



Toxicity and Repellency Potential of Silver Nano-Formulated Phytoextracts Against Rice Weevil *Sitophilus oryzae* (L.) (Coleoptera: Curculionidae)

Muhammad Salman Saeed¹, Umar Farooq¹, Muhammad Zeeshan Majeed^{1*}, Lamya Ahmed Alkeridis², Samy Sayed³, Abu Bakar Muhammad Raza¹ and Asma Akram¹

¹Department of Entomology, College of Agriculture, University of Sargodha, Sargodha 40100, Pakistan

²Department of Biology, College of Science, Princess Nourah Bint Abdulrahman University, P.O. Box 84428, 11671 Riyadh, Saudi Arabia

³Department of Economic Entomology and Pesticides, Faculty of Agriculture, Cairo University, Giza 12613, Egypt

ABSTRACT

Rice weevil, *Sitophilus oryzae* (L.) (Coleoptera: Curculionidae), is one of the most widely distributed destructive pests of stored rice and wheat grains. Synthetic insecticides and fumigants are predominately used to control this pest. However, wide use of these chemicals manifest pesticidal residues and pest resistance necessitating to look for some other environmental-friendly alternatives such as nano-formulated botanical extracts. This study evaluated nano-formulated leaf extracts of some promising plant species against *S. oryzae* adults. Initial screening of the acetone extracts of neem (*Azadirachta indica* A. Juss.), guava (*Psidium guajava* L.), dhraik (*Melia azedarach* L.), tobacco (*Nicotiana tabacum* L.) and eucalyptus (*Eucalyptus camaldulensis* Dehn.) using grain treatment bioassay method showed maximum and significant weevil mortality by the extracts of *A. indica* and *P. guajava* i.e. 98 and 94%, respectively at 120 h post-exposure. These extracts were further nano-formulated using silver nitrate (AgNO₃). Toxicity and repellency of different concentrations (i.e. 10, 5, 2.5 and 1.25%) of these nano-formulated extracts were determined against *S. oryzae* weevils. Results showed significant mortality of the exposed weevil individuals by all treatments and this mortality response was directly proportional to the extract concentration and exposure time. Nano-formulated *A. indica* extract showed highest mortality (98.7%), followed by *M. azedarach* (97%), *N. tabacum* (96%) and *P. guajava* (84%) at 96 h post-exposure. Probit analysis exhibited minimum LC₅₀ for *A. indica* (0.62%), followed by *P. guajava* (0.66%), *N. tabacum* (0.71%) and *M. azedarach* (0.94%) at 96 h post-exposure. In case of repellency, 10% concentration of the nano-formulated *N. tabacum* extract exhibited highest repellency (54%), followed by *M. azedarach* (48%) and *P. guajava* (28%) after 24 h of exposure. Overall study results demonstrated the effectiveness of these nano-formulated phytoextracts against *S. oryzae* advocating their integration in eco-friendly management of stored grain insect pests.

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Authors' Contribution

MZM and ABMR conceived the idea and designed the study. MSS and UF performed experimentation and recorded data. AA and LAA analyzed the data and prepared results. MSS, UF and AA wrote the initial draft of manuscript. SS and ABMR technically proofread the manuscript. MZM and ABMR supervised the research work. LAA and SS provided technical and financial assistance for the study. All authors have read and approved the final version of the manuscript.

Key words

Stored grain pests, Rice weevil, Silver nanoparticles, Nano-formulation, Phytoextracts, Repellency bioassay, In-vitro toxicity

INTRODUCTION

Rice (*Oryza sativa* L. Poaceae) is one of the most imperative and essential staple crops, particularly in

Asian countries (Chandio *et al.*, 2016). Rice weevil, *Sitophilus oryzae* (L.) (Coleoptera: Curculionidae), is one of the most destructive pests of stored grains worldwide including rice. This pest causes substantial damage to stored rice in tropical and sub-tropical regions with losses reaching up to 40% (Rani *et al.*, 2019; Covele *et al.*, 2020). Synthetic insecticides and fumigants such as methyl bromide, magnesium and aluminum phosphides are the prime source to control *S. oryzae* infestations both at domestic and commercial levels (Nayak *et al.*, 2020; Wakil *et al.*, 2021). However, their use and effectiveness in stored grain sector have been limited due to their residual toxicity, health hazards, environmental contamination and problems of pest resistance and resurgence (Chayengia *et*

* Corresponding author: zeeshan.majeed@uos.edu.pk
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al., 2010; Hubert *et al.*, 2018).

