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PHYTOCHEMICAL SCREENING AND GREEN SYNTHESIS OF SILVER AND IRON NANOPARTICLES USING VANDA ROXBURGI EXTRACT

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Abstract:
In the present investigation iron and silver nanoparticles were synthesized by green synthesis method and were characterized with the help of UV vis spectrometer. Also various phytochemical test were performed. This research starts with the brief introduction of nanoparticles which elaborates it's chemistry as well as its importance. Further green synthesis method was explained which involves the two main methods which were top-down approach and bottom up approach. Literature of nanotechnology was discussed which shows its importance and beneficial points .Motivation point was covered which indicates how nanoparticles can be formed through various substance such as DNA, RNA etc. A brief description was given on the extract of Vanda rox burghii which illustrates it's family, botanical discription etc. Further various chemicals and equipments used in the research were explained. Green synthesis process of silver and iron nanoparticles was explained. Here the characterization was done with the help of UV vis spectroscopy where the samples were scanned at the speed of 480 nm. After this the phytochemicals test were discussed which involved various test they are glycosides test , test for steroids which was carried through salkowski's test , test for phenol and tannins, test for alkaloids which was performed through dragendroffs test , test for saponins and glycosides which was carried through foam test and solubility test all the test showed a positive results.

Keywords: *- Silver nanoparticles, iron nanoparticles, green synthesis, nanotechnology, UV- Vis spectroscopy*

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INTRODUCTION:

Nanotechnology involves the design, synthesis, and manipulation of particles with sizes ranging from approximately 1-100 nanometers. These nanoparticles (NPs) are widely used in areas such as health care, cosmetics, food and feed, environmental health, mechanics, optics, biomedical sciences, chemical industry, electronics, space industry, drug delivery, energy science, optoelectronics, catalysis, single electron transistors, light emitters, and nonlinear optical tool. [1] Research in nanoparticle chemistry is a relatively new area of chemistry. Many early nanoparticle studies may be found, but much of the field's growth happened during the 1980s and 1990s, Metal and semiconductor nanoparticles first came into widespread attention because of their plasmonic and excitonic properties, followed by their potential applications in new technologies. As nanoparticles in gold and silver nanoparticles or semiconductor nanorods show luminescence polarization in many cases, the shape of the particles directly impacts their physical resource. [2] It is widely accepted that nanoparticles are being used in a wide variety of applications including medical treatment, solar and oxide fuel batteries for energy storage, as well as inclusion in a wide variety of everyday items including cosmetics, clothes, and optics devices, catalytic, bactericidal, electronic, sensor technology, biological labelling and treatment of some cancers. Due to their excellent properties, such as high thermal conductivity, high resistance to oxidation and antibacterial activity, nanoparticles have been used for various applications including catalysis, bactericidal, electronic and sensor technology. Metal nanoparticles that have immense industrial applications include gold, silver, alloys, and magnetic nanoparticles, among others. Nanoparticles can be made chemically or biologically. [3] An approach to synthesizing nanoparticles can be top-down or bottom-up. Nucleating atomic-size materials into nanoparticles is a bottom-up approach. In addition to the Turkevich method (citrate reduction), gas phase synthesis, block copolymer synthesis, and more recently, microbial synthesis, there are some other common synthesis approaches. Methods such as milling, laser ablation, and spark ablation involve physically breaking down a bulk material into smaller molecules. [4, 5, 6] Since the past few years, green chemistry has attracted considerable attention. Because of the looming energy crisis and its profound effect on the developing economies, the need to develop alternative traditional chemistry has received significant boosts from multidisciplinary and interdisciplinary fields of study, which have given conventional chemistry a serious consideration. In this regard, nanoscience offers an amazing range of opportunities since it enables chemical, biochemical, and biophysical processes in an easier and more reliable manner through a number of chemical, biochemical, and biophysical transformations. In the fields of catalysis, synthesis, enzyme immobilization, and molecular interactions, nanoparticles have made the process simpler, faster, and more controllable. The different options for synthesis of nanoparticles, especially the use of microbes, plants, and chemical routes, as well as sonication, microwaving, and other methods are available. [7]

