Antimicrobial efficacy of certain insect toxins in inhibiting *E. coli* and *Staphylococcus aureus* compared to synthetic antibiotics

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Abstract

The present investigation was carried out at the College of Science at Tikrit University. Its goal was to investigate how well certain insect poisons affected the human-pathogenic microorganisms *E. coli* and *Staphylococcus aureus*. Using an antibiotic susceptibility test, this inhibition was contrasted with the capacity of a number of antibiotics, such as Amoxillin, Gentamicin, and Pencillin, to stop the growth of harmful bacteria. The results demonstrated significant inhibitory effectiveness against the two bacterial species studied. The findings demonstrated that the venom of the red wasp (Vespa orientalis) was the most effective against E. coli, with an average inhibitory diameter of 22 mm. The results also showed that the venom of the yellow jacket wasp was the least effective against the same bacteria, with an average inhibition diameter of 10 mm.

The results showed that black ant venom was the most effective on *S.aureus* bacteria with an average inhibition diameter of 13.5 mm, while the effect of yellow jacket venom was the least effective on the same bacteria with an average inhibition diameter of 10 mm. In comparison with the effectiveness of antibiotics against the bacteria under study, it was noted that there was an inhibitory effect of Gentamicin on *E.coli* and *S.aureus* bacteria, while the inhibitory effect of Pencillin was on *E.coli* bacteria. As for Amoxillin, it showed an inhibitory effect on *Staphylococcus aureus* bacteria only, and *E.coli* bacteria were not affected by it.

Key words: Antimicrobial activity, insect toxins, E. coli, *Staphylococcus aureus*, antibiotics.

الفاعلية ضد مايكروبية لسموم بعض الانواع الحشرية في تثبيط E. Coli. و *Staphylococcus aureus*
مقارنة بالمضادات الحيويه الصناعية

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أُجريت الدراسة الحالية في كلية العلوم بجامعة تكريت، وهدفت إلى تقصّي كفاءة بعض سموم الحشرات في التأثير على الأحياء المجهرية الممرِضة للإنسان، وهما بكتيريا Escherichia coliو *Staphylococcus aureus*. وتمت مقارنة هذا التأثير التثبيطي مع قدرة عدد من المضادات الحيوية، مثل الأموكسيسيلين (Amoxicillin)والجنتاميسين (Gentamicin) والبنسلين(Penicillin) ، على تثبيط نمو هذه البكتيريا المرضية، وذلك باستخدام اختبار الحساسية للمضادات الحيوية .أظهرت النتائج وجود فعالية تثبيطية معنوية ضد كلا النوعين من البكتيريا المدروسة. حيث بيّنت النتائج أن سم دبور الزنبور الأحمر (Vespa orientalis) كان الأكثر فاعلية ضد بكتيريا E. coli، إذ بلغ متوسط قطر منطقة التثبيط 22 ملم. كما أوضحت النتائج أن سم دبور السترة الصفراء (Yellow jacket wasp) كان الأقل فاعلية ضد البكتيريا نفسها، بمتوسط قطر تثبيط بلغ 10 ملم.

كما أظهرت النتائج أن سم النملة السوداء كان الأكثر تأثيرًا على بكتيريا *Staphylococcus aureus*، بمتوسط قطر تثبيط بلغ 13.5 ملم، في حين كان سم دبور السترة الصفراء الأقل فاعلية ضد البكتيريا نفسها، بمتوسط قطر تثبيط قدره 10 ملم .وعند مقارنة فعالية المضادات الحيوية ضد البكتيريا قيد الدراسة، لوحظ أن للجنتاميسين تأثيرًا تثبيطيًا على كل من *E. coli* و *Staphylococcus aureus*، في حين اقتصر التأثير التثبيطي للبنسلين على بكتيريا *E. coli* فقط. أما الأموكسيسيلين، فقد أظهر تأثيرًا تثبيطيًا على بكتيريا *Staphylococcus aureus* دون أن يكون له تأثير على بكتيريا E. coli.

الكلمات المفتاحية: الفعالية المضادة للبكتريا ، سموم الحشرات، الإشريكية القولونية، المكورات العنقودية الذهبية، المضادات الحيوية.

