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Non-Disulfide-Bridges Peptides from Scorpion Venoms: Targets for Novel Drugs Against Antibiotic-Resistant Pathogens

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ABSTRACT:

Overuse of antibiotics in recent decades has led to the emergence of antibiotic-resistant infections. Drug resistance is now regarded as a major threat to global public health. For this reason, the phenomenon of antibiotic-resistance caused that health care professionals looking for new types of effective antimicrobial agents against the threat of traditional antibiotic-resistant pathogens. In their role as effector molecules in the innate immune response, antimicrobial peptides (AMPs) carry out numerous important tasks. Through interactions with negatively charged phospholipids, cationic AMPs have the ability to damage microbial cell membranes. Positively charged cationic AMPs reduce the risk of resistance, which is a major advantage compared to available antibiotics. In recent decades, a large number of non-disulfide-Bridges peptides (NDBPs) with medicinal properties have been isolated from the venom of the scorpion. Therefore, scorpion venom is a tremendous source of raw AMPs for design and development of novel drugs against antibiotic-resistant pathogens. NDBPs are effective against different types of antibiotic-resistance bacterial strains, including methicillin-resistant *staphylococcus aureus* (MRSA). In this review, a diversity of NDBPs that are effective against the resistant pathogens was discussed.

Keywords: Non-disulfide-bridges peptides, Scorpion venom, Antimicrobial activity, Novel drugs, Antibiotic-resistant pathogens.

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1. Introduction

Antimicrobial peptides (AMPs) have been identified in a wide range of organisms, including fish, birds, plants, amphibians, insects, and other invertebrates. AMPs have also been found in unicellular microorganisms, particularly in humans, who have a more developed immune system than other animals. [1-7]. AMPs are part of innate immune system, which act as effectors of the innate immune response and have several major activities [1, 5, 7]. These peptides have lack of dependence to cofactors and are not recognized by inhibitors. Therefore, they act very specific against targets. These characterizations make minimize the adverse effects of drugs, which be formulated by these peptides [13].

Initially, AMPs were isolated from the insects' lymph, frog skin and mammalian neutrophil granules [2, 7]. These peptides were tested against a variety of infectious diseases [9] and showed antimicrobial activities [2,5]. These activities have been caused to isolate more than 1700 AMPs from different animal species [10].

AMPs show different sequences, structures and specific targets. These peptides based on their sequence, can be divided into three main groups: 1) Linear amphipatic peptides lacking Cys and often with α-helical structure; 2) Peptides with three disulfide-bridges; 3) Peptides with unusual frequency in certain amino acids such as proline, arginine, tryptophan or histidine [7].

Some AMPs have a series of common characteristics, including small size (10 to 50 amino acid residues), net positive charge of +2 to +9 and molecular weight of less than 10KDa. These peptides are known cationic AMPs [2, 7, 10]. Despite the small size and common physiochemical characteristics, cationic AMPs have

structural diversity. According to their secondary structure, these peptides are divided to following four groups:

1) Amphipatic and α-helical peptides
2) Amphiphilic peptides with 2 to 4 β-strand
3) Loop structures
4) Extended structure [2].

Despite the intense variety in primary and secondary structures, AMPs have widespread effects against broad range of infectious agents including Gram-positive and Gram-negative bacteria, fungi, viruses, parasites and also tumor cells [1, 5, 6, 10, 11]. In addition, some cationic AMPs have inhibitory activity against antibiotic-resistant bacteria such as methicillin-resistant staphylococcus aureus (MRSA) [1, 2, 6].

Specific feature of AMPs is selective interaction with lipid membrane of bacterial cells, which have negative charge [5, 12]. The net positive charge of cationic AMPs causes to binding on bacterial surface, consisting anionic polymers such as lipoteichoic acids (LTA), interaction with bacterial membrane and membrane breaking [4]. This feature can reduce the possibility of resistance against AMPs, and is considered a major advantage compared to antibiotics [5]. The majority of available antibiotics are usually metabolic enzyme targets inhibitors. This feature makes possible a drug resistance [1]. In addition, the different internal signaling pathways are affected by AMPs. This character also reduces the possibility of drug resistance [6]. Therefore, it is likely that AMPs are promising alternatives to traditional antibiotics, particularly against antibiotic-resistant pathogens [1, 3, 6, 7, 12]. Due to broad antimicrobial spectra and difficulty in causing resistance, AMPs extracted from scorpion venoms show great promise as novel antibiotics. Developing strong AMPs with less toxicity to host cells is crucial for clinical applications.

