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Review Article

Applications of Spider Venom in Medical Science and Agriculture

HAFIZ MUHAMMAD TAHIR[1], AROOJ ZAHEER[1], RABIA YAQOOB[1], ABDUL KHALIQ[2] & KHANUM ZAHRA[1]

[1]Department of Zoology, University of Sargodha, Sargodha, Pakistan
[2]University College of Agriculture, University of Sargodha, Pakistan

ABSTRACT

Spider venom comprises of 10 million bioactive peptides but only 800 peptides are pharmacologically characterized. Despite this limited number of characterized peptides, the range of biological activities they perform is incredible. Their venom is able to potently block a variety of receptors, channels, enzymes and other various target sites. Therefore, it is not surprising that the peptides extracted from spider venom possess substantial potential for therapeutic applications. This review article encompasses the potential applications of spider venom as analgesics, anti-fungal, anti-parasitic and anti-bacterial agent, and bio-insecticides. Moreover, some of the venom peptides have marked antiarrhythmic and neuroprotective potentials. With these potentials spider venom could be utilized in development of prototypes for pharmaceutical applications.
Key Words: Spider venom, therapeutic agents, bio-insecticides, AMPs, antibacterial potential.

INTRODUCTION

Spiders like other venomous animals, such as scorpions, centipedes, cone snails and snakes, are capable of producing venom that holds a diverse array of biologically active compounds. Spiders are considered as one of the most successful and abundant terrestrial predator with approximately 45,840 extant species (http://wsc.nmbe.ch, version 17.0). Among the animals, employing venom for their existence, spiders are the most cosmopolitan predator with a wide range of distribution throughout the globe. They potentially devour the most diverse and varied range of prey (Saez *et al.,* 2010). Spiders are adept and proficient predators that produce venom containing a large variety of toxins. These toxins target a diverse range of sites including membranes, receptors, channels, and enzymes in a vast variety of invertebrate and vertebrate species (Saez *et al.*, 2010). Spiders venom comprises of heterogeneous compounds of different chemical nature, that can traditionally be divided into three groups on the basis of molecular mass: (i) substances with low molecular weight (less than 1 kilo Dalton) including organic acids, amino acids, amines, polyamines, nucleotides and nucleosides; (ii) peptides (ranging from 1-10 kilo Dalton), with two main subgroups-linear cytolytic peptides and disulfide containing neurotoxins; (iii) substances with high molecular weight (more than 10 kilo Dalton) including different proteins, enzymes and neurotoxins (Escoubas *et al.,* 2000; Adams, 2004; Vassilevski *et al.,* 2009).

Previously, relatively little work has been done on the identification and potentials of peptides isolated from spider venom. That's why, the reported number of pharmacologically characterized peptides in spider venom is comparatively less. But despite of being less studied spiders stood at the top, with its venom having the most diverse range of impressive biological activities including the potential application in therapeutics (Vassilevski *et al.,* 2009). Although the spider venoms are generally thought to be rich in neurotoxic peptides, they also holds various peptidic toxins having anti-cancerous, anti-malarial, anti-microbial, analgesic, cytolytic, hemolytic, and enzyme inhibitory activity (Gao *et al.,* 2005). Currently, spider venom peptides are extensively used in biological research purposes. Scientific innovations have led to development of better techniques for purification of proteins and synthesis of recombinant toxin. Also, at present the researchers utilize the novel technologies to overcome conventional problems of low venom volumes by synthesizing its analogs. Now, the spider venom peptides are largely incorporated for the development of novel therapeutics agents against a large number of medical conditions including cardiovascular disorders, chronic pain, bacterial and fungal infections and chronic neurological diseases. Spider venom peptides also exhibit selective insecticidal potential. The peptidic toxins of spider venom have high target specificity and selectivity. This potential makes spider-venom peptides as lead compound for discovery of novel therapeutic agents.

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**Correspondence author: hafiztahirpk1@yahoo.com*

Analgesic Potential

Our exposure to possibly damaging or life-threatening incidents is limited by common nociceptive pain. This ordinary pain is our adaptive response. Conversely, eccentric long-lasting pain transmutes this essential response into a poorly administered and devastating malady. Chronic pain affects about 15% of the elderly population worldwide (Darrell & Patrick, 2012) and this figure increases up to 50% for those who are more than 65 years old (Brennan *et al.,* 2007). This renders chronic pain as one of the major health problems of the present era. Nevertheless, some drugs are available as medication for chronic pain but these drugs have limited usage due to less efficacy and relatively significant side-effects. Therefore, there is a substantial need to introduce more effective analgesics for the treatment of chronic pain. Currently, various ion-channels have shown to play a significant role in the pathophysiology of pain including acid sensing ion channels, calcium (CaV) and sodium (NaV) channels and purinergic receptors.

Acid sensing ion channels (ASICs) that opens accordingly with low pH are the proton-gated sodium channels. These are characterized by predominantly neuronal distribution. As a novel therapeutic target, ASICs have been employed for a wide range of pathophysiological ailments including chronic pain (Wemmie *et al.,* 2006; Xiong *et al.,* 2008; Sluka *et al.,* 2009; Gründer & Chen, 2010). π-theraphotoxin-Pc1a (π-TRTX-Pc1a), a potent and specific inhibitor of ASIC is isolated from the venom of tarantula, *Psalmopoeus cambridgei*. π-theraphotoxin-Pc1a is proven to be an effective analgesic (Pignataro *et al.,* 2007).

Voltage-gated sodium (NaV) are threshold channel that intensifies pain signals, conducted beyond a certain level (Clare, 2010). Out of 9 NaV, usually three NaV subtypes: NaV1.3, NaV1.7, and NaV1.8 are frequently involved in pain signaling (Krafte & Bannon, 2008; Priest, 2009). However, human NaV1.7 (hNaV1.7) emerged as best analgesic target followed by various notable genetic studies. Spider venom has a potential to modulate several peptidic NaV channel (Herzig *et al.,* 2011; Gilchrist *et al.,* 2013). β-TRTX-Tp2a (Protoxin II) isolated from venom of *Thrixopelma pruriens* (a green velvet tarantula) is believed to be most potent blocker of human NaV1.7 (Maggio *et al.,* 2010). It is a 30-residue inhibitor cystine knot (ICK) peptide. This toxin is 100 times more selective for human NaV1.7 as compared to other NaV subtypes i.e., NaV1.2, NaV1.3, NaV1.5, NaV1.6 and NaV1.8 (Middleton *et al.,* 2002).

P2X purinergic receptors are the ion channels that are known to be non-selective ion channels. They are ATP-gated and have permeability for sodium, potassium and calcium ions (Khakh & North, 2006). Presently, 7 subtypes of P2X receptors (P2X1 to P2X7) are identified (Skaper *et al.,* 2010). In relation to pain P2X3 receptor subtype is most thoroughly studied (Wirkner & Sperlagh, 2007). It was declared to be implicated not only in inflammatory, acute, and chronic pain but is also involved in inducing cancer pain along with visceral and migraine pain (North, 2004; Wirkner & Sperlagh, 2007; Saez *et al.,* 2010). Recently, however, purotoxin-1 (PT1) from the venom of the central Asian spider *Geolycosa* sp. is isolated. It is potent and it selectively modulates the P2X3 receptor subtype (Grishin *et al.,* 2010). Therefore, PT1 seems to be a lead compound that is capable of producing analgesics, targeting P2X3 receptors.

One of calcium channel subtype Cav2.2 is extensively distributed at nerve terminals together with nociceptive fibers. It controls vital physiological processes, including contraction of muscles and secretion or release of hormones and neurotransmitters (McDavid & Currie, 2006). Cav2.2 also plays its role in ascending pathways of acute and chronic pain (nociceptive transmission) (McDonough *et al.,* 2002). In response to painful stimulus from nocireceptors, the neuronal membrane potential is changed. This forces Cav2.2 gates to open resulting in propagation of pain signals. Therefore, the toxins that blocks Cav2.2 have significant analgesic effect (Cizkova *et al.,* 2002; Wallace *et al.,* 2010). Venoms of almost all the poisonous animals including cone snails, snakes, centipedes, assassin bugs, scorpions and spiders are loaded with extremely potent and selective Cav2.2 inhibitors. More specifically Cav inhibitors constitute the major portion of spider venom (Klint *et al.,* 2012). Variety of different peptide toxins such as agatoxins, grammotoxin and DW13.3 found in spider venom potently obstructs the activities of voltage-dependent calcium gates (Zamponi, 1997). ω-agatoxin IIIA is a 76 residue peptide extracted from *Agelenopsis aperta*, a funnel web spider with broad specificity across various voltage-sensitive subtypes of calcium channel (Cohen *et al.,* 1992; Mintz, 1994). DW13.3 from venom of spider *Kukulcania hibernalis* is another peptide with 74 residues. It causes a potent and reversible inhibition of Cav2.1, Cav2.2, Cav1.2 and Cav2.3 channels (Sutton *et al.,* 1998). Ω-ctenitoxin-Pn4a is a toxin extracted from the spider *Phoneutria nigriventer*. It non-selectively blocks neuronal Cav channel (Cordeiro *et al.,* 1993; Cardoso *et al.,* 2003;

Vieira *et al.,* 2005) and displays prolonged analgesia (Souza *et al.,* 2008). Moreover, ω-ctenitoxin-Pn4a does not interfere with average arterial blood pressure, heart beat or neuronal performance. This suggests that Cav channel inhibitors present in spider venoms could also find therapeutic applications as efficient and long lasting analgesic agents (De-Souza *et al.,* 2011). Also Phα1β, a potent calcium channel blocker isolated from *P. nigriventer* has a potential application in managing postoperative pain. It can produce longer pain reducing effects in postoperative phase as compared to ω-conotoxin MVIIA (cone snail) or morphine (De-Souza *et al.,* 2011). Ω-ctenitoxin-Pn2a is another non-selective toxin from venom of *P. nigriventer*, (with sequence of potency: Cav2.1 > Cav2.3 > Cav1 > Cav2.2) (Leao *et al.,* 2000). It displays predominant anti-nociceptive effects and does not cause undesirable motor effects in the neuropathic pain models (Dalmolin *et al.,* 2011). Spider toxins such as ω-Agatoxins IVA6 (Bourinet *et al.,* 1999) and ω-grammotoxin (Li-Smerin & Swartz, 1998) consisting of 60-90 and 30-40 residue peptides respectively have also shown analgesic effects by inhibiting CaV channel (McDonough *et al.,* 1997).

