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GREEN SYNTHESIS OF SILVER NANOPARTICLES FROM EXTRACT OF *PADINA TETRASTROMATICA* LEAF

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The synthesis, characterization and application of biologically synthesized nanomaterials are an important aspect in nanotechnology. The present study deals with the synthesis of silver nanoparticles (Ag-NPs) using the aqueous extract of brown seaweed *Padina tetrastromatica* leaf extract. The complete reduction of silver ions was observed after 72 h of reaction at 300 ºC under shaking conditions. The colour changes in reaction mixture (pale yellow to dark brown) was observed during the incubation period, because of the formation of silver nanoparticles in the reaction mixture enables to produce particular colour due to their specific properties. The formation of Ag-NPs was confirmed by UV-Visible Spectroscopy, X-Ray Diffraction (XRD) pattern, Scanning Electron Microscopy (SEM) and Transmission Electron Microscopy (TEM) with Energy dispersive X-ray (EDX) patterns. The synthesized Ag-NPs were predominately spherical in shape and polydispersed. Fourier Transform Infra-Red (FT-IR) spectroscopy analysis showed that the synthesized nano-Ag was capped with bimolecular compounds which are responsible for reduction of silver ions. The approach of plant-mediated synthesis appears to be cost efficient, eco-friendly and easy alternative to conventional methods of silver nanoparticles synthesis.

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Keywords: Bioreduction, Silver nanoparticles, *Padina tetrastromatica,* UV-Vis, XRD,SEM, TEM and FT-IR.

1. Introduction

Generally, metal nanoparticles are synthesized and stabilized through chemical and mechanical methods [1, 2], electrochemical techniques [3], photochemical reactions in reverse micelles [4] and nowadays via green chemistry method [5]. Synthesis of nanoparticles through biological method is a good, environment friendly and economically alternative method. Synthesis of green nanomaterials and their characterization is an emerging field of nanotechnology from the past few decades, because of their applications in the fields of physics, chemistry, biology and medicine.

Application of green chemistry to the synthesis of nanomaterials has vital importance in medicinal and technological aspects [6, 7]. Biologically synthesized silver nanoparticles (Ag-NPs) have wide range of applications because of their remarkable physical and chemical properties. The literature on the extra cellular biosynthesis of Ag-NPs using plants and pure compounds from plants are insignificant [8-10]. Specifically, while there is relatively little or no literature on the extra cellular synthesis of Ag-NPs by using seaweeds [11].

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In this article, we describe a simple onestep method for the synthesis of Ag-NPs by the reduction of aqueous Ag-ions using extracts of brown sea weed, at direct sunlight conditions. The amount of nanoparticles synthesized and qualitative differences between synthesized nanoparticles were also investigated by various analytical methods.

Padina tetrastromatica (Forsskal) (Class: Phaeophyceae, Order: Dictyotales, Family: Dictyotaceae, Genus: *Padina*, Species: *P. tetrastromatica*) is a striped, yellowish brown, fan shaped algae, which becomes olive green on drying and proliferates in tropical waters; its thalli are irregularly cleft into narrow lobes, having involuted apical margin. *Padina tetrastromatica* is abundantly growing seaweed in Coastal areas of south India. It occurs in inter tidal region of Gulf of Mannar, Southeast coast of India. It grows in shallow sand covered rocky pools of mid- and lower littoral zones [12]. There are about 50 taxa of *Padina* distributed worldwide, although most are poorly known and many would prove to be synonymous, according to Guiry and Dhonncha [13], only 30 of these are currently accepted.

2. Experimental details

2.1. Screening and selection of plants

Fresh plants of *Padina tetrastromatica* (fig 1) brown seaweed was collected from Mandapam coastal region (78°8'E, 9°17'N), in Gulf of Mannar, Tamilnadu, South India. Samples were brought to laboratory in polythene bags and cleaned thoroughly with fresh water to remove adhering debris and associated biota. The algae were cleaned using brush for the removal of the epiphytes with distilled water. After cleaning, the algae were dried in shade at room temperature for a week.