Introduction

About 75% of animal toxins are confined to insects, where three animal groups: insects of the order Hymenoptera, snakes, and spiders are often responsible for deaths attributed to venomous animals [1]. The Hymenoptera order is the third largest insect order and perhaps the most beneficial to humans, comprising approximately 115,000 distributed species, including wasps, bees, ants, etc. This order is important to human life as pollinators of cultivated and wild flowering plants, parasitizing destructive insects, and useful in the production of honey and wax [2].

Wasps belong to the suborder Apocrita, which is divided into two groups, Parasitica and Aculeata. Approximately 95% of the 15,000 species of wasps in the Aculeata group are solitary [3]. The solitary (asocial) lifestyle of wasps means they do not form colonies. Wasps of the family Vespidae are distributed worldwide and contain over 5,000 species. Isolated components of wasp venom have been shown to have numerous benefits, including antibacterial, anticancer, and anti-inflammatory properties [4]. Wasp venom is a mixture of biologically active components, which include enzymes such as phospholipase A (PLA) and hyaluronidase, amines such as histamine, serotonin, and chatholamine, peptides such as mastoparans and wasp kinin, and other components such as acetylcholine (an antigen) [5].

Bees belong to the species Apis mellifera, which participates in many activities closely related to humans, including pollination and the production of some components of honey such as ricin, wax, royal jelly, propolis, polyene, and apitoxin (bee venom) [6]. Among the most important substances produced by honeybees is apitoxin. Glands in the insect's abdomen produce this chemical compound. Apitoxin consists of 88% water, while the remaining 12% contains compounds such as hyaluronidase, phospholipase A2, histamine, melittin, and some peptides such as apamin and secapin [7]. Honey has been known for its diverse medicinal uses since ancient civilizations [8]. One of the well-established properties of honeybee venom and its main components is its microbiological activity against bacteria and fungi [9], viruses [10], and parasites [11]. Honeybee venom is a clear, colorless liquid with a pleasant smell and a pungent flavor. It has a basic mass of 1.13 and contains a group of proteins, carbohydrates, amino acids, and volatile oils [12].

Bee venom dries quickly, even at room temperature, and tends to be acidic because its pH is between 4.5 and 5.5. Bees use it for self-defense. When it comes into contact with the eyes or mucous membranes, bee venom causes severe burning and irritation [13].

Ant venom is composed of a complex mixture of chemicals such as proteins, enzymes, peptides, biogenic amines, hydrocarbons, formic acid, and alkaloids [14]. All of these components are produced by the venom gland, which consists of two elongated and twisted cylindrical tubules connected to a venom reservoir. The venom is secreted by the tubular glands, which are stored in the reservoir and connected to the delivery system, where up to 130 mg of venom can be delivered after each bite [15]. Ants use venom for several purposes, including colony defense against predators/competitors and pathogens, as well as for social communication . Ant venom contains agents that are lethal and paralytic to a wide variety of arthropods [16]. Many ants are generalist predators, feeding on a wide variety of invertebrates. Some ant species are specialized, feeding only on certain prey species [18]. The current study aims to detect the effectiveness of venoms and secretions from honeybees, ants, the oriental wasp, and the yellow leaf wasp in inhibiting the growth of *E. coli* and *Staphylococcus aureus* bacteria. Then compare the effectiveness of insect venoms with antibiotics

Materials and Methods

A- Collecting insect samples under study: Insect samples (red wasps, oriental wasps, and honeybees) were collected using a locally made netting method consisting of a wire clip fixed to cubic metal poles with an opening on one side and wooden nets for baiting. Wasps entered through this opening to feed on the bait. Meanwhile, black ants were collected by hand and placed in a killing bottle. They were identified in the Entomology and Parasitology Laboratories, Department of Life Sciences, College of Science, Tikrit University, and preserved for use by removing the stinger and gland, making concentrations of its toxins, and then treating the bacteria.

Table (1) Collection of insect samples

Location of Catch/Collecton	Specimen type
Tikrit/Al-Qadisiyah/Al-Balaj Neighborhood	Red wasp
Tikrit/Al-Alam District	Yellow wasp
Baghdad/Abu Ghraib	Honybees
Tikrit/Al-Diyum Neighborhood	Black Ants

B- Collection and diagnosis of bacterial strains:

E.coli and *Staphylococcus aureus* samples were collected from Salah Al-Din General Hospital from different types of samples (urine, wounds and burns, discharge), for both sexes and of different ages. They were transferred in special preservation conditions to Tikrit University / Microbiology Laboratory in the College of Science, and were permanently preserved in the laboratory in a special refrigerator to preserve bacterial isolates until they are used.

