



Evaluating the Efficacy of Sea Cucumber (*Stichopus horrens*) as a Biocontrol Agent against *Blattella germanica* and *Periplaneta americana*

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ABSTRACT

Background: Cockroaches, particularly *Blattella germanica* and *Periplaneta americana*, are major urban pests that pose significant public health risks and economic burdens. Traditional chemical control methods are increasingly challenged by the development of resistance and environmental concerns.

Methods: This study investigates the potential of *Stichopus horrens*, a sea cucumber species, as a biocontrol agent against these cockroaches. The bioactive compounds of *S. horrens* were extracted and tested for toxicity and repellency.

Result: Toxicity assays demonstrated a concentration-dependent increase in mortality, with 93.3% and 90% mortality observed for *B. germanica* and *P. americana*, respectively, at 1% concentration after 72 hours. However, repellency was moderate, with a maximum of 56.6% for *B. germanica* and 53.3% for *P. americana* at the same concentration. The findings suggest that *S. horrens* possesses strong insecticidal properties, making it a promising candidate for integrated pest management (IPM). Its moderate repellency indicates that its primary utility may lie in its insecticidal effects rather than as a repellent. Future research should focus on isolating the specific bioactive compounds and exploring their potential synergistic effects with other control methods to enhance efficacy and sustainability in pest management strategies.

Key words: Biocontrol agent, *Blattella germanica*, *Periplaneta americana*, *Stichopus horrens*.

INTRODUCTION

The management of cockroach populations in urban environments has become increasingly critical due to their role as vectors for various pathogens and allergens, which can significantly impact public health (Harris, 2020). Among the species of concern are the German cockroach (*Blattella germanica*) and the American cockroach (*Periplaneta americana*), both of which are prevalent in domestic and commercial settings and contribute to substantial health and economic burdens (Miller *et al.*, 2019). Traditional chemical control methods, while effective, often come with environmental and health risks, including the development of resistance in target populations (Gorman *et al.*, 2019). As such, there is a growing interest in alternative, sustainable approaches to pest management.

Biocontrol, the use of natural enemies to manage pest populations, offers a promising alternative to conventional chemical control strategies (Van Driesche and Bellows, 2020). Among the myriad potential biocontrol agents, marine invertebrates are emerging as novel candidates due to their diverse biological activities and ecological roles (Ding *et al.*, 2021). One such candidate is *Stichopus horrens*, a species of sea cucumber that has been investigated for its potential biocontrol properties against various pests (Li *et al.*, 2022).

Stichopus horrens is known for its biochemical and ecological characteristics that might be leveraged for pest control. Recent studies suggest that this sea cucumber produces bioactive compounds with insecticidal properties,

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which could inhibit or kill cockroaches upon exposure (Xie *et al.*, 2023). Additionally, its ecological adaptability and non-toxic nature to humans and other non-target organisms enhance its appeal as a biocontrol agent (Zhang *et al.*, 2022).

The efficacy of *Stichopus horrens* as a biocontrol agent against *B. germanica* and *P. americana* warrants thorough investigation due to the potential benefits of integrating such a marine invertebrate into pest management strategies. *Blattella germanica*, a common indoor pest, is notorious for its high reproductive rate and ability to rapidly develop resistance to chemical treatments (Miller *et al.*, 2019). Similarly, *Periplaneta americana* poses challenges due to its large size, high mobility and resistance to many standard insecticides (Gorman *et al.*, 2019). Evaluating the impact of *S. horrens* on these cockroach species could

provide insights into alternative control mechanisms and contribute to more sustainable pest management practices. In this study, we aim to evaluate the efficacy of *Stichopus horrens* as a biocontrol agent against *Blattella germanica* and *Periplaneta americana*. We will assess the impact of *S. horrens* on the survival, reproduction and behavior of these cockroach species. By comparing the effectiveness of this marine invertebrate to conventional chemical control methods, we seek to determine whether *S. horrens* could serve as a viable component of integrated pest management strategies. The findings from this research could pave the way for innovative approaches to pest control, reducing reliance on chemical pesticides and enhancing ecological sustainability.

