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Isolation and characterization of the insecticidal compounds in *Anacardium Occidentale* (cashew nut) shell liquid against the rice weevil, *Sitophilus oryzae* L. (Coleoptera: Curculionidae)

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Abstract
In the present study, insecticidal compounds in cashew nut shell liquid against *Sitophilus oryzae* were isolated and characterized. Bioassay guided isolation led to the isolation and identification of three major cardanol derivatives, (*Z*)-3-(8- pentadecenyl)phenol (1), (8*Z*,11*Z*)-3-(8,11-pentadecadienyl)phenol (2) and (8*Z*,11*Z*,14*Z*)-3-(8,11,14-pentadecatrienyl)phenol (3). Of the three compounds, compound 3 with a yield of 36.55 mg in the 1 g equivalent of extract, showed the highest insecticidal activity with LC_{50} value 60.36 mg/mL (95% CL = 64.52 – 126.41) against adult *S. oryzae*. These results suggest that cashew nut shell liquid has insecticidal potential and can be used in the management stored product beetles especially *S. oryzae*.

Keywords: *Sitophilus oryzae*, Cashew Nut Shell Liquid, Cardanol, Bioactivity, (8*Z*,11*Z*,14*Z*)-3-(8,11,14-pentadecatrienyl) phenol

1. Introduction
Stored product insect pests pose several threats to global food security. Food losses due to insect infestation during storage are a serious problem in agricultural practices especially in developing countries [1]. These pests feed on the stored grains, reducing them to powder and in some cases, infesting them with their webbings and frass, thereby reducing their economic and nutritive values [2]. The rice weevil, *Sitophilus oryzae* L. (Coleoptera: Curculionidae) is a major pest of stored rice in tropical and temperate regions of the world [3]. The female uses strong mandibles to chew a hole into the rice after which it deposits a single egg within the hole, sealing it with secretions from its ovipositor [2]. The larva develops within the rice, hollowing it out while feeding. It then pupates and emerges 2–4 days after eclosion [2]. Thus, both adult and larval stages contribute to the damage caused by this insect.
Several control measures have been employed in managing stored product insect pests including the use of botanicals. Some plants such as *Zanthoxylum zanthoxyloides* Lam, *Moringa oleifera* Lam, *Ocimum canum* Sims and *Securidaca longepedunculata* Fresen among others have been found to be insecticidal active against stored product pests [4-7].
The cashew tree *Anacardium occidentale* L. is a tropical tree of great economic importance. It is native to tropical America, from Mexico to West Indies to Brazil and Peru and currently being cultivated in other tropical regions especially in coastal areas [8]. The most important product of the cashew tree is probably the nut, which is used as confectionery. While the other parts of the tree have traditionally been used for snake-bites and other folk remedies the cashew nut shell yields derivatives that can be used in many applications from lubricants to paints [9].
One of such derivatives is the cashew nut shell liquid (CNSL). This is a caustic, viscous, dark liquid and is a natural source of saturated and unsaturated long chain phenols and constitutes about 25% of the cashew weight and 30% - 35% of the nut shell weight [10]. The CNSL can be classified into two types per the extraction method utilized: solvent - extracted immature CNSL and heat - extracted technical CNSL [11] and the composition of the CNSL varies depending on which extraction method used [12]. However, CNSL whether cold solvent extracted or heat extracted is known to contain anacardic acid, cardol, cardanol and other

polymeric materials [13]. The common feature of all the components of CNSL is the presence of hydrophobic side chain differing in the degree of unsaturation, approximately 48% - 49% monoene, 16% - 17% diene and 29% - 30% triene [14].

The biological activities of the various components of CNSL have attracted considerable attention, in the areas of fungicidal activity [15], anti-inflammatory activities [16], anti-oxidant activity [17] and xanthine oxidase inhibition [18]. However very little work has been done on the insecticidal activities of the components of CNSL against stored product beetles.

In the present study using bioassay guided isolation, we isolated and identified the bioactive components of the cashew nut shell liquid responsible for insecticidal activities against *S. oryzae*.

