



Green synthesis, Characterization and Antimicrobial activity of ZnS using Syzygium aromaticum extracts

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Abstract : This present work reported biosynthesized ZnS nanoparticles using methanol extract Syzygium aromaticum as an antimicrobial agent. The soxhlet apparatus was used to extract Syzygium aromaticum and As-biosynthesized ZnS nanoparticles were characterized by using different analysis techniques. The nanoparticles structural properties and surface morphology formation were investigated using X-Ray diffraction (XRD), Scanning Electron Microscopy (SEM) and average grain size also calculated. The optical absorption and different functional group of biosynthesized ZnS studied by UV-Visible spectroscopy (UV-Vis) and Fourier transform infrared spectroscopy (FTIR). The antimicrobial activity was evaluated by agar well disc diffusion method against various microorganisms.

Keyword : ZnS, Syzygium aromaticum, Antimicrobial Activity, XRD, SEM, UV-Vis, FTIR.

1. Introduction

ZnS is one of the promising semiconductor materials in II-VI group because of their wide band gap energy (3.7 eV), Dielectric constant, Refractive index (2.35)^[1-3]. In photovoltaic and optoelectronics devices ZnS is resolvable material because of their Nano and polycrystalline structure^[4]. ZnS have to different zinc blende and wurtzite crystalline structure^[5] there is different methods are used to synthesis ZnS, chemical bath deposition^[6] spray pyrolysis, vacuum evaporation and pulsed laser deposition^[7] in health and food industries silver, titanium gold nanoparticles are widely used because of their antimicrobial properties^[8] synthesized ZnS and other sulphide metals are broadly compared and determined their antibacterial, Fungal and viral activity^[9-10] Syzygium aromaticum is good medicinal plant its commonly known clove and widely used novel drug medicine, green pharmacy due to the antimicrobial activity^[11] Syzygium aromaticum (clove) have excellent antimicrobial activity against food spoilage bacteria^[12] methanol extract Syzygium aromaticum (clove) increase zone of inhibition to compare ethanol extract^[13] Syzygium aromaticum (clove) is family of myrtaceae and it's a unopened buds of flower in plant^[14-15] the objective of the study is biosynthesis of ZnS NP's using Syzygium aromaticum (clove) methanol extract and investigate their characterization and Antimicrobial activity.

2. Materials and Methods

2.1 Plant Extract

Syzygium aromaticum(clove) buds are purchased from agriculture university, Coimbatore and well growth Syzygium aromaticum (clove) was conform by Mr.R.Gokulavasan, seed certificate officer, Tharapuram

02. The obtained *Syzygium aromaticum* (clove) were washed running tap water followed DI water five times for removing soil and dusts, Washed material treated shadow dried, open air dried at room temperature more than ten days and grinder used to crushed powder form of dried plant *Syzygium aromaticum*. The Soxhlet apparatus used to extract plant, 25 g of coarse powder was treated 250 ml of methanol to extract successful solvent and stored, used for further investigations.

2.2 BIOSYNTHESIS OF ZnS

Biosynthesis of ZnS used from *Syzygium aromaticum* extract, zinc sulphate obtained from Merck Industries in analytical grade used for without purification, 14.4 g zinc sulphate was dissolved in 100 ml DI water and 20 ml of *Syzygium aromaticum* extract was treated in 20 min stirred. Then 20 ml of stirred extract was added drop by drop in 80 ml (1 mM) of aqueous ZnS solution were kept from continuous stirring at 2 hours. The formation of nanocolloid solution was formed at result and dried for 60°C at 5 hours.

2.3 Preparation of Inoculums and Antimicrobial Test

One of the best antimicrobial susceptibility tests is disc diffusion method and Muller Hinton Agar (MHA) plates are obtained from Hi-Media Mumbai used to screening *In vitro* antimicrobial activity all the stock culture was maintained 4°C, The loopful process were used transfer stock culture into MHA plates and all stocks cultures were kept 24 hours at 24°C to 35°C in incubator

Before transfer the cultures the MHA plates are treated different process for purification, 15 ml pouring of molten media use to prepare MHA plates and allow 5 min for solidify plates and the minimum amount 0.1% of stock cultures inoculums were swapped in MHA plate surface and allowed 5 min to dry, The 60 mg/disc concentration of disc are placed MHA plate surface each disc have 6mm width. The prepared biosynthesized ZnS solution were loaded all disc surface with different concentration and allow 5 min to diffuse extracts and diffused MHA plates are kept Incubation at 37°C for 24 hrs during the incubator zone of inhibition (ZOI) was formed around the disc, transparent millimeter ruler use to measure ZOI.