These ecological consequences of extensive and continuous use of synthetic fumigants and other chemicals in stored grain sector necessitate looking for some biorational and environmental-friendly pest control options such as plant-based extracts (Isman, 2020). Phytoextracts can be effective alternatives to synthetic insecticides against stored grain insect pests (Dubey *et al.*, 2008). Due to their plant origin, they exhibit low persistency, low mammalian toxicity, high volatility and effective toxicity against stored grain insect pests (Regnault, 1997; Suteu *et al.*, 2020).

Moreover, nanotechnology is also being perceived as a promising field of research with a significant potential for future pest management (Muraisi *et al.*, 2022). Nano-formulations, particularly silver nanoparticles (AgNPs)-based formulations, have significant implications in various agricultural disciplines due to their high surface area to volume ratio, physical and chemical stability, low toxicity and simple surface chemistry (Scrinis and Lyons, 2007; Ale *et al.*, 2023). AgNPs synthesized through green methods can be effective carriers to transport agrochemicals to the targeted sites (Sankar and Abideen, 2015; Abbas *et al.*, 2024). They have proven to be highly effective against insect pests demonstrating considerable insecticidal, antifungal and antiviral properties (Chandran *et al.*, 2006; Srinivasan *et al.*, 2023). Therefore, insecticidal phytoextracts formulated with AgNPs could be an efficient strategy against stored grain insect pests including *S. oryzae* weevils.

The present research work aimed to evaluate and compare the nano-formulated phytoextracts against *S. oryzae* weevils under laboratory conditions. Firstly, available promising local phytoextracts were assessed for their effectiveness against *S. oryzae* adults. Secondly, the most effective phytoextracts were nano-formulated using AgNPs and were evaluated against *S. oryzae* adults to determine their LC_{50} and LT_{50} values. Lastly, the repellency of these nano-formulated phytoextracts was also assessed against *S. oryzae* adults.

MATERIALS AND METHODS

Rearing of *S. oryzae*

S. oryzae population was taken from the local grain market of Sargodha, Punjab, Pakistan from stored rice and was maintained in the Laboratory of Entomology, College of Agriculture, University of Sargodha. Culture was feed on clean and disinfected white fine rice (super basmati, unpolished). Fifty adult weevils were released in glass containers (20×30 cm) and the containers were closed with a fine-mesh muslin cloth. These containers

were kept in a growth chamber at $28\pm 2^{\circ}\text{C}$ temperature and $65\pm 5\%$ relative humidity. Weevil culture was reared up to F3 generation until its use in downstream bioassays.

Preparation of phytoextracts

Fresh leaves of neem (*Azadirachta indica* A. Juss.), guava (*Psidium guajava* L.), eucalyptus (*Eucalyptus camaldulensis* Dehn.), dharaik (*Melia azedarach* L.) and tobacco (*Nicotiana tabacum* L.) were collected from the plants in the vicinity of College of Agriculture, University of Sargodha during spring 2023. These leaves were washed with tap-water and were air-dried at room temperature (27°C) for two weeks. Shade dried leaves were powdered using an electric bleeder and were put in hermetic plastic bags. Using pure acetone as extraction solvent, the extraction of plant samples was carried out through Soxhlet apparatus (DH.WHM-12393, Daihan Scientific, South Korea) following the protocol described by Majeed *et al.* (2020) and Tayyab *et al.* (2022). Prepared phytoextracts were stored in dark-colored hermetic glass vials in refrigerator at 4°C until their use in experimentation.

Synthesis and characterization of AgNPs-based nano-formulated phytoextracts

Silver nanoparticles of effective phytoextracts were synthesized using a chemical reduction method. First of all, a solution was prepared by dissolving 0.017g of silver nitrate (AgNO_3) in 100 mL of deionized water. The solution was then heated to 80°C , while continuously stirring. Subsequently, the solution was transferred to a reflux condenser. To this solution, 10 mL of phytoextracts was added using a graduated pipette. The entire solution was heated in an oil bath at 100°C for duration of 30 min, while maintaining constant stirring. Finally, the solution was allowed to cool to room temperature (27°C). The formation of silver nanoparticles of both phytoextracts was confirmed and characterized using UV-visible spectrophotometer (UV-Vis, Perkin Elmer, Lambda 35), Raman Spectrometer (Peak Seeker Pro-785; Agiltron, USA) and Scanning Electron Microscope (SEM) (FEI Quanta 250, Hillsboro, OR, USA) following the protocols described by Jafir *et al.* (2021).

