FABRICATION OF AN ANTIMICROBIAL PAPER

Sherine Mathew[1], Prof.Chinnamma[2]
Malabar College of Engineering and Technology, Kerala Technology University, Thrissur (Dist),Kerala,India

Abstract

The word antimicrobial can be termed to any substance that is active against microbes. Infections can be occurred by any means, special care must be taken in preventing the spread of these microbes which may lead to serious treats. Antimicrobial paper can be termed as a paper that is active against microbes. In this project, an attempt has been made, for the production of an antimicrobial paper out of hay. The paper was traditionally made out of hay and antimicrobial properties were imparted by the synthesis of ZnO and Curcumin Longa. Synthesis can be defined as the combination of components or elements to form a connected whole. In synthesis the product impact all the properties of the components mixed. This antimicrobial paper is beneficial in various areas such as food covering, hospitals, sanitary etc. This would therefore prevent the spread of infections. Agar disc diffusion was also conducted to study the antimicrobial property.

Index Term- ***Antimicrobial, ZnO, Curcumin Longa***

I. INTRODUCTION

The word “antimicrobial” can be defined as any substance that is active against microbes. Infections can be spread by any means, special care must be taken in preventing the spread of these microbes or it would lead to serious treats. Antimicrobial paper can be simply explained as a paper that is active against microbes. Paper is widely used in all areas such as the food industry, as coverings, industries etc and at once when these papers are made antimicrobial their scope increases to a wider area. In this project an antimicrobial paper is manufactured from hay, turmeric powder and zinc dioxide. All the materials used are cheap, easily available, safe and economical. Here zinc dioxide that is synthesized with turmeric powder is embedded on paper that is made out of hay.An antimicrobial is an agent that kills microorganisms or stops their growth. Antimicrobial medicines can be grouped according to the microorganisms they act primarilyagainst.Forexample, antibiotics ar e used against bacteria and antifungals are used against fungi. They can also be classified according to their function. Agents that kill microbes are called microbicidal, while those that merely inhibit their growth are called biostatic. The use of antimicrobial medicines to treat infection is known as antimicrobial chemotherapy, while the use of antimicrobial medicines to prevent infection is known as antimicrobial prophylaxis.

The main classes of antimicrobial agents are disinfectants ("nonselective antimicrobials" such as bleach), which kill a wide range of microbes on non-living surfaces to prevent the spread of illness, antiseptics (which are applied to living tissue and help reduce infection during surgery), and antibiotics (which destroy microorganisms within the body). The term "antibiotic" originally described only those formulations derived from living microorganisms but is now also applied to synthetic antimicrobials, such as the sulphonamides, or fluoroquinolones. The term also used to be restricted to antibacterials (and is often used as a synonym for them by medical professionals and in medical literature), but its context has broadened to include all antimicrobials. Antibacterial agents can be further subdivided into bactericidal agents, which kill bacteria, and bacteriostatic agents, which slow down or stall bacterial growth. In response, further advancements in antimicrobial technologies have resulted

in solutions that can go beyond simply inhibiting microbial growth. Instead, certain types of porous media have been developed to kill microbes on contact. Antimicrobial use is known to have been common practice foratleast2000years. AncientEgyptians and ancientGreeks used specific molds and plant extracts to treat infection.

Antibacterials are used to treat bacterial infections. The drug toxicity to humans and other animals from antibacterials is generallyconsideredlow.(depends) Prolong ed use of certain antibacterials can decrease the number of gut flora, which may have a negative impact on health. Consumption of probiotics and reasonable eating can help to replace destroyed gut flora. Stool transplants may be considered for patients who are having difficulty recovering from prolonged antibiotic treatment, as for recurrent Clostridium difficile infections.

Antifungals are used to kill or prevent further growth of fungi. In medicine, they are used as a treatment for infections such as athlete's foot, ringworm and thrush and work by exploiting differences between mammalian and fungal cells. They kill off the fungal organism without dangerous effects on the host. Unlike bacteria, both fungi and humans are eukaryotes. Thus, fungal and human cells are similar at the molecular level, making it more difficult to find a target for an antifungal drug to attack that does not also exist in the infected organism. Consequently, there are often side effects to some of these drugs. Some of these side effects can be life-threatening if the drug is not used properly.

