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Efficacy of synthesized silver nanoparticles using *Ocimum sanctum* (L.) leaf extract against *Corcyra cephalonica* (S.)

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Abstract
The rice moth, *Corcyra cephalonica* (Stainton) (Lepidoptera: Pyralidae), is an important stored-product pest all over the world. Biosynthesis of pesticides from plant extract is most promising new approach for pest control. The present study was carried out for two years 2015-16 and 2016-17 to ascertain the larvicidal activity of synthesized silver nanoparticles (AgNP's) using plant leaf extracts from *Ocimum sanctum* (L.). Synthesis was confirmed through UV-Vis spectrophotometer in wavelength range of 200-700 nm. The peak was recorded at 422 nm which confirmed the formation of silver nanoparticles, zeta potential values were found to be -16.4 mV, FT-IR analysis showed strong peaks at ranges of 800-4000cm^{-1} which exhibited presence of different types of functional groups *viz.*, (O-H, H-H, C-H, C-C and N-H). TEM analysis determined the size and shape of *Ocimum sanctum* (L.) AgNP's with average size of 64.04 nm which is roughly circular or spherical in shape. Efficacy of green synthesized silver nanoparticles (AgNP's) from *Ocimum sanctum* (L.) against *Corcyra cephalonica* (S.) was evaluated in dilutions range 200, 300, 400 and 500 ppm. Larval mortality of *C. cephalonica* was found to be (80% and 83.33%) both the years 2015-16 and 2016-17. The LC_{50} was also calculated both in 2015-16 and 2016-17; the lowest LC_{50} value was recorded to be 244.04 ppm after fifth day of the treatment. As the nanoparticles concentration and days after treatment increased, larval mortality also increased. There were statistically significant differences in larval mortality between concentrations. The result suggested that the use of plants for synthesis of silver nanoparticles may be considered as environmentally safer and greener approach for pest control.

Keywords: Nanotechnology, *C. cephalonica*, silver nanoparticles, green synthesis

1. Introduction
In every year, one third of the world grain crop is loss during storage, much of this is particularly due to insect attack. Worldwide losses in stored products, caused by insects, have been estimated to be between five and ten percent [1]. Losses of grain in storage due to insects, limits the final agricultural production. The rice moth, *Corcyra cephalonica* (S.) is an important stored-product pest all over the world. It is probably one of the most catholic feeders among the storage pests which feeds on a wide variety of materials viz., rice, corn, cocoa, dried fruits like almonds, date palm, nuts, chocolates, biscuits, oilcakes, coffee and other seeds [2] and also when they become fully grown, larvae contaminate the grain by producing dense webbing containing their faecal material and cast skins [3]. The current methods for managing stored grain pests depend heavily on synthetic pesticides, but the repeated use of certain chemical fungicides in storage houses has led to the appearance of fungicidal-resistant populations of storage pathogens. Unfortunately this leads to contamination of food with toxic residues [4]. To overcome this situation, different forms of insecticides have been advocated. In recent years, pest management strategies are gradually bending towards eco-friendly agriculture without interrupting ecological balance [5]. Recently, nanotechnology has being embraced in the world of pesticides and pest contro [16], has the potential to revolutionize modern day agriculture by including management of insect pests through the formulations of nanomaterials-based insecticides.

In particular, silver nanoparticles (AgNP's) are proved to have potential antibacterial, antifungal, larvicidal properties [7]. Synthesis of nanoparticles using microorganisms, enzymes, plants or plant extracts has been suggested as possible eco-friendly alternatives to chemical and physical methods [8]. Using plants for nanoparticles synthesis can be advantageous over other biological processes because it eliminates the elaborate process of maintaining cell cultures and can also be suitably scaled up for large-scale nanoparticles synthesis [9]. The use of plants for synthesize of nanoparticles are rapid, low cost, eco-friendly and a single-step method for biosynthesis process [10]. In present study, *Ocimum sanctum* (L.) has been used on the basis of its insecticidal properties. There are two kinds of *Ocimum sanctum* available, Telugu Purple variety is called Krishna Tulsi and green variety is called Sri Tulsi, the common name is Holy basil. Their chemical constituents are similar and also have common medicinal properties. The leaf volatile oil contains eugenol. Methyl eugenol is a phenylpropene, a type of phenylpropanoid compound, the methyl ether of eugenol which is used in insect traps for insect attraction. Even today, the research on the application of nanopesticides against pest larvae is in infancy. The present study was designed with a novel, rapid and cost effective route of biosynthesis of AgNP's using *Ocimum sanctum* (L.) leaf extract. The synthesized AgNP's obtained by the green method are under evaluation of their effect against *Corcyra cephalonica* (S.).

2. Materials and Methods

2.1 Insects Rearing

The stock culture of *Corcyra cephalonica* (S.) was obtained from Biological Control Laboratory, Assam Agriculture University. For preparation of wooden boxes and rice medium a particular method was followed [11]. The stock cultures were maintained at 28±1 °C. Fourth instar stage of the larvae were used for the experiments.

2.2 Collection and preparation of plant leaves extract

Fresh plant leaves of *Ocimum sanctum* (L.) was collected and washed several times with distilled water to remove the dust particles and then sun dried to remove the residual moisture. The leaf extracts used for synthesis of nanoparticles were prepared by placing 10 gram of the washed, dried and fine cut leaves in 200ml glass beaker along with 100 ml of sterile distilled water. The aqueous mixture was then boiled for 5-10 minutes until the colour of aqueous solution changes to light yellow or brown. Then the aqueous extracts were cooled to room temperature and filtered with Whatman filter paper No. 1 before centrifugation at 1200 rpm for 5 minutes to remove the heavy biomaterials. The leaf extracts were stored at room temperature in order to be used for further studies as per the method [12].

2.3 Synthesis of silver nanoparticles

Silver nanoparticles were synthesized by modified method [12]. For synthesis of silver nanoparticles 5-10 ml of plant leaf extract *Ocimum sanctum* (L.) was added separately into 100 ml of 1 mM aqueous solution of silver nitrate ($AgNO_3$) (Sigma Aldrich) with stirring magnetically at room temperature. After adding 0.0017gm/ml of silver nitrate solution in 100 ml of sterile distilled water which contained 5-10gm of plant extract the flasks were put into the dark condition for 1-2 days. After 1-2 days the resulting light solution changed to dark brown in colour, indicating the formation of silver nanoparticles (AgNPs).

2.4 Characterization of green synthesized silver nanoparticles (AgNPs) of *Ocimum sanctum* (L.)

Characterization of green synthesized AgNPs of *Ocimum sanctum* (L.) were done by the following equipments *viz.*, UV-Vis spectrophotometer, Zeta potentiometer, Transmission Electron microscopy and Fourier transform infrared spectroscopy. The silver nanoparticles formation were confirmed by measuring the wave length of reaction mixture in the UV-Vis spectrum of the Shimadzu UV 1601 spectrophotometer at a resolution of 1 nm (from 300 to 700 nm) in 2 ml quartz cuvette with 1 cm path length (Nanotechnology Lab, AAU, Jorhat). Zeta potential used for determined the surface charge of nanoparticles in order to study their coagulation properties of green synthesized nanoparticles. Zeta potential of the silver nanoparticles was determined by Malvern Zetasizer ZEN 3600 (NEIST, Jorhat). Fourier transform infrared spectroscopy (FT-IR), used for the characterization of functional groups on the surface of AgNPs by plant extracts. The analysis was done by Shimadzu at (NEIST, Jorhat). The spectra were scanned in the range of 4000-400 cm^{-1} range at a resolution of 4 cm^{-1}. The samples were prepared by dispersing the AgNPs uniformly in a matrix of dry KBr, compressed to form an almost transparent disc. Morphological analysis of the nanoparticles was done with Transmission Electron Microscopy (TEM). TEM Micrograph was done at Sophisticated Analytical Instrument Facility (SAIF), North Eastern Hill University (Meghalaya) at accelerating voltage of 200kv with 2000x magnification. The TEM micrograph images were recorded on a JEOL 1200 EX instrument on carbon coated copper grids with an accelerating voltage of 100 to 200 kV.

2.5 Efficacy of green synthesized silver nanoparticles against *Corcyra cephalonica* (S.)

The experiment was conducted with 5 treatments and 5 replications for synthesized (AgNPs) from *Ocimum sanctum* (L.).To determine lethal concentration of green synthesized nanoparticles solution, serial dilution ranging from 200, 300, 400 and 500 ppm were prepared. The required amount of larvae were directly collected from field and put into the clean, air dried petriplates, rice grains were coated with different concentrations of silver nanoparticles solution and air dried. The control grains were treated with water alone. The 3 hrs starved different larvae were fed with different concentrations of silver nanoparticles treated and untreated plant parts [13]. After every 24 hrs, the uneaten plant parts were removed and placed with fresh treated and untreated ones, up to fifth day of the treatment. Then the percent larval mortality data were subjected to ANOVA analysis.

2.6 Observations

Corrected Larval Mortality (%)

Mortality rates (MR) were measured[14] for larvae of *Corcyra cephalonica* (L.). Series of controls *Corcyra cephalonica* (L.) larvae were allowed to complete their development under the room conditions of light and temperature. Then the MR were estimated as,

$$\text{Larval mortality } (\%) = \frac{\text{Number of dead larvae}}{\text{Initial number of larvae}} \times 100$$

2.7 Statistical analysis

Data were analyzed by Completely Randomized Design (CRD). Result with $P>0.05$ was considered to be statistically significant. The data sets were subjected to Analysis of Variance (ANOVA) and the data sets were also used for Probit Analysis [15] to determine the LC_{50} of the above mentioned synthesized nanoparticles using, (SPSS, Inv., version 20.00).

3 Results and Discussion

3.1 Characterization of green synthesized silver nanoparticles (AgNPs) of *Ocimum sanctum* (L.)

For synthesis of silver nanoparticles, *Ocimum sanctum* (L.) leaves extract were exposed to 1 mM aqueous solution of silver nitrate and observed that colour of plant extracts changes from light colour to dark brown colour at 72-96 hrs of reaction. This colour changed from light to dark which confirmed the synthesis of silver nanoparticles. Gradual increase in colour development from light to dark brown was observed in synthesized materials, which indicated the formation of silver nanoparticles [16]. In the present study characteristics SPR absorption peak (band) was observed in plant extract of *Ocimum sanctum* (L.) treated with 1 mM $AgNO_3$ at 422 nm. Green synthesis of silver nanoparticles using *Ocimum sanctum* (L.) and found the absorbance peak at 430 nm, silver nanoparticles exhibits dark yellowish brown colour in the aqueous solution due to the SPR phenomenon [17]. Zeta potential was determined and recorded the charge of green synthesized silver nanoparticles of *Ocimum sanctum* (L.) was -16.4 mV. It indicated that the silver nanoparticles were stable and did not have ability to agglomerate. Tulasi nanoparticles zeta potential value -14.9 mV was recorded [18].

FT–IR analysis of silver nanoparticles synthesized from *Ocimum sanctum* (L.) extract showed strong peaks at 1370 cm^{-1} could assigned to C-O vibration of alcohol, phenols and C-N vibration of amides. Band at 1530 cm^{-1} and 1800 cm^{-1} indicate fingerprinting region of O-H and C- O group. Strong absorption peaks at 3590 cm^{-1} -3336 cm^{-1} result from stretching of the N-H band of amino groups or are indicative of bonded O-H hydroxyl group. Similarly FT-IR measurements of *O. sanctum* (L.) silver nanoparticles, showed sharp absorption peaks located at about 1635 and 3430 cm^{-1}, absorption peak at 1635 cm^{-1} might be assigned to the amide I bond of proteins raised due to carbonyl stretch in proteins and peaks at 3430 cm^{-1} are assigned to O-H stretching in alcohols and phenolics compounds [19]. The TEM images of the silver nanoparticles prepared at 50, 20 and 2 nm scales. TEM analysis revealed that *Ocimum sanctum* (L.) AgNPs were circular in shape with maximum particles in size 16-127 nm, average size 64 nm. TEM micrograph also indicates that green synthesized silver nanoparticles are relatively polydisperse in nature [20]. Tulasi nanoparticles are polydispersed in nature with size 50 nm[1].

3.2 Efficacy of green synthesized silver nanoparticles from *Ocimum sanctum* (L.) against *Corcyra cephalonica* (S.)

Efficacy of green synthesized AgNPs from *Ocimum sanctum* (L.) at different concentration (200, 300, 400, 500 ppm and control) was tested against *Corcyra cephalonica* (S.) for both year 2015-16 and 2016-17, showed in table 1.*Ocimum sanctum* (L.) AgNPs gave mortality of 60%, 66.66%, 76.66% and 80%, respectively at 500 ppm concentrations during 2015-16 were showed in table 1. In 2016-17, *O. sanctum* AgNPs gave mortality of 63.33%, 66.66%, 70% and 83.33% fifth day of the treatment, respectively at 500 ppm concentration. As the concentration and days after treatment increased, the larval mortality of forth instar larvae of *Corcyra cephalonica* (S.) also increased. The results of larvicidal activity clearly indicate that the percentage of mortality being directly proportional to the concentration of the extract. Smaller size of the nanoparticles gave higher efficacy. There were highly significant differences between treated and control larvae. Many natural products from plants having high insecticidal activities against stored-product pests and nanoparticles extracted from these plants were proved to be effective against *C. cephalonica* [21]. The effect of silica nanoparticles against *C. cephalonica* and observed that amorphous silica nanoparticles were found to be highly effective against *C. cephalonica* causing 100% mortality on 8th day of the treatment [22].

Nanoparticles extracted from *Ocimum sanctum* (L.) gave the lowest LC_{50} value of 244.04 ppm against fourth instar larvae of *Corcyra cephalonica* (S.) after fifth day of the treatment during 2016-17, while in 2015-16 LC_{50} was 247.84 ppm after fifth day of the treatment which showed in table 2.Maximum $LC_{50\ 317.86}$ ppm after first day of the treatment during 2015-16 and during 2016-17, probit value 316.59 ppm after first day of the treatment. Probit analysis showed variations in virulence. These LC_{50} values had minimum fiducial limits. Chi-square test revealed statistically non-significant differences. This indicates that the probit analysis supplemented the ANOVA result. Concentration plays important role in larvicidal activity. Present findings are in agreement with earlier reports, bio efficacy of inorganic nanoparticles CdS, Nano-Ag and Nano-TiO_2 against *Spodoptera litura*. The LC_{50} (216.91 to 938.95) was increased as the larval age increased. Probit analysis showed variations in virulence. DNA tagged gold nano particles gave the lowest LC_{50} of 256.31 ppm against 2nd instar *S. litura* larvae on the 5th day and the maximum LC_{50} of 645.75 ppm on the 3rd day after treatment. However, on the 4th day, the LC_{50} of 377.21 ppm was realized [23].

Table 1: Effect of green synthesized silver nanoparticles from *Ocimum sanctum* (L.) on per cent mortality of *Corcyra cephalonica* (L.) under laboratory conditions during 2015-16 and 2016-2017

Treatment (AgNP's)	Dose (PPM)	Per cent larval mortality at different days after treatment		
		1 DAT	3 DAT	5 DAT
Tulasi $AgNO_3$ (2015-16)	200	30 (30.21)	46.66 (43.05)	60 (50.77)
	300	33.33 (35.24)	50 (45)	66.66 (54.70)
	400	36.66 (37.23)	53.33 (46.89)	76.66 (61.07)
	500	46.66 (43.05)	60 (50.77)	80 (63.44)
S. Ed ±		2.24	2.76	3.05
CD (5%)		5.17	5.90	6.30
Tulasi $AgNO_3$ (2016-17)	200	33.33 (35.24)	40 (39.23)	63.33 (52.71)
	300	36.66 (37.23)	50 (45)	66.66 (54.70)
	400	40 (39.23)	53.33 (46.89)	70.00 (56.79)
	500	50 (45)	60 (50.77)	83.33 (65.88)
Control (Water spary)	-	0.0	0.0	0.0
S. Ed ±		2.48	2.90	3.16
CD (5%)		5.22	6.10	6.48

Mean of 10 larvae/replication/treatment; figures in the parentheses are angular transformed values, DAT= Days after treatment.

Table 2: LC_{50} value on per cent mortality of *Corcyra cephalonica* on exposure to different concentrations of green synthesized AgNPs, during 2015-16 and 2016-17

AgNP's from Plants	DAT	X^2(n-1)	Regression equation	LC_{50} (PPM)	Fiducial Limits (con. at 95% CL)
Tulasi $AgNO_3$ (2015-16)	1	1.12	Y=-4.01+1.60x	317.86	269.33 to 365.69
	3	1.21	Y=-4.05+1.65x	281.56	228.41 to 321.18
	5	1.77	Y=-4.27+1.78x	247.84	194.04 to 294.20
Tulasi $AgNO_3$ (2016-17)	1	1.38	Y=-4.10+1.64x	316.59	269.24 to 362.74
	3	1.57	Y=-4.14+1.69x	281.20	229.58 to 329.90
	5	2.32	Y=-4.39+1.83x	244.04	190.57 to 280.59

LC_{50}= Lethal concentration that kills 50% of the exposed larvae, UCL = Upper confidence limit, LCL= Lower confidence limit, x^2 = Chi-square values, significant at P<0.05 level, DAT= Days after treatment.

3.3 Behavioural changes in *Corcyra cephalonica* (S.) larvae after treatment with green synthesized silver nanoparticles synthesized from *Ocimum sanctum* (L.)

After exposure of larvae to the test concentrations dilutions of 200, 300, 400 and 500 ppm of green synthesized AgNPs from *Ocimum sanctum* (L.) several changes have been noticed. The *C. cephalonica* larvae almost became 'C' shaped and black in colour and then died on third to fifth day after the treatment at 400-500 ppm concentration. At 200 and 300 ppm concentration larvae turned black and then died. DNA tagged gold nanoparticles against *S. litura* showed cessations of active movement, the skin and entire body became stiff and hard and oozing of the body content (fleshy white), lysis after third day of the treatment. Four days after treatment, the body became swollen, pulpy and fragile. It attained almost a 'C'-shaped and body turned into dark brown [24]. Five days after treatment, the larvae showed premature moulting and the body became discoloured and turned brown. Leaf aqueous extract and synthesized silver nanoparticles of *Nerium oleander* against *Anopheles stephensi* became restlessness, sluggishness, tremors, and convulsions followed by paralysis after exposure to different test concentrations [25].

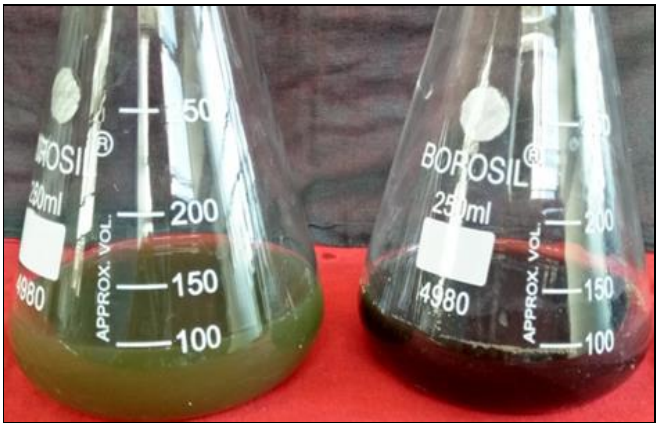

Plate 1: Plant extracts, (A) before addition of $AgNO_3$, (B): after formation of silver nanoparticles

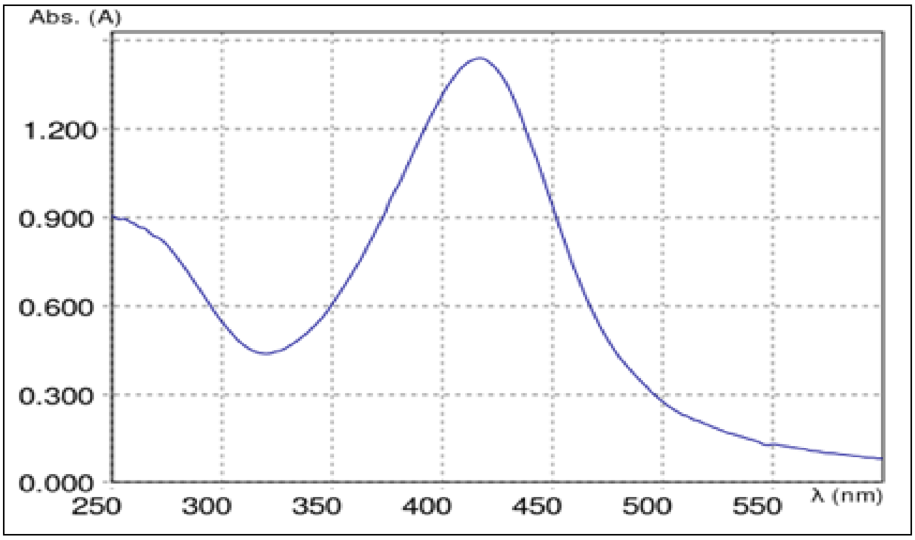

Plate 2: UV-Vis absorption spectra obtained from synthesized silver nanoparticles using *Ocimum sanctum* (L.)

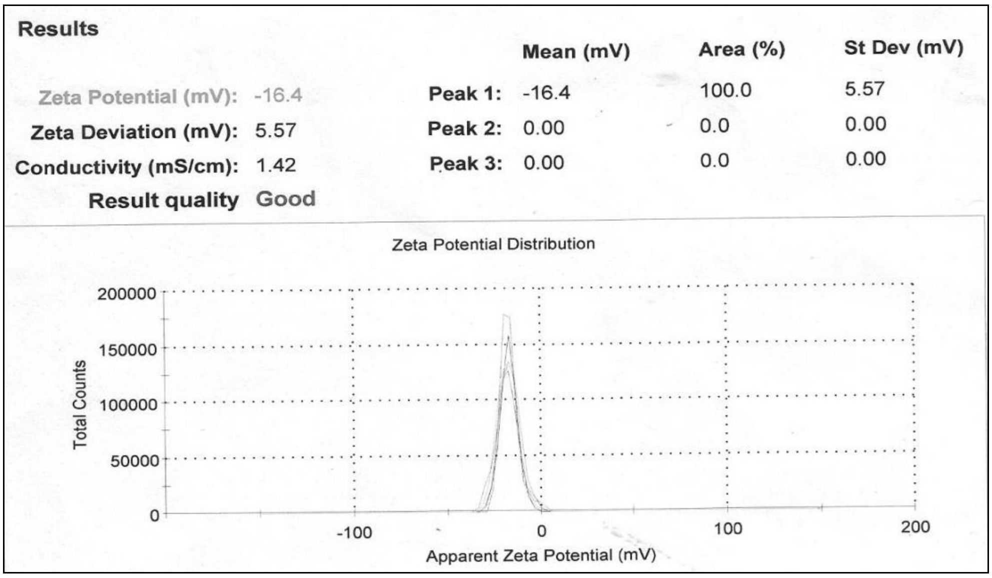

Plate 3: Zeta potential analysis of green synthesized silver nanoparticles from *Ocimum sanctum* (L.)

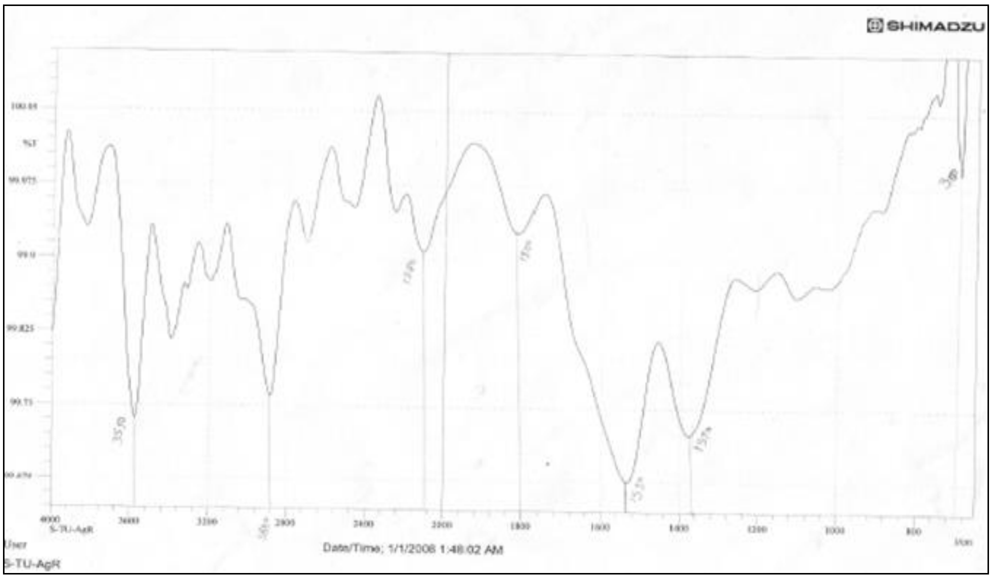

Plate 4: FT-IR analysis of green synthesized silver nanoparticles from *Ocimum sanctum* (L.)

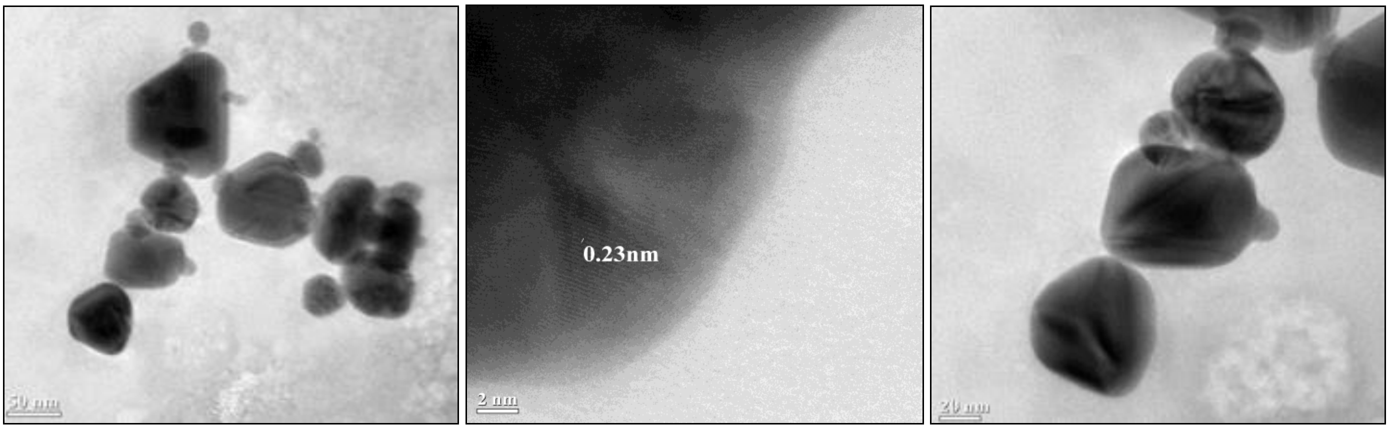

Plate 5: TEM images of *Ocimum sanctum* (L.) AgNPs at different scales showing shape and size

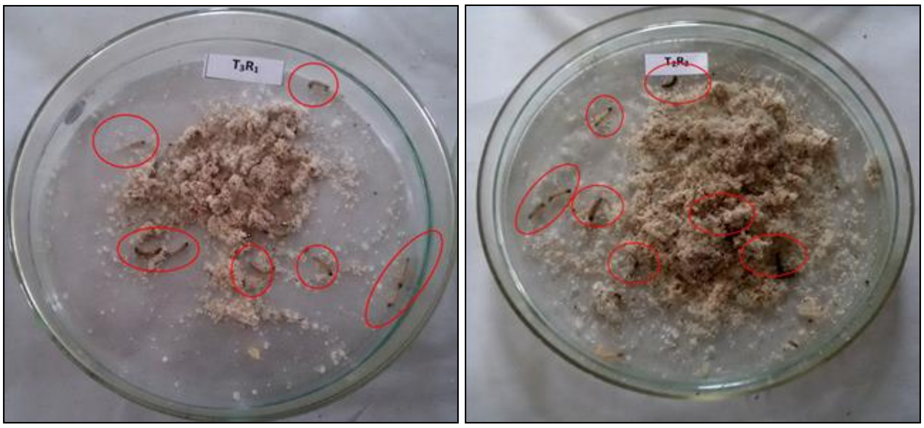

Plate 6: Behavioural changes after treatment of green synthesized nanoparticles 3 days after treatment from *Ocimum sanctum* (L.) AgNPs

Plate 7: Behavioural changes after treatment of green synthesized nanoparticles 5days after treatment from *Ocimum sanctum* (L.) AgNP's

Plate 8: Behavioural changes after spraying of water 5 days after treatment

4. Conclusions

Biological synthesis of silver nanoparticles would be a boon for the development of eco-friendly way of controlling lepidopteran pests instead of toxic insecticides. The results suggested that the use of plants for synthesis of silver nanoparticles may be considered as environmentally safer and greener approach for pest control. Green synthesis of silver nanoparticles can be used as a valuable tool in pest management strategies against *Corcyra cephalonica* (S.). However the environmental tracking of silver nanoparticles, if applied in the field then the impact on environment and health need to be assessed. More detailed study required to understand the actual mechanism of effect of silver nanoparticles inside the insect's body.

5. References

1. David KW, Reeves PA. Introduction-Stored Grain Pest Management. 3rd Edition, Macmillan, New York, 1982.
2. Jagadish PS, Nirmala P, Rashmi MA, Neelu N. Biology of rice moth, *Corcyra cephalonica* (Stainton) on foxtail millet. Karnataka Journal of Agricultural Sciences. 2009; 22:674-675.
3. Fangneng H, Bhadriraju, S. Responses of *Corcyra cephalonica* (Stainton) to pirimiphos-methyl, spinosad, and combinations of pirimiphos-methyl and synergized pyrethrins. Pest Management Science. 2003; 60:191-198.
4. Dubey SC, Suresh M, Singh B. Evaluation of *Trichoderma* species against *Fusarium oxysporum* for integrated management of chickpea. Biological Control. 2007; 40:118-127.
5. Dubey NK, Bhawana S, Ashok K. Current Status of Plant Products as Botanical Pesticides in storage pest management. Journal of Biopesticides. 2008; 1:182-186.
6. Harper S. New Approaches Needed to Gauge Safety of Nanotech-Based Pesticides, Researchers Urge. Published in physics and chemistry, 2010, 4(33).
7. Saxena AP, Tripathi RM, Singh RP. Biological synthesis of silver nanoparticles using onion (*Allium cepa*) extract and their antibacterial activity. Digital Journal of Nanomater Bioscience. 2010; 5(2):427-432.
8. Mohanpuria P, Rana NK, Yadav SK. Biosynthesis of nanoparticles: technological concepts and future applications. Journal of Nanoparticle Research. 2008; 10:507-517.
9. Shankar S, Rai A, Ankamwar B, Singh A, Ahmad A, Sastry M *et al*. Nature Material. 2004; 3:482.
10. Huang J, Li Q, Sun D, Lu Y, Su Y, Yang X *et al*. Biosynthesis of silver and gold nanoparticles by novel sun dried *Cinnamomum camphora* leaf. Nanotechnology. 2007; 18:104-105.
11. Chaudhuri N, Senapati S. Development and reproductive performance of rice moth *Corcyra cephalonica* Stainton (Lepidoptera: Pyralidae) in different rearing media. http://dx.doi.org/10.1016/j.jssas.2011.11.004
12. Araj A, Nida M, Ihab H, Akl M. Toxicity of Nanoparticles against *Drosophila melanogaster* (Diptera: Drosophilidae). Hindawi Publishing Corporation. Journal of Nanomaterials, 2015. Article ID 758132.
13. Devi GD, Murugan K, Selvam P. Green synthesis of silver nanoparticles using *Euphorbia hirta* (Euphorbiaceae) leaf extract against crop pest of cotton bollworm, *Helicoverpa armigera*. Journal of Bio pesticides. 2014; 7:54-66.

14. Abbott WS. A method of computing the effectiveness of insecticides. Journal of Economical Entomology. 1925; 18:265-267.
15. Finney DJ. Probit analysis. Cambridge University, London, 1971, 68-78.
16. Mukunthan KS, Elumalai EK, Patel TN, Murty VR. *Catharanthus roseus:* a natural source for the synthesis of silver nanoparticles. Asian Pacific Journal of Tropical Biomedicine. 2011; 1(4):270-274.
17. Rout Y, Behera S, Ojha A, Nayak P. Green synthesis of silver nanoparticles using *Ocimum sanctum* (Tulashi) and study of their antibacterial and antifungal activities. Journal of Microbiology and Antimicrobials. 2011; 4(6): 103-109.
18. Shende SS, Gaikwad ND, Bansod SD. Synthesis and evaluation of antimicrobial potential of copper nanoparticles against agriculturally important phytopathogens. International Journal of Biological Research. 2016; 1(4):41-47.
19. Jilie K, Shaoning YU. Fourier transforms infrared spectroscopic analysis of protein secondary structures. Acta Biochimica ET Biophysica Sinica. 2007; 39(8):549-559.
20. Rout Y, Behera S, Ojha A, Nayak PL. Green synthesis of silver nanoparticles using *Ocimum sanctum* (Tulashi) and study of their antibacterial and antifungal activities. Journal of Microbiology and Antimicrobials. 2011; 4(6):103-109.
21. Isman MB. Botanical insecticides, deterrents, and repellents in modern agriculture and an increasingly regulated world. Annual Review of Entomolgy. 2005; 51:45-66.
22. Vani C, Brindhaa U. Silica nanoparticles as nanocides against *Corcyra cephalonica* (S.), the stored grain pest. International Journal of Pharma and Bio Science. 2013; 4(3):1108-1118.
23. Chakravarthy AK, Chandrashekhraiah Kandakoor SB, Bhattacharya A, Dhanabala K, Gurunatha K, Ramesh P *et al*. Bio efficacy of inorganic nanoparticles CdS, Nano-Ag and Nano-TiO2 against *Spodoptera litura* (Fabricius) (Lepidoptera: Noctuidae). Current Biotica. 2012a; 6(3):271-281.
24. Chakravarthy AK, Bhattacharyya A, Shashank PR, Epidi TT, Doddabasappa B, Mandal SK *et al*. DNA-tagged nano gold: A new tool for the control of the armyworm, *Spodoptera litura* Fab. (Lepidoptera: Noctuidae). African Journal of Biotechnolgy. 2012b; 11(38):9295-9301.
25. Roni M, Murugan K, Panneerselvam C, Subramanian J, Jiang-shiou H. Evaluation of leaf aqueous extract and synthesized silver nanoparticles using *Nerium oleander* against *Anopheles stephensi* (Diptera: Culicidae). Parasitological Research. 2013; 112:981-990.