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Evaluation of Phytochemicals Profile and Antioxidant Activities of *Asperagus racemosus* using *in vitro* and *in silico* Approach

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ABSTRACT

Asparagus racemosus belongs to family Liliaceae and commonly known as Satawar, Satamuli, Satavari. A. racemosus is an important medicinal plant and its root paste or root juice has been used in various ailments and as health tonic. The management of burn wound victims is often considered a difficult task due to frequent infections. Moreover, the treatment with conventional antibiotics is associated with the upsurge of drug resistant bacterial strains; for the current study, A. Racemosus was collected, authenticated, and subjected for the preparation of extracts using ethanol as solvent for extracting phyto constitutes present in them. Further in Phytochemical analysis of Asparagus racemosus the quantitative estimation of phenolic and flavonoid content was done. Results obtained were TPC (Total Phenolic Content) and TFC (Total Flavonoid Content) 22.2514 µg GAE/mg and 5.5028 µg QE/mg respectively which showed the presence of good amount of phenolic and flavonoid content. In chemical analysis the plant extract was analysed via. GC-MS and FTIR. The GC-MS study showed many Phytochemicals such as Hydroxypyranone, Tetradecanoic acid, Octadecadienoic acid, n-Hexadecanic acid etc. (24 compounds). These compounds show antimicrobial, antitumor, anticonvulsant, anti-inflammatory and ty-rosinase inhibitory effects, Anticancer, Antioxidant properties. The FTIR analysis of ethanol plant extracts of A. racemosus confirmed the presence of alcohols, phenols, alkanes, carboxylic acids, alkenes, aromatics, esters, ethers, allenes compounds etc., which showed major peaks.

Keywords: Phytochemical analysis; Tetradecanoic acid; Hydroxypyranone; Octadecadienoic acid

INTRODUCTION

In silico approach yield was also determined to be useful. Among the phytoconstituents obtained from GC-MS, majority has showed the useful ADMET profiles i.e. they showed the exhibition of properties such as human intestinal absorption, blood brain barrier penetration, human oral bioavailability, Caco^{-2}, and plasma binding properties. ProTox tool that determines the toxicity of the chemical compounds has resulted that among 24 compounds, 17 compounds are in the range of 4-5 which indicates that they are non-toxic in nature. Thus, compounds from the plant extract of *Asparagus racemosus* can be effectively used in the various formulation of drug in order to treat burn wounds and overcome the difficulties raised in the treatment of burn wounds as future application [1].

Extraction

Cold maceration: Maceration is a technique use in wine making and has been adopted and widely used in medicinal plants research. Maceration involved soaking plant materials (coarse or powdered) in a stoppered container with a solvent and allowed to stand at room temperature for a period of minimum 3 days with frequent agitation. The processed intended to soften and break the plant's cell wall to release the soluble phytochemicals. Collection of Plant materials (Medicinal part: Root). Washing it with distilled water and drying it at 25°C-30°C. Crushing dried root material to powder and keeping it in refrigerator at 4°C until use. Preparation of plant extracts with 100 ml distilled water, ethanol by maceration. Filtration of plant extract through a Buchner funnel with What man filter paper number after 24 hrs for distilled water and 48 hrs for ethanol. Evaporation of extract under reducing pressure to dryness at 40°C in hot air oven. Collection and storage of crude extracts at 4°C until using. Re-dissolving of extract in sterile distilled water [2].

Microwave assisted extraction: Microwave-assisted extraction technology is now becoming an emerging technology to obtain useful compounds from plant biomass. Microwaves directly generate heat by initiating molecular motions of water and electrolytes in plant biomass; therefore, the irradiated materials are quickly heated from within. In addition, microwaves enhance the diffusion of the target compounds by directly heating biomass from within and facilitating non equilibrium mass transfer from inside of the plants to the extracting solvent. 0.5 g of dried plant root powder was dissolved in 10 ml of distilled water followed by dissolving the mixture was subjected to microwave radiation in micro oven for two set of time intervals i.e. 2 minutes (1 min heating+1 min cooling), 6 minutes (1 min heat+1 min cool repetition: 3 times). Further the mixture was filtered using Whatman filter paper the filtrate was dried for 24 hrs in hot air oven at 40°C. Dried extract was scrape and dry weight of extract was determined [3].

