BIOSYNTHESIS, CHARACETERIZATION AND ANTIBACTERIAL ACTIVITY OF ZINC OXIDE NANOPARTICLES USING *Citrullus Lanatus* (WATERMELON) PEEL EXTRACT

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ABSTRACT

The Present study is focused on synthesis, characterization and antibacterial activity of nanoparticle from watermelon (*citrullus lanatus).* The plant part used in this study is watermelon peel (*citrullus lanatus).* The watermelon peel allowed interacting under the sun. It was dried under sun then its extract was prepared. Nanoparticles were prepared by green synthesis method. The synthesized nanoparticle was characterized by UV- Spectroscopy, FTIR, and SEM analysis. This nanoparticle has got antibacterial activity against *Enterococcus faecalis, Escherichia coli, Staphylococcus aureus, Bacillus cereus, proteus and Salmonella typhi,* this study aims to conclude that aqueous extract of watermelon peel has got antibacterial activity.

KEYWORDS: Zinc oxide nanoparticles, Antibacterial, Watermelon peel (*citrullus lanatus*).

I.INTRODUCTION

Nanotechnology is a contemporary and become visible field of science that is bound to have tremendous impact on man humanity by helping solve major challenges facing humanity in health and energy. This is due to the practical application of green and eco-friendly way of synthesis of zinc nanoparticles will play a critical role in many key technologies. It is importance in such areas in medical field, medical industry, drug delivery, photo electrochemical applications in sensors and catalytic properties. Nanoparticles using plant extract that have a size of 1-100 nm in at least one dimension and possess unique biological properties due to their large surface area to volume ratio and smaller size. Recently, agricultural wastes such as watermelon peel, banana peel, and custard apple peel. (Roopan et al .2011). And various plant extract have been successfully used in the synthesis of zinc nanoparticles (Vilchis- Nestor et al., 2008). Some of the plant extract used include Aloe vera extract, Banana peel extract, etc.

Green chemistry is the utilization set of principles that will help reduce the use and generation of hazardous substance during the manufacture and application of chemical products. Green chemistry aims to protect the environment not by cleaning up, but by originate new chemical processes that do not pollute. It is a rapidly developing and importance area in the chemical science. (Badami et al., 2008). Extensive review of available literature reveals that very few studies have been done to investigate the potential of agriculture waste synthesizing zinc oxide nanoparticles. The cheapest and environmental- friendly source of zinc nanoparticles would ideally use biomass that has no other useful competing applications (Ahamed et al., 2016).

The kingdom of watermelon is plantae - plant and subkingdom of watermelon Tracheobionta – Vascular plants and superdivision of watermelon is spermatophyte – seed plants, division of watermelon magnoliophyte – flowering plants. Class of watermelon magnoliosida – dicotyledons, order of watermelon is *Cucurbitaceae.* Genus of watermelon is citrullus. And species of watermelon is *citrullus lanatus*. (Ambreen Naz et al., 2014).

Storage conditions are also cardinal that significantly affect the concentrations of lycopene, phenolics and vitamin C contents. The higher ratio of lycopene to carotene in watermelon 1:12 yields remarkable antioxidant capacity. Besides the presence of lycopene, it is a source of B vitamins, especially B1 and B6, as well as Watermelon contains phenolics quite comparable with that of other fruits It is an inexpensive and nutritious source that is readily available to all socio-economic groups of Pakistan throughout the summer season. Its consumption depends on number of factors e.g. availability, income, age, gender, racial and ethnic norms. In this context, per capita consumption in Asian communities is almost 3 times greater as compared to other part of globe. (Ambreen Naz et al., 2014).

Watermelon *(Citrullus lanatus*) has association with cucumber, pumpkin, squash and gourds; belonging to family *Cucurbitaceae*). Fruit of this plant is major consumed portion and variations in growth characteristics determine its end use quality considering the nutritional profile, consumption of 100 g watermelon provides 30 kcal. It contains almost 92 % water and 7.55 % of carbohydrates out of which 6.2 % are sugars and 0.4 % dietary fiber. It is enriched with carotenoid, vitamin C, citrulline, carotenoids and flavonoids and fat and cholesterol free, thus considered as low caloric fruit Additionally, watermelon is rich source of β-carotene acts as an antioxidant and precursor of vitamin A. (Ambreen Naz et al.,2014).