A new class of innovative clinical targets has emerged from Persistent efforts with unique properties and many promising applications: nanoparticles (NPs). They are, however, produced in an environmentally hazardous manner due to the primary methods currently used in their production. By describing eco-friendly methods for the preparation of biogenic NPs and the mechanisms involved in their biosynthesis, this paper is aiming to promote a green approach for the synthesis of NPs. Flavonoids, alkaloids, terpenoids, phenolic compounds, enzymes, and flavonoids are among the secondary metabolites that are abundant in natural plant extracts.. In one-step eco-friendly synthetic processes, secondary metabolites can reduce metal ions to nanoparticles. Furthermore, NPs synthesised from plant extracts typically do not require stabilizing or capping agents, but rather produce bioactive products based on shape and size. [8] From simple molecules, nanoparticles can be grown via techniques such as chemical vapor deposition, sol-gel processes, spray pyrolysis, and laser pyrolysis, all of which require a bottom-up approach. The capability of living systems to modify inorganic metals into nanoparticles through their intrinsic organic chemistry processes has opened up a previously unexplored area of biochemistry research. A new area of nanobiotechnology was created when nanotechnology was combined with biology and living organisms, such as algae, cyanobacteria, actinomycetes, bacteria, viruses, yeasts, fungi, and plants, contributed to nanobiotechnology. Every biological system varies in its capabilities to supply metallic nanoparticles. Because of their enzymatic activities and intrinsic metabolic processes, however, not all biological organisms are capable of producing nanoparticles. Bio-reduction of metallic particles then leads to the creation of nanoparticles through the use of biological entities or their extracts. A range of pharmaceutical applications are possible with these biosynthesized metallic nanoparticles, such as delivery of drugs or genes, detection of pathogens, and tissue engineering. Nanotechnology has made an important contribution to translational research related to pharmaceutical products and their applications through the effective delivery of drugs and tissue engineering. [9, 10, 11,12]. There are a number of applications for silver nanoparticles, including catalysis, chemical sensing, biosensing, photonics, electronics, and pharmaceuticals. [13]. Biological applications of silver nanoparticles include antimicrobial activity [14]. Silver nanoparticles have antimicrobial properties, making them suitable for a wide range of household applications, including textiles, food storage containers, home appliances, and medical devices [15]. Silver exhibits low toxicity while being an effective antimicrobial agent [16]. Silver nanoparticles and silver are the most commonly used materials in the medical industry in ointments that prevent infection against burns and open wounds. [17]. As a result of their attractive physiochemical properties, silver nanoparticles play a profound role in biology and medicine. A variety of silver products have been known for centuries to have strong inhibitory and bactericidal properties, as well as antimicrobial properties, which have been used to prevent and treat a variety of diseases, most notably infections [18]. Nanoparticles of silver have been reported to have antifungal, anti-inflammatory, antiviral, antiangiogenesis, and antiplatelet properties [19]

MATERIAL AND METHODS:

Present Study:**-** In the present study we synthesis silver nanoparticles (AgNPs) and Silver from aqueous Extract of Vanda Rox Burghi Extract using microwave assisted method and Phyto chemical Screening analysis.

Collection of plant extract: **-** The Vanda roxburghii Extract/Rasna Extract is fresh prepared extract from Plant part used-root collected from the Herbals and Formulations of Rutvik Enterprises from Vasai Mumbai.