2. Materials and methods

2.1 Insect rearing

The adult beetles were cultured in brown rice under a temperature of 26 ± 2 ºC, 70% relative humidity and 12 L: 12D photo regime in the insect breeding house of the chemical ecology laboratory, Kochi University, Japan.

2.2 Extraction of cashew nut shell liquid

The cashew nuts with their shells were harvested from the University Farms, University of Ghana, Legon in March 2014. The shells were removed from the nuts and were dried in the laboratory under room temperature. The dried shells (100.13 g) were blended and the shell liquid was extracted twice using 4.0 L of methanol. The concentrations of the crude extract (22.58 g) were adjusted with methanol for bioassay at 0.5 g of the extract equivalent (eq.)/mL, 1.0 g eq./mL and 2.0 g eq./mL.

2.3 Insecticidal activity bioassay method

Ten adult insects were each dipped in turns into each sample and then transferred into clean Petri dishes containing brown rice. Six replicates were made and these were observed daily until there were no changes in insect mortalities. An insect was considered dead if it did not respond to probing of a blunt probe [19]. All the isolated and separated components of the methanol extract whose bio activities were tested were maintained at a concentration of 2.0 g eq./mL.

2.4 Isolation of bioactive components

The methanol extract (11.71g) of the CNSL was dissolved in 281.0 mL of H_2O, and then extracted twice with 197 mL each of hexane, diethyl ether and ethyl acetate (AcOEt).

The hexane layer (dry weight 5.85g) was chromatographed on a Silica gel open column (175.5g, 30 mm i.d. x 496.6 cm). The column was eluted with 3.6 L each of hexane, 5% AcOEt in hexane, 30% AcOEt in hexane, 70% AcOEt in hexane, AcOEt and methanol. The 5% AcOEt in hexane fraction (dry weight 4.28 g) was submitted to a normal phase Silica gel HPLC (Column: Cosmosil 5SL Waters, 10 mm i.d. x 250 mm) eluted with 2% AcOEt in hexane at a flow rate of 3 mL/min and UV detector (254 nm). Compounds 1(t_R = 20.05 min - 22.99 min; yield: 54.2 mg), 2 (t_R = 23.00 min - 24.99 min; yield: 25.5 mg) and 3 (t_R = 25.00 - 28.99 min; yield: 73.1 mg) were isolated.

2.5 Dose-response relationship and LC_{50} of isolated compounds

Six different concentrations (316.0 mg/mL, 174.0 mg/mL, 87.0 mg/mL, 43.5 mg/mL, 21.75 mg/mL and 10.88 mg/mL) of the isolated compounds 1, 2 and 3 were prepared, and used for the bioassay. The control was made up of only methanol and the set up was observed periodically for insect mortalities until there were no changes in insect mortalities over the period. Data recorded on insect mortalities were analyzed statistically and the LC_{50} values of the isolated compounds were determined.

2.6 Instrument

GC-MS data (EI-positive) were recorded with a GC 2010 Plus, (Shimadzu) equipped with HP-5 MS Column (Cross linked 5% PH ME Siloxane, 30 m x 0.32 mm i.d. x 0.25 µm Film thickness). The carrier gas was Helium, at a flow rate of 11.7 mL/min, the split ratio was 1:10, detector and vaporizer temperature was held at 300 ºC. The column temperature was initially at 150 ºC for 6 minutes, raised to 300 ºC at 5 ºC / min and held for 10 minutes. The identification of the components was confirmed by comparison with the mass spectra of compounds documented by the National Institute of Standards and Technology (NIST) 14 online library. ^{1}H and ^{13} C- NMR spectra ($CDCl_3$) were measured with a Jeol JNM-ECX500, TMS as internal standard.

2.7 Statistical and analysis

Data collected was analyzed using GenStat Statistical Package 9.2 (9th Edition, VSNi, London, UK) and SPSS Statistics (IBM, USA). Analysis of variance was run at 95% confidence level and means separation was done using Tukey's HSD. Data involving counts were transformed using square root ($y = \sqrt{x}$) transformation while those involving percentages were transformed using arcsine ($y = \sin^{-1}\sqrt{x/100}$ $\sqrt{x/100}$) transformation before analysis. Mean (± SE) of untransformed data are reported.

