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Insecticidal characteristics and mechanism of a promising natural insecticide against saw-toothed grain beetle

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The prevention of grain storage pests is a universal concern all over the world. It is in high demand to explore novel, safe and green insecticidal techniques to address such concerns. In this work, both raw and calcined diatomite were used as a natural insecticide to remove common grain storage pests with improved lethal effect on the saw-toothed grain beetle. Interestingly, the raw diatomite showed higher insecticidal efficiency than the calcined diatomite, and its associated insecticidal properties and preparation conditions were also optimized through orthogonal tests. The optimal conditions for processing the raw diatomite insecticide were identified as follows: the diatomite dust was 500 mesh (A_3), the temperature was 25 °C (B_1), the relative humidity was 65% (C_2), the diatomite dosage was 20 g m⁻² (D_2), the influence factor order was $C \geq D > A > B$. The observation of surface morphology indicated that the raw diatomite had a complete, multi hole surface morphology and good adsorption performance, whereas the structure of the calcined diatomite was uncomplete with collapsed pores, resulting in poor adsorption performance. The special pore structure and excellent adsorption capacity of diatomite make the stored grain pests lose water to lethal effect. Acute toxicity and long-term toxicity tests in mice showed that diatomite has no harmful effects on mammals. The findings from our work led to a green and effective approach in producing a highly efficient and safe storage grain insecticide.

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1. Introduction

With natural disasters and epidemics in 2020, locust infestations in India and Pakistan, bushfires in Australia, influenza virus in the United States and Covid-19 around the world, having sufficient is essential for human health.^{1,2} Enough food, feed, fiber, and biofuel to meet society's needs has been the goal of agriculture for thousands of years.³ As global populations continue to increase and the climate changes,⁴ agricultural productivity will be challenged.⁵ Since the early 1990s, a number of countries have begun to build up grain storage networks, enabling the establishment of grain reserve systems.⁶ However, food storage has some common problems, such as heating inside the grain pile,⁷ insect pests,⁸ mildew,⁹ mites¹⁰ and so on. In Brazil, losses in grain yield may range from 40 to 100%; annual losses ranging between 90 000 and 280 000 tons of grain would be enough to feed 6–20 million adult Brazilians. According to statistics, every year the food loss world-wide caused by pests and microorganisms accounts for 10–18% of the total grain production, and the food loss caused by pests in developed countries might rise up to

30%.¹¹ Thus, the control of stored grain pests is universal concern. Most of the stored grain pests belong to the order Coleoptera and Lepidoptera of the class insecta. There are about 600 species of known grain pests in the world, and more than 30 kinds are common and important types. Among these stored grain pests, the most common ones in grain depots in China include the saw-toothed grain beetle, *Oryzaephilus surinamensis* (L.) (Coleoptera:Silvanidae);¹² corn weevil *Oryzaephilus surinamensis* (L.) (Coleoptera:Silvanidae);¹³ grain beetle *Oryzaephilus surinamensis* (L.) (Coleoptera:Silvanidae) and *Tribolium ferrugineum Paracarida* (L.) (Coleoptera:Silvanidae)¹⁴ etc. For stored grain pests, researchers have done a lot of research on natural insecticide. For instance, plant extracts and essential oils prepared from the aerial parts of *Cinnamomum camphora*, *Ocimum basilicum*, *Chenopodium ambrosioides*, and seeds of *Pimpinella anisum* can be used for stored-product insect control because they constitute a rich source of bioactive chemicals.¹⁵ There are also many studies on natural pesticides. *E.g.*, datura leaf extract (DLE) is used to store two important insect pests on stored rice, *Trogoderma granarium* and *Sitophilus oryzae*, under laboratory conditions.¹⁶ Furthermore, spraying kaolin on cotton plants was used to prevent boll weevil damage.¹⁷ And new biopesticide water-based formulations based on sol-gel microencapsulated orange oil is used to cotton aphid *Aphis gossypii* Glover (Hemiptera:Aphididae).¹⁸

When many insect pests occur, the water content of grain will be changed with the insect pests, causing heat, mildew or

