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# ANTICANCER POTENTIAL OF GREEN SYNTHESIZED SILVER NANOPARTICLES OF SARGASSUM WIGHTII AGAINTS HUMAN PROSTATE CANCER (PC-3) CELL LINE.

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#### **ABSTRACT**

The purpose of this study is to evaluate the anticancer potential of different concentrations of silver nanoparticles biosynthesized by *S. wightii* on human prostate cancer cell (PC-3) *in vitro*. Silver nanoparticles were synthesized by the reduction of silver nitrate in the algal aqueous extract. The green synthesis of silver nanoparticles through algal extract was monitored by colour change and various spectroscopic studies. Results of MTT assay showed a decrease in viability of PC-3 cells by increasing the concentration of silver nanoparticles in the tested algae. The maximum inhibitory concentration was found to be 8.84 µg/ml at 48 hrs of incubation when compared to aqueous extract (40.59 µg/ml). Silver nanoparticles formed biologically by *S. wightii* characterized by a uniform shaped and very small in size and these properties making these silver nanoparticles to possess marked cytotoxic activity.

**KEYWORDS:** S. wightii, green synthesis, PC-3 cells and MTT assay.

#### INTRODUCTION

The field of nanotechnology is one of the most active areas of research in modern materials science. Nanotechnology is emerging as a cutting edge technology interdisciplinary with biology, chemistry and material science. [1] New applications of nanoparticles and nanomaterials are emerging rapidly in biomedical sciences. [2,3] This decade has witnessed the inception of new significant technological products particularly based on nanotechnology; nanoparticle synthesis is being widely explored since they exhibit unique size and shape dependent properties for applications in optics, electronics, catalytic systems, magnetic and biomedical such as HIV inhibition, cancer cell cytotoxicity and genotoxicity. [4-6] Apart from this, recently the anti-tumor effect of AgNPs has been reported against different cancerous cell lines. [7-9] Nanoparticles with the size range between 1 and 1000 nm are mainly explored for the diagnosis and treatment of human cancers, which led to the new discipline of nano-oncology. [10]

However, there is still need for economic, commercially viable as well as environmentally clean synthesis route to synthesize nanoparticles. A number of approaches are available for the synthesis of AgNPs. Among the various known synthesis methods, plant-mediated nanoparticle synthesis is preferred as it is rapid, cost-effective, eco-friendly, and safe for human therapeutic uses. [11-15] Cancer is one of most important scourges of mankind

and responsible for major mortality. The rapidly developing field of nanoscience has raised the possibility of using therapeutic nanoparticles in the diagnosis and treatment of human cancers. [10,16] Prostate cancer is the second most common cancer in men. An estimated 9,00,000 men worldwide were diagnosed with prostate cancer in 2008, accounting for almost one in seven (14%) cancers diagnosed in men (7% of the total in men and women). [17]

Innumerous studies from across the world have demonstrated that marine algae also possess a number of biological activities beneficial for human health, including antimicrobial, cytotoxic, antimitotic, anticancer, and anti-mutagenic activities. [18-24] Review of previous literature revealed that synthesis of nanoparticle using algae as source has been unexplored and unraveled. Recently there are a few reports regarding the use of algae as a biofactory for synthesis of metallic nanoparticles. Synthesis of gold nanoparticles using Sargassum wightii and Kappaphycus alvarezii. [25,26] Likewise, synthesized silver nanoparticles using Sargassum wightii, Gelidiella acerosa and Sargassum tenerrimum, has also been reported. [27-29] Therefore the present study was aimed to investigate the green synthesis silver nanoparticles of Sargasssum wighti and to evaluate its anticancer effect against human prostate cancer (PC-3) cell line.

#### MATERIALS AND METHODS

#### Collection and Processing of Sargassum wightii

Sargassum wightii was collected from Central Salts and Marine Chemicals Research Institute-Marine Algal Research Station, (CSMCRI-MARS), Mandapam Coast, Tamil Nadu. The sample was identified as Sargassum wightii (Phaeophyta) by Dr. K. Krishnamoorthy of Krishnamoorthy Institute of Algalogy, Chennai, Tamil Nadu, India.

#### **Preparation of Plant Extract**

The fine powder of the plant was obtained from the dried material by using kitchen blender. Then, 20 g of dried powder was taken and mixed with 200 ml of the respective solvents (methanol, chloroform, ethyl acetate, hexane and distilled water) and kept in a boiling water bath at 60°C for 10 min. The extract was filtered with Whatman filter paper No. 1. The filtered extract was stored in refrigerator at 4°C until further use.

