



## Colorimetric Detection of Pesticide Residues in Water Using Zinc oxide Nanoparticles

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### ABSTRACT

Zinc oxide nanoparticles were synthesized through green chemistry using lemon extract as reducing agent. The sensitivity of the synthesized nanoparticles against pesticide residues in water was determined. The nanoparticles were characterized based on the absorbance. Characteristic peaks were observed at 281nm and 328nm attributed to the exciton absorption peak and maximum absorbance that confirmed the size of the product. The nanoparticles were mixed with different concentration of pesticides in water at varying ratio. The color change was observed in relation to its sensitivity to the solutions. Evident change in color from pale yellow to colorless were observed for both 400ppb of clothianidin and imidacloprid mixed with ZnO nanoparticles at 1:1 ratio. Absorbance spectra revealed that there is a difference in the interaction of the two pesticides with ZnO nanoparticles. However, same visual color change were observed for both pesticides from pale yellow to colorless. This suggests that the ZnO nanoparticles were not selective as colorimetric detector for clothianidin and imidacloprid.

**Keywords:** Colorimetric detection, Zinc oxide nanoparticles, Pesticide residues, Water pollutant, Nanotechnology.

### INTRODUCTION

Pesticide residues occurrence in water systems is one of the persistent environmental problems worldwide which also pose great risk to health and safety. Occurrence of pesticides in water systems are mainly due to soil erosion and run off from fields. A review on pesticide residues detection worldwide based on published researches from 2000 to 2020, revealed that 31 countries including the Philippines have water systems contaminated

with pesticide residues<sup>1</sup>. This not only suggests the need for the regulations regarding pesticide use and additional systems for water purification but also the need for continuous monitoring and evaluation of pesticide residues. The Philippines is considered as an agricultural country where almost 50% of its land area is being utilized by agriculture<sup>2</sup>. As such, rampant use of pesticides in the country that seeps to water systems are also evident<sup>3,4</sup>.

Detection of pesticide residues in food and



water has been the focus of several researches due to the excessive use of pesticides parallel to the emergence of resistance of pests to pesticides. It is a known fact that pesticides have acute and chronic health risks associated to its exposure and intake. Pesticide residues in water usually is detected using sophisticated equipment such as LC-MS and GC-MS as reflected in the previous studies. Recent advances in the field of nanotechnology provided an alternative on the use of nanoparticles in colorimetric detection of pollutants. Several methods have been developed with the aim of simplifying the procedure of detection without the use of sophisticated equipment. Thus, colorimetric detector for the detection of pesticide residues attracted attention for research and innovation. Colorimetric detection aids in on-site detection of analyte real-time using the naked-eye. This offers the advantages of being economical, practical, with comparable sensitivity and selectivity, and it does not require a trained technician or specialized equipment for the detection<sup>5,6</sup>. There are several methods and strategies involved in the development of colorimetric sensors. Detection of pesticide residues based on nanoparticles is one of these.

Nanoparticles have strong extinction coefficient and localized surface plasmon resonance (LSPR) that enables it to change color in different environment. The size changes the surface plasmon resonance (SPR) of metal nanoparticles. It is the depiction of changes that occurs in the quantum scale of the resonance effect created due to the interaction of conduction electrons of metals<sup>7</sup>. It is dependent on the size, the shape or assembly and the composition of the dispersion medium. This phenomenon causes the nanoparticles to have a different optical, chemical and electronic properties as compared to the bulk material. The SPR serves as a measuring aid in determining changes in the refractive index on the surface of the nanoparticle<sup>8</sup>. It enables these changes to be visible to the naked-eye through exhibition of different colors in the visible region in the electromagnetic spectrum. Thus, nanoparticles have broad applications as colorimetric sensors for detection of analyte. Studies have been conducted in the development of silver nanoparticles modified with citrate in the detection of fungicide residues in water, potato and wheat samples<sup>9,10</sup> and silver nanoparticles reduced using sucrose as sensor probe for hydrogen peroxide<sup>11</sup>.

Gold nanoparticles have also been developed as colorimetric sensors for organophosphates<sup>12,13</sup> and malathion<sup>14</sup>. The color change observed was from red to grey or purple attributed to the aggregation of gold nanoparticles in the presence of organophosphates. Metal oxides nanoparticles have also been used both for sensing and remediation in the environment field due to its superconducting properties<sup>15</sup>. These nanoparticles tend to have photocatalytic activities that is being utilized for both sensing and remediation of pesticide residues. Zinc oxide nanoparticles have attracted attention worldwide due to its ease of synthesis and characteristics which can be utilized in various industries. It was determined to possess high chemical and thermal stability as well as optical transparency and luminescent properties<sup>16</sup>. Several studies have been conducted for its antimicrobial<sup>17</sup> and therapeutic activity<sup>18</sup>. Recently, it was also determined to have the advantages as source of zinc for soil as nanofertilizers<sup>19,20,21</sup> owing to its bioavailability and low toxicity in comparison to other metal oxide nanoparticles. Zinc oxide nanoparticles also showed significant photocatalytic activity and affinity to organic dyes<sup>22,23</sup>. ZnO nanoparticles have been modified with 1-butyl-3-methylimidazolium tetrafluoroborate for the removal of toxic dyes<sup>24</sup>. The same modification on ZnO nanoparticles was conducted and was determined to be effective adsorbent of naphthalene and efficient removal of pesticides<sup>25</sup>. ZnO nanoparticles have also been used and determined to be effective as nanophotocatalyst in the degradation of methyl parathion and parathion<sup>26</sup> and permethrin<sup>27</sup>. These are considered as neurotoxic pesticides. These physical and chemical properties of zinc oxide nanoparticle provides it with advantages as a colorimetric detector application without having harmful threat to living organisms and posing as a pollutant in the future.

Green synthesis involves the use of organic materials or less toxic alternative to chemicals in the synthesis of a product. Plant materials such as stem, leaf, root, fruit and seeds extract have been utilized for green synthesis of nanoparticles due to its economical advantage in addition to its environmental viability in large scale production<sup>28</sup>. Several nanoparticles have been synthesized utilizing this advantages of green synthesis such as the synthesis of magnesium oxide nanoparticles using *Tridax procumbens* and *Myristica fragrance*<sup>29</sup> and copper oxide nanoparticles using

*Illicium verum* and *Polianthes tuberosa* extracts as anticancer agent<sup>30</sup>. However, the efficiency of green synthesis is dependent on its optimization due to variations caused in changes in temperature, pH and incubation time for the synthesis<sup>31</sup>. Green synthesis of ZnO nanoparticles is usually confirmed by the appearance of a yellowish mixture. Several studies have been conducted in synthesizing ZnO nanoparticles using extracts from NPs synthesized from extracts of *Illicium verum*<sup>32</sup>, *Agathosma betulina*, *Moringa oleifera*, *Pongamia pinnata*, *Plectranthus amboinicus*, *Nephelium lappaceum* and *Calatropis gigantea*.

The study explores the characteristics of zinc oxide (ZnO) nanoparticles as a less toxic alternative for detecting pesticide residues in water. It includes the characterization of ZnO, sensitivity to different pesticide concentration and selectivity based on absorbance changes. The study is limited to determination of the sensitivity and selectivity of zinc oxide nanoparticles against the two pesticides commonly used in the production of palay. Particularly, the study focused only on the determination of two active ingredients of the pesticides, Clothianidin and imidacloprid.

## MATERIALS AND METHODS

### Chemicals and equipment

Zinc oxide (ZnO) and other chemical reagents were used without any further purification. Distilled and deionized water was used as the solvent to prepare all the solutions.

A Perkin Elmer Lambda 365 UV-Vis Spectrophotometer was used for the characterization, sensitivity and selectivity studies.

### Synthesis and Characterization of Zinc oxide Nanoparticles

Zinc oxide nanoparticles were synthesized using the optimized conditions determined by Hossain *et al.*,<sup>33</sup> where lemon (*Citrus limon*) extract was used as reducing agent. The lemon fruits were cut into pieces and dried at 60°C for 10 hours. The dried lemon pieces were then esterized to obtain powder. The powder was mixed with distilled water at a ratio of 100 mL per 1 g of lemon powder. The suspension was stirred continuously using a magnetic stirrer at 60-70°C for 4 hours. The

suspension was allowed to cool at room temperature. It was then filtered using Whatman No. 1 filter paper. The extract was used as reducing agent for the synthesis of the nanoparticles.

About 4.07 g of ZnO was dissolved in 10 mL of ethylene glycol and diluted using distilled water to 100 mL. The solution was mixed with the lemon extract with 1:1 ratio gradually at room temperature for 4 h until a colloid was observed. The colloid was diluted with distilled water with 1:10 ratio prior to its characterization.

The absorbance of the synthesized ZnO nanoparticles was determined using Perkin Elmer Lambda 365 UV-Vis Spectrophotometer. The absorbance at wavelength 250 to 500nm at 0.5nm resolution and scan rate of 120nm/minute was obtained.

### Sensitivity and Selectivity of Zinc oxide Nanoparticles

The two most commonly used pesticides for palay in the region were used in the sensitivity studies. Active ingredients are clothianidin and imidacloprid. Different concentrations of each pesticide were prepared using distilled water as solvent in accordance with the reported United States Environmental Protection Agency (US EPA) toxic dose as presented in Table 1.

**Table 1: Concentrations of pesticides used for the sensitivity studies**

Pesticide	US EPA toxic dose	Concentrations used
Clothianidin	320ppb	200, 250, 300, 350, and 400ppb
Imidacloprid	283ppb	200, 250, 300, 350, and 400ppb

\*ppb = parts per billion

The pesticide solutions were mixed with ZnO nanoparticles with 1:1 ratio. Changes in the absorbance of the ZnO nanoparticles and the initial spectra of the pesticide were determined by scanning the absorbance at wavelength 200 to 500nm at 0.5nm resolution.

## RESULTS AND DISCUSSION

### Zinc oxide Nanoparticles Synthesis and Characterization

Zinc oxide nanoparticles were synthesized

through green synthesis approach using lemon extract as reducing agent. The use of lemon extract offered the advantage of using a non-toxic and readily available reducing agent. Based on the review study of Basnet *et al.*,<sup>34</sup> (2018), the polyphenols such as the tannins, glycosides, and flavonoids present in the plant extract forms complex with zinc ions precursors reducing zinc ions to be in zero valency. Consequently, the hydroxyl groups in the polyphenols causes the occurrence of hydrolysis reactions leading to the formation of zinc hydroxide. Zinc oxide nanoparticles result from the calcination and decomposition reactions that occurs afterwards. Lemon peels extract is determined to be rich in phytochemicals including carbohydrates, alkaloids, tannins, oils, proteins, cardiac glycosides, steroids, phenols, flavonoids and amino acids<sup>35,36</sup>.

The absorbance spectrum of the synthesized ZnO nanoparticles was obtained thru UV-Vis spectrophotometer from 300 to 500nm as shown in Figure 1.

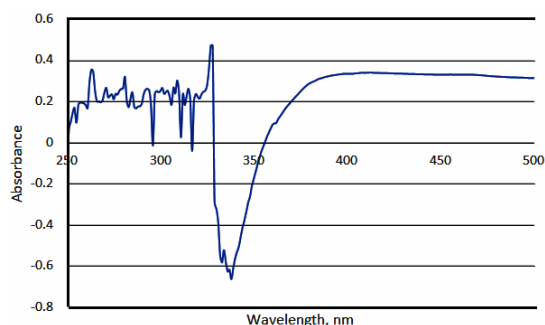


Fig. 1. Absorbance spectrum of ZnO nanoparticles

The characteristic peak of ZnO nanoparticles at 328nm confirmed its size in the nanometer range. Another peak was observed at 281nm. This peak can be considered as an excitonic absorption peak which occurred due to ZnO nanoparticles that lie below the band gap wavelength<sup>37</sup>. It can be gleaned that the several small absorption peaks are evident aside from the two normal absorption peaks of ZnO nanoparticles. This is due to the polydispersed nature of the sample which closely resembles a suspension. A side peak at around 362nm was also evident in the spectrum. The side peak is due to the intrinsic band-gap absorption of the nanoparticles upon its electron transitions from the valence band to the conduction band<sup>38,39</sup>. These transitions due to band gaps are characteristics of ZnO nanoparticles that are absent in other nanoparticles.

### Sensitivity of ZnO Nanoparticles to Pesticides

Sensitivity studies of the ZnO nanoparticles was conducted using different concentrations of pesticides commonly used in the rice fields. The pesticides used contained two different active chemical; clothianidin and imidacloprid.

Clothianidin solutions with concentration ranging from 200 to 400ppb were prepared and were mixed with ZnO nanoparticles at 1:1 ratio. The appearance as obtained from a Huawei Leica smartphone with a white background was shown in Figure 2.

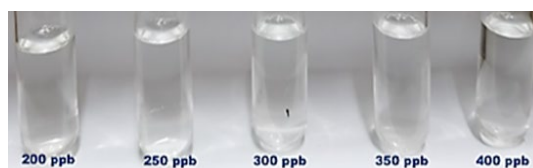


Fig. 2. Clothianidin and ZnO nanoparticles at 1:1 ratio

The color of the solutions ranges from pale yellow to almost colorless at increasing clothianidin concentration. Without a white background, the change in color cannot be seen clearly. Visible color change was observed at the solution with highest clothianidin concentration (Figure 3).

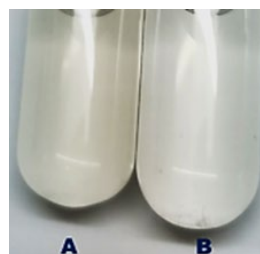


Fig. 3. Difference in color of solutions at 1:1 ratio of (A) pure ZnO nanoparticles with distilled water and (B) ZnO nanoparticles with 400ppb clothianidin

Thus, visible limit of detection was determined at around 400ppb. This value is higher than the EPA determined toxicity limit of clothianidin for chronic exposure which is 320ppb. This suggests that the limit of detection of ZnO nanoparticles still needs to be improved for application in colorimetric field testing.

Imidacloprid solutions with concentration ranging from 200 to 400ppb were prepared and were mixed with ZnO nanoparticles at 1:1 ratio. The appearance as obtained from a Huawei Leica smartphone with a white background was shown in Figure 4.

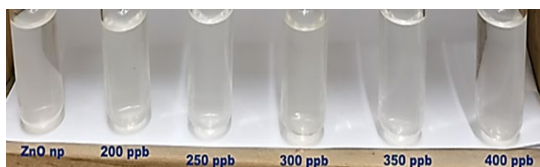


Fig. 4. Imidacloprid and ZnO nanoparticles at 1:1 ratio

The color of the solutions ranges from pale yellow to almost colorless at increasing imidacloprid concentration. Similar to that of clothianidin, without a white background, the change in color cannot be seen clearly. However, the change in color is more gradual in comparison to clothianidin. Visible color change was observed at the solution with highest imidacloprid concentration (Figure 5).

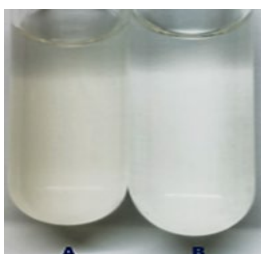


Fig. 5. Difference in color of solutions at 1:1 ratio of (A) pure ZnO nanoparticles with distilled water and (B) ZnO nanoparticles with 400ppb imidacloprid

The visible limit of detection was determined at around 400ppb. This value is higher than the EPA determined toxicity limit of imidacloprid for chronic exposure which is 283ppb. This suggests that the limit of detection of ZnO nanoparticles still needs to be improved for application in colorimetric field testing.

#### Selectivity studies of ZnO nanoparticles

As based on the color change observed for both pesticides, it can be noted that the ZnO nanoparticles is not selective for it provided similar change from pale yellow to colorless for both pesticides. This non-selectivity observed was further confirmed by analyzing the absorbance spectra of the samples.

The change in color can be explained upon looking at the changes in the absorbance spectrum of ZnO nanoparticles upon addition of clothianidin (Figure 6).

As can be seen in Fig. 7, evident change in part of the absorbance spectra of ZnO nanoparticles occurred upon addition of 400ppb of clothianidin.

The characteristic excitonic absorption peak at 281 nm disappeared. This suggests that the positively charged electron hole created due to the absorption of energy that causes the electron to move from the valence band to the conduction band was placed with possibly an electron from clothianidin. This further suggests that the electronic properties of the nanoparticles will also change. While the disappearance of the pale yellow color may be attributed to the decrease in the absorbance of the solution at 420 to 450nm.

The change in color can be explained upon looking at the changes in the absorbance spectrum of ZnO nanoparticles upon addition of imidacloprid (Figure 7).

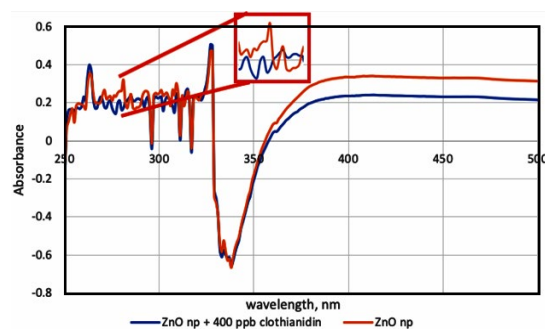


Fig. 6. Absorbance spectra of ZnO nanoparticles (orange) and ZnO nanoparticles with 400ppb of clothianidin (blue)

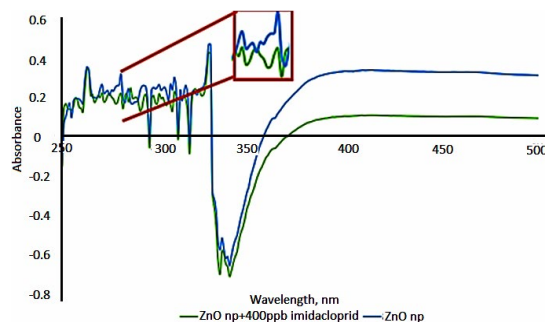


Fig. 7. Absorbance spectra of ZnO nanoparticles (blue) and ZnO nanoparticles with 400ppb of imidacloprid (green)

It can be gleaned that even if the change in color for both the pesticides was from pale yellow to colorless, the change in absorbance spectra were not exactly similar. The exciton absorption peak at 281nm was still evident in the case of ZnO nanoparticles mixed with imidacloprid. The peak however, prominently decreased in absorbance. The results suggest that the positively charged electron holes were still present. This may be attributed to the larger structure of imidacloprid in comparison with clothianidin (Figure 8).

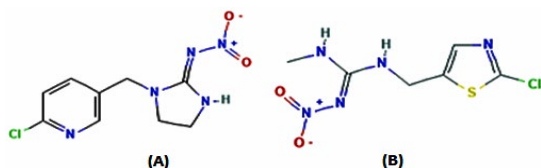


Fig. 8. Chemical structure of (A) imidacloprid and (B) clothianidin

Moreover, it can be gleaned from the structures that imidacloprid has only 2 rotatable bond count which is lower than the 4 rotatable bond count for clothianidin. This implies that the structure of clothianidin may twist more readily and squeeze into the holes in the nanoparticles better than imidacloprid.

However, despite the difference in the interaction of the two pesticides with ZnO

nanoparticles, the absorbance spectra of the samples at 420 to 450nm were the same. Hence, the change in color observed was the same for both. In terms of colorimetric detection using the naked-eye, the ZnO nanoparticles were not selective against the two pesticides used in the study. Thus, modification of the nanoparticle may be explored to develop a more selective colorimetric detector. Other confirmatory analyses such as the use of IR, XRD, SEM techniques may also be conducted for future studies.

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#### REFERENCES

1. El-Nahhal, I.; & El-Nahhal, Y. *J. Environ. Manage.*, **2021**, *299*, 113611.
2. Yamagishi, K.; Gantalao, C. & Ocampo, L. *J. Tour. Futures.*, **2021**.
3. Bonmatin, J. M.; Mitchell, E. A.; Glauser, G.; Lumawig-Heitzman, E.; Claveria, F.; van Lexmond, M. B. & Sánchez-Bayo, F. *Sci. Total Environ.*, **2021**, *757*, 143822.
4. Cubelo, J. E. C. & Cubelo, T.A. *JNS.*, **2021**, *1*, 25-43
5. Singh, R.; Kumar, N.; Mehra, R.; Kumar, H.; Singh, V. P. *Trends Environ. Anal. Chem.*, **2020**, *26*, e00086.
6. Che Sulaiman, I. S.; Chieng, B. W.; Osman, M. J.; Ong, K. K.; Rashid, J. I. A.; Wan Yunus, W. M. Z.; Mohamad, A. *Mikrochim Acta.*, **2020**, *187*(2), 1-22.
7. Jana, J.; Ganguly, M.; Pal, T. *RSC Adv.*, **2016**, *6*(89), 86174-86211.
8. Homola, *J. Chem. Rev.*, **2008**, *108*(2), 462-493.
9. Keller, A. A.; Vosti, W.; Wang, H.; Lazareva, A. *J. Nanoparticle Res.*, **2014**, *16*(7), 1-10.
10. Ma, S.; He, J.; Guo, M.; Sun, X.; Zheng, M.; Wang, Y. *Colloids Surf. A Physicochem. Eng. Asp.*, **2018**, *538*, 343-349.
11. Reyes D. F. *Orient. J. Chem.*, **2020**, *36*(4).
12. Li, H.; Guo, J.; Ping, H.; Liu, L.; Zhang, M.; Guan, F.; Zhang, Q. *Talanta.*, **2011**, *87*, 93-99.
13. Liu, W.; Zhang, D.; Tang, Y.; Wang, Y.; Yan, F.; Li, Z.; Zhou, H. S. *Talanta.*, **2012**, *101*, 382-387.
14. Bala, R.; Dhingra, S.; Kumar, M.; Bansal, K.; Mittal, S.; Sharma, R. K.; Wangoo, N. *J. Chem. Eng.*, **2017**, *311*, 111-116.
15. Rawtani, D.; Khatri, N.; Tyagi, S.; Pandey, G. *J. Environ. Manage.*, **2018**, *206*, 749-762.
16. Fakhari, S.; Jamzad, M.; Kabiri Fard, H. *Green Chem Lett Rev.*, **2019**, *12*(1), 19-24.
17. da Silva, B. L.; Abuçafy, M. P.; Manaia, E. B.; Junior, J. A. O.; Chiari-Andréo, B. G.; Pietro, R. C. R.; Chiavacci, L. A. *Int J Nanomedicine.*, **2019**, *14*, 9395.
18. Wiesmann, N.; Tremel, W.; Brieger, J. *J Mater Chem B.*, **2020**, *8*(23), 4973-4989.
19. Bala, R.; Kalia, A.; Dhaliwal, S. S. *J. Soil Sci. Plant Nutr.*, **2019**, *19*(2), 379-389.
20. Yusefi-Tanha, E.; Fallah, S.; Rostamnejadi, A.; Pokhrel, L. R. *Sci. Total Environ.*, **2020**, *738*, 140240.
21. Awan, S.; Shahzadi, K.; Javad, S.; Tariq, A.; Ahmad, A.; Ilyas, S. *J. Saudi Soc. Agric. Sci.*, **2021**, *20*(1), 18-24.
22. Al-Rawashdeh, N. A.; Allabadi, O.; Aljarrah, M. T. *ACS omega.*, **2020**, *5*(43), 28046-28055
23. Dodoo-Arhin, D.; Asiedu, T.; Agyei-Tuffour, B.; Nyankson, E.; Obada, D.; Mwabora, J. M. *Mater. Today: Proc.*, **2021**, *38*, 809-815.
24. Chaudhary, S.; Kaur, Y.; Umar, A.; Chaudhary, G. R. *J. Mol. Liq.*, **2016**, *224*, 1294-1304.

25. Kaur, Y.; Bhatia, Y.; Chaudhary, S.; Chaudhary, G. R. *J. Mol. Liq.*, **2017**, *234*, 94-103.
26. Sharma, A. K.; Tiwari, R. K.; Gaur, M. S. *Arab. J. Chem.*, **2016**, *9*, S1755-S1764.
27. Dehaghi, S. M.; Rahmanifar, B.; Moradi, A. M.; Azar, P. A. *J. Saudi Chem. Soc.*, **2014**, *18*(4), 348-355.
28. Qu, J.; Yuan, X.; Wang, X.; Shao, P. *Environ. Pollut.*, **2011**, *159*(7), 1783-1788.
29. Kalaimathi, M.; Hariharan, K.; Vishnu, S. & Chinnasamy, R. *CRGSC.*, **2021**, *4*, 100185.
30. Sisira, S.; Hithisha, K. S.; Sankar, J. S.; Nazirin, N.; Vimalraj, R. K. & Kalaimathi, M. *Inorg. Chem. Commun.*, **2022**, *145*, 109961.
31. Ochieng, P. E.; Iwuoha, E.; Michira, I.; Masikini, M.; Ondiek, J.; Githira, P.; Kamau, G. N. *Int. J. Biochem. Phys.*, **2015**, *23*, 53-61.
32. Kalaiamthi M.; Maheshwaran, A.; Hariharan, K; Poovarasana, B & Chandru P. *Orient. J. Chem.*, **2021**, *37*(4).
33. Hossain, A.; Abdallah, Y.; Ali, M.; Masum, M.; Islam, M.; Li, B.; An, Q. *Biomolecules.*, **2019**, *9*(12), 863.
34. Basnet, P.; Inakhunbi Chanu, T.; Dhruvajyoti, S. & Somenath, C. *J. Photochem. Photobiol. B: Biol.*, **2018**, *183*, 201-221.
35. Mathew, B.; Suresh, B.; Jatawa, K. & Tiwari, A. *Int J Pharm Pharm Sci.*, **2012**, *2*, 369-71.
36. Duru, I.A. & Enyoh, C.E. *J. Medicinal Plants.*, **2020**, *8*(5), 178-182.
37. Talam, S.; Karumuri, S. R.; Gunnam, N. *Int. Sch. Res. Notices.*, **2012**.
38. Zak, A.K.; Razali, R.; Abd Majid, W.H.; Darroudi, M. *Int J Nanomedicine.*, **2011**, *6*, 1399.
39. Sangeetha, G.; Rajeshwari, S.; Venckatesh, R. *Mater. Res. Bull.*, **2011**, *46*(12), 2560-2566.