2.4 Characterization Techniques

Biosynthesized zinc sulphide nanoparticles were investigated different techniques for analysis the characterization, X-ray diffractometer (XPERT-PRO) used to analysis crystalline structure of ZnS and average grain size, Hitachi S-4500 scanning electron microscope (SEM) help study surface morphology and formation of nanoparticles, The optical absorbance was determine using UV-Visible spectrometer, JASCO FT/1 IR-6600 instrument was use to measure different function group of synthesized ZnS.

3. Result and Discussion

3.1 Structural Analysis

Synthesized ZnS nanoparticles using *Syzygium aromaticum* extract crystalline structure characterization was carried out by using X-Ray powder diffraction analysis. Figure 1 show the different intensity peaks ($2\theta = 28.55, 33.08, 47.51, 56.28, 59.13$) for corresponding reflective plane (111, 200, 220, 311, 222) it conform ZnS cubic zinc blende structure because the major peak were absorb at ($2\theta = 28.55$ and 47.51) for (111,220) plane (JCPDS 05-0566)^[16], Average crystalline size (D) calculated by using Scherrer's formula.

$$D = 0.9\lambda / \beta \cos \theta \quad \text{----- (1)}$$

Here 0.9 is the shape factor constant value, $\lambda = 1.5405 \text{ \AA}$ wave length of the incident beam CuK α 1, Full width half maximum is β , diffraction angle θ in radian, average grain size of ZnS is ~46 nm