Chemicals used: -

Table1: - chemicals used in the green synthesis

Sr.No	Chemical name
1	Silver Nitrate
2	Ferric Chloride
3	Distilled water
4	Vanda Rox Burghi Extract
5	Tollens reagent
6	Sulphuric acid
7	Potassium Permanganate
8	Dragendroff's reagent

Equipment's used: -

Table2: - Equipments used in the green synthesis

Sr.No.	Equipments
1	U. V visible spectrometer
2	IR spectrometer
3	Centrifuge
4	Hot Air Oven

Glassware and Apparatus: -

All glass wares (Conical flasks, Thermometer, Measuring cylinders, Beakers, Petri plates and Test tubes, fennel, Stirrer)

Test tube holder, burner, filter paper, Tripod stand, wire gauge.

Green Synthesis of Silver Nanoparticles

Take about 10 gm of extract (Vanda Roxburghii) then take 100ml of distilled water in a beaker, mixed well with the used of glass rod and then heat for10 minutes. Extracts were then **filtered** and stored at 4°C for later experiments.

Fig.1. samples of Vanda *Roxburghii(rasna))*

Synthesis of Vanda *Roxburghii* –AgNPs

For the synthesis of AgNPs, take about 5 mL of *Vanda Roxburghii* aqueous extract was added to 5 mL of 0.1M $AgNO_3$ aqueous solution and kept in oven at 60 degree Celsius for 90 seconds. Rapid reduction of Ag^+ ions to Ag**-** was observed by the change in the color of the solution from yellowish brown to dark brown colour of AgNPs synthesized were taken up for further study. [20]

Figure 2:-color changes after adding $AgNO_3$ solution. (A) Sample (B) $AgNO_3$ (C) silver nanoparticles

UV-Vis Spectroscopy:

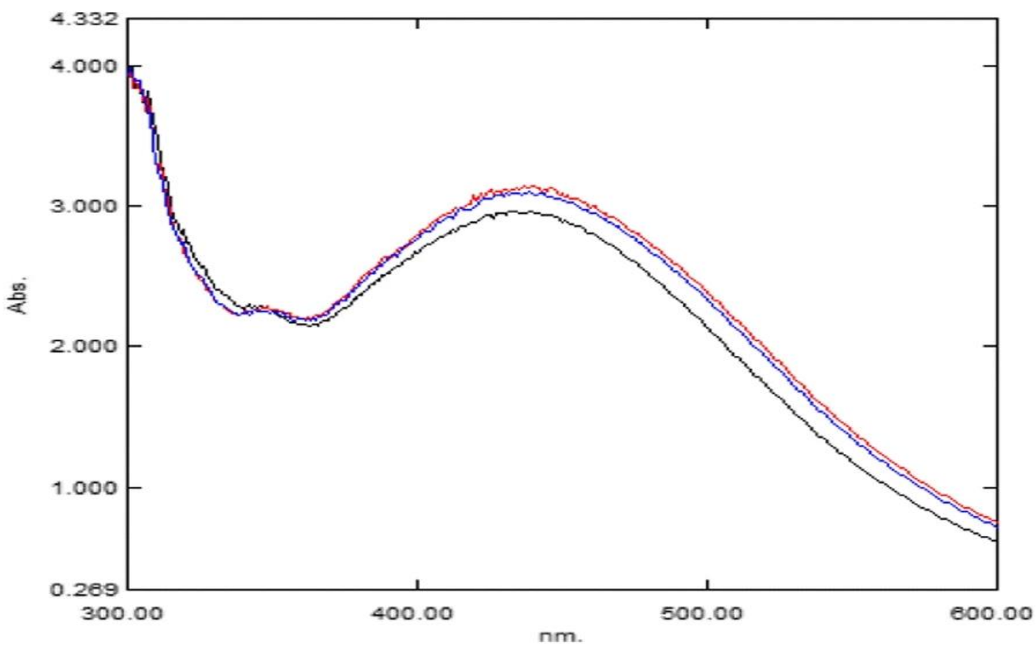

Figure 3:- Absorption spectra of silver nanoparticles obtained at different time intervals

The wavelength scale was fixed between 300 and 600 nm, and the solution was scanned in this range. Maximum absorbance at 440 nm was observed, which is characteristic of silver nanoparticle. The curve shows an increase in absorbance with the increase in incubation time (30 min, 45 min, and 1 h) of silver nitrate and extract.