C - Preparation of insect poison concentrations:

A 0.05% concentration of insect poisons under study was prepared after removing the stinger of each group of insects, one by one, and getting rid of the cuticle and stinger, by taking 0.05 grams of insect poison and dissolving it in 1 ml of distilled water and mixing it well in a vortex device.

Result and Discussion

The findings demonstrated that amoxillin and pencilin had different inhibitory effects on the pathogenic bacteria utilized in the investigation. Using the same antibiotic, pencillin clearly inhibited *E. coli* bacteria with an inhibition rate of 10 mm, but it had no effect on Staph. aureus bacteria. With an inhibition rate of 14 mm, the results also demonstrated that amoxillin clearly inhibited Staph. aureus bacteria but had no effect on *E. coli* bacteria. As displayed in (Table 2).

Table (2) The inhibitory effect of antibiotics on *Staphylococcus aureus* and *E. coli* bacteria

Median Inhibition Diameter		Antibiotic
E.coli	*Staphylococcus aureus*	
10	——	Pencillin
18	22	Gentamycin
——	14	Amoxillin

According to the data, a solution of red wasp venom had the strongest inhibitory impact on *Staphylococcus aureus* bacteria, with an inhibition rate of 25 mm. In contrast, a solution of yellow wasp venom had the least effect, with an inhibition rate of 10 mm. While the honeybee venom solution had no inhibitory effect on the same bacteria, black ant venom demonstrated inhibition at a rate of 13.5 mm.

All of the insect toxins utilized in the study demonstrated an inhibitory impact on bacteria with varying inhibitory diameters when compared to *E. coli* bacteria. With an inhibition rate of 22 mm, the red wasp venom had the greatest effect on the same bacteria, whereas the yellow wasp venom had the lowest inhibitory value, averaging 10 mm. The inhibition rates of the honeybee and black ant venoms were 15 mm and 16.5 mm, respectively. As shown in Table (3) and Figure (1 and 2)

Table (3): Average diameters of inhibition for the growth of the two types of bacteria, *Staphylococcus aureus* / *E. coli*, treated with the insecticides under study.

Inhibition diameter by bacteria type mm				
Black Ants	**Red Wasp**	**Yellow Wasp**	**HoneyBees**	**Type of Bacteria**
16.5	22	10	15	*E.coli*
13.5	25	10	-	*Staphylococus aureus*

