of chloroform and 2 milliliters of concentrated solution. Sulfuric acid with a chemical formula of H2SO4. Mix thoroughly. The chloroform layer changed to a red color as the acid layer showed a fluorescence that was greenish yellow.[5, 6]

Test for Cardiac Glycosides:

Baljet's Test: A portion displayed a yellowish orange after being treated using sodium picrate.[5, 6]

Test for Saponins:

Foam Test: Agitate filtrate vigorously using water. Stable foam that lasts for a long time was noticed.[5, 6]

Test for Flavonoids:

A small residue quantity is subjected to a solution of lead acetate. A solid formed in a chemical reaction is yellow color was produced.[5, 6]

Test for Tannins:

A white precipitate formed in the lead acetate solution.[5, 6]

Test for Alkaloids:

Dragendorff's test: Put 2-3 ml of the filtrate and a small amount of Dragendorff's reagent together. Orange brown precipitate has been produced.[5, 6]

Total Flavonoids and Phenolic Content

a) Determination of Total Flavonoids Content:

Ribarova et al. (2005) utilized the aluminum chloride colorimetric assay to quantify the overall flavonoid content in the sample. Fill a 10 ml volumetric flask containing 4 ml of distilled water, either 1 milliliter of sample (1000 µg/ml) or varying concentrations of quercetin standard solution (25-1000 µg/ml) were added. The above mixture received an the inclusion of 0.3 ml of sodium nitrite (NaNO2) with a concentration of 5%. 5 minutes later, 0.3

milliliters of aluminum chloride (AlCl3) solution with a concentration of 10% was introduced. Administer 2 ml of sodium hydroxide solution with a concentration of 1 M gradually over a period of 6 minutes and adjust the final volume to 10 milliliter using purified water. Thoroughly combine the resolution and measure the absorbance measurement at 510 nm compared to a prepped reagent blank. Extracts are measured in milligrams of quercetin to each gram of extract determine total flavonoid content.[7]

b) Measurement of Overall Phenolic Amount:
The entire phenolic content of the extract was measured using spectrometry (Singleton and Rossi, 1965). Mix 1 ml of tannic acid (10-100 µg/ml) with 1 milliliter of previously weakened (1:20) Reagent known as Folin-Ciocalteu before adding to the 1 milliliter sample (1000 µg/ml) and stirring thoroughly. Pour 4 ml of sodium carbonate (75g/L) into the mixture, then add purified water to reach the overall amount of 10 milliliter in the container and mix well. The blend was abandoned to sit at ambient two hours of exposure to heat. The materials underwent Spin at a speed of 2000 revolutions per minute for 5 minutes, and the the liquid layer above the sediment absorbance occurred measured at a wavelength of 765 nm. Various levels of a substance of tannic acid were accustomed to generate regular curves. Findings were stated to be milligrams of tannic acid equivalents (TAE) for each gram of the extract.[8]

Evaluation of Invitro pharmacological activity

Assessment of antioxidant capacity

The ability of DPPH (1, 1-Diphenyl-2-picryl-hydrazyl) to scavenge free radicals:
The Brand-Williams et al. [2005] described the DPPH method was accustomed to measure the extract's ability to hunt down free radicals. In short, a solution containing 0.1 millimoles of DPPH was made using ethanol. Approximately 3.5 milliliter of DPPH solution was mixed with 0.5 milliliter of obtain the solution at different concentrations dissolved underwater. The combination was vigorously stirred and then left to settle for 30 minutes at normal room temperature. Next, the device known as a spectrophotometer was utilized to assess the absorbance reading at 517 nm. A lower absorbance in the reaction mixture showed increased free radical scavenging activity. Butylated hydroxytoluene (BHT) was used as the benchmark antioxidant. The antioxidant properties of DPPH percentage was determined by applying this formula.[8, 9]