#### Green Synthesis of Silver Nanoparticles (AgNPs)

Biological synthesis of AgNPs was carried out in the five different polar solvent extracts followed by the method of Song *et al.* (2009). [30]

#### **Bio-reduction of Silver Ions**

The bio-reduction of silver ions was monitored by UV-Visible Spectrophotometer at a range between 200 nm to 800 nm. [31,32] Synthesis of AgNPs was seen only in aqueous extract at 420 nm, in all other extracts AgNPs synthesis was not that much successful. Based on the above findings, aqueous extract and it's AgNPs were taken for further studies.

### Phytochemical Analysis

#### **Qualitative Phytochemical Analysis**

Preliminary phytochemical analysis of aqueous extract was carried out by method of Harborne (1973) and Parekh and Chanda (2007). [33,34]

#### **Characterization of AgNPs**

The synthesized AgNPs were characterized by the following methods.

#### **Transmission Electron Microscope (TEM)**

Transmission Electron Microscopic (TEM) analysis of the synthesized AgNPs of *S. wightii* were done according to the methods of Sondi and Sondi (2004). [35]

### Scanning Electron Microscope with Energy Dispersive X-Ray Spectroscopy (SEM & EDX)

SEM (this study was undertaken to know the size and shape of the AgNPs) analysis was done using Carl Zeiss MA15 / EVO 18. The presence of elemental silver was confirmed through EDX. Energy dispersive X-ray spectroscopy (EDX) was carried out by the same instrument and employed to confirm the presence of silver in the particles as well as to detect the other elementary compositions of the particles. [36,37]

#### Fourier Transform Infrared Spectroscopy (FT-IR)

The purified AgNPs were examined for the presence of biomolecules using FT-IR analysis by the method of Sivaraman *et al.* (2009). The spectrum obtained from the dried sample was recorded on FTIR spectrum (Perkin-Elmer, USA) in the diffuse reflectance mode at a resolution of 4 cm-1 in KBr pellets.

#### X-Ray Diffraction (XRD)

Crystalline AgNPs were determined by X-ray diffraction analysis. Briefly, the biosynthesized AgNPs were laid onto glass substrates on a Phillips PW 1830 instrument operating at a voltage of 40 kV and a current of 30 mA with Cu  $K\alpha 1$  radiation, and the diffracted intensities were recorded from 10′ to 70′ of 2  $\theta$  angles. [39]

#### **Atomic Force Microscope (AFM)**

Size and the surface topography of the drop coated film of the AgNPs were investigated with Atomic Force Microscope (Park XE-100) by the method of Sadhasivam *et al.* (2010). [40]

#### **Dynamic Light Scattering (DLS)**

Particle size was measured by laser diffractrometry using a Nano Size Particle Analyzer (ZEN 3600 MALVERN USA) in the range between 0.6 nm to 6.0  $\mu$ m, under the following conditions: particle refractive index 1.590, particle absorption coefficient 0.01, water refractive index 1.33, viscosity 0.8872, temperature 25°C and general calculation model for irregular particles. [41]

#### Cytotoxicity assay of aqueous extract and AgNPs

Cytoxicity assay PC-3 cell line was assessed by MTT following the method Mosmann  $(1983)^{[42]}$  and Manikandan *et al.* (2012). [43]

#### **Statistical Analysis**

The data with five replicates were subjected to statistical analysis and the mean value along with its respective standard error was calculated. The per cent change between control and experimental data were calculated. The data were analyzed statistically using Two Way Analysis of Variance (ANOVA). [44]

#### **RESULTS**

### Green Synthesis of silver nanoparticles using different solvent extracts of *S. wightii*

On mixing methanol, chloroform, hexane, ethyl acetate and aqueous extracts of *S. wightii* with 1mM AgNO<sub>3</sub> solution, the colour change of solution was not found to be clear in all the above mentioned extracts, which might be due poor solubility of these solvent extracts in 1mM AgNO<sub>3</sub> solution. On the other hand, aqueous extract alone showed clear solubility and colour formation with the appearance of dark brown colour from pale yellow solution. The colour formation occurred within 20 minutes itself. This might be due to the reduction of silver ions, indicating the formation of silver nanoparticles (Fig. 1).