3. Results

The three concentrations (0.5 g eq./mL, 1.0 g eq./mL and 2.0 g eq./mL) of the crude extracts of the CNSL showed significantly ($Fpr < 0.05$) different levels of insecticidal activities against *S. oryzae* (Table 1). As the concentration of CNSL extract increases, higher percentage mortalities were observed. The highest percentage mortality of 80.0 ± 13.3% was observed in the 2.0 g eq./mL treatment after 48 hours while the control which consisted of only methanol had no mortalities. These results indicate the presence of potential insecticidal compounds present in the crude extracts of the CNSL.

Table 1: Percentage mortality of *S. oryzae* to the different concentrations of methanol extracts of Cashew nut shell liquid

Treatment (g eq./mL)	*% Mortality					
	0 h	24 h	48 h	72 h	96h	120 h
2.0	0.0 ± 0.0^a	33.3 ± 11.1^c	80.0 ± 13.3^c	80.0 ± 13.3^d	80.0 ± 13.3^d	80.0 ± 13.3^d
1.0	0.0 ± 0.0^a	23.3 ± 9.7^b	23.3 ± 9.2^b	30.0 ± 6.3^c	36.7 ± 9.2^c	36.7 ± 9.2^c
0.5	0.0 ± 0.0^a	3.3 ± 3.3^a	6.7 ± 3.3^a	13.3 ± 3.3^b	13.3 ± 3.3^b	13.3 ± 3.3^a
Control	0.0 ± 0.0^a	0.0 ± 0.0^a	0.0 ± 0.0^a	0.0 ± 0.0^a	0.0 ± 0.0^a	0.0 ± 0.0^a

Values with different small letters within the columns are significantly different ($Fpr. < 0.01$) based on Tukey's HSD test following ANOVA. * Each value is expressed as mean ± S. E (n=6 x 10)

When the crude CNSL extract was dissolved in H_2O and extracted with hexane, diethyl ether and AcOEt used for bioassay, no mortalities were observed in the AcOEt layer, water layer as well as in the control (Table 2A). On the other hand, the hexane layer exhibited the highest activity recording 100.0 ± 0.0% mortality after 24 hours while the ether layer showed a weaker activity with a percentage mortality of 10.0 ± 0.0% after 72 hours. These results showed that the activity of the crude CNSL extracts was retained in the hexane layer.

The insecticidal activities of the various fractions obtained from the silica gel open column chromatography of the active hexane layer are shown in Table 2B. The 100% hexane, 30% AcOEt in hexane, 70% AcOEt in hexane, 100% AcOEt and 100% methanol fractions showed no insecticidal activity of 0.0 ± 0.0% mortality against *S. oryzae*. The 5% AcOEt in hexane fraction on the other hand, showed percentage mortality of 93.3 ± 4.4% after 24 hours which was significantly ($Fpr < 0.01$) higher than the other isolated fractions. This was not significantly different ($Fpr > 0.01$) from the percentage mortality 100 ± 0.0% yielded by the combination of all the fractions (in equal volumes) after 24 h.

Table 2A: Percentage mortality of S. oryzae to the four separated layers from liquid-liquid partitioning of the methanol extracts of Cashew nut shell liquid

Treatment (Per 2g eq./mL)	*% Mortality					
	0 h	24 h	48 h	72 h	96 h	120 h
Hexane Layer	0.0 ± 0.0^a	100.0 ± 0.0^b	100.0 ± 0.0^b	100.0 ± 0.0^c	100.0 ± 0.0^c	100.0 ± 0.0^c
Diethyl Ether Layer	0.0 ± 0.0^a	3.3 ± 3.3^a	6.7 ± 3.3^a	10.0 ± 0.0^b	10.0 ± 0.0^b	10.0 ± 0.0^b
Ethyl Acetate Layer	0.0 ± 0.0^a	0.0 ± 0.0^a	0.0 ± 0.0^a	0.0 ± 0.0^a	0.0 ± 0.0^a	0.0 ± 0.0^a
Water Layer	0.0 ± 0.0^a	0.0 ± 0.0^a	0.0 ± 0.0^a	0.0 ± 0.0^a	0.0 ± 0.0^a	0.0 ± 0.0^a
All layers	0.0 ± 0.0^a	100.0 ± 0.0^b	100.0 ± 0.0^b	100.0 ± 0.0^c	100.0 ± 0.0^c	100.0 ± 0.0^c
Control	0.0 ± 0.0^a	0.0 ± 0.0^a	0.0 ± 0.0^a	0.0 ± 0.0^a	0.0 ± 0.0^a	0.0 ± 0.0^a