$$100\% \text{ of DPPH scavenging effect} = \frac{(A0 - A1)}{A0} \times 100$$

Where,

A0 = control's absorbance

Al= absorbance of the test

Reducing Power Assay:
(Yildirim method) was employed to evaluate the reducing power in the assay conducted. 2.5 ml of phosphate buffer (0.2 M, pH 6.6) and 2.5 ml K3Fe (CN) 6 were both included to a mixture of 1 milliliter of extract and its sub-fractions at a concentration of 25-100 micrograms per milliliter. The last combination was left to keep at 50 °C for 20 minutes for incubation. 2.5 ml of Trichloro acetic acid was introduced, then centrifuged at 3000 rpm for 30 minutes. In the end, 2.5 milliliters of the uppermost portion of the mixture, known as the liquid layer above the sediment, was gathered and combined using 2.5 ml of distilled water and 0.5 ml of ferric chloride. The measurement of measurement of absorption was carried out at a wavelength of 700 nanometers. Vitamin C and BHT served according to the usual practice, while buffer containing phosphate ions was utilized as the empty resolution. Increasing optical density levels indicate rising reducing power.[9]

Assessment of Anti-Microbial Activity

Disk Diffusion Assay:
The plant's methanolic extract exhibits antibacterial properties of Nelumbo nucifera was studied by standard paper method of disk diffusion. Mueller-Hinton agar was prepared and filled the Petri dishes that are free from microorganisms. Bacteria in suspension were introduced in petri-plates with final cell concentration of 10^8CFU/ml (colony-forming unit per millilitre) with reference to the McFarland turbidometry. Plant extract was prepared in 100 mg/mL concentrations were used to prepare filter paper discs that are sterilized and have a diameter of 6mm were saturated with 30 µl of different concentrations of extract and were set down onto the agar plate surface made with Mueller-Hinton agar. Filter paper discs that have been filled with 50 µg/ml gentamicin and streptomycin were used as positive control and discs loaded with diluting solvent was employed as the control group that received no treatment. The petri dish were left at 25ºC for 1 h and subsequently petridishes were Each disc was kept in an incubator set at 37ºC for a period of 24 hours, and the diameter of the zone of inhibition measured recorded in ml. The test was conducted repeated thrice and mean was recorded in table no 8.[10, 11]

Determination of MIC is the lowest concentration of a substance that inhibits the growth of a microorganism:
The smallest amount that stopped growth was evaluated by Mustafa et al. (2018) with some modifications. Lowest concentration that prevents visible growth of a microorganism is the least possible quantity of the substance that inhibits the growth of microorganisms following 24 hours of incubation. Different concentrations of the extract from the plant (100, 200, 300, 400 and 500 mg/ml) preparation of samples was done through serial dilution and 30 µl of The sample was placed on extensively sterilized filter paper discs. Sterile Petri dishes were filled with Mueller-Hinton agar and inoculated with bacterial suspension. The filter paper discs containing various concentrations of plant extract were positioned atop the agar plates. The dishes were stored at a temperature of 25ºC for 1 hour before being kept in an incubator at a temperature of 37ºC for a duration of 24 hours. The measurement and recording of the inhibition zones were documented in table number 8. The minimum inhibitory concentration was measured to be the smallest quantity of sample in which no apparent signs of growth were seen. The process was done thrice.[11, 15]

Assessment of antifungal Activity

Standard (Fluconazole) Disks:
30µg disks were created by placing fluconazole at a concentration of 2 mg/ml onto clean sterile disks. The disks were dried and kept at 4°C until they were used within 1 to 2 weeks.

Inoculum:
The yeast samples were kept in sterile deionized water at room temperature, transferred to Sabouraud dextrose agar for purity and viability maintenance, and sub-cultured to choose isolated colonies. Five colonies were mixed in 5.0 ml of sterile deionized H2O and thoroughly vortexed. The suspension was set to a turbidity standard of 0.5 McFarland (106 CFU/ml) with the help of a spectrophotometer. Serial dilutions of the suspension were made in log10 increments, then cultured to count CFU. Confluent growth for lawn formation was achieved using a 1-ml inoculum from a suspension containing 104 CFU/ml. 200µl of the inoculums was added onto Sabouraud dextrose agar followed by the application of antifungal disks, with a maximum of five 90 mm petri dishes filled with disks on each plate. Table 9 shows the zone sizes (in millimeters) established 48 hours following incubation at a temperature of 35 degrees Celsius and assessment at that specific moment of complete stunted growth. Every test was carried out three times.[12, 14, 15]

