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DEVELOPMENT OF HYDROGEL BEAD NANOCOMPOSITE USING TEA DUST FOR THE AQUEOUS BIOMEDICAL WASTE MANAGEMENT

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Keywords:

Aqueous Biomedical waste, Nanocomposite, Tea dust, Hydrogel Bead, Adsorption, Nanoparticles

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ABSTRACT: Most of the Bio-medical wastes got dumped in highly eminent aquatic sources. To control this pollution due to biomedical wastes, the current study focused on the development of hydrogel bead nanocomposite incorporated with the adsorbent and nanoparticles along with biopolymer. The formed hydrogel bead nanocomposite acts as an effective adsorbent of infectious microbes, heavy metals and pollution causing materials. The conjugation efficiency of the activated tea dust, zinc oxide nanoparticles, and bioconjugated system was analyzed using UV-Vis spectroscopy which shows the maximum absorption at 330nm, 379nm, and 580nm in the conjugation system. Further, this was analyzed using FTIR shows good functional groups stretching, Dynamic Light Scattering analysis depicts the particle size as 5.203µm and has good stability. The effectiveness of the hydrogel bead nanocomposite on the wastewater sample was characterized through solubility test and heavy metal adsorption ability. The bead does not solubilize in the water sample over a long time which shows the high stability of the conjugated system. Whereas the adsorption efficiency (%R) of the hydrogel bead in the Sengulam lake water sample was found to be 90.29% and from Kurichi lake it was found to be 4.21%. This hydrogel bead containing nanocomposite acts as an eco-friendly product, it can be bio-degradable and does not cause any threat to the marine source. It can be act as a good adsorbent of various forms of biological wastes.

INTRODUCTION: Any solid or liquid waste which may be found as a threat of infection to humans and aquatic organisms termed as bio-medical waste (BMW). It is mainly established from healthcare sectors such as hospitals, blood banks, laboratories and research institutes, veterinary hospitals.

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The biomedical wastes mostly composed of blood solution, sharps, microbiological culture and stalks, human or animal tissue, used bandages, dressings, and discarded gloves [1]. And also, it contains some toxic heavy metals like Cu, Pb and Zn [2]. Most of the biomedical wastes are disposed off in aqueous regions [3]. This causes genetic modification in marine organisms, and this consecutively leads to change in the human genome. To control the water pollution and reducing the toxicity of heavy metals in water bodies the study focused on the development of nanocomposite hydrogel beads derived from waste tea dust as a natural adsorbent of Various infectious threats [4, 5, 6].

To overcome this problem, the nanocomposite of hydrogel bead was synthesized using naturally extracted polymer from marine source and was polymerized with activated tea dust and nanoparticle to form a nanocomposite [7]. The Zinc oxide nanoparticles have the property of anti-microbial and adsorb heavy metals which are synthesized using the Green synthesis method by the extraction of *Eichhornia crassipes* (Water hyacinth). The bio-conjugate of adsorbent and nanoparticles was prepared by the physical binding method. The biopolymer chitosan material was synthesized using chitin from marine sources [8, 9]. The hydrogel bead was formed by the incorporation of bio-conjugate into a biopolymer along with cellulose for a rigid structure. The present study is mainly focused on the usage of waste tea dust as adsorbent, the zinc oxide as a nanoparticle, and chitosan cellulose as biopolymer material. The formation of rigid hydrogel bead can be useful for the adsorption of aqueous biomedical waste [10]. At present, a series of studies have been undergoing for adsorption of wastes. This study will provide the characteristics of developed nanocomposite beads and the adsorption property of waste materials on to the developed beads. The hydrogel bead can be a useful product for the prevention and adsorption of biomedical waste from contaminated water sources.

MATERIALS AND METHODS:

Materials: All chemicals used in this process are of an analytical grade of Indian Research Products. The chemicals used are Zinc Oxide, Sulphuric acid, Hydrochloric acid, Sodium Hydroxide, PBS Buffer (Sodium chloride, Potassium chloride, Di-Sodium Hydrogen Phosphate, Potassium di-Hydrogen Phosphate), Acetic acid, Cellulose, Sodium Alginate in a small ratio.

