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Green Synthesis of Iron Nanoparticle, their Characterization Using Melia Azedrach Leaves Extract and its Phytochemical Analysis

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Abstract

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Keywords: Melia Azedarach, Iron Nanoparticle, FeCl3 Salt, UV-Vis Spectroscopy, FTIR

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Introduction

Nanotechnology is the influence of the all features of the human being living. A lot of interest is currently paid to the synthesis of strong, constant, and custom-made nanoparticles by utilizing biological sources due to the drawback of linking the production of nanoparticles using chemical and physical processes (Zhu et al., 2000) (Mafuné et al., 2000). The process of nanotechnology has advanced significantly in the disciplines of health and remedial sciences, and it has broader applicability across other scientific and technological domains.

Nanotechnology is a rapidly expanding scientific field that studies nanoparticles and their applications in various fields..growing and rise in the approximately in every field of life and making a great force in all of mankind (Khatami et al., 2018). Several biological agents are broadly demoralized for the synthesis of the nanoparticle (Klaus et al., 1999)- (Kumar & Yadav, 2008). The biosynthesized nanoparticle is currently well-known to be utilized in biosensors (Kirubaharan et al., 2012). The process is used for water purification (Jain & Pradeep, 2005). In the extra the nanometer chip among green and environmental associates' characteristics is extensively utilized for the features of Nano grinding in the semiconductor industry (Zhang et al., 2012), (Zhang, Song, et al., 2012). The new enlargement of the diamond wheels and machining approach is also working on nanoparticles (Zhang et al., 2017)-(Zhang et al., 2015). That is applied in return to the traditionally polluted techniques (Zhang, Huang, et al., 2017). The utilization of the developed green slurries techniques, and high-act devices is made-up (Zhang, Huo, et al., 2012b), (Zhang et al., 2013). The nanoparticles are extensively utilized in biomedical applications such as antimicrobial, anticancer, mosquitocidal, ovicidal, and larvicidal which is budding on malaria, dengue, and filariasis mosquito vectors (Benelli & Mehlhorn, 2016). The green synthesized nanoparticles also take action as inhibitors of the dengue development (Murugan et al., 2015). There are several advantages of plant nanoparticle synthesis than other methods (Kumar & Yadav, 2008), (Iravani, 2011). The numbers of the plants were shown successfully to produce different metals of nanoparticles like silver, copper, gold, and others (Yashni et al., 2019). The silver nanoparticles have been well established and are found in wide application in several areas like optical receptors (Schultz et al., 2000), bioactive materials (Blaker et al., 2004), signal enhancers in enzyme immunoassay based on SERS

(Chen et al., 2009). The silver nanoparticle is known to have antimicrobial activity (Morones et al., 2005b). One significant component that makes silver nanoparticles efficient against bacteria and low in toxicity is that they are specifically intended to heal burn burns.

The antibacterial activity is due to the broad spectrum. The silver nanoparticles are utilized for therapeutic purposes and have been focused on increasing interest (Bellantone et al., 2000).

In the biosynthesis of nanostructured materials, a novel technique is presented that is referred to as "green techniques." Utilizing local natural resources and using eco-friendly practices to reduce pollution and the risks brought on by numerous harmful compounds are the key goals in developing this method. Antioxidant bacteria, chemicals, and active compounds found in the extract of numerous indigenous medicinal plants enable the use of natural resources (Mittal et al., 2013). Thus, it is wanted to believe that using the green method of Synthesis is essential to scientific study in the realm of nanoscience and nanotechnology.

Nanostructures are being increasingly used in medical treatments for various diseases, offering benefits such as reduced biohazards, toxicity risks, and biocompatibility.

(Vahabi & Dorcheh, 2014). Additionally, silver nanoparticles are used in a variety of spectroscopic methods such as SERS or surface-enhanced Raman spectroscopy.

(Khan et al., 2015). Gold In addition to silver nanoparticles, other nanoparticles with a size range of 20 to 25 nanometers also possess catalytic characteristics (Saravanakumar et al., 2015).

