Eco-Friendly Synthesis of CuO Nanoparticles by Using *Ulva Fasciata* Algae Extract for Antibacterial and Supercapacitor Application

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Abstracts: An eco-friendly, biogenic synthesis of copper oxide nanoparticles (CuO NPs) using *ulva fasciata* extract was reported. XRD, FTIR, UV-Vis spectroscopy, SEM, TEM, EDX, antibacterial activity and electrochemical analysis were used to characterize the biosynthesized CuO NPs. XRD spectrum reveals the monoclinic structure. The FTIR spectrum confirms the functional group of CuO NPs. The optical study gives the band gap values of CuO at different concentrations of *ulva fasciata algae* extract. The antibacterial activities were tested against one gram-positive bacteria *S. aureus* and one gram-negative bacteria *E. coli.* The CV curve exhibits a high specific capacitance of 132 F/g, at the scan rate of 10 mV/S.

Keywords: Green Synthesized CuO, Ulva Fasciata Algae Extrct, Antibacterial Activity and Electrochemical Analysis.Introduction

1.INTRODUCTION

The environmentally friendly synthesis of nanoparticles (NPs) is a challenging field in nanobiotechnology that is presented as a promising alternative to chemical pathways because it avoids the production of secondary contaminants that harm the environment [1-4]. The green synthesis of NPs engages reductant agents derived from bacteria, yeast, plants, algae, fungi, or plants, which contain secondary metabolites such as sugars, alginates, proteins, some amino acids, and other molecules used to reduce metals and lead the nucleation process [5-8]. These eco-friendly and cost-effective methods generate NPs with varying compositions, sizes, morphologies and dispersion, which may affect their final property and application. The optical, magnetic, catalytic, thermal and antibiotic activity of copper-based NPs is attracting attention due to their enhanced physicochemical properties due to their small surface-to-volume ratio when compared to their bulk material [9-10]. A few authors have investigated the use of raw aqueous algal extracts as reductant and capping sources for various metal oxide nanoparticles (Cdo, CuO, Co_3O_4, etc.). among them Copper oxide nanoparticles (CuO NPs) have been widely used in antimicrobials, gas sensors, supercapacitors and thermal conductivity appliances due to their superior stiffness, electrical conductivity, alloy strength, and ductility. In this, we prepared CuO NPs by using *Ulva fasciata algae* extract [11-13]. This *Ulva fasciata algae* extract is derived from ulva fasciata species and it is used widely in various industries as food, medicine, agriculture and etc. usually this *Ulva fasciata algae* is grown in the coastal areas [14-15]. By this process we collected *Ulva fasciata algae* from Rameswaram costal area and it's preserved it for further process.

2. EXPERIMENTAL PROCEDURE

2.1. Ulva Fasciata Extract Preparation

For this preparation, *Ulva fasciata algae* were collected from Rameswaram costal area (Tamil nadu, India). And the collected *Ulva fasciata algae* were washed with DI water until it reaches the value of pH 7. Then washed *Ulva fasciata algae* was dried oven at 60˚C for 12 hrs. After that, the dried *Ulva fasciata algae* were grinded by using agate mortar. For the extract preparation, 10g of dried *Ulva fasciata algae* were dissolved in 100 ml of DI water and stirred continuously for 1hr. after that homogeneous solution is heated at 100˚C for 15 min. then extract is filtered with whatman No. 1 filter paper. The filtered ulva fasciata extract is used for CuO NPs.

2.2 Preparation of CuO Nanoparticles

0.1 M of Copper nitrate (II) trihydrate is dissolved in 20 ml solvent of *Ulva fasciata algae* extract and stirred continuously for an hour. After that the homogeneous product was dried at 100˚C for 4 hrs. Then the dried product was collected and calcinated at 300˚C for 3 hrs. The same procedure is repeated for 30 ml and 40 ml of *Ulva fasciata algae* extract.

2.3 Electrode Preparation

The working electrode (CuO) is prepared by the following method. 80:10:10 ratio of CuO, PVDF and carbon black was taken and grind it smoothly, after that NMP solution was added with the well grind composite. The composite was grind until it reaches slurry form. The prepared slurry was coated on (1X1) cm nickel plate evenly. Then the coted plate was dried in oven at 100˚C for 6 hrs. The working electrode was analyzed by Biologic – SP 300 instrument. 1M KOH was used as an electrolyte solution.