Methodology:

Activation of Tea dust Adsorbent: The domestic tea waste was collected, cleansed to remove the impurities. The tea dust was boiled with distilled water at 80º C to remove tannin and caffeine. It was decolorized by washing several times through distilled water. The decolorized waste tea dust was oven-dried at 50 ºC for 12 h and sieved (100µm). The sieved tea dust was further activated by agitating vigorously in Sulphuric acid [11]. It was washed with distilled water to obtain neutral pH and oven-dried at 50 ºC for 10 h. The adsorbent was stored in a bottle for further use [12].

Green Synthesis of Zinc Oxide Nanoparticles: *Eichhornia crassipes* (Water hyacinth) were collected from the lakes in abundance. The leaves of *Eichhornia crassipes* (Water hyacinth) was cut into pieces and boiled to collect extract at 70 ºC [13]. The extract was filtered and added with Zinc Oxide powder. The solution was stirred and boiled for about 30 min at 70 ºC. The extract was cooled at room temperature and centrifuged at 5000 rpm. The pellet collected was oven-dried at 50 ºC overnight. The Zinc Oxide nanoparticles were collected and stored at room temperature for further use.

Physical Binding of Nanoparticles with Adsorbent: The Zinc Oxide nanoparticles and adsorbent were physically bound in the ratio of 1:2 respectively. The binding solution PBS Buffer [14] was prepared and added with an adsorbent: nano-particles mixture. The solution was allowed to stir overnight. The collected sample was centrifuged; the pellet collected was oven-dried. The Bio-conjugated nanoparticles were stored at room temperature for further use [15].

Preparation of Chitosan from Chitin Flakes: The exoskeleton of the crab shell was collected, washed, and sundried. The dried shell was mechanically grounded. The sample powder was deproteinized using 1.2M NaOH by boiling at 70 ºC for about 1 h. The obtained sample was washed severally with distilled water [16, 17]. The sample was demineralized using 0.7M HCl, boiled at 70 ºC for 15 min, and washed severely using distilled water. Deacetylation of the sample was done using 50% of NaOH, boiled at 100 ºC for 3 hours. Wash the sample with distilled water until the pH adjusted to neutral [18]. The sample was oven-dried, sieved and stored at room temperature for further use.

Development of Hydrogel Bead: The 100ml of 2% acetic acid and 2g of chitosan were mixed and stirred at 70 ºC for about 6 h. 2g of cellulose is then added to the solution along with bio-conjugated powder and stirred at 200rpm at room temperature for 6 h. The blended solution was injected into droplets into a 1M Sodium hydroxide/Calcium chloride to form hydrogel beads and allowed to stay in the solution for hardening for 12 h [19, 10].

The beads were then separated from the solution and stored at room temperature for further adsorption studies.

Characterization of Nanocomposites and Hydrogel Bead: The developed hydrogel bead was characterized by different spectroscopic methods.

UV-Visible Spectrophotometer: The Ultra-Violet spectroscopy works under the principle of Beer Lambert's law. The visible spectrum was taken for the quantitative analysis of absorbance of waste tea dust, zinc oxide, chitosan, bioconjugate and hydrogel bead adsorption. Using Systronics UV-Spectrophotometer 119, the analytics was found between the ranges of 300-700 nm [20].

Fourier-Transform Infrared Spectroscopy (FTIR): FTIR is an analytical technique used to identify functional groups and chemical properties. The resultant spectrum was analyzed over a width of 4000-400cm $^{-1}$. The capability and stabilization of samples of tea dust, zinc oxide, chitosan, and bio-conjugate were obtained [20].

Particle Size Analysis: The particle size works under the principle of Beam of Light, scattered by a group of particles, the angle of light scattering is inversely proportional to particle size. It measures the average particle diameter in nm. The measurement of Zinc Oxide nanoparticles was observed [21].

Zeta Potential Analysis: The zeta potential works under the principle of Laser Doppler Electrophoresis. It measures the electrical charges on the surface of the particle in suspension. The charge of Zinc Oxide and Bio-conjugate was measured [21].

Scanning Electron Microscopy (SEM): A scanning electron micrograph is used to detect the topology of the surface morphology of a particle under the scattering of light by focusing on different magnifications. The topography of Zinc Oxide and Bioconjugate samples were identified.

RESULTS AND DISCUSSION:

UV-Visible Spectroscopy: The ultra-violet spectrophotometer was used to analyze the concentration of a particular compound in a mixture and was given below in the **Fig. 1(A)** - zinc oxide, (B) - tea dust, (C) – bio-conjugate, and (D) - chitosan.