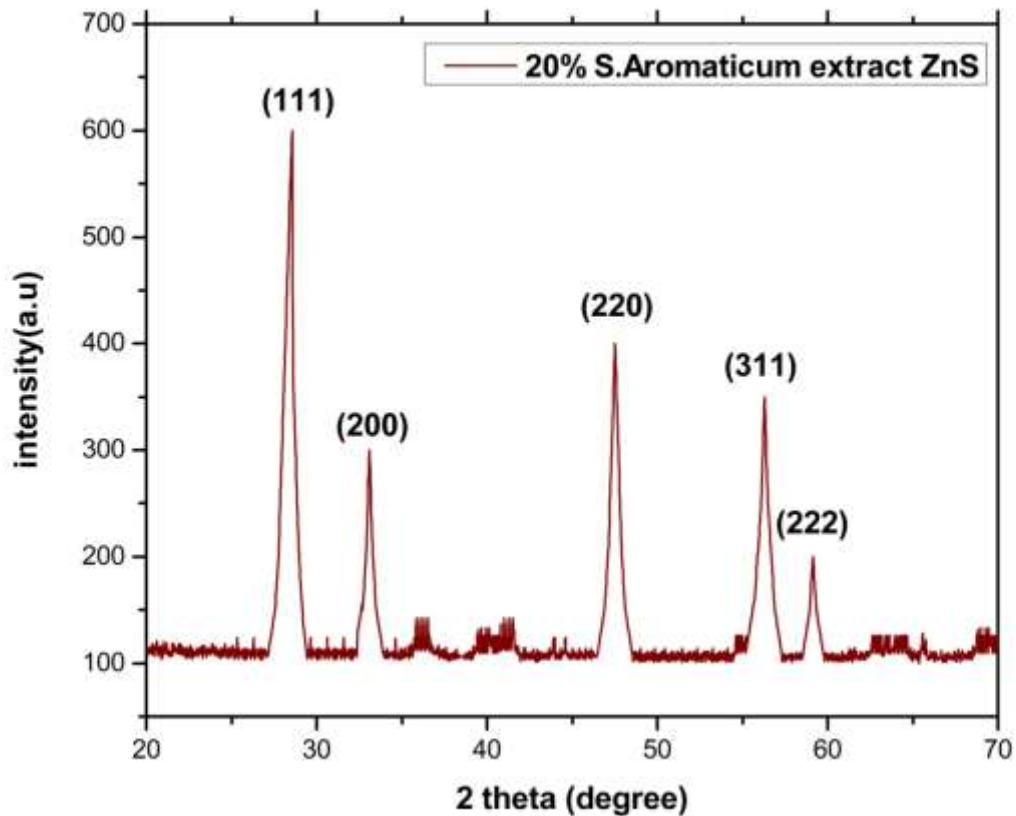


Figure 1: XRD pattern of ZnS using Syzygium aromaticum extract

3.2 Morphology Analysis

Surface morphology of synthesized ZnS nanoparticles using Syzygium aromaticum extract, powder sample were analysis by using SEM, Figure 2 clearly show the different particle size and zinc blend and spherical microstructure on the surface and estimate size was found less than 2 μm .

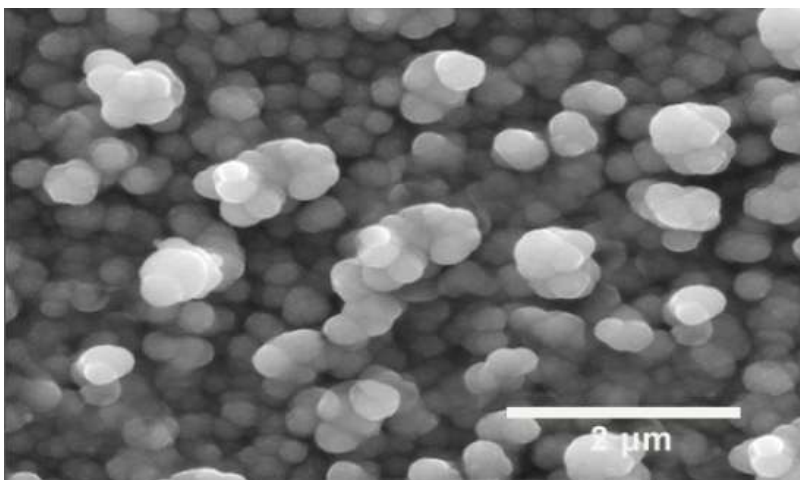


Figure 2: SEM analysis of ZnS using Syzygium aromaticum extract

3.3 UV-Visible Analysis

Figure 3 show the UV-Visible spectrum of synthesized ZnS nanoparticle using *Syzygium aromaticum* extract. The ZnS nanocolloidal solutions optical absorbance examined from 400-600 nm range. The sharp absorbance of UV-Vis region at 492 nm and decrease up to 600 nm during the absorbance green ZnS nanocolloidal solution transition in to white color.

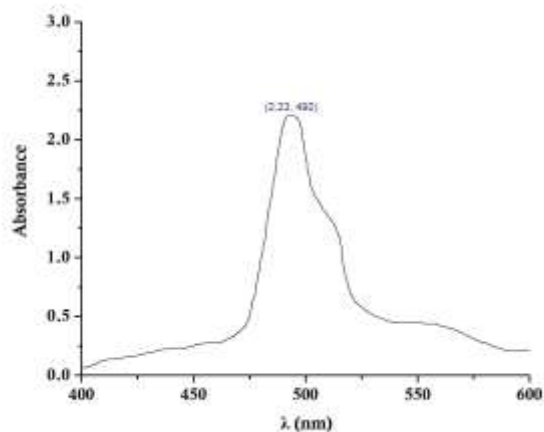


Figure 3: UV-Visible spectrum of ZnS using *Syzygium aromaticum* extract

3.4 Functional Group Analysis

FTIR analysis use to evaluate different functional group of biosynthesized ZnS nanoparticles using *Syzygium aromaticum* extract. Figure 4 shows various IR spectra of biosynthesized ZnS sharp peaks, all functional group exhibit range from 500 to 4000 cm^{-1} absorption.

