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Biological evaluation of nano synthesized particles from the root extract of violet tree (Securidaca longepedunculata)

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Abstract

The research was carried out to evaluate the biological activities of nano synthesized particles from the root extract of violet tree (securidaca longepedunculata). In this study, the root sample was subjected to extraction by increasing polarity index of solvents (hexane, ethyl acetate, acetone and water). The synthesized nanoparticles were characterized and tested for their antimicrobial activity for E. coli, Klebsiella pneumonia and Shigella sp. UV-visible spectrophotometer and Fourier Transform Infrared (FTIR) Spectrometer were used to analyze the sample extracts and the nano synthesized particles. Colour changes were observed for each extract indicating nanoparticles formation. The UV-Vis spectra did not show a distinctive peak. The FTIR analysis indicated the presence of active functional groups for all the samples analyzed. The antibacterial test showed that the plant has a minimal zone of inhibition and that the nano synthesized particles showed a higher zone of inhibition for all the samples except for the water extract. The synthesized nanoparticles via biological method could be further explored for use in drug discovery.

Keywords: Biological activities, drug discovery, nanosynthesized particles, therapeutic properties, Securidaca longepedunculata

Full length article * Corresponding Author, e-mail: terversase@gmail.com

1. Introduction

Plants have great potential uses, especially as traditional medicine and pharmaceutical drugs. A large proportion of the world's population depends on traditional medicine because of the scarcity and high costs of orthodox medicine [1-2]. Natural products play a dominant role in the development of novel drugs for the treatment and prevention of diseases [3-4]. Plants are remarkable factories of bioactive chemical compounds referred to as phytochemicals [20]. Bioactive plant compounds have served as templates for several synthetic drugs and as precursors. They are used in the production of semisynthetic drugs [3]. The need to screen plants for pharmaceutical uses is particularly urgent in the light of rapid deforestation and the concurrent loss of biodiversity throughout the world [5]. There is a serious need to develop new antimicrobial agents that are very effective in the treatment of diseases caused by pathogens and other

ailments, particularly because antimicrobial resistance has become a global health challenge [21].

The plant Securidaca longepedunculata belongs to the family Polygalaceae. Its leaves are oblanceolate and obtuse at apex. The flowers are purple or blue in colorat and the seeds are winged [6]. In northern Nigeria, the Nupe and the Hausa tribes utilize S. longepedunculata ethnomedicinally as a remedy for numerous human and animal ailments [7]. According to Dapar et al. (2007), [8] the aqueous extracts of its roots are used as psychopharmaceutical agents. It is also used as a sexual boost for men [9]. This plant is also used for the treatment of every conceivable ailment such as headache, rheumatism, tuberculosis, cancer, venereal diseases, diabetes as well as an abortifacient [10] and probably the reason why the Hausas (from Nigeria) refer to it as "uwar magunguna" (the mother of all medicines). The flowers are in abundance at the beginning of the rainy season, sweetly scented, bright purple or violet racemes and the fruit is winged. The fruit is more or less a round nut, somewhat heavily veined

occasionally smooth, bearing a single, oblong, rather curved, membranous wing up to 4 cm long; purplish-green when young, becoming pale, straw-colored when mature. It is a beautiful flowering tree with potential as an ornamental plant for use in beautifying parks, gardens and along the roadsides [22].

S. longepedunculata has in recent times been reported to be used to cure a variety of ailments. It has been described as the "mother" of all plants in Northern Nigeria and the root bark extract is reported to possess nephrotoxic and hepatotoxic activities. The aqueous root bark extract of the plant has also been shown to have significant prooxidant activity. NSPs have a broad antibacterial effect on a range of Gram-negative and Gram-positive bacteria and antibiotic-resistant bacteria strains [22].

Antimicrobial efficacy of NSPs depends on their size and concentration. Normally, a high concentration leads to more effective antimicrobial activity, while particles of small sizes can kill bacteria at a lower concentration. Apart from size and concentration, shape also influences the antimicrobial efficiency of NSPs. Sadeghi et al (2017) [11] investigated the antimicrobial activity of different nano silver shapes, which included silver nanoplates, silver nanorods, and silver nanoparticles, on Staphylococcus aureus and E. coli. They found that silver nanoplates had the best antimicrobial activity. It has also been reported that NSPs combined with various antibiotics have better antimicrobial effects than NSPs or antibiotics alone. It is widely accepted that NSPs can anchor to and subsequently penetrate the bacterial cell wall, thereby causing structural change of the cell membrane and increasing cell permeability, leading to cell death.