Values with different small letters within the columns are significantly different ($Fpr. < 0.01$). based on Tukey's HSD test following ANOVA. * Each value is expressed as mean ± S. E (n=6 x 10)

Table 2B: Percentage mortality of *S. oryzae* to the various fractions from the Silica-gel open column chromatography of the hexane layer of Cashew nut shell liquid

Treatment (Per 2g eq./mL)	*% Mortality					
	0 h	24 h	48 h	72 h	96 h	120 h
100% Hexane	0.0 ± 0.0^a	0.0 ± 0.0^a	0.0 ± 0.0^a	0.0 ± 0.0^a	0.0 ± 0.0^a	0.0 ±0.0^a
5% AcOEt in hexane	0.0 ± 0.0^a	93.3 ± 4.4^b	93.3 ± 4.4^b	93.3 ± 4.4^b	93.3 ± 4.4^b	93.3 ±4.4^b
30% AcOEt in hexane	0.0 ± 0.0^a	0.0 ± 0.0^a	0.0 ± 0.0^a	0.0 ± 0.0^a	0.0 ± 0.0^a	0.0 ±0.0^a
70% AcOEt in hexane	0.0 ± 0.0^a	0.0 ± 0.0^a	0.0 ± 0.0^a	0.0 ± 0.0^a	0.0 ± 0.0^a	0.0 ±0.0^a
100% AcOEt	0.0 ± 0.0^a	0.0 ± 0.0^a	0.0 ± 0.0^a	0.0 ± 0.0^a	0.0 ± 0.0^a	0.0 ±0.0^a
100% MeOH	0.0 ± 0.0^a	0.0 ± 0.0^a	0.0 ± 0.0^a	0.0 ± 0.0^a	0.0 ± 0.0^a	0.0 ±0.0^a
All fractions	0.0 ±0.0^a	100.0 ± 0.0^b	100.0 ± 0.0^b	100.0 ± 0.0^b	100.0 ± 0.0^b	100.0 ±0.0^b
Control	0.0 ± 0.0^a	0.0 ± 0.0^a	0.0 ± 0.0^a	0.0 ± 0.0^a	0.0 ± 0.0^a	0.0 ±0.0^a

Values with different small letters within the columns are significantly different ($Fpr. < 0.01$). based on Tukey's HSD test following ANOVA. * Each value is expressed as mean ± S. E (n=6 x 10)

The 5% AcOEt in hexane fraction was submitted to HPLC and three compounds {1 (t_R = 20.05 min - 22.99 min), 2 (t_R = 23.00 min - 24.99 min) and 3 (t_R = 25.00 - 28.99 min)} were isolated (Fig 1).