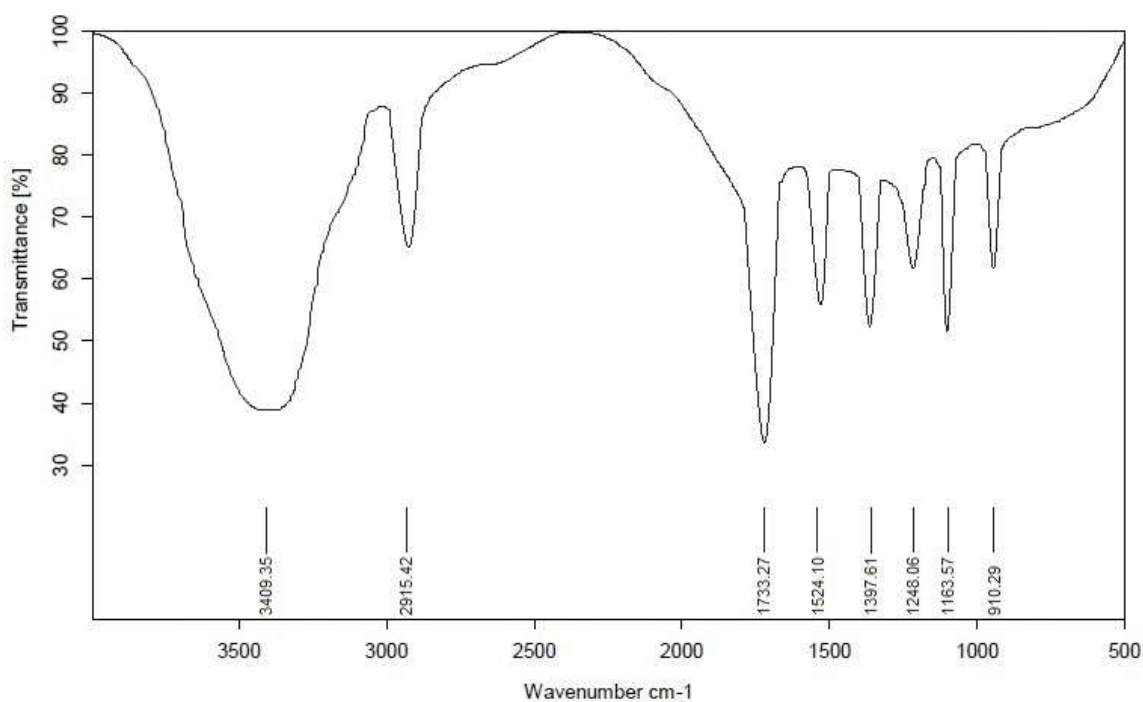


Figure 4: FTIR spectrum of ZnS using *Syzygium aromaticum* extract

Broad absorption range in 3409 cm^{-1} exhibit O-H alcohol bonding due to high stretching of Zinc, capping agent of *Syzygium aromaticum* extract were low exhibit stretching at 2915 cm^{-1} due to C-H methyl and methylene group, capping agent of sulphide high stretching exhibit at 1733 cm^{-1} C=O carbonyls, 1524 cm^{-1} is C=C aromatic of extract 1397 cm^{-1} is C-H Cellulose, hemicelluloses of extract, again zinc stretching at 1248 cm^{-1} O-H Phenolic and 1163 cm^{-1} O-H Alcohols (Primary and secondary) of sulphide exhibits, at 910 cm^{-1} is C=C alkanes indicate water stretching of nanocolloidal ZnS solution

3.5 Antimicrobial Activity Analysis

Biosynthesized ZnS using *Syzygium aromaticum* extract antimicrobial activity were examined by in vitro disc diffusion method using MHA plates, during the incubation zone of inhibition (ZoI) was formed around the disc, figure 5 and table 1 clearly show their formation. Here both gram positive, gram negative bacteria and fungus are tested. *Staphylococcus aureus* and *Bacillus subtilis* are gram positive, *Escherichia coli* and *Pseudomonas aeruginosa* are gram negative bacterial, only cell membranes differ for both bacteria's and *Candida albicans*, *Aspergillus niger* are fungus culture. The 30 μl to 60 μl different four concentration was tested in MHA plate surface.

Table 1: Antimicrobial activity of ZnS using *Syzygium aromaticum* extract

Organism	C	30 μl	40 μl	50 μl	60 μl
<i>Staphylococcus aureus</i>	16	12	14	22	24
<i>Bacillus subtilis</i>	13	11	16	20	21
<i>Escherichia coli</i>	15	9	14	18	20
<i>Pseudomonas aeruginosa</i>	15	10	16	18	22
<i>Candida albicans</i>	14	11	15	23	25
<i>Aspergillus niger</i>	12	9	18	20	21

At 30 μl concentration ZoI were formed against all tested microorganisms *Staphylococcus aureus* (12 mm) lead at this concentration followed by *Bacillus subtilis*, *Candida albicans* (11 mm), *Pseudomonas aeruginosa* (10 mm), *Escherichia coli* and *Aspergillus niger* (9 mm) but 40 μl concentration *Aspergillus niger* (18 mm) lead and 50 μl concentration *Candida albicans* (23 mm) formed high ZoI.