Toxicity bioassay against *S. oryzae* weevils

Treated grain bioassay method was used to determine the toxicity of phytoextracts against *S. oryzae* adults. First of all, 10% extracts were screened and then the most effective phytoextracts were formulated with silver (AgNO_3) nanoparticles. Experimental design was completely randomized with five replications for each of the four concentrations (*i.e.* 10, 5, 2.5 and 1.25%) of nano-formulated phytoextracts and a control (acetone)

treatment. Five grams of raw white fine rice (Super Basmati, unpolished) were treated with one milliliter of each treatment in 60 mm glass Petri plates and 10 individuals of *S. oryzae* (containing both male and female ones) were released in each Petri plate and these plates were incubated in dark in a growth chamber at $28 \pm 2^\circ\text{C}$ temperature and $65 \pm 5\%$ relative humidity. Mortality of the exposed insect individuals was recorded at 12, 24, 48, 72, 96 and 120 h post-exposure and was corrected using Abbott formula (Abbott, 1925).

Repellency bioassay

Half filter-paper disc method was utilized to assess the repellency potential of nano-formulated phytoextracts. In brief, Whatman filter paper (No.1) discs were cut into two halves according to glass Petri plate (60 cm). One half of each paper was treated with 1 ml of either 10 or 5% concentration of each treatment (phytoextracts) and other half was treated with acetone alone (control). After treating, both halves were placed at room temperature (27°C) for drying up to 10 min and were rejoined in 60 cm glass Petri plate. Ten adults of *S. oryzae* were released in each Petri plate and were covered with lid and were incubated $28 \pm 2^\circ\text{C}$ temperature and $65 \pm 5\%$ relative humidity. Data regarding the repellency of exposed insect individuals were recorded at 3, 18 and 24 h post-exposure (Elbrense *et al.*, 2022).

Statistical analysis

The percent mortality and repellency of tested *S. oryzae* adults are presented graphically. Apart from this graphical representation, factorial analysis of variance (ANOVA) was run to analyze the mortality and repellency data by keeping exposure time and treatment concentration as factors, followed by Honestly significant different (HSD) to compare treatment means at 95% probability level ($P < 0.05$). Median lethal concentration (LC_{50}) and median lethal time (LT_{50}) values were calculated by Probit analysis using POLO[®] statistical regression software. Prior to Probit analyses, data were corrected using Abbott's formula (Abbott, 1925). Percent repellency was determined by using a previously described equation (Elbrense *et al.*, 2022). Statistical interpretation of data was done using Statistix 8.1[®] (Abbott, 1925).

$$\text{PR} = \left[\frac{(C - T)}{C + T} \right] \times 100$$

Where PR is percent repellency, C is numbers of insects on control and T is numbers of insects on treatment.

RESULTS

Characteristics of nano-formulated phytoextracts

The most effective phytoextracts *i.e.* *A. indica*,

M. azedarach, *P. guajava* and *N. tabacum* were nano-formulated using silver (AgNO_3) particles and then the most effective two nano-formulations *i.e.* *A. indica* and *P. guajava* were further characterized using Raman spectroscopy, ultraviolet-visible (UV-vis) spectroscopy and scanning electron microscopy (SEM) techniques. All these determinations confirmed the formation of silver nanoparticles (AgNPs). The absorption spectra of nano-formulated *A. indica* and *P. guajava* extracts were determined by UV-visible spectroscopy in the wavelength range of 300–800 nm by taking deionized water as blank. Due to surface plasmon resonance, the color of solution was transformed from yellow to dark brown, which is thought to be the most important indicator of synthesis of AgNPs. The maximum absorbance spectrum of synthesized *A. indica* and *P. guajava* AgNPs was observed at 417 and 430 nm, respectively (Figs. 1, 2). Furthermore, the spectral measurement of AgNPs of nano-formulations of both extracts in liquid form was performed with Raman spectrometer (Peak Seeker Pro-785; Agiltron, USA) by a 785 nm laser as a source of excitation transferring a laser power of 50 mW with the help of a 40X objective lens.

The size and morphology of AgNPs of both nano-formulated extracts were determined by scanning electron microscope (SEM) at 100,000X magnification. The average size of both *A. indica* and *P. guajava* synthesized AgNPs was approximately 19 and 18 nm, respectively (Figs. 1, 2). Energy dispersive X-ray spectroscopy (EDX) related to SEM corroborated the presence of elemental silver by indicating the conversion of silver ions to silver nanoparticles. Furthermore, peaks of carbon, silver and oxygen appeared in case of aqueous extract of *A. indica* (Fig. 1), while the peaks of oxygen, potassium, chlorine, silicon and sodium appeared from the aqueous extract of *P. guajava* extract (Fig. 2).