Nanoparticles of zinc oxide ranging in size from 9.6 to 25.5

nanometers are used in photocatalytic processes and exhibit antimicrobial properties (Nayak et al., 2015). Gold nano-structured particles were created by Pooja*etal.*, using karaya gum in the field of anti-cancer drugs. These gold nanostructures had full biological compatibility (Rajan et al., 2015).

The herbal remedy history is an older human civilization. The therapeutic plant is utilized as a stable source of medicaments for various diseases. Plants are supplied and the sources of botanical antibacterial, anthelmintic, and insecticides (Satyavati et al., 1976). The plant Melia is a small genus of 2 species. i.eAzadirachta and azedarach. The family of the Melia is Meliaceae and the best-known species. Due to the resemblance of the leaves to those of the manna ash or blossoming ash plant, it has acquired its name from the classical Greek term Melia for the plant, and azedarach from the name of an extinct deadly tree called Azadirachta. It originated in the area of upper Burma (Nahak & Sahu, 2010).

It is a native species of South Asia (Iran, India, and South China) that was brought to the New World, domesticated there, and then naturally spread throughout tropical America from Mexico to Argentina (Pennington, 1981). Its large geographic range 11 includes China, India, Japan, Indonesia, Northern Australia, Africa, North America, Tropical South America, and Southern Europe (Ntalli et al., 2010), (G et al., 2009).

Melia azedarach is a small to medium-sized deciduous tree, growing up to 45 meters tall, used in coffee and tea plantations as a decorative avenue or shade tree.

. The tree is resilient and draught-proof (Seth, 2004).

It may be cultivated extensively up to 2000 meters above sea level in the sub-Himalayan zone. Under normal circumstances, the plant regenerates without restriction from seeds during rain. The plant can be artificially multiplied through cuttings, root suckers, direct sowing, or transplanting seedlings. Its bark is smooth, but ages, and its leaves are bipinnate or tripinnate.

When crushed, they emit a strong aroma. A 20 cm long, axillary panicle serves as the inflorescence. Flowers range from white to lilac to purple and are fragrant; sepals are 5-lobed and are 1 cm long; pentamerous petals are 5-lobed and measure 0.9 cm long; stamens are deep purple, blue, and brown fruit, also known as berries, is a small, yellow drupe with 4 to 5 black seeds, measuring 15mm in diameter and 3.5mm x 1.6mm.

Traditional uses for Melia Azedarach L. include anthelmintic, antilithic diuretic, astringent, and stomachic (Warrier et al., 1995). Scientific investigations have revealed the antitumor (Ntalli et al., 2010), antimalarial activity, analgesic and anti-inflammatory activity (Vishnukanta, 2010).

Materials and Methods:

Studied Area

Tarbela, located in northwestern Pakistan, features mountainous terrain with the Margalla Hills to the east and the Hindu Kush foothills to the north. It experiences a subtropical climate with hot summers (over 40°C) and mild winters (5-15°C). Humidity levels vary, peaking during the July-September monsoon season, crucial for agriculture and the Tarbela Dam's water reservoir.

Study Design and Period

This work uses the extract of Melia azedarach leaves to create iron nanoparticles (FeNPs) using an experimental approach. The process of synthesis will be maximized through the manipulation of variables including temperature, pH, and reaction time. FeNPs will be thoroughly characterized by UV-Vis spectroscopy, XRD, SEM, TEM, and FTIR methods. The Melia azedarach extract will also undergo a phytochemical investigation.

Period

From August 1, 2021, to March 1, 2022, a study was conducted to investigate the environmentally friendly production of iron nanoparticles from Melia azedarach leaf extract.

The study concentrated on examining the phytochemical makeup of the plant extract and characterizing the nanoparticles.

This period involved the synthesis process, characterization techniques, and phytochemical analysis, contributing to understanding eco-friendly nanoparticle synthesis and the potential bioactive compounds in Melia azedarach leaves.