GREEN SYNTHESIS OF IRON NANOPARTICLES:

Preparation of aqueous extract

The extract of the Vanda *Roxburghii* was prepared by taking 20 g of Vanda *Roxburghii powder* in 1000 ml of *distilled* water. The solution was heated at 80°C in the water bath to get the Vanda *Roxburghii* extract. The extract was collected and filtered and stored in a clean, dried beaker for further use.

Synthesis of iron nanoparticles

For The synthesis of Iron Nanoparticles, prepared 0.01 M Ferric Chloride in a beaker then take Vanda Roxburghii extract and Ferric Chloride in 1:1 proportion in a clean flask. Then the solution of Vanda *Roxburghii* extract and 0.01 M Fecl3 was change black in color.Then resultant mixture was stirred using a magnetic stirrer for 30 min.The solution was centrifuged at 8000 rpm for 20 min and the supernatant was discarded and the pellet was washed with deionized water and was centrifuged again to remove any impurities as show in fig.5. [21]

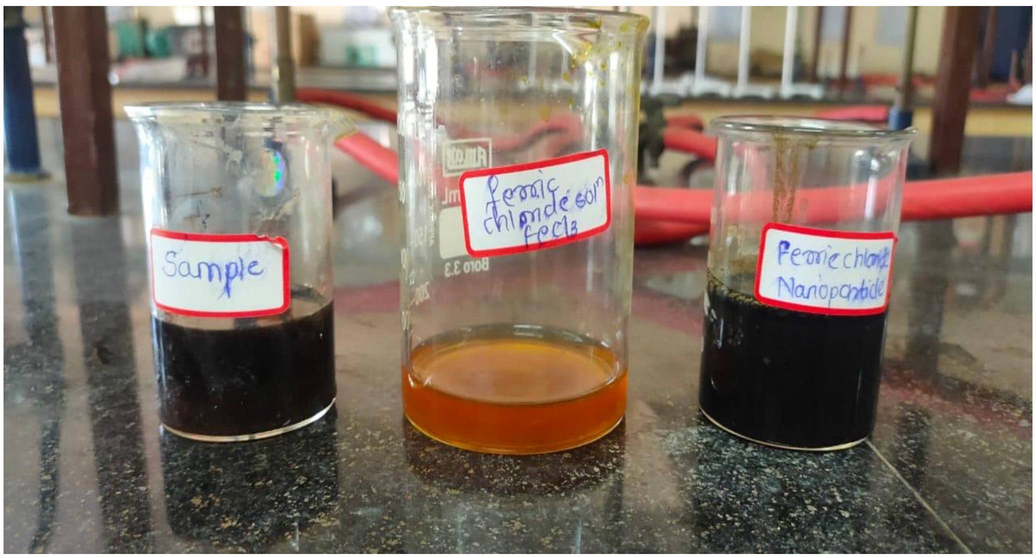

Figure 4:- color change after adding AgNO3 solution. (A) Sample (B) AgNO3 (C) Silver Nanoparticles.

Pallets formation

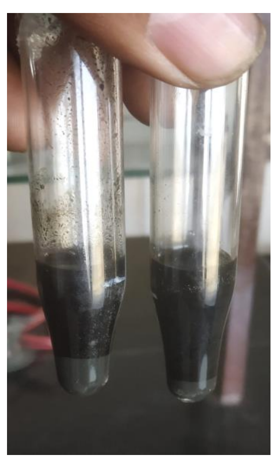

Figure 5:- The black pellets that settled during centrifugation were formed as a result of iron nanoparticle synthesis.