### Characterization of Silver Nanoparticles UV-Vis Spectra Analysis

Evaluating the synthesis of silver nanoparticles using different solvent extracts of S. wightii showed poor solubility with 1mM AgNO<sub>3</sub> solution resulted in unassigned peaks in UV-Vis spectrophotomer and all the extracts also showed no much changes in the peak and the colour also remained same during different time intervals of UV observation (Fig. 2). In contrary, aqueous extract alone showed prominent peak around  $\lambda$ max 420 nm within 20 minutes (Fig. 3) with elevated dark brown colour formation. Based on colour change and UV-Vis spectral analysis, aqueous extract-based synthesized S. wightii AgNPs were taken for further analysis.

#### **Qualitative Phytochemical Analysis**

The phytochemical characteristics of *S. wightii* aqueous extract are summarized in the Table 1. The results revealed the presence of phenols, flavonoids, alkaloids, saponins and tannins in the aqueous extract. Carbohydrates, starch, acids, coumarins, quinones, terpenoids and steroids were absent in the aqueous extract.

#### **Transmission Electron Microscope (TEM)**

The spherical and plate like nanostructures were examined and size distribution of silver nanoparticles were clearly observed in the TEM analysis. Fig. 4 demonstrates that the TEM ascertained morphology and size of *S. wightii* silver nanoparticles in the optimized circumstance. The shape of the nanoparticles was mostly spherical and some are in slightly rounded rectangle like structures. The size distribution of silver nanoparticles was below 100 nm.

## Scanning Electron Microscope with Energy dispersive X-ray spectroscopy (SEM & EDX)

Scanning electron microscopy (SEM) was also used to investigate the morphology and size of the synthesized silver nanoparticles. SEM images were recorded at different magnifications and the SEM images showed high density of silver nanoparticles synthesized by *S. wightii* aqueous extract, which was further confirmed by EDX. Elemental silver can be seen in the graph presented by the EDX analysis in support of SEM results, which indicated the reduction of silver ions to elemental silver. The presence of Al signal might be due to the thin film made on the glass slide taken for the EDX (Fig. 5 A and B).

#### Fourier Transform Infrared Spectroscopy (FT-IR)

The FT-IR spectrum was used to identify the functional groups of the active components based on the peak value in the region of infrared radiation. The results of FT-IR peak values and functional groups are presented in Table 2 and 3. The FT-IR spectra of *S. wightii* aqueous extract shows interaction of biomolecules having intensive peak at 496 to 3127 Cm<sup>-1</sup> (Fig.6) and after the reduction of silver nanoparticles the biomolecules such as alkenes,

carboxylic acid, nitro groups, amines, ethers and alkyl halides are responsible for the formation of silver nanoparticles (Fig. 7).

#### X-Ray Diffraction (XRD)

The XRD patterns had three main diffraction features corresponding to the planes and were indexed with (111), (200) and (220) and all the three peaks were indexed to standard cubic phase of silver (JCPDS file no. 04-0783). The XRD spectra of our experiment indicated the formation of silver nanoparticles as crystalline in nature and aggregation might have formed due to the action of stabilizing agents present in the algal extract (Fig. 8).

#### **Atomic Force Microscope (AFM)**

The 3D images of the silver nanoparticles shown in Fig. 9 possess separated spherical particles. The sizes of particles are in the range of 19.643 to 24.888 nm which are clearly indicated with the scales in the figure 9.

#### **Dynamic Light Scattering (DLS)**

Dynamic light scattering (DLS) analysis showed the size distribution of particles with an % intensity 90.4 and width of 84.4 (d nm) (Fig. 10). As expected, the particle size obtained from TEM and DSL scattering is marginally different due to the varying principles used for measurement.

#### Cell viability assay against PC-3 cancer cell line

The S. wightii aqueous extract and biosynthesized silver nanoparticles were evaluated for their effect on cell viability at concentrations of 25, 50, 75, 100 and 125 ug/ml by MTT method against PC-3 cancer cell lines. The tested aqueous extract and biosynthesized silver nanoparticles showed a dose-dependent decrease in cell viability at the end of 24 hrs. Increasing the time of incubation to 48 hrs showed a further decrease in cell viability (Table 4). Statistical treatment of the data by two-way ANOVA showed that all values were significant at 5 % level. Inhibition of cell viability to 50% was observed at 109.88 µg/ml with an exposure time of 24 h and 40.59 µg/ml of 48 h in aqueous extract, On the other hand, AgNPs showed 50% inhibition of cell viability at 49.48 µg/ml in 24 h and 8.84 µg/ml at 48 h (Fig. 11), and the exact IC<sub>50</sub> values are showed in Table 5. Further studies were carried out at 40.59 µg/ml of aqueous extract and 8.84 µg/ml of AgNPs; being the 48 h IC<sub>50</sub> concentrations. ANOVA analysis revealed that all the values were significantly different.