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caking in the grain storage pile, which in turn form a beneficial living environment for the late emergence of meat-eating insect pests. At the same time, the feces produced by moth-eaten grain will increase the possibility of fungus infection in grain. Under the appropriate temperature and humidity, microorganisms multiply in large numbers and then cause grain discoloration, odor, heat and mold. It not only causes serious economic losses, but also threatens the health of people and animals after ingestion. The common methods to kill insects are often chemical related.¹⁹ So, in the grain storage system, particularly where the grain is stored in bulk, the control strategy requires that the grain be routinely treated with a residual insecticide at intake.²⁰ However, non-polluting grain storage methods are best. Due to these challenges, much effort has been devoted to the development of ecologically compatible methods of pest control using natural-occurring compounds or materials. It has been found that the changes in environmental humidity affect the activity behavior of insects and can kill pests under appropriate conditions.²¹ On the other hand, utilizing insecticide still remains as an effective approach. Diatomite is a natural mineral powder material with special pore structure, large number of orderly micropores on the shell and perfect adsorb ability, allowing the diatomite to be an ideal environmental humidity regulator.^{22–27} In addition, diatomite showed higher sorption capacity than that of lots of the sorbents reported in the literature we surveyed.^{28,29} Such as, diatomite earth-based insecticides (DE), Protect-It and SilicoSec, a nano-structured silica product AL06, used as an insecticide for two stored product pests *Callosobruchus maculatus* and *Sitophilus oryzae*.³⁰ Commercially available diatomite earth (DE) formulations could be satisfactorily on wheat stored pests *Sitophilus granarius*, *Rhyzopertha dominica* and *Tribolium confusum*.³¹

In this work, the diatomite was used as an insecticide to remove grain storage pests. The lethal effect on common stored grain pests was observed as well. The insecticidal process conditions were optimized along with exploring the insecticidal mechanism.³² Furthermore, the toxicity analysis of the diatomite was conducted.

2. Materials and methods

2.1 Materials

Two kinds of diatomite were used as insecticide in this work, raw diatomite (RD) and calcined diatomite (CD) were from Baishan City, Jilin China. The diatomite materials were screened by different mesh sieve. The diatomite of 100 mesh, 300 mesh and 500 mesh was collected and reserved.

The common grain pests selected in this work were as follows, saw-toothed grain beetle (SGB), corn weevil (CW), grain beetle (GB) and *Tribolium ferrugineum* (TF) from specialized purchasing agency.

2.2 Characterization of insecticidal effect

In the experiment, SGB, CW, GB, and TF were selected as the test objects. The single factor test conditions were: temperature 30 °C, relative humidity 65%, diatomite dosage 5 g m⁻², 10 g

m⁻², 15 g m⁻², 20 g m⁻², 25 g m⁻², 30 g m⁻², 35 g m⁻². The size of diatomite was 100 mesh. Each repeat is done three times, and a blank test is performed for analysis. Put 20 pests into each Petri dish (90 mm) and record the weight. The weight loss rate was calculated according to the following formula:

$$\text{Weight loss rate} = (W_b - W_a)/W_b \times 100\%$$

W_b : weight before the experiment, g; W_a : weight after the experiment, g.

2.3 Optimization of insecticide process conditions

The factors affecting diatomite insecticidal activity and the optimal factor level were identified through an orthogonal experiment, and the corresponding factor levels were shown in Table 1. All experiments were carried out in triplicate, and average values are reported. Mean separation and significance were analyzed using the IBM SPSS Statistics (Statistical Product and Service Solutions, USA).

2.4 Characterization of the diatomite

Scanning electron microscopy analysis (JEOL, JSM-6460LV), energy spectrum analysis (Oxford Instruments, X-MAX50), BET analysis (McMerritik Instruments, ASAP2050), FT-IR analysis (PE, One-B) and contact angle analysis of diatomite materials were carried out to reveal the chemical composition, morphology and surface properties.