Fourier Transform infrared spectroscopy (FTIR):
The FTIR spectroscopy of Iron nanoparticles was done from Konkan gyanpeeth Rahul darker college of pharmacy and research which is located in karjat.

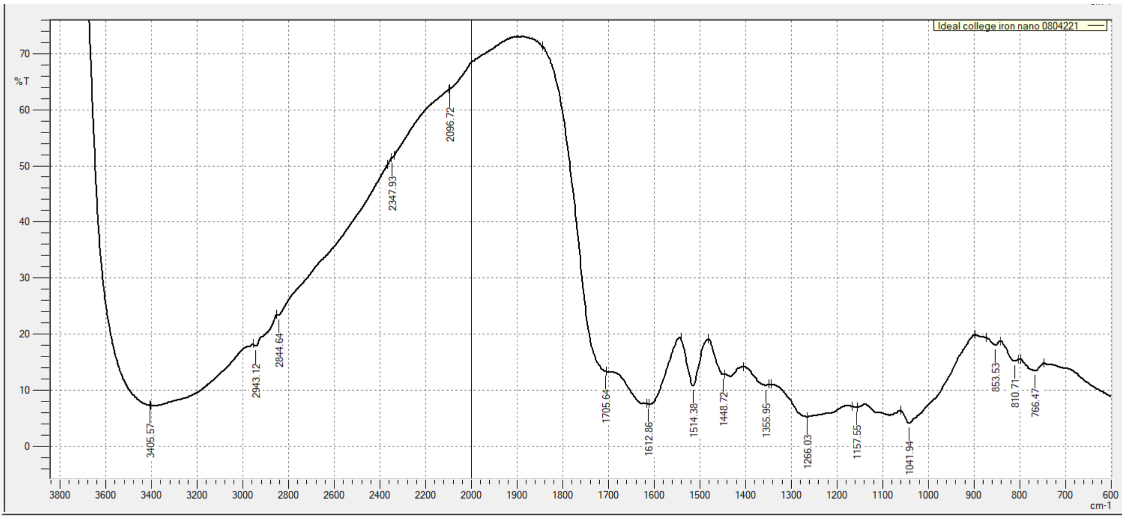

Figure 6:- FT-IR Spectra for Vanda Roxburgi Extract

PHYTOCHEMICAL SCREENING ANALYSIS: -
Preparation of Vanda Rox Burghi Extract:-Vanda Rox Burghi Extract was taken and then under running tap water to remove dust.The fruit was crushed and inti breaker and distilled water was added to the solution and the solution was boiled for 30 minutes .Now,the solution was filtered with the help of filter paper and filtered extract were taken and used for further phytochemical Analysis of Extract.[22,23]

1. Test for glycosides: - The 4 ml of extract solution was dried till up to 2ml and then 1ml of ammonium hydroxide was added and shaken. Appearance of cherish red colour indicates the presence of glycosides.

Figure 7:- glycosides test

2. Test for steroids: -
Salkowski's test:-The Extract was mixed with 2ml of Chloroform and add 2 ml of concentrated H2SO4 was added carefully and shaken gently .A reddish brown colour indicates the presence of glycosides.

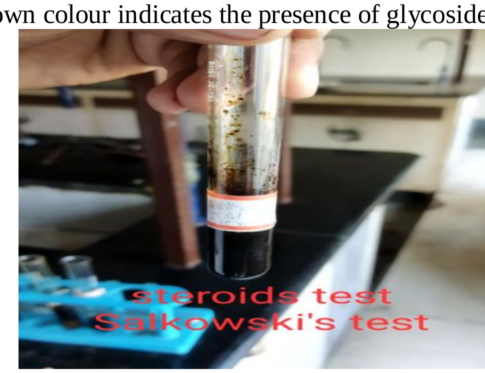

Figure 8:- steroids test of Salkowski's test

3. Test for Phenols and Tannins: - Extract was mix with 2 ml of 2% Solution of Fecl3 and a blue-greenish or blue-black colour indicates the presence of polyphenols and Tannins.