2. Isolated AMPs from scorpion venoms

Scorpions are specifically resistant against bacterial infections. This resistance has always been of interest [5]. Scorpion venom is a rich source of peptides and proteins with diverse biological and pharmacological activities [10, 11, 15, 16]. Numerous peptides with therapeutic qualities, pharmacological probes, or physiological probes have been extracted from scorpion venom in recent decades [1, 2, 6, 8]. So scorpion venom is a rich source of raw AMPs [1] with tremendous potential for the new drugs development [10, 16].

In general, the scorpion venom includes different compounds that can be divided into three groups based on molecular weight: a) The first group consists of proteins with enzymatic activity such as hyaluronidase, phospholipase and Sphangomyelinases; b) The second group consists of peptide fractions with molecular weight less than 10 KDa such as cytolitic agents and toxins; c) The third group consists of various components such as ions, free amino acids, biogenic amines, neurotransmitters, acylpolyamines, heterocyclic compounds and alkaloids [5]. According to the primary structure, these peptides themselves are also classified into two major groups: 1) Cys-rich peptides with disulfide bridges (DBPs: Disulfide-Bridges Peptides); 2) Non-Disulfide-Bridges peptides (NDBPs) [3,11,16]. Most DBPs are responsible for the toxicity of scorpion venom [11, 16]. They affect the functions of ion channels and other membrane receptors in target tissues or prey [14, 17]. Most of these toxins have 28 to 98 amino acid residues (2-8 KDa) [18] with three to four disulfide-bridges. This backbone was established to interact with different ion channels [10,14,16]. In addition, these peptides can be divided into four groups according to their targets [3, 11, 14, 16]:

Specific sodium channels toxins: These toxins are responsible for lethality of scorpion venom [19]. These toxins containing 53 to 78 amino acid residues (5-8 KDa) that are stabilized by four disulfide-bridges [15, 19, 20]. The gating mechanisms of Na+ channels in excitable and non-excitable cells are affected by these toxins [15, 16].

Specific potassium channel toxins: These toxins are short-chain peptides containing 20 to 43 amino acid residues (KDa 3-5), which are stabilized by three or four disulfide-bridges [15, 19, 20].

Specific Cl- channels toxins

Specific calcium channels toxins

Recent studies were primarily concentrated on the properties of disulfide bridge-rich ingredients [16]. But the scorpion venom also contains peptides with low molecular weight (1-4 KDa) [11] which are non-disulfide-bridges peptides (NDBPs) [14, 16]. These components have been less studied [16]. The structure, evolution, and pharmacological activities of these peptides have high difference than disulfide bridges- rich peptides. These peptides have normally 13 to 74 amino acid residues [16] and are divided into two categories: cationic peptides, and highly acidic peptides.

Cationic peptides of scorpions have typically α-helical and amphipatic structure [14] with high variation in primary and secondary structures, and a variety of activities and functions [3, 11]. The majority of these peptides exhibit antimicrobial activity [3, 17] and may interact with the membranes of bacteria and mammals and/or form trans-membrane pores [3]. A number of these peptides also interfere directly with the human innate immune system functions [3] and have immunomodulatory effects by altering signaling pathways [14, 17].

Non-Disulfide Bridge Peptides (NDBPs), a complex combination of AMPs lacking disulfide bands found in scorpion venom, potentiate bradykinin by blocking the conversion of angiotensin 1 to angiotensin 2 through the inhibition of Angiotensin-Converting Enzyme activity,

ultimately lowering blood pressure in the victims. This NDBP characteristic is proposed as biological drug potential [3, 14, 17].

2. Discussion

Based on the primary sequences and Cys structures, the isolated AMP molecules from scorpion venoms are categorized into three groups:

1) The first is scorpine and its homologues. This AMP with antibacterial and anti-malarial activity was isolated from *P.imperator* venom. It has 75 to 78 amino acid residues with three Disulfide-Bridges [20].

