








GREEN SYNTHESIS, CHARACTERIZATION AND LARVICIDAL ACTIVITY OF CU/Ni BIMETALLIC NANOPARTICLES USING FRUIT EXTRACT OF PALMYRA PALM

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ABSTRACT

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The mixture of Cu²⁺/Ni²⁺ solution was reduced to Cu/Ni BMNPs using fruit extract of Palmyra palm as a green reducing agent. The formation of Cu/Ni BMNPs noticed based on the visual change in color of the reaction mixture at room temperature from sky-blue to light blue within 15 min. UV-Visible and Fourier Transform Infra Red spectrophotometers were applied to investigate the Optical properties as well as the interaction between the formed Cu/Ni hybrid nanoparticles and the raw fruit extract respectively. These Cu/Ni BMNPs were evaluated for larvicidal potency against first, second and third instars of *Culex quinquefasciatus*. The larvicidal studies were carried out at the concentration range of 5–50 mg/L (Cu/Ni BMNPs) and the %mortality increases with increase in concentration for all the instars. The result showed good activity where the lethal concentration values herein for the three instars were LC₅₀=14.75, LC₉₀=83.96; LC₅₀=18.25, LC₉₀=258.83; LC₅₀=18.50, LC₉₀=331.50 mg/L respectively. Therefore, Cu/Ni BMNPs could be a potential future nanolarvicide for *Culex quinquefasciatus*.

Contribution/Originality: The study is one of a very few studies which have investigated fast green synthesis of Cu/Ni bimetallic nanoparticles and its larvicidal activity against larvae of *Culex quinquefasciatus*.

1. INTRODUCTION

Mosquitoes are vectors for many tropical and subtropical diseases that cause destructive effects to human health [1, 2]. These effects include malaria, filariasis, yellow fever and encephalitis [3, 4]. *Culex quinquefasciatus* was reported to transmit filariasis, Japanese encephalitis and other mosquito borne diseases. However, in the quest for synthesizing nanolarvicides, various physical, chemical and biological routes were employed [5, 6]. The chemical methods such as chemical vapour deposition, sol-gel technique, aerosol technology, sono chemical method and photochemical reduction are expensive and associated with vector resistance, environmental pollution have potential health hazards. This necessitated the significance in the search for plant-based insecticide which in contrast serves as an alternative green approach that are simple, rapid, eco-friendly and target effective [7]. The Plant materials are rich in active pharmacological ingredients whose usage are not only limited to reducing agents, but also act as capping agents for NPs which in turn intensify their biomedical efficacies [8].

Nanotechnology involves the production and design of nanoparticles which helps in reducing the toxicities of the metals [9]. Monometallic and hybrid bimetallic nanoparticles (NPs) possess unique properties which find many applications such as catalysis, photonics, in solving engineering problems that cannot be solved with conventional alloyed metals [10-12]. Nanoparticles were also used as bactericidal [13] larvicidal agents [14] core-shell bimetallic nanoparticles also have high sensitivity and have application to be used in electrode modification [15]. Individual metallic nano solution has peculiar properties. Thus, a combination of two metallic nanoparticles will obtain special characteristics or increase the special properties of them due to synergy [12, 16].

Palmyra palm belonging to the genus *Borassus* is a tropical plant commonly known as the Palm. Saranya and Vijayakumar [17] reported the phytochemical synthesis of the fruit extract. The research revealed the presence of flavonoids, terpenes glycosides, saponins, phenols and steroid among others but showed negative test for proteins and suggested the possible extraction of these compounds in drug development. This work is aimed at the eco-friendly synthesis of nanosized particles of hybrid, silver and nickel from the secondary metabolites of Palmyra palm fruit, partial characterization using Ultraviolet - Visible Spectrophotometer and Fourier Transform Infrared Spectroscopy to confirm the formations as well as evaluation of larvicidal activity.

2. MATERIALS AND METHODS

2.1. Collection of Plant Materials

Fresh fruits of Palmyra palm were collected from Shongom Local Government, Gombe State. The fruit sample was identified and authenticated at the Taxonomy Section in Gombe State University.