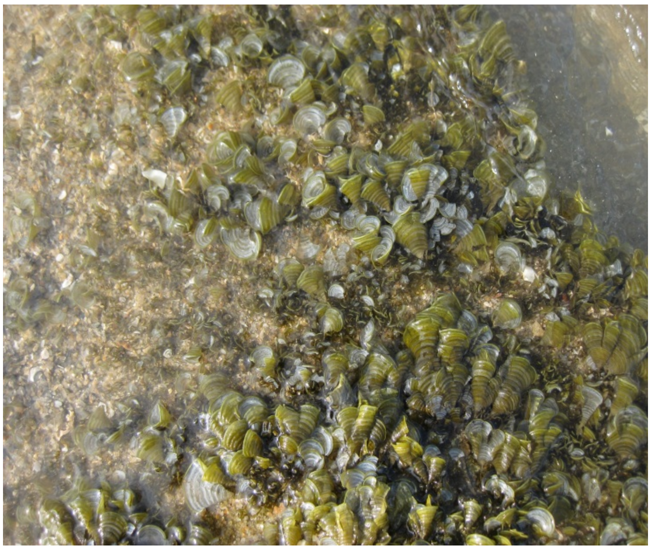


Fig 1. Padina tetrastromatica

2.2. Preparation of aqueous extract

The whole plant of *Padina tetrastromatica* were initially rinsed thrice in distilled water and dried on paper toweling, and samples (25 *g*) were cut into fine pieces and boiled with 100 ml of sterile distilled water for 5 *min*. The crude extract was passed through Whatman No.1 filter paper and the filtrate was stored at 4*°C* for further use.

2.3. Synthesis of silver nanoparticles

Silver nitrate ($AgNO_3$) was of analytical grade (AR) and purchased from E. Merck (India). In the typical synthesis of silver nanoparticles, 10 *ml* of the aqueous extract of *Padina tetrastromatica* was added to 90 *ml* of 1 *mM* aqueous $AgNO_3$ solution in 250 *ml* conical flask and kept at room temperature for 72 *h* at 120 *rpm*. Suitable controls were maintained throughout the experiments.

2.4. UV-visible spectroscopy analysis

The colour change in reaction mixture (metal ion solution + seaweed extract) was recorded through visual observation. The bio reduction of silver ions in aqueous solution was monitored by periodic sampling of aliquots (0.5 *ml*) and subsequently measuring UV-Vis spectra of the solution. UV-vis spectra of these aliquots were monitored as a function of time of reaction on UV-Vis spectrophotometer UV-2450 (Shimadzu).

2.5. XRD measurement

The Ag-NPs solution thus obtained was purified by repeated centrifugation at 5000 *rpm* for 20 *min* followed by redispersion of the pellet of Ag-NPs in 10 *ml* of deionized water. After freeze drying of the purified Ag-NPs, the structure and composition were analyzed by XRD. The dried mixture of Ag-NPs was collected for the determination of the formation of Ag-NPs by an X'Pert Pro x-ray diffractometer (PAN analyticalBV, The Netherlands) operated at a voltage of 40 *kV* and a current of 30 *mA* with Cu Kα radiation in a θ- 2 θ configuration. The crystallite domain size was calculated from the width of the XRD peaks, assuming that they are free from non-uniform strains, using the Scherrer's formula.

$$D= 0.94 \lambda / \beta \cos \theta$$

where D is the average crystallite domain size perpendicular to the reflecting planes, λ is the X-ray wavelength, β is the full width at half maximum (FWHM), and θ is the diffraction angle. To eliminate additional instrumental broadening the FWHM was corrected, using the FWHM from a large grained Si sample.

$$\beta \text{ corrected} = (\text{FWHM}^2 \text{sample- FWHM}^2 \text{si})^{1/2}$$

This modified formula is valid only when the crystallite size is smaller than 100 *nm* [14].

2.6. SEM analysis of silver nanoparticles

Scanning Electron Microscopic (SEM) analysis was done using Hitachi S-4500 SEM machine. Thin films of the sample were prepared on a carbon coated copper grid by just dropping a very small amount of the sample on the grid, extra solution was removed using a blotting paper and then the film on the SEM grid were allowed to dry by putting it under a mercury lamp for 5 *min*.

2.7. TEM analysis of silver nanoparticles

The structural characterization of the silver nanoparticles was carried out by Transmission Electron Microscopy (TEM) using JEOL JEM 2100 high resolution transmission electron microscope. The sample was prepared by air-drying drops of diluted solutions of the preparations on carbon films supported by copper grids.

2.8. FTIR measurement

To remove any free biomass residue or compound that is not the capping ligand of the nanoparticles, the residual solution of 100 *ml* after reaction was centrifuged at 5000 *rpm* for 10 *min*. The supernatant was again centrifuged at 10000 *rpm* for 60 *min* and the pellet was obtained. This is followed by redispersion of the pellet of Ag-NPs into 1 *ml* of deionized water. Thereafter, the purified suspension was freeze dried to obtain dried powder. Finally, the dried nanoparticles were analyzed by FTIR Nicolet Avatar 660 (Nicolet, USA).