2) The second group is long non-disulfide-bridges peptides (NDBPs), with 41 to 44 amino acid residues. These peptides have antibacterial effects along with cytolitic activities [14].

3) The third group is short NDBPs with 13 to 24 amino acid residues. Most members have both anti-bacterial and cytolitic activities [6].

Reduced hemolytic activity and increased antimicrobial activity of AMPs isolated from scorpion venom are valuable solution to design new drugs.

The therapeutic index (TI) can be used as an indicator of selectivity toward bacterial cells. TI is proportional of the concentration of peptide that induces 50% hemolysis at its MIC. The greater index shows more selectivity against bacterial cells [12].

One of most deadly antibiotic-resistant pathogens is methicillin-resistant *staphylococcus aureus* (MRSA) [1, 4]. This pathogen is causing hospitalization of more than 5500 people in the United States, yearly. In a 6-years period, the number of people who have been hospitalized due to the infection was more than twice and high costs of treatment have been spent [2, 4]. Vancomycin is one of the most effective antibiotics against MRSA with many side-effects. The prevalence of vancomycin using against MRSA and existing vancomycin-resistant entrococci has led to emergence Vancomycin-intermediate *S.aureus* [1, 2].

So far, various effective antimicrobial compounds have been found against different types of bacterial strains, including MRSA and other resistant bacteria like:

Penicillin-resistant *S. aureus* (PRSA)
Penicillin-resistant *S. epidermidis* (PRSE)
Methicillin-resistant *coagulase-negative Staphylococcus* (MRCNS)
Penicillin-sensitive *S. epidermidis* (PSSE)
Penicillin-resistant *Enterococcus faecalis* (PREF)
Multidrug-resistant *Pseudomonas aeruginosa* (MDRPA)

Many antimicrobial compounds have been isolated from scorpion venom. Names and details of some of these effective compounds against a variety of antibiotic-resistant pathogens are given in Table 1. As shown in Table 2, most of these compounds have potent effects (as for their MIC_s) against various strains of antibiotic-resistant pathogens.

According to the table (2), isolated AMPs appears to have a greater impact on methicillin-resistant *staphylococcus aureus* strains compared to similar antibiotics, such as cefotaxime, penicillin and equal to vancomysin. All AMPs have more potency than penicillin. Among the different types of AMPs, vejovine has specific effect on multidrug-resistant bacteria such as *Escherichia coli*, *Enterobacter cloacae*, *Klebsiella pneumoniae*, *Pseudomonas aeruginosa* and *Acinetobacter baumanii*.

Kn2-7 has the greatest impact against MRSA, MRCNS and PSSE, and this effect is much stronger than that of vancomycin. BmKn2 shows its maximal antimicrobial effect against PSSE. This effect is equal to cefotaxime and more than penicillin. Citriporin has greatest effect on MSSA. The effect against antibiotic-resistant pathogens was less than vancomycin and more than penicillin. Although imcroporin has been effective on antibiotic-resistant pathogens, the effect is much smaller than vancomycin. The maximum antimicrobial effect of imcroporin was shown against PSSE. IsCT and its derivative compounds, IsCT-P and IsCT-a, have been tested against the multidrug-resistant pathogens. The antimicrobial effectiveness was significant against MRSA and also other pathogens. It should be noted that the change of amino acids of IsCT with positively charged amino acids to enhance the flexibility of α-helix structure of this peptide has not had much influence on their performance. Mucroporin-M1 peptide isolated from scorpion *Lychas muronatus* showed greatest impact on PRSE, MRSA, MRCNS and PSSE. It should be noted that this peptide has shown better performance against methicillin-resistant pathogens than cefotaxime. The highest effects of StCT2 were shown on MRSA, MRCNS and PSSE. As shown in Table 2, Kn2-7 is likely the most potent antimicrobial peptide isolated from scorpion venom that is effective against the four classes of resistant strains to methicillin and penicillin. This peptide has a large number of positively charged amino acids [1, 2, 4, 6, 12, 22-24].

The majority of AMPs have common motif of WEGXKS (X can be any amino acid residue), which indicates the important role of this motif for antimicrobial effects [6]. It also appears that the N-terminal α-helical region is essential for the antimicrobial activity of AMPs [21].