3.RESULT AND DISCUSSION

3.1. XRD Analysis

The structural parameter of green synthesized CuO NPs was characterized by XRD analysis. Fig 1(a-c) shows the XRD pattern of CuO NPs prepared by using *Ulva fasciata algae* extract at different solvent ratios (20 ml, 30 ml and 40 ml). From the fig, eleven diffraction planes were observed at (32˚, 35˚, 38˚, 48˚, 53˚, 58˚, 61˚, 66˚, 68˚, 72˚ and 75˚) and the corresponding hkl planes are (110), (-111), (111), (-202), (020), (202), (-113), (-311), (220), (311) and (004) respectively [16-19]. The prepared CuO NPs shows the monoclinic structure all the diffraction planes are very well coincide with JCPDS card number 89-5895.

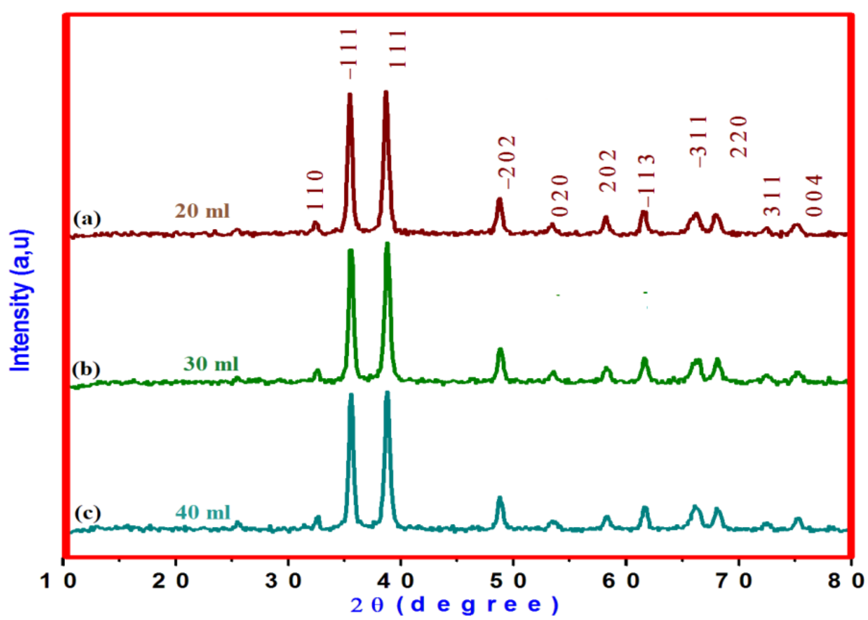

Figure 1: XRD pattern of CuO NPs (a) 20 ml of ulva fasciata extract, (b) 30 ml of ulva fasciata extract and (c) 40 ml of ulva fasciata extract.

The average crystal size of the prepared CuO NPs was calculated by Scherrer formula

$$D = \frac{k\lambda}{\beta cos\theta} \quad \ldots (1)$$

Where D is the average crystal size, k is a constant equal to 0.9, λ is the wave length of X-ray radiation (1.54060 Å), β is the full-width at half maximum (FWHM) of the peak (in radians) and 2θ is the Bragg angle (degree).

The other structural parameters of microstrain and dislocation density was calculated by the following equations,

$$\delta = \frac{1}{D^2} \quad \ldots (2)$$

Where δ is the dislocation density of the CuO NPs.

$$\varepsilon = \frac{B\cos\theta}{4} \quad \ldots (3)$$

Where $\boldsymbol{\varepsilon}$ is the microstrain of the CuO NPs.

The calculated structural parameters are tabulated in table 1.