Soxhlet extraction: Soxhlet extraction is only required where the desired compound has a limited solubility in a solvent, and the impurity is insoluble in that solvent. If the desired compound has a high solubility in a solvent, then a simple filtration can be used to separate the compound from the insoluble substance. In this method, the finely ground crude powder of plant material is placed in a porous bag or "thimble" made of strong filter paper, which is placed in chamber E of the Soxhlet apparatus. The extracting solvent in flask A is heated, and its vapours condense in condenser D. The condensed extractant drips into the thimble containing the crude powder of plant material and extracts it by contact. When the level of liquid in chamber E rises to the top of siphon tube C, the liquid contents of chamber E siphon into flask A. This process is continuous and is carried out until a drop of solvent from the siphon tube does not leave residue when evaporated. The advantage of this system is that instead of many portions of warm solvent being passed through the sample, just one batch of solvent is recycled. This method cannot be used for thermolabile compounds as prolonged heating may lead to degradation of compounds [4].

MATERIALS AND METHODS

Phytochemical Analysis

Determination of TPC (Total Phenolic Content): Materials used are as

- Gallic acid in water (100 µg/ml)-5 ml
- Distilled water-5 ml
- 10 % Folin- Cicocalteu's phenol reagent-5 ml
- 1M sodium carbonate solution-5 ml
- Ethanolic extract (500 mg/ml)-2 ml each
- Soxhlet extract (500 mg/ml)-2 ml each

Principle: The Folin-Ciocalteu (FC) assay is such a method and has been proposed as a standardized method for use in the routine quality control and measurement of antioxidant capacity of food products and dietary supplements. The F-C assay relies on the transfer of electrons in alkaline medium from phenolic compounds phosphomolybdic/phosphotungstic acid complexes to form blue complexes that are determined spectroscopically at approximately 760 nm. Although the exact chemical nature of the F-C reaction is unknown, it is believed that sequences of reversible one- or two-electron reduction reactions lead to blue species (possibly (PMoW11O40)4-). Major considerations in the interpretation of the F-C assay are that the chemistry is non-specific and that other oxidation substrates in each extract sample can interfere in an inhibitory, additive or enhancing manner [5].

Method: The Total Phenolic Content (TPC) of all extracts was determined by using the Folin Ciocalteu method. Standard solutions of Gallic acid of concentration 1.56-100 µg/ml were prepared in water. 20 µl of extracts or standard solution were added to 20 µl of distilled water. 30 µl of 10 % Folin-Cicocalteu's phenol reagent and 50 µl of 1 M sodium carbonate solution were added to the mixture in a 96 well plate. Distilled water was used as blank. The reaction mixture was incubated for 60 min at room temperature and protected from light. The absorbance was measured at 750 nm with a microplate reader. TPCs were expressed as µg Gallic Acid Equivalents (GAE)/mg of dry plant material [6].

Determination of TFC (Total Flavonoid Content): Materials used are as

- Quercetin in 80 % ethanol (100 µg/ml)-5 ml
- 10 % aluminium chloride solution-5 ml
- 95% ethanol-5 ml
- 1M sodium acetate-5 ml
- Ethanolic extract (500 mg/ml)-2 ml each
- Soxhlet extract (500 mg/ml)-2 ml each

Principle: The principle involved in aluminium chloride ($AlCl_3$) colorimetric method is that $AlCl_3$ forms acid stable complexes with the C-4 keto groups and either the C-3 or C-5 hydroxyl group of flavones and flavanols. In addition, it also forms acid labile complexes with the ortho-dihydroxyl groups in the A- or B-ring of flavonoids.

Methods: Total Flavonoids Content (TFC) of all extracts were determined by the aluminium chloride colorimeter assay. Standard solutions of quercetin of concentration 1.56-100 µg/ml were prepared in 80% ethanol. 30 µl of extracts (1 mg/ml) or standard solution was added to 20 µl of 10% the aluminium chloride solution and followed by 100 µl of 95% ethanol. 10 µl of 1 M sodium acetate was added to the mixture in a 96-well plate. 80% ethanol was used as the reagent blank. All reagents were mixed and incubated for 40 min at room temperature protected from light. The absorbance was measured at 415 nm with a microplate reader i.e. multimode reader. TFCs were expressed as µg Quercetin Equivalents (QE)/mg dry of plant material [7].

Chemical Analysis

Preparation of extract: Tubers of *Asparagus racemosus* were shade dried. 20 g of the powdered tubers were soaked in 95% ethanol for 12 h. The extracts were then filtered through Whatman filter paper No.41 along with 2 gm sodium sulphate to remove the sediments and traces of water in the filtrate.

Before filtering, the filter paper along with sodium sulphate was wetted with 95% ethanol. The filtrate was then concentrated by bubbling nitrogen gas into the solution. The extract contained both polar and non-polar phytocomponents of the plant material used. 2 µl of these solutions was employed for GC/MS analysis [8].