As well as their use in medicine, antifungals are frequently sought after to control mold growth in damp or wet home materials. Sodium bicarbonate (baking soda) blasted on to surfaces acts as an antifungal. Another antifungal serum applied after or without blasting by soda is a mix of hydrogen peroxide and a thin surface coating that neutralizes mold and encapsulates the surface to prevent spore release. Some paints are also manufactured with an added antifungal agent for use in high humidity areas such as bathrooms or kitchens. Other antifungal surface treatments typically contain variants of metals known to suppress mold growth e.g. pigments or solutions containing copper, silver or zinc. These solutions are not usually available to the general public because of their toxicity.

Antiviral drugs are a class of medication used specifically for treating viral infections. Like antibiotics, specific antivirals are used for specific viruses. They are relatively harmless to the host and therefore can be used to treat infections. They should be distinguished from viricides, which actively deactivate virus particles outside the body.

Many antiviral drugs are designed to treat infections by retroviruses, mostly HIV. Important antiretroviral drugs include the class of protease inhibitors. Herpes viruses, best known for causing cold sores and genital herpes, are usually treated with the nucleoside analogue acyclovir. Viral hepatitis is caused by five unrelated hepatotropic viruses and can be treated with antiviral drugs depending on the type of infection. influenza A and B viruses have become resistant to neuraminidase inhibitors such as oseltamivir and the search for new substances is on.

Antiparasitics are a class of medications indicated for the treatment of infection by parasites,suchas nematodes, cestodes, tr ematodes, infectious protozoa, and amoebae. Like all antimicrobials against intracellular microbes, they must kill the infecting pest without serious damage to the host.

II.MATERIALS AND METHODS

1. Curcuma Longa

Curcuma Longa is a medicinally important plant. Curcuma longa L. (turmeric) of ginger family (Zingiberaceae) belongs to the group of oldest cultivated spice plants in the south-east Asian countries. For many years rhizome of this plant has been used also as a safe and active drug for the

treatment of various.chronic diseases, especially of diabetes mellitus (DM). The active substance of turmeric - curcumin (diferuloylmethane), possesses multiple therapeutic properties. At the molecular level it has been stated that curcumin inhibits cell proliferation, metastasis creation and apoptosis. The leaves of Curcuma Longa required for this experiment were collected from a nearby area.

2. Hay

Hay is grass, legumes, or other herbaceous plants that have been cut and dried to be stored for use as animal fodder, particularly for large grazing animals raised as livestock, such as cattle, horses, goats, and sheep. However, it is also fed to smaller domesticated animals such as rabbits and guinea pigs. Even pigs may be fed hay, but they do not digest it as efficiently as herbivores. Hay can be used as animal fodder when or where there is not enough pasture or rangeland on which to graze an animal, when grazing is not feasible due to weather (such as during the winter), or when lush pasture by itself would be too rich for the health of the animal. It is also fed when an animal is unable to access pasture. Lv, E., Xia, W., Tang, M., and Pu, Y. in their paper "Preparation of an efficient oil-spill adsorbent based on wheat straw," explained about the adsorbent properties of wheat straw. And it was found successful.

3. Zinc Acetate

Zincacetate isa salt withthe formula $Zn(CH_3CO_2)_2$, which commonly occurs as the dihydrate $Zn(CH_3CO_2)_2 \cdot 2H_2O$. Both the hydrate and the anhydrous forms are colorless solids that are commonly used in chemical synthesis and as dietary supplements. Zinc acetates are prepared by the action of acetic acid on zinc carbonate or zinc metal. When used as a food additive, it has the E number E650. In anhydrous zinc acetate the zinc is coordinated to four oxygen atoms to give a tetrahedral environment, these tetrahedral polyhedra are then interconnected by acetate ligands to give a range of polymeric structures. In contrast, most metal diacetates feature metals in octahedral coordination with bidentate acetate groups.In zinc acetate dihydrate the zinc is octahedral, wherein both acetate groups are bidentate.the molar mass of Zinc Acetate is 83.48 g/mol and its solubility in water is 43 g/100 mL and if methanol it is 1.5 g/100 mL .