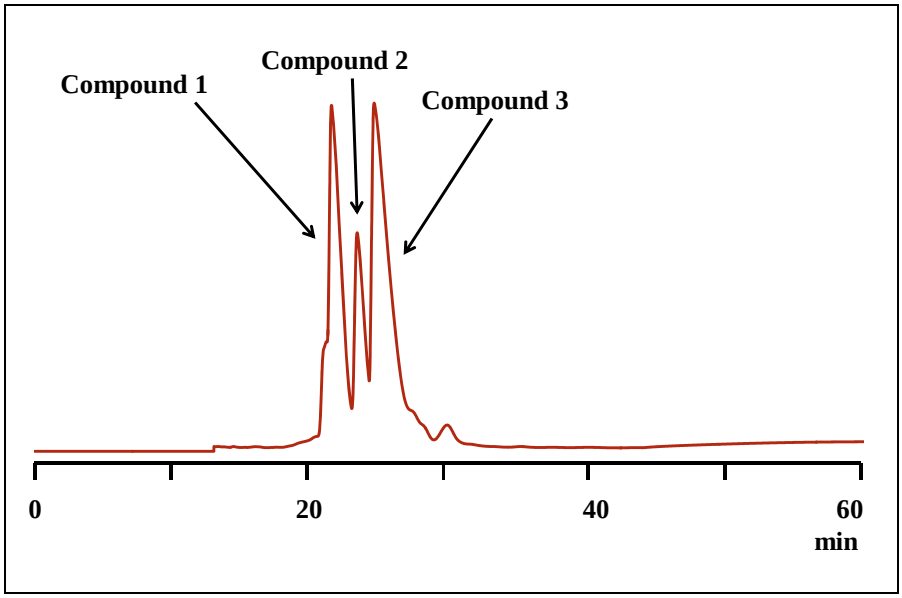

Fig 1: HPLC profile for the 5% AcOEt in hexane fraction showing the peaks for isolated compounds 1, 2 and 3. [1 (t_R = 20.05 min - 22.99 min), 2 (t_R = 23.00 min - 24.99 min) and 3 (t_R = 25.00 - 28.99 min)]

The amount of compounds 1, 2 and 3 isolated from the 2.0 g equivalent extract of the CNSL are 48.2 mg, 25.5 mg and 73.1 mg respectively. This means that the 5% AcOEt in hexane fraction that was submitted to the normal phase HPLC was rich in compound 3.

All the isolated compounds at 2.0 g eq./mL showed significantly different (F*pr* < 0.01) insecticidal activities against the adult *S. oryzae* (Table 3). Compound 2 as well as the control showed 0.0 ± 0.0% mortality each, whiles compound 1 showed a weaker insecticidal activity of 23.3 ± 4.4%. Compound 3 on the other hand showed a significantly (F*pr* < 0.01) higher insecticidal activity of 83.0 ± 8.9% mortality. These results therefore suggest that the main compound responsible for the insecticidal activity of CNSL against *S. oryzae* is compound 3.

Table 3: Percentage mortality of *S. oryzae* to the HPLC fractions isolated from the 5% AcOEt in hexane fraction

Treatment (Per 2 g eq./mL)	*% Mortality					
	0 h	24 h	48 h	72 h	96 h	120 h
Compound 1	0.0 ± 0.0^a	23.3 ± 4.4^b	23.3 ± 4.4^b	23.3 ± 4.4^b	23.3 ± 4.4^b	23.3 ± 4.4^b
Compound 2	0.0 ± 0.0^a	0.0 ± 0.0^a	0.0 ± 0.0^a	0.0 ± 0.0^a	0.0 ± 0.0^a	0.0 ± 0.0^a
Compound 3	0.0 ± 0.0^a	83.3 ± 8.9^c	83.3 ± 8.9^c	83.3 ± 8.9^c	83.3 ± 8.9^c	83.3 ± 8.9^c
All Compounds	0.0 ± 0.0^a	100.0 ± 0.0^d	100.0 ± 0.0^d	100.0 ± 0.0^d	100.0 ± 0.0^d	100.0 ± 0.0^d
Control	0.0 ± 0.0^a	0.0 ± 0.0^a	0.0 ± 0.0^a	0.0 ± 0.0^a	0.0 ± 0.0^a	0.0 ± 0.0^a

Values with different small letters within the columns are significantly different (F*pr*. < 0.01). based on Tukey's HSD test following ANOVA.* Each value is expressed as mean ±S. E (n=6 x 10).