GC-MS analysis: GC-MS analysis was carried out on a GC clarus 500 Perkin Elmer system comprising a AOC-20i autosampler and gas chromatograph interfaced to a mass spectrometer (GC-MS) instrument employing the following conditions: Column Elite-1 fused silica capillary column (30 × 0.25 mm ID × 1EM df, composed of 100% Dimethyl poly siloxane), operating in electron impact mode at 70 eV; helium (99.999%) was used as carrier gas at a constant flow of 1 ml/min and an injection volume of 0.5 EI was employed (split ratio of 10:1) injector temperature 250°C; ion-source temperature 280°C. The oven temperature was programmed from 110°C (isothermal for 2 min), with an increase of 10°C/min, to 200°C, then 5°C/min to 28°C, ending with a 9 min isothermal at 280°C. Mass spectra were taken at 70 eV; a scan interval of 0.5 s and fragments from 40 Da to 550 Da.

FTIR (Fourier Transform Infrared Spectrophotometer) Analysis: Fourier Transform Infrared Spectrophotometer (FTIR) is perhaps the most powerful tool for identifying the types of chemical bonds (functional groups) present in compounds. The wavelength of light absorbed is characteristic of the chemical bond as can be seen in the annotated spectrum. By interpreting the infrared absorption spectrum, the chemical bonds in a molecule can be determined. Dried powder of different solvent extracts of each plant materials was used for FTIR analysis. 10 mg of the dried extract powder was encapsulated in 100 mg of 397 KBr pellet, in order to prepare translucent sample discs. The powdered sample of each plant specimen was loaded in FTIR spectroscope, with a Scan range from 400 nm to 4000 nm with a resolution of 4 cm [9].

***In silico* Analysis**

ProTox: ProTox, a web server for the prediction of rodent oral toxicity. The prediction method is based on the analysis of the similarity of compounds with known median Lethal Doses (LD 50) and incorporates the identification of toxic fragments, therefore representing a novel approach in toxicity prediction. In addition, the web server includes an indication of possible toxicity targets which is based on an in-house collection of protein-ligand-based pharmacophore models (‘toxophores’) for targets associated with adverse drug reactions [10].

Protox Interface

Input: The ProTox web server has an easy-to-use interface and the only requirement is the 2D structure of the molecule for which the toxicity is to be predicted. The user has the possibility to draw the structure with an embedded chemical editor or use the compound name.

Output: Typically, a toxicity prediction report for one compound is generated within seconds. The report can be divided into two parts: The prediction of the acute oral toxicity and the indication of possible toxicity targets. The oral toxicity prediction results are based on the analysis of 2D similarities and the recognition of toxic fragments, as described below. In addition to the prediction of the LD50 in mg/kg, the input compound is classified into a toxicity class ranging from I to VI, according to the globally harmonized system of classification of labelling of chemicals. The prediction accuracy derived from cross-validation results is also given. Additionally, the structure of the input compound as well as its physicochemical properties are displayed. In the similarity section, the three most similar compounds of the database which are used for the prediction of the oral toxicity, their properties and classification are shown. Furthermore, the fragment section indicates if and where toxic fragments occur in the input compound. In the second part, the possible binding to defined toxicity targets is indicated. Currently, 15 toxicity targets which have been associated with adverse drug reactions are covered, but future updates of the target list are planned. Wherever available, further information about the protein targets is given as links to the Super Target database. The toxicity target alert is based on ligand mapping to pharmacophore models developed from targets with known experimental structures and the similarity of the input molecule to known ligands of a target, as described below. For every input compound, a toxicity target profile is provided in form of a 4-coloured heat map. The more intense the colour, the higher is the probability of binding. Moreover, details are provided for every indicated toxicity target, including the similarity to known ligands of that target and the average mapping to the toxicophoric models [11].

AdmetSAR

AdmetSAR description: In total, the admetSAR database included more than 210 000 annotated measurements of 95 629 unique compounds including thousands of FDA approved and experimental drugs, pesticides, environmental agents, and industrial chemicals. The data fields of each compound included three types: The general information, the physicochemical properties, and the ADMET associated profiles. The general data of each molecule includes IUPAC name, formula, CASRN, common name, Drug-bank ID, SMILES. The physicochemical properties include MW, Log P, the number of hydrogen bond acceptor’s and donors, and TopoPSA. More than 45 kinds of ADMET-associated properties, proteins, species, and organisms, such as water solubility, human intestinal absorption, oral bio-availability, blood-brain barrier penetration, P-glycoprotein substrate and inhibitor, renal organic cation transporter, plasma protein binding, volume of distribution, CYP450 substrates and inhibition (CYP1A2, 2C9, 2C19, 2D6 and 3A4), drug-induced liver injury, Human Ether-A-Go-Go-Related Gene (hERG) inhibition, rat acute toxicity, skin sensitivity, AMES mutagenicity, carcinogens, fish toxicity, Tetrahymena pyriformis toxicity, honey bee toxicity, quail toxicity, reproductive toxicity, biodegradability, bio-concentration factors, etc., were stored in admetSAR database [12].