When designing peptides with increased biological activity, the relationship between the structure and function of AMPs is crucial [2]. The antimicrobial potency may increase with increased positive charges [11]. The increased number of positively charged amino acids raises the affinity of peptides to the bacterial membrane [4].

Table 1: Isolated AMPs from scorpion venom affecting antibiotic-resistant pathogens**.**

Name	Source	Amino acids (aa) Sequence	Number of aa	Molecular mass (Da)	Charge	Disulfide-bridge	Hc50 (µg/ml)	Ref.
Ctriporin	*Chaerilus tricostatus*	FLWGLIPGAVTSLIAISKK-NH2	19	2014.2	2	0		1
Imcroporin	*Isometrus maculates*	FFSLLPSLIGGLVSAIK-NH2	17		1	0	84	2
Kn2-7	derived of BmKn2 from *Mesobuthus martensii Karsch*	FIKRIARLLRKIF	13		5	0	90.27	4
BmKn2	*Mesobuthus martensii Karsch*	FIGAIARLLSKIF	13		2	0	17.13	4
StCT1	*Scorpiops tibetanus*	GFWGSLWEGVKSVV-NH2	14	1549.2	0	0		6
IsCT	*Opisthacanthus madagascariensis*	ILGKIWEGIKSLF-NH2	13		1	0	18 µM	12
IsCT-P	derived of IsCT from *Opisthacanthus madagascariensis*	ILKKIWKPIKKLF-NH2	13	1654.5	5	0	400 < (0%)*	12
IsCT-a	derived of IsCT-P from *Opisthacanthus madagascariensis*	ILKKIWKaIKKLF-NH2	13	1628.7	5	0	400 < (0%)*	12
Mucroporin-M1	derived of Mucroporin from *Lychas muronatus*	LFRLIKSLIKRLVSAFK-NH2	17	2031.57	5	0		22
StCT2	*Scorpiops tibetanus*	GFWGKLWEGVKSAI-NH2	14	1576.9	1	0	80.3	23
Vejovine	Vaejovis mexicanus	GIWSSIKNLASKAWNSDIGQSL RNKAAGAINKFVADKIGVTPSQAAS	47	4873	4	0	100 µM	24

*: Parenthesis indicates % hemolysis of the peptides at 400 µM

Positively charged amino acids interaction with the negatively charged outer membrane of bacteria leads small opening formation in bacterial membrane and cell lysis [21]. So, replacing a neutral or negatively charged amino acid with positively charged amino acids such as Lys using site-directed mutagenesis resulted in increased antimicrobial activity [2, 10, 16].

It is known that the relative potency of AMPs is related to the content of α-helical [11]. Thus, the antimicrobial activity is dependent to α-helicity of the peptide. The α-helicity is increased by replacing neutral and acidic amino acids on the hydrophilic α-helix surface [2]. In the active membrane of α-helical AMPs, proline plays an important role in the effect on ion channels, the pore-forming in biological membranes and increase of α-helix flexibility. This amino acid cannot form intermolecular hydrogen bonds. Like proline, peptoide also is an imino acid. This imino acid may disrupt the stable α-helical structure of a peptide. Peptoide is unable to create backbone-to-backbone hydrogen bonds because it does not have an amide proton. So replacing one amino acid with an amino acid, such as Pro or peptoide, improve the structural flexibility [12].

Despite all the advantages of AMPs towards available antibiotics, their therapeutic potential is limited due to high hemolytic activity and their non-specific binding to the mammalian cell membrane [4, 12]. These binding leads hemolysis and loss the normal cell function [3]. Increasing the net positive charge of AMPs may be an appropriate way to reduce the high hemolytic activity [4]. Increased hydrophobicity is also strongly associated with increased hemolytic activity. Zwitterionic mammalian membranes contain large amounts of cholesterol and less negative charge than bacterial membranes. There is no cholesterol in bacterial membranes. Therefore, increasing the hydrophobicity raises the interaction between peptides and mammalian membrane [11]. Hemolytic activity of scorpion AMPs is associated with chain length, so that the hemolytic activity of long-chain amphipatic peptides is much weaker than short-chain amphipatic peptides [3].