Table 1: structural parameters of CuO nanoparticles

S.No.	Extract (ml)	Crystal size (nm)	Dislocation density (δ) 10^{15} (lines/m^2)	Microstrain X 10^{-3}	Lattice parameters (Å)		
					a	b	c
1	20	12	75.20	2.26	4.57	3.33	5.04
2	30	11	86.42	2.47	4.62	3.37	5.07
3	40	10	99.44	2.52	4.67	3.41	5.12

As observed from table 1, the crystallite size of CuO NPs decreases from 12 to 10 nm when the ratio of the solvent increases, whereas microstrain and dislocation density increases, Which is due to the inverse relationship with the crystalline size of the NPs.

3.2 FTIR Analysis

Functional group analysis of CuO NPs is carried out by FTIR technique. The analysis is carried out from 400 cm^{-1} to 4000 cm^{-1}. Fig 2 (a-c) shows the FTIR spectrum of CuO NPs. There are seven characteristic peaks were observed. The peaks 3421 cm^{-1} and 2921 cm^{-1} denote the O-H group present in the CuO NPs which were attributed to the presence of water or phenolic compounds from plants. The peak observed at 2840 cm^{-1} represents the C-H stretching vibration. The peak spotted at 1625 cm^{-1}, 1384 cm^{-1} and 1112 cm^{-1} are confirms the presence of C=C, CH_2 and C-O bonds respectively[20]. The characteristic peak CuO appeared at 531 cm^{-1} which evidently confirms the formation of CuO nanoparticles. According to the results of the FTIR analysis, *Ulva fasciata algae* phytochemicals as phenolics, tannins, and proteins were involved in the formation of CuO NPs.

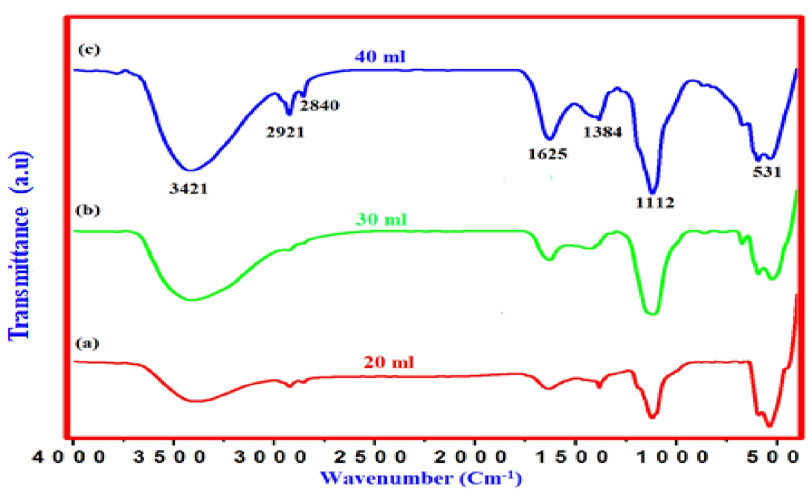

Figure 2: FTIR spectrum of CuO NPs (a) 20 ml of ulva fasciata extract, (b) 30 ml of Ulva fasciata algae extract and (c) 40 ml of Ulva fasciata algae extract

3.3 UV- Vis Analysis

Fig 3 (a-c) shows the UV vis spectrum of green synthesized CuO NPs. three absorption peaks are observed at 245 nm, 246 nm and 252 nm corresponds to 20 ml, 30 ml and 40 ml of CuO NPs using *Ulva fasciata algae* extract respectively. These peaks are appear due to the collective oscillation of free conduction band electrons excited by incident electromagnetic radiation causing surface plasmon absorption in CuO nanoparticles [22-24]. The tauc plot is used to identify the band gap energy of CuO nanoparticles. Fig 4 (a-c) shows the tauc UV spectrum of CuO nanoparticles prepared by 20 ml, 30 ml and 40 ml ulva fasciata leaf extract respectively.