Prediction of ADMET properties of new chemicals: In total, 22 highly predictive qualitative classification models were implemented. These models include human intestinal absorption, blood-brain barrier penetration, Caco-2 permeability, P-glycoprotein substrate and inhibitor, CYP450 substrate and inhibitor (CYP1A2, 2C9, 2D6, 2C19, and 3A4), hERG inhibitors, AMES mutagenicity, carcinogens, fathead minnow toxicity, honey bee toxicity, and Tetrahymena Pyriformis toxicity. The range of the area under the receiver operating characteristic curve (AUC) is from 0.638 to 0.956 for 22 classification models *via* 5-fold cross validation. In addition, all classification models were given a probability output described in our previous work instead of simple binary output. In scientific community of ADMET prediction, quantitative predictions are more useful. Therefore, five highly predictive quantitative regression models including $Caco^{-2}$ permeability, water solubility, rat acute toxicity, Tetrahymena pyriformis toxicity, and fathead minnow acute toxicity were built using the Support Vector Machine (SVM) regression algorithm and implemented in admet SAR. The range of the square of correlation coefficient (R_2) is from 0.564 to 0.810, and the range of Root-Mean-Square Deviation (RMSE) is from 0.256 to 0.283 using 5-fold cross validation. To ensure usefulness of admetSAR, it will be updated monthly with additional computational models based on available data, especially for quantitative models using My SQL database management system. If data sets with new end points are reported, new models will be built and implemented in our database. If data sets with known end points in admet SAR are reported, the old models will be updated and replaced by new ones with more diverse chemical space coverage.

The generalization ability of a model decides the usefulness and reliability of models. In order to test the actual predictive ability of admet SAR, several models were validated using the available external validation sets. For example, 27 novel chemicals were predicted first using the admet SAR server and were further assayed using the MITI-I test protocol. The detailed experimental and predicted results were given in Supporting Information Scheme S1 and Table S3. The overall predictive accuracy of admet SAR was 88.9%; that is, 24 chemicals were predicted correctly. The admetSAR outperformed Biowin 5 and Biowin 6 implemented in the EPISuite v4.10. In addition, above data sets of 27 computational models were released. All data sets were carefully curated by us and can be directly used to build ADMET models and methodology assessment, which can be downloaded from the download module. However, a registration is required for all users. And we encourage the user to provide new comments about how to improve the next version of admetSAR or upload new data sets to admetSAR [13].

Synthesis of Silver Nano-Particles (SNPs) and characterization

Synthesis of Silver Nano Particles (SNPs): For the synthesis of AgNPs, 50 ml of aqueous Root extract of *A. raemosus* and 50 ml of 1 mM silver nitrate ($AgNO_3$) solution were mixed with 400 ml of distilled water in the conical flask. Immediately the flask was kept in the sunlight. A change in the colour was observed indicating the beginning of the mechanism of the formation of silver nanoparticles the colour change of the mixture from light to yellowish to reddish to colloidal was monitored periodically. After the synthesis and completion of reaction the solution was centrifuged at 10,000 rpm for 20 min. The transparent solution was discarded and the pellets of silver nanoparticles were collected. The pellets were dried in the oven at 45°C to 50°C to obtain constant weight.

UV-Vis spectra analysis: The bioreduction of Ag^+ ions in each sample was observed periodically followed by the dilution with 2 ml of double distilled water. This AgNP synthesized mixture was scan, in the range of 200 nm to 800 nm wavelengths under UV-visible spectrophotometer the spectra were recorded at the intervals of 1 min to 10 min and the distilled water was used as a baseline [14].

RESULTS

Phytochemical and Chemical Analysis

Further this prepared extract was subjected to phytochemical and chemical analysis where in Total Phenol Content (TPC), Total Flavonoid Content (TFC) was determined. Chemical analysis for the same extracts was done *via* GC-MS and FTIR to determine the phytochemical components within the extracts. (Figures 1-4) [15-18].