Toxicity bioassays against *S. oryzae* adults

In preliminary screening, acetone extracts of five indigenous plant species were tested for their insecticidal potential against *S. oryzae* adults. Results of this bioassay in which weevils were exposed to 10% extracts of the plants showed that all extracts exhibited significant mortality ($F_{4,20} = 6.68$; $P = 0.0014$) of exposed weevils as compared to the control treatment (Supplementary Table I). Maximum mortality of *S. oryzae* adults was observed in case of extracts of *A. indica* (98%) and *P. guajava* (89%), followed by *M. azedarach* (87%) and *N. tabacum* (84%). Minimum weevil mortality (74%) was recorded for the extract of *E. camaldulensis* (Fig. 3A) Weevil mortality remained less than 5% in control treatment.

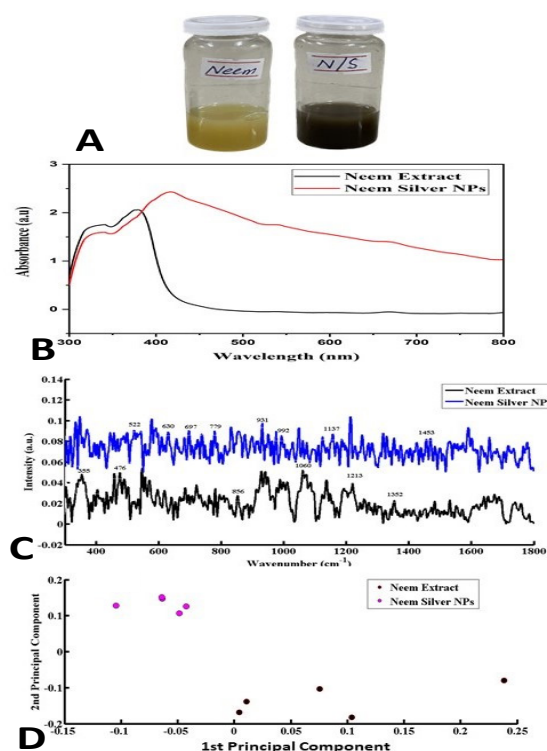


Fig. 1. Acetone extract of neem (*Azadirachta indica* L.) and its silver nanoparticles-based formulation (A) (aqueous solution of 1.0 mM AgNO₃ before (neem) and after (N/S) adding the botanical extract) and their Raman UV spectra (B), mean plots (C) and scatter plots (D) and (E) Scanning electron microscopic (SEM) image and energy dispersive x-ray spectroscopic (EDX) spectra and surface morphology (at 700 μ m resolution) of silver nanoparticles of acetone extract of neem (*Azadirachta indica* L.).