One of other restrictions on scorpion venom using is that the antimicrobial activity is only seen with fresh venom. This activity doesn't show in lyophilized or stored venom [15].

Table 2: Minimal inhibitory concentration (MICs) of Isolated AMPs from scorpion venom against different clinical antibiotic-resistant bacterial strains (µg/ml)**.**

	Kn2-7	BmKn2	StCT1	Ctriporin	Imcroporin	IsCT	IsCT-P	IsCT-a	Mucroporin-M1	StCT2	Vejovine	Vancomycin	Penicillin	Cefotaxime
Penicillin resistant														
PRSA (P1383)	6.25	12.5	--	--	--	--	--	--	10	12.5	--	6.83	10000	6
PRSA (P838)	--	--	50	--	--	--	--	--	--	--	--	--	200	12.5
PRSE (P1389)	6.25	12.5		10	50	--	--	--	8	12.5	--	6.5	10000	4.5
PREF (P675)	--	--	50	--	--	--	--	--	--	--	--	--	100	12.5
Methicillin resistant														
MRSA (P1374)	3.13	12.5	200	10	50	--	--	--	8	6.25	--	6.5	4000	400
MRSA (P1381)	6.25	12.5	500	--	--	--	--	--	20	25	--	5.8	5000	400
MRSA (P1386)	6.25	12.5	250	10	50	--	--	--	25	12.5	--	4.3	10000	100
MRSA (CCARM 3001)	--	--	--	--	--	2 µM	2 µM	2 µM	--	--	--	--	--	--
MRSA (CCARM 3543)	--	--	--	--	--	1 µM	1 µM	1 µM	--	--	--	--	--	--
MRCNS (P1369)	3.13	12.5	125	10	50	--	--	--	8	6.25	--	6.5	20000	400
Multidrug resistant														
MDRPA (CCARM 2095)	--	--	--	--	--	2 µM	1 µM	2 µM	--	--	--	--	--	--
Escherichia coli 170	--	--	--	--	--	--	--	--	--	--	4.4 µM	--	--	--
Enterobacter cloacae 2524	--	--	--	--	--	--	--	--	--	--	4.4 µM	--	--	--
Klebsiella pneumoniae 913	--	--	--	--	--	--	--	--	--	--	17.7 µM	--	--	--
Pseudomonas aeruginosa 4667	--	--	--	--	--	--	--	--	--	--	17.7 µM	--	--	--
Acinetobacter baumanii 7847	--	--	--	--	--	--	--	--	--	--	5 µM	--	--	--
Penicillin sensitive														
PSSE (P1111)	3.13	6.25	--	--	20	--	--	--	20	6.25	--	5.4	25	6.25
PSSE (P1368)	--	--	--	--	--	--	--	--	8	--	--	8	40	10
PSSA (P969)	--	--	--	--	--	--	--	--	40	--	--	8	4	5
Methicillin sensitive														
MSSA (AB94004)	--	--	--	5	--	--	--	--	--	--	--	3	6.25	3

penicillin-resistant *S. aureus* (PRSA), penicillin-resistant *S. epidermidis* (PRSE), methicillin-resistant *S. aureus* (MRSA), penicillin-sensitive *S. epidermidis* (PSSE), penicillin-resistant *Enterococcus faecalis* (PREF), Methicillin-sensitive *S. aureus* (MSSA), Multidrug-resistant *Pseudomonas aeruginosa* (MDRPA), penicillin-sensitive *S. aureus* (PSSA), methicillin-resistant coagulase-negative Staphylococcus (MRCNS).

Other limitation is that the direct purification of venom is difficult. In addition, the production of recombinant peptide less than 50 residues is difficult. The molecular weight of less than 5 KDa complicates their purification far from many peptides and biological molecules produced by *E. coli* or other organisms. So the best way is peptide synthesis [3].

4. Conclusion

Since microbial infections have a high global rate of morbidity and mortality, they pose a serious threat to public health. Microorganisms develop resistance through a variety of molecular mechanisms, which diminishes their efficacy. Therefore, it's necessary to look for fresh opportunities in naturally occurring sources that have antimicrobial activity. Using peptides found in various scorpion venoms is one substitute.