Collection and Preparation Plant Extract

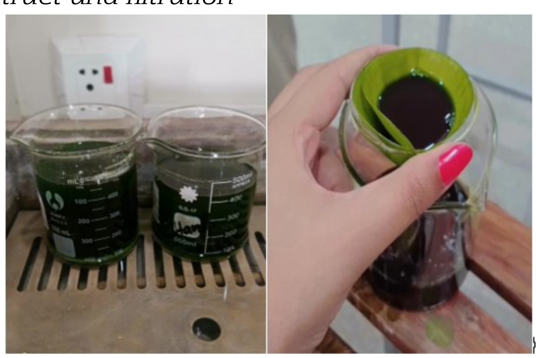
The leaves of Melia Azedarach were gathered from the Tarbela neighborhood. After that, the fresh leaves were repeatedly cleaned with tap water and then deionized water. After three days of drying in a shaded place, the leaves were ground into a fine powder. 500 milliliters of methanol and 20 grams of fine powder are combined, and the mixture is then heated to a moderate temperature in a water bath for two hours. The extract is then filtered through the Whatman filter paper. To get the pure extract, the filtration process was repeated four times. A rotary evaporator was used to concentrate the filtrate, which was then kept for later use at 40°C.

Figure 1

Dry leaves and powder of Melia Azedarach plant

Figure 2

Methanolic preparation of extract and filtration



Biosynthesis of Iron Nanoparticle Using Methanolic Extract of Melia azedarach

The prepared extract was diluted to 100ml.$FeCl_3$ 0.1mM solution was made with deionized water. The preparation of different fractions was prepared utilizing iron and plant extract in different ratios. Such as 1:1, 1:2, 1:4, 1:6, 1:8, 1:10, 1:12, 1:14, 1:16, 1:18, 1:20,2:1, 4:1, 6:1, 8:1, 10:1, 12:1, 14:1, 16:1, 18:1, 20:1 The iron extract was used to synthesize nanoparticles, with a 2:1 ratio, as confirmed by a UV-Visible spectrophotometer, indicating the optimal reaction combination for nanoparticle formation.

Figure 3

Different ratios of plant extract and salt solution

Phytochemical Analysis:

Cardiac Glycosides

Additions included 1 mL of glacial acetic acid, 2 mL of plant extract, and 5% ferric chloride. The presence of a greenish-blue tint when a few drops of concentrated H2SO4 were added suggests the presence of cardiac glycosides.

Flavonoids

Plant extract (0.5 g) was dissolved in a diluted sodium hydroxide solution and then a few drops of hydrochloric acid solution were added a yellowish solution turned colorless which indicates the presence of flavonoids.

Steroids

Each sample weighed 0.5 g, and 2 ml of H2SO4 was added along with 2 ml of acetic anhydride. In several samples, Steroids were present as the hue altered from violet to blue or green.

Tannins

Each extract was diluted with a small amount of water, brought to a boil in a water bath, and then filtered. Ferric chloride was added to the filtrate in little drops. The presence of tannins is indicated by a dark green solution.

Anthraquinones

About 0.5 g of plant extract and its small fractions were cooked with 10 HCl for a short while. The resulting liquid was then filtered and allowed to cool. A small amount of CHCl3 was then added to each filter, along with a few drops of 10% ammonia, and heated. Anthraquinones are indicated by the presence of a rose-pink color.

Saponins

After shaking 0.5 g of plant extract with 5 mL of distilled water and boiling it, the presence of saponins is shown by the formation of white foam.

Phlobatanins

The extract (0.5 g) was dissolved in distilled water and then filtered. The filtrate was boiled in a 2% HCl solution. Phlobatanins are present as a red precipitate.

Terpenoids

To make a layer, 0.5 g of each extract in 2 ml of chloroform was carefully mixed with 3 ml of concentrated H SO. The interface successfully indicated the presence of terpenoids by developing a reddish-brown tint.

Characterization of Iron Oxide Nanoparticle:

Visual Analysis

In visual analysis, the color change indicates the formation of iron oxide nanoparticles.

UV-Visible Spectroscopy

The formation of nanoparticles was analyzed using wavelengths from 200 to 800 due to iron absorption observed in this range, and the absorption spectrum 411 was also measured. By utilizing wavelength ranges of 190 to 800 nm to examine the absorption spectrum, the creation of the iron nanoparticles was verified. Using newly generated fractions at 50°C and an optical path of 1 cm length of quartz cuvettes using a spectrometer (300 Plus Optima Japan), the spectroscopic examination for iron nanoparticles was performed.