The GC-MS analysis of the three compounds indicates the presence of an aromatic ring (specifically a phenol group) in each compound with different degrees of saturation on the adjoining aliphatic carbon chain. Together with the ^{13}C and ^{1}H NMR data which was consistent with literature data, [17, 18] the compounds were identified. Compound 1 was identified as (*Z*)-3-(8-pentadecenyl)phenol, compound 2 was identified as (8*Z*,11*Z*)-3-(8,11-pentadecadienyl)phenol and compound 3 was identified as (8*Z*,11*Z*,14*Z*)-3-(8,11,14-pentadecatrienyl) phenol (Fig. 2) These three compounds are collectively called cardanol.

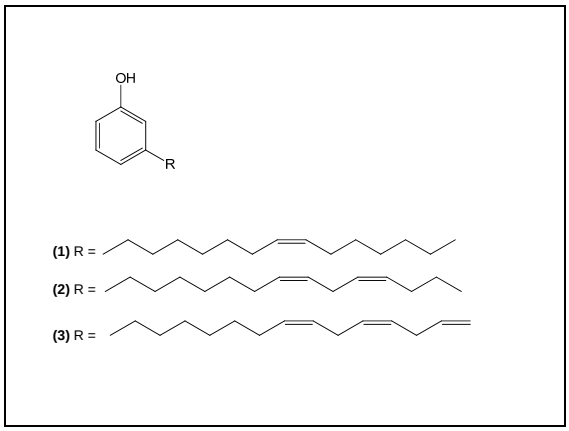

Fig 2: Chemical structures of cardanols [(*Z*)-3-(8- pentadecenyl) phenol (1), (8*Z*,11*Z*)-3-(8,11-pentadecadienyl)phenol (2) and (8*Z*,11*Z*,14*Z*)-3-(8,11,14 –pentadecatrienyl)phenol (3)].

The three compounds, 1, 2 and 3 showed dose dependent toxicities towards the mortalities of *S. oryzae* (Table 4). Percentage mortalities of *S. oryzae* increased with increasing concentrations of each of the compounds however at the same dose of each of the three compounds, compound 3 was found to be more toxic to *S. oryzae* than 1 and 2. For instance, at a concentration of 316.0 mg/mL for all three compounds, percentage mortalities of 85.0 ± 0.0%, 50. 0 ± 5.0% and 100 ± 0.0% were observed for compound 1, 2 and 3 respectively. LC_{50} values were determined to be 121.56 mg/mL (95% CL = 399.78 - 1041.13) for compound 1, 325.53 mg/mL (95% CL = 201.22 - 526.67) for compound 2, and 60.36 mg/mL (95% CL = 64.52 - 126.41) for compound 3 (Table 5). These results also suggest that, against *S. oryzae*, compound 3 was 5.4-folds more toxic than compound 2 which showed the least insecticidal activity. Therefore, among the three compounds, compound 3 (8*Z*, 11*Z*, 14*Z*)-3-(8,11,14-pentadecatrienyl) phenol is the most active insecticidal compound.

Table 4: Effect of different amounts (mg/mL) of compounds 1, 2 and 3 on the mortalities of *S. oryzae*.

Concentration (mg/mL)	*% Mortality		
	Compound 1	Compound 2	Compound 3
316.0	85.0 ± 0.0^a	50.0 ± 5.0^b	100.0 ± 0.0^c
174.0	85.0 ± 0.0^a	25.0 ± 5.0^b	100.0 ± 0.0^c
87.0	25.0 ± 0.0^a	10.0 ± 0.0^b	70.0 ± 5.0^c
43.5	0.0 ± 0.0^a	0.0 ± 0.0^a	30.0 ± 0.0^b
21.75	0.0 ± 0.0^a	0.0 ± 0.0^a	10.0 ± 0.0^b
10.88	0.0 ± 0.0^a	0.0 ± 0.0^a	0.0 ± 0.0^a
Control	0.0 ± 0.0^a	0.0 ± 0.0^a	0.0 ± 0.0^a

Values with different small letters within the same row are significantly different (*P*<0.05). Those with same small letters means no significant difference (*P*>.05) based on Tukey's HSD test following ANOVA. * Each value is expressed as mean ± Standard error of 6 replicates after 168 h.