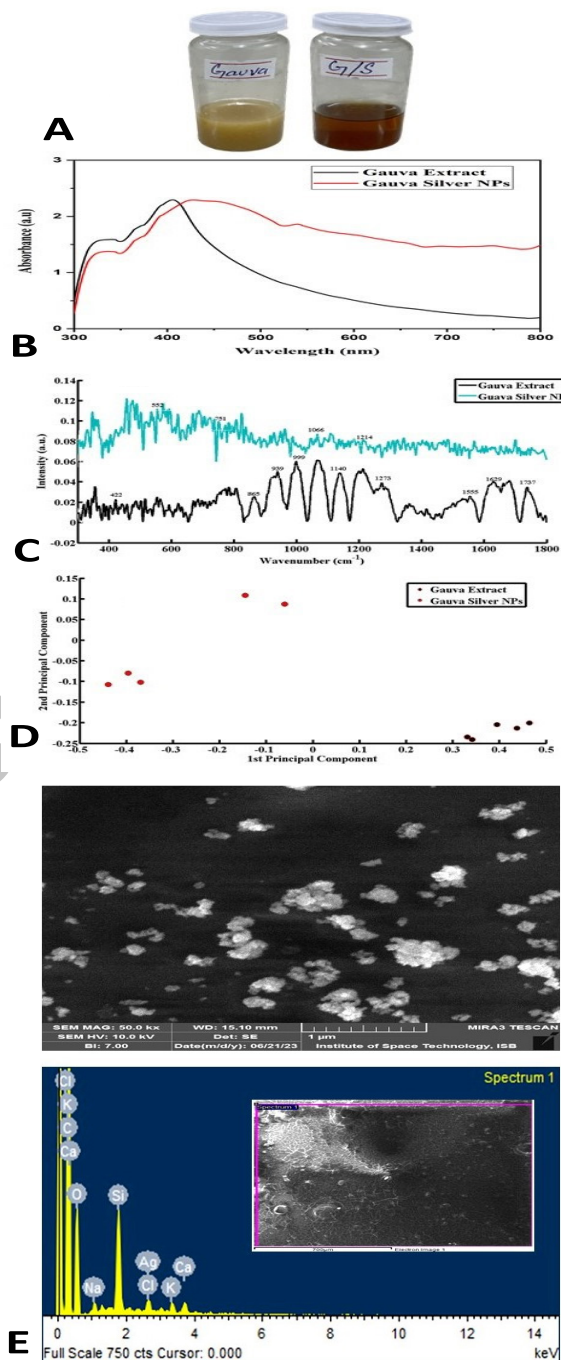


Fig. 2. Acetone extract of guava (*Psidium guajava* L.) and its silver nanoparticles-based formulation (A) (aqueous solution of 1.0 mM AgNO₃ before (guava) and after (G/S) adding the botanical extract) and their Raman UV spectra (B), mean plots (C) and scatter plots (D) and (E) Scanning electron microscopic (SEM) image and energy dispersive x-ray spectroscopic (EDX) spectra and surface morphology (at 700 μ m resolution) of silver nanoparticles of acetone extract of guava (*Psidium guajava* L.).

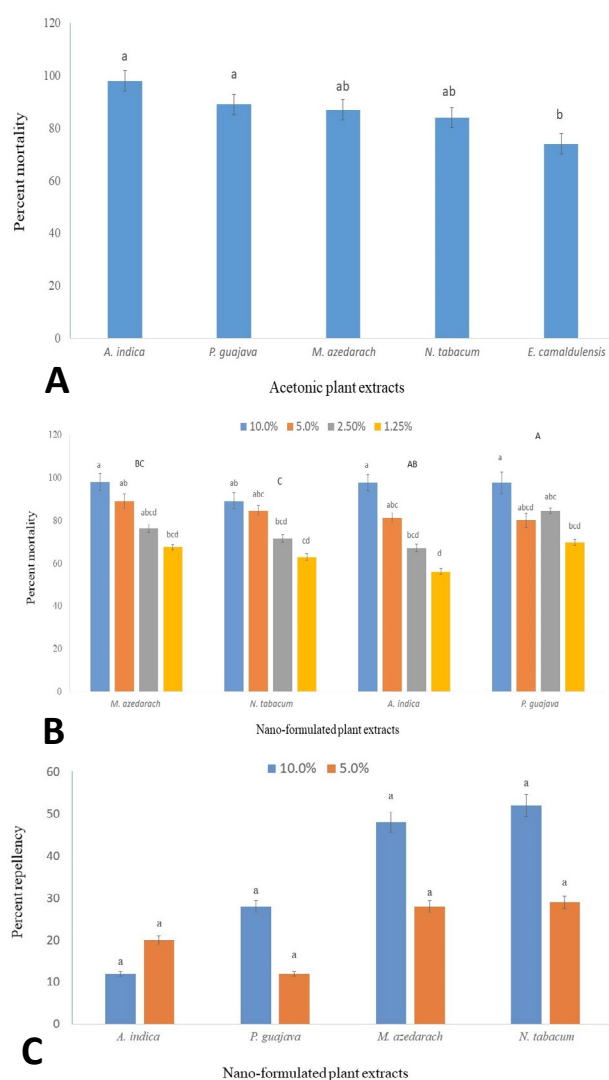


Fig. 3. Cumulative percent mortality (mean $\pm$ SE; n = 5) of adult individuals of rice weevil (*S. oryzae*) exposed to 10% acetone phytoextracts (A) different concentrations of nano-formulated acetone phytoextracts recorded at 96 h (B) and 24 h (C) post-exposure. Different small and capital letters at column tops indicate statistical difference between all treatments (one-way ANOVA; Tukey's HSD test at $\alpha=0.05$) and overall among the treatments (factorial ANOVA; Tukey's HSD test at $\alpha=0.05$), respectively.