Fourier Transform Infrared (FTIR) Spectroscopy Analysis

The Shimadzu FTIR - 8400-S (AIOU) Fourier transform spectrometer was used to assess the presence of functional groups on iron nanoparticles. The powdered sample was used to prepare the samples. The pulverized specimens were inserted into NaCl and KBr pellet cells. The computer's bands that were identified displayed the outcomes. The 4000-400 cm-1 range was employed.

Results and Discussion

Phytochemical Analysis Result

Table 1.

Result of phytochemical analysis

Phytochemicals analysis of crude extracts and various fractions of Melia Azedarach

Phytochemicals	Aqueous extract
Cardiac glycosides	+
Flavonoids	+
Steroids	+
Tannins	-
Anthraquinones	-
Saponins	-
Phlobatanins	-
Terpenoid	-

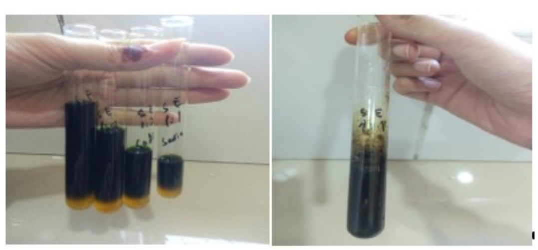
Result of Nanoparticle:

Visual Analysis

The $FeCl_3$ solution and the methanol extract solution were used to synthesize Fe2O3NPs. The color of the mixture changed from light green to dark blackish-brown, and transparent yellow to dark black in 10 minutes at 60°C, demonstrating the synthesis of Fe2O3NPs. The surface plasmon vibrations of the Fe2O3NPs were excited, resulting in a dark color.

Figure 4

Before stirring salt and extract solution is separated and after stirring the mixture is homogenous and dark brownish in color.



UV-Vis Analysis

In UV analysis at a specific absorption range and on a specific wavelength samples are given different peaks that show whether the nanoparticle has formed or not. Accordingly in this research, the sample has given 2 Adsorption values on 430nm wavelength at Raito 2:1. So, according to this research results, the best absorption peak is on 2: 1 ratio among all raito that can be shown (Fig12).