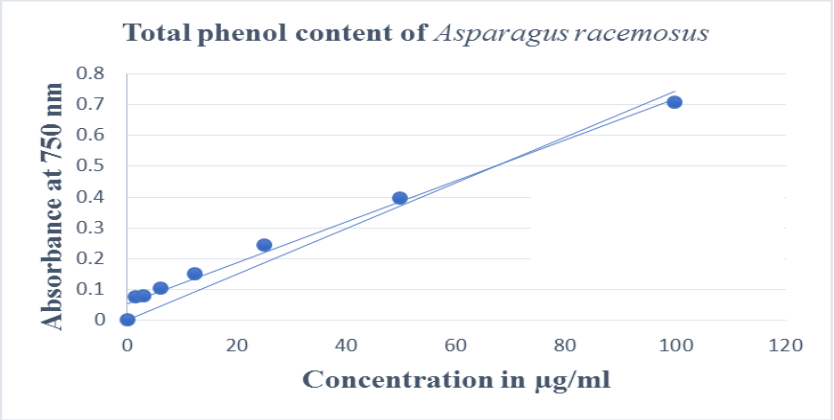

Figure 1: Total Flavonoid content (TFC). Note: y=0.0074x, R2=0.9507, y=0.0066x+0.0537.

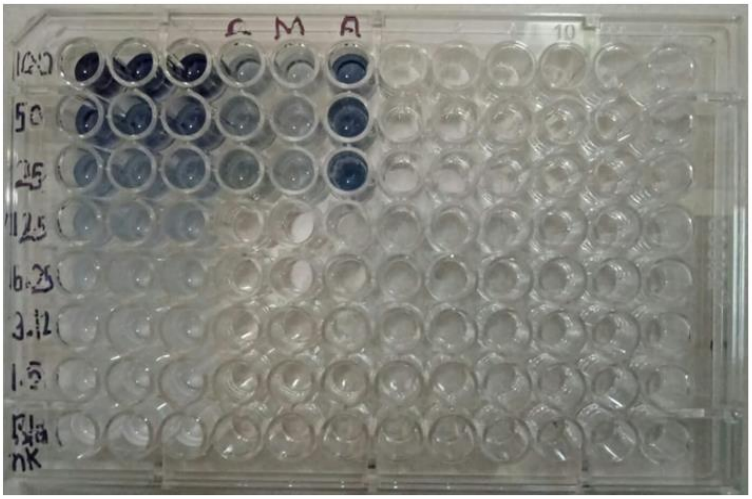

Figure 2: Total phenol content (96 well plate).

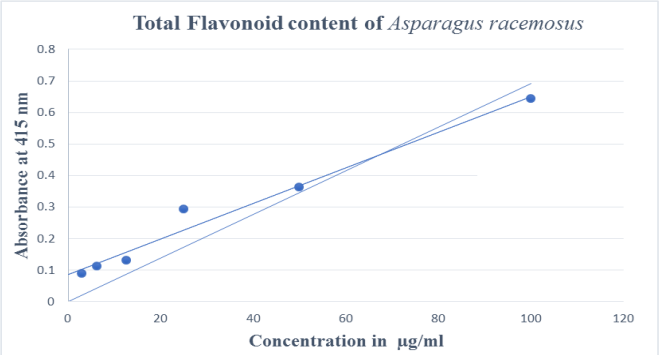

Figure 3: Total Flavonoid Content (TFC). Note: y=0.0069x, R2=0.8762, y=0.0057x+0.0855.

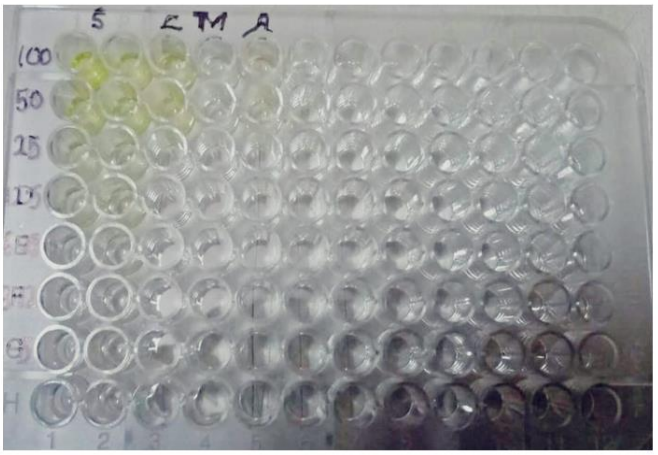

Figure 4: Total Flavonoid content (96 well plate).