In detailed toxicity bioassay, four phytoextracts (*i.e.* *A. indica*, *P. guajava*, *M. azedarach* and *N. tabacum*) which showed maximum and significant mortality of *S. oryzae* weevils were further formulated with silver nanoparticles (AgNPs) and were tested against *S. oryzae* adults using same protocol as done for previous bioassay. In this bioassay, different concentrations of the nano-formulated phytoextracts were used. Results of this bioassay clearly

demonstrated a significant effect on the mortality of *S. oryzae* adults by the treatments ($F_{3,64} = 7.53$, $P < 0.001$), their concentrations ($F_{3,64} = 39.70$, $P < 0.001$) and by their interaction ($F_{9,64} = 2.32$, $P = 0.0248$) (Supplementary Table II). On overall basis, nano-formulated extracts of *A. indica* and *P. guajava* exhibited highest weevil mortality (*i.e.* 98%) at 96 h post-exposure and were statistically different from other two extracts (Fig. 3B).

Probit analysis of the weevil mortality data demonstrated almost similar trend of toxicity of nano-formulated phytoextracts against *S. oryzae* and this trend was directly proportional to concentration of the extract and exposure time (Tables I and II). According to probit analysis, *A. indica*, *P. guajava* and *N. tabacum* appeared to be the most toxic extracts exhibiting minimum LC_{50} and LT_{50} values *i.e.* 0.60, 0.66 and 0.71% at 96 h of exposure, and 4.97, 7.57 and 13.7 h for 10% concentration of extracts, respectively (Tables I and II). While, minimum LC_{50} and LT_{50} values were recorded for the extract of *M. azedarach*.

Table I. Median lethal concentration (LC_{50}) values of nano-formulated acetone leaf extracts of indigenous plant species evaluated against adults of rice weevil *S. oryzae* under laboratory conditions.

Botanical extracts	Observation time (h)	LC_{50} (%)	95% upper and lower fiducial limits	X ² -value	P value*
<i>A. indica</i> (Neem)	12	13.96	7.29-158.81	83.27	< 0.05
	24	4.32	6.64 -7.65	109.18	< 0.05
	48	1.16	0.64-1.63	94.17	< 0.05
	72	0.72	0.28-1.13	93.22	< 0.05
	96	0.62	0.38-1.03	72.85	< 0.05
<i>M. azedarach</i> (Dhraiak)	12	3.78	2.83-5.06	117.32	< 0.05
	24	2.11	1.33- 2.86	161.44	< 0.05
	48	1.41	0.83 -1.91	148.40	< 0.05
	72	1.30	0.66 -1.84	211.82	< 0.05
	96	0.94	0.26 -1.49	242.40	< 0.05
<i>N. tabacum</i> (Tobacco)	12	3.49	1.99-6.06	122.91	< 0.05
	24	1.81	0.72- 2.77	146.35	< 0.05
	48	1.33	0.57-1.97	168.11	< 0.05
	72	0.93	0.29-1.49	196.05	< 0.05
	96	0.71	0.29-1.27	324.11	< 0.05
<i>P. guajava</i> (Guava)	12	5.83	3.43 -7.63	148.54	< 0.05
	24	4.11	2.53-7.65	147.20	< 0.05
	48	1.40	0.13-2.53	76.57	< 0.05
	72	0.79	0.29-1.27	74.44	< 0.05
	96	0.66	0.12- 1.14	117.10	< 0.05

*Since the significance level is less than 0.05, a heterogeneity factor is used in the calculation of confidence limits α denotes water used as solvent.

Table II. Median lethal time (LT₅₀) values of nano-formulated acetone leaf extracts of indigenous plant species evaluated against adults of rice weevil *S. oryzae* under laboratory conditions.

Botanical extracts	Concentration (%)	LT ₅₀ (h)	95% upper and lower fiducial limits	X ² -value	P value*
<i>A. indica</i> (Neem)	10	4.97	42.61-66.31	83.27	< 0.05
	5	19.74	15.73-37.47	109.18	< 0.05
	2.5	26.69	12.01-26.67	94.17	< 0.05
	1.25	52.15	7.3-19.61	93.22	< 0.05
<i>M. azedarach</i> (Dhraiik)	10	14.85	0.82-9.39	270.41	< 0.05
	5	12.90	2.69-23.61	300.85	< 0.05
	2.5	14.10	3.48-19.78	197.07	< 0.05
	1.25	36.85	27.69-48.13	94.19	< 0.05
<i>N. tabacum</i> (Tobacco)	10	13.75	3.79-11.02	168.77	< 0.05
	5	12.15	2.79-23.94	306.13	< 0.05
	2.5	14.10	3.47-19.78	197.07	< 0.05
	1.25	36.85	27.68-48.13	94.19	< 0.05
<i>P. guajava</i> (Guava)	10	7.57	7.34-19.61	180.56	< 0.05
	5	21.44	12.01-26.67	178.74	< 0.05
	2.5	34.69	15.73-37.47	162.15	< 0.05
	1.25	55.15	42.61-66.31	87.250	< 0.05

*Since the significance level is less than 0.05, a heterogeneity factor is used in the calculation of confidence limits α denotes water used as solvent.