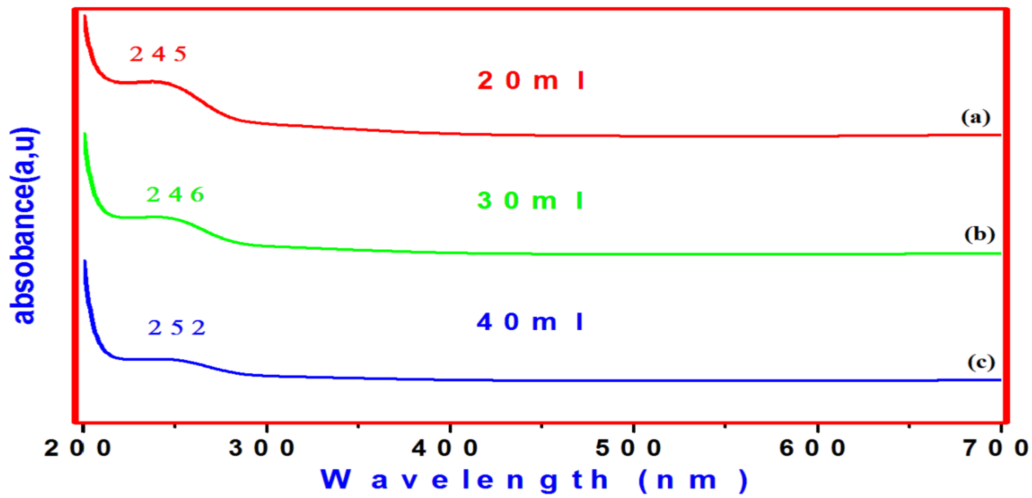

Figure 3: UV vis spectrum of CuO NPs (a) 20 ml of ulva fasciata extract, (b) 30 ml of ulva fasciata extract and (c) 40 ml of ulva fasciata extract

The following tauc equation is used find out the band gap of CuO nanoparticles.

Tauc's plot is used to calculate the energy band gap. You can find Tauc's equation using

$$\alpha h\nu = A\,(h\nu - Eg)\, n \qquad \ldots(4)$$

Where A is the constant, hv is the photon energy, is the absorption coefficient, and Eg is the energy bandgap. Depending on whether the transition from the valence band to the conduction band is direct or indirect, the value of n is either 1/2 or 2. If the transition is direct, the value is 1/2 and if it is indirect, it is 2. CuO has a straight band structure, therefore n is equal to 1/2. The equation then has the following form.

$$(\alpha h\nu)^{1\backslash 2} = B\,(h\nu - Eg) \qquad \ldots(5)$$

where B is a constant referring to the charge carriers' effective masses in relation to the valence and conduction bands.

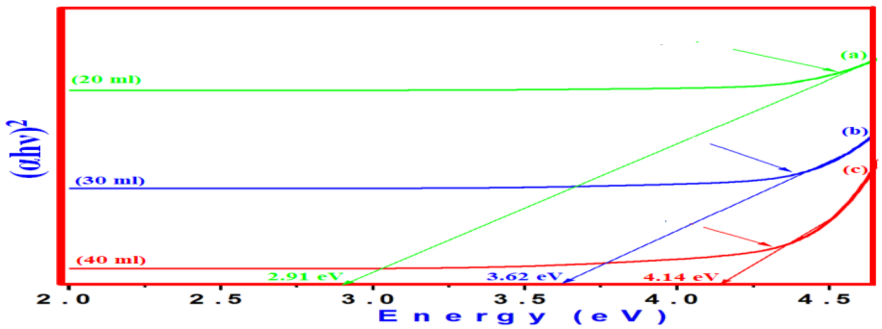

Figure 4: Tauc plot of CuO NPs (a) 20 ml of ulva fasciata extract, (b) 30 ml of ulva fasciata extract and (c) 40 ml of ulva fasciata extract

From the graph, the band gap values of 20 ml, 30 ml and 40ml ulva fasciata algae extract CuO NPs and 2.91 eV, 3.62 eV and 4.14 eV respectively.

3.4 Microstructural and Elemental composition analysis of CuO Nanoparticles

The microstructural and compositional analysis of bio synthesized CuO nanoparticles is carried out by SEM, HRTEM with EDX spectrum. Fig 5(a&b) shows the different magnifications SEM images of CuO nanoparticles. The SEM images clearly show that the CuO nanoparticles are closely packed and exhibit agglomerated nanosheets [25]. This wrinkled nature of the CuO nanosheets increases the surface area, which may promote more ion interaction between electrode and electrolyte in CV studies.