In this study, a green method has been developed for the synthesis of silver nanoparticles (AgNPs) where AgNPs were synthesized using water-based facile hydrothermal method. Silver nitrate (AgNO₃) and polyvinylpyrrolidone (PVP) were used as precursor and reducing agents respectively to produce AgNPs [12]. This research work therefore tried to look at the biological efficacy of *S. longepedunculata* by evaluation of nano synthesized particles from the root extract of the violet tree.

2. Materials and methods

2.1. Sample collection and identification

S. longepedunculata root extract was collected from wildlife park forest behind Solomon Lar amusement park, Jos, Plateau state Nigeria, with the help of Mr. Gabriel Sunday, a staff member and identified at the Federal College of Forestry Jos, Plateau state-Nigeria. The method by Baru *et al* (2014) was used with slight modifications. The sample was chopped into small sizes and dried at room temperature for six (6) weeks. The dry sample was crushed into smaller sizes using mortar and pestle and was stored in a clean dry polythene bag for further analyses.[23]

2.2. Preparation of plant extract

The dried crushed sample (200g/800ml) was extracted successively with n-hexane, ethylacetate, acetone and water using a sequential extraction method. The solvent was left in contact with the plant for 72 hours, in each case. The extract was decanted and allowed to stand, after which it was filtered through a Whatman No. 42 filter paper. The extracts were concentrated using a rotary evaporator to recollect the solvents leaving the extract with a little amount of the solvents. The extract was transferred into a preweighed beaker and placed on a water bath at 40°C until a plastic form of the extract (soft extract) was obtained.[25]

2.3. Synthesized nanoparticles

Synthesis silver nanoparticles (AgNPs) were synthesized using a facile hydrothermal method. The synthesis was carried out based on the previous study by Zhou and Wang (2012) [13] with some modifications. In this process, all the materials were added together at room temperature and the reaction was carried out by heating in a closed hydrothermal system [12]. 2g of concentrated crude extracts were dissolved in 20mL of its solvent in a 2:20 ratio (at room temperature). 100ml of 0.001M silver nitrate solution was measured into a beaker and placed on a magnetic stirrer hot plate, 10mL of the extract solution was transferred into the beaker and left to stir for one hour. The extract solution was transferred and mixed into an aqueous solution of the silver ion complex. Colour changes were observed concerning phenomena surface plasmon resonance (SPR). This was done separately with each type of plant extract [14].

2.4. Determination of antibacterial activity

Antimicrobial activities of the crude extracts and their synthesized colloidal nano synthesized particles were determined using the agar well diffusion assay method reported by Prasad et al., (2011) [15]. Approximately 20 mL of molten and cooled media (MHA) was poured into sterilized Petri dishes. The plates were left overnight at room temperature to check for sterility. The test organisms were grown in selected broth for 24 h. About 1 mL broth culture of each test organism containing approximately 1×105 cfu/mL was used to prepare bacterial lawns. Agar wells of 5 mm diameter were prepared with the help of a sterilized stainless-steel cork borer labelled as A and B. "A" well was loaded with 30 µL of colloidal nano synthesized particles suspended "hydrosols" and "B" well was loaded with 30 µL of positive control drugs (ciprofloxacin) used as positive controls. The plates containing the test organism and colloidal nano synthesized particles were incubated at 37°C for 24-48 hrs. The plates were examined for evidence of zones of inhibition, which appeared as a clear area around the wells. The diameter of such zones of inhibition was measured using a meter ruler, and the mean value for each organism was recorded and expressed in millimetre [14,24].

3. Results and discussion

We observed from this study that an aqueous root extract of S. longipenduculata for both the crude and in nanosynthesized particles showed potential the management of microbial infections. But the nanosynthesized particles seem to have higher antimicrobial activities than the crude extracts on a general note except for the water crude extracts. The extracts seemed to possess both bacteriostatic and bactericidal effects against. E. coli, Klebsiella and Shigella sp., suggesting that a therapeutic concentration could be attained in a living host. The high antimicrobial activity of the aqueous root extract observed is similar to the findings of Ajali and Chukwurah (2004), [16] which attributed these activities to the high content of flavonoids. Besides, Abubakar et al. (2005) [17] and Okoli et al. (2006) [18] reported that S. longipenduculata possess antitrypanosomal and anti-inflammatory activities. respectively. This might be because the extracts can exhibit remarkable activities. Antifungal activities of the aqueous extracts appeared to be more effective than acetone extracts

since aqueous solutions could extract a wide variety of active component as compared to acetone. It has been reported that the aqueous root and acetone extracts yielded alkaloids, cardiac glycosides, flavonoids, saponins, tannins, volatile oils, terpenoids and some steroids [19]. Flavonoid together with the other secondary metabolites identified in some related study has been severally reported to show curative activity against diverse pathogens, used traditionally such as analgesic antimicrobial, anti-tumour headache, venereal diseases, constipation and coughs. The antibacterial test showed that the plant has a minimal zone of inhibition and that the nanosynthesized particles showed a higher zone of inhibition for all the samples except for the water extract.