Repellency bioassay against *S. oryzae* weevils

Analysis of variance (ANOVA) performed on the results of repellency bioassay revealed that the treatments (phytoextracts) ($F_{3,32} = 3.50$; $P = 0.0509$) and exposure time ($F_{1,32} = 3.21$; $P = 0.0225$) exhibited a significant effect on weevils repellency, while the interaction of both these factors exerted a non-significant effect ($F_{3,32} = 1.90$; $P = 0.4521$) on the repellence property of *S. oryzae* adults (Supplementary Table III). High concentration (10%) of nano-formulated *N. tabacum* extract exhibited the highest repellency effect (52%), followed by *M. azedarach* (48%), *P. guajava* (24%) and *A. indica* (12%). While, about 29, 28, 20 and 12% repellency was exhibited by the 5% extracts of *N. tabacum*, *M. azedarach*, *P. guajava* and *A. indica*, respectively (Fig. 3C).

DISCUSSION

Rice weevil *S. oryzae* has been a challenging stored products' pest being primarily controlled by the extensive applications of synthetic fumigants and other insecticides. Pest resistance, environmental contamination and health hazards are being manifested by this approach necessitating to look for other alternate pest control methods which

would be safe and environment-friendly such as using pesticidal phytoextracts (Isman, 2020; Wakil *et al.*, 2021).

This study therefore aimed to determine the insecticidal and repellency potential of some selected phytoextracts against *S. oryzae* adults. As nano-formulations of pesticidal compounds is being focused as a novel and effective approach (Fraceto *et al.*, 2016), the effective phytoextracts were further nano-formulated using silver nanoparticles. Results of preliminary bioassay with 10% of acetone phytoextracts demonstrated a significant mortality of *S. oryzae* adults by the extracts of *A. indica*, *P. guajava*, *N. tabacum* and *M. azedarach* causing considerable weevil mortality as compared to control. These results are in accordance with the findings of Chayengia *et al.* (2010), Khani *et al.* (2011) and Hashim *et al.* (2017) who reported considerable mortality of *S. oryzae* weevils by these phytoextracts. Particularly, neem (*A. indica*) and guava (*P. guajava*) appeared to be the most toxic to *S. oryzae* in our bioassays. *A. indica* is a well-known plant species with insecticidal properties (Chaudhary *et al.*, 2017; Ferreira and Alves, 2021).

Later on, AgNPs-based formulations of these phytoextracts were synthesized by green-route chemistry which is an eco-friendly and promising approach for stored grains insect pest management (Rehman *et al.*, 2021; Jasrotia *et al.*, 2022). The results of bioassay with nano-formulated phytoextracts against *S. oryzae* adults revealed a significant increase in percent mortality with the increase of concentration and exposure time. Our findings are consistent with many previous research reports (Debnath *et al.*, 2011; Stadler *et al.*, 2012; Zahir *et al.*, 2012; Rouhani *et al.*, 2013; Carbone *et al.*, 2020; Srinivasan *et al.*, 2023; Abdel-Megeed *et al.*, 2024). Stadler *et al.* (2012) evaluated nano-structured alumina (NSA) against *S. oryzae* and *Rhyzopertha dominica* and reported high mortality of *S. oryzae* adults 3 days post-treatment to NSA-treated wheat. Goswami *et al.* (2010) reported the toxicity of different surface functionalized hydrophilic nanoparticles *i.e.* silver (AgNPs), silica (SNPs), aluminum oxide (ANPs), zinc oxide (ZNPs) and titanium dioxide (TNPs) nanoparticles against *S. oryzae*. Insect mortality due to AgNPs treatment was about 85%, obtained at dose rates of almost 10 ml/kg of grain. Similarly, Rani *et al.* (2019) and Adnan *et al.* (2022) reported a significantly high mortality of adults of *S. oryzae* and other stored product insect pests with green-synthesized AgNPs mediated with the leaf extracts of *Moringa oleifera* (Moringaceae) and *Withania somnifera*, respectively.

The results of repellency bioassay revealed that all botanicals exhibited significant repelling effect to *S. oryzae* adults. In the present study, all nano-formulated phytoextracts showed moderate repellency at minimum exposure time of 3 h, while high repellency as recorded at

24 h post-exposure. These results are consistent with the findings of previous studies (Nerio *et al.*, 2009; Jayakumar *et al.*, 2017; Yeguerman *et al.*, 2022; Arafah *et al.*, 2023). The order of the repellency of phytoextracts at 10% was *N. tabacum* > *M. azedarach* > *P. guajava* > *A. indica* after 24 h of exposure time. Our results are in line with those of Kanmani *et al.* (2021) who reported 100% repellent effects of nano-formulated *N. tabacum* to *S. oryzae* and *C. maculatus* at 100 ppm of concentration. Zia-ul-Haq *et al.* (2014) reported that the repellent effect of *N. tabacum* and *M. azedarach* may be due to metabolic activities and sensitivity of insects to these botanicals. Similarly, Reeba and Helen (2021) and Shahzadi *et al.* (2019) reported highest repellency against *S. oryzae* and *T. castaneum* by plant mediated AgNPs prepared by aqueous extract of grapefruit (*Citrus paradisi*) and acetone essential oil of clove (*Syzygium aromaticum*), respectively.

CONCLUSION

On the basis of overall study results, it is concluded that silver nitrate (AgNO₃) based nano-formulated extracts of all tested plant species, particularly of neem (*A. indica*) and guava (*P. guajava*), exhibited a significant mortality of adults of rice weevil *S. oryzae*, while nano-formulated extracts of dhraik (*M. azedarach*) and tobacco (*N. tabacum*) caused considerable repellency of *S. oryzae* weevils as compared to control. These findings suggest that the commercial formulations of green nanoparticles could be developed to manage *S. oryzae* and other coleopterous stored grain pests. Moreover, the characterization of active components of phytoextracts and the validation of these phyto-formulations at large scale constitute the future perspectives of the study.

DECLARATIONS

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Data availability

All data taken and analyzed in this study are available

in the published manuscript.

Supplementary material

There is supplementary material associated with this article. Access the material online at: <https://dx.doi.org/10.17582/journal.pjz/20241008140626>

Statement of conflict of interest

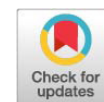
The authors have declared no conflict of interest.

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Supplementary Material

Toxicity and Repellency Potential of Silver Nano-Formulated Phytoextracts Against Rice Weevil *Sitophilus oryzae* (L.) (Coleoptera: Curculionidae)

Muhammad Salman Saeed¹, Umar Farooq¹, Muhammad Zeeshan Majeed^{1*},
Lamya Ahmed Alkeridis², Samy Sayed³, Abu Bakar Muhammad Raza¹ and
Asma Akram¹

¹Department of Entomology, College of Agriculture, University of Sargodha,
Sargodha 40100, Pakistan

²Department of Biology, College of Science, Princess Nourah Bint Abdulrahman
University, P.O. Box 84428, 11671 Riyadh, Saudi Arabia

³Department of Economic Entomology and Pesticides, Faculty of Agriculture, Cairo
University, Giza 12613, Egypt

Supplementary Table I. Analysis of variance (ANOVA) comparison table for mean mortality of adult weevils of *S. oryzae* exposed to 10% plant extracts recorded at 96 h post-exposure under laboratory conditions.

Source	DF	SS	MS	F	P
Treatment	4	1478.00	369.500	6.68	0.0014
Error	20	1106.00	55.300		
Total	24	2584.00			

Supplementary Table II. Analysis of variance (ANOVA) comparison table for mean mortality of adult weevils of *S. oryzae* exposed to different concentrations of nano-formulated plant extracts recorded at 96 h post-exposure under laboratory conditions.

Source	DF	SS	MS	F	P
Treatment	3	2575.7	858.57	7.53	0.0002
Concentration	3	13578.6	4526.20	39.70	0.0000
Treatment × concentration	9	2384.3	264.92	2.32	0.0248
Error	64	7296.1	114.00		
Total	79	25834.7			

Supplementary Table III. Analysis of variance (ANOVA) comparison table for mean repellency of adult weevils of *S. oryzae* exposed to different concentrations of nano-formulated plant extracts recorded at 96 h post-exposure under laboratory conditions.

Source	DF	SS	MS	F	P
Plant extract (PE)	3	1267.5	422.50	3.50	0.0509
Concentration (Conc.)	1	2722.5	2722.50	3.21	0.0225
PE × Conc.	3	2287.5	762.50	1.90	0.4521
Error	32	27120.0	847.50		
Total	39	33397.5			

* Corresponding author: zeeshan.majeed@uos.edu.pk
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