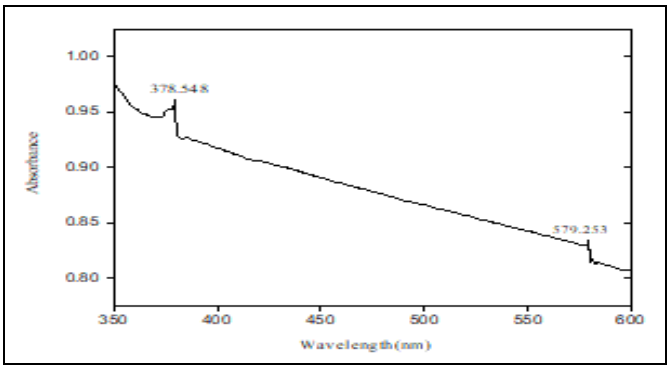

FIG. 1A: UV-VISIBLE SPECTRA OF ZINC OXIDE NANOPARTICLES

The ultra-violet spectrophotometer for Zinc Oxide nanoparticle was analyzed and the adsorption peak was observed between ranges of 378nm and 578nm. It was due to agglomeration and settling of nanoparticles in the cuvette, decreasing the radiation of absorbance [22].

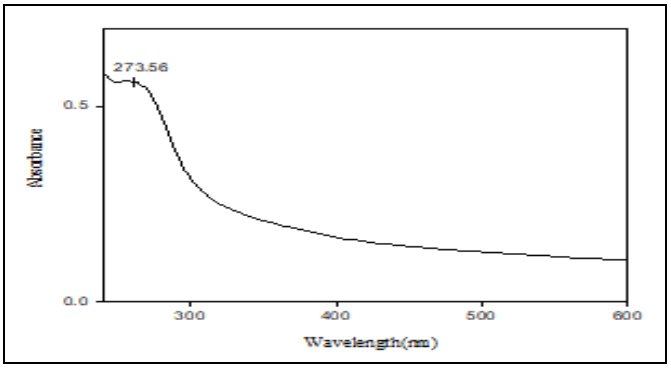

FIG. 1B: UV-VISIBLE SPECTRA OF TEA DUST

The ultraviolet spectrophotometer of waste tea dust was analyzed and the adsorption peak was observed as 273nm. It was due to the excitation of tea dust nanoparticle from the ground state to an excited state.

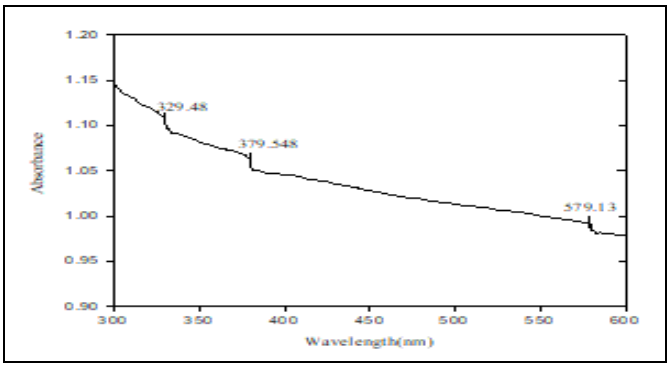

FIG. 1C: UV-VISIBLE SPECTRA OF NANOPARTICLE AND TEA DUST BIO-CONJUGATE

The ultra-violet spectrophotometer for bio-conjugate was analyzed and the adsorption peak was observed between the range of 330nm, 379nm, and 580nm. This bioconjugate has the absorption peak as of tea dust absorption peak at 330 nm and peak as of Zinc oxide nanoparticle at 379nm and 580nm. There was a slight shift in the peak from the un-bioconjugated particles, which was due to the excitation of the nanoparticles during conjugation.

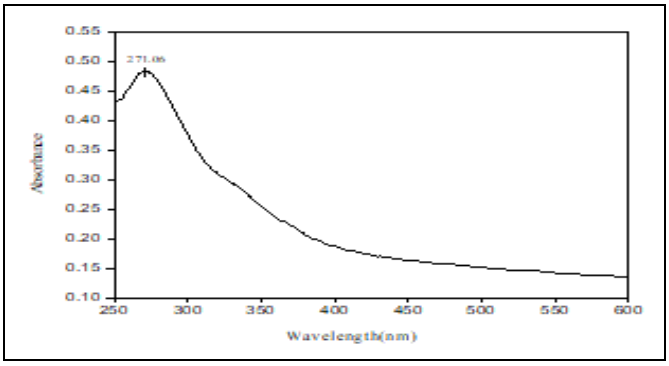

FIG. 1D: UV-VISIBLE SPECTRA OF CHITOSAN

The ultra-violet spectrophotometer for chitosan was analyzed, and the adsorption peak was observed at 271nm. It was due to the excitation of chitosan particles from the ground state to an excited state.