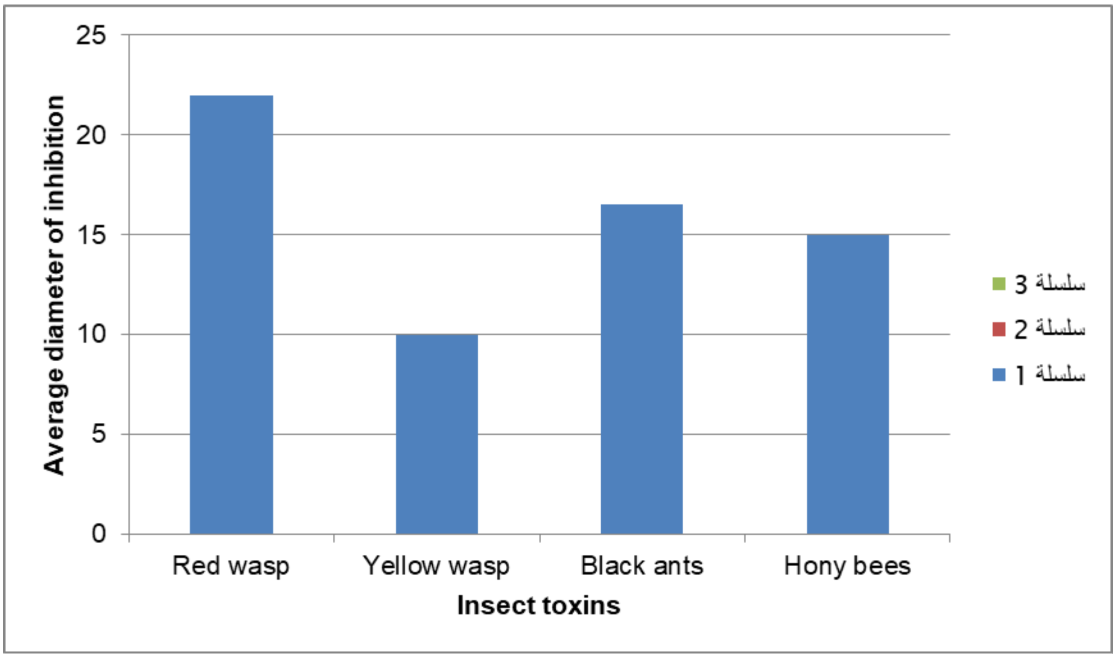

Figure (1) Average diameter of inhibition of growth of *E.coli* bacteria treated with the insect toxins under study

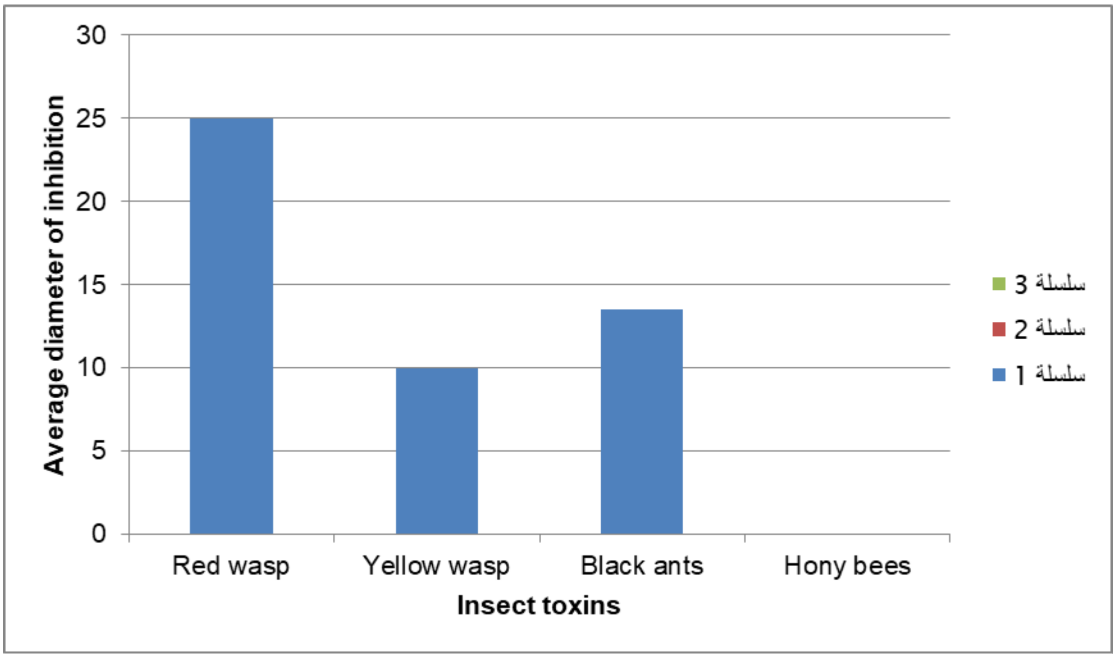

Figure (2) Average diameter of inhibition of growth of Staphylcoccus aureus bacteria treated with the insect toxins under study

The effectiveness of insecticide solutions in inhibiting two types of bacteria, 0.1% of the bacterial suspension was put on the medium, and then the holes were drilled with a cork drill. After that, a 0.05 mM insecticide solution was added to the perforations. After that, the plates were incubated for 18 to 24 hours at 37°C. Using a measuring ruler, the diameter of the inhibition circle was measured to determine how well the insecticide solutions worked against the bacterial isolates [18].

Conclusions

The present study concluded the most efficient wasp venom against *E. coli* was red, whereas the least effective was yellow. In addition The most efficient venom against *Staphylococcus aureus* was that of black ants, whereas the least effective was that of yellow wasps. When it came to blocking both kinds of bacteria, Gentamicin outperformed Amoxillin and Pencillin.

Recommendation

1- Chemical analysis of the insect poisons being studied and evaluation of each compound's capacity to inhibit these human and other harmful bacteria and fungi.

2. Researching how these toxins work to render these infections inactive

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