Figure 9:-Test for Phenols and Tannins

4. Test for alkaloids:-
Dragendroff's test:-2-3ml extract and add few drops of Dragendroff's reagent. Orange brown precipitate is formed.

Figure 10:- Alkaloids test Drafonfendroff's test

5. Test for Saponin glycosides: - Foam Test:-Shake the Drug Extract or Dry powder vigorously with the water and persistent Foam is observed.

Figure 11: - Test for Saponin glycosides Foam Test

CERTIFICATE OF ANALYSIS OF VANDA ROX BURGHI EXTRACT: -

Vanda Roxburghii also known as Rasna the extract used in the research was collected in the powder form through Rutvik enterprises which is well known manufacturer of herbal extract as well as of ayurvedic extract this enterprise is located in Mumbai at Vasai West.

Tables 3:- Analysis of Vanda Rox Burghi Extract

Sr. No.	Test	Standard	Result
1	Description	Light brown colored fine powder	Light brown colored fine powder
2	Odour	Characteristic	Complies
3	Taste	Characteristic	Complies
4	Loss on drying	<5%	3.2%
5	Sieve Size	Pass 60 mesh	Complies
6	Bulk Density	45-50g/100ml	49.8g/100ml
7	Herb Ratio	10:1	Complies
8	Heavy Metal	<20ppm	Complies
9	Arsenic	<2ppm	Complies
10	Total Plate count	<500 cfu/g	Complies
11	Yeast Moulds	<1000 cfu/g	Complies
12	E. Coli	Negative	Complies
13	Salmonella	Negative	Complies

RESULT AND DISCUSSION:

Colour change of solution:

As the colour changes sequentially, Ag Nanoparticles are formed by our plant materials. In order to test the formation of AgNPs, this is the primary test. The colour change observed after leaf extract was added to a silver nitrate solution demonstrated the colour reduction of AgNO3 into nanoparticles. A brown colour appeared within a few minutes, indicating the formation of AgNPs. The color was changed in brown. The change in color indicates the formation of AgNPs.

UV- Visible spectroscopy:

As a technique for analyzing the formation of nanoparticles and their stability in aqueous solutions, UV-visible absorbance spectroscopy has been proven to be extremely useful. We recorded the spectra of AgNO3 reacting with extract to determine if the reaction was complete. Fig. 11 shows Silver nanoparticles synthesized under UV-visible light, which is visible at 440 nm, is responsible for the dark brown colour of AgNPs.

FT-IR study:

Figure 14, the FT-IR spectrum of the extract, gives information regarding the chemical transformation of the functional groups involved in the reduction of the irons. Some pronounced absorbance bands centered at 766, 810, 853, 1041, 1157, 1266, 1355, 1448, 1514, 1612, 1705, 2096, 2347, 2844, 2943, and 3405 cm-1 were observed in the region 600 - 3800 cm^{-1}. Among them, the absorbance bands at 766, 810, 853, 1041, 1157, 1266, 1355, 1448, 1514, 1612, 1705, 2096, 2347, 2844, 2943, and 3405 cm-1 were associated with the stretch vibration of C-C, C-CL, C-O-C, C-C-C, C-N, C-H, CH3CH, C=C, C-C, C=O, C≡N & C≡C, C-H, and O-H respectively. There were bands of absorbance at 1705 cm-1 which could be associated with reducing sugars, flavonoids, saccharides, and proteins in the extract. The band observed at 3405 cm-1 may have resulted from –O-H groups in reducing sugars, flavonoids, saccharides and proteins in the extract. All saccharides have reduction sugars that act as a reduction agent, while the other saccharides act as a protection agent. The functional groups associated with these saccharides as well as the protein Fe+ can be reduced to Fe2+ by matter. These functional groups mediate the reduction of metal salts to nanoparticles by biological components.

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