Determination of minimum fungicidal concentration (MFC) refers to the lowest concentration of a substance that is able to kill fungi:
The Minimum fungicidal concentration was determined using the broth micro dilution method value of the products of the ten strains tested in the biological tests. 100 microliters of RPMI-1640 liquid substance was dispensed down into the depths of a U-shaped bottom 96-well micro dilution plate. After that, Citral emulsion in a volume of 100 µl was added to the initial line in a straight direction wells on the dish. Concentrations ranged

from 1024 to 1 µg/ml through two-fold serial dilutions, where 100 µl was moved from the most concentrated well to the next. In the end, 10 µl of suspension containing C. albicans was added to each individual well of the plate, with each column representing a distinct yeast strain. Simultaneously, yeast viability and sensitivity to amphotericin B were assessed. Incubation of plates at 35°C lasted 24-48 hours. Following a suitable period of incubation, the visibility of growth was observed. Cell clusters or "buds" were observed forming in the disc pits. The MIC was established as the minimum inhibitory concentration smallest amount of citral that visibly halted the expansion of yeast. To calculate minimum fungicidal concentration (MFC), we inoculated small amounts (1 µl) of citral products, amphotericin B (at MIC, twice MIC, and four times MIC levels) onto SDA plates and observed yeast growth. After a 24-48 hour incubation at 35°C, the MFC was estimated by monitoring the growth of the control samples. Table 9 represents the minimum fungicidal concentration (MFC) as the level of a substance that hinders the growth of yeast or allows for less than three colony forming units (CFU) to be present, indicating fungicidal activity.[13, 14, 15]

RESULTS

Technique and process

Percentage yield (%) of soxhlet extraction

The percentage yield of Nelumbo nucifera was found to be 5.89%.

Physicochemical Properties of Plant Material

The various physico chemical properties isolated of plant material from seeds of Nelumbo nucifera have been characterized under the specification as per Pharmacopoeial guidelines. The ash value measured was determined to be 4.75% by weight, The ethanol-soluble extractive value was determined to be 14.75%. All the other physicochemical parameters results are shown in (table 1) and these parameters conferred as per pharmacopoeia guidelines.

Table 1: Physicochemical screening of Nelumbo nucifera

S. NO.	Test	Observation
1.	Loss on drying (%w/w)	8.33 ± 0.22
2.	Total ash value (%w/w)	4.75 %
3.	Acid insoluble ash(w/w)	2 %
4.	Percentage of Ethanol soluble extractive value	14.75 %
5.	Percentage of extractives that dissolve in water value	41%

Phytochemical Properties of Nelumbo Nucifera

Phenolic compounds are commonly belongs to the secondary metabolites. These type of phenolic componds are commonly found in other plants also. The isolated phenolic compounds from the seeds of Nelumbo nucifera was examined for various phytochemical constituents present in it. (Table 2) show data of other phytoconstituents of isolated extract.

Table 2: Phytochemical analysis

S. NO.	Phytoconstituents	Test	Result
1.	Amino acids	Ninhydrin test	Positive
2.	Alkaloids	Dragendorff's test	Positive
3.	Cardiac Glycosides	Baljet's test	Positive
4.	Steroids	Salkowski reaction	Positive
5.	Saponins	Foam test	Positive
6.	Protein	Million's test	Positive
7.	Tannins	Lead acetate solution	Positive
8.	Carbohydrates	Molisch's test	Positive
9.	Flavonoids	Lead acetate solution	Positive

Total Flavonoids and Phenolic Contents

The data in table 3 shows that the extract has a high concentration of total flavonoids, at 34.23 ± 4.84 mg QE/g quercetin equivalent per gram of extract, and total phenolics, at 25.12 ± 3.58 mg TAE/g tannic acid equivalent per gram of extract, in powdered drug form.