The UV-visible spectra were performed to identify the compounds containing σ -bonds, π -bonds and lone pair of electrons, chromophores, and aromatic rings. The results showed that no maximum peaks were clearly identified in the curves this is because of the variable functional groups found in the extracts.

N-HEXANE		ETHYL ACETATE		ACETONE		WATER	
Wavelength	Absorbance	Wavelength	Absorbance	Wavelength	Absorbance	Wavelength	Absorbance
(nm)	(A)	(nm)	(A)	(nm)	(A)	(nm)	(A)
380	1.201	460	2.553	480	2.602	500	2.585
420	1.091	500	2.292	520	1.979	540	2.347
460	0.975	540	2.081	560	1.660	580	2.137
500	0.919	580	2.161	600	1.416	620	1.910
540	0.921	620	1.213	640	1.238	660	1.684
580	0.927	660	1.947	680	1.416	700	1.516
620	0.915	700	1.876	720	0.910	740	1.360
660	0.909	740	0.186	760	0.800	780	1.223
700	0.882	780	1.845	800	0.720	820	1.103
740	0.851	820	1.830	840	0.657	860	1.000
780	0.837	860	1.818	880	0.609	900	0.906
820	0.823	900	1.801	920	0.556	940	0.827
860	0.807	940	1.785	960	0.505	980	0.755
900	0.780	980	1.788	1000	0.476	1020	0.694
940	0.758	1020	1.764	1040	0.450	1060	0.643

Table 1. Result of U.V-Vis showing wavelength and absorbance of the samples of S. longepedunculata

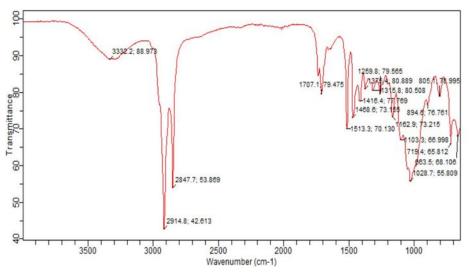


Figure 1. F.T.I.R spectrum of S. longepedunculata for n-Hexane Extract showing wavelength for specific functional groups

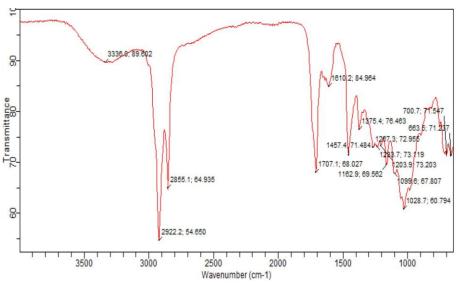


Figure 2. F.T.I.R spectrum of S. longepedunculata for Ethylacetate extract showing wavelength for specific functional groups

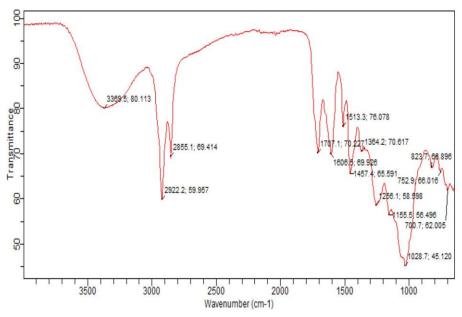


Figure 3. F.T.I.R spectrum of S. longepedunculata for acetone Extract showing wavelength for specific functional groups

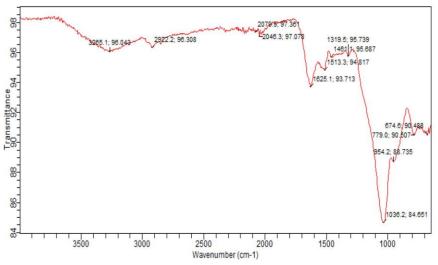


Figure 4. F.T.I.R spectrum of S. longepedunculata for n-Hexane Extract showing wavelength for specific functional groups

Wavelength of	Appearance	Group	Compound class	
Absorbance				
3332.2	Medium	N-H stretching	Aliphatic primary amine	
2914.8	Medium	C-H stretching	Alkane	
2847.7	Medium	C-H stretching	Alkane	
1707.1	Strong	C-O stretching	Conjugated aldehyde	
1513.3	Strong	N-O stretching	Nitro compound	
1259.8	Strong	C-O stretching	Aromatic ester	
1375.4	Medium	O-H bending	Phenol	
1315.8	Strong	S=O stretching	Sulfone	
894.6	Strong	C=C bending	Alkene	
805.1	Medium	C=C bending	Alkene	
719.4	Strong	C=C bending	Alkene	