Table 5: LC_{50} of isolated compounds against *S. oryzae*

Compound	LC_{50} [a] mg/mL (95% CL[b])	Slope ± SE	RT[c]	[d]X^2 (df)	R^2
1	121.56 (399.78 – 1041.13)	3.12 ± 0.07	2.7	14.0 (12)	0.79
2	325.53 (201.22 – 526.67)	2.28 ± 0.11	1.0	21.0 (18)	0.99
3	60.36 (64.52 – 126.41)	3.00 ± 0.08	5.4	24.0 (24)	0.99

[a] LC_{50} values were calculated after 120 h of exposure at 27± 2 °C and 70% relative humidity (n=6 x10)

[b] 95% lower and upper confidence limits are shown in parenthesis

[c] Relative toxicity = LC_{50} value of Compound **2**/LC_{50} value of each compound

[d]X^2 (df): Chi-square values (degrees of freedom)

4. Discussion

Cold solvent extracted CNSL consists of various amounts of the mixtures of anacardic acids, cardols and cardanols [22]. The common feature of all the components is the presence of an aromatic ring and hydrophobic side chains, differing in the degree of unsaturation, approximately 5% - 8% saturated, 48% - 49% monoene, 16% - 17% diene and 29 – 30% triene [14].

The present study led to the isolation of three derivatives of cardanol with various insecticidal activities against adult *S. oryzae*. Even though anacardic acids and cardols are also known to show antibacterial, larvicidal, and insecticidal activities [12, 23, 24], they were not isolated in this experiment. This may suggest that although they are present in CNSL; they do not show any insecticidal activities against *S. oryzae* as per the results of the present study. Therefore, the bioactivities of compounds in CNSL are considered be specific to organisms involved.

Cardanols refer to the decarboxylated derivatives of naturally occurring anacardic acid found in CNSL. The cardanol mixture includes more than one compound because the composition of the side chain varies in its degree of saturation [25]. As shown by the results of the present studies, the three cardanol derivatives have the same phenol ring but varied degrees of saturation on the alkyl side chain. They have been isolated and identified as the main bioactive compounds in CNSL showing a positive response to 2,2-diphenyl-1-picrylhydrazyl (DPPH) scavenging assay, xanthine oxidase assay as well as Salmonella cerevisiae assay [11, 26,27,28].

Bioactivities of naturally occurring compounds are often associated with the presence of one or two functional groups present in them. The antioxidant properties of phenols are due to their oxyreduction properties, which allow them to act as reducing agents, hydrogen donors and singlet oxygen elimination [29]. The linear side chain and its degree of unsaturation are both required for enhancing bioactivities of the compounds found in CNSL.

Kubo *et al*. [30] indicated that anacardic acid with alkyl triene side chain showed a higher bioactivity against *Streptococcus mutans* (ATCC 2517) than anacardic acid with a saturated alkyl side chain. Oliviera *et al*. [31] also concluded that the unsaturated side chain of 15 carbon atoms of anacardic acid is more strongly associated with the higher larvicidal activities of anacardic acid. In the current experiment, the cardanol with tri- unsaturated alkyl chain showed the highest insecticidal activity against *S. oryzae* at the same concentration with the other two derivatives. This is consistent with the findings of Lomonaco *et al.* [32] who showed that the hydrogenation of the double bonds in the side chain of cardanol and cardol resulted in decreased larvicidal activities against *Aedes aegypti*. Andrade *et al*. [26] indicated that the unsaturation on the long side chain could be an important factor in the activity of cardanol and therefore inferred that the allyl substituents could trap both alkyl and peroxy radicals and lead to an increase in activity. Therefore, it is important to note that the linear alkyl side chain and the degree of its unsaturation are both required for enhancing the insecticidal activity of cardanol against *S. oryzae* as shown by the results of the present study.

5. Conclusion

Considering the continual search for renewable and biodegradable ways to manage stored product pests, we believe that, (8*Z*, 11*Z*, 14*Z*)-3-(8,11,14–pentadecatrienyl) phenol justifies CNSL as an insecticide for managing stored product pests especially *S. oryzae*, based on the results of present studies. However, it is important to consider the toxic and corrosive properties of CNSL during its handling

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