#### **Cytomorphology Observations**

Treatment of PC-3 cells with aqueous extract and AgNPs even at low doses induced morphological changes in the PC-3 cells, which had similar effect on cells morphology. Microscopic observations were made using Nikon light inverted microscope, wherein treated cells showed distinct cellular morphological changes indicating unhealthy cells, whereas the control appeared normal in shape (Fig. 12). Control cells were irregular confluent aggregates with rounded and polygonal cells.

Aqueous extract and biosynthesized AgNPs treated cells appeared to shrink, became spherical in shape and cell

spreading patterns were restricted when compared to control.

Table 1: Qualitative phytochemicals of S. wightii aqueous extract

S.No.	Phytochemicals	Aqueous extract
1.	Carbohydrate	=
2.	Starch	=
3.	Tanins	+
4.	Phenols	+
5.	Acids	=
6.	Alkaloids	+
7.	Flavonoids	+
8.	Coumarins	=
9.	Quinones	=
10.	Terpenoids	=
11.	Steroids	-
12.	Saponins	+

Table 2: FT-IR spectral peak values and functional groups obtained for the aqueous extract of S. wightii

PEAK VALUES	FUUNCTIONAL GROUPS	
3127 Cm-1	Alkynes ≡C-H Stretch	
3064 Cm <sup>-1</sup>	Alkenes C=C-H Asymmetric Stretch	
3025 Cm <sup>-1</sup>	Alkenes C=C-H Asymmetric Stretch	
2920 Cm <sup>-1</sup>	Alkanes H-C-H Asymmetric & Symmetric Stretch	
1951 Cm <sup>-1</sup>	C=N Stretch	
1893 Cm <sup>-1</sup>	C=O	
1844 Cm <sup>-1</sup>	Ketones C=O Stretch	
1711 Cm <sup>-1</sup>	C=O Carbonyl group	
1699 Cm <sup>-1</sup>	C=O Carbonyl group  Ketones C=O Stretch	
1659 Cm <sup>-1</sup>	Ketones C=O Stretch	
1621 Cm <sup>-1</sup>	KetonesC=O Stretch	
1530 Cm <sup>-1</sup>		
1487 Cm <sup>-1</sup>	Aromatic Rings C-C=C Asymmetric Stretch	
1408 Cm <sup>-1</sup>	Aldehydes C-H	
1337 Cm <sup>-1</sup>	Aldehydes C-H bending	
1286 Cm <sup>-1</sup>	Ethers (C-O Stretch)	
1231 Cm <sup>-</sup>	Ethers (C-O Stretch)	
1198 Cm <sup>-1</sup>	Ethers (C-O Stretch)	
1173 Cm <sup>-1</sup>	Ethers (C-O Stretch)	
1028 Cm <sup>-1</sup>	Ethers (C-O Stretch)	
969 Cm <sup>-</sup>	Alkenes =C-H bend	
989 Cm <sup>-1</sup>	Alkene C=C bendin	
884 Cm <sup>-1</sup>	Alkene C=C bending	
849 Cm <sup>-1</sup>	Halo compound C-Cl	
828 Cm <sup>-1</sup>	Alkene C=C bending	
787 Cm <sup>-1</sup>	C-H bending	
691 Cm <sup>-1</sup>	<b>Phenylgroup</b> Strong (look for =C-H & C=C first)	
665 Cm <sup>-1</sup>	alkylhalides C-Br stretch	
650 Cm <sup>-1</sup>	alkylhalides C-Br stretch	
607 Cm <sup>-1</sup>	alkylhalides C-Br stretch	
629 Cm <sup>-1</sup>	alkylhalides C–Br stretch	
550 Cm <sup>-1</sup>	alkylhalides C-Br stretch	
515 Cm <sup>-1</sup>	Alkylhalides C–Br stretch	
528 Cm <sup>-1</sup>	alkyl halides C–Br stretch	
496 Cm <sup>-1</sup>	alkyl halides C-Br stretch	

Table 3: FT-IR spectral peak values and functional groups obtained for the AgNPs of S. wightii