4. Paper Production

Papermaking, regardless of the scale on which it is done, involves making a dilute suspension of fibres in water, called "furnish", and forcing this suspension to drain through a screen, to produce a mat of interwoven fibres. Water is removed from this mat of fibres using a press. The method of manual papermaking changed very little over time, despite advances in technologies. The process of manufacturing handmade paper can be generalized into five steps:

a. Separating the useful fibre from the rest of raw materials.
b. Beating down the fibre into pulp
c. Adjusting the colour, mechanical, chemical, biological, and other properties of the paper by adding special chemical premixes
d. Screening the resulting solution
e. Pressing and drying to get the actual paper

Screening the fibre involves using a mesh made from non-corroding and inert material, such as brass, stainless steel or a synthetic fibre, which is stretched in a wooden frame similar to that of a window, this tool being knows as a paper mould. The size of the paper is governed by the open area of the frame. The mould is then completely submerged in the furnish, then pulled, shaken and drained, forming a uniform coating on the screen. Excess water is then removed, the wet mat of fibre laid on top of a damp cloth or felt in a process called "couching". The process is repeated for the required number of sheets. This stack of wet mats is then pressed in a hydraulic press. The fairly damp fibre is then dried using a variety of methods, such

as vacuum drying or simply air drying. Sometimes, the individual sheet is rolled to flatten, harden, and refine the surface. Finally, the paper is then cut to the desired shape or the standard shape (A4, letter, legal, etc.) and packed. So here the entire process can be simply explained as, Raw plant materials must be cut or crushed, retted that is, soaked in water to soften the fibers, and then cooked in an alkaline solution here washing soda is used. The duration of soaking and cooking and the strength and kind of alkaline all influence the condition of the plant fibers. The cooked fibers are rinsed thoroughly and bleached traditionally outdoors under the sun, but today often using chemical bleach. The fibers are then beaten and hydrated that is soaked in and mixed with water to form paper pulp. Both the natural minerals in and the temperature of the water influence the qualities of the paper produced. The cooked fibers are rinsed thoroughly and bleached traditionally outdoors under the sun, but today often using chemical bleach. The fibers are then beaten and hydrated that is soaked in and mixed with water to form paper pulp. Both the natural minerals in and the temperature of the water influence the qualities of the paper produced. . The first paper proved to be thicker, however consequent papers made became thinner. The paper produced had a brown shade to it which could be cleared to certain extend with the help of bleaching powder.

Fig.1.Homemade Paper

5. Preparation of Plant Extract

For the preparation of plant extract 5g of sundried Curcuma Longa powder was taken and soaked in a beaker containing 100ml water. The solution was boiled at 70˚C for 8min. The leaf extract was allowed to cool to room temperature, filtered through Whatman number-1 filter paper, and the filtrate was stored for further experimental use.

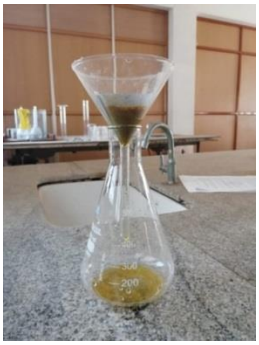

Fig.2.Collection of Filtrate

6. Synthesis of ZnO and Curcuma Longa

22g of Zinc Acetate was dissolved in 100ml water and kept in stirrer for 1hr. Then 20ml NaOH solution was slowly added into the Zinc Acetate solution and 25ml of plant extract was added to the same. The mixture was kept in stirrer for 3hrs and then was kept for incubation. The color of the reaction mixture was changed after 1hr of incubaton.

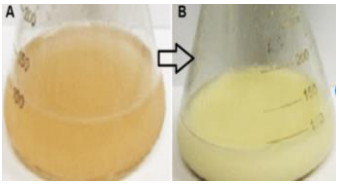

Fig.3.Confirming end point of synthesis

7. Charachterisation

Following were the methods that were used to characterize the synthesized product. Each and every test was conducted so that a detailed study could be conducted. X-ray powder diffraction (XRD) is a rapid analytical technique primarily used for phase identification of a crystalline material and can provide information on unit cell dimensions. The analyzed material is finely ground, homogenized, and average bulk composition is determined.A scanning electron microscope (SEM) is a type of electron microscope that produces images of a sample by scanning the surface with a focused beam of electrons. The electrons interact with atoms in the