2.5 Diatomite toxicity tests

Chinese Kunming (CK) mice were selected as the experimental animals, and an outbred mouse strain, which has been widely used in toxicology studies, was used in product safety testing and also in reproductive toxicity studies. However, acute toxicity test and long-term toxicity test were carried out separately. All animal procedures adopted in this work received prior approval by the local animal protection committee and were in agreement with the China's law of animal protection.

Mice were randomly divided into groups of five, with a total of four groups, ten males and ten females. Animals were fed with a mixture of diatomite and mice food (diatomite : mice food = 2 : 25) at 23 ± 2 °C with a 16 h : 8 h L : D photoperiod. For acute toxicity test, the diatomite was injected into the CK mice by gavage according to 20% of body weight twice a day for two weeks. For long-term toxicity test, animals were fed at regular intervals every day according to 15% by body weight for

Table 1 Factor and level of orthogonal experiment

Factors	Level		
	1	2	3
A, Diatomite size	100	300	500
B, Temperature (°C)	25	30	35
C, Relative humidity (%)	55	65	75
D, Diatomite dosage (g m ⁻²)	10	20	30

six months. Mortality was recorded daily and the drinking water was renewed every day as prescribed.

3. Results and discussion

3.1 Preliminary study on insecticidal effect of diatomite

The influences of different diatomite on lethal time and weight loss rate of the four selected grain pests were investigated at 30 °C temperature and 65% relative humidity.

It showed that the lethal time of the four pests decreased with the increase of the amount of different diatomite (Fig. 1). The effect of the raw diatomite was more profound than that of the calcined diatomite. When the diatomite dosage was increased from 5 g m⁻² to 10 g m⁻², the lethal time decreased sharply. When the amount of diatomite was higher than 30 g m⁻², the lethal time remained almost unchanged and leveled off. In conclusion, the increase of diatomite dosage can accelerate the lethal rate of the pests to a certain extent. Under the conditions of this experiment, the appropriate dosage is between 10–30 g m⁻².

As can be seen from Fig. 2, the weight loss rates of the four pests changed irregularly. Among them, the weight loss rate of the *saw-toothed grain beetle* was the highest, followed by the *Tribolium ferrugineum*, *corn weevil* and *grain beetle*. Similarly, the effect of the raw diatomite is significantly better than that of the calcined diatomite. Based on the obvious change of lethal time and weight loss rate of *saw-toothed grain beetle*, the insecticidal characteristics of the raw diatomite is more significant, so that the raw diatomite and the *saw-toothed grain beetle* were selected as the insecticide and the pest subject, respectively, in the following work.

Fig. 2 Weight loss rate of different diatomite to different pests @ 30 °C and RH 65%; TF@RD: the effect of raw diatomite on *Tribolium ferrugineum* at 30 °C and RH 65%; CW@RD: the effect of raw diatomite on *corn weevil* at 30 °C and RH 65%; TF@CD: the effect of calcined diatomite on *Tribolium ferrugineum* at 30 °C and RH 65%; CW@CD: the effect of calcined diatomite on *corn weevil* at 30 °C and RH 65%; GB@RD: the effect of raw diatomite on *grain beetle* at 30 °C and RH 65%; SGB@RD: the effect of raw diatomite on *saw-toothed grain beetle* at 30 °C and RH 65%; GB@CD: the effect of calcined diatomite on *grain beetle* at 30 °C and RH 65%; SGB@CD: the effect of calcined diatomite on *saw-toothed grain beetle* at 30 °C and RH 65%.

3.2 Optimization of insecticidal process

According to the information presented in Table 1, orthogonal experiment was conducted to optimize the insecticidal process conditions of the raw diatomite insecticide. Under the same living conditions, pests can survive more than 1 month in the grain without diatomite insecticide.

Table 2 shown that the raw diatomite can accelerate the lethal time of the pest subject and the effect is obvious compared with the pest subjects without using insecticide. Range analysis of the lethal time and the weight loss rate were used to identify the optimal process conditions (Table 3).