2.2. Preparation of Aqueous Plant Extract

The fruit sample was thoroughly washed under running tap water and rinsed severally with distilled water followed by sun-drying to remove residual moisture. The dried materials were cut into fine pieces and 40 g of it was weighed and dispersed in 800 ml of sterile distilled water in a 1000 ml glass beaker and boiled at 200 °C for 15 min and was allowed to cool. The solution was filtered through Whatman No. 1 filter paper.

2.3. Green Synthesis of Cu/Ni Bimetallic Nanoparticles (Cu/Ni Bmnps)

Two hundred milliliters of the filtrate obtained was measured in a measuring cylinder and transferred into a 2000 ml beaker. A 500 ml each of 10^{-2} M $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$ and 10^{-2} M $\text{NiCl}_2 \cdot 6\text{H}_2\text{O}$ were mixed in a standard Erlenmeyer flask and added gradually to the boiling fruit extract with occasional stirring using a glass rod 20 minutes. There was a visual color change to light blue throughout, nanoparticles were allowed to settle and decanted and dried in an oven.

2.4. UV-visible Spectroscopy Analysis

The bioreduction process of Cu/Ni BMNPs in aqueous solution was measured by sampling of 1 ml aliquot compared with 1 ml of distilled water used as blank and subsequently measuring the UV-visible spectrum of the solution. UV-visible spectrum was monitored on Cary Series UV-Vis spectrophotometer Agilent Technology, operated within the wavelength range of 200 to 800 nm.

2.5. Infra-Red Spectral Analysis

The dried Cu/Ni BMNPs were taken and made into a pellet using pelletizer and hydraulic presser then placed into FTIR sample holder and introduced into the infrared spectrophotometer for analysis. The FT-IR spectrum was recorded in the range of $450\text{-}4000\text{cm}^{-1}$ using PerkinElmer Spectrum version 10.03.09.

2.6. Larvicidal Bioassay

A solution of 100 mg/L of the NPs was prepared. From the stock suspension, 5, 10, 20, 25 and 50 mg/L were prepared subsequently by serial dilution. A 100 mL of these concentrations were placed together with twenty larvae each of first, second and third instars in plastic cups. A test of each concentration against each instar was repeated two times. After 24 h exposure, larval mortality was recorded.

3. RESULTS AND DISCUSSION

3.1. UV-Visible Spectroscopic Analysis of Cu/Ni BMNPs

The formation of Cu/Ni BMNPs was first noticed based on the visual change in color of the reaction mixture at room temperature from sky-blue to light blue within 15 min. This was followed by UV-Vis spectroscopy frequently used to characterize the BMNPs. The UV-Vis absorption spectrum of the synthesized Cu/Ni BMNPs is shown in Figure 1. The maximum absorption peak was shown at absorbance of 0.07 with corresponding wavelength (λ_{\max}) at 500 nm. The change in color of the reaction mixture was also due to surface Plasmon resonance phenomenon which provides a convenient indication of the formation of Cu/Ni BMNPs. Less is known on the characterization of these NPs using this equipment, however, the λ_{\max} is consistent with 510nm reported by Ramasamy, et al. [18] and Gopinath, et al. [7] both for Au/Ag BMNPs.

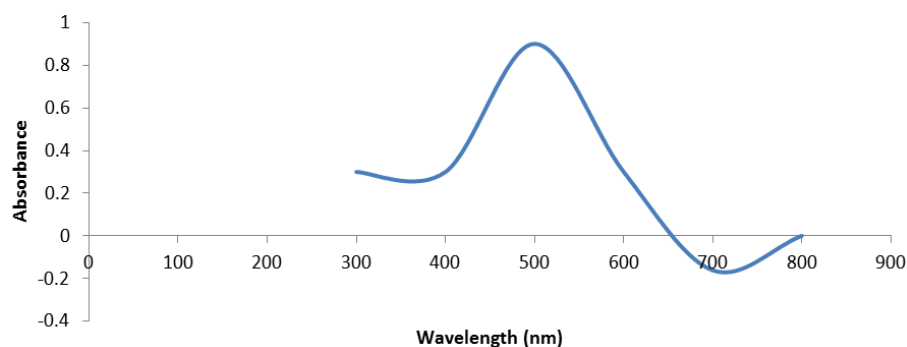


Figure-1. UV-Visible spectra of Cu/Ni bimetallic nanoparticles.