FTIR Spectra: The synthesized samples were analyzed using FTIR spectra for the prediction of functional groups present in the ranges of 4000-400 cm $^{-1}$. The stretching of the functional groups for each sample was analyzed using the obtained graphs [23]. The study of capability and stability of functional groups in the sample graphs are given below in **Fig. 2** (A) – Zinc oxide, (B) – tea dust, (C) – bio-conjugate, and (D)-chitosan.

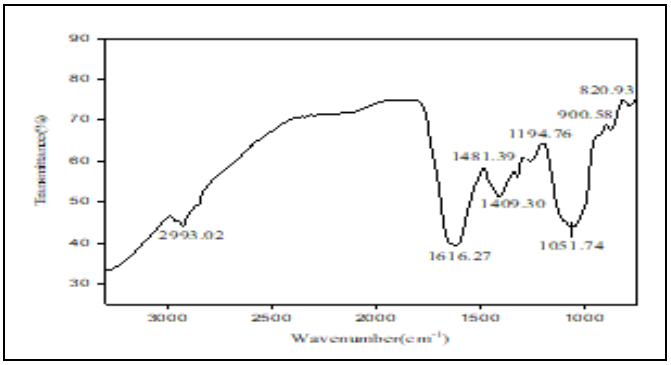

FIG. 2A: FTIR SPECTRA OF ZINC OXIDE NANOPARTICLES

The stretching of functional groups of Zinc oxide nanoparticles include =C-H bending from the range of 800-1000 cm $^{-1}$. The strong stretching of the C-O functional group was found between the ranges of 1000-1200 cm^{-1}. Strong bending was found between the peaks of 1400-1600cm^{-1} and the strong stretch of the C-H functional group was found between the ranges of 2900-3000 cm $^{-1}$ [24].

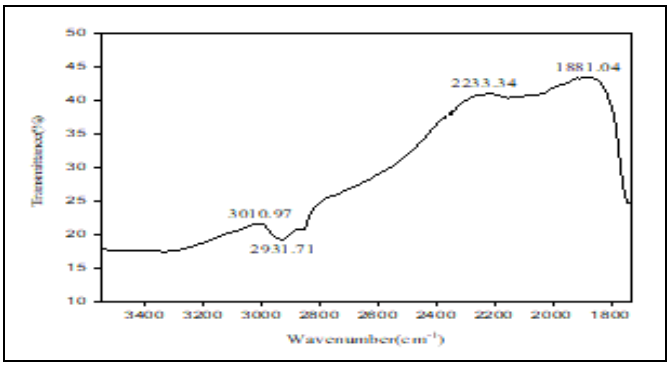

FIG. 2B: FTIR SPECTRA OF TEA DUST

The stretching of the two bands of functional group C=O was found between the ranges 1800-1900 cm^{-1}. The very strong bond was formed by the stretching of functional group O-H ranges from 2700-3100 cm^{-1}. This shows the presence of carbon bonding in tea dust nanoparticles which acts as an adsorbent.