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PEAK VALUES	FUUNCTIONAL GROUPS			
3058 Cm <sup>-1</sup>	Alkenes C=C-H Asymmetric Stretch			
2971 Cm <sup>-1</sup>	Alkanes H-C-H Asymmetric & Symmetric Stretch			
2926 Cm <sup>-1</sup>	Carboxylic Acids Hydrogen-bonded O-H Stretch			
2841 Cm <sup>-1</sup>	Carboxylic Acids Hydrogen-bonded O-H Stretch			
1682 Cm <sup>-1</sup>	Alkenes C-C=C Symmetric Stretch			
1605 Cm <sup>-1</sup>	Alkenes C-C=C Symmetric Stretch			
1573 Cm <sup>-1</sup>	Nitro Groups N=O Stretch			
1542 Cm <sup>-1</sup>	Nitro Groups N=O Stretch			
1507 Cm <sup>-1</sup>	Nitro Groups N=O Stretch			
1445 Cm <sup>-1</sup>	Amines—Secondary N-H Bend			
1334 Cm <sup>-1</sup>	Amides N-H Stretch (similar to amines)			
1302 Cm <sup>-1</sup>	Nitro Groups N=O Bend			
1293 Cm <sup>-1</sup>	Ethers (C-O Stretch)			
1253 Cm <sup>-1</sup>	Ethers (C-O Stretch)			
1205 Cm <sup>-1</sup>	Ethers (C-O Stretch)			
1171 Cm <sup>-1</sup>	Esters (C-O Stretch)			
1108 Cm <sup>-1</sup>	Esters (C-O Stretch)			
1083 Cm <sup>-1</sup>	Esters (C-O Stretch)			
1022 Cm <sup>-1</sup>	Esters (C-O Stretch)			
836 Cm <sup>-1</sup>	Aromatiques C-H			
813 Cm <sup>-1</sup>	Aromatiques C-H			
782 Cm <sup>-</sup>	Aromatiques C-H			
768 Cm <sup>-</sup>	Aromatiques C-H			
722 Cm <sup>-1</sup>	( <b>CH</b> <sub>2</sub> <b>-</b> ) <b>n</b> C-H			
659 Cm <sup>-1</sup>	Alkylhalides C–Br stretch			
630 Cm <sup>-1</sup>	alkyl halides C-Br stretch			
598 Cm <sup>-1</sup>	alkyl halides C-Br stretch			
548 Cm <sup>-1</sup>	alkyl halides C-Br stretch			
527 Cm <sup>-1</sup> alkyl halides C–Br stretch				
498 Cm <sup>-1</sup>	alkyl halides C-Br stretch			

Table 4: Per cent cell viability of PC-3 cells for 24 and 48 hrs when treated with aqueous extract and AgNPsof *S. wightii*.

Time	24 hrs incubation		48 hrs incubation	
Concentration	Aqueous extract	Silver nanoparticles	Aqueous extract	Silver nanoparticles
Control	$100 \pm 0$	$100 \pm 0$	$100 \pm 0$	$100 \pm 0$
10a/ml	95.35± 0.49*	82.29 ± 0.37*	70.99 ±0.35*	43.41 ±0.29*
10 μg/ml	(-4.65)	(-17.71)	(-29.01)	(-56.59)
20a/ml	90.16 ±0.37*	78.91 ±0.401*	62.61 ± 0.61*	40.76 ±0.33*
20 μg/ml	(-9.844)	(-21.09)	(-37.39)	(-59.24)
20a/ml	84.71±0.43*	65.43 ± 0.34*	54.69 ±0.35*	37.48 ± 0.33*
30 μg/ml	(-15.29)	(-34.57)	(-45.31)	(-62.52)
40 μg/ml	77.32 ±0.44*	57.87±0.42*	$50.36 \pm 0.76$ *	33.29 ±0.22*
40 μg/III	(-22.68)	(-42.13)	(-49.64)	(-66.71)
50 ug/ml	$73.42 \pm 0.31*$	$49.56 \pm 0.27*$	44.37 ±0.52*	29.58 ±0.30*
50 μg/ml	(-26.58)	(-50.44)	(-55.63)	(-70.42)

Values are mean  $\pm$  S.E. of five individual observations.

Values in parentheses are per cent change over control.

Table: 5 Exact IC<sub>50</sub> values of aqueous extract and AgNPs of S. wightii against PC-3 cell line.