3.2. Fourier-Transform Infrared Spectroscopy

FT-IR spectroscopy was applied to identify the functional groups involved in the bio-fabrication of copper-nickel nanoparticles. The FTIR spectrum of the Cu/Ni bimetallic nanoparticles is shown in Figure 2. It showed absorptions at 3426.76cm^{-1} due to O-H stretching which may be due to phenol, a peak at 2924.37 cm^{-1} due to hydrogen attached to a saturated carbon, a C=C absorption at around 1632.89 cm^{-1} , a C-O absorption at 1112.62 cm^{-1} and a cluster of peaks in the aromatic region around $500\text{-}600\text{ cm}^{-1}$ plausibly and likely due to metal-carbon bonds.

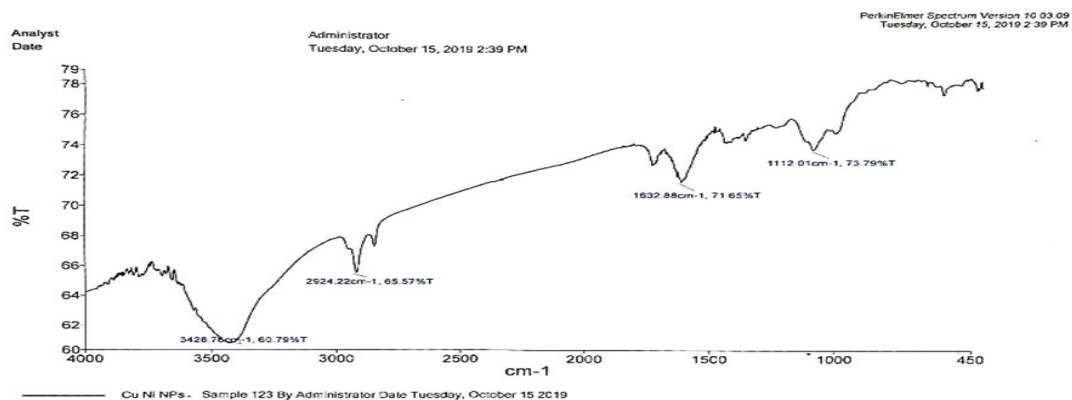


Figure-2. FT-IR spectra for Cu/Ni bimetallic nanoparticles.

3.3. Larvicidal Activity Result

Larvicidal effects of different concentrations of Cu/Ni bimetallic particles exposed to instar I, II and III larvae of *Culex quinquefasciatus* was studied by recording the mortality rates after a day. The mortality rate increased with increasing concentration and attenuated with increasing larval stage. Probit result evaluated using statistical software for 5, 10, 20, 25 and 50 mg/L dose exposure is depicted in Table 1 and represented in form of a histogram for better comparative understanding, Figure 3. The lethal concentrations obtained were ($LC_{50}=14.75$, $LC_{90}=83.96$) for instar I, ($LC_{50}=18.25$, $LC_{90}=258.83$) for instar II and ($LC_{50}=18.5$, $LC_{90}=331.5$) for third instars respectively. Hassanain, et al. [19] synthesized Cu nanoparticles using Lantana Camara leaves extract and its larvicidal activity was tested on the 4th instar of Anopheles multicolor. The LC_{50} and LC_{90} were 12.6 and 18.4 mg/L respectively. This result is comparable with the current. Also, Elango, et al. [20] studied larvicidal activity of Ni nanoparticles using aqueous extract of Coir dust against 4th instar of *Aedes aegypti* in which the LC_{50} and LC_{90} were 259.24 and 446.99 mg/L respectively. Similarly, Elango, et al. [21] studied larvicidal activity of Ni-Pd nanoparticles using aqueous extract of Coir dust against 4th instar of *Aedes aegypti* in which the LC_{50} and LC_{90} were 288.88 and 483.06 mg/L respectively. It could be concluded that the studied larvicidal activities of Ni NPs and Ni-Pd bimetallic NPs does not yield significant result as compared to Cu/Ni BMNPs of the current study. Recently, Cu-Zn and Cu-Ag bimetallic NPs were synthesized using leaf extract of *Oscimum sanctum* (Linn) and its larvicidal activities were evaluated on the 3rd instar of Anopheles stephesis. For, Cu-Zn BMNPs, the LC_{50} and LC_{90} were 444.734 and 1077.953 mg/L respectively. While for the Cu-Ag BMNPs, the LC_{50} and LC_{90} were 888.792 and 192.93 mg/L respectively, Minal and Prakash [22]. It is quite obvious to say that the toxicity of both Cu-Zn and Cu-Ag BMNPs on larvae of anopheles stephesis is low as compared to Cu/Ni BMNPs in the current study. However, in our previous report, Ag-Co bimetallic nanoparticle was synthesized using root extract of palmyra plant and the larvicidal activity was studied on *Culex quinquefasciatus* larvae. The LC_{50} and LC_{90} were 5.237 and 49.240 mg/L for first instar, 9.310 and 94.969 mg/L for second instar as well as 13.626 and 105.542 mg/L for third/fourth instar respectively, Danbature, et al. [23]. Though within the same range, our previous report yielded good activity than the current study.