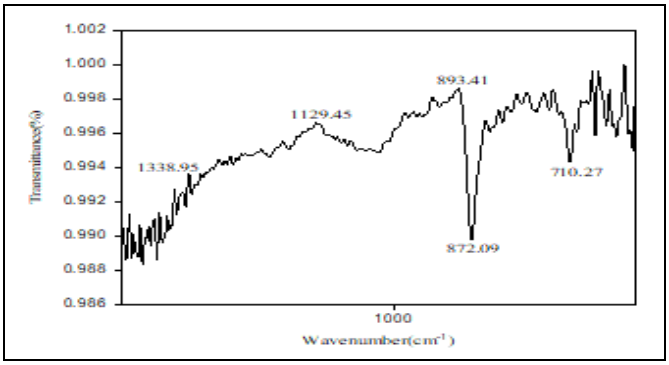

FIG. 2C: FTIR SPECTRA OF CHITOSAN

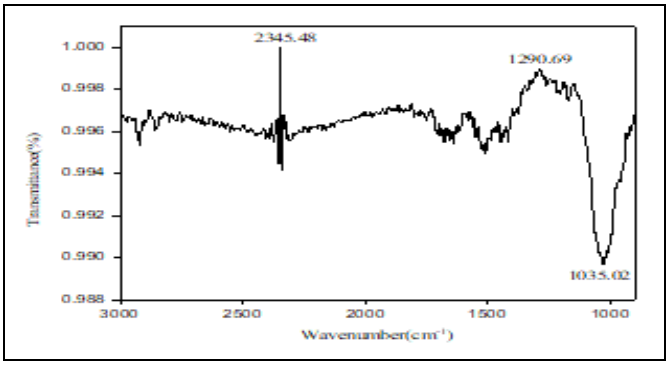

FIG. 2D: FTIR SPECTRA OF NANOPARTICLE AND TEA DUST BIO-CONJUGATE

The strong stretching of functional group C-Cl was found between the ranges 600-800 cm^{-1}. The

stretch between the ranges 800-1000 cm^{-1} consists of the functional group of strong bond =C-H. The strong stretching bond was formed between the ranges 1000-1400 cm^{-1} with functional group C-O. The noise found in the graph is due to the absorbance of water as moisture during analysis. So the noise has been neglected in the graph.

The Bio-conjugate consists of a combination of tea dust and zinc oxide nanoparticles. The results consist of the binding of adsorbent and nanoparticles thoroughly by analyzing graphs obtained. The stretching and functional groups were found similar to both the nanoparticle samples. The stretching of the strong bond was found between the ranges 1000-1200 cm^{-1} with functional group C-O. The strong bond of functional group O-H was found between the ranges 2000-3000 cm^{-1}. The noise found in the graph was neglected.

Particle Size Measurement: The particle size analyzed to any of the solid or liquid, or gaseous particles. The particle size of a nanoparticle is generally measured between 1-100 nm in diameter. Sometimes the diameter can be the extent to 100-500 nm. The particle size of zinc oxide nanoparticles was measured at 25 ºC. The Z-Average valve of the nanoparticle is 5203.0 nm which was determined as the averaged particle diameter.

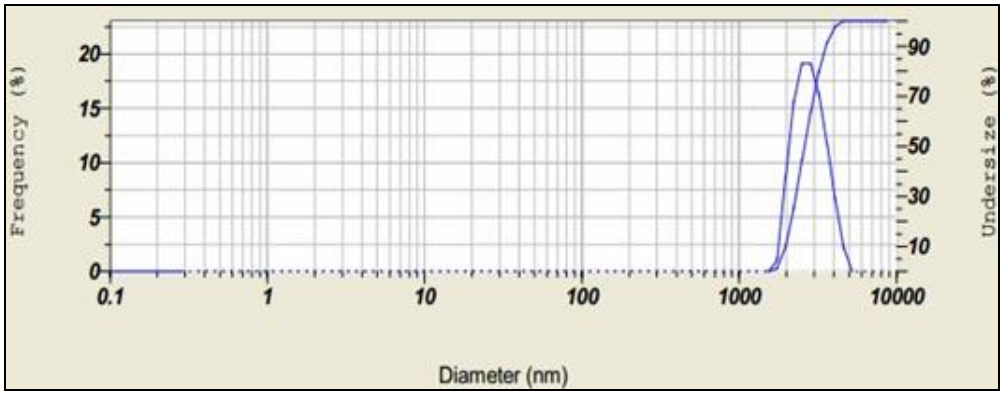

FIG. 3: PARTICLE SIZE DISTRIBUTION OF ZINC OXIDE NANOPARTICLES

The polydispersity index of the nanoparticle was measured as the ratio of the square of the standard deviation to the mean diameter of the particle size. The PI index of the zinc oxide nanoparticle was found to be 77.442 nm. Hence the Z-Average and PI index of zinc oxide were found to be 5.203µm and 0.077µm respectively.

Zeta Potential Measurement: Zeta potential for zinc oxide and bio-conjugate are shown in **Fig. 4 (A)** and **(B)**. The zeta potential is the measure of electric charge or potential difference on the surface of the colloidal particle immersed in colloidal dispersions. The zeta potential for the Zinc Oxide nanoparticle [25] and bio-conjugate was carried out at 25 ºC temperatures. The valve of Zeta potential for zinc oxide showed at the peak of -1.1 mV and the value ranges between -35 mV to +35 mV.