Figure 5

Graph 1 of UV

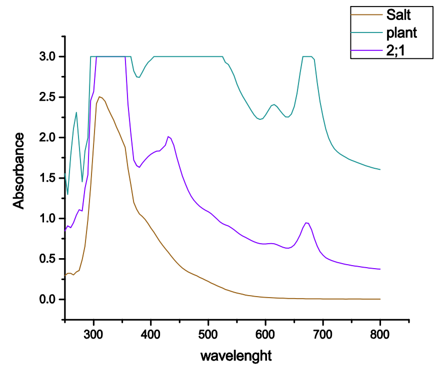


Figure 6

Graph 2 of UV

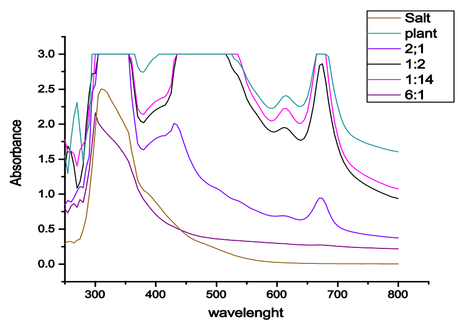


Figure 7

Graph 3 of UV

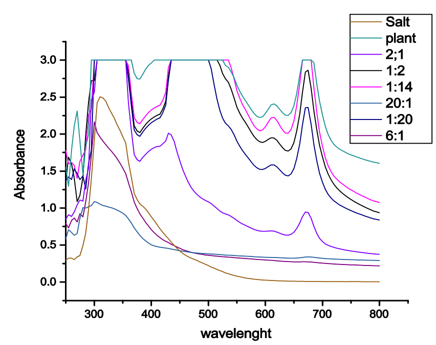


Figure 8

Graph 4 of UV

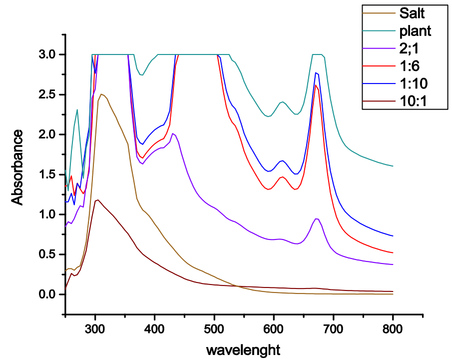


Figure 9

Graph 5 of UV

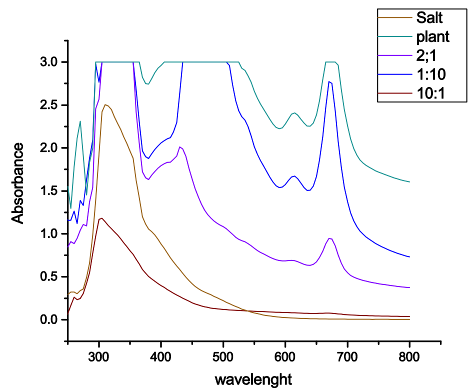


Figure 10

Graph 6 of UV

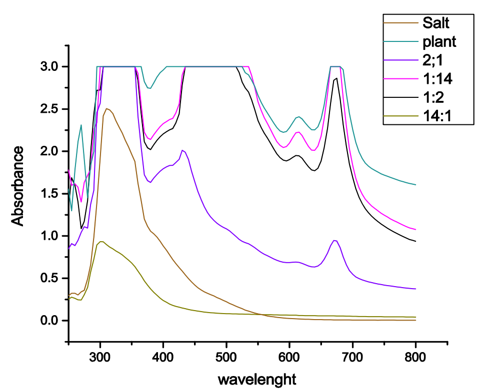


Figure 11

Graph 7 of UV

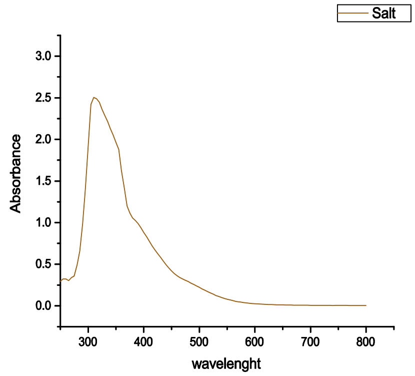


Figure 12

Graph 8 of UV

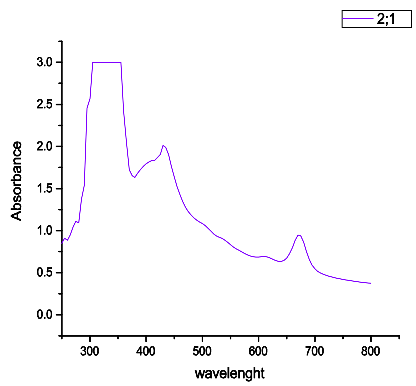
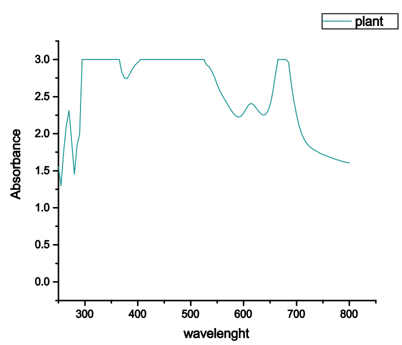


Figure 13

Graph 9 of UV



FTIR Analysis

The molecular and functional groups involved in the stabilization as well as in the formation of nanoparticles can be found by FT-IR. So, FT-IR is basically a procedure utilized to find out the main functional group that gets involved in nanoparticle synthesis (S. Saranyan, K. Viayarani, and S. Pavithra 2017). According to the result from previous research., the FTIR peak values are different for plant extract, but they decrease when they are checked for nanoparticle samples e.g in previous research Based on the FTIR spectra, the O-H stretch at 3318cm-1 was ascribed to the O-H stretch in the phenol contained in the banana peel extract extract, and it decreased in the FTIR of FeNP. It is likely that the synthesis of FeNP involved the usage of these phenolic chemicals.