Table 3: Quantitative estimation of total flavonoids and phenolic contents

Phytoconstituents	Total content
Total Flavonoids (mg quercetin equivalent/g extract)	34.23 ± 4.84 mg QE/g
Total Phenolics (mg tannic acid equivalent/g extract)	25.12 ± 3.58 mg TAE/g

Assessment of Antioxidant Activity of Nelumbo Nucifera

DPPH Scavenging Assay

There is concentration dependent decrease in DPPH free radical formation by the extract. The extract showed the significant percent inhibition at different concentration when compared to standard antioxidant, extract IC50 value were described in table 4a.

Table 4a: Effect of Nelumbo nucifera seeds extract and BHT on DPPH free radical scavenging activity

Concentration (µg/ml)	% DPPH Inhibition	IC 50 (µg/ml)
Extract		
10	4.185 ± 0.234	
20	20.147 ± 1.865	
30	30.285 ± 0.769	34.05342
40	48.249 ± 1.775	
50	82.887 ± 0.363	
BHT	**% DPPH Inhibition**	
2	35.756 ± 1.654	
4	48.789 ± 1.034	
6	60.143 ± 0.857	4.21242238
8	67.453 ± 0.231	
10	75.565 ± 0.357	

Value are expressed as mean ± SEM (n=3);

IC 50 is the concentration that inhibits 50% of a substance.

Reducing Power Assay

Figure 1 compares the curve of NNSE at various concentrations (100, 200, 400, 600, 800, 1000 µg/ml) with the curves of ascorbic acid and Butylated hydroxytoluene in this test. There was an increase in antioxidant activity as the absorbance of the reaction mixture increased. In table 4b, The antioxidant capacity at a concentration of 100µg/ml was determined to be NNSE (0.30±0.04), Vitamin C (0.51±0.06), and Butylated hydroxytoluene (0.471±0.01). As the sample concentration increased, the extract's reducing power also increased. Eventually, it was determined that the reductive potential order was Ascorbic acid > BHT > NNSE.

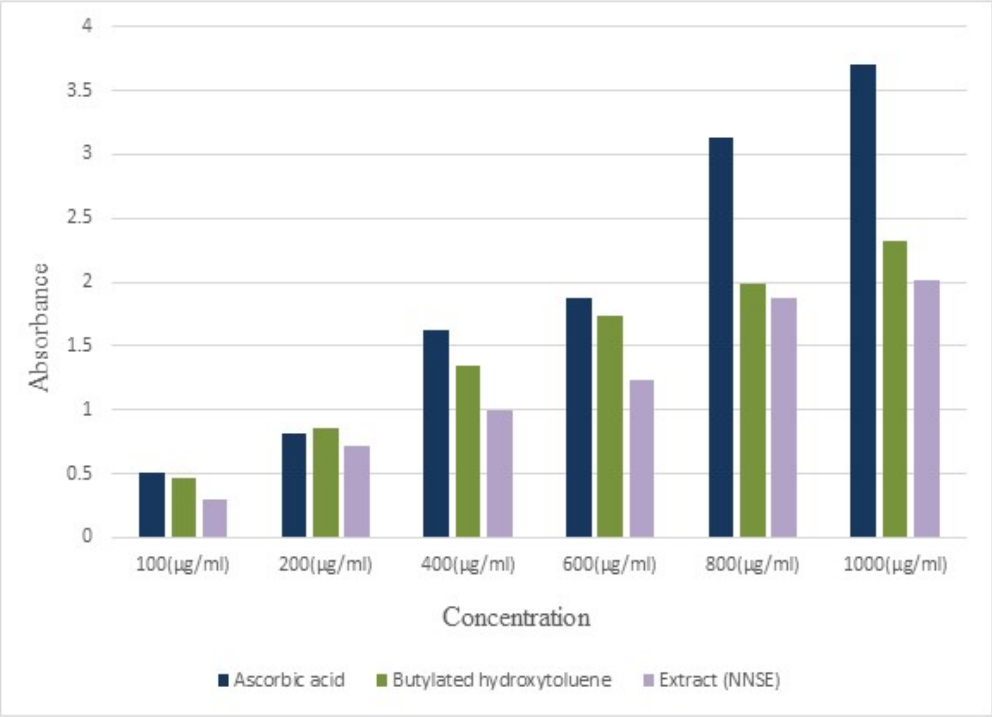

Figure 1

Table 4b: Effect of Nelumbo nucifera seeds extract, BHT and Ascorbic acid on reducing power activity