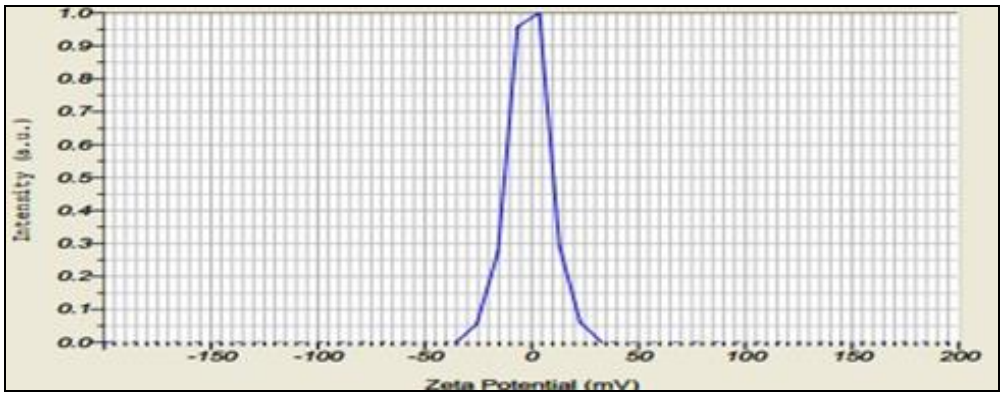

FIG. 4A: ZETA POTENTIAL DISTRIBUTION OF ZINC OXIDE NANOPARTICLE

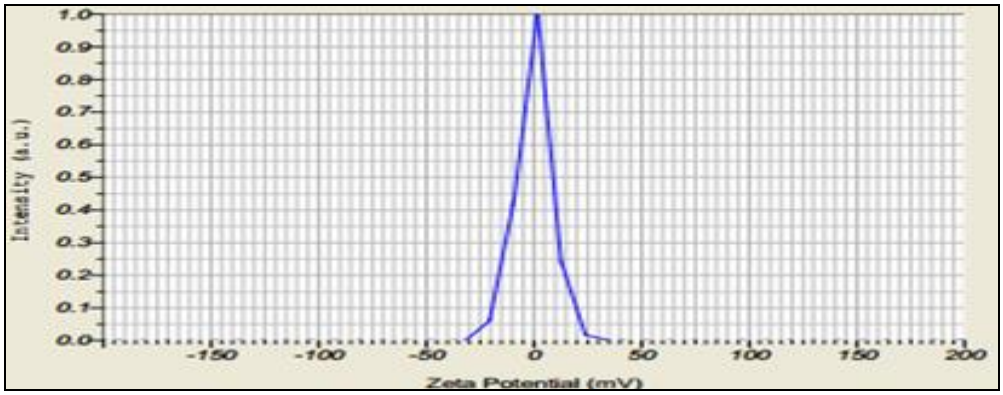

FIG. 4B: ZETA POTENTIAL DISTRIBUTION OF NANOPARTICLE AND TEA DUST BIO-CONJUGATE

Similarly, the zeta potential value of bio-conjugate showed at sharp peak of -0.2 mV and the value ranges between -35 mV to +35 mV. The negative value of the zeta potential indicates the stability of nanoparticles in zinc oxide and bio-conjugate.

Scanning Electron Microscopy (SEM): The SEM micrograph measures the surface topology of the nanoparticle. The SEM with Energy-dispersive X-ray spectroscopy (EDX) is measured together for the chemical analysis of compounds present in the sample particles. The SEM with EDX was analyzed for Zinc oxide nanoparticle and bio-conjugate [26]. The SEM for zinc oxide nanoparticles and bio-conjugate were measured under different magnifications of 10k, 20k, 55k and 5k, 10k, 20k, 30k respectively showed in the below **Fig. 5(A), (B), (C)** and **Fig. 6 (A), (B), (C), (D)**. The EDX analysis consists of the chemical compounds present in the samples which are represented below in **Fig. 7(A)** and **(B)**.

