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ROLE OF MICROWAVE ASSISTED EXTRACTION FOR ISOLATION OF SAPONINS FROM *SAPINDUS MUKORROSAI* AND SYNTHESIS OF ITS STABLE BIOFUNCTIONALIZED SILVER NANOPARTICLES AND ITS HYPOLIPIDAEMIC ACTIVITY

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ABSTRACT: This research emphasis on microwave assisted extraction of fruits of *Sapindus mukorrosai*. Aqueous fruit extract is subjected for the isolation of saponins, focusing only on higher molecular weight of saponins using dialysis membrane. On treatment of aqueous solution of silver nitrate with isolated saponin, stable silver nanoparticles were rapidly formed. The green synthesized silver nanoparticles were characterized by UV-vis spectroscopy, scanning electron microscope (SEM), energy dispersive X-ray (EDX). The SEM confirms the synthesis of square shape of nanocrystalline particles with the size range of 60-90 nm. The presence of saponins has confirmed by FTIR spectra. These nanoparticle has been subjected for hypolipidaemic activity in animal model. It showed significant reduction in total cholesterol, triglyceride level, HDL and VLDL parameters.

INTRODUCTION: *Sapindus mukorossi* Gaertn., a member of the family Sapindaceae, is commonly known by several names such as soapnut, soapberry, washnut, reetha, aritha, dodan and doadni. The fruit is valued for the saponins (10.1%) present in the pericarp and constitutes up to 56.5% of the drupe known for inhibiting tumor cell growth.[1] The major constituents of Sapindus mukorossi fruit are saponins (10%-11.5%), sugars (10%) and mucilage.[2]

Saponins are secondary metabolites synthesized by many different plant species [3]. Their name is derived from Latin word "sapo" meaning soap, due to their surfactant properties which allows forming stable soap-like foam upon shaking in aqueous solution.[4, 5]

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They have many medicinal uses including, microbial, anti-tumor, anti-insect[6] hepatoprotective, haemolytic [7], and anti-inflammatory activities. They also decrees blood cholesterol level and may be used as adjuvant in vaccines.[8-14] In addition, saponins are used in preparation of soaps, detergents, fire extinguishers, shampoos, beer and cosmetic.[15] Many saponins exhibit haemolytic activity, have a bitter taste and are toxic to fish.[16]

They are large molecules and contain a hydrophobic part, composed of a triterpenoid (30 carbon atoms) or steroid (27 carbon atoms with a 6-ring spirostane or a 5-ring furostane skeleton) backbone and a hydrophilic part consisting of several saccharide residues, attached to the hydrophobic scaffold through glycoside bonds. Terpenoid and steroid saponins are usually found in dicotyledonous and monocotyledonous plants, respectively. [4, 9, 17] Saponins with a glucuronic acid moiety at C-3 of oleanolic acid are found in the flowers, while saponins with a glucose moiety at the same position are found in the roots [18]. Due to their amphiphilic nature, saponin molecules form

micelles in aqueous solutions. The size, shape, and structure of the saponin micelles depend on their plant origin, pH, temperature and the presence of electrolyte in the solution.[9]

Saponins are a large family of structurally-related compounds of steroid or triterpenoid aglycone (sapogenin) linked to one or more oligosaccharide moieties by glycosidic linkage. The aglycone, or sapogenin, may contain one or more unsaturated C-C bonds. The oligosaccharide chain is normally attached at the C3 position (monodesmosidic), but many saponins have an additional sugar moiety at the C2,6 or C2,8 position (bidesmosidic).[2]

Sapindus mukorossai possess anti-inflammatory activity, piscicidal activity, anti-platelet aggregation activity, molluscicidal activity, fungicidal activity, tyrosinase inhibition and free radical scavenging, anxiolytic activity, hepatoprotective activity, anti-cancer activity, anti-*Trichomonas* activity, spermicidal activity, insecticidal activity, anti-bacterial activity etc.[19]

In many previous reports, green synthesis of nanoparticles was done in conical flasks utilizing H_2O as a solvent.[20] The bioreduction of silver from silver ions due to their surface Plasmon resonance (SPR) has been achieved using plants such as tamarind [21] *Helianthus annus, Basella alba, Cinnamomum camphora,* [22, 23] alfalfa, [24] *Capsicum annuum,* [25] *Avena sativa,* [26] *Azardirachta indica,* [27] *and Pelargonium graveolens,*[28] have been reported for the presence of saponin. Due to the various medicinal properties of *Sapindus mukorossai*, thus commercial production of the saponin is quite high and thereby shows potential for the commercial synthesis of nanoparticles. In this study we describe for the first time, the isolation of higher molecular weight saponins from *Sapindus mukorossai* and its formulation of silver nanoparticle.

MATERIAL AND METHOD:

Plant collection& chemicals:

Chemicals used for synthesis of silver silver nitrate ($AgNO_3$) (Sigma-Aldrich, MO). Fresh fruits of *Sapindus mukorossai* collected from local market the Kanpur were used as a saponin source for green nanoparticle synthesis. Equipment including an Orbitol shaker (Labquake shaker; Labindustries, Berkeley, CA) and a Perkin Elmer spectrophotometer (Boston, MA) were used initially for nanoparticle synthesis. Nanoparticles size was confirmed by using TEM (1200 EX; JOEL USA, Inc, Peabody, MA) and SEM (FEI Quanta FEG 200; FEI Company, Hillsboro, OR) with high and low vacuum. The functional groups in the synthesized nanoparticles were confirmed by using FTIR (6700 spectrum; Thermo Nicolet, Madison, WI).

Extraction and isolation of saponins:

Fresh fruits of *Sapindus mukorossai* were cleaned, dried properly, extracted with double distilled water (200 ml) in a 250 ml wide neck borosil conical flask and were exposed to microwave for 180 s. Then the raw extract obtained was filtered in hot condition with 10 micron mesh to remove fibrous impurities. This extract is treated with n-butanol, kept for some time. Finally di ethyl ether was added into it to isolate saponins from the extract. This extract is passed through dialysis membrane into the water solution for getting high molecular weight of saponins.

FTIR spectroscopy analysis of saponin: [29]

A carefully weighed quantity of saponin was subjected to FTIR analysis. It was centrifuged at 10,000 rpm for 15 minutes and the pellet was washed three times with 20 mL of deionized water. The resulting purified suspension was completely dried and ground with KBr pellets and analyzed by FTIR.

Green synthesis of silver nanoparticle using saponin: [30]

In the typical synthesis of silver nanoparticles, 1mL of saponin was treated with 9mL of 1mM silver nitrate solution and kept in room temperature. Subsequently the synthesis of silver nanoparticles was initially identified by brown colour formation and further monitored by measuring UV-vis spectra of the reaction mixture. Silver nitrate and saponin fraction reaction mixture was kept at room temperature and formation of nanoparticles was recorded at different functional times. Influences of silver nitrate concentration were performed to find their effects on nanoparticles synthesis.

Characterization of Synthesized Silver Nanoparticles:

Energy dispersive X-ray (EDX) analysis:

Analysis through energy dispersive X ray (EDX) spectrometers confirmed the presence of elemental silver signal of the silver nanoparticles. The vertical axis displays the number of X ray counts whilst the horizontal axis displays energy in keV. This analysis revealed that the nanostructures formed were solely of silver.

Scanning electron microscopy:

The prepared silver nanoparticle were characterized using high resolution SEM analysis. The samples were prepared by simple drop coating of the suspension of silver onto an electric clean glass and allowing the solvent to evaporate. The samples were left to dry completely at room temperature.

UV- VIS adsorbance spectroscopy analysis:

The bioreduction of nanoparticles was monitored periodically by UV-vis spectroscopy. The samples used for analysis were diluted with 2ml deionized water and measured by the UV-vis spectrum at different interval time intervals. The UV-vis spectrometric readings were recorded at a scanning speed of 200 to 750 nm.

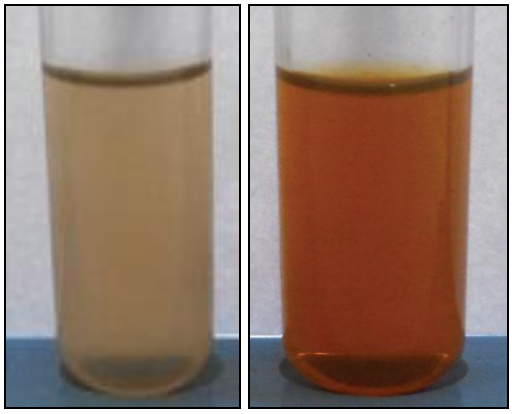


FIG.1: SHOWING GREEN SYNTHESIS OF SILVER NANOPARTICLE USING SAPONIN ON THE BASIS OF CHANGE IN COLOUR

Evaluation of hypolipidemic activity:

The fractionated part of saponin were administered in the form of suspension p.o. at two dose levels of 200 and 400 mg/kg body weight to rats as 1 % (w/v) sodium carboxy methyl cellulose (SCMC) for evaluating their hypolipidemic and antihyperlipidemic effects. Triton was purchased from SD Fines Chemical and Kits for lipid profile analyses were purchased from a local supplier, viz; TC and HDL (Span Diagnostics), TG (Diassays, Germany). Male Albino rats of the Sprague-Dawley strain (average weight 150 g), bred and maintained in our Institute's Animal House were used for the study after prior scrutinization and approval from Institutional Animal Ethical Committee (IAEC). Antihyperlipidemic potential of LSFE in Triton-induced hyperlipidemic rats and in normal rats was evaluated as per the method described by Moss [31], Vogel [32] and Hirsch, et al [33].

Effects in Triton-induced hyperlipidemic rats:

The antihyperlipidemic effects of the above extracts were evaluated in 45 Triton-induced hyperlipidemic rats starved for 18 hr. The rats were divided into 9 groups of 5 each and then injected, ip, with Triton at a dosage of 100 mg/kg body weight except rats of Group I, which served as normal vehicle treated and Group II as control treated with 1% SCMC, po. Group III were treated daily with saponin nanoparticles in two divided doses of 200 and 400 mg/kg, respectively immediately after the Triton injection by ip administration. Blood samples were collected after 6, 24 and 48 hr of Triton injection to evaluate the lipid profile.

Data analysis:

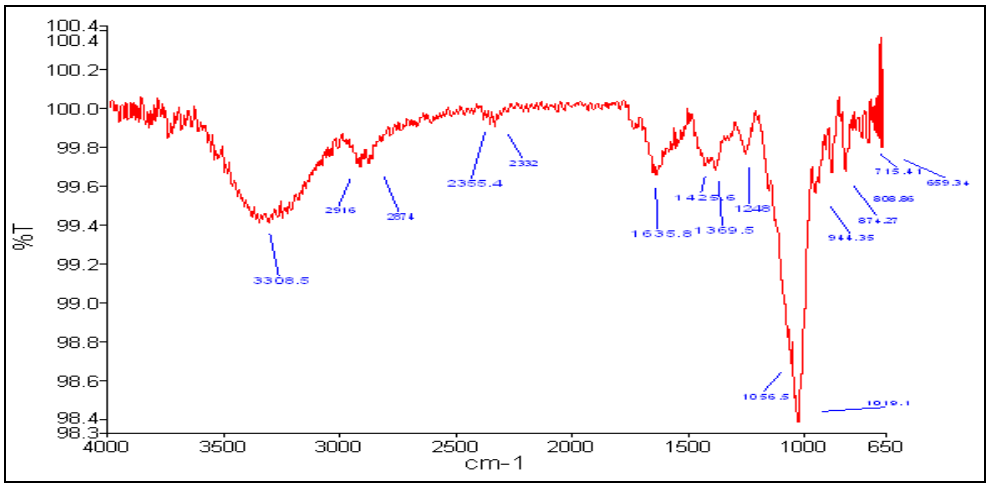
Data were statistically analyzed as mean ±SE and expressed as non-significant $P > 0.05$, just significant $P < 0.05$ and significant $P < 0.01$ as the case may be using ANOVA followed by Dunnett's t - test and unpaired t- test with Welch correction[34]. Total cholesterol was determined by one-step method of Wybenga and Pillegi 11 based on the reaction between cholesterol and cholesterol reagent (ferric chloride, ethyl acetate and sulphuric acid). HDL from blood was determined by a two-step method i.e. initial separation of HDL from blood using a precipitating agent and then the precipitated HDL was determined by using colorimetric reaction with cholesterol reagent [35].

Triglyceride (TG) was determined colorimetrically by enzymatic reaction using glycerol-3-phosphate oxidase. Enzymatic splitting of lipoprotein lipase along with reaction between 4-aminoantipyrin, 4-chlorophenol and H_2O_2 under catalytic action of peroxidase generates quinimine which is used as

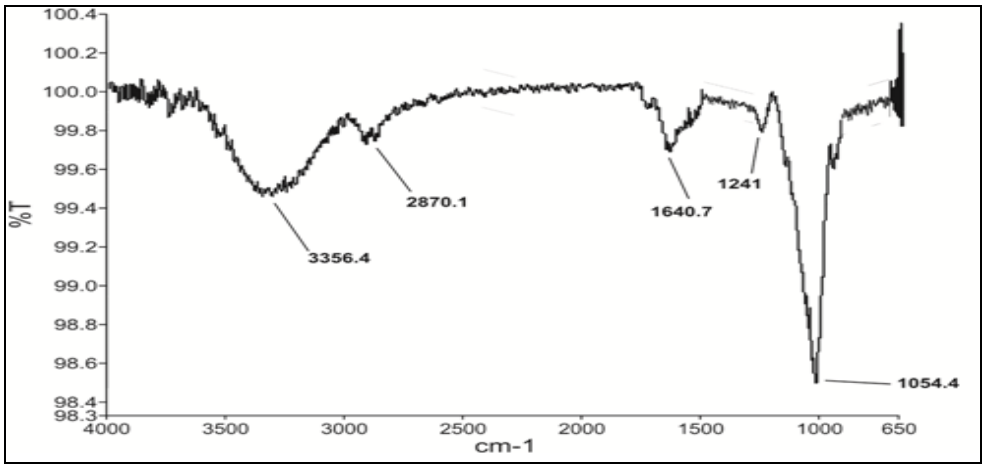
internal indicator in this colorimetric determination. LDL was determined by using Friedelwald's formula LDL=TC − (TC − VLDL) [36]

RESULTS AND DISCUSSION:

Confirmation of saponin existence by IR analysis [29] The FTIR 80% ethanol revealed the presence of broad and strong signal oh hydroxyl group (3308 cm^{-1}), C-H (2874 cm^{-1}), signal olefinic (C=C) (1635.8 cm^{-1}), strong absorption signals sulfate group (C-O-C) (1248 cm^{-1}) were assigned in figure. This spectra has been matched with the standard spectra of Quillaija saponin (Elaheh Amini et al). Existence of –OH, -C=O, C-H and C=C bands in absorption peak of FTIR spectrum was characteristic of saponin.



GRAPH1: IR SPECTRA OF ISOLATED SAPONIN OF *SAPINDUS MUKORROSAI*



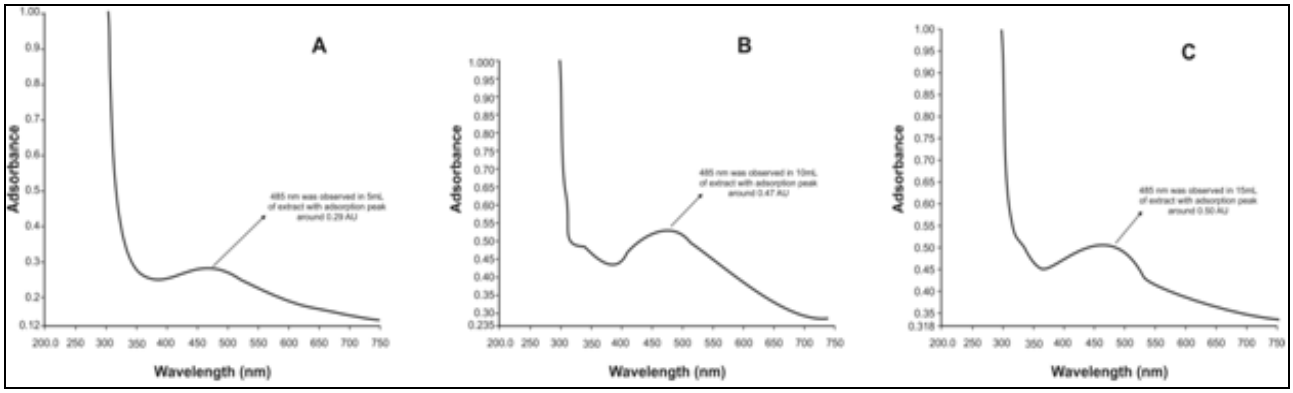
GRAPH 2: IR SPECTRA OF STANDARD SAPONIN OF *QUILLAIJA SAPONARIA*

UV- vis adsorbance spectroscopy analysis:

The kinetics of reduction of aqueous silver ions during reaction with the saponin of *Sapindus mukorossai* were easily analyzed by the UV Visible spectroscopy. The optical adsorption UV-vis spectra of silver nanoparticles produced by different amounts (5, 10, 15 ml) of the dried saponin of *Sapindus mukorossai.* The result show the production of silver nanoparticles within 18 hours after silver ions came in contact with the saponin. The addition of isolated saponin solution to the aqueous $AgNO_3$ solution resulted in the colour of the solution changing to dark brown due to the SPR. The colour change has been noted because of the production of silver nanoparticles. (**Fig.1**)

Graph 3 clearly depicts the strong adsorption peak at 0.50 (AU) over increasing time of reaction by gently reducing $AgNO_3$ with 15 ml of saponin fraction at around 485 nm. A weak adsorbance peak was observed at 0.29 AU in 5 ml of saponin fraction and mild reduction of adsorption of silver ions at 0.47 AU with 10 ml of saponin fraction. The evolution of UV-Vis spectrum of synthesized silver nanoparticles have an increasingly sharp

adsorbance peak identified at around 485 nm in adsorption spectra of silver nanoparticles. Adsorption peak was not clear in the 5 ml and 10ml of saponin fraction part, but a sharp peak was observed at the higher dosage level 15 ml and peak resolution was also very clear.



GRAPH 3: UV- VIS ADSORPTION SPECTRA OF SILVER NANOPARTICLE AFTER BIOREDUCTION KINETICS OF THE REACTION OF *SAPINDUS MUKOROSSAI* WITH AQUEOUS SILVER IONS AT 5 ML (A), 10 ML (B), 15 ML (C) IN THE CONCENTRATION RANGE OF 2000 TO 750 NM WITH DIFFERENT TIME INTERVALS.

Scanning electron microscopy:

The SEM image showing high density Ag-NPs synthesized by using the isolated saponin fraction further confirmed the development of silver nanostructures. Finally nanoparticles obtained are square in shape and of uniform size. Higher magnification showed the average diameter of these square nanoparticles to be about 60- 90 nm. (**Fig.2**). The SEM analysis of silver nanoparticle supports the results of Govindaraju et al in *S. platensis.*

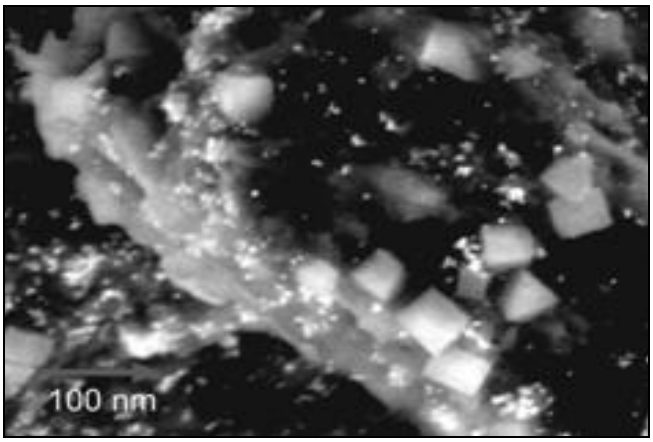
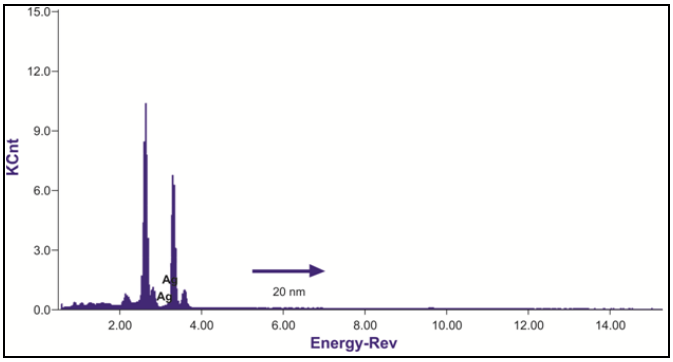


FIG.2: SEM IMAGE OF SYNTHESIZED NANOPARTICLE

Energy dispersive X-ray (EDX) analysis:

The EDAX pattern thus clearly shows that the silver nanoparticles are crystalline in nature by the reduction of silver ions. The EDAX analysis obtained in the present study confirmed the presence of silver nanoparticles of *Sapindus mukorossai* and mostly showed showed strong signal energy peaks for silver atoms in the range 2-4 keV. (**Graph 4**)



GRAPH 4: EDAX PROFILE OF SILVER NANOPARTICLE RESULTING FROM THE EXPERIMENT BY USING 15 ML OF SAPONIN FRACTION OF *SAPINDUS MUKOROSSAI.*

Hypolipidemic activity:

Saponins nanoparticles showed decrease in blood lipids in hyperlipidemic rats when compared with their respective controls. It showed significant ($P<0.05$) decrease in TC, TG and LDL along with an increase in HDL level, dose-dependently at the tested dose (400 mg/kg body weight), when administered in Triton-induced hyperlipidemic rats (**Tables 1** and **2**).

Saponins also act as antihyperlipidemics by one of the following mechanisms as by binding with cholesterol in intestinal lumen, so that cholesterol is less readily absorbed or bile acids causing reduction in its extra hepatic circulation and increasing metabolism of cholesterol to sterols

through their fecal excretion. Increase in bile acid excretions offset by enhanced synthesis from cholesterol in the liver consequently lowers the plasma cholesterol. Saponins are also reported to increase the lipoprotein lipase activity (LPL), which is considered as helpful in faster removal of free fatty acid from circulation that causes in turn a decrease in total cholesterol [37, 38, 39]. Soluble dietary fiber contents are having the beneficial effect in the promotion of bile acid formation and their excretion in the stool or in the blockage of cholesterol absorption [40].

TABLE 1: EFFECT OF SAPONIN ON TOTAL CHOLESTEROL AND TRIGLYCERIDE LEVELS IN TRITON-INDUCED HYPERLIPIDEMIC RATS

Groups/ treatment	Cholesterol			Triglycerides		
	6 hr	24 hr	48 hr	6 hr	24 hr	48 hr
control	18.70±0.63	25.33±0.88	31.00±1.06	75.50±1.17	80.66±0.71	80.08±0.45
Triton WR 1339 control	56.20±1.23	77.10±2.08	87.12±3.36	62.30±1.08	71006±1.07	88.04±2.19
Standard drug fenofibrate	49.50±1.86	82.30±1.19*	49.05±1.29	36.20±1.05	53.10±1.62	41.08±1.52**
silver nanoparticle	19.76±2.12	30.91±1.01*	25.58±2.11	74.02±3.13	81.11±3.14	76.31±2.90

Values are expressed as mean ± SEM for 6 animals in each group.
* = P<0.05, ** = P<0.01, * = significant, ** = more significant, P<0.05(comparison of group II with group III and IV)

TABLE 2: EFFECT OF SAPONIN ON HDL AND LDL IN TRITON-INDUCED HYPERLIPIDEMIC RATS [VALUES, EXPRESSED AS mg/dl, ARE MEAN ±SE OF 6 ANIMALS IN EACH GROUP]

Groups/ treatment	HDL			LDL		
	6 hr	24 hr	48 hr	6 hr	24 hr	48 hr
I. Control	82.70±4.15	115.53±4.23	87.98±2.68	15.10±0.23	16.13±0.14	16.68±0.62
II. Triton WR 1339 control	58.00±2.36	91.06±3.76	110.04±3.21	32.09±1.02	46.00±1.15	87.06±2.01
III. Standard drug fenofibrate	28.90±2.72	59.01±1.26	38.08±0.29**	40.00±1.66	36.22±1.19*	66.75±1.98
IV. Silver nanoparticle	89.80±3.30	96.50±3.88**	93.15±4.47	15.26±0.37	18.10±0.15**	15.01±0.05

Values are expressed as mean ± SEM for 6 animals in each group.
* = P<0.05, ** = P<0.01, * = significant, ** = more significant, P<0.05(comparison of group II with group III and IV)

CONCLUSION: This research is an attempt to prove that saponin could be an excellent bioreductant and can be easily isolated from plant source for synthesis of silver nanoparticle. This method seems to be ecofriendly and therefore this protocol could be used for the rapid production of silver nanoparticles. These saponin nanoparticles shows significant effect in hypolipidaemic activity.

REFERENCES:

1. Tanaka O, Tamura Y, Masuda H, Mizutani K. Application of saponins in food and cosmetics: saponins of Mohova Yucca and Sapindus mukorossi Gaertn, saponins used in food and agriculture. New York: Plenum Press; Waller GR and Yamasaki K; 1996. p.1-11.
2. Francis G, Kerem Z, Makkar H, Becker K. The biological action of saponins in animal systems: a review. Br J Nutr. 2002; 88:587-605.
3. Arif T, Bhosale JD, Kumar N, Mandal TK, Bendre RS, Lavekar GS, Dabur R. Natural products – antifungal agents derived from plants. J Asian Nat Prod Res. 2009; 11(7):621–638.
4. Faizal A, Geelen D. Saponins and their role in biological processes in plants. Phytochem Rev. 2013; 12:877–893.
5. Hosamath PV. Evaluation of antibacterial activity of *Litsea glutinosa*. Int J Pharmaceut Appl. 2011;2(1):105-114.
6. Dixon RA, Sumner LW. Legume natural products: Understanding and manipulating complex pathways for human and animal health. Plant Physiol. 2003; 131:878–885.
7. Bink A, Pellens K, Cammue BPA, Thevissen K. Anti-biofilm strategies: How to eradicate *Candida* biofilms? The Open Mycology J. 2011;5:29-38.
8. Hu X, Neil SJ, Cai W, Tang Z. Nitric oxide mediates elicitor-induced saponins synthesis in cell cultures of *Panax ginseng*. Funct Plant Biol. 2003;30:901-907.
9. Stanimirova R, Marinova K, Tcholakova S, Denkov ND, Stoyanov S, Pelan E. Surface rheology of saponin adsorption layers. Langmuir. 2011;27:12486–12498.
10. Oboh HA, Omofoma CO. The effects ofheat treated lima beans (*Phaseolus lunatus*) on plasma lipids in hypercholesterolemic rats. Pak J Nutr. 2008;7(5):636-639.

11. Bhargava D, Shivapuri JN, Kar S, Pandit BR, Sidhiqie A, Upadhyay A, Thakur S, Mondal KC. Evaluation of antigonorrhoeal activity of saponins extract of *Sapindus mukorossi* Gaertn. Res J Pharm Biol Chem Sci. 2012;3(2):459-470.
12. Meesapyodsuk D, Balsevich J, Reed DW, Covello PS. Saponin biosynthesis in *Saponaria vaccaria.* cDNAs encodingbamyrin synthase and a triterpene carboxylic acid glucosyltransferase. Plant Physiol. 2007; 143:959–969.
13. Soetan KO, Oyekunle MA, Aiyelaagbe OO, Fafunso MA. Evaluation of the antimicrobial activity of saponins extract of *Sorghum Bicolor* L. Moench. Afr J Biotechnol. 2006; 5:2405-2407.
14. Jyothi TC, Sindhu Kanya TC, Appu Rao AG. Influence of germination on saponins in soybean and recovery of soy sapogenol I. J Food Biochem. 2007; 31:1–13.
15. Bhargava A, Shukla S, Ohri D. *Chenopodium quinoa*-an Indian perspective. Ind Crop Prod. 2006; 23:73– 87.
16. Ceyhun Sezgin AE, Aruk N. Determination of saponin content in Turkish Tahini Halvah by using HPLC. Adv J Food Sci Technol. 2010; 2(2):109-115.
17. Mert-Turk F. Saponins versus plant fungal pathogens. J Cell Mol Biol. 2006; 5:13-17.
18. Hostettmann K, Marston A. Chemistry and pharmacology of natural product: Saponins. University press.UK. 1995; 18.
19. Aparna Upadhyay & D.K. Singh, Pharmacological effects of *Sapindus mukorossi*, Rev. Inst. Med. Trop. Sao Paulo September-October, 2012,54(5):273-280,
20. Narayanan KB, Sakthivel N. Coriander leaf mediated biosynthesis of gold nanoparticles. *Mater Lett*. 2008; 62:4588–4590.
21. Kumar V, Yadav SK. Plant-mediated synthesis of silver and gold nanoparticles and their applications. *J Chem Technol Biotechnol*. 2009; 84:151–157.
22. Parashar V, Parashar R, Sharma B, Pandey AC. *Parthenium* leaf extract mediated synthesis of silver nanoparticles, a novel approach towards weed utilization. *Digest Journal of Nanomaterials and Biostructures*. 2009; 4:45–50.
23. Narayanan KB, Sakthivel N. Coriander leaf mediated biosynthesis of gold nanoparticles. *Mater Lett*. 2008; 62:4588–4590.
24. Huang J, Li Q, Sun D, et al. Biosynthesis of silver and gold nanoparticles by novel sundried *Cinnamomum camphora* leaf. *Nanotechnology*. 2007; 18:105–104.
25. Gardea-Torresdey JL, Gomez E, Peralta-Videa JR, Parsons JG, Troiani H, Jose-Yacaman M. *Alfalfa* sprouts a natural source for the synthesis of silver nanoparticles. *Langmuir*. 2003:19: 1357–1361.
26. Li S, Shen Y, Xie A, et al. Green synthesis of silver nanoparticles using *Capsicum annuum L*. extract. *Green Chem*. 2007; 9:852–858.
27. Armendariz V, Herrera I, Peralta-videa JR, et al. Size controlled gold nanoparticle formation by *Avena sativa* biomass use of plants in nanobiotechnology. *J Nanopart Res*. 2004; 6:377–382.
28. Sharma NC, Sahi SV, Nath S, Parsons JG, Gardea-Torresdey JL, Pal T. Synthesis of plant- mediated gold nanoparticles and catalytic role of biomatrix-embedded nanomaterials. *Environ Sci Technol*. 2007; 41: 5137–5142.
29. Elaheh Amini, Mohammad Nabiuni, Javed Baharara, Kazem Parivar, Javed Asili, Hemolytic and cytotoxic effects of saponin like compounds isolated from Persian Gulf brittle Star (Ophiocoma erinaceus), Journal of coastal Life Medicine, 2014,2(10),762-768.
30. M. Vanaja, K. Paulkumar, G. Gnanajobitha, S. Rajeshkumar, C. Malarkodi, and G. Annadurai, Herbal Plant Synthesis of Antibacterial Silver Nanoparticles by *Solanum trilobatum* and Its Characterization, Hindawi Publishing Corporation International Journal of Metals Volume 2014,
31. Moss J N & Dajani E Z, Antihyperlipidemic agents, *In screening methods in pharmacology*, edited by R A Turner, P A Hebben (Academic Press, New York) 1971, 121.
32. Vogel G & Vogel W H, Influence of lipid metabolism, in Drug Discovery and Evaluation: Pharmacological assay (Springer- Verlag, Berlin) 1997, 604.
33. Hirsch R L & Keller A, the pathogenesis of hyperlipidemia induced by means of surface active agents. II failure of exchange of cholesterol between the plasma and liver in rabbits given Triton WR 1339, J Exp Med, 104 (1956)1.
34. Wybenga D R, Pileggi V J, Dirstine P H & Di Giorgio J, Direct manual detremination of serum total cholesterol with a single stable reagent, Clin Chem, 16 (1970), 980.
35. Demacker P N m, Vos Jansees H E, Jansen A P & Von't Laar A, Evaluation of the dual- precipitation method by comparison with the ultracentrifugation method for measurement of lipoproteins in serum, Clin Chem, 23 (1977) 1238.
36. Friedwald W T, Levy R I & Priedrickson D S, Estimation of concentration of LDL cholesterol in plasma without preparation or ultracentrifugation, Clin Chem, 18 (1972) 449.
37. Guimarese P R, Galavao A M P, Batista, C M, Azovedo G S, Oliveira R D, Lamounier R P, Freire N, Barros A M D, Sakurai E, Olivera J P, Vieira E C & Aalvarez J I, Eggplant (*Solanum melongena*) infusion has modest and transitory effect on hypercholesteremic subjects, Braz J Med Biol Res, 33 (2000) 1027.
38. Sidhu G S & Oakenful D G, A mechanism for the hypocholesterolaemic activity of saponins, Br J Nutrition, 55 (1986) 643.
39. Sidhu G S & Oakenful D G, Could saponins be a useful treatment for hypocholesterolaemia? Eur J Clin Nutr, 44 (1990) 79.
40. Prasannakumar G, Sudheesh S, UshaKumari B & Valsa A K J, A comparative study on the hypolipidaemic activity of eleven different pectins, Food Sci Technol, 34 (1997) 103.

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