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	Time	Aqueous extract (con)	AgNPs (con)	
	24 Hrs	109.88 μg/ml	49.48 μg/ml	
	48 Hrs	40.59 ug/ml	8.84 ug/ml	

<sup>-</sup> Denotes per cent decrease over control.

<sup>\*</sup> Denotes that values are significant at P<0.05.

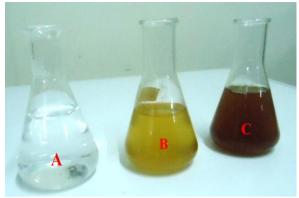


Fig. 1: A. Color intensity of 1 mM AgNO<sub>3</sub> solution.

B. Aqueous extract of Sargassum wighhtii (pale yellow colour).

C. AgNPs synthesized at different hours with pale yellow colour to dark brown colour.

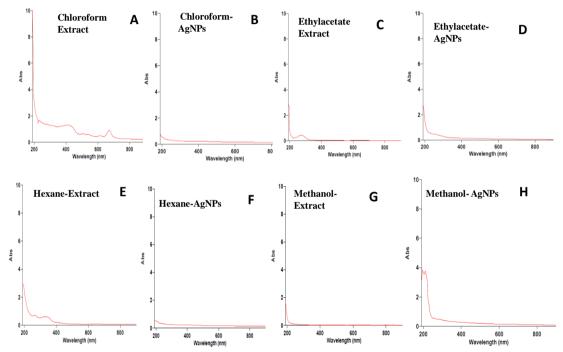


Fig. 2: UV-Vis spectral image of different solvent extract synthesized silver nanoparticles (A) Chloroform extract, (B) ChlorofomAgNPs, (C) Ethyl acetate extract, (D) Ethyl acetate AgNPs, (E) Hexane extract, (F) Hexane AgNPs, (G) Methanol extract and (H) Methanol AgNPs

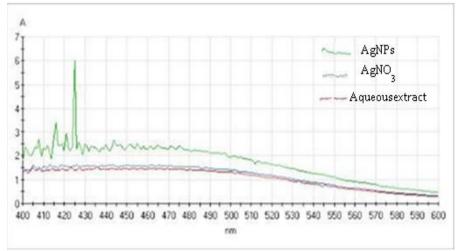


Fig. 3: UV-Vis spectral image of aqueous extract based synthesized silver nanoparticles.

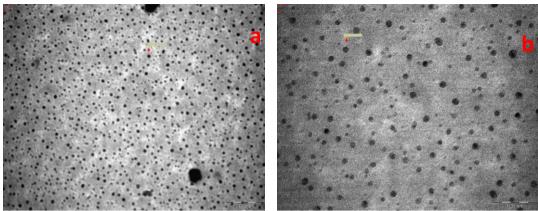


Fig: 4. TEM images of silver nanoparticles formed by reduction of silver nitrate using *S. wightii* (a) 200 nm (b) 100 nm.

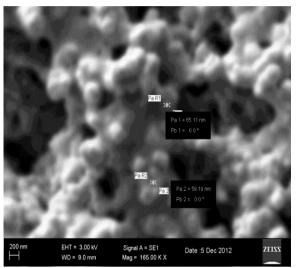


Fig. 5A: SEM micrograph of silver nanoparticles synthesized from the aqueous extract of *S. wightii* 

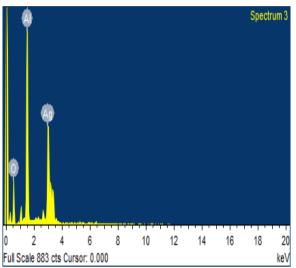


Fig. 5B: Energy dispersive spectrometer analysis of silver nanoparticles synthesized from the aqueous extract of *S. wightii* 

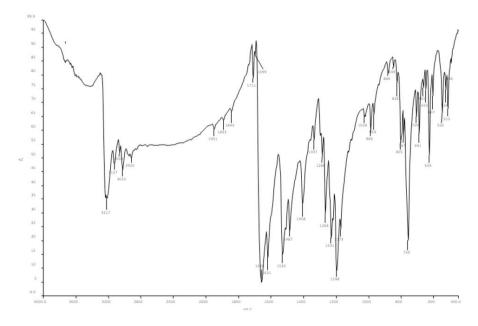


Fig. 6: FT-IR spectral image of various functional groups ( $4000 \text{ to } 400 \text{ cm}^{-1}$ ) obtained for aqueous extract of S. wightii