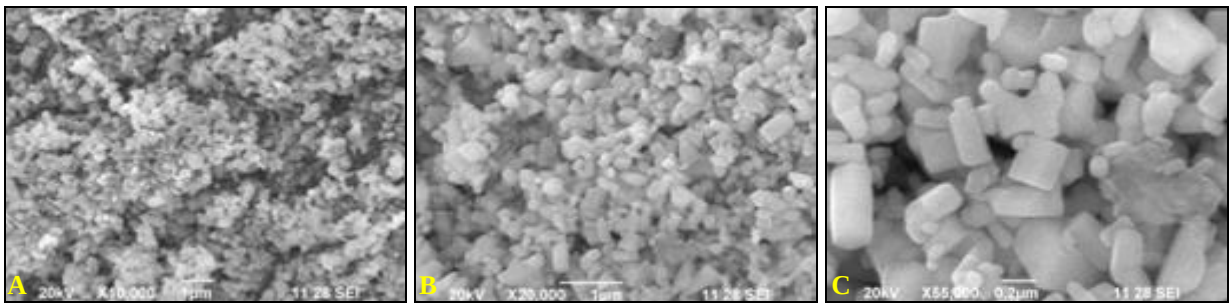

FIG. 5: SEM IMAGE OF ZINC OXIDE NANOPARTICLE AT MAGNIFICATIONS OF (A)-10k, 1µm, (B)- 20k, 1µm and (C)-55k, 2 µm

The above results of SEM for zinc oxide nanoparticles were found to be rod-shaped. The size of the nanoparticle was found to be between 100-200nm.

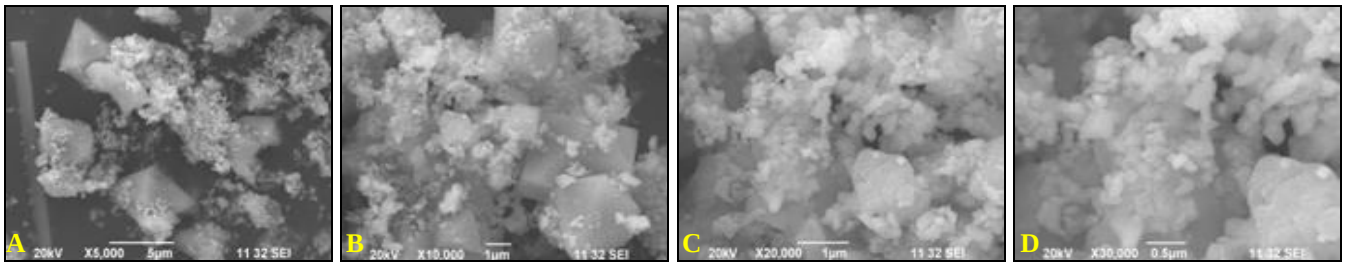

FIG. 6: SEM IMAGE OF BIO-CONJUGATE AT MAGNIFICATIONS OF (A)-5k, 5 µm, (B)-10k, 1 µm, (C)-20k, 1 µm and (D)-30k, 0.5µm

The above results of SEM for Bio-conjugate were found to be in cuboids shape. The surface morphology refers to the binding of tea dust particles with the zinc oxide nanoparticles. Hence, the result shows the physical binding of nanoparticles to form a bio-conjugate. The size of the nanoparticle was found to be 100-200 nm.

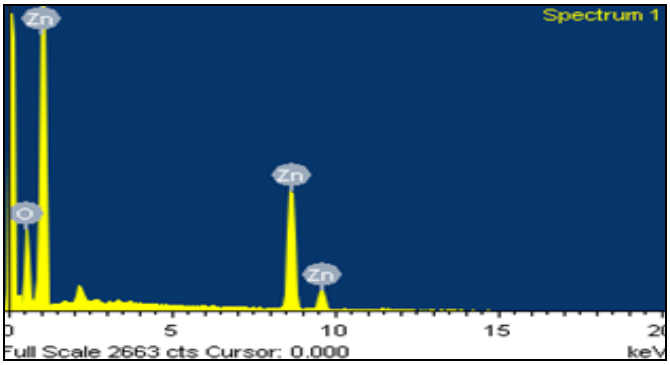

FIG. 7A: EDX MICROGRAPH OF ZINC OXIDE NANOPARTICLE

EDX graph shows the elemental composition of Zinc Oxide nanoparticles. It was found that the spectrum has high concentration of Zn and O [27]. The higher intensity of other peaks was due to an increase of calcinations. The presence of high concentration of Zn and O molecules determines the purity of the zinc oxide nanoparticles.