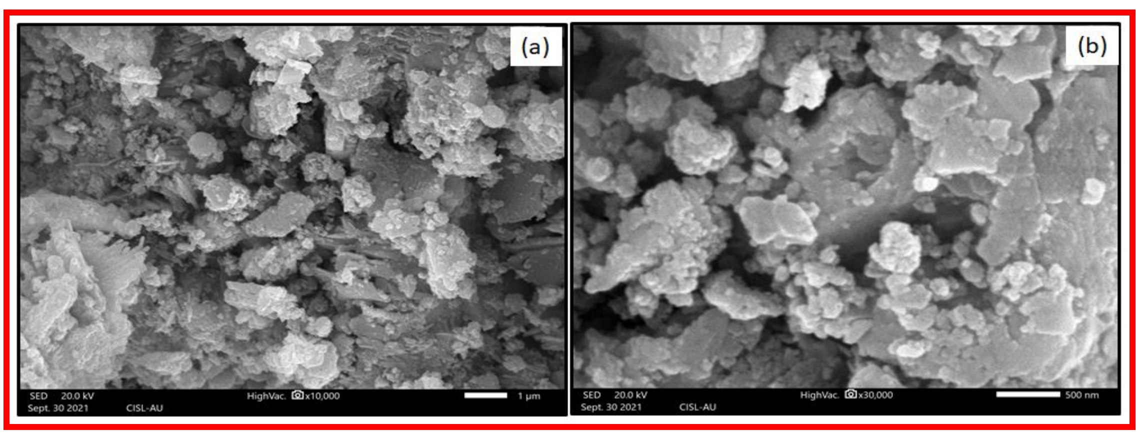

Figure 5: (a&b) SEM images of bio synthesized CuO nanoparticles

Fig. 6(a&b) shows the HRTEM images of green synthesized CuO NPs at different magnification. The images evidently confirm the SEM images. Fig. 6(c&d) shows the d spacing and SAED patterns of CuO NPs. the d spacing and hkl planes are very well matched with the XRD resultant values. Fig 7 shows EDX spectrum of CuO nanoparticles. The prominent components of Cu and O are majority present in the spectrum which confirms the purity of CuO nanoparticles. Other minor elements appeared due to the chemical composition of Ulva fasciata algae extract. Table 2 shows the elemental composition of green synthesized CuO nanoparticles.

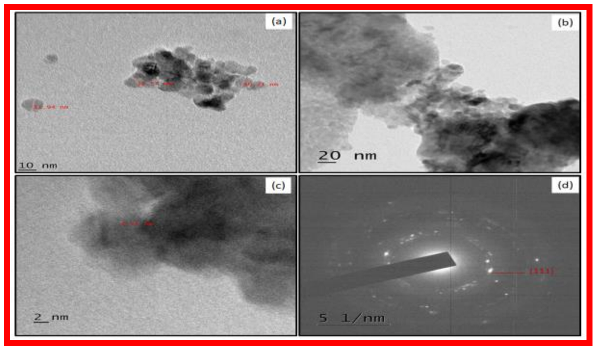

Figure 6: (a&b) HRTEM images of CuO NPs. (c) d spacing fringes and (d) SAED pattern CuO nanoparticles

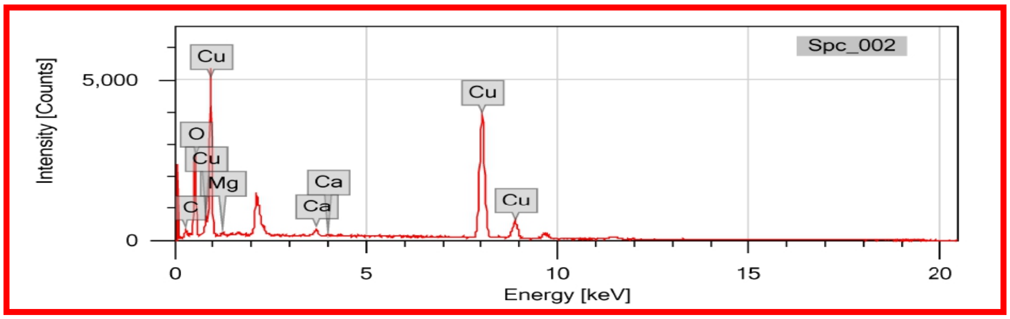

Figure 7: EDX spectrum of CuO nanoparticles

Table 2: Elemental composition of CuO nanoparticles

Element	Line	Mass%	Atom%
C	K	3.96±0.06	13.76±0.22
O	K	11.37±0.10	29.64±0.26
Mg	K	0.64±0.05	1.10±0.08
Ca	K	0.90±0.03	0.93±0.04
Cu	K	83.13±0.45	54.56±0.30
Total		100.00	100.00