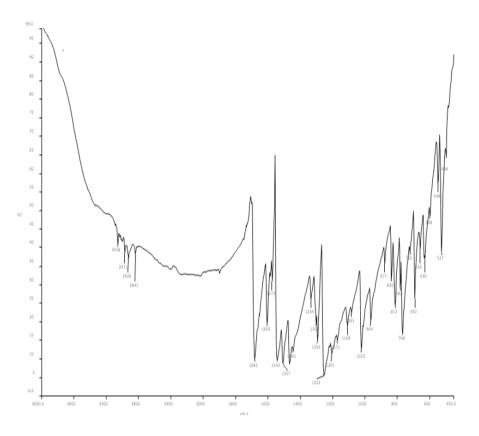


Fig. 7: FT-IR spectral image of various functional groups (4000 to 400 cm<sup>-1</sup>) obtained for AgNPs of S. wightii

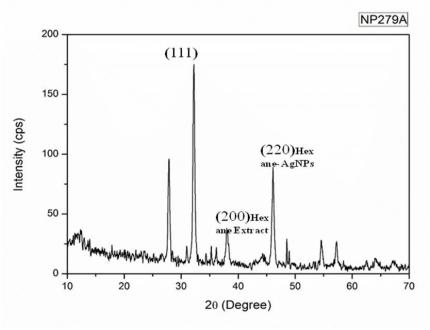


Fig: 8. XRD pattern for S. wightii mediated silver nanoparticles

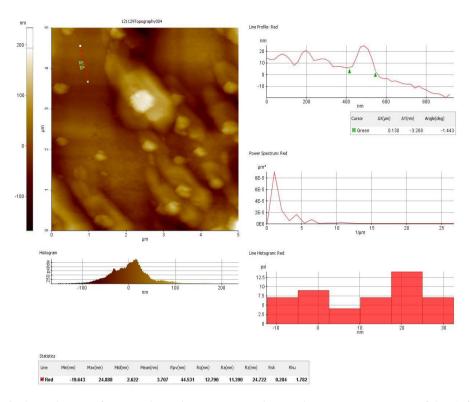


Fig. 9: AFM image of synthesized silver nanoparticles using aqueous extract of S. wightii

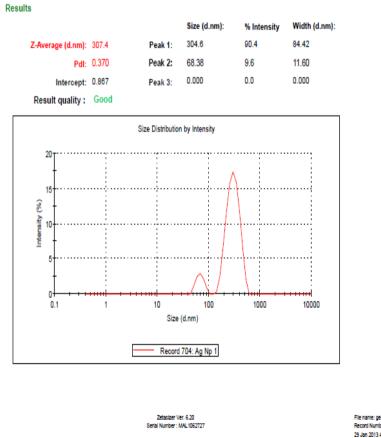


Fig: 10. DLS image of synthesized silver nanoparticles using aqueous extract of S. wightii

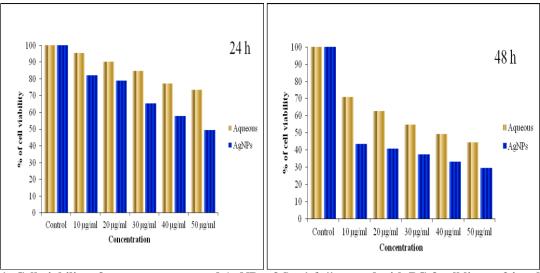


Fig. 11: Cell viability of aqueous extract and AgNPs of S. wightii treated with PC-3 cell line at 24 and 48 h.

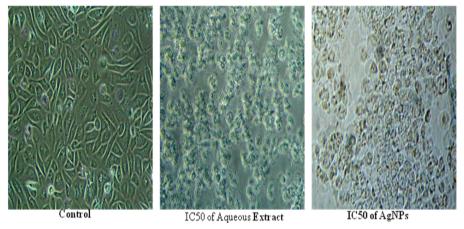


Fig. 12: Cytomorphology of PC-3 cells

#### DISCUSSION

The aqueous silver ions when exposed to Sargassum wightii aqueous extract resulted in the reduction of silver ions, there by leading to the formation of silver hydrosol. The aqueous extract was pale yellow colour before addition of silver nitrate solution and this changed to dark brown colour suggesting the rapid formation of silver nanoparticles. The time duration of change in colour was 20 minutes. The change of colour indicates biosynthesis of silver nanoparticles and this might be due to surface plasmon resonance phenomenon. The synthesized silver nanoparticles had been confirmed by measuring the UV-Vis spectrum of the reaction media. The UV-Vis spectrum of colloidal solutions of silver nanoparticles synthesized from the aqueous extract of Sargassum wightii showed an absorbance peak at 420 nm.