Finally at 60 μl concentration fungus culture of *Candida albicans* (25 mm) formed high zone of inhibition followed by *Staphylococcus aureus* (24 mm), *Pseudomonas aeruginosa* (22 mm), *Bacillus subtilis* and *Aspergillus niger* (21 mm) minimum zone was formed *Escherichia coli* (20 mm) because ZnS and *Syzygium aromaticum* extract damaged cell membrane of bacteria and fungus basically cell membrane of microorganisms affect by iron source [15] and sulphide source. So the biosynthesized ZnS nanoparticles using *Syzygium aromaticum* extract have excellent antimicrobial activity against entire tested microorganism.

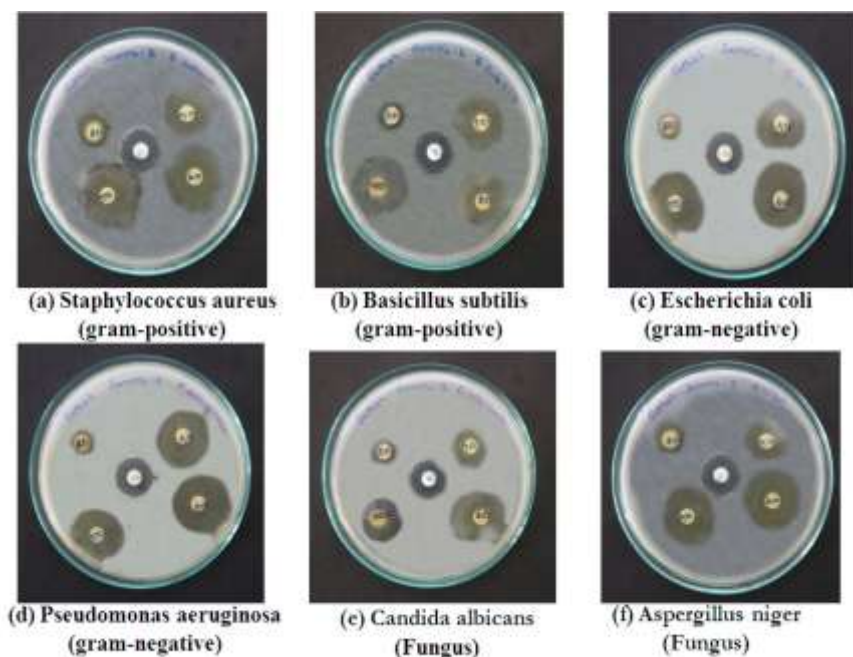


Figure 5: Antimicrobial activity of ZnS using *Syzygium aromaticum* extract

4. Conclusion

Biosynthesized ZnS nanoparticles using *Syzygium aromaticum* extract in first time we reported, characterization of ZnS nanoparticles were analyzed by using XRD, SEM, UV-Visible and FTIR. The antimicrobial activity of ZnS tested in vitro disc diffusion method using MHA plates obtained from Hi-Media Mumbai. Biosynthesized ZnS nanoparticles using *Syzygium aromaticum* are cubic zinc blende structure and average grain size was calculated ~ 46 nm using XRD analysis. Surface morphology of synthesized ZnS was found different zinc blende shapes of surface, size less than $2 \mu\text{m}$ by using SEM analysis, the sharp optical absorbance at 492 nm and different functional group of *Syzygium aromaticum* extract were found by using UV-Visible and FTIR spectrum. The antimicrobial activity of biosynthesized ZnS nanoparticles are tested different microorganisms like bacteria and fungus the maximum ZoI was formed *Candida albicans* (25 mm), minimum ZoI formed *Escherichia coli* (20 mm) at concentration $60 \mu\text{l}$. It was evident that biosynthesized ZnS have an excellent antimicrobial activity against all tested microorganisms.

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