Concentration (µg/ml)	Ascorbic acid	Butylated hydroxytoluene (BHT)	Extract (NNSE)
100	0.51 ± 0.06	0.471 ± 0.01	0.30 ± 0.04
200	0.81± 0.02	0.854 ± 0.02	0.71 ± 0.04
400	1.63 ± 0.03	1.339 ± 0.02	0.99 ± 0.03
600	1.88 ± 0.08	1.737 ± 0.03	1.23 ± 0.05
800	3.13 ± 0.02	1.984 ± 0.04	1.87 ± 0.03
1000	3.71 ± 0.03	2.324 ± 0.04	2.02 ± 0.05

Assessment of Activity Against Microorganisms of Nelumbo Nucifera

Methanol extract from the Nelumbo nucifera utilized for measurement of antibacterial activity and the zone diameter for bacteria are show on table 5. Of the bacteria strains with a diameter of 12.6 mm, pseudomonas aeruginosa exhibited the highest level of Nelumbo nucifera's antibacterial activity, Mentioned in figure no. 2.

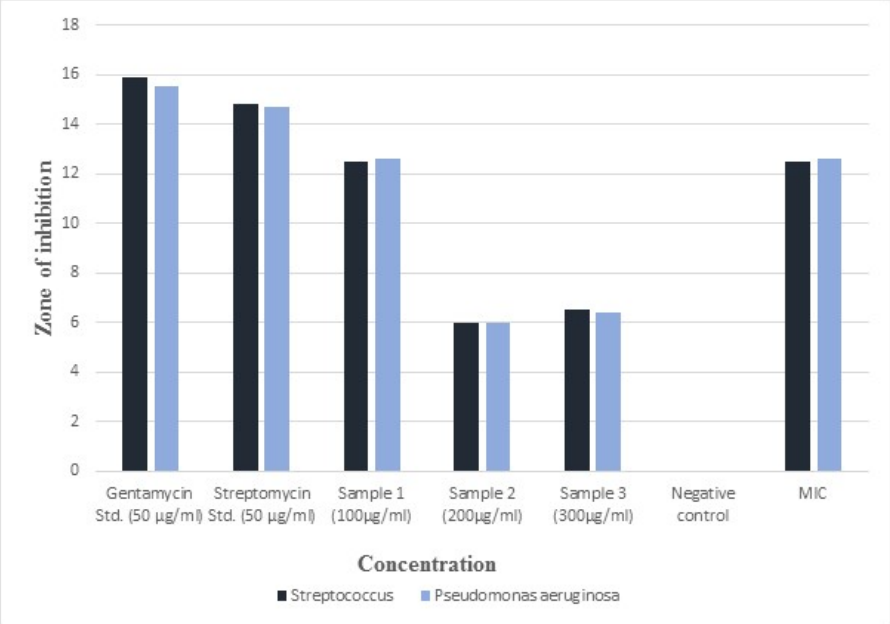

Figure 2

Table 5: Impact of Nelumbo nucifera seed extract on the proliferation of bacteria (Zone of inhibition) in different concentrations

Name	Concentration	Straptococcus	Pseudomonas aeruginosa	MIC
Gentamycin (Std)	50 µg/ml	15.9	15.5	
Streptomycin (Std)	50 µg/ml	14.8	14.7	
Blank	-	-	-	
Sample 1	100µg/ml	12.5	12.6	100µg/ml
Sample 2	200µg/ml	6	6	
Sample 3	300µg/ml	6.5	6.4	

