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STUDY THE EFFECT OF BEE VENOM (ABEVAC) ONPATHOGENIC GRAM-POSITIVE BACTERIA

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ABSTRACT

Bee venom has been used in treatment of many inflammatory diseases, some autoimmune diseases and bovine mastitis. There are some commercially available drugs prepared from bee venom that are used for medical purposes. Abevac is one of these bee venom drugs. The present study show the effect of Abevac against some reference strains of pathogenic Gram-positive bacteria (*S.aureus*, MRSA, *B.cereus* and *S.pyogenes*) usingdisc diffusion method and micro dilution assay against standard antibiotic (penicillin).Abevac is more potent than penicillin on Gram-positive bacteria.

Key words:

Abevac, Bee venom, *S. aureus*, MRSA, *B. cereus, S .pyogenes.*

INTRODUCTION

The frequencies of development and spread of antibiotic resistance among pathogenic bacteria are increasing worldwide and constitute a serious public health concern. This necessitates the continuous search for new antibiotics or discovery of new antibacterial agents like antimicrobial peptides of natural products **(Levy and Marshall, 2004)**. The majority of bacterial species such as *Pseudomonas, Klebsiella, Enterobacter, Acinobacter, Salmonella, Staphylococcus,* Methicillin resistant *Staphylococcus aureus* (MRSA), *Enterococcus*spp, Penicillin Resistant *Streptococcus pneumoniae* (PRSP) and VancomycinResistant enterococci have developed several ways to resist antibiotics. Such bacteria are becoming a serious clinical problem throughout the world **(Ang *et al*., 2004).** Bees have been appreciated for their medicinal purposes and Apitherapy can be traced back to ancient Egypt and has been practiced in China for 3000-5000 years **(Urtubey, 2005)**.The ancient Greek doctor Hippocrates used bee stings and bee venom in his medical practices **(Kim *et al.,* 1992**) and Doctor Charlemagne received bee stings for therapy against Gout **(Clark *et al.,* 1999)**. The modern use of bee venom in apitherapy was initiated by Philip Terk who published its

results on rheumatism **(Terk, 1888)**. The use of bee venom therapy in the USA was initiated by **Mraz, (1995)**.Bee venom therapy, which is a part of Apitherapy that utilize bee hive products, has a long history in folk medicine in the treatment of human health conditions. It is a medical treatment that injects the purified venom extracted from the natural honey bees in the affected area or acupuncture points **(Sturm *et al.,* 2002 and Choi *et al.,* 2003)**. It has been recorded by **Simics (2001)** that bee venom therapy is most affective when it comes directly from the live bee during the late spring to early fall season when bees have a good pollen source to produce potent venom. Bee venom, also called Apitoxin, is an odorless liquid with pH 4.5-5.5 and has a specific gravity of 1,131 **(Schmidt *et al.,* 1999**). It is synthesized in venom glands of worker bee and queen and stored in venom sac **(Kerr *et al.,* 1962 and Benton *et al.,* 1963).** It is composed of 88% water and 12% pharmacologically active components including peptides, amines and various enzymes and low components like carbohydrates and lipids **(De Abreu *et al.,* 2010 and Hundstad and Gjersoe, 2010)**. Hyaluronidase is among the bee venom enzymes that are responsible for breakdown of hyaluronic acid. Also phospholipase A, which destroys the phospholipids causing the death of the cell and is considered the most harmful allergic component of bee venom **(Habermann, 1972; Urtubey, 2005 and Jeong *et al.,* 2011**). Bee venom peptides include melittin, apamin, adolpain and mast cell degranulating peptide (MCD). Melittin is the main biological active component accounting 50-55% of dry bee venom. It has a powerful antibacterial action against many organisms and is more active against Gram-positive bacteria than Gram-negative bacteria **(Fennel*et al.,*1967 and Piek,1986)**.Melittin and apamin stimulate the body adrenal and pituitary system to produce cortisol **(Vick *et al.,* 1972 and Rudenko and Nipot, 1996)**.The aim of the present study was to elucidate the antimicrobial potentialsofAbevac drug against some reference strains of pathogenic Gram-positive bacteria.