For lethal time, the shorter the lethal time, the better the insecticide effect of the diatomite. Range analysis showed that the optimal insecticidal process combination of the raw diatomite insecticide was A₃B₁C₂D₂. The diatomite size was 500 mesh, the temperature was 25 °C, the relative humidity was 65% and the diatomite dosage was 20 g m⁻². For weight loss rate, the greater

Fig. 1 Lethal time of different diatomite to different pests at 30 °C and RH 65%; TF@RD: the effect of raw diatomite on *Tribolium ferrugineum* at 30 °C and RH 65%; CW@RD: the effect of raw diatomite on *corn weevil* at 30 °C and RH 65%; TF@CD: the effect of calcined diatomite on *Tribolium ferrugineum* at 30 °C and RH 65%; CW@CD: the effect of calcined diatomite on *corn weevil* at 30 °C and RH 65%; GB@RD: the effect of raw diatomite on *grain beetle* at 30 °C and RH 65%; SGB@RD: the effect of raw diatomite on *saw-toothed grain beetle* at 30 °C and RH 65%; GB@CD: the effect of calcined diatomite on *grain beetle* at 30 °C and RH 65%; SGB@CD: the effect of calcined diatomite on *saw-toothed grain beetle* at 30 °C and RH 65%.

Table 2 Insecticidal effect of the raw diatomite

Number	Factors				Test results	
	A	B	C	D	Lethal time (h)	Weight loss rate (%)
1	100	25	55	10	28	39.55
2	100	30	65	20	27	62.84
3	100	35	75	30	50	73.42
4	300	25	65	30	31	53.27
5	300	30	75	10	50	60.20
6	300	35	55	20	48	47.37
7	500	25	75	20	23	54.26
8	500	30	55	30	31	30.92
9	500	35	65	10	43	47.18

Table 3 Range analysis of the lethal time and the weight loss rate^a

Range analysis of the lethal time				
	A	B	C	D
ΣK_{1j}	315	247	318	363
ΣK_{2j}	387	321	303	295
ΣK_{3j}	289	423	370	333
$\bar{K}_{1j}$	105	82.33	106	121
$\bar{K}_{2j}$	129	107	101	98.33
$\bar{K}_{3j}$	96.33	141	123.33	111
R	32.67	58.67	22.33	22.67
Range analysis of the weight loss rate				
	A	B	C	D
ΣX_{1j}	527.43	441.24	353.52	440.79
ΣX_{2j}	482.52	461.88	489.87	493.41
ΣX_{3j}	397.08	503.91	563.64	472.83
$\bar{X}_{1j}$	175.81	147.08	117.84	146.93
$\bar{X}_{2j}$	160.84	153.96	163.29	164.47
$\bar{X}_{3j}$	132.36	167.97	187.88	157.61
R	43.45	20.89	69.99	17.54

^a ΣK_{ij} : the sum of the i -level experiment in the j column; $\bar{K}_{ij}$: the average value of the experimental results of the i -level in the j column. ΣX_{ij} : the sum of the i -level experiment in the j column; $\bar{X}_{ij}$: the average value of the experimental results of the i -level in the j column.

the weight loss rate, the better the insecticide effect. The optimal insecticidal process condition of raw diatomite was $A_1B_3C_3D_2$. The diatomite mesh was 100, the temperature was 35 °C, the relative humidity was 75%, and the diatomite dosage was 20 g m⁻². Overall, the dominant influencing factors are different for different indexes. The variance analysis is shown in Table 4.

For the lethal time (Table 4), the four factors were all significant. For the weight loss rate, the relative humidity is the most significant factor, followed by the diatomite mesh and the temperature, and the diatomite dosage is the least significant. The variance analysis results are consistent with those from the range analysis.

In this work, the lethal time is the main index of insecticidal activity of the diatomite insecticide. It suggested that the influence factors order was $C \geq D > A > B$. Based on the range analysis and variance analysis, the optimal insecticidal process conditions of the raw diatomite insecticide were as follows: the diatomite size was 500 mesh, the temperature was 25 °C, the relative humidity was 65% and the diatomite dosage was 20 g m⁻².