2.9. EDX observation of silver nanoparticles

Energy-dispersive X-ray (EDX) analysis was carried using JEOL JEM 2100 high resolution transmission electron microscope to confirm the presence of silver in the particles as well as to detect other elementary compositions of the particles.

3. Results and discussion

It is well known that Ag-NPs exhibit reddish-brown in water [15]. The formation of Ag-NPs by reduction of the aqueous Ag+ during exposure to the aqueous extract of *Padina tetrastromatica* showed reddish-brown colour (fig 2), which suggested the formation of Ag-NPs in solution. The colour arises due to excitation of surface plasmon vibrations in the silver metal nanoparticles [16].

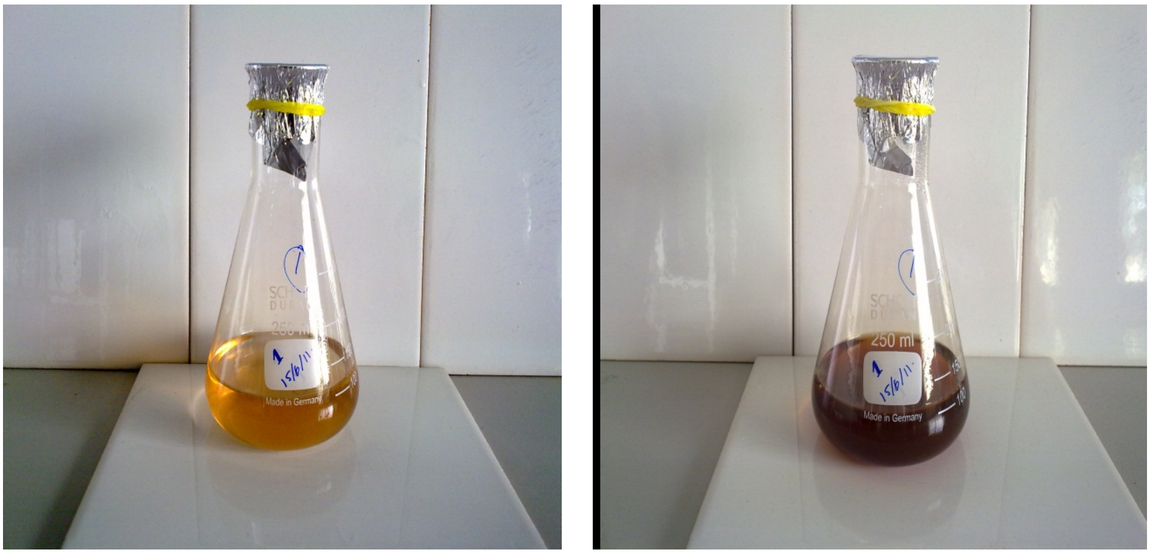
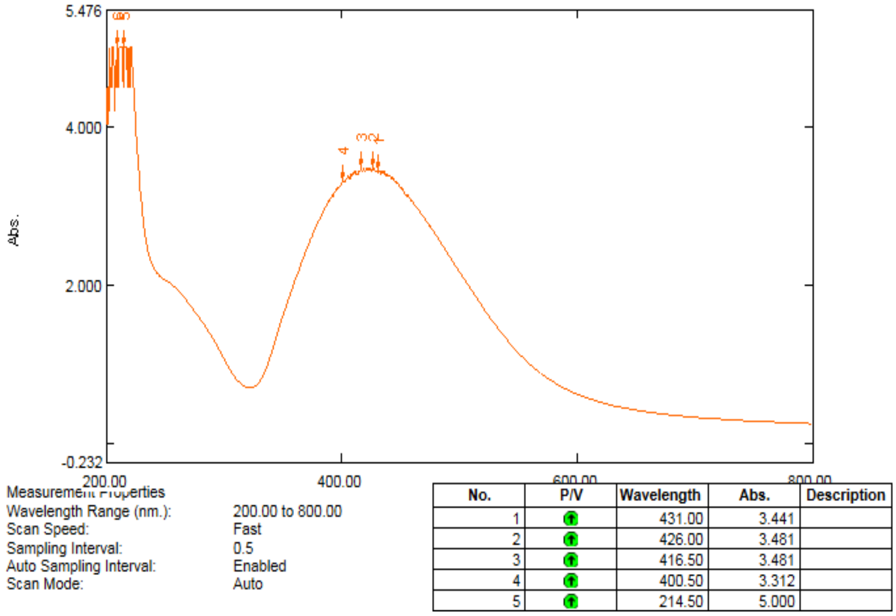