MATERIAL AND METHODS

Abevac is a purified bee venom drug produced by Vacsera (the Egyptian holding company for vaccine and biological preparations production). Each 1 ml Abevac contains: -

Bee venom : 1 mg of purified *Apismellifera* venom

Sod.chloride : 9 mg

Deionized Water for injection (WFI): 1 ml

Abevacwas studied againstsome reference strains of pathogenic Gram-positive bacteriakindly obtained from the Department of microbiology, Faculty of veterinary medicine, Cairo

University. It includes (*S.aureus*, MRSA, *B. cereus* and *S.pyogenes*) usingdisc diffusion method **(Lin *et al.,* 2003)** and micro dilution assay **(Wu and Hancock, 1999)** against standard antibiotic (penicillin).

RESULTS

Preliminary screening of antibacterial activities of Abevac for each standard bacterial strain using:

Disc diffusion method:

Abevac treatment showed wide inhibition zone diameters against the seven bacterial mastitis pathogens screened as summarized in (Table 1). The highest inhibitory zones of 21.4 mm were observed from *S. aureus*, followed by MRSA at 18.2 mm against the standard of 10.5 and 7.4 respectively.The Abevac exhibited the maximum inhibitory zones with potency almost up to that of standard drugs against *S. aureus*, MRSA, *B. cereus, S .pyogenes.*

Table(1): Preliminary screening of antibacterial activities of Abevac against standard bacterial strain.

Bacterial species	Inhibition zone in diameter (mm) *	Antibiotic standard †
S.aureus	**21.4 ± 0.71**	**10.5 ± 0.91**
MRSA	**18.2 ± 1.23**	**7.4 ± 1.13**
B. cereus	**15.0 ± 0.7**	**9.6 ± 0.43**
S.pyogenes	**16.1 ± 1.63**	**14.8 ± 0.88**

The values are presented as mean±SD (n=4) and represent Abevac inhibition zone in mm.
†Penicillin was used as standard (10 unit/disc).

Minimum inhibitory concentrations (MIC):

The highest inhibition expressed at MIC values of 15.5µg/ml was recorded against *S. aureus* (Table 2). The MIC values against MRSA, *B. cereus, S.pyogenes*, were 16.7, 17.1 and 16.1µg/ml respectively.

Table (2): Minimum inhibitory concentrations (MIC) of Abevac against standard bacterial strain.

Bacterial species	MIC values (µg/ml)
S.aureus	**15.1 ± 0.33**
MRSA	**16.7 ± 0.44**
B. cereus	**17.1 ± 0.25**
S.pyogenes	**16.1 ± 1.63**

DISCUSSION

Honey bee venom contains at least 18 active substances. Mellitin, the most prevalent, is one of the mostpotent anti-inflammatory agents known(100 times more potent than hydrocortisol). Adolapin is another strong anti-inflammatory substance, and inhibits cyclooxidase. Apamin inhibits complement C3 activity, and blocks calcium-dependent potassium channels, thus enhancing nerve transmission. Other substances, such as Compound X, hyaluronidase, phospholipase A2, histamine and mast cell degeneration protein, are also involved in the inflammatory response of the venom. The antibacterial mechanism of bee venom was the action potential of melittin, which had a high affinity with the cell membrane lipid, through pores on the cell membrane and it has strong antibacterial and antifungal actions **(Matsuzaki, 1997**). Abevac is one of these bee venom drugs that is produced by Vacsera Company for vaccine and biological preparations production and is registered in Ministry of Health in Egypt. Abevac is formulated from honey bee venomand each 1 ml of Abevac contains 1mg of purified *Apismellifera* venom and 9 mg sod. Chloride and 1ml water for injection. Abevac has been used as a human therapy for rheumatism and some autoimmune disorders. The study reveals that Abevac is more potent on Gram-positive bacteria (*S.aureus*, MRSA, *B. cereus and S.pyogenes*) with some variations. **Attalla *et al.*, (2007**) revealed that three bee products; bee venom, proplis and royal jelly had antibacterial activity against certain microorganisms. ThreeGram-positivebacteria;*S.aureus,B.subtilis*and *L.monocytogenes* and two Gram-negative bacteria; *E. coli* and *S. enteritidis*were compared for sensitivity to these products by determining the MLCs. The results indicated that bee venom was the most effective followed by propolis then royal jelly. Gram-positive bacteria were more sensitive to these products than Gram–negative ones. In the present research, the highest inhibitory zones of 21.4 mm were observed from *S. aureus* (Table 1) and the highest inhibition expressed at MIC values of 15.5µg/ml was recorded against *S. aureus* (Table 2).

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