It is challenging to categorize scorpion NDBPs based on sequence or structural similarity because of the high divergence of their primary and secondary structures. This review's primary objective is based on a comparison of their pharmacological and antimicrobial properties.

In this review, for the first time, a summary of NDBPs with penicillin-resistant S. aureus (PRSA) Penicillin-Resistant S. Epidermidis (PRSE), Methicillin-Resistant S. aureus (MRSA) Penicillin-Sensitive S. epidermidis (PSSE), Penicillin-Resistant Enterococcus faecalis (PREF), Methicillin-Sensitive S. aureus (MSSA), Multidrug-Resistant Pseudomonas aeruginosa (MDRPA), Penicillin-Sensitive S. aureus (PSSA), Methicillin-Resistant Coagulase-Negative Staphylococcus (MRCNS) properties isolated from the scorpion venoms are described. Although non-disulfide bond-containing peptides (NDBPs) make up to 5% of the composition of scorpion venom, short, linear, NDBPS have received less research attention than neurotoxins. These peptides are more effective than available antibiotics against the majority of resistant pathogens to antibiotics. Therefore, these peptides may be regarded as a lead compound for the treatment of pathogens resistant to antibiotics or as a prospective anti-infective medication.

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Conflict of interest

There is no conflict of interest.

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References

[1] Fan Z, Cao L, He Y, Hu J, Di Z, Wu Y, W Li, et al. Ctriporin, a new anti-methicillin-resistant Staphylococcus aureus peptide from the venom of the scorpion Chaerilus tricostatus. *Antimicrob. Agents Chemother* (2011) 55(11):5220-9.
[2] Trentini MM, DasNeves RC, Santos BDPO, aSilva RA, Barros de Souza AC, Mortari MR, Schwartz EF, et al. Bridge pepide 5.5 from the scorpion Hadrurus gertschi inhibits the growth of mycobacterium abscessus subsp. massilliense. *Front. Microb*. (2017) 8:273.
[3] Pedron CN, Araujo I, da Silva Junior PI, da Silva FD, Torres MDT, Junior VXO. Repurposing the scorpion venom peptide VmCT1 into an active peptide against Gram-negative ESKAPE pathogens. *Bioorg. Chem.* (2019) 90:103038.
[4] Cao L, Dai C, Li Z, Fan Z, Song Y, Wu Y, Cao Z, et al. Antibacterial activity and mechanism of a scorpion venom peptide derivative in vitro and in vivo. *PloS One*. (2012) 7(7):e40135.
[5] Baradaran M, Jalali A, Jolodar A, Ghasemian S. A new caerin-like antibacterial peptide from the venom gland of the Iranian scorpion Mesobuthus eupeus: cDNA amplification and sequence analysis. *Afr. J. Biotech*. (2012) 11(44):10176-81.
[6] Yuan W, Cao L, Ma Y, Mao P, Wang W, Zhao R, Wu Y, et al. Cloning and functional characterization of a new antimicrobial peptide gene StCT1 from the venom of the scorpion Scorpiops tibetanus. *Peptides* (2010) 31(1):22-6.
[7] Amorim-Carmo B, Adriana MS, Souza PES, Silva-Junior AA, Araújo RM, Fernandes-Pedrosa MF. Antimicrobial Peptide Analogs from Scorpions: Modifications and Structure-Activity. *Front. Mol. Biosci*. (2022): 887763
[8] Shao JH, Wang YQ, Wu XY, Jiang R, Zhang R, Wu CF, Zhang JH. Cloning, expression, and pharmacological activity of BmK AS, an active peptide from scorpion Buthus martensii Karsch. *Biotech. Lett.* (2008) 30(1):23-9.
[9] Gao B, Xu J, Rodriguez Mdel C, Lanz-Mendoza H, Hernandez-Rivas R, Du W, Zhu S. Characterization of two linear cationic antimalarial peptides in the scorpion Mesobuthus eupeus. *Biochimie* (2010) 92(4):350-9.
[10] Velasco-Bolom JL, Corzo G, Garduño-Juárez R. Folding profiles of antimicrobial scorpion venom-derived peptides on hydrophobic surfaces: a molecular dynamics study. *J. Biomol. Struct. Dyn*. (2020) 38(10):2928-2938.
[11] Almaaytah A, Zhou M, Wang L, Chen T, Walker B, Shaw C. Antimicrobial/cytolytic peptides from the venom of the North African scorpion, Androctonus amoreuxi: biochemical and functional characterization of natural peptides and a single site-substituted analog. *Peptides* (2012) 35(2):291-9.
[12] Lim SS, Yoon SP, Park Y, Zhu WL, Park IS, Hahm KS, Shin SY. Mechanism of antibacterial action of a synthetic peptide with an Ala-peptoid residue based on the scorpion-derived antimicrobial peptide IsCT. *Biotech. Lett.* (2006) 28(18):1431-7.
[13] Bringans S, Eriksen S, Kendrick T, Gopalakrishnakone P, Livk A, Lock R, Lipscombe R. Proteomic analysis of the venom of *Heterometrus longimanus* (Asian black scorpion). *Proteomics* (2008) 8(5):1081-96.
[14] Li F, Lang Y, Ji Z, Xia Z, Han Y, Cheng Y, Liu G, et al. A scorpion venom peptide Ev37 restricts viral late entry by alkalizing acidic organelles. *J. Biol. Chem.* (2019) 294: 182-194.
[15] Garcia-Gomez BI, Coronas FI, Restano-Cassulini R, Rodriguez RR, Possani LD. Biochemical and molecular characterization of the venom