3.3 BET and particle size distribution analysis

The pore structure of diatomite determined its adsorption properties. The particle size and specific surface area of diatomite also affected its adsorption performance. BET surface area, pore size and pore volume was presented in Table 5.

It can be seen from Table 5 that the specific surface area, pore volume and pore diameter of the RD were all better than that of the CD. The pore structure of the CD was partially destroyed by high temperature during calcination. Therefore the RD has good adsorption and higher insecticidal rate which consistent with the previous results. In addition, the surface properties of the RD and the CD are different with different particle sizes. However overall, the performance of 100 mesh is better than the other two. Hence, 100 mesh diatomite was selected as the preliminary experiment.

Particle size analysis was carried out to understand the physical characteristics of samples RD and CD, the results are tabulated in Fig. 3.

Fig. 3 showed that the particle size distribution of RD was relatively uniform, which also confirmed the characteristics of uniform particle distribution and high quality of RD. The results were consistent with the surface morphology results mentioned above. But Fig. 4 showed that the particle distribution uniformity of CD was much worse than that of RD, and there were too large and too small particles.

In the insecticidal process of diatomite, the small particles have a high specific surface area, which was conducive to adsorption. However, although the size of CD 500 mesh was small and the specific surface area larger, the adsorption effect was poor because the pores have been melted and collapsed during the calcination process. So it is better to choose a larger size for the CD. Therefore, the size and pore morphology should be comprehensively determined in the selection process of the

Table 5 BET data of RD and CD

Sample	Surface area (m ² g ⁻¹)	Pore volume (cm ³ g ⁻¹)	Average pore diameter (nm)
100 mesh RD	22.3047	0.07449	12.4483
300 mesh RD	23.7537	0.068817	11.5885
500 mesh RD	24.9753	0.079349	12.7085
100 mesh CD	11.7211	0.013042	4.4506
300 mesh CD	9.2493	0.010826	4.6820
500 mesh CD	10.5597	0.014150	5.3598

Table 4 Variance analysis of lethal time and weight loss

Variance analysis of lethal Time						Variance analysis of weight loss				
Factors	S	f	$\bar{S}$	F	Significance	S	f	$\bar{S}$	F	Significance
A	572.74	2	286.37	171.48	**	943.10	2	471.55	26.26	**
B	1735.41	2	867.71	519.59	**	195.40	2	97.70	5.44	*
C	274.74	2	137.37	82.26	**	2494.05	2	1247.03	69.43	**
D	258.07	2	129.04	77.27	**	124.99	2	62.50	3.38	
Error	30.01	18	1.67			323.35	18	17.96

Fig. 3 Particle size distribution of the RD.

Fig. 4 Particle size distribution of the CD.

mesh number of diatomite insecticides. Therefore, the insecticidal effect of CD 100 mesh is better.

3.4 FT-IR spectroscopy

In order to investigate the influence of diatomite surface group structure on the insecticidal effect, infrared analysis of diatomite was carried out, and the results are shown in Fig. 5.

From Fig. 5, it can be seen that there are subtle differences in the infrared images of different diatomite. RD has strong absorption bands near 472 cm^{-1} , 800 cm^{-1} and 1093 cm^{-1} , which belong to the vibration absorption peak of opal. The peak at 472 cm^{-1} was belonged to the Si-O-Si antisymmetric bending vibration in the silicon-oxygen tetrahedron. The peak at 800 cm^{-1} was belonged to the Si-O symmetrical stretching vibration. The broad asymmetric strong absorption band near 1093 cm^{-1} was attributed to the Si-O-Si stretching vibration. There was a weak absorption band near 1637 cm^{-1} , which belonged to the surface hydroxyl vibration absorption peak of free water. A broad absorption band appears near 3406 cm^{-1} was attributed to the vibrational absorption peak of the surface hydroxyl groups bound to water.³³ The groups represented by the absorption bands in the infrared images of CD were consistent with RD. The infrared spectrum shown that there were a large number of hydroxyl groups on diatomite, which indicated that diatomite can be absorbed a large number of water molecules. These water molecules form silanols with

Fig. 5 The FT-IR spectrum of diatomite.

silica in the form of hydrogen bonds, and are connected with silanols by hydrogen bonds, so a large amount of water is stored between the pores of diatomite. The surface hydroxyl vibration intensity of the bound water of CD was lower than that of RD, which was due to the fact that high temperature calcination would reduce the active hydroxyl groups on the surface of opal, resulting in a decrease in the adsorption performance of diatomite.^{34,35} Therefore, when designing diatomite insecticides, diatomite with rich hydroxyl structure should be selected as much as possible.