GC-MS: In GC-MS analysis sample was first diluted at 1% and further 1 micro-litre is loaded which got rinse two times and then passes through the column of 30 m. Cut time for elimination of solvent peak was maintained for 2-3 minutes and then based on retention time of components respective compounds from the sample was predicted. Further the sample passed on to MS *via* interface and the components within the samples gets fragmented and based on the number of ions and charge on it mass to charge ratio was determined (Table 1). The obtained charge to mass ration for each compound was further compared with the NIST (MS) data which has 62,000 patterns and accordingly possible components are predicted. (Figures 5 and 6) [19].

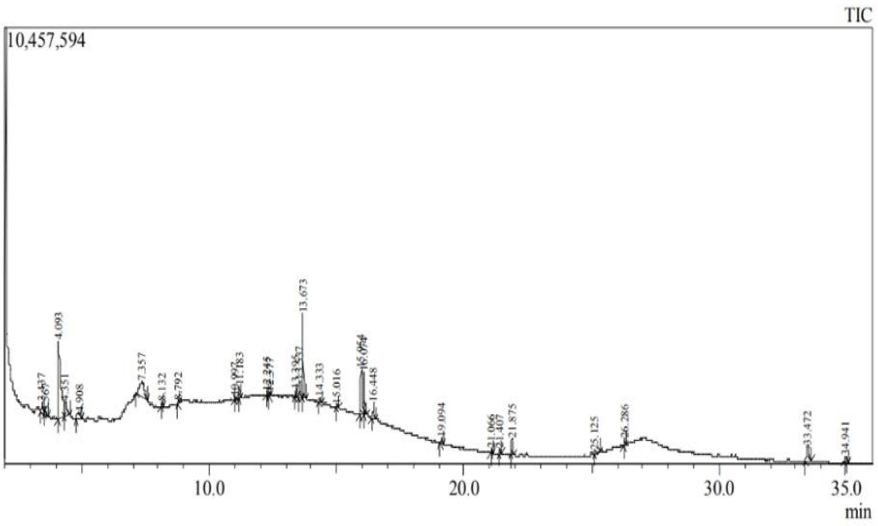

Figure 5: Chromatogram of GCMS for *Asparagus racemosus*.

Table 1: Peak report of GCMS for *Asparagus racemosus.*

Peak report TIC					
Peak#	**R. Time**	**Area**	**Area%**	**Height**	**Name**
1	3.437	993877	2.24	256219	4H-Pyran-4-one,2,3-dihydro-3,5-dihydroxy-6-method
2	3.567	310639	0.7	65981	4-Benzoyloxy-1-morpholinocyclohexene
3	4.093	13009012	29.31	1884526	5-Hydroxymethylfurfural
4	4.351	1897915	4.28	340768	Maltol
5	4.908	1150851	2.59	146154	5-Amino-1-pentanol,N2O-diacetyl-
6	7.357	4636709	10.45	352208	Sucrose
7	8.132	241014	0.54	120468	Phenol,2,4-bis(1,1-dimethylethyl)
8	8.792	134675	0.3	75196	Dodecanoic acid
9	10.997	106258	0.24	61367	Phenol,Pentachloro-
10	11.183	546434	1.23	284274	Tetradecanoic acid
11	12.245	202245	0.46	94671	1,2-Benzenedicarboxylic acid,bis(2-methylpropyl)
12	12.377	171652	0.39	83721	Pentadecanoic acid
13	13.395	403274	0.91	177622	Dibutyl Phthalate
14	13.537	1381278	3.11	412102	n-Hexadecanoic acid
15	13.673	4927791	11.1	2016249	n-Hexadecanoic acid
16	14.333	320832	0.72	130818	Oxazole,2,5-diphenyl-
17	15.016	197292	0.44	87347	Heptadecanoic acid
18	15.954	3595305	8.1	1100923	cis-Linoleic acid
19	16.074	2875304	6.48	1036883	Oleic Acid
20	16.448	1460028	3.29	396968	Octadecanoic acid
21	19.094	489026	1.1	152089	Phenol,4,4-(3-ethenyl-1-propene-1,3-diyl)bis-,€
22	21.066	270601	0.61	95132	Androsta-1,4,6-triene-3,17-dione
23	21.407	341178	0.77	118718	n-Nonadecanoic acid
24	21.875	1060978	2.39	394761	Bis(2-ethylhexyl)phthalate
25	25.125	162050	0.37	68623	9-Octadecenamide,(Z)-
26	26.286	476276	1.07	157788	supraene
27	33.472	2255890	5.08	363561	stigmasterol
28	34.941	770522	1.74	144039	Gamma-Sitosterol

FTIR: The FTIR spectrum of plant extracts (prepared in Ethanol) of *Asparagus racemosus.* Chromatogram of FTIR for *Asparagus racemosus.* The data on the peak values and the probable functional groups (obtained by FTIR analysis) present in plant extracts (prepared in Ethanol) of *Asparagus racemosus* (Figure 7 and Table 2).