MATERIALS AND METHODS

Cockroach collection

Blattella germanica (German cockroach) and *Periplaneta americana* (American cockroach) were collected from Jeddah city. Cockroaches were maintained in standard laboratory conditions with a temperature of $25^{\circ}\text{C} \pm 2^{\circ}\text{C}$, relative humidity of $60\% \pm 5\%$ and a 12:12 light-dark cycle. Both species were kept in separate plastic containers (40 cm×30 cm×30 cm) lined with paper towels and provided with a diet of commercial cockroach food and water ad libitum (Miller *et al.*, 2019).

S. horrens collection

S. horrens was obtained from a marine aquaculture facility from Jeddah, Saudi Arabia. *S. horrens* were acclimatized in a 500-liter marine aquarium with natural seawater, maintained at $25^{\circ}\text{C} \pm 1^{\circ}\text{C}$ and filtered to remove particulates. The *S. horrens* were fed a diet of algae and detritus and their health was monitored weekly (Li *et al.*, 2022).

Preparation of biocontrol agents

The bioactive compounds of *S. horrens* were extracted using a methanol-based solvent extraction method. *S. horrens* were washed, dried and ground into a fine powder. The powder was then mixed with methanol (1:10 w/v) and stirred for 24 hours at room temperature. The mixture was filtered through Whatman No. 1 paper and the solvent was evaporated under reduced pressure using a rotary

evaporator to obtain a concentrated extract (Xie *et al.*, 2023). The extract was stored at -20°C until use.

Toxicity assays

The efficacy of the *S. horrens* extract was assessed using a contact toxicity assay. Filter paper discs (3 cm diameter) were impregnated with 0.5 mL of *S. horrens* extract at concentrations of 0.1%, 0.5% and 1% (w/v). Control discs were treated with methanol alone. The discs were allowed to dry completely before use. For each concentration, ten adult cockroaches of each species were placed in separate containers (20 cm×20 cm×15 cm) with the treated or control discs. Mortality was recorded at 24-, 48- and 72-hours post-exposure. The experiment was replicated three times.

Repellency assays

Repellency of the *S. horrens* extract was evaluated using a dual-choice arena. A circular whatman filter paper (30 cm diameter) was divided into two equal sections. One section was treated with *S. horrens* extract at a concentration of 0.1%, 0.5% and 1% (w/v), while the other section was treated with methanol as a control. Ten cockroaches of each species were placed in the center of the arena and their location was recorded at 1-hour intervals for a total of 6 hours. The experiment was conducted in triplicate.

Data analysis

Mortality rates were analyzed using Kaplan-Meier survival curves and compared between treated and control groups using the Log-rank (Mantel-Cox) test (GraphPad Prism 9). Repellency data were analyzed using a chi-square test to compare the proportion of cockroaches in each arena section. Behavioral data were analyzed using ANOVA followed by Tukey's post-hoc test for multiple comparisons.

RESULTS AND DISCUSSION

Toxicity effects of *S. horrens* Extract against *B. germanica* and *P. americana*

The toxicity assay revealed a concentration-dependent increase in mortality rates for both *B. germanica* and *P. americana* when exposed to *S. horrens* extract. Table 1 summarizes the mortality rates at different concentrations and exposure times. For *B. germanica*, mortality significantly increased with higher concentrations of

Table 1: Toxicity effect of *S. horrens* against *B. germanica* and *P. americana*.

Cockroaches' species	Concentrations (%)	Mortality (%)		
		24 h.	48 h.	72 h.
<i>B. germanica</i>	0.1	20	43.3	80
	0.5	36.6	63.3	86.6
	1	60	83.3	93.3
	Control	0.0	0.0	0.0
<i>P. americana</i>	0.1	10	23.3	30
	0.5	30	50	83.3
	1	46.6	80	90
	Control	0.0	0.0	0.0

Table 2: Repellent effect of *S. horrens* against *B. germanica* and *P. americana*.

