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BIOGENIC SYNTHESIS OF COPPER NANOPARTICLES FROM BLUE GREEN ALGA – ARTHROSPIRA MAXIMA AND ITS CHARACTERIZATION

Savitha. M. Murthy*, Shravya , S and Shalini, B.U.

*__Corresponding Author__: Dr. Savitha M Murthy, Associate Professor, Department of Botany, Mount Carmel College, Autonomous, Bengaluru, Karnataka

e-mail: dr.savithamurthy@mccblr.edu.in

ABSTRACT

The nanoparticles which are synthesized in metallic form are very important in the present scenario because of its properties and its uses in various industries. In the present study, the cyanobacteria, species was used to synthesize the copper oxide nanoparticles by green method. The nanoparticles were synthesized from both wet and dry extracts of naturally grown cultures. The nanoparticles synthesized were characterized by UV – Visible absorption spectroscopy, Energy Dispersive X-ray (EDAX) and Scanning Electron Microscopy (SEM) studies. From the EDAX studies it was found that the elemental compounds present in the synthesized nanoparticles were copper and oxygen about 6.07% and 40.53% and 27.255 and 47.57% for wet and dry samples respectively. The size of the nanoparticles was observed from SEM analysis and it ranges from 82 nm to 106nm and 115nm to 125nm for wet and dry algal samples respectively. From the present study it was concluded that the nanoparticles synthesized using dry extract performed better compared to the wet extract.

Key words: *Arthrospira*, Biogenic synthesis, Copper oxide, Elements, Nanoparticles.

Nanotechnology is a most developing area of science which deals with the synthesis of nanoparticles which show different sizes and shapes and finds use in various applications. The size of the nanoparticles vary in size and the diameter between 1 to 100nm. The nanoparticles which could be categorized under three groups *viz* natural, incidental and engineered nanoparticles.

The synthesis of nanoparticles is the principal branch in nanoscience and technology. The nanoparticles can be synthesized chemically or biologically using natural extracts from plants, bacteria, fungi and algae. These NPs can be in the form of metals, metal oxides, silicates, etc (*Sunil et al., 2017*). The nanoparticles are environmental friendly and using green method for its synthesis, which has wide uses in diverse fields (Awwadl et al., 2012 and Ramgir et al., 2013). The nanoparticles which are synthesized in metallic form are receiving cumulative importance in a various fields. Metallic nanoparticles have its own uniqueness in its optical, electronic and magnetic properties (Abboud et al., 2014). They are good semi conductors in solar energy transformation, electronics, etc (Ramgir et al., 2013; Jani et al., 2013 and Shalana et al., 2013).

Nanoparticles have unique physical, chemical, thermal, biological properties and for this reason this field has gained importance in the present technological world (D*aniel and Astruc, 2004*).

Two diverse types of nanoparticles are present , which are inorganic and organic. The inorganic nanoparticles include metal and metal oxides (*Oscar et al., 2016*), which are potent antibacterial agents (*Loomba and Scarabelli, 2013*).

Copper nanoparticles synthesized from natural source are used as antimicrobial agents and have shown a good antibacterial activity by membrane disruption and Radical Scavenging species production (Huh and Kwon, 2011).

Algae are predominantly aquatic organisms which includes both micro and macro forms, which are important both economically and ecologically (Oscar et al., 2016). They have a vital role to play in pharmaceutical, agriculture, cosmetic industries. They are also a good source of biofuels and dyes. Algae have the capacity of up taking metals and also when the biological method is used; it will be economically challenging *(Bilal et al, 2018)*. As the algae are rich in having reducing agents as they have the capacity to reduce the metal salts to their respective metallic nanoparticles without liberating any harmful substances as by products *(Arya et al, 2018)*.

Hence, Algae can be called as "bionanofactories" because the nanoparticles can be synthesized from both the living as well as dead biomass (*Arya et al, 2018*).

The synthesis of nanoparticles from algae can be followed in different methods like algal extract preparation, molar solution of desired ionic metallic compound preparation and incubation of algal and ionic molar solution by vigorous shaking for certain time under controlled conditions (*Thakkar et al., 2010 and Rauwel et al., 2015).* The synthesis of nanoparticle depends on the type of algal species used (*Vincy et al., 2017*). *Spirulina platensis* has been utilized for the extra cellular synthesis of gold, silver nanoparticles (*Chakraborty et al., 2009*).

In the present study, the *Arthrospira maxima* (= *Spirulina maxima)* were selected for the green synthesis of copper nanoparticles and its characterization through different analytical techniques.

MATERIALS AND METHODS

Cyanobacteria, *Arthrospira maxima* were procured from E2E Biotech Pvt Ltd., Bengaluru, Karnataka, India.