Fig 2. Change of color from pale yellow to brown by the addition of silver nitrate

The reduction of silver was subjected to analysis by using the UV-Vis Spectrophotometer. Absorption spectra of Ag-NPs formed in the reaction media had absorbance peak at 426 *nm*, broadening of peak indicated that the particles are polydispersed (fig 3). The frequency and width of the surface plasmon absorption depends on the size and shape of the metal nanoparticles as well as on the dielectric constant of the metal itself and the surrounding medium [17-19].



Measurement Properties
Wavelength Range (nm.): 200.00 to 800.00
Scan Speed: Fast
Sampling Interval: 0.5
Auto Sampling Interval: Enabled
Scan Mode: Auto

No.	P/V	Wavelength	Abs.	Description
1	↑	431.00	3.441	
2	↑	426.00	3.481	
3	↑	416.50	3.481	
4	↑	400.50	3.312	
5	↑	214.50	5.000	

Fig 3. UV-Vis absorption spectra of silver nanoparticles synthesized from Padina tetrastromatica extract by treating 1mM silver nitrate after 72 hr

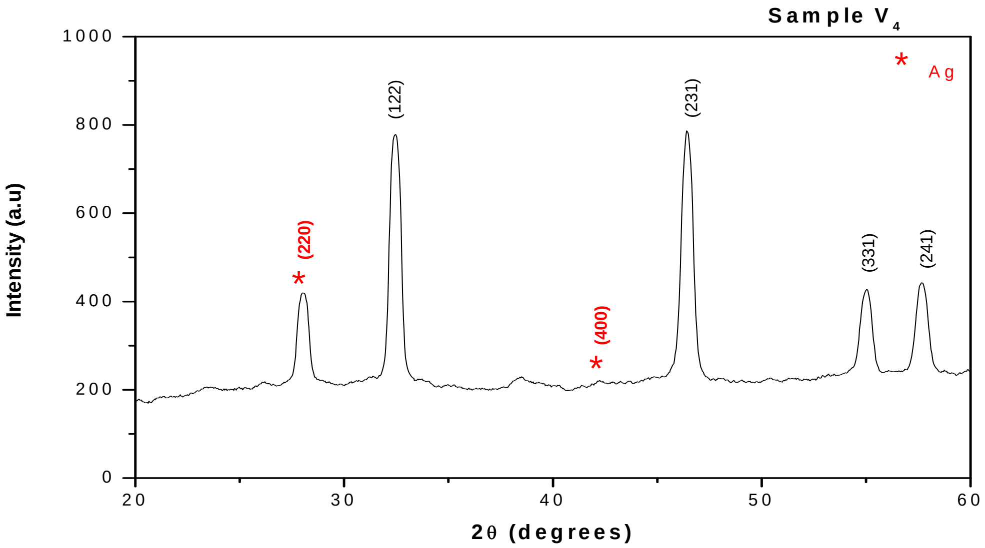


Fig 4. XRD patterns of capped silver nanoparticles synthesized by treating Padina tetrastromatica extract with 1 mM silver nitrate

The biosynthesised silver nanostructure by using *Padina tetrastromatica* extract was further demonstrated and confirmed by the characteristic peaks observed in the XRD image at $\theta = 28.09^{o}$, marked with (220). A number of Bragg reflections corresponding to the (220) sets of lattice planes were observed which may be indexed based on the face-centred crystal structure of silver. The XRD pattern thus clearly showed that the Ag-NPs are crystalline in nature (Figure 4).