The Watermelon (*Citrullus lanatus)* being the largest and heaviest fruit, is one of the most great popular and low price available fruits in India with 3 lakh tones produced every year. Red flesh of watermelon rich in vitamin B6 and antioxidants lycopene and beta-carotene (Oseni et al., 2013). The cheapest and environmental- friendly source of zinc nanoparticles would ideally use biomass that has no other useful competing applications. Fleshy pulp fruit present inside is sweet, edible and used for juices, tasty summer fruity but outer rind is considered as waste and has no commercial value. Watermelon rind (WR) consists of pectin, citrulline, cellulose, proteins, and carotenoids (Rimando et al. 2007). And the seeds contain significant amounts of trace elements such as zinc, magnesium, and calcium, the rind though edible, is usually discarded as Agriculture wastes would offer such a source, for example, watermelon rind. The watermelon rind may potentially be used to synthesize Znic Oxide Nanoparticles. (Michael Ndikau 2017 et.al). Studies have shown that the *Citrullus lanatus (*watermelon) rind extract in this work, we show that the potential of using *citrullus lanatus* (watermelon) rind extract as a green synthesis.

II. MATERIALS AND METHODS

2.1 Collection of aqueous water melon (*Citrullus lanatus)* Peel extract;

The water melon fruit will be purchased from the local Tirupattur market and used its rind as a sample for our study. The water melon was collected from Tirupattur local vegetable market.

Figure: 1
Watermelon (*Citrullus lanatus*)

Figure: 2
Watermelon peel (*Citrullus lanatus*)

2.2 Preparation of aqueous watermelon *(Citrullus lanatus*) peels extract;

After collection of *Citrullus lanatus* (water melon rind) flesh were separated and sliced. Then dried in sunshade to remove the residual moisture. The extract used for the reduction of Zinc ions (Zn2) to Zinc Oxide Nanoparticles (ZnO) was prepared by placing 80g of dried fine flesh power along with 1litre of sterile distilled water and then boiled for 20minutes at 60c until the colour of the aqueous solution change from watery to brown. The extract was allowed to cool and kept in room temperature and filtered using Whatman filter paper. Whatman filter paper. The extract was stored in the refrigerator in order to be use in further experiments.

2.3 Synthesis of Zzinc nanoparticles;

For the synthesis of Zinc Oxide Nanoparticles, of zinc Oxide Nanoparticles, 500 ml of (*Citrullus lanatus)* flesh extract was taken in a clean conical flask. And 10 g of Zinc nitrate was added to the solution and mixed thoroughly and kept in shaker incubator for 2 hours at 60C. After incubation that mixture was allowed to cool down to room temperature. And the solution was centrifuged for 20minutes at 400 rpm. After centrifugation supernatant was discarded and obtain pellet were separated and kept in Hot air oven for 6-7 hours at 80c. The resultant sample was collected and smashed in a mortar and pestles so, as to get a finger nature for further characterization of zinc Oxide nano particles and stored in air tight container (Mishra and Sharma 2015).

2.4. CHARACTERIZATION OF ZNIC OXIDE NANOPARTICLES;

2.4.1. UV-VIS spectrophotometer

The nanoparticles synthesized are confirmed by recording the UV- Vis spectra at periodic time

2.4.2. FTIR Analysis

Then the FTIR analysis was performed to classify the groups that are responsible for reduction of the material and for the stabilization of nanoparticles.

2.4.3 SEM Analysis;

Scanning electron microscopy (SEM) will be done that gives the morphology of the synthesized ZnO nanoparticles. The characterization of Scanning electron microscope analysis is employed to determine the size, shape & morphologies of formed nanoparticle SEM gives high resolution images of the surface of a sample is desired.

2.4.4 ANTIBACTERIAL ACTIVITY;

Many systemic method have been adopted to evaluate the antibacterial activity of ZnO Nanoparticles. One of the most used is broth dilution method, followed by colony count, in which the plates serial culture broth dilutions containing the bacteria and ZnO Nanoparticles incubated at proper condition, in suitable agar medium. Presently, *Esherichia coli (*gram negative), *Staphylococcus aureus* (gram positive), *Enterococcus faecalis* (gram positive), proteus mirabilis (gram negative), *Bacillus cereus* (gram positive), *Salmonella typhi* (gram negative) are mainly chosen as model bacteria to evaluate the antibacterial activity of ZnO nanoparticles. Twenty four hours old culture of selected bacterial broth prepared by inoculating a loop full of mother culture in to the test tubes containing 5 ml of broth (nutrient agar) which were incubated at appropriate time and temperature (37^{o}C for 24 hours).

2.5.1 Preparation of test solution

The test solution was prepared with know weight of sample dissolved in 5% Dimethyl Sulphoxide (DMSO) (Corresponding to 50µl, 100µl, 150µl, 200µl).