sample, producing various signals that contain information about the surface topography and composition of the sample. The electron beam is scanned in a raster scan pattern, and the position of the beam is combined with the intensity of the detected signal to produce an image. In the most common SEM mode, secondary electrons emitted by atoms excited by the electron beam are detected using an Everhart-Thornley detector. The number of secondary electrons that can be detected, and thus the signal intensity, depends, among other things, on specimen topography. SEM can achieve resolution better than 1 nanometer. Ultraviolet–visiblspectroscopy or ultraviolet–visible spectro spectrophotometry (UV–Vis or UV/Vis) refers to absorption spectroscopyor reflectance spectroscopy in part of the ultraviolet and the full, adjacent visible spectral regions. This means it uses light in the visible and adjacent ranges. The absorption or reflectance in the visible range directly affects the perceived color of the chemicalsinvolved. In this region of the electromagnetic spectrum, atoms and molecules undergo el ectronic transitions. Absorption spectroscopy is complementary to fluorescence spectroscopy, in that fluorescence deals with transitions from the excited state to the ground state, while absorption measures transitions from the ground state to the excited state.Antimicrobial activity refers to the process of killing or inhibiting the disease causing microbes. Various antimicrobial agents are used for this purpose. Antimicrobial may be anti-bacterial, anti-fungal or antiviral. They all have different modes of action by which they act to suppress the infectionThe disk diffusion test, or agar diffusion test, or Kirby–Bauer test (disc-diffusion antibiotic susceptibility test, disc-diffusion antibiotic sensitivity test, KB test), is a test of the antibiotic sensitivity of bacteria. It uses antibiotic discs to test the extent to which bacteria are affected by those antibiotics. In this test, wafers containing antibiotics are placed on an agar plate where bacteria have been placed, and the plate is left to incubate. If an antibiotic stops the bacteria from growing or kills the bacteria, there will be an area around the wafer where the bacteria have not grown enough to be visible. This is called a zone of inhibition.

III. RESULTS AND DISCUSSIONS

1.UV-Visible Analyses

UV-visible spectroscopy is usually conducted to confirm the synthesis of ZnO NPs. Conducting electrons start oscillating at a certain wavelength range due to surface plasmon resonance (SPR) effect.Fig.4 represents the UV-visible spectra of freshly prepared ZnO NPs. Peak obtained at 378 nm clearly demonstrates the presence of ZnO NPs in the reaction mixture. Initial peak obtained at range of 420 nm got further raised due to oscillation of more electrons after 5 h which depicts the continuous synthesis of ZnO NPs.

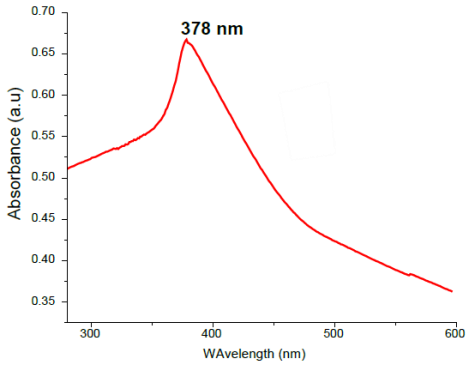

Fig.4. UV-Vis absorption spectra of ZnO nanoparticles synthesized using Curcumin Longa

2. SEM

SEM analysis is done to visualize shape and size of nanoparticle. Carl-Zeiss scanning electron microscope was used to study the morphological characteristics of synthesized zinc oxide. Magnification ranges of 10μm clearly demonstrated the presence of spherical shaped nanoparticle.