Naturally, spider-venom has great affinity and selectivity for a varied range of molecular targets. Therefore, it is not surprising that various peptides isolated from spider venom potently blocks activity of a wide variety of therapeutic targets including analgesic targets.

Treatment of Neurological Diseases

Neurological disorders have attracted great interest due to their high impact on society (WHO, 2006). Most of these disorders have a chronic profile. Even with the increasing search for new treatments, there is still a lack of pharmacological therapy that is able to efficiently control the progression of these diseases. Considering this dilemma, research is underway of using natural products for modern drug discovery processes (Monge-Fuentes *et al.,* 2015). On this aspect, the attention of researches has been attracted by peptides and acylpolyamines isolated from the arthropod venom. These have analgesic, antiepileptic and neuroprotective effects (Estrada *et al.,* 2007; Mortari *et al.,* 2007; Mortari & Cunha, 2013; Youdim *et al.,* 2014). These venom components have been tested for the treatment of the four most prevalent neurological disorders: Stroke, Alzheimer's disease (AD), Epilepsy, and Pathological anxiety (Monge-Fuentes *et al.,* 2015).

It is estimated that throughout the world about 16.9 million people suffer from stroke every year. Stroke is also mentioned as the second leading cause of death (Mehndiratta *et al.,* 2015). Stroke results in neuronal cell death. Excessive activation of glutamate receptors is involved in brain damage following stroke. Synthetic glutamate antagonists have failed in neuroprotection against stroke with considerable side effects (Leoni *et al.,* 2000; Xiong *et al.,* 2004). Therefore, the researchers have focused on natural glutamate receptor antagonists found in majority of spider's venoms. Acylpolyamines found in the venom of *A. aperta*, a funnel web spider, are selective non-competitive glutamate receptor (Glu-R) antagonist. They blocks glutamate receptors, thus preventing excessive Ca2+ influx which produces neuroprotective effects (Mueller *et al.,* 1999). Peptides toxins PhTx3-3 and PhTx3-4, extracted from venom of spider *P. nigriventer* also have neuroprotective action against neuronal damage (Cordeiro *et al.,* 1993). These compounds block the Ca2+ channel resulting in decreased neuronal death (Prado *et al.,* 1996) and loss of neurotransmission in hippocampus (Pinheiro *et al.,* 2009). In the light of above discussion it can be concluded that acylpolyamines and peptides present in spider venom can serve as a platform to design new drugs for the treatment of stroke in humans (Monge-Fuentes *et al.,* 2015).

Alzheimer's disease (AD) is a form of severe dementia characterized by memory loss and a lower capacity for recognizing objects and making plans. Dementia affects 35.6 million people worldwide, out of which 60-70% cases are of AD (WHO, 2012). The main pathological hallmark found in AD patients is the presence of amyloid-β (Aβ) aggregates and neurofibrillary tangles of tau protein (Ross & Poirier, 2004). Due to these neuropathological aggregations, neuronal loss occurs, especially in the hippocampus and basal forebrain. Therapeutic approaches remain focused on the symptomatic treatment of the disease, as there is no approved drug able to effectively stop the progression of disease (Huang & Mucke, 2012; Mancuso & Gaetani, 2014). The effect of peptide Tx3-1, a selective blocker of K+ currents, extracted from venom of spider *P. nigriventer*, was evaluated in mice. Results showed the ability of the toxin to enhance both short- and long-term memory. Moreover, Tx3-1 restored memory of injected mice and exhibited higher potency to improve memory of injected mice when compared to control group (Gomes *et al.,* 2013).

Epilepsy is characterized by uncontrolled spontaneous epileptic seizures. Antiepileptic drugs (AEDs) are given to epileptic patients to eliminate or reduce seizures to the maximum degree. Despite

drug availability, approximately 30% of patients of epilepsy are regarded as therapy-resistant and they continue to live with uncontrolled seizures (Bialer, 2012). In this scenario, arthropod venoms may represent an extraordinary source of bioactive molecules that act with selectivity and specificity in the mammalian CNS (Monge-Fuentes *et al.,* 2015). Among arthropods, spider's venom contains a diverse array of such bioactive compounds that produce neuroprotective and antiepileptic effects by acting on voltage-sensitive Na+ and Ca2+ channels or on glutamate receptors (GluR) (Rajendra *et al.,* 2004). Neuroactive effects are observed for polyamine toxins obtained from venom of *Nephilia clavata* spiders (Kawai *et al.,* 1982). The venom of *Argiope lobata* and other members of the Araneidae family holds a biologically important compound named Argiotoxins. Argiotoxins are neuroprotective and inhibit cell death mechanisms that involve ionotropic GluR (Green *et al.,* 1996; Grishin, 1999Rash & Hodgson, 2004). Moreover, argiotoxin-636 (from *Argiope lobata*), Parawixin2 (Liberato *et al.,* 2006) and parawixin10 (from *Parawixia bistriata* spider) have an antiepileptic effect by blocking the seizures inducing receptors (Gelfuso *et al.,* 2014). A neuroactive fraction obtained from spider *Lycosa erythrognatha* (SrTx1) also exhibit antiepileptic effects (Cairrão *et al.,* 2002). Agatoxin-489 a peptide isolated from *A. aperta* is used as antiepilectic due to its ability to block KA-induced seizures (Williams, 1993).

Pathological anxiety corresponds to the most prevalent psychiatric disorder including: panic disorders, obsessive-compulsive disorder, generalized anxiety disorder, various phobias and disorders due to post-traumatic stress. These disorders are characterized by excessive worrying, uneasiness, and fear of some future events. This includes several neurotransmitters executing their role in numerous receptors inside the limbic system of cerebral cortex (Durant *et al.,* 2010). In addition to benzodiazepines, antiepileptics, antidepressants, and antipsychotics, have also been used to treat anxiety disorders (Gelfuso *et al.,* 2014). Although in recent decades the pharmacological treatment for anxiety disorders has become increasingly more available, there is still a great need to develop new drugs with greater efficacy and tolerability, limited abuse potential, and fewer side effects (Gelfuso *et al.,* 2014). In this sense, arthropod venoms may be useful for the development of novel pharmacological tools directed toward the treatment of pathological anxiety. Neurological activity of parawixin-2, isolated from the spider *P. bistriata*, demonstrated anxiolytic effects (Beleboni *et al.,* 2006; Liberato *et al.,* 2006). It blocks gamma-aminobutyric acid (GABA) receptors which are responsible for anxiolytic-like behaviors due to an increase in endogenous GABAergic activity (Schmitt *et al.,* 2002).

The scarcity of studies directly correlating neurotoxins from arthropod venoms and anxiety disorders, coupled with the great potential of some molecules for the treatment of this pathology, reveals a highly promising and still underexplored scientific area.

Antiarrhythmic Drugs From Spider Venoms

Cardiac arrhythmia is a problem with the rate or rhythm of the heartbeat. Most of arrhythmia are asymptomatic and are generally considered as harmless. But severe arrhythmias arising from weakened or damaged heart can even lead to stroke, heart attack and cardiac arrest (Andrew & Epstein, 2015). Arrhythmia is correlated with the activity of mechanosensitive channels that causes stretching of atrial chamber. Mechanosensitive channels (MSCs), also known as stretch-activated channels, have distribution among all the cells (Martinac & Kloda, 2003). M-TRTX-Gr1a (GsMTx4) and κ-TRTX-Gr2a isolated from the venom of tarantula, *Grammostola rosea*, are the 2 MSC's selective blockers (Suchyna *et al.,* 2000; Oswald *et al.,* 2002). κ-TRTX-Gr2a has a low affinity for inhibition of MSCs. Whereas, M-TRTX-Gr1a potently blocks the MSCs (Suchyna *et al.,* 2000). M-TRTX-Gr1a cannot be itself used as therapeutic agent as it is unable to directly interact with MSCs (Saez *et al.,* 2010). Yet, M-TRTX-Gr1a can be used as a valuable tool for revealing the abilities of MSCs as a therapeutic target for the medication of ailments such as cardiac arrhythmias, dystrophy of muscles, spinal cord injuries and gliomas (Bowman *et al.,* 2007).

GsMtx-4 is an active peptide isolated from the venom of Chilean spider *Grammostola spatulata*. This peptide is found to be useful in regulating and adjusting rapid and irregular electrical activity in the atrial chamber of heart. It is also shown that this peptide produces no effect on normal heart that is unstretched (Dobson, 2001) suggesting the side effects to be minimal. Another peptide, PhKv (Tx3-1), isolated from the venom of spider *P. nigriventer* is a neurotoxin that act as potent antiarrhythmic agent by blocking voltage activated A-type K+ currents in the GH3 neuroendocrinal cell line (Cordeiro *et al.,* 1993; Almeida *et al.,* 2011), This raises the possibility that antiarrhythmic effects produced by PhKv is due to its direct effect on electrical properties of cardiomyocytes (Almeida *et al.,* 2011).

Anticancerous Potential

Cancer is categorized as the set of diseases that involves aberrant and unchecked cell growth. Cancerous cells possess the ability to invade and proliferate to the other body parts**.** In year 2012, approximately 14.1 million new cancer cases were reported globally (WHO, 2014) along with 8.2 million reported deaths that constitutes nearly 14.6% of all human fatalities. In 2010, cancer has been estimated to cause the financial cost of more than $1.16 trillion US$ (WHO, 2014). Several options are available for the treatment of cancer, including surgical removal of tumors, chemotherapy, radiotherapy and targeted and hormonal therapy. However, their usage is generally restricted by the fact that toxicity is also induced in the tissues other than the target tissue. Therefore, there is great need of an alternative medication for cancer.

In this context the toxins present in spider venom bear great anti-tumor potential. The venom of brown spider contains a toxic substance named as phospholipase-D. It exhibits significant hemolytic activity (Silva *et al.,* 2000) along with recognized anticancerous effect. Another compound, hyaluronidase present in the venom of various spiders, potentially boosts up the tissue permeability. This facilitates the penetration of certain drugs. Also, due to enhancing effect of spider venom on tissue permeability, it can even directly be used as anti-cancer agent (Matsushita & Okabe, 2001; Girish & Kemparaju, 2007). The venom of *Oxyopes kitabensis*, contains certain peptides known as oxyopinins that develops perforations in lipid membranes (Corzo *et al.,* 2002; Belokoneva *et al.,* 2003). This property of oxyopinins nominates it as a good candidate for the therapeutic treatment of cancer (Duke *et al.,* 1994; Shaposhnikova *et al.,* 1997). The venom components isolated from *Macrothele raveni* spider (Hexathelidae) affect the human cervical carcinoma cell's (HeLa) invasion and cytotoxicity. At precise doses of 10, 20, and 40 milligram per liter, spider venom considerably reduces cell proliferation and invasion in HeLa cells; furthermore, the same doses significantly increase cytotoxicity leading to apoptosis of treated cell (Gao *et al.,* 2005). The same venom components are shown to be very effective on the carcinoma cell line MCF-7 that are involved in human breast cancer (Gao *et al.,* 2007). The venom at doses of 40, 20, and 10 microgram per milliliter, leads to death of these cells by either necrosis or apoptosis. At low doses it suppresses tumor growth (Heinen & Veiga, 2011). The venom of a West Indies tarantula possesses a potent toxin known as Psalmotoxin 1. This peptide inhibits cation currents that are facilitated by acid-sensing ion channels (ASIC) (Escoubas *et al.,* 2000; Bubien *et al.,* 2004). More specifically, it causes inhibition of sodium ion currents in GBM (glioblastoma multiforme) also known as high-grade astrocytoma cells of human. Also in normal human astrocytes, is fails to obstruct the whole-cell current, indicating the usage of Psalmotoxin 1 in the diagnosis and treatments of malignant gliomas. The venom of the spider *M. raveni* has an inhibitory effect on the proliferation and invasion of human hepatocellular carcinoma cell line BEL-7402. It inhibits the cell proliferation and DNA synthesis of the treated cells resulting in arresting of cancerous cells in the G (0) or G (1) phase of cell cycle. Ultimately leading cancerous cells to apoptosis (Gao *et al.,* 2005).

The hemocytes of *Acanthoscurria gomesiana*, contains a potent peptide named as gomesin. It exhibits a potential of anti-tumor activity (Rodrigues *et al.,* 2013). It induces apoptosis in target cell by potently effecting the growth of cancerous cells along with the blood vessels providing nutrients to cancerous cells. It significantly reduces tumor growth and displays cytotoxic effects on a diverse array of tumor cell lines including breast cancer, melanoma and carcinoma of colon (Heinen & Veiga, 2011).

Whereas the other clinical drugs employed for treatment of cancer i.e., Etoposide (Vp-16) and Paclitaxel (Taxol), destroy normal cells along with the cancerous cells. This shortcoming of the clinical treatment severely reduces the efficacy of these drugs in treating cancer. Therefore, the peptides and other vital anticancerous compounds of spider venom provide a workable solution for the development of anti-cancerous therapies having minimal toxic effects.

Antimalarial Toxins

Malaria has been regarded as one of the most contagious and prevalent mosquito-borne diseases existing in the world. In 2015 an estimated 214 million cases of malaria occurred globally with 438,000 reported deaths mainly including children, less than 5 years of age. The economic cost of malaria is estimated to be 12 billion US$ per year (World Malarial Report, 2015). Most of the cases of malaria are reported from tropical and subtropical areas where the rainfall is sufficient and temperature is suitable for the development and multiplication of both the mosquito and the protozoa (Sachs & Malaney, 2002). The protozoan responsible for causing malaria belongs to genus *Plasmodium*. It spreads by means of female *Anopheles* mosquitoes which act as a vector for the disease. Among the five malaria causing genera of *Plasmodium*, *Plasmodium falciparum* is the most

virulent one (Wellem & Plowe, 2001; Choi *et al.,* 2004). In the last few decades, the malarial infections caused by *P. falciparum* could effectively be averted by the use of antimalarial drug, chloroquine. Particularly, in the 20th century chloroquine proved to be the most successful drug ever deployed against malaria. But its heavy usage during the past few decades ultimately led to widespread resistance in *P. falciparum* against multiple drugs particularly chloroquine (Wellem & Plowe, 2001; Enayati & Hemingway, 2010).

This increased resistance of the malarial parasite to the conventional antimalarial drugs refers to the urgent need for novel chemotherapeutic approaches (Price & Nosten, 2001). Besides the therapies centering small molecules i.e. artemisinins, atovaquone and quinine, the development of new classes of molecules based on proteins is an active field of research (Bastianelli *et al.,* 2011). In this aspect the peptides isolated from spider venom can prove as good candidate for production of antimalarial drugs. The venom of *P. cambridgei,* a Trinidad chevron tarantula contains two ICK (inhibitor cystine knot) peptides namely U1-TRTX-Pc1a (Psalmopeotoxin I) and U2-TRTX-Pc1a (Psalmopeotoxin II) that are shown to be effective against the *P. falciparum*. These ICK peptides cause inhibition of intra-erythrocyte development of *P. falciparum* with ED50 (median effective dose) values of low micro molar range, 1.1–1.6 µM (Saez *et al.,* 2010). Moreover, they also do not exhibit cytotoxic, hemolytic or neurotoxic activities in mammals suggesting them as potential tools for antimalarial drug discovery (Choi *et al.*, 2004). Although these peptides have unknown mode of action, one possibility is that they may be targeting the permeability routes of erythrocyte membrane that are recently generated by parasitic penetration (Staines *et al.,* 2005). Consequently, these ICK peptides may help in finding novel target sites for anti-malarial drug in addition to being valuable therapeutic leads.

Antifungal Potential

Mycotic infections that are developing multidrug resistance, pose a major emerging challenge to the medical field. Fungal infections are frequently becoming a serious threat to the immunocompromised patients suffering from AIDS, cancer, or persons undergoing transplantation (Venkatesan *et al.,* 2005). Over the last 20 years percentage of mortality and morbidity because of invasive mycosis had been increasing day by day (Matejuk *et al.,* 2010). In the past few years, Amphotericin B and azoles have generally been employed as effective fungicidal pharmacological drugs but their irrational use caused many fungal strains to become resistant to them. Moreover, there are considerable side effects associated with its use, including nephrotoxicity. Therefore, In the light of emerging resistance to conventional fungicides, novel antifungal approaches are urgently required. Presently, in pharmacology antifungal peptides are attracting substantial interest. They are universally present in plant and animal kingdoms. Spider venom is also rich in these antifungal peptides. Therefore, the spider toxins are extensively being used in pharmacological studies for the development of prototypes for antifungal drugs.

The venom of a tarantula, *Avicularia juruensis,* (Theraphosidae) contains a compound named juruin. It shows remarkable antimicrobial action when treated against yeast and filamentous fungi, i.e. *Candida albicans*. Juruin is a peptide with 38 amino acids having a molecular weight of 4005.83. Juruin is found to be successful in inhibiting the growth of majority of the fungi and yeast strains, with minimum inhibitory concentration between 2.5–5 micro molar, except for *Aspergilus niger* having MIC values between 5–10 µM (Ayroza *et al.,* 2012). Although, Amphotericin B is effective even when used in 6 times lower concentrations (µM) than the concentration of Juruin. Yet Juruin is much effective against Amphotericin B-resistant fungal strains such as *Candida glabrata* and *C. albicans* (Khan *et al.,* 2008; Krogh-Madsen *et al.,* 2006). Even at the higher concentrations up to 10 µM, no hemolytic activity caused by Juruin is reported. This implies that Juruin does not work by rupturing the cell membranes. Moreover, Juruin has greater specificity to the charged targets having higher values of electronegativity, i.e. cells of prokaryotic origin (Silva *et al.,* 2000), nucleic acids (DNA and RNA) and intracellular proteins (Nguyen *et al.,* 2011). Hence, it can be inferred that in Juruin, the amino acids residues having positive charge are involved in recognition of target receptors. Moreover the selectivity of Juruin against pathogens is also due to presence of specific positive charged amino acids (Ayroza *et al.,* 2012).

The crude venom of *P. bistriata* shows distinct antifungal activity for *C. albicans*. It shows no recorded hemorrhagic activities along with minimal genotoxicity (Gimenez *et al.,* 2014). Moreover, LyeTx I extracted from the venom of *Lycosa erythrognatha* along with its pronounced antibacterial activity is effective against some strains of fungi, i.e. *Candida krusei* and *Cryptococcus neoformans* (Santos *et al.,* 2010). Furthermore, the venom of *Hogna carolinensis*, a wolf spider (Lycosidae) contains two antimicrobial peptide toxins namely lycotoxins I and II. These peptides

are potent growth inhibitors of *Escherichia coli* and *Candida glabrata* (Yan & Adams, 1998). Also, antimicrobial peptides (AMP) are identified in the venom of wolf spider, *Lycosa singoriensis* and named lycocitin 1, 2 and 3. These AMPs retards fungal growth (*C. albicans*) besides inhibiting the growth of gram-positive (*Staphylococcus aureus, Bacillus subtilis*) and gram-negative (*E. coli, Pseudomonas aeruginosa*) bacteria (Budnik *et al.,* 2004).

Along with several potent antifungal peptides that have selectivity for pathogenic fungi, the spider venom is expected to play an important role in the treatment of fungal infections. Although the antimicrobial peptides (AMPs) isolated from the spider venom created the hope for the effective treatment of opportunistic fungal infections. Yet there is great need for addressing the safety concerns of new therapies on biological systems.

Antibacterial Potential

The massive use of antibiotics in the mid-20th century resulted in dramatic decline in the death rate across the globe (Bi *et al.,* 2003). However, prolonged and intensive use of broad-spectrum antibiotics has eventually led to the rise of multi-resistant bacterial types (Wright, 2007). Antibiotic resistance refers to a general term that means the ability of a microbe to endure the lethal antibiotic effects. Particularly, bacteria are the most efficient microbe at adjusting according to their environmental conditions. According to an estimate, bacteria have developed at least one resistance mechanism for all the seventeen classes of antibiotics (Sutti *et al.,* 2015). Antibiotic resistance is emerging as one of the world's most tenacious and alarming concern relating to health (WHO, 2014; Hoffman *et al.,* 2015). These days, almost all the clinically important bacterial pathogens in Pakistan and throughout the world are increasingly becoming resistant to antibiotics (Riaz *et al.,* 2011). Antimicrobial resistance (AMR) is commonly spreading owing to overuse, misuse, and irrational use of antibiotics by doctors, pharmacists and quacks, self-medication by patients and its heavy usage in agriculture (Hussain *et al.,* 2011). On an average 70-80% of antibiotic prescriptions are perhaps advised unreasonably by the medical practitioners (Mansourian *et al.,* 2007). Resistance evolves in bacteria as an outcome of natural resistance in various types of bacteria, genetic mutations in microbes, one species attaining resistance from another and selection pressure from antibiotic use that offers a competitive edge for mutated strains (Raghunath, 2008). Emerging antimicrobial resistance in clinically important bacterial infections (diarrhoeal and respiratory infections, meningitis, sexually transmitted diseases, and hospital-acquired infections) are a matter of great concern these days (Tauxe *et al.,* 1990). Today, there are many bacterial strains that remain uninfluenced by the application of conventional antibiotics i.e*., Staphylococcus epidermidis* and *S. aureus* that are resistant to methicillin, vancomycin-resistant *Enterococci*, multi-resistant *Streptococcus pneumoniae*, *Neisseria gonorrhea, Shigella dysenteriae, Salmonella typhi,* and *Mycobacterium tuberculosis* (Wattal *et al.,* 2010). This decline in efficacy of antibiotics in treating ordinary infections is exponentially accelerating in modern age. It seems that we are standing at the verge of post antibiotic era (Huang *et al.,* 2002; Alanis, 2005).

The solution to this dilemma and to cope with the upcoming challenges related to increased resistances among microbes, the adoption of novel therapeutic approaches and drugs is urgently required. In this aspect, the small polypeptides known as antimicrobial peptides (AMPs) have attracted much attention in recent years as novel antimicrobial agents (Izadpanah & Gallo, 2005; Benli & Yigit, 2008; Ali & Dacheng, 2013). Mainly, AMPs causes structural and functional disruptions in cell membranes even at micromolar concentrations by directly binding to plasma membrane of the target cell (Jenssen *et al.,* 2006). The AMPs causes permeation and lysis of the cell followed by the interaction of microbe's receptor with the peptide (Zasloff, 2002). Also, to some extent it seems unlikely for bacteria to acquire resistance against AMPs (Ding & Ho, 2004).

Moreover, the recent successful introduction of the daptomycin that is lipopeptide antibiotic (Robbel & Marahiel, 2010), recreated interest in antimicrobial peptides (Vooturi & Firestine, 2010). So far, approximately forty MAMPs (membrane-acting antimicrobial peptides) have been identified in the venom of 4 different families of araneomorphs. These MAMPs shows a diverse range of antimicrobial activities, being potently active against bacterial as well as fungal agents (Yan & Adams, 1998). Anti-trypanosomal activity is also reported from some MAMPs (Kuhn-Nentwig *et al.,* 2002). MAMPs are amphipathic peptides having α-helical configuration. MAMPS effectively yield their antimicrobial effects by causing lysis of plasma membranes (Kuhn-Nentwig, 2003; Nomura & Corzo 2006; Pukala *et al.,* 2007; Dubovskii *et al.,* 2008).

Interestingly, several biologically active antimicrobial peptides (AMPs) have been identified from diverse prokaryotic and eukaryotic origins (Kastin, 2006; Kuhn-Nentwig, 2009; Peters *et al.,* 2010). AMPs are also abundantly found in natural

venoms (Vassili *et al.,* 2011). In particular, spider venoms is simultaneously loaded with dozens of various AMPs having distinct structures and diverse mode of actions (Vassilevski *et al.,* 2009). In addition, a wide range of pathogenic organisms such as Gram-positive and Gram-negative bacteria, fungi and even viruses are targeted by these AMPs (Mor & Nicolas, 1994; Zasloff, 2002; Kastin, 2006).

The first description of anti-microbial activity of spider venom was published in 1989 by Xu *et al.* (1989) in the venom of *L. singoriensis*, a wolf spider. The venom of *L. singoriensis* contains antimicrobial peptides (AMP) which are named as lycocitin 1, 2 and 3 (Budnik *et al.,* 2004). Both lycocitin 1 and 2 peptides are shown to inhibit the growth of gram-positive bacteria such as *S. aureus* and *B. subtilis* along with gram-negative bacteria i.e. *E. coli* and *P. aeruginosa* bacteria (Xu *et al.,* 1989; Liu *et al.,* 2009).

In another study conducted by Benli and Yigit (2008) on antibacterial effect of venom of *Agelena labyrinthica* (Agelenidae) showed that out of all 9 bacterial species i.e. *B. subtilis, Enterococcus gallinarium, Enterococcus faecalis, E. coli, Listeria monocytogenes, Shigella* sp., *Streptococcus pyogenes, S. aureus,* and *P. aeruginosa*, the one tenth dilution of venom was effective on five bacterial types (*B. subtilis, E. coli, Shigella* sp., *S. aureus,* and *P. aeruginosa*). The bacterial cells treated by dilution of venom undergo shrinkage and their cell wall displayed depression at multiple sites. This is probably due to the excessive loss of cytoplasm from bacterial cell. Moreover, the venom of another wolf spider *L. carolinensis* is found to have two antibacterial toxins namely, Lycotoxins I and II. Both of these peptidic toxins effectively constrain the growth of Gram-negative bacteria (*E. coli*) as well as yeast *C. glabrata* (Yan & Adams, 1998). Furthermore, the venom of *Lachesana tarabaevi* spider (Zodariidae) possesses peptides known as latarcins. Latarcins are the cytolytic, antibacterial peptides that causes lysis in cells of various organisms including cells of gram-positive and gram-negative bacteria along with fungal cell i.e. yeast (Kozlov *et al.,* 2006). Also, the venom of *A. labyrinthica* is proven to be very effective against *S. aureus* which is major cause of infections acquired in hospitals and is resistant to many antibiotics like methicillin (Vizioli & Salzet, 2002).

In addition, Haeberlia *et al.* (2000) isolated 5 antimicrobial peptides from the venom of a neotropical wandering spider (Ctenidae), *Cupiennius salei*. In a study conducted by Kuhn-Nentwig *et al.* (1998), the amphipathic structure of peptides isolated from the venom of same spider, *C. salei* were demonstrated to have marked antibacterial activity. These peptides were tested against five different bacterial species including Gram positive bacteria i.e. *S. epidermidis* and *B. subtilis* and gram negative bacteria i.e. *E. coli, Pseudomonas putida and Paracoccus denitrificans*. All the 5 different bacterial strains were found to be prone to these antibacterial peptides with MICs ranging between 0.18 to 18 mM. These antibacterial peptides inhibit growth of bacteria by causing lysis of the bacterial cells.

The venom of *L. erythrognatha* (wolf spider) contains an antimicrobial peptide known as LyeTx I. It shows marked inhibitory activity against Gram-positive and Gram-negative bacteria including *S. aureus* and the *E. coli* respectively. Its permeability decreases about five times in plasma membranes having sterols associated with POPC (1-palmitoyl-2-oleoyl-sn-glycero-3-phosphocholine) that contains cholesterol (vertebrates) but remains unchanged in membrane containing ergosterol (bacteria, fungi and other protozoans). Also, LyeTx I shows quite low haemolytic activity, implying that the preferential targets of LyeTx I are membranes of microbes rather than vertebrate membranes. This renders LyeTx I peptide as a good candidate for further antibacterial drug development. Moreover, LyeTx I is anticipated to be very useful in the treatment of skin and mucosal infections of bacteria (Chen *et al.,* 2005).

The venom of tarantula, *Vitalius dubius*, contains an acylpolyamine called VdTX-I. The VdTX-I toxin has a significant antimicrobial activity (fungi, yeast and bacteria), with broad spectrum, and is experimentally inert to mammalian blood cells. This toxin also acts relatively fast against the tested bacteria (Sutti *et al.,* 2015). Moreover, some antimicrobial compounds are also identified in the venom of tarantula spider, *Haplopelma hainanum* (Zhao *et al.,* 2011; Kuhn-Nentwig *et al.,* 2013). Besides the usual neurotoxins and cytotoxins, venom of the lynx spider *Oxyopes takobius* contains modular toxins having 2 domains. These modular toxins are named as spiderines: OtTx1a, OtTx1b, OtTx2a and OtTx2b. These modular toxins possess potent antimicrobial potential along with enormous insecticidal activity. OtTx1a-AMP is a bactericidal toxin that restrains bacterial growth at MICs value in the range of 0.1-10 µM (Vassilevski *et al.,* 2013). Furthermore, the venom of the *O. kitabensis* contains amphipathic peptides known as oxyopinins. Oxyopinins are shown to have marked antimicrobial activity along with hemolytic and insecticidal potential (Corzo *et al.,* 2002).

Therefore, by keeping in sight the above discussion we can conclude that the biologically

active components of spider venom can be used as lead therapeutic agents having targeted action and significantly reduced side effects.

Bioinsecticidal Potential

More than 10,000 species of arthropods are believed to be pest organisms worldwide (Windley *et al.*, 2012). It is estimated that arthropod pests contributes nearly 14% to the destruction of the world's annual crop and 20% damage to the stored food grains (Oerke & Dehne, 2004; Peshin *et al.,* 2007). This results in approximately 100 billion US$ loss per year (Carlini & Grossi-de-Sá, 2002). Also many of arthropod pests especially mosquitoes (Gratz, 1999), midges and flies (Gubler, 2002; Hall & Gerhardt, 2009) transmit the contagious diseases by acting as a vehicle for the pathogens (Nauen, 2007). At present, chemical insecticides are chiefly used to control arthropod pest populations. The use of chemical insecticides was first established in the 1940s and it remained the primary method for controlling pest organisms. At that time, DDT (Organochlorine) emerged as an eminent synthetic organic pesticide. Its use quickly spread over the globe as it was inexpensive and was very effective at killing pests. But in 1960s, bioaccumulatory and biomagnifying effects of DDT arose concern over its use. The chemical industry responded to this concern with new classes of pesticides, which are less persistent and causes acute toxicity. For that reason organophosphates and carbamates were introduced in the 1960s as an effective tool against pest populations (Casida & Quistad, 1998). Afterwards the new generation of chemical insecticides including pyrethroids, neonicotinoids, ryanoids, formamidines, dinitrophenols, pyridazinones and quinazolines were extensively used for controlling pest populations. This massive use of chemical insecticides in agriculture and for controlling vectors of infectious diseases, offered an effective, rapid and comparatively cheap solution for resolving the problems related to pest. However, major problems with the use of agrochemicals rapidly sprung up including (i) hazard to human health due to non-target specificity of chemical pesticides, (ii) harmful environmental and ecological impacts (iii) insecticidal resistance owing to nonexistence of diversity in bioactivity of these chemicals (Windley *et al.*, 2012). Owing to these detrimental impacts on environment and human health, 169 pesticides were de-registered from Jan 2005 to Dec 2009, with barely nine new pesticides being registered within the same phase (Dale, 2012).

These problems signify the requirement to pinpoint new and safer insecticidal agents. Therefore, it is pivotal to identify novel insecticides that are target specific and eco-friendly. Currently researchers are being focused on bio-insecticides as an effective and safer alternative to the chemical insecticides. They employ organisms or their derivative products i.e., recombinant baculoviruses, toxin-fusion proteins, transgenic plants, and peptidomimetics for controlling pest populations (Windley *et al.*, 2012). The promising sources of bio-insecticides include microbial agents (viruses, bacteria, fungi), insect eating (entomophagous) round worms, plant-derived compounds, pheromones and resistance genes against insects incorporated in crops (Copping & Menn, 2000). Particularly, insecticidal toxins extracted from insect predators i.e., peptide neurotoxins extracted from the venom of scorpions (Froy *et al.,* 2000), parasitic wasps (Quistad & Skinner, 1994), straw itch mite (Tomalski *et al.,* 1988), and spiders (King, 2007; Nicholson, 2007), is of rising interest in the production of bio-insecticides. At present, spider venom is receiving a great deal of attention as it comprises of a huge and diverse array of potent neurotoxic peptides (Windley *et al.*, 2012). Their venom comprises of rich source of hyper stable mini-proteins that are selectively insecticidal (Herzig *et al.,* 2011). These mini protein induce lethality and paralysis in insects by modulating ion channels, receptors or enzymes. Among the 800 characterized bioactive spider-venom peptides, 136 peptides are considered to have insecticidal potential. It is also indicated that out of these 136 peptides, the potent insect-selectivity is exhibited by thirty eight peptides, thirty four are shown to be non-selective while sixty four have unspecified selectivity (Windley *et al.*, 2012). The most familiar recognized insecticidal targets of spider-venom are voltage gated sodium and calcium channels, calcium-activated potassium channels, receptors of N-methyl-D-aspartate (NMDA), lipid bilayer, and presynaptic nerve terminals (Vetter *et al.,* 2011). The spiders in most families of araneomorph and mygalomorph produce toxins that target NaV channels. Spider toxins alter the neuronal excitability that results in paralysis and ultimately death of insects (Catterall *et al.,* 2007). Peptidic components of spider venom have high affinity and specificity for the neurotoxin receptor sites on NaV channels of insects. Therefore, they have potential to be utilized as bio-insecticides.

ω-Hexatoxin-Hv1a extracted from the venom of Australian funnel-web spiders has high selectivity for the insect's CaV channels (Fletcher *et al.*, 1997; Wang *et al.,*1999; Chong *et al.,* 2007). Therefore, it can simultaneously block all of the subtypes of insect's high voltage activated (HVA)

CaV channel. Furthermore, the calcium currents in the nervous system of rats are not affected by it (Atkinson *et al.,* 1996). Hence, it is shown to produce no toxic effects in vertebrates even when used in 10,000 times greater concentrations (Khan *et al.,* 2006). Voltage-activated potassium channels are concerned with the excitability of neurons, contraction of smooth muscles, regulation of heart rate and volume of cell, neurotransmitter release and processes such as cell signaling (Wei *et al.,* 2005). Insect potassium channels are selectively targeted by κ-Hexatoxin-1 toxin family, extracted from venom of Australian funnel-web spiders (Gunning *et al.,* 2008). Also several cytolytic peptides having antimicrobial potential have been successfully isolated from the venom of araneomorph spider. These peptides are categorized as MAMPs (membrane-acting antimicrobial peptides). Short MAMPS show high activity against Gram positive and negative bacteria whereas long, linear M-ZDTX-Lt toxins (cytoinsectotoxins) are more effective for insecticidal activity (Vassilevski *et al.,* 2008). Furthermore, the venom of widow spider of the genus *Latrodectus* (Theridiidae) contains 5 insect-specific peptides, recognized as latroinsectotoxins (LIT) α, β, γ, δ and ε. These latroinsectotoxins have phylum-selective insecticidal activity (Grishin, 1998; Graudins *et al.,* 2011). Also two of these latroinsectoxins α-LIT-Lt1a (Molecular mass 111 kDa) (Kiyatkin *et al.,* 1993) and δ-LIT-Lt1a (molecular mass 130 kDa) (Dulubova *et al.,* 1996) have been completely sequenced and cloned. It is believed that these high-molecular mass proteins induce paralysis in insects by promoting enormous exocytosis of neurotransmitter from the nerve terminals. Two insect ionotropic anion gates have been discovered in insect CNS neurons named as GluCls (Raymond *et al.,* 2000). The L-glutamate receptors that gate chloride channels (GluCls) in insects CNS neurons are prominent target sites for the action of insecticides. Acylpolyamines are significant low molecular weight toxins in the venoms of some spiders. Up till now more than 100 acylpolyamines have been characterized in spider venom (Atta-ur-rehman, 2012). It is thought that their primary purpose in venom is to paralyze insects by blocking glutamate receptors.

Therefore, we can conclude from above literature that variety of toxins and peptides isolated from spider-venom are efficient bio-insecticides. They can develop a combination of desired features of high potency, phyletic selectivity, specific target action, and stable structure. Furthermore, pharmacological characterization of spider venom also revealed novel and unique target sites in insects. These sites were not exploited by traditional agrochemicals in past. Thereby discovering novel insecticide targets for further screening programs in future. These attributes when combined with the future prospects recommend spider venom as lead compound for the generation of bio-insecticides.

REFERENCES

Adams, M. E., 2004. Agatoxins. *Toxicon*, **43**: 509-525.

Alanis, A. J., 2005. Resistance to antibiotics: Are we in the post-antibiotic era. *Arch Med. Res*., **36**: 697–705.

Ali, A. B. & Dacheng, R., 2013. Antimicrobial Peptides. *Pharmaceuticals (Basel)*, **6**(**12**): 1543–1575.

Almeida, A. P., Andradea, A. B., Ferreirab, A. J., Piresc, A. C. G., Damascenoa, D. D., Alvesa, M. N. M., Gomesa, E. R. M., Kushmericka, C., Limaa, R. F., Pradod, M. A. M., Pradod, V. F., Richardsone, M., Cordeiroe, M. N., Guatimosima, S. & Gomezf, M. V., 2011. Antiarrhythmogenic effects of a neurotoxin from the spider *Phoneutria nigriventer*. *Toxicon,* **57**(**2**): 217–224.

Andrew, E. & Epstein, M. D., 2015. The Wearable Cardioverter-Defibrillator in Newly Diagnosed Cardiomyopathy: Treatment on the Basis of Perceived Risk. *J. Am. Coll. Cardiol*., **66**(**23**): 2614–2617.

Atkinson, R., Vonarx, E. & Howden, M., 1996. Effects of whole venom and venom fractions from several Australian spiders, including Atrax (Hadronyche) species, when injected into insects. *Comp. Biochem. Physiol*., **114**: 113–117.

Atta-ur-rehman., 2012. *Studies in natural product chemistry*. 1st edition, Elsevier Science Publishers, Amsterdam, Netherlands. 36 (series number**)**. pp: 28.

Ayroza, G., Ferreira, I. L. C., Sayegh, R. S. R., Tashima, A. K. & Silva, P. I., 2012. Juruin: an antifungal peptide from the venom of the Amazonian Pink Toe spider, *Avicularia juruensis*, which contains the inhibitory cystine knot motif. *Front Microbiol*., **3**: 324.

Bastianelli, G., Bouillon, A., Nguyen, C., Crublet, E., Pêtres, S., Gorgette, O., Le-Nguyen, D., Barale, J. C. & Nilges, M., 2011. Computational reverse-engineering of a spider-venom derived peptide active against

Plasmodium falciparum SUB1. *PLoS One*., **6**(**7**): e21812.

Beleboni, R. O., Guizzo, R., Fontana, K., Pizzo, A. B., Oliveira, R. & Carolino, G., 2006. Neurochemical characterization of a neuroprotective compound from *Parawixia bistriata* spider venom that inhibits synaptosomal uptake of GABA and glycine. *Mol. Pharmacol*., **69**(**6**): 1998–2006.

Belokoneva, O. S., Villegas, E., Corzo, G., Dai, L. & Nakajima, T., 2003. The hemolytic activity of six arachnid cationic peptides is affected by the phosphatidylcholine to-sphingomyelin ratio in lipid bilayers. *Biochim. Biophys. Acta*., **1617**: 22–30.

Benli, M. & Yigit, N., 2008. Antibacterial activity of venom from funnel web spider *Agelena labyrinthica* (Araneae Agelenidae). *J. Venom. Anim. Toxin Incl. Trop. Dis*., **14**: 641-650.

Bi, P., Whitby, M., Walker, S. & Parton, K.A., 2003. Trends in mortality rates for infectious and parasitic diseases in Australia: 1907–1997. *Intern. Med. J*., **33**: 152–162.

Bialer, M., 2012. Chemical properties of antiepileptic drugs (AEDs). *Adv. Drug Deliv. Rev*., **64**(**10**): 887–895.

Bourinet, E., Soong, T. W. & Sutton, K., 1999. Splicing of α1A subunit gene generates phenotypic variants of P- and Q-type calcium channels. *Nat Neurosci*., **2**: 407–415.

Bowman, C. L., Gottlieb, P. A., Suchyna, T. M., Murphy, Y. K. & Sachs, F., 2007. Mechanosensitive ion channels and the peptide inhibitor GsMTx-4: history, properties, mechanisms and pharmacology. *Toxicon*, **49**: 249–270.

Brennan, F., Carr, D. B. & Cousins, M., 2007. Pain management: A fundamental human right. *Anesth. Analg*., **105**: 205–221.

Bubien, J. K., Ji, H. L., Gillespie, G.Y., Fuller, C.M., Markert, J.M. & Mapstone, T. B., 2004. Cation selectivity and inhibition of malignant glioma Na+ channels by Psalmotoxin 1. *Am. J. Physiol. Cell Physiol*., **287**: 1282–1291.

Budnik, B. A., Olsen, J. V., Egorov, T. A., Anisimova, V. E., Galkina, T. G., Musolyamov, A. K., Grishin, E. V. & Zubarev, R. A., 2004, De novo sequencing of antimicrobial peptides isolated from the venom glands of the wolf spider *Lycosa singoriensis. J. Mass. Spectrom*., **39**: 193–201.

Cairrão, M., Ribeiro, A. M., Pizzo, A. B., Beleboni, R. O., Miranda, A. & Santos, W. F., 2002. Anticonvulsant and GABA uptake inhibition properties of venom fractions from the spiders *Parawixia bistriata* and *Scaptocosa raptoria*. *Pharm. Biol*., **40**(**6**): 472–729.

Cardoso, F. C., Pacifico, L. G., Carvalho, D. C., Victoria, J. M., Neves, A. L., Chavez-Olortegui, C., Gomez, M. V. & Kalapothakis, E., 2003. Molecular cloning and characterization of *Phoneutria nigriventer* toxins active on calcium channels. *Toxicon*, **41**: 755–763.

Carlini, C. R. & Grossi-de-Sá, M. F., 2002. Plant toxic proteins with insecticidal properties. A review on their potentialities as bioinsecticides. *Toxicon*, **40**: 1515–1539.

Casida, J. E. & Quistad, G. B., 1998. Golden age of insecticide research: past, present, or future? *Annu. Rev. Entomol*., **43**: 1-16.

Catterall, W. A., Cestèle, S., Yarov-Yarovoy, V., Yu, F. H., Konoki, K. & Scheuer, T., 2007. Voltage-gated ion channels and gating modifier toxins. *Toxicon*., **49**(**2**): 124-141.

Chen, Y., Mant, C. T., Farmer, S. W., Hancock, R. E., Vasil, M. L. & Hodges, R. S., 2005. Rational design of a-helical antimicrobial peptides with enhanced activities and specificity/therapeutic index. *J. Biol. Chem*., **280**: 12316–12329.

Choi, S., Parent, R., Guillaume, C., Deregnaucourt, C., Delarbre, C., Ojcius, D. M., Montagne, J. J., Célérier, M. L., Phelipot, A., Amiche, M., Molgo, J., Camadro, J. M. & Guette, C., 2004. Isolation and characterization of Psalmopeotoxin I and II: two novel antimalarial peptides from the venom of the tarantula *Psalmopoeus cambridgei*. *FEBS Lett*., **527**: 109–117.

Chong, Y., Hayes, J. L., Sollod, B., Wen, S., Wilson, D. T., Hains, P. G., Hodgson, W. C., Broady, K. W., King, G. F. & Nicholson, G. M., 2007. The ω-atracotoxins: selective blockers of insect M-LVA and HVA calcium channels. *Biochem. Pharmacol*., **74**: 623–638.

Cizkova, D., Marsala, J., Lukacova, N., Marsala, M., Jergova, S., Orendacova, J. & Yaksh, T. L., 2002. Localization of N-type Ca2+ channels in the rat spinal cord following chronic constrictive nerve injury. *Exp. Brain Res*., **147**: 456–463.

Clare, J. J., 2010. Targeting voltage-gated sodium channels for pain therapy. *Expert Opin. Invest. Drugs*., **19**: 45–62.

Cohen, C. J., Ertel, E. A. & Smith, M. M., 1992. High affinity block of myocardial L-type calcium channel gating currents with spider toxin ω-Aga-IIIA: advantages over 1,4-dihydropyridines. *Mol. Pharmacol*., **42**: 947–951.

Copping, L. & Menn, J., 2000. Bio pesticides: a review of their action, applications and efficacy. *Pest Manag. Sci*., **56**: 651–676.

Cordeiro, N., de Figueiredo, S. G., Valentim, C., Diniz, C. R., von Eickstedt, V.R., Gilroy, J. & Richardson, M., 1993. Purification and amino acid sequences of six tx3 type neurotoxins from the venom of the brazilian 'armed' spider *Phoneutria nigriventer*. *Toxicon*, **31**: 35–42.

Corzo, G., Villegas, E., Gomez-Lagunas, F., Possani, L.D., Belokoneva, O.S. & Nakajima, T., 2002. Oxyopinins, large amphipathic peptides isolated from the venom of the wolf spider *Oxyopes kitabensis* with cytolytic properties and positive insecticidal cooperativity with spider neurotoxins. *J. Biol. Chem*., **277**: 23627–23637.

Dale, K., 2012. Personal communication. U.S. Environmental Protection Agency, Washington, DC, USA.

Dalmolin, G. D., Silva, C. R., Rigo, F. K., Gomes, G. M., Cordeiro, M. N., Richardson, M., Silva, M. A., Prado, M. A., Gomez, M. V. & Ferreira, J., 2011. Antinociceptive effect of brazilian armed spider venom toxin tx3–3in animal models of neuropathic pain. *Pain*, **152**: 2224–2232.

Darrell, J. G. & Patrick, R., 2012. The Economic Costs of Pain in the United States. *J. Pain*., **13**(**8**): 71-724.

De Souza, A. H., Lima, M. C., Drewes, C. C., da Silva, J. F., Torres, K. C., Pereira, E. M., de Castro Junior, C. J., Vieira, L. B., Cordeiro, M. N. & Richardson M., 2011. Antiallodynic effect and side effects of pα1β, a neurotoxin from the spider *Phoneutria nigriventer*: Comparison with ω-conotoxin mviia and morphine. *Toxicon*, **58**: 626–633.

Ding, J. L. & Ho, B., 2004. Antimicrobial peptides: Resistant-proof antibiotics of the new millennium. *Drug Dev. Res*., **62**: 317–335.

Dobson, R., 2001. Spider venom may prevent atrial fibrillation. *West J. Med*., **174**(**3**): 164.

Dubovskii, P., Volynsky, P. E., Polyansky, A. A., Karpunin, D. V., Chupin, V. V., Efremov, R. G. & Arseniev, A. S., 2008. Three-dimensional structure/hydrophobicity of latarcins specifies their mode of membrane activity. *Biochemistry*, **47**: 3525–3233.

Duke, R. C., Witter, R. Z., Nash, P. B., Young, J. D. E. & Ojcius, D. M., 1994. Cytolysis mediated by ionophores and pore-forming agents: role of intracellular calcium in apoptosis. *FASEB J*., **8**: 237–246.

Dulubova, I. E., Krasnoperov, V., G., Khvotchev, M. V., Pluzhnikov, K. A., Volkova, T. M., Grishin, E. V., Vais, H., Bell, D. R. & Usherwood, P. N., 1996. Cloning and structure of δ-latroinsectotoxin, a novel insect-specific member of the latrotoxin family: functional expression requires C-terminal truncation. *J. Biol. Chem*., **271**: 7535–7543.

Durant, C., Christmas, D. & Nutt, D., 2010. The pharmacology of anxiety. In: Stein MB, Steckler T, (eds). Current Topics in Behavioral Neurosciences. 2nd Edition. Germany: Springer Berlin Heidelberg. pp: 303–330.

Enayati, A. & Hemingway, J., 2010. Malaria management: past, present, and future. *Annu. Rev. Entomol*., **55**: 569–591.

Escoubas, P., De Weille, J. R., Lecoq, A., Diochot, S., Waldmann, R., Champigny, G., Moinier, D., Menez, A. & Lazdunski, M., 2000. Isolation of a tarantula toxin specific for a class of proton-gated Na+ channels. *J. Biol. Chem*., **275**: 25116–25121.

Estrada, G., Villegas, E. & Corzo, G., 2007. Spider venoms: a rich source of acylpolyamines and peptides as new leads for CNS drugs. *Nat. Prod. Rep*., **24**(**1**): 145–161.

Fletcher, J. I., Smith, R., O'Donoghue, S. I., Nilges, M., Connor, M., Howden, M. E., Christie, M. J. & King, G. F., 1997. The structure of a novel insecticidal neurotoxin, ω-atracotoxin-HV1, from the venom of an Australian funnel web spider. *Nat. Struct. Biol*., **4**: 559–566.

Froy, O., Zilberberg, N., Chejanovsky, N., Anglister, J., Loret, E., Shaanan, B., Gordon, D. & Gurevitz, M., 2000. Scorpion neurotoxins: structure/function relationships and application in agriculture. *Pest Manag. Sci*., **56**: 472–474.

Gao, L., Yu, S., Wu, Y. & Shan, B., 2007. Effect of spider venom on cell apoptosis and necrosis rates in MCF-7 cells. *DNA Cell Biol*., **26**: 485–489.

Gao, T., Furnari, F. & Newton, A.C., 2005. PHLPP: a phosphatase that directly dephosphorylates Akt, promotes apoptosis,

and suppresses tumor growth. *Mol. Cell.*, **18**(**1**): 13–24.

Gelfuso, E. A., Rosa, D. S., Fachin, A. L., Mortari, M. R., Cunha, A. O. S. & Beleboni, R. O., 2014. Anxiety: a systematic review of neurobiology, traditional pharmaceuticals and novel alternatives from medicinal plants. *CNS Neurol Disord Drug Targets*., **543**: 150–165.

Gilchrist, J., Das, S., Petegem, F. V. & Bosmans, F., 2013. Crystallographic insights into sodium-channel modulation by the β4 subunit. *Proc. Natl. Acad. Sci*., **110**: 5016– 5024.

Gimenez, G. S., Coutinho-Neto, A. & Kayano, A. M., 2014. Biochemical and Functional Characterization of *Parawixia bistriata* Spider Venom with Potential Proteolytic and Larvicidal Activities. *Bio. Med. Res. International*, **2014:** 1-13.

Girish, K. S. & Kemparaju, K., 2007. The magic glue hyaluronan and its eraser hyaluronidase: a biological overview. *Life Sci*., **80**(**21**): 1921-1943.

Gomes, G. M., Dalmolin, G. D., Cordeiro, M. N., Gomez, M. V., Ferreira, J. & Rubin, M. A., 2013. The selective A-type K + current blocker Tx3-1 isolated from the *Phoneutria nigriventer* venom enhances memory of naïve and Aβ 25–35 -treated mice. *Toxicon*, **76**: 23–27.

Gratz, N. G., 1999. Emerging and resurging vector-borne diseases. *Annu. Rev. Entomol*., **44**: 51–75.

Graudins, A., Little, M. J., Pineda, S. S., Hains, P. G., King, G. F., Broady, K. W. & Nicholson, G. M., 2011. Cloning and activity of a novel α-latrotoxin from red-back spider venom. *Biochem. Pharmacol*., **83**: 170–183.

Green, A. C., Nakanishi, K. & Usherwood, P. N. R., 1996. Polyamine amides are neuroprotective in cerebellar granule cell cultures challenged with excitatory amino acids. *Brain Res*., **717**(**1**): 135–46.

Grishin, E. V., Savchenko, G. A., Vassilevski, A. A., Korolkova, Y. V., Boychuk, Y. A., Viatchenko-Karpinski, V. Y., Nadezhdin, K. D., Arseniev, A. S., Pluzhnikov, K. A., Kulyk, V. B., Voitenko, N. V. & Krishtal, O. O., 2010, Novel peptide from spider venom inhibits P2X3 receptors and inflammatory pain. *Ann. Neurol*., **67**: 680–683.

Grishin, E.V., 1998. Black widow spider toxins: the present and the future. *Toxicon*, **36**: 1693–1701.

Grishin, E.V., 1999. Polypeptide neurotoxins from spider venoms. *Eur. J. Biochem*., **264**(**2**): 276–80.

Gründer, S. & Chen, X., 2010. Structure, function, and pharmacology of acid-sensing ion channels (ASICs): Focus on ASIC1a. *Int. J. Physiol. Pathophysiol. Pharmacol*., **2**: 73–94.

Gubler, D. J., 2002. The global emergence/resurgence of arboviral diseases as public health problems. *Arch. Med. Res*., **33**: 330–342.

Gunning, P., O'Neill, G. & Hardeman, E., 2008. Tropomyosin-based regulation of the actin cytoskeleton in time and space. *Physiol. Rev*., **88**(**1**): 1-35.

Haeberlia, S., Kuhn-Nentwig, L., Schaller, J. & Nentwig, W., 2000, Characterization of antibacterial activity of peptides isolated from the venom of the spider *Cupiennius salei* (Araneae: Ctenidae). *Toxicon*, **38**: 373-380.

Hall, R. D. & Gerhardt, R. R., 2009. Flies (Diptera). In: Medical and Veterinary Entomology, 2nd Edition.; Mullen, G.R., Durden, L.A., (Eds).; Elsevier: Burlington, N J, USA. pp: 127–161.

Heinen, T. E & Veiga, A. B. G., 2011. A Review: Arthropod venoms and cancer. *Toxicon*, **57**(**4**) :497–511.

Herzig, V., Wood, D. L. A., Newell, F., Chaumeil, P. A., Kaas, Q., Binford, G. J., Nicholson, G. M., Gorse, D. & King, G. F., 2011 ArachnoServer 2.0, an updated online resource for spider toxin sequences and structures. *Nucleic Acids Res*., **39**: 653–657.

Hoffman, S. J., Outterson, K., Rottingen, J. A., Cars, O., Clift, C., Rizvi, Z., Rotberg, F., Tomson, G. & Zorzet, A., 2015. "An international legal framework to address antimicrobial resistance". *Bull World Health Organ*., **93**(**2**) : 66.

Huang, G. T., Zhang, H. B., Kim, D., Liu, L. & Ganz, T., 2002. A model for antimicrobial gene therapy: demonstration of human beta-defensin 2 antimicrobial activities in vivo. *Hum. Gene Ther*., **13**: 2017–2025.

Huang, Y. & Mucke, L., 2012. Alzheimer mechanisms and therapeutic strategies. *Cell*, **148**(**6**): 1204–22.

Hussain, S., Malik, F., Hameed, A., Parveen, G., Raja, F. Y. & Riaz, H., 2011. Pharmacoepidemiological studies of prescribing practices of health care providers of Pakistan: A cross-sectional

survey. *Afr. J. Pharm. Pharacol*., **5**: 1484–1493.

Izadpanah, A. & Gallo, R. L., 2005. Antimicrobial peptides. *J. Am. Acad. Dermatol*., **52**: 381-390.

Jenssen, H., Hamill, P. & Hancock, R. E. W., 2006. Peptide antimicrobial agents. *Clin. Microbiol. Rev*., **19**: 491–511.

Kastin, A., 2006. Handbook of Biologically Active Peptides. Academic Press; San Diego, USA.

Kawai, N., Niwa, A. & Abe, T., 1982. Spider venom contains specific receptor blocker of glutaminergic synapses. *Brain Res*., **247**(**1**): 169–171.

Khakh, B. S. & North, R. A., 2006. P2X receptors as cell-surface ATP sensors in health and disease. *Nature*, **442**: 527–532.

Khan, S. A., Zafar, Y., Briddon, R. W., Malik, K. A. & Mukhtar, Z., 2006. Spider venom toxin protects plants from insect attack. *Transgenic Res*., **15**: 349–357.

Khan, Z. U., Ahmad, S., Al-Obaid, I., Al-Sweih, N. A., Joseph, L. & Farhat, D., 2008. Emergence of resistance to amphotericin B and triazoles in *Candida glabrata* vaginal isolates in a case of recurrent vaginitis. *J. Chemother*, **4**: 488–491.

King, G. F., 2007. Modulation of insect Ca V channels by peptidic spider toxins. *Toxicon*, **49**: 513–530.

Kiyatkin, N. I., Dulubova, I. E. & Grishin, E. V., 1993. Cloning and structural analysis of α-latroinsectotoxin cDNA. Abundance of ankyrin-like repeats. *Eur. J. Biochem*., **213**: 121–127.

Klint, J. K., Sebastian, S., Rupasinghe, D. B., Er, S. Y., Herzig, V., Nicholson, G.M. & King, G. F., 2012. Spider-venom peptides that target voltage-gated sodium channels: Pharmacological tools and potential therapeutic leads. *Toxicon*, **60**(**4**): 478–491.

Kozlov, S. A., Vassilevski, A. A., Feofanov, A. V., Surovoy, A. Y., Karpunin, D. V. & Grishin, E. V.,, 2006. Latarcins, antimicrobial and cytolytic peptides from the venom of the spider *Lachesana tarabaevi* (Zodariidae) that exemplify biomolecular diversity. *J. Biol. Chem*. **281**: 20983–20992.

Krafte, D. S. & Bannon, A. W., 2008. Sodium channels and nociception: Recent concepts and therapeutic opportunities. *Current Opin. Pharmacol*., **8**: 50–56.

Krogh-Madsen, M., Arendrup, M. C., Heslet, L. & Knudsen, J. D., 2006. Amphotericin B and caspofungin resistance in *Candida glabrata* isolates recovered from a critically ill patient. *Clin. Infect. Dis*., **7**: 938–944.

Kuhn-Nentwig, L., 2003. Antimicrobial and cytolytic peptides of venomous arthropods. *Cell. Mol. Life Sci*., **60**: 2651–2668.

Kuhn-Nentwig, L., 2009. Cytolytic and antimicrobial peptides in the venom of scorpions and spiders. In: De Lima, M.E., Pimenta, A.M.C., Martin-Eauclaire, M.F., Zingali, R. &Rochat, H (eds). Animal toxins: state of the art. Perspectives in health and biotechnology, 1st Edition. Editora UFMG, Belo Horizonte. pp: 153–172.

Kuhn-Nentwig, L., Bücheler, A., Studer, A. &Nentwig, W., 1998. Taurine and histamine: low molecular compounds in prey hemolymph increase the killing power of spider venom. *Naturwissenschaften*, **85**: 136-138.

Kuhn-Nentwig, L., Muller. J., Schaller, J., Walz, A., Dathe, M, & Nentwig, W., 2002. Cupiennin 1, a new family of highly basic antimicrobial peptides in the venom of the spider *Cupiennius salei* (Ctenidae). *J. Biol. Chem*., **277**: 11208–11216.

Kuhn-Nentwig, T., Sheynis, S., Kolusheva, W. & Nentwig, R., 2013. Jeline N-terminal aromatic residues closely impact the cytolytic activity of Cupiennin1a, a major spider venom peptide. *Toxicon*, **75**: 177–186.

Leao, R. M., Cruz, J. S., Diniz, C. R., Cordeiro, M. N. & Beirao, P. S., 2000. Inhibition of neuronal high-voltage activated calcium channels by the omega-phoneutria nigriventer tx3–3peptide toxin. *Neuropharmacology*, **39**: 1756–1767.

Leoni, M. J., Chen, X., Mueller, A. L., Cheney, J., McIntosh, T. K. & Smith, D. H., 2000. NPS 1506 Attenuates cognitive dysfunction and hippocampal neuron death following brain trauma in the rat. *Exp. Neurol*., **166**(**2**): 442–449.

Liberato, J. L., Cunha, A. O. S., Mortari, M. R., Gelfuso, E. A., Beleboni, R. O. & Coutinho-Netto, J., 2006. Anticonvulsant and anxiolytic activity of FrPbAII, a novel GABA uptake inhibitor isolated from the venom of the social spider *Parawixia bistriata* (Araneidae: Araneae). *Brain Res*., **1124**(**1**): 19–27.

Li-Smerin, Y. & Swartz, K. J., 1998. Gating modifier toxins reveal a conserved structural motif in voltage-gated Ca2+ and K+ channels. *Proc. Natl. Acad. Sci. USA*, **95**: 8585–8589.

Liu, Z. H., Qian, W., Li, J., Zhang, Y. & Liang, S., 2009. Biochemical and pharmacological study of venom of the wolf spider *Lycosa singoriensis. J. Venom Anim. Toxins incl. Trop. Dis*., **15**: 79–92.

Maggio, F., Sollod, B. L., Tedford, H. W., Herzig, V. & King, G. F., 2010. Spider toxins and their potential for insect control. In: Gilbert L.I., Gill S.S., (eds). Insect Pharmacology: Channels, Receptors, Toxins and Enzymes. Academic Press; London, UK. pp. 101–123.

Mancuso, C. & Gaetani, S., 2014. Preclinical and clinical issues in Alzheimer's disease drug research and development. *Front Pharmacol*., **5**: 234.

Mansourian, A., Saifi, A., Vakili, M., Marjani, A., Ghaemi, E. & Moradi, A., 2007. Prescribing Antibiotics by General and Specialist Physicians: A Pharmacist Administrated Survey. *J. Med. Sci*., **7**: 427–431.

Martinac, B. & Kloda, A., 2003. Evolutionary origins of mechanosensitive ion channels. *Prog. Biophys. Mol. Biol*., **82**: 11–24.

Matejuk, A., Leng, Q., Begum, M. D., Woodle, M. C., Scaria, P., Chou, S. T. & Mixson, A. J., 2010. Peptide-based Antifungal Therapies against Emerging Infections. *Drugs Future*, **35**(**3**): 197.

Matsushita, O. & Okabe, A., 2001. Clostridial hydrolytic enzymes degrading extracellular components. *Toxicon*, **39**(**11**): 1769-1780.

McDavid, S. & Currie, K. P. M., 2006. G-Proteins Modulate Cumulative Inactivation of N-Type (Ca V 2.2) Calcium Channels. *J. Neurosci*., **26**(**51**): 13373–13383.

McDonough, S. I., Boland, L. M., Mintz, I. M. & Bean, B. P., 2002. Interactions among toxins that inhibit N-type and P-type calcium channels. *J. Gen. Physiol*., **119**: 313–328.

McDonough, S. I., Lampe, R. A. & Keith, R. A., 1997. Voltage-dependent inhibition of N- and P-type calcium channels by the peptide toxin ω-grammotoxin-SIA. *Mol. Pharmacol*., **52**: 1095–1104.

Mehndiratta, P., Smith, S. C. & Worrall, B. B., 2015. Etiologic stroke subtypes: updated definition and efficient workup strategies. *Curr. Treat. Options Cardiovasc. Med*., **17**(**1**): 357.

Middleton, R. E., Warren, V. A., Kraus, R. L., Hwang, J. C., Liu, C. J., Dai, G., Brochu, R. M., Kohler, M. G., Gao, Y. D., Garsky, V. M., Bogusky, M. J., Mehl, J. T., Cohen, C. J. & Smith M. M., 2002. Two tarantula peptides inhibit activation of multiple sodium channels. *Biochemistry*, **41**: 14734–14747.

Mintz, I. M., 1994. Block of Ca channels in rat central neurons by the spider toxin ω-Aga-IIIA. *J. Neurosci*., **14**: 2844–2853.

Monge-Fuentes, V., Gomes, F. M. M., Campos, G. A. A., Silva, J. C., Biolchi, A. M., Anjos, L. L., Gonçalves, J. C., Lopes, K. S. & Mortari, M. R., 2015. Neuroactive compounds obtained from arthropod venoms as new therapeutic platforms for the treatment of neurological disorders. *J. Venom. Anim. Toxins. Trop. Dis*., **21**: 31.

Mor, A. & Nicolas, P., 1994. Isolation and structure of novel defensive peptides from frog skin. *Eur. J. Biochem.*, **219**: 145–154.

Mortari, M. R. & Cunha, A. O. S., 2013. New perspectives in drug discovery using neuroactive molecules from the venom of arthropods. In: Baptista GR, (eds). An Integrative View of the Molecular Recognition &Toxicology - From Analytical Procedures to Biomedical Applications. pp: 91–117.

Mortari, M. R., Cunha, A. O. S., Ferreira, L. B. & Dos Santos, W. F., 2007. Neurotoxins from invertebrates as anticonvulsants: from basic research to therapeutic application. *Pharmacol. Ther*., **114**(**2**): 171–183.

Mueller, A. L., Artman, L. D., Balandrin, M. F., Brady, E., Chien, Y. E. & Delmar, E. G., 1999. NPS 1506, a novel NMDA receptor antagonist and neuroprotectant. Review of preclinical and clinical studies. *Ann. N. Y. Acad. Sci*., **890**(**1**): 450–457.

Nauen, R., 2007. Insecticide resistance in disease vectors of public health importance. *Pest. Manag. Sci*., **63**: 628–633.

Nguyen, L. T., Haney, E. F. & Vogel H. J., 2011. The expanding scope of antimicrobial peptide structures and their modes of action. *Trends Biotechnol*., **9**: 464–472.

Nicholson, G. M., 2007. Fighting the global pest problem: preface to the special Toxicon issue on insecticidal toxins and their potential for insect pest control. *Toxicon*., **49**: 413–422.

Nomura, K. & Corzo G., 2006. The effect of binding of spider-derived antimicrobial peptides, oxyopinins, on lipid membranes. *Biochim. Biophys. Acta*., **1758**: 1475–1482.

North, R. A., 2004. P2X3 receptors and peripheral pain mechanisms. *J. Physiol*., **554**: 301–308.

Oerke, E. & Dehne, H., 2004. Safeguarding production-losses in major crops and the role of crop protection. *Crop Prot*., **23**: 275–285.

Oswald, R. E., Suchyna, T. M., McFeeters, R., Gottlieb, P. & Sachs, F., 2002. Solution structure of peptide toxins that block mechanosensitive ion channels. *J. Biol. Chem*., **277**: 34443–34450.

Peshin, R., Dhawan, A. K., Kalra, R. & Tript, K., 2007. Evaluation of insecticide resistance management based integrated pest management programme. *AI & Society J. Human Centred Systems*, **21**: 358–381.

Peters, B. M., Shirtliff, M. E. & Jabra-Rizk, M. A., 2010. Antimicrobial peptides: Primeval molecules or future drugs? *PLoS Pathog*., **6**: e1001067.

Pignataro, G., Simon, R. P. & Xiong, Z. G., 2007. Prolonged activation of ASIC1a and the time window for neuroprotection in cerebral ischaemia. *Brain*, **130**: 151–158.

Pinheiro, A.C., da Silva, A.J., Prado, M.A., Cordeiro, M.N., Richardson, M., Batista, M.C., de Castro Junior, C.J., Massensini, A.R., Guatimosim, C., Romano-Silva, M.A., Kushmerick, C. & Gomez, M.V., 2009. Phoneutria spider toxins block ischemia-induced glutamate release, neuronal death, and loss of neurotransmission in hippocampus. *Hippocampus,* **19**:1123–1129.

Prado, M. A. M., Guatimosim, C., Gomez, M. V., Diniz, C. R., Cordeiro, M. N. & Romano-Silva, M., 1996. A novel tool for the investigation of glutamate release from rat cerebrocortical synaptosomes: the toxin Tx3-3 from the venom of the spider *Phoneutria nigriventer*. *Biochem. J*., **314**: 145–150.

Price, R. N. & Nosten, F., 2001. Drug resistant falciparum malaria: clinical consequences and strategies for prevention*. Drug Resist. Updat*., **4**: 187–196.

Priest, B. T., 2009. Future potential and status of selective sodium channel blockers for the treatment of pain. *Curr. Opin. Drug Discov. Devel*., **12**: 682–692.

Pukala, T. L., Boland, M. P., Gehman, J. D., Kuhn-Nentwig, L., Separovic, F. & Bowie, J. H., 2007. Solution structure and interaction of cupiennin 1a, a spider venom peptide, with phospholipid bilayers. *Biochemistry*, **46**: 3576–3585.

Quistad, G. B. & Skinner, W. S., 1994. Isolation and sequencing of insecticidal peptides from the primitive hunting spider, *Plectreurys tristis*. *J. Biol. Chem*., **269**: 11098–11101.

Raghunath, D., 2008. Emerging antibiotic resistance in bacteria with special reference to India. *J. Biosci*., **33**: 593–603.

Rajendra, W., Armugam, A. & Jeyaseelan, K., 2004. Neuroprotection and peptide toxins. *Brain Res. Rev*., **45**(**2**): 125–141.

Rash, L. D. & Hodgson, W. C., 2004. Pharmacology and biochemistry of spider venoms. *Toxicon*, **40**(**3**): 225.

Raymond, V., Sattelle, D. B. & Lapied, B., 2000. Co-existence in DUM neurones of two GluCl channels that differ in their picrotoxin sensitivity. *Neuroreport*, **11**: 2695–2701.

Riaz, H., Malik, F., Raza, A., Hameed, A., Ahmed, S. & Shah, P. A., 2011. Assessment of antibiotic prescribing behavior of consultants of different localities of Pakistan. *Afr. J. Pharm. Pharmacol*., **5**: 596–601.

Robbel, L. & Marahiel, M. A., 2010. Daptomycin, a bacterial lipopeptide synthesized by a nonribosomal machinery. *J. Biol. Chem*., **285**(**36**): 27501–27508.

Rodrigues, M. L., Franzen A. J., Nimrichter, L. & Miranda. K., 2013. Vesicular mechanisms of traffic of fungal molecules to the extracellular space. *Curr. Opin. Microbiol*., **16**(**4**): 414-420.

Ross, C. A. & Poirier, M. A., 2004. Protein aggregation and neurodegenerative disease. *Nat. Med*., **10**: 10–17.

Sachs, J. & Malaney, P., 2002. The economic and social burden of malaria. *Nature*, **415**: 680-685.

Saez, N. J., Senff, S., Jensen, J. E., Er, S. Y., Herzig, V., Rash, L. D. & King, G. F., 2010. Spider-Venom Peptides as Therapeutics. *Toxins* (Basel), **2**(**12**): 2851–2871.

Santos, D. M., Verly, R. M., Pilo-Veloso, D., de Maria, M., de Carvalho, M. A., Cisalpino, P. S., Soares, B. M., Diniz, C. G., Farias, L. M., Moreira, D. F., Frezard, F., Bemquerer, M. P., Pimenta, A. M. & de Lima, M. E., 2010. LyeTx I, a potent antimicrobial peptide from the venom of the spider *Lycosa erythrognatha*. *Amino Acids*, **39**(**1**): 135-144.

Schmitt, U., Lüddens, H. & Hiemke, C., 2002. Anxiolytic-like effects of acute and chronic GABA transporter inhibition in rats. *J. Neural Transm*., **109**(**5–6**): 871–880.

Silva, P. I. Jr., Daffre, S. & Bulet, P., 2000. Isolation and characterization of gomesin, an 18-residue cysteine-rich defense peptide from the spider *Acanthoscurria gomesiana* hemocytes with sequence similarities to

horseshoe crab antimicrobial peptides of the tachyplesin family. *J. Biol. Chem*., **43**: 33464–33470.

Skaper, S. D., Debetto, P. & Giusti, P., 2010. The P2X7 purinergic receptor: from physiology to neurological disorders. *FASEB J*., **24**: 337–345.

Sluka, K. A., Winter, O. C., & Wemmie, J. A., 2009. Acid-sensing ion channels: a new target for pain and CNS diseases. *Curr. Opin. Drug Discov. Devel*., **12**: 693–704.

Souza, A. H., Ferreira, J., Cordeiro, M. N., Vieira, L. B., De Castro, C. J., Trevisan, G., Reis, H., Souza, I. A., Richardson, M. & Prado, M. A., 2008. Analgesic effect in rodents of native and recombinant ph α1β toxin, a high-voltage-activated calcium channel blocker isolated from armed spider venom. *Pain*, **140**: 115–126.

Staines, H. M., Ellory, J. C. & Chibale, K., 2005. The new permeability pathways: targets and selective routes for the development of new antimalarial agents. *Comb. Chem*., High Throughput Screen., **8**: 81–88.

Suchyna, T. M., Johnson, J. H., Hamer, K., Leykam, J. F., Gage, D. A., Clemo, H. F., Baumgarten, C. M. & Sachs, F., 2000. Identification of a peptide