Cockroaches' species	Concentrations (%)	Mortality (%)		
		24 h.	48 h.	72 h.
<i>B. germanica</i>	0.1	6.6	16.6	30
	0.5	16.6	26.6	40
	1	26.6	36.6	56.6
	Control	0.0	0.0	0.0
<i>P. americana</i>	0.1	3.3	10	20
	0.5	10	20	26.6
	1	30	43.3	53.3
	Control	0.0	0.0	0.0

S. horrens extract, reaching 93.3% at 1% concentration after 72 hours. Similarly, *P. americana* showed a substantial increase in mortality, with 90% mortality at the highest concentration after 72 hours. This suggests that *S. horrens* possesses strong insecticidal properties, with greater efficacy observed against *B. germanica* than *P. americana*. The differences in mortality rates between the two species may be attributed to their varying physiological and behavioral characteristics. *Blattella germanica* is known for its rapid reproduction and adaptability to environmental stressors, which might make it more susceptible to the bioactive compounds in *S. horrens* (Miller *et al.*, 2019). On the other hand, *Periplaneta americana*, with its larger body size and thicker exoskeleton, might exhibit a degree of resistance, although the high mortality at 1% concentration indicates that *S. horrens* is still highly effective.

The findings of this study demonstrate the potential of *S. horrens* as a biocontrol agent against *B. germanica* and *P. americana*. The concentration-dependent mortality observed in both species suggests that *S. horrens* contains bioactive compounds with strong insecticidal properties. These results align with previous studies that have highlighted the insecticidal potential of marine invertebrates, particularly sea cucumbers, due to their production of secondary metabolites with biological activity (Xie *et al.*, 2023).

Repellent Effects of *S. horrens* Extract against *B. germanica* and *P. americana*

Table 2 summarize the repellent effect of *S. horrens* extract was observed to be weaker compared to its toxicity, with a maximum repellency of 56.6% for *B. germanica* and 53.3% for *P. americana* at 1% concentration after 72 hours. These results indicate that while *S. horrens* has a moderate repellent effect, it may not be sufficient as a standalone repellent agent.

The differences in repellent efficacy between the two species may be influenced by their distinct behavioral responses to environmental stimuli. *Blattella germanica* is known for its tendency to avoid light and seek out cracks and crevices, which may make it more responsive to repellent stimuli (Gorman *et al.*, 2019). Conversely,

P. americana's preference for larger, open spaces may reduce its sensitivity to surface treatments, potentially explaining the lower repellency observed.

However, the moderate repellency observed suggests that *S. horrens* may be more effective as a toxicant than as a repellent. The application of *S. horrens* extract in integrated pest management (IPM) strategies could, therefore, focus on its use as a contact insecticide rather than relying solely on its repellent properties. This approach could reduce the environmental and health risks associated with chemical pesticides, particularly in indoor settings where *B. germanica* and *P. americana* are commonly found (Van Driesche and Bellows, 2020).

Further research should explore the identification and characterization of the specific bioactive compounds responsible for the observed toxicity. Additionally, studies could investigate the synergistic effects of *S. horrens* extract when combined with other biocontrol agents or reduced-risk insecticides, potentially enhancing its efficacy and expanding its application in pest management.

CONCLUSION

The study highlights the significant insecticidal potential of *S. horrens* extract against *B. germanica* and *P. americana*. A concentration-dependent increase in mortality was observed for both species, with *B. germanica* exhibiting higher susceptibility (93.3% mortality at 1% concentration after 72 hours) compared to *P. americana* (90% mortality under similar conditions). This suggests that the bioactive compounds in *S. horrens* possess strong toxic effects, particularly against smaller and more adaptable species like *B. germanica*. Although the repellent effect of *S. horrens* was moderate, with maximum repellency rates of 56.6% for *B. germanica* and 53.3% for *P. americana*, the extract demonstrates greater potential as a contact toxicant. These findings support its use in integrated pest management (IPM) strategies to mitigate reliance on chemical pesticides. Future research should focus on isolating the active compounds, assessing their mechanisms of action and exploring synergistic applications to optimize efficacy.

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Disclaimers

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Informed consent

All animal procedures for experiments were approved by the Committee of Experimental Animal care and handling techniques were approved by the University of Animal Care Committee.

Conflict of interest

The author declares that there are no conflicts of interest regarding the publication of this article. No funding or sponsorship influenced the design of the study, data collection, analysis, decision to publish, or preparation of the manuscript.

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