3.5 Insecticidal mechanism of the diatomite

The insecticidal mechanism of diatomite was studied by analyzing the surface morphology and surface energy spectrum of the dead pest.

Fig. 6 shows that there are many diatomite particles gathered near the pores and hairs of the subjects. Pores are the important part of pests' survival activities, through which water and nutrients are absorbed. Furthermore, pores can also discharge the toxins and waste produced by metabolism from the body and stabilized the body temperature. The diatomite can absorb the *saw-toothed grain beetle's* body fluid upon exposure to the surface and internal of the pores, leading to dehydration, physiological disorder and lethal in the end. Results showed that the disc pore structure on the surface of the raw diatomite was well constructed, compared to that of the calcined diatomite. It is because that the calcination process induces the collapse of pore structure, forming the fragmented structure.^{36,37} Moreover, the adsorption efficiency and the insecticidal rate of the raw diatomite were also higher than those of the calcined diatomite, which was consistent with the orthogonal

Fig. 6 Surface morphology of SGB @RD (a) and SGB@CD (b).

Fig. 7 Element distribution and surface morphology of the raw diatomite (a) and calcined diatomite (b).

results mentioned above.^{38,39} Therefore, the lethal time of the raw diatomite was shorter than that of the calcined diatomite.

The difference of element distribution between the raw diatomite and the calcined diatomite was revealed by energy spectrum analysis (Fig. 7). During the calcination process, the high pyrolysis temperature caused the migration and dissolution of element silicon. The silicon content of the calcined diatomite decreased and the inorganic element content increased due to the carbonization during pyrolysis. The change of elements directly affected the adsorption properties. The reduction of silicon element reduced the formation of silicon-oxygen bond and lowered the adsorption and resolution of water, thus influencing the moisture-regulating performance of diatomite. Therefore, the high adsorption ability of the raw diatomite ensures its excellent humidification and realizes the lethal effect, which is in agreement with the above results.

The absorption of pest body fluid by the diatomite is an adsorption phenomenon as well as a mass transfer process, which is closely related to the migration of water vapor or liquid water.⁴⁰ It has been reported that the diatomite can not only absorb the moisture in the pest body, but also weaken the absorption of moisture in the air by the pest.⁴¹ The properties of diatomite's surface, such as hydrophobia, charge, ion exchange and adsorption capabilities, are highly governed by the presence of water, which is partially structurally connected to the crystal mesh of the diatomite, forming active hydroxyl groups on it.⁴² Which makes its adsorption capacity stronger and accelerates the death of pests. In the process of insecticide, mass transfer process is governed by the partial pressure of water vapor and the transfer process of body fluid absorbed by diatomite, as well as the migration process of free water in the porous structure. Unevenness of the surface of diatomite makes the forces of atoms on the surface of diatomite asymmetrical compared with those in the interior, so that the surface of diatomite has residual surface free energy. And, the gas or liquid molecules collide with the surface of diatomite and adsorbed by unbalanced forces.^{43,44} In the insecticidal process, the hygroscopic absorption of diatomite towards the pest body fluid *via* physical adsorption driven by van der Waals forces and the formation of hydrogen bonding.^{45,46} This unique characteristic enables diatomite to absorb the pest body fluid and dehydrates the pest to lethal.

The results shown in Fig. 8 indicated that the surface of the tested pest is hydrophobic regardless of whether diatomite is used. After treated with different diatomite insecticides, the contact angle decreased to a certain extent, and the contact

Fig. 8 Contact angle of SGB dead body without diatomite (a), @RD (b) and @CD (c).