Similar to that study, there was a prominent peak at 1620 cm-1 and a stretching vibration of the C=O group in ketones, aldehydes, and carboxylic acid. Represent a higher concentration of the carbonyl group compound in the extract. This peak, however, was incredibly weak and only suggested the presence of a very small amount of the carbonyl compound in the FTIR of FeNPs.

(Tyagi *et al.*,2021). Accordingly, in current research, the FTIR spectra were mainly located at 3304.25cm^{-1}(Fig14) for the plant extract and decreased to 3326.17cm^{-1}(Fig15). The presence of a peak at 1633.83cm^{-1} points to a potential phenol group O-H stretching vibration that could be in charge of the creation and stabilization of nanoparticles.

While the FTIR spectra for plant extract were also located at 1635.83cm^{-1} (Fig 14) and decreased to 1635.18cm^{-1}(Fig 15) for nanoparticle samples. The presence of a peak at 1635.18cm^{-1} indicates the C=O stretching of the carbonyl group which is also responsible for the synthesis as well as stabilization of the nanoparticle produced.

Figure 14

FTIR Graph of plant extract

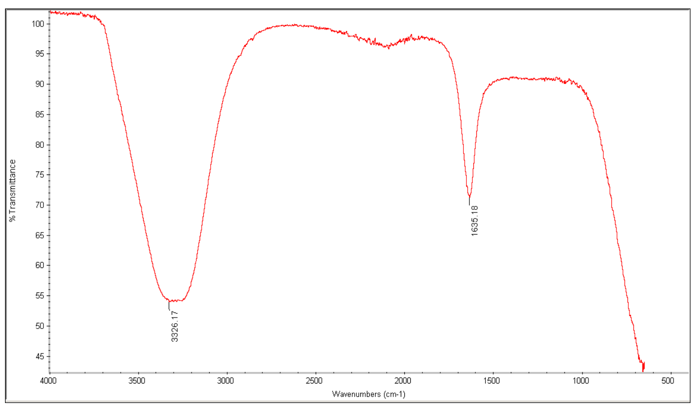
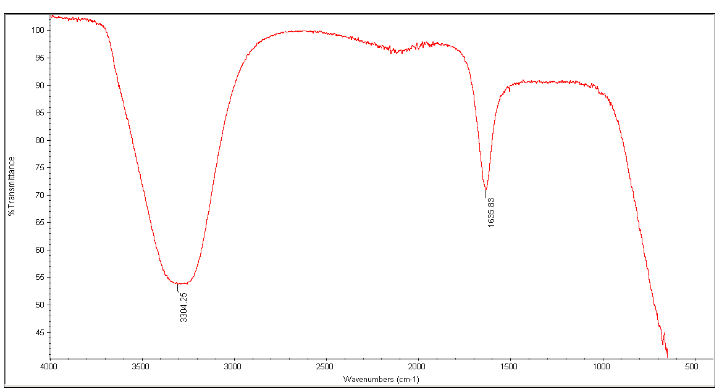


Figure 15

FTIR Graph of NPs



Conclusion

In the current research, work is done on the synthesis of iron nanoparticles utilizing *Melia Azedarach* plant leaf extract. Initially, the color changed from greenish to dark brownish or blackish confirming the formation of iron nanoparticles. Further, UV-Vis spectroscopy analysis also confirmed the synthesis of the iron nanoparticles by giving the best absorption peak at 430nm wavelength with 2 a.u absorbances. Moreover, according to the current research, the plant *Melia Azedarach* extract also contains some biomolecules like cardiac Glycoside, Flavonoids, and Steroids as reducing and capping agents and help to produce toxic-free, eco-friendly, and stable nanoparticles. Conclusively, the current research proves that the *Melia Azedarach* plant leaf extract has the potential to develop simple, low-cost, eco-friendly, stable, and biocompatible iron oxide nanoparticles.

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