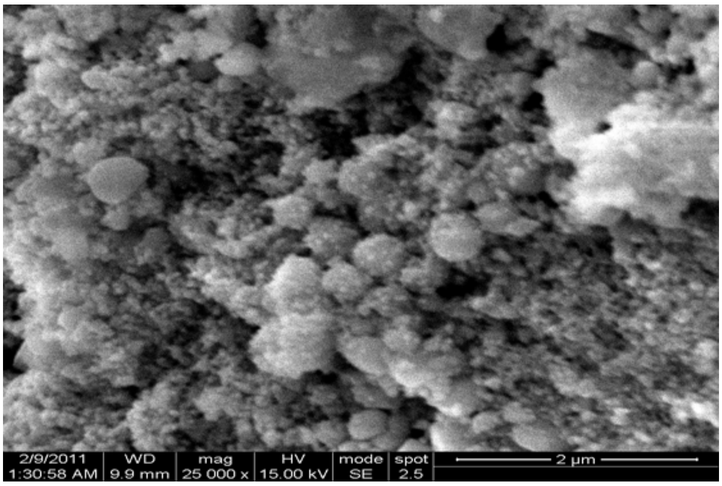
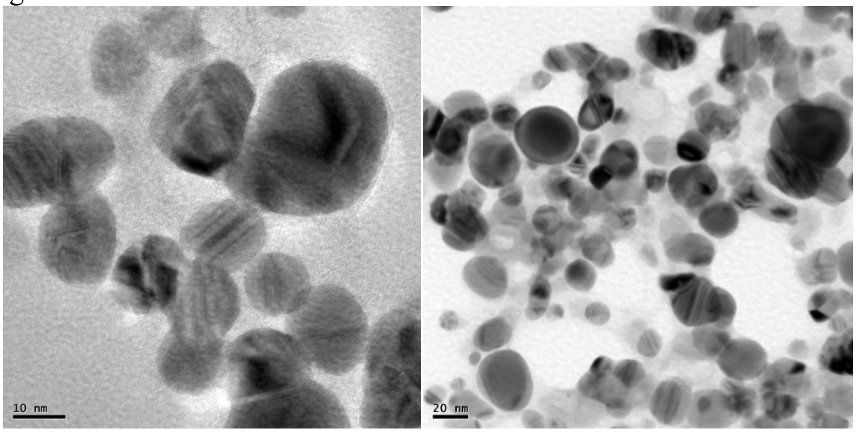


Fig 5. SEM micrograph of silver nanoparticles synthesised by the reaction of 1 mM silver nitrate with Padina tetrastromatica extract

The SEM image (Figure 5) showing the high density Ag-NPs synthesized by the *Padina tetrastromatica* further confirmed the development of silver nanostructures. The SEM micrographs of nanoparticle obtained in the filtrate showed that Ag-NPs are spherical shaped, well distributed without aggregation in solution.



(a) ***(b)***

Fig 6. TEM (a and b) micrograph of silver nanoparticles synthesised by the reaction of 1 mM silver nitrate with Padina tetrastromatica extract

The silver nanoparticles synthesized by *Padina tetrastromatica* extract were scanned using TEM from which the average mean size of the silver nanoparticles was 20 *nm* and seems to be spherical in morphology as shown in figure 6.

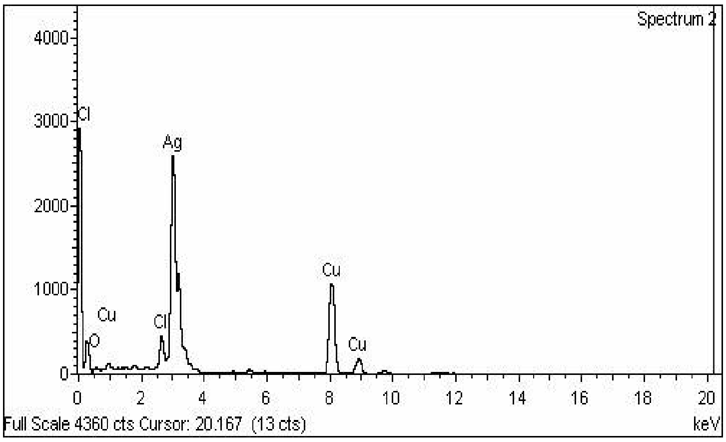


Fig 7. EDX spectrum recorded showing sharp peak between 2.7and 4 keV confirming the presence of silver.

Analysis through Energy dispersive X-ray (EDX) spectrometers confirmed the presence of elemental silver signal of the silver nanoparticles (Fig. 7). The vertical axis displays the number of x-ray counts whilst the horizontal axis displays energy in KeV. Identification lines for the major emission energies for silver (Ag) are displayed and these correspond with peaks in the spectrum, thus giving confidence that silver has been correctly identified.