2.5.2. Determination of antibacterial activity

Agar well diffusion method was trailed for antibacterial activity. The Muller Hinton agar was prepared, poured on petriplates and allowed to solidify. After solidification, 0.1 ml of standardized microbial inoculum suspension was poured and uniformly spread. The excess inoculum was drained and the plates were allowed to dry for 5 minutes. After drying, samples were placed on the plates with sterile pipette. Biosynthesized sample (50 µl, 100 µl, and 150 µl,200 µl) were used as the positive control and the DMSO (5%) was used as a blind control. Finally the inoculated plates were incubated at 37 c for 24 hours. The zone of inhibition was observed and measured in millimetres. This experiment was repeated for two times.

III. RESULTS AND DISCUSSION

3.1. UV-Visible Spectra Analysis

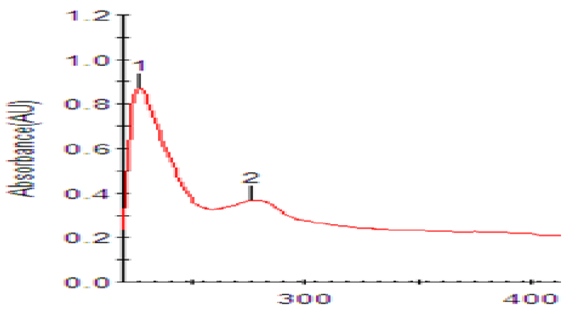


Figure: 3

S.No.	Peak(nm)	Peak(%T)
1	226.75	0.871
2	277.40	0.368

The synthesized ZnO nanoparticles illuminate the optical properties by UV-Vis Spectrum at room temperature as displayed in figure 1. The UV band around 200-400 nm express the exhaustive absorption which can be in figure 1. The peak of maximum absorption was visualized approximately at 277 nm which indicates analogous to the results of (Belay et al., 2017) where they had absorbance at 278 nm.

3.2 FTIR Analysis

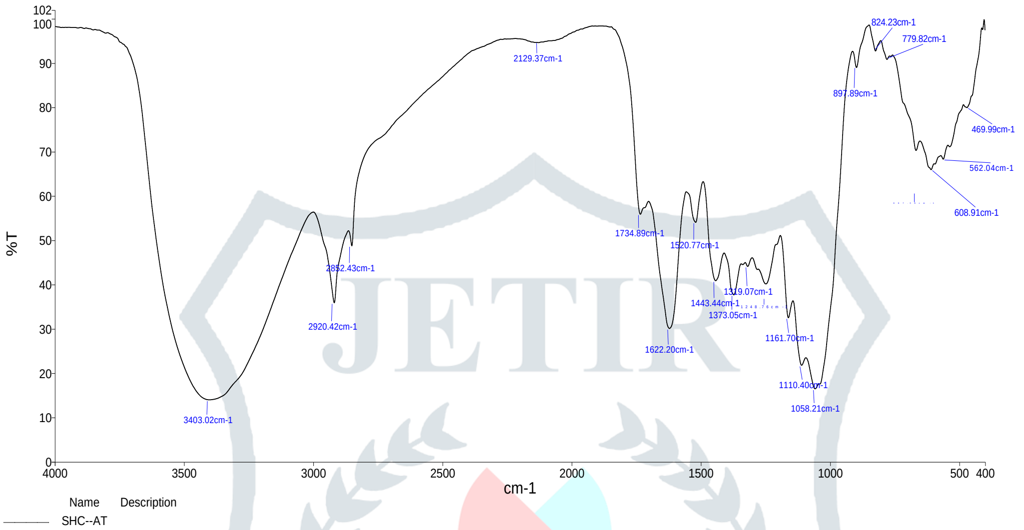
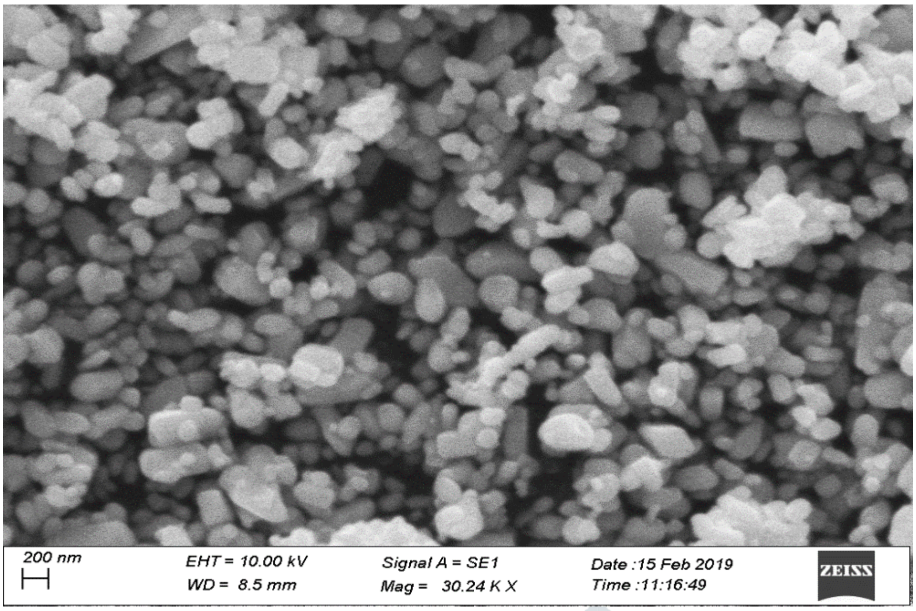