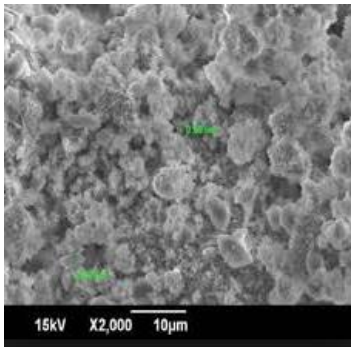

Fig.5. SEM image 10µm magnification

3. XRD Analysis

XRD Spectra provides an insight about the crystallinity of nanoparticle. Fig.6. represents XRD Spectra of ZnO NPs synthesized using curcumin longa extract. Size of the nanoparticle was calculated using Debye–Scherrer equation. X-ray diffraction peaks obtained at 31.8 °, 34.44 °, 36.29 °, 47.57 °, 56.61 °, 67.96 ° and 69.07 ° corresponded to the lattice plane of (1 0 0), (0 0 2), (1 0 1), (1 0 2), (1 1 0), (1 1 2), (2 0 1) Suggesting the face-centered cubic (fcc) crystal structure of the nanoparticle. Joint Committee on Powder Diffraction Standards (JCPDS) was used as a reference to assign the lattice planes according to the peaks obtained. Average size of the synthesized nanoparticle was found to be. 37.67 nm. $D = K\lambda\ \beta cos\theta$ where, D- particle size in nm, λ- X-ray wavelength, β- FWHM, θ- Bragg's angle of reflection. Table.1.represents the FWHM value for every peak assigned for particle size calculation.

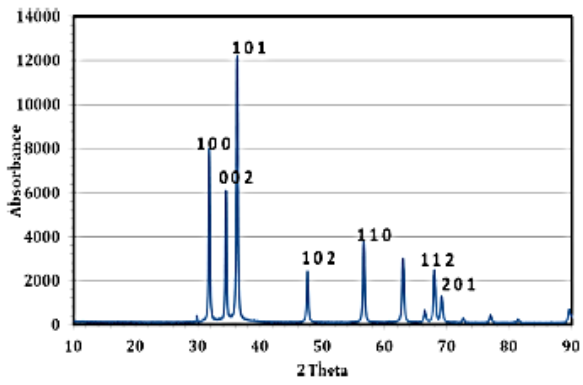

Fig.6. XRD Spectrum of ZnO NPs

Table.1. Parameter Calculation for Average Size Calculation for Nanoparticle

2Θ	hkl	FWHM(β)	D(nm)
31.8	100	0.152	56.77
34.44	002	0.192	45.26
36.29	101	0.238	36.7
47.57	102	0.309	29.36
56.61	110	0.217	43.44
67.96	112	0.37	27.05
69.07	201	0.401	25.11

4. Antimicrobial Activity

Figures 7 and 8 show the antibacterial activity of ZnO nanoparticles synthesised from curcumin longa on *E. coli, S. aureus, Streptococcus sp and, Enterococcus sp.* bacteria respectively. Antibacterial activity results showed that the ZnO nanoparticle synthesised using Curcumin Longa has exhibited strong antibacterial activity against all bacteria. Moreover, ZnO nanoparticles performed better antibacterial activity on gram-positive (*S. aureus*) than the gram-negative (*E. coli*) bacteria in comparison to standard antibiotics. The previous research report also indicated antibacterial activity of ZnO particles were greater on gram-positive than gram-negative bacteria. They suggested that the outer thick peptidoglycan layer and other surface components of grampositive bacteria may promote ZnO attachment onto the cell wall whereas the components of gram-negative bacteria may repeal this attachment.This discussion points out the importance of understanding the mechanism of ZnO antimicrobial activity. Generally, the growth inhibition of all the bacteria can be increased by increasing the concentration of ZnO nanoparticles in discs. But it was also noted that more than 25µl of the fluid could not be carried by the discs.

As the paper discs couldn't carry more than 25µl, readings were taken from lesser value. That is the concentration taken were of four different ranges that had lesser

value than 25 µl, the values taken were 5 µl ,10 µl ,15µl, 20µl

From table 2 it would be clear that the highest effectiveness was found for gram positive bacteria. Maximum zone of inhibition of 20mm was found in gram positive bacteria whereas the antibiotics could give only 11mm. Hence the best result was found in S. aureus at 20µl, and the least for E.coli bacteria.

Table.2. Zone of Inhibition reading(mm)

Sample name	Antibiotics	5µl	10 µl	15 µl	20 µl
E.coli	10	-	3	4	5
S.aureus	11	-	8	17	20
Streptococus. Sp	10	-	6	10	12
Entrococus. Sp	11	-	11	12	13