from the Cuban scorpion Rhopalurus junceus. *Toxicon* (2011) 58(1):18-27.
[16] Dias NB, de Souza BM, Cocchi FK, Chalkidis HM, Dorce VAC, Palma MS. Profiling the short, linear, non-disulfide bond-containing peptidome from the venom of the scorpion *Tityus obscurus*. *J. Proteomics* (2018) 170:70-79.
[17] Harrison PL, Heath GR, Johnson BRG, Abdel-Rahman MA. Strong PN, Evans SD, Miller K. Phospholipid dependent mechanism of smp24, an α-helical antimicrobial peptide from scorpion venom. *Biochim. Biophy*. *Acta* (2016) 1858: 2737-2744.
[18] Borges A, Silva S, Op den Camp HJ, Velasco E, Alvarez M, Alfonzo MJ, Jorquera A, et al. In vitro leishmanicidal activity of Tityus discrepans scorpion venom. *Parasito. Res.* (2006) 99(2):167-73.
[19] Luna-Ramirez K, Quintero-Hernandez V, Vargas-Jaimes L, Batista CV ,Winkel KD, Possani LD. Characterization of the venom from the Australian scorpion Urodacus yaschenkoi: Molecular mass analysis of components, cDNA sequences and peptides with antimicrobial activity. *Toxicon* (2013) 63:44-54.
[20] Feng J, Yu C, Wang M, Li Z, Wu Y, Cao Z, Li W, et al. Expression and characterization of a novel scorpine-like peptide Ev37, from the scorpion *Euscorpiops validus*. *Protein Expr. Purifi.* (2013) 88(1):127-33.
[21] Miyashita M, Sakai A, Matsushita N, Hanai Y, Nakagawa Y, Miyagawa H. A novel amphipathic linear peptide with both insect toxicity and antimicrobial activity from the venom of the scorpion *Isometrus maculatus*. *Biosci Biotech. Biochem.* (2010) 74(2):364-9.
[22] Dai C, Ma Y, Zhao Z, Zhao R, Wang Q, Wu Y, Cao Z, et al. Mucroporin, the first cationic host defense peptide from the venom of Lychas mucronatus. *Antimicrob. Agents Chemother*. (2008) 52(11):3967-72.
[23] Cao L, Li Z, Zhang R, Wu Y, Li W, Cao Z. StCT2, a new antibacterial peptide characterized from the venom of the scorpion Scorpiops tibetanus. *Peptides* (2012) 36(2):213-20.
[24] Hernandez-Aponte CA, Silva-Sanchez J, Quintero-Hernandez V, Rodriguez-Romero A, Balderas C, Possani LD, Gurrola GB. Vejovine, a new antibiotic from the scorpion venom of *Vaejovis mexicanus*. Toxicon (2011) 57(1):84-92.