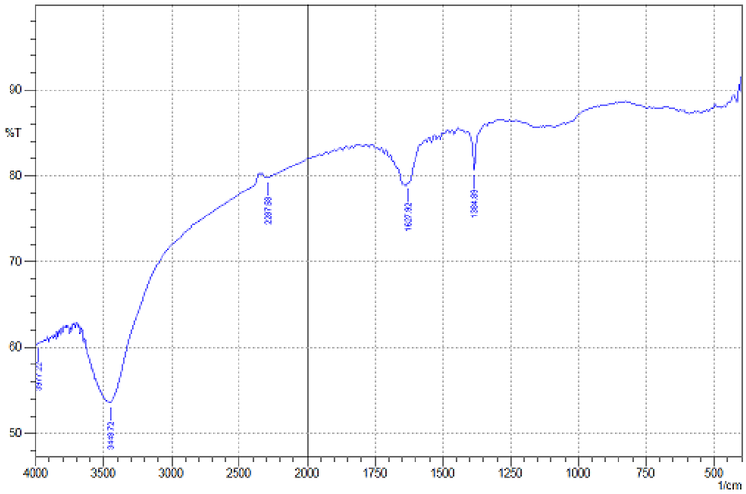


Fig 8. FTIR spectra of silver nanoparticles synthesized by the reduction of 1mM silver nitrate with the Padina tetrastromatica extract

FTIR analysis was used for the characterization of the extract and the resulting nanoparticles. Absorbance bands seen at 3448,1627 and 1384 *cm*$^{-1}$ were assigned to the NH stretching vibrations of free NH group, C=C Stretching vibrations of non conjugated group and CH bending vibrations of CH_3 group respectively (Fig 7). The results revealed that the capping ligand of the Ag-NPs may be an aromatic compound or alkanes or amines.

4. Conclusion

In this paper we have successfully synthesized silver nanoparticles using *Padina tetrastromatica* leaves extract. The morphology of silver nanoparticles was characterized by Transmission electron microscope. The TEM images reveal that the particles are spherical in shape and their size varies from 10-100nm. The XRD result confirms that the particles have a face centered cubic crystalline structure.

References

[1] K.Balantrapu, D. Goia, J. Mater Res. **24(**9), 2828 (2009).
[2] R.M.Tripathi, A. Saxena, N. Gupta, H. Kapoor,R.P. Singh, Digest J. Nanomat. Biostruct. **5**(2), 323 (2010).
[3] R. Patakfalvi, I. Dekany, Colloid Polym Sci. **280**(5), 461 (2010).
[4] L. Rodriguez-Sanchez, M.C. Blanco, M.A. Lopez-Quintela, J. Phys Chem B **104**, 9683(2000).
[5] A. Taleb, C. Petit, M.P. Pileni, J. Phys Chem B **102**(12), 2214 (1998).
[6] S. Mondal, N. Roy, R.A. Laskar, I. Sk, S. Basu,D. Mandal, Colloids Surf B. Biointerfaces. **82**(2), 497 (2011).
[7] N.A. Begum, S. Mondal, S. Basu, R.A. Laskar, D. Mandal, Colloids Surf B Biointerfaces **71**(1), 113 (2009).
[8] V. Kattumuri, K. Katti, S. Bhaskaran, E.J.Boote, S.W. Casteel, G.M. Fent, Small **3**(2), 333 (2007).
[9] J.Y. Song, B.S. Kim, Korean J Chem Eng. **25**(4), 808 (2008).
[10] M. Gilaki, J. Biol Sci.**10**(5), 465 (2010).
[11] K. Govindaraju, V. Kiruthiga, V. Ganesh Kumar, G. Singaravelu, J. Nanosci Nanotechnol. **9**, 5497 (2009).
[12] R. Schnetter, Marine Algen der karibischen Kusten von Kolumbein,. Phaeophyceae, Band 24. J. Cramer , Germany, **1**, 125. (1976).
[13] M.D. Guiry, D. Nic, E. Honncha, Algae Base version.,World wide electronic publications, National University of Ireland , Galway, (2003).
[14] B. D. Cullity. Elements of X-ray Diffraction, Edison-Wesley Publishing Company Inc, USA, (1978).
[15] M. Sastry, V. Patil, S.R. Sainkar, J. Phys Chem B. **102**, 1404 (1998).
[16] P. Mulvaney, Langmuir. **12,** 788 (1996).
[17] P. Mukherjee , A. Ahmad, D. Mandal, S. Senapati, S.R. Sainkar, M.I. Khan, Nano Lett, **1**(10) 515 (2001).
[18] P. Mukherjee, S. Senapati, D. Mandal, A. Ahmad, M.I. Khan, R. Kumar, M. Sastry, Chem. Bio. Chem. **3**, 461 (2002).
[19] J. Gonzalo, R. Serna, J. Sol, D. Babonneau, C. N. Afonso, J. Phys.: Condens. Matter **15** 3001 (2003).