Figure: 4

S.NO	Absorption Band	Indication
1	**1520 cm -1**	**-OH stretching**
2	**1622cm-1**	**C-O stretching of acetyl group**
3	**2920 cm-1**	**=C-H stretching**
4	**1058 cm-1**	**Fingerprint region**
5	**1319-1373 cm-1**	**C-H bending**

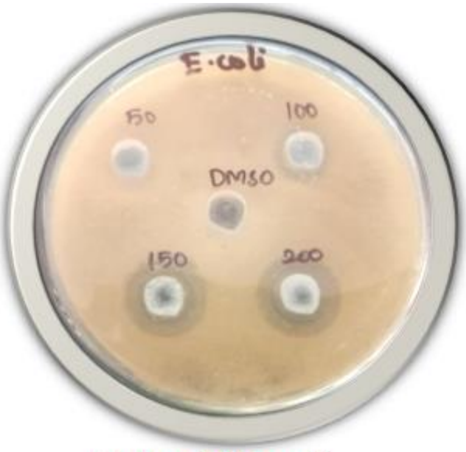
3.3. SEM Analysis

The figure- shows the SEM micrograph of the ZnO Nanoparticles. The nature of nanoparticles was clearly visible. The spherical shape particles formed are predominant; some of the ZnO nanoparticles are aggregated. The size of smallest ZnO Nanoparticles is around 25 nm and the size of aggregated ZnO Nanoparticles is around 100 nm. These results are agreement with earlier studies.



3.4. Antibacterial Activity

Antibacterial Agent	Concentration of sample				
	DMSO	50µl	100µl	50µl	200µl
Bacillus cereus	NIL	10mm	12 mm	12 mm	12 mm
Enterococcus faecalis	NIL	12 mm	12 mm	12 mm	12 mm
Escherichia coli	NIL	12 mm	12 mm	12 mm	12 mm
Salmonella typhi	NIL	12 mm	12 mm	12 mm	12 mm
Staphylococcus aureus	NIL	12 mm	12 mm	12 mm	12 mm
proteus vulgaris	NIL	10 mm	10 mm	11mm	12mm



Escherichia coli

Enterococcus faecalis

Salmonella typhi

Staphylococcus aureus

Proteus Vulgaris

Bacillus cereus

CONCLUSION:

The present study was aim to conclude that when compared to ZnO nanoparticles by green synthesis method is very low cost effective and eco-friendly. In recent study determines the aqueous extract of watermelon peel or rind (*Citrullus lanatus*). Which acts a reducing and stabilizing agent in ZnO Nanoparticles synthesis? Synthesized ZnO Nanoparticles and analysed the maximum peak range, it was noted that highest peak range at 277nm by using UV-Visible Spectroscopy.

The FTIR- (Fourier transform infrared) Analysis the various functional group present in the watermelon (*Citrullus lanatus*) peel extract. The band was observed and it is considered as a region of ZnO metal nanoparticles.

In SEM analysis it was determined that the synthesized nanoparticles are in spherical shape thus it confirmed the presence of ZnO Nanoparticles (Ahamed et al., 2018).

Antibacterial activity was carried out by using aqueous extract of watermelon peel (*Citrullus lanatus*). It shows good zone of inhibition against bacterial pathogens. ZnO Nanoparticles has sufficient inhibition activity against for both Gram positive and Gram-negative bacteria

Antibacterial activity was carried out by using aqueous extract of watermelon peel (*Citrullus lanatus*). ZnO nanoparticles has got antibacterial activity against *Enterococcus faecalis*, *Escherichia coli*, *Staphylococcus aureus, Bacillus cereus, proteus and Salmonella typhi*, from my project study it was entitled that Aqueous extract of watermelon peel found to have antibacterial activity property.

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