Fig.7.Zone of inhibition for S. aureus and Entrococcus.sp

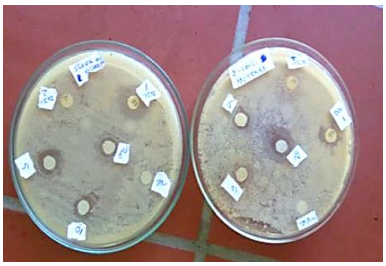

Fig.8. Zone of inhibition for Streptococcus Sp and E.coli

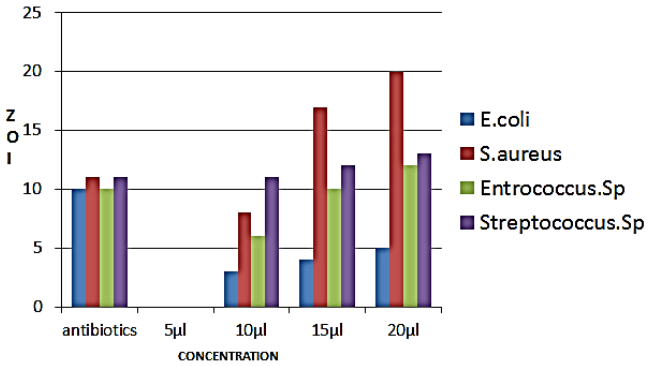

Fig.9. Graphical representation of antimicrobial activity

IV. CONCLUSION

Thus after successful completion of the work it can be concluded that the antimicrobial paper produced out of hay as an easy, cost-effective, environmental friendly product. It is easy to produce and has got widespread application. However commercializing of this product would require further study on various factors such as type of hay, concentration, amount to be sprayed per each paper, size, and economical part. Various other facts can also be concluded from this project. Such as the effectiveness of synthesis, and how effective is biological synthesis compared to mechanical and chemical synthesis. Zinc Oxide as an very effective antimicrobial was also studied. The medicinal importance of curcumin longa was also studied. XRD sujjested FCC structure and SEM suggested spheroidal shape of Zinc and UV-analyses confirmed the presence of Zinc Oxide after synthesis with curcumin longa. Thus as a whole, the entire process can be concluded as an effective, eco-friendly approach.

REFERENCES

1) *Amna Sirelkhatim, Shahrom Mahmud , Azman Seeni,, Noor Haida Mohamad Kaus , Ling Chuo Ann , Siti Khadijah Mohd Bakhori,,Habsah Hasan, Dasmawati Mohamad, "Review on Zinc Oxide Nanoparticles:*

Antibacterial Activity and Toxicity Mechanism" ,11 March,2015.

2) *E S Mones , "Fabrication of Zinc Oxide embedded Kapok Paper", IOP Conference series: Material Science and Engineering, 2017.*
3) *Rajani Bharati, S. Suresh, " Biosynthesis of ZnO/SiO_2 nanocatalyst with palash leaves' powder for treatment of petroleum refinery effluent", Resource Efficient Technologies, Vol 3, pg 528-541, 2017.*
4) *Happy Agarwal, S. Venkat Kumar, S. Rajeshkumar , ' A review on green synthesis of zinc oxide nanoparticles –An eco-friendly approach" , Resource Efficient Technologies, Vol 3, pg 406-413, 2017.*
5) *Ibrahim Khan, Khalid Saeed, Idrees Khan, "Nanoparticles: Properties, applications and toxicities", Arabian Journal of Chemistry, 18 March 2017.*
6) *Soumya Menon, Rajeskumar S, Venkat kumar S, "A review on biogenic synthesis of gold nanoparticles, characterization, and its applications" ,journal on Resource-Efficient Technologies,Vol 3, pg.516-527,14 August 2017.*
7) *Arumugam Sudha, Jeyaraman Jeyakanthan, Pappu Srinivasan, "Green synthesis of silver nanoparticles using Lippia Nodiflora aerial extract and evaluation of their antioxidant, antibacterial and cytotoxic effects" Journal on Resource-Efficient Technologies, Vol 3, pg 506-515, 14 August 2017.*
8) *J. Santhosh Kumar, S. Venkat Kumar, S. Rajesh Kumar, Synthesis of zinc oxide nanoparticle using plant leaf extract against urinary tract infection pathogen, journal Resource-Efficient Technologies, Vol 3, pg 459-465, 22 May 2017*

Sherine Mathew *received B.tech degree in civil engineering from Tejus Engineering College, Vellarakad in 2016 and currently pursing M.tech environmental engineering from Malabar College of Engineering and Technology,Desamangalam.*