Isolation and Culturing of Cyanobacteria:

The pure cultures of *Arthrospira maxima* were multiplied on CFTRI medium, using 1.5% Agar-agar as a solidifying agent (Venkataraman, 1985). The algae grown was maintained in the culture room at 27 ± 1°C, 1.2 ± 0.2 Lux light intensity and 16: 8 hrs photoperiod, following all the aseptic conditions. Periodic culturing was maintained following the standard aseptic conditions and the algal biomass was used for further experimental work.

Algal Extract Preparation:

The algal extracts were prepared using wet and dry extracts of *Arthrospira maxima*. The wet algal extract was prepared by homogenization of the pure algal cells. 2ml of the algal culture was taken and homogenized with the help of glass beads. Then, the extract was used to synthesize the copper nanoparticles.

Dry algal extract was prepared by collecting the algal sample in a petridish and oven dried at 60°C overnight. The dried algae were scrapped with the help of scalpel and powdered in a mortar and pestle to get fine powder. 2g of the algal powder were dissolved in 20ml of distilled water and was centrifuged at 1800 rpm for 20 minutes. From the obtained supernatant, 2 ml was used for the green synthesis of copper nanoparticles (*Abbouad et al, 2014*).

Synthesis of Copper Oxide Nanoparticles:

2ml of algal extract was added drop wise to 20ml of 1mM aqueous copper acetate solution with constant stirring at 100 - 120°C on a shaker for few hours. Within few hours, the blue color changes to brown and slowly to brick red color indicative of the synthesized CuO nanoparticles (Abbouad et al., 2014).

Characterization of Copper Oxide Nanoparticles:

The synthesized copper nanoparticles were characterized by UV-Visible spectroscopy, SEM and EDAX studies.

The reduction of copper ion in the solution was observed using Shimadzu UV-1700 double beam UV-Visible spectrophotometer. The spectral range from 200nm to 1100nm were recorded with a time interval of 5 minutes for all the samples against distilled water as blank at the resolution of 5nm *(Mahdieh et al, 2012)*.

SEM: A scanning electronic microscope (TESCAN – VEGA 3) was used to record the micrograph images of synthesized copper nanoparticles to know the size and surface morphology of NPS *(Biradar et al, 2012)*.

EDAX: Energy Dispersive X-Ray analysis was done using X1 Analyzer AMETEX to know the chemical composition and its percentage presence in the sample.

RESULTS AND DISCUSSION

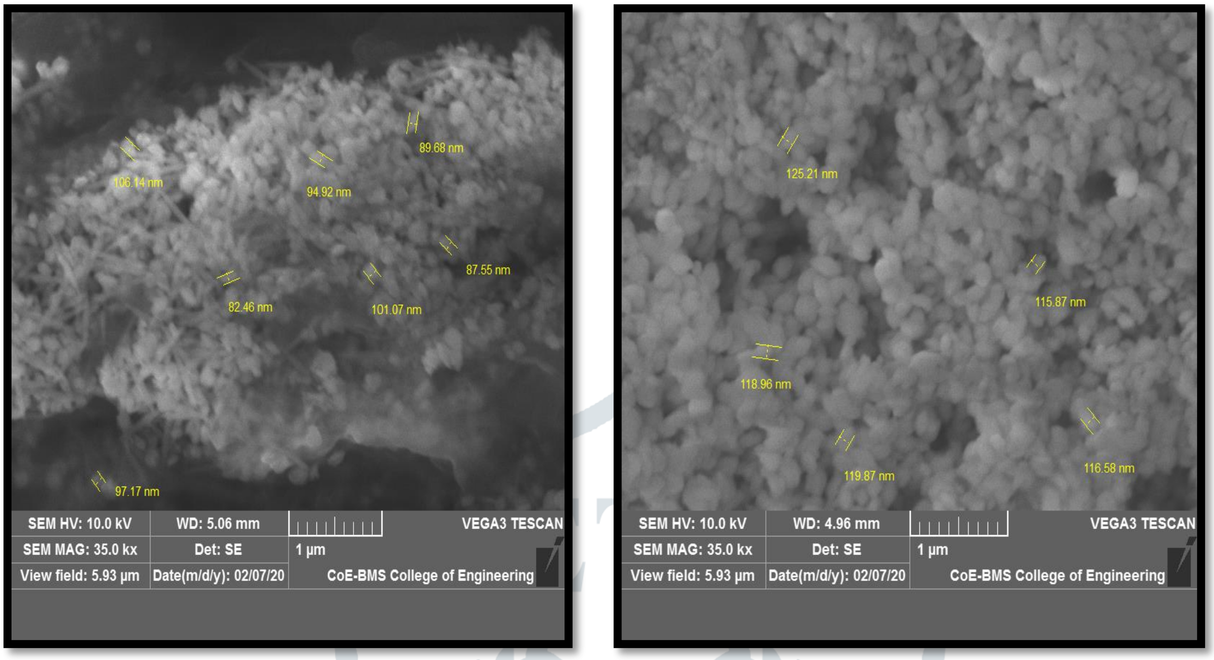
The green synthesis of copper nanoparticles was initially confirmed by changing in color from blue to brown and to brick red. Later, was characterized by using UV-Visible spectrophotometer. The change in the color of the reaction mixture (**Figure 1**), indicates the formation of copper NPs *(Abboud et al, 2014)*.