Silver nanoparticles synthesized by using aqueous extract of *Sargassum polycystum* showed absorbance at 430 nm<sup>[45]</sup>, *Sargassum longifolium* showed absorbance peak at 460 nm<sup>[46]</sup>, *Urospora sp.* showed absorbance at 430 nm<sup>[47]</sup>, and of *Cochlospermum religiospum* showed absorbance between 200-400 nm.<sup>[48]</sup> In many other cases,

like silver nanoparticles synthesized leaf extracts of *Eucalyptus hybrid*<sup>[49]</sup>, *Acalypha indica*<sup>[50]</sup>, *Solanum tarvum*<sup>[51]</sup>, *Helianthus annus*<sup>[52]</sup>, and *Cassia auriculata*<sup>[53]</sup>, the absorbance peaks were between 400 and 450 nm. When compared with these plants and seaweeds, silver nanoparticles synthesized from aqueous extract of *Sargassum wightii* were active at relatively lower wavelength.

The AFM analysis of aqueous extract of *Sargassum wightii* showed spherical shaped silver nanoparticles formed with diameter ranging from 24.888 nm. The AFM analysis of leaf extract of *Citrulus colocynthus* showed the size of silver nanoparticles as 31 nm<sup>[54]</sup>, of *Syzygium cumini* as 30 nm<sup>[55]</sup> and of *Citrulus limon*as 50 nm.<sup>[56]</sup> The particle size of silver nanoparticles synthesized from the leaf extract of *Glycine max* was from 25 to 100 nm<sup>[57]</sup>, that of *Moringa olifera* was from 5 to 80 nm.<sup>[58]</sup> Sasikala and Savithramma (2012)<sup>[48]</sup> have stated that the size of the nanoparticles varies with the plant by altering the pH, strength of elements, plant sources, incubation temperature of the nanoparticle synthesis reaction mixture and the synthesis methods, ans so it is possible to create a wide range of different

nanoparticles. Nanoparticles of various sizes and properties can be obtained by further tapping the plant bioresources of diverse type in wild environment.

The cytotoxicity of S. wightii aqueous extract-based synthesized silver nanoparticles showed profound effect on PC-3 cells when compared to aqueous extract and this might be due to phytocompounds such as tannins, phenols, alkaloids, flavanoids and saponins that responsible for the reduction of silver nanoparticles. Zhang et al. (2011)<sup>[59]</sup> stated that brown seaweeds with low molecular weight fucoidan, mediated the broadspectrum growth inhibition of human carcinoma cells like HeLa, HT1080, K562, U937, A549 and HL-60. Bousarghin et al. (2003)<sup>[60]</sup> reported that sulfated polysacchardies such as heparin, cellulose sulfate and dextran sulfate block the infectivity of papllomavirus. Taskin et al. (2010) [61] also stated that polysacchardies and terpenoids from brown algae are considered as promising bioactive molecules with anticancer activity. Marine algae are important sources of non-animal sulfated polysaccharides and these biomolecules are widely studied on the therapeutic applications such as anti-thrombotic, anticoagulant, antioxidant, inflamatory and anti-proliferative effects as opined by Barroso et al. (2008). [62]

In recent years, S. wightii has becoming a source of medicine due to the medicinal properties, which is attributed to its unique chemical library polysacchardies, vitamins, minerals, polyunsaturated fatty acids. Moreover they can also serve as good source of healthy food. Also seaweeds may solve the problems of deficiency of protein, carbohydrates and minerals in human nutrition by consuming them in daily life. The present study suggests that the seaweed extract and extract-based synthesized silver nanoparticles S.wightii possess potent anticancer property. It is suggested that S.wightii could be a potent source of natural anticancer which are of great importance as therapeutic agent in inhibiting the growth of cancerous cells. Further seaweeds will lead as novel candidates in pharmaceuticals to develop a natural compounds as an anticancer agent for production of potential anticancer drug and it is necessary to revitalize the use of seaweed in the newly health conscious consumer environments of several countries.

#### CONCLUSION

The present investigation reveal that aqueous extract-based synthesized silver nanoparticles showed high anti-proliferative activity against PC-3 cells, due to suppression of the cancer cell growth. Hence, *Sargassum wightii* based synthesized silver nanoparticles can be used as a potential therapeutic agent for human prostate cancers.

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