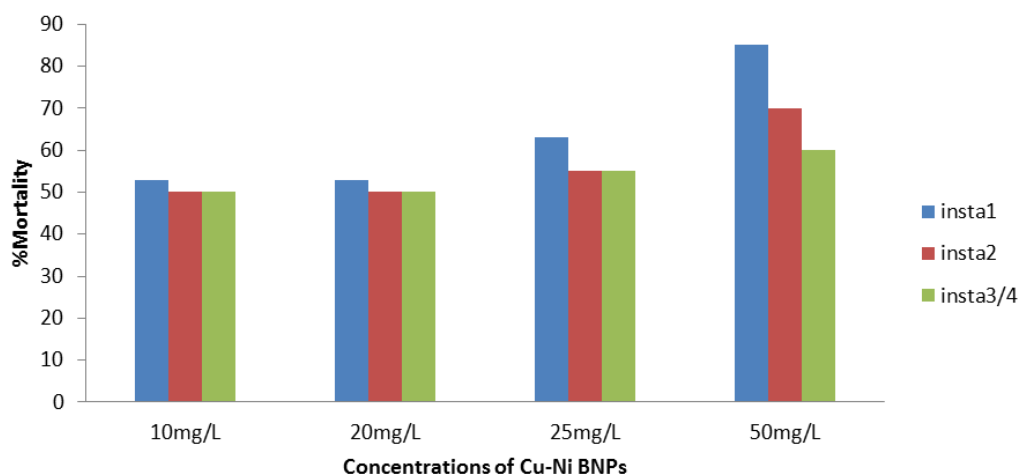


Figure-3. Chart for Cu/Ni BMNPs larvicidal result.

4. CONCLUSION

Fruit extract of Palmyra palm were used as substrates in the synthesis of Cu/Ni bimetallic nanoparticles through a cost effective green route which is associated with less toxicity and does not involve use of harsh chemicals as with other conventional methods. The visual color change and the spectroscopic techniques confirmed the formation of NPs. The result of the larvicidal result suggests that they might have applications as nanolarvicide if more investigation is done to establish more baseline information.

Table-1. Probit result for larvicidal activity.

Larva stage	Conc. (mg/L)	%mortality	LC ₅₀	LC ₉₀	95%confidence		X ²	r
					LC ₅₀	LC ₉₀		
1 st Instar	5	25	14.746	83.959	11.815-16.777	64.049-155.159	3.426	0.969
	10	43						
	20	53						
	25	63						
	50	85						
2 nd Instar	5	20	18.251	258.831	15.746-24.125	110.613-468.908	1.235	0.925
	10	40						
	20	50						
	25	55						
	50	70						
3 rd Instar	5	15	18.496	331.496	16.957-25.381	105.462-386.851	4.542	0.873
	10	40						
	20	48						
	25	58						
	50	67						

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