Fig. 9 Insecticidal mechanism of the diatomite insecticide.

angle of the sample treated by the raw diatomite was slightly lower than that of the one treated by the calcined diatomite. Therefore, the diatomite particles destroy the protective layer and the joints of the pest but cannot destroy them completely. This kind of destruction accelerates the body liquid loss rate of the pest.

Fig. 9 illuminates the process and mechanism of the inactivation of pest caused by the diatomite. When the diatomite is attached to the pests, the adhesion of the diatomite on the pest body can stimulate its sensory system which triggers the movement of the pest more frequently. With the accumulation of the diatomite on the body surface, part of the diatomite will puncture the body surface of the pest, thereby damaging the joints. This promotes the body liquid loss of the pests faster and slows down the activity, dying eventually.

3.6 Diatomite toxicity analysis

Protection of stored-grain against insect pests is essential to secure a continuous and safe food supply. The insecticides should be guaranteed to have no residual risks for consumers and no the concomitant risks for human health. Therefore, the acute toxicity and the long-term toxicity analyses of raw and the calcined diatomite were carried out. The results from acute toxicity analysis indicated that there was no death or other abnormality in the tested mice within two weeks, suggesting that the diatomite had not a noteworthy effect on the physiological activities of mice. Long-term toxicity analysis found that none of the mice died after six months, demonstrating that the diatomite is safe and non-toxic to mammals.

4. Conclusion

The diatomite was used as an insecticide to remove grain storage pest. The insecticidal effect and mechanism were explored *via* well-designed experiments. Single factor experiment showed that the diatomite had excellent lethal effect on four common stored grain pests, *i.e.*, saw-toothed grain beetle, corn weevil, grain beetle and *Tribolium ferrugineum*. The lethal time was shortened with the increase of diatomite insecticide dosages. Among four types of pests, the diatomite exhibited the best activity towards saw-toothed grain beetle in terms of the weight loss rate and sensitivity. Moreover, the insecticidal efficiency of the raw diatomite was obviously higher than that of the calcined diatomite. The optimal conditions of the raw diatomite insecticide were as follows: the diatomite mesh was 500 (A_3), the temperature was 25 °C (B_1), the relative humidity was 65% (C_2), the diatomite dosage was 20 g m⁻² (D_2), the

influence factors order was $C \geq D > A > B$. The different performance between the raw diatomite and the calcined diatomite is attributed to their different structure and silicon content. The raw diatomite has complete porous and uniform disk structure, high silicon and hydroxyl content, large specific surface area, high mesopore content, good water absorption and retention. However, the pore structure of the calcined diatomite is collapsed with the decrease of silicon content the amorphous structure, as well as the disappearing of mesoporous structure and weak adsorption capacity. The special pore structure, water absorption and retention ability of the diatomite caused the body liquid loss and joint damage of the pest which made pest move slowly gradually and eventually dying. Toxicity analysis suggested that the diatomite was non-toxic to mammals. The difference between this paper and the former researchers⁴⁷ were that the strong adsorption of diatomite be used to control the stored grain pests to achieve pure physical insecticide and can be removed from the grain during the milling process, reducing secondary pollution. However, currently commercially available diatomite insecticides all contain a certain amount of plant-derived insecticides or chemical insecticides. Insecticides were carried out by chemical methods, and the loaded pesticides had certain residual side effects. Therefore, the diatomite is a kind of promising natural insecticide by regulating body fluids and physiological system, which is low toxicity, worker safety, low risk of food residues, environmentally friendly and perfect for green grain storage.

Ethical statement

All animal procedures were performed in accordance with the Guidelines for Care and Use of Laboratory Animals of "Hunan Animal Experiment Center" University and experiments were approved by the Animal Ethics Committee of "Beijing Forestry University".

Author contributions

Chunyan Li: data curation, writing – original draft preparation. Xueru Sheng, Na Li, Qingwei Ping and Peng Lu: supervision. Jian Zhang: funding acquisition, writing – reviewing and editing.

Conflicts of interest

The authors declare no competing financial interest.

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