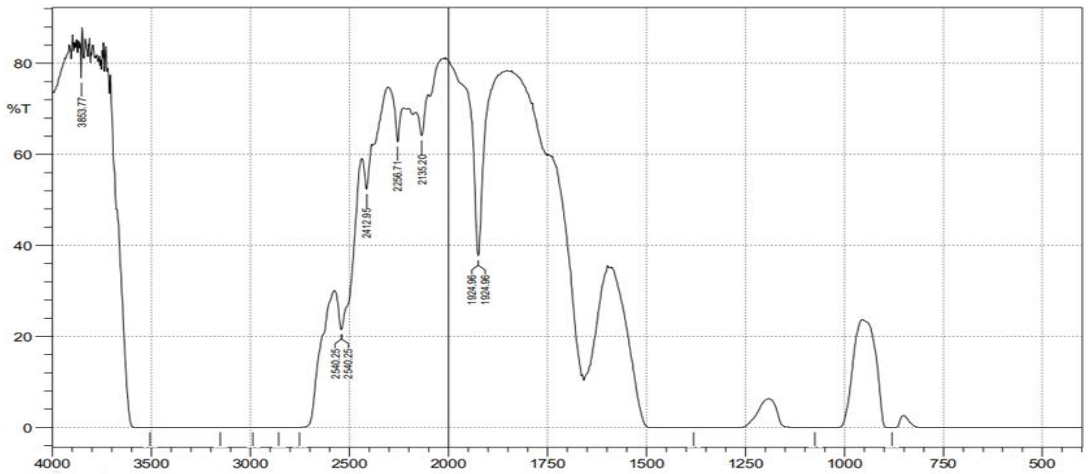

Figure 7: Chromatogram of FTIR for *Asparagus racemosus*.

Table 2: FTIR spectral peak values and functional groups obtained for *Asparagus racemosus* extract.

Peak	Functional group	Types of compound
879.54	C-H Group	Aromatic compounds
1074.35	C-O Group	Alcohol, ethers, anhydrides, carboxylic acids compounds
1381.03	C-H Group	Alkanes compound
1924.96	C=C Group	Allene compounds
1924.96	C=C Group	Allene compounds
2135.2	C≡C Group	Alkynes compounds
2256.71	C≡C Group	Alkynes compounds
2412.95	O-H Group	Alcohol, phenol compounds
2540.25	O-H Group	Alcohol, phenol compounds
2540.25	O-H Group	Alcohol, phenol compounds
2752.42	O-H Group	Alcohol, phenol compounds
2858.51	C-H Group (weak)	Alkane, Aldehyde compounds
2987.74	C-H Group	Alkane compounds
3153.61	O-H Group	Alcohol, phenol compounds
3508.52	O-H Group	Alcohol, phenol compounds
3852.77	O-H Group	Alcohol, phenol compounds

Synthesis and Characterization of SNPs

UV-Vis Spectra analysis: Synthesis of silver nanoparticles was done by photo reduction method. This AgNP synthesized mixture was scan, in the range of 200 to 800 nm wavelengths under UV–visible spectrophotometer *via* multimode reader the spectra were recorded (Figures 8 and 9).

Figure 8: Dried silver nanoparticles.

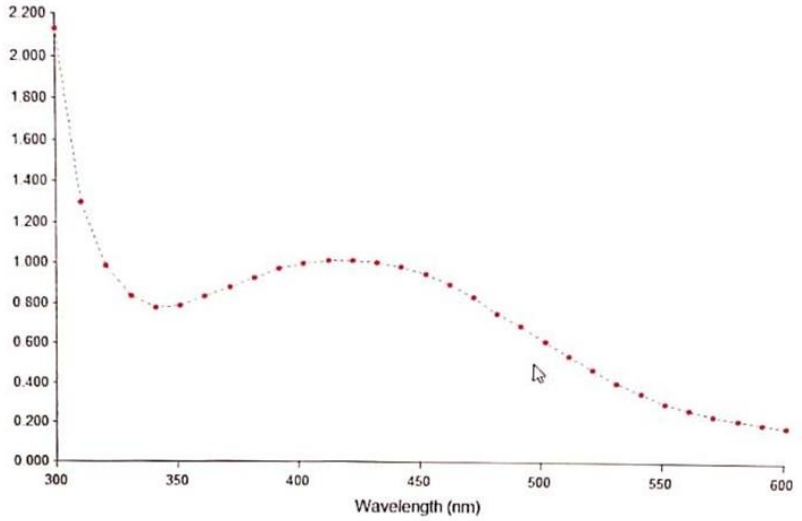

Figure 9: Characterization of SNPs by UV-Visible spectra analysis.

DISCUSSION

Asparagus racemosus is a species of asparagus common throughout Sri Lanka, India and the Himalaya. *Asparagus racemosus* belongs to family Liliaceae and commonly known as Satawar, Satamuli, Satavari. The management of burn wound victims is often considered a difficult task due to frequent infections. Moreover, the treatment with conventional antibiotics is associated with the upsurge of drug resistant bacterial strains. *A. racemosus* species have been widely used as antioxidant, antiulcer, antimicrobial agent in Indian system of traditional medicine for treatment of various infection. Thus, current study aims to determine the effect of *A. racemosus* plant extract on the clinical isolates to overcome the above difficulties in treatment of burn wounds. Further for the study, *A. racemosus* was collected, authenticated, and subjected for the preparation of extracts using ethanol as solvent for extracting phyto constitutes present in them. Further in Phytochemical analysis of *Asparagus racemosus* the quantitative estimation of phenolic and flavonoid content was done which showed the presence of good amount of phenolic and flavonoid content. In chemical analysis the plant extract was analysed *via* GC-MS and FTIR. The GC-MS study showed many Phytochemicals such as Hydroxypyranone, Tetradecanoic acid, Octadecadienoic acid, n-Hexadecanic acid, 5-Amino-1-pentanol, n-Nonadecanol-1, etc. compounds that shows antimicrobial, antitumor, anticonvulsant, anti-inflammatory and ty-rosinase inhibitory effects, Anticancer, Antioxidant, Hypercholesterolemic, Lubricant, Nematicide, Hepatoprotective, Antiacne, 5 alpha reductase inhibitors, Antiarthritic agent, emulsifying agents in dry-cleaning soaps, wax removers, cosmetics, paints and insecticides, surfactant properties. Certain phytosterol was also found such as Stigmasterol, Gamma-Sitosterol which act as anticancer, anticoronary in function. The FTIR spectroscopic studies revealed different characteristic peak values with various functional compounds in the extracts. The FTIR analysis of ethanol plant extracts of *A. racemosus* confirmed the presence of alcohols, phenols, alkanes, carboxylic acids, alkenes, aromatics, esters, ethers, allenes compounds etc., which showed major peaks [20].

CONCLUSION

In silico approach yield was also determined to be useful. Among the phytoconstituents obtained from GC-MS, majority has showed the useful ADMET profiles i.e. they showed the exhibition of properties such as human intestinal absorption, blood brain barrier penetration, human oral bioavailability, $caco_2$, and plasma binding properties. Among the 24 compounds obtained 13 compounds has not shown aquatic toxicity, all 23 compounds except maltol has showed Ames mutagenesis negative. Further in ProTox tool that determines the toxicity of the chemical compounds has resulted that among 24 compounds, 17 compounds are in the range of 4-5 which indicates that they are non-toxic in nature. thus, from the above results obtained *via* the phytochemical analysis by *In vitro* as well as *In silico* approach, it can be concluded that these compounds from the plant extract of *Asparagus racemosus* can be effectively used in the various formulation of drug in order to treat burn wounds and overcome the difficulties raised in the treatment of burn wounds. However further studies need to be done i.e. *via* docking where in structure of this component can be docked on the target site of antibiotics that are being currently used in the hospitals. These antibiotics being used are day by day gaining the resistance from the bacterial strains isolated from the clinical burn isolates. If the docking of these components on the respective target site eventually succeeds definitely the plant extract can be used as effective alternate to synthetic medicines. Moreover, if this succeeds, this will provide a resource which is relatively cheaper, safer to use and with less or no side effects. Further based on the results obtained the nanoparticle synthesis was done using plant extracts and characterization was done. On the basis of the result obtained it can be concluded that the approach to photosynthesis of AgNPs from root extract of *A. racemosus* is simple cost effective, precise and eco-friendly method. It is a fast and convenient method as well as has great medical importance and on the other hand AgNPs arepotentially biocompatible, the result described here will be useful in biomedical applications as well as in the area of further research activities in nanobiotechnology. A colour change from light yellow to yellowish to reddish to colloidal during the formation of silver nanoparticles is confirmed by uv–vis spectroscopy and however further characterization has to be done using NTA, XRD, FTIR, and TEM analysis.

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