Assessment of Antifungal activity of Nelumbo nucifera

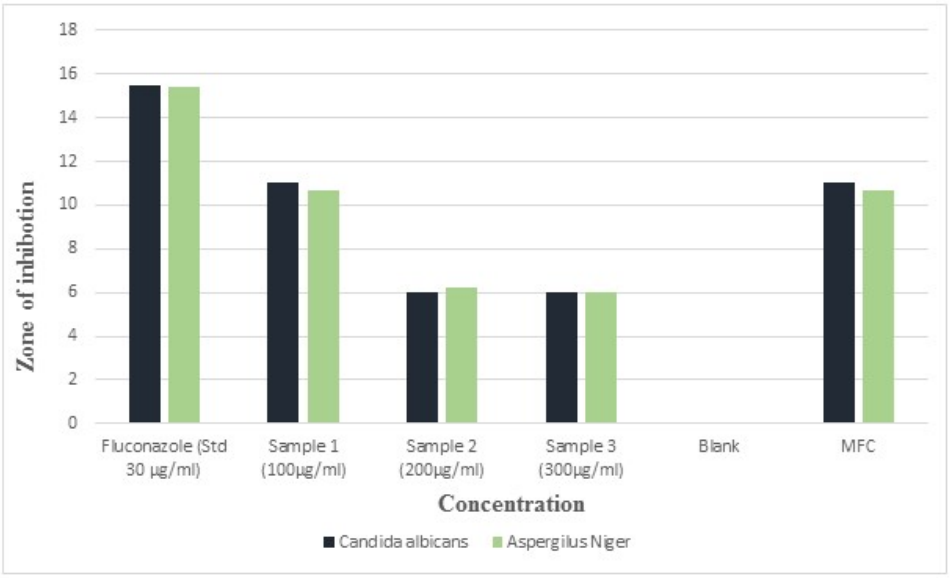

Figure 3

Figure 3 displays the methanolic extract of Nelumbo nucifera, which was utilized to measure the antifungal activity. The fungus zone diameter, of the bacterial strains with an 11 mm, Nelumbo nucifera exhibited the antifungal efficacy against Candida albicans. Shoe in table no. 6.

Table 6: Impact of Nelumbo nucifera seed extract on the growth of fungi (Zone of inhibition) in different concentrations

Sample name	Concentration	Zone of inhibition Candida albicans	Zone of inhibition Aspergilus Niger	MFC
Fluconazole	30µg/ml	15.5	15.4	
Blank	-	-	-	
Sample 1	100µg/ml	11	10.7	100µg/ml
Sample 2	200µg/ml	6	6.2	
Sample 3	300µg/ml	6	6	

DISCUSSION & CONCLUSION

The current research aimed to gather plant material from N. nucifera, create an extract, characterize it, and assess its antioxidant, anti-microbial, and antifungal properties. Samples of N. nucifera were made and assessed. N. nucifera seeds were extracted with methanol using the Soxhlet extraction technique. The samples were examined for different beneficial elements. Phytochemical analysis showed that the methanolic extract contained carbohydrates, glycosides, proteins, amino acids, steroids, saponins, alkaloids, flavonoids, and tannins. The N. nucifera seed extract showed promising antioxidant properties in a laboratory setting. The research revealed that the N. nucifera seed extract showed effective activity against various bacterial and fungal strains in the laboratory. Nelumbo nucifera extract showed efficacy against Streptococcus and Pseudomonas Aeruginosa, with Streptomycin and Gentamycin used as controls. The extract showed the same level of effectiveness against both candida albicans and Aspergillus niger. Fluconazole was utilized as the norm. Overall, antibacterial activity and antifungal activity was possessed by extract of seeds. This study showed the the ability to fight against oxidation, microorganisms, and fungi N. nucifera seeds extract. To conclude, further investigations are necessary to find out the active ingredients in charge of its antioxidant, antimicrobial, and antifungal properties.

In conclusion: This study demonstrated the antioxidant, antimicrobial, and antifungal qualities of extracts from Nelumbo nucifera seeds. To conclude, further investigations are necessary to find out the active ingredients In charge of its ability to act as an antioxidant, antimicrobial, and antifungal agent. Nevertheless, additional in vivo research is required in animals for confirmatory evidence.

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CONFLICTS OF INTEREST

The authors make it clear that they have no competing interests.

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