UV – Visible Spectroscopy: The copper nanoparticles synthesized by *Spirulina* species and commercial powder *Spirulina* exhibited the absorption peaks ranging from 340nm and 1083nm respectively, when observed under UV-Visible spectroscopy. The synthesized copper nanoparticles from wet sample show absorbance at 2 points 750 nm and 290 nm. Similarly, the dry algal sample show absorbance peaks at 720 nm and 275nm. There is not much difference in the 1st peak between 2 extracts. Formation of CuO nanoparticles showed absorbance peak at 260nm (*Borgohain et al., 2002*) and also as reported in the brown alga *Bifurcata*, the formation of copper nanoparticles between 245 and 290nm (Abboud et al., 2014)].

UV-Visible spectrum of the copper NPs formed and the transition of color change in the solution mixture from blue to brick red indicates the biotransformation of ionic copper to reduced copper, which can be attributed.

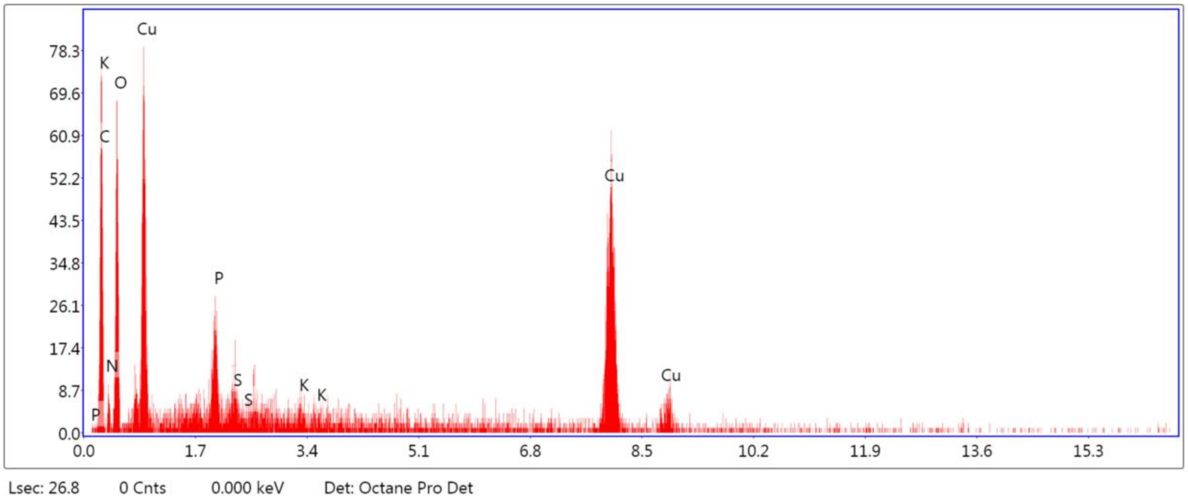
Scanning Electron Microscopy (SEM): Green synthesis of copper nanoparticles from the samples were studied for their shape and size with the help of SEM (**Figure 2**) in the view field of 5.93µm and 1 µm resolution. The diameter of NPs in the wet algal sample were around 82nm to 106nm. For the dry algal sample, the diameter was 115nm to 125nm. By observing the images of surface morphology of the nanoparticles in the three samples, it was clear that the nanoparticles surface looked like crystalline form. The larger sized nanoparticles were seen in the dry algal extract compared to the wet algal extract.

Fig 2: SEM image of wet and dry samples showing the size of CuO Nanoparticles



EDAX: Estimation of EDAX confirmed the presence of copper oxide nanoparticles in all the three algal extracts (**Figure 3**). The extracts contained elements like C, N, O, Si, P, S, K, Cu and the atomic percentage of elements present in the extracts was also detected by this analysis. The wet algal extract contained around 6.07% of Cu atoms and 40.53% of O atoms and the dry extract had 27.25% of Cu and 47.57% O. In contrast to this the commercial extract contained around 6.23% Cu and 29.92% of O. The dry algal extract had more Cu compared to the other two extracts.

Fig. 3: Chemical composition of chemicals in nanoparticles



Over all, in the present study, the dry algal extract can be selected to synthesize the copper oxide nanoparticles and those NPs can be used for various applications.

CONCLUSION

The present work is a good environment friendly method to synthesize the copper nanoparticles from the blue green alga *Arthrospira maxima*. The aqueous extract of the algal sample confirms the reduction of copper ions to copper nanoparticles. The synthesized NPs were also characterized by analytical techniques. This is the preliminary investigation on the green synthesis and characterization of copper oxide nanoparticles. These synthesized nanoparticles will be used in various industries.

Declaration: The authors declare that there is no conflict of interests regarding the publication of paper in this journal.

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