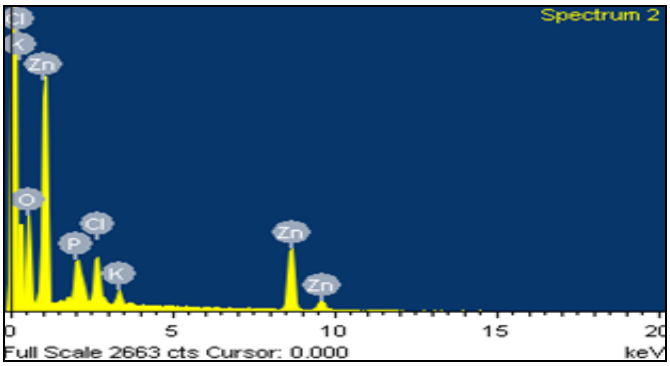

FIG. 7B: EDX MICROGRAPH OF NANOPARTICLE AND TEA DUST BIO-CONJUGATE

From the EDX micrograph, the elemental composition of the bioconjugate was analyzed. The various molecules present in the bioconjugate micrograph were due to the physical binding of tea dust and zinc oxide nanoparticles. The bioconjugate shows the higher concentration of the following elements, which include K, Cl, Zn, O, and P. During the physical binding, the PBS buffer was used; hence the K, Cl and P molecules are traced. The rest Zn and O are from the zinc oxide nanoparticle. As the tannin, caffeine, and other molecules are removed and made into adsorbent like activated carbon; hence tea dust does not trace for any chemical compounds.

This also referred the purity of adsorbent used. The increased peaks showed the high intensity of the presence of chemical compounds in the bio-conjugate.

Solubility Test: The prepared hydrogel bead through a simple gel entrapment method was tested with the solubility in water. The hydrogel bead acts in water so this test was done using the samples collected from the various water sources. The hydrogel bead was allowed to stay in the water for 24 h and tested with the solubility in water. The ratio of solubility was found to be negligible. Hence the product stands good for the solubility in water solutions. The adsorption test can be carried out after the solubility test.

Adsorption of Impurities by Hydrogel Bead: The developed hydrogel bead with a nanocomposite of treated tea dust has effective adsorption of various biological impurities present in water bodies. As this study concentrated on adsorption of biomedical wastes present in water bodies, the developed hydrogel bead was immersed in water collected from water sources from Sengulam and Kurichi lakes which is highly contaminated with biological wastes at Coimbatore. These water samples were treated with the developed hydrogel bead for 24 h. After 24 h the sample was analyzed using a UV-Vis spectrophotometer at 200nm. On zeroth hour the water sample taken from Sengulam Lake has its concentration as 0.886 mg/ml. After 24th h its concentration was reduced to about 0.086 mg/ml.

Also, the sample from Kurichi Lake has its zeroth concentration of 0.570 mg/ml. After 24th h its concentration was found to be 0.546 mg/ml. In both cases, the concentration was reduced. This reduction indicates the adsorption of heavy metals present in the water samples by the developed hydrogel bead. Using the impurities concentration at a zeroth hour and 24th hour the adsorption efficiency can be calculated with the following formula [28, 29].

$$\%R = [(C0 - Cf)/ C0] \times 100$$

TABLE 1: ADSORPTION OF BIOLOGICAL IMPURITIES BY HYDROGEL BEAD IMMERSED IN THE WATER

S. no.	Water sample	Concentration of impurities at 0th hour (mg/ml) (C0)	Concentration of impurities at 24th hour (mg/ml) (Cf)	% of adsorption (%R)
1	Sengulam Lake	0.886	0.086	90.29
2	Kurichi Lake	0.570	0.546	4.21

CONCLUSION: The results were discussed for each characterization technique and found that hydrogel beads may be a source for the removal of aqueous biomedical waste from the contaminated water bodies. The synthesized hydrogel bead on reaction with different lake sources had shown a different range of adsorption. This diversion may be due to the selectivity of the materials that can be adsorbed onto the hydrogel bead. But it does not show any ill effects on the water sources as it contains only natural compounds. This work forms the preliminary concept of using hydrogel bead in vast water bodies to adsorb the biological wastes. So, the extension of this work can study the different metal adsorption and different biological systems adsorption onto the hydrogel bead. Since the combination of the various compounds as a nanocomposite, the hydrogel bead makes a better source for aqueous biomedical waste management.

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CONFLICTS OF INTEREST: Nil

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