3.5 Antibacterial Activity Of Cuo Nanoparticles

The antibacterial activity of green synthesized CuO NPs was tested against one gram positive bacteria *S. aureus* and one gram negative bacteria *E. coli* by disc diffusion method. Fig. 8 shows the antibacterial activity of green synthesized CuO NPs against (a) *S. aureus* and (b) *E. coli* at two different concentrations (50 µl and 100 µl). The resultant parameters are shown in table 3. The appearance of an inhibition zone around the wells in all of the petri plates clearly demonstrated the antibacterial potential of biosynthesized CuO NPs. CuO nanoparticles have antibacterial activity against both gram positive and gram negative test strains. From the result CuO NPs had a strong bactericidal effect on gram negative bacteria (*E.coli*). The antibacterial activity mechanism is based on the penetration of CuO NPs through the bacterial cell membrane, which causes membrane damage and cell death. CuO NPs interacted with the bacterial membrane due to their nanosize and high surface-to-volume ratio. In this study, the inhibition effect of CuO NPs on gram negative bacteria was found to be greater than that on gram positive bacteria, which could be attributed to differences in bacterial membrane structures [26-29]. As a result, CuO NPs synthesised from uluva algae extract have been found to be an effective antibacterial agent against gram negative (E. coli) bacteria.

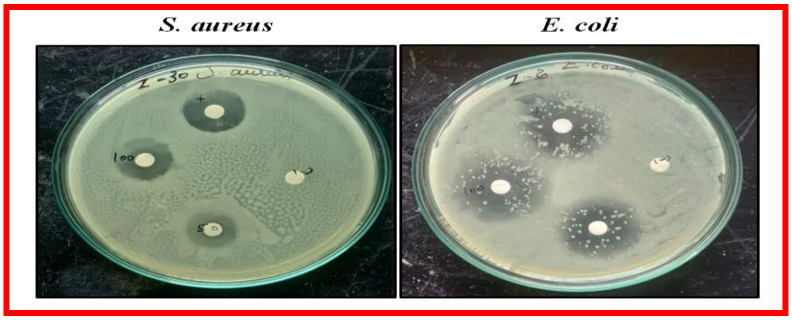

Figure 8: shows the antibacterial activity of green synthesized CuO NPs against (a) *S. aureus* and (b) *E. coli*

Table 3: Antibacterial activity result of CuO nanoparticles

Test bacterial pathogens	Zone of inhibition (mm)	
	50 µg	100 µg
S. aureus	7.08±0.58	8.01±0.18
E. coli	12.08±0.18	13.42±0.54

3.6 Electrochemical Performance of Cuo Nanoparticles

The electrochemical performance of CuO nanoparticle is carried out by cyclic voltammetry and galvanostatic charge/discharge analysis. The fabricated CuO electrodes were subjected to 1M KOH electrolyte solution and characterized by cyclic voltammograms. Fig.9 represents the CV curve of Cuo NPs at different scan rate (10 mV/s, 25 mV/s, 50 mV/s, 75 mV/s and 100 mV/s). The CV curves of both of these electrodes show the oxidation and reduction peaks and it displays pseudocapacitive nature.

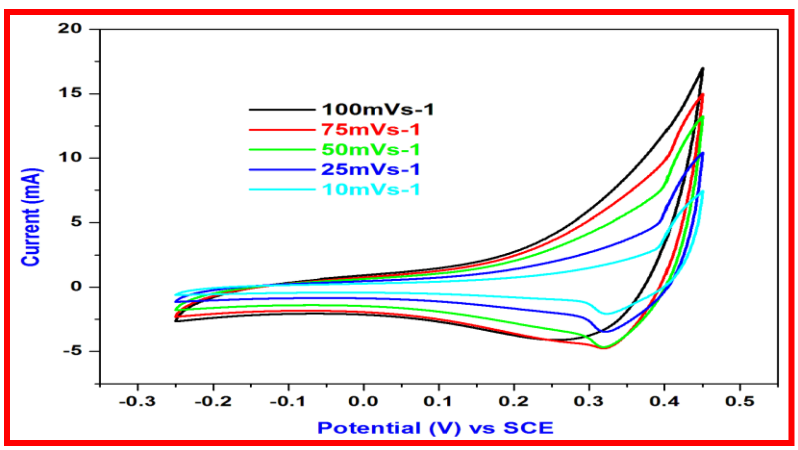

Figure 9: CV cure of CuO nanoparticles

The specific capacitance CuO electrodes can be calculated by using the equation

$$C_p = \frac{A}{m\Delta V} \quad \ldots (6)$$

Where C_p, A, m and ΔV represent the cathodic and anodic charge on each scan rate, the area under the CV curve, the mass of the active material (mg) and the scan rate in mV/s. Using the above equation, the C_p values of the CuO electrode are calculated as 132 F/g, 95 F/g, 70 F/g, 56 F/g and 43 F/g for the scan rate of 10, 25, 50, 75 and 100 mV/s, respectively. At the lower scan rate the specific capacitance of CuO exhibit high, which indicates more ion interaction between electrode and electrolyte. At higher scan rate the interaction between electrode and electrolyte is very less so it exhibits low specific capacitance [30]. The GCD analysis also confirms the specific capacitance of green synthesized CuO NPs. Fig. 10 represents the GCDs curves of CuO electrodes measured at the current density of 1 A/g and 2 A/g with the potential window varied from -0.25 V to 0.45 V.

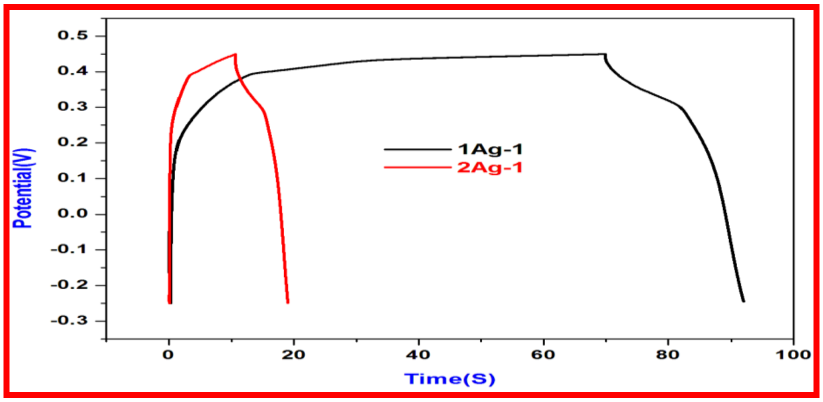

Figure 10: GCD cure of CuO nanoparticles

The C_p of CuO electrodes using GCDs can be calculated from the following equation

$$C_{p\ (GCD)} = \frac{i\Delta t}{m\Delta V} \quad \ldots (7)$$

Where i is the current density (A/g), Δt is the discharging time (s), m is the mass of the active material (mg) and ΔV is the potential window (volts). The calculated C_p values of CuO electrode are 28 F/g and 20F/g for current density of 1 A/g and 2 A/g respectively. As the current density increases, the specific capacitance decreases due to increase in ohmic drop during the charge/discharge process.

CONCLUSION

The green synthesized CuO NPs were prepared by using *Ulva fasciata algae* extract. The XRD spectrum confirms the monoclinic structure of CuO nanoparticles and the diffraction planes are very well coincide with JCPDS card number 89-5895. The FTIR spectrum confirms the functional group present in CuO NPs. The band gap values of CuO NPs is increased when the crystallite size of the NPs are decreased. The green synthesized CuO NPs was act effective antibacterial agent against gram negative (E. coli) bacteria. From the electrochemical analysis result, the CuO NPs is highly suitable for supercapacitor application.

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