 Table 2. Showing functional group identified in the n-hexane extract of S. longepedunculata

Table 3. Showing functional group identified in the Ethylacetate extract of S. longepedunculata

Wavelength of	Appearance	Group	Compound class
Absorbance			
3336.0	Medium	N-H Stretching	Secondary amine
2922.2	Medium	C-H Stretching	Alkane
2855.1	Medium	C-H Stretching	Alkane
1707.1	Strong	C=O Stretching	Conjugated aldehyde
1610.2	Strong	C=C Stretching	α,β-unsaturated ketone
1375.4	Medium	O-H bending	phenol
1267.3	Strong	C-O Stretching	Alkyl aryl ether
1233.7	Medium	C-N Stretching	Amine
1203.9	Strong	C-O Stretching	Tertiary alcohol
1099.6	Strong	C-O Stretching	Secondary alcohol
700.7	Strong	C=C bending	Alkene

Wavelength of	Appearance	Group	Compound class
Absorbance			
3369.5	Medium	N-H Stretching	Aliphatic primary amine
2922.2	Medium	C-H Stretching	Alkane
2855.1	Medium	C-H Stretching	Alkane
1707.1	Strong	C=O Stretching	Conjugated aldehyde
1606.5	Medium	C=C Stretching	Conjugated alkene
1513.3	Strong	N-O stretching	Nitro compound
1364.2	Medium	O-H bending	Phenol
1256.1	Strong	C-O Stretching	Aromatic ester
700.7	Strong	C=C bending	Alkene

Table 4. Showing functional group identified in the acetone extract of S. longepedunculata

Table 5. Showing functional group identified in the water extract of S. longepedunculata

Wavelength of	Wavelength of Appearance		Compound class
Absorbance			
3265.1	Strong, broad	O-H stretching	Carboxylic acid
2922.2	Medium	C-H Stretching	Alkane
2046.3	Strong	N=C=S Stretching	Isothiocyanate
1625.1	Medium	C=C Stretching	Conjugated alkene
1513.3	Strong	N-O stretching	Nitro compound
1319.5	Strong	S=O Stretching	Sulfone
1036.2	Strong	S=O Stretching	Sulfoxide
674.6	Strong	C=C bending	alkene

Table 6. Showing Bacterial Zone of Inhibition by different samples of S. longepedunculata

		Hex	Hexane Eth		vlacetate Ace		tone	Wa	Water	
S/No	Bacteria	CE	NP	CE	NP	CE	NP	CE	NP	
1	E. coli	3.0	4.2	-	-	3.0	5.0	5.9	5.0	
2	Shegella spp	-	4.3	2.3	2.3	4.0	5.0	8.3	5.0	
3	Klebsiella spp	1.0	-	1.0	2.4	3.3	4.3	5.2	4.3	
3	Klebsiella spp	1.0				3.3			5.2	

CE= Crude Extract NP= Nanosynthesized particles

Zone of inhibition is taken in mm

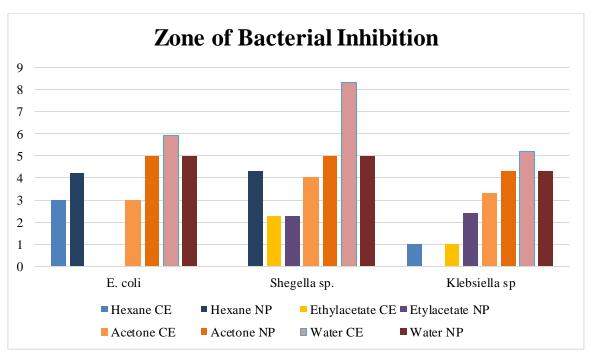


Figure 5. Zone of Inhibition by Crude extract and Nanosynthesized particles

4. Conclusion

We have presented the biological potentials of the root extracts of S. longepedunculata in the present study, using different solvents for the extraction. The crude extracts have been synthesized and evaluated for antimicrobial activities and a comparative study between the crude extracts of the plant and the nano synthesized particles from the various solvent extracts studied. These were evaluated for antimicrobial activities against selected resistant pathogens, and we observed that the synthesized particles had better antimicrobial potentials than the crude extracts. Antimicrobial resistance has become a global health challenge and of course a public concern [21, 23], therefor it has become imperative to search for more, better and effective antimicrobial agents. It is hope that this work will serve as lead to drug discovery and development of potent antimicrobial drugs to solve the global menace of antibacterial resistance.

5. Conflict of interest

The authors declare no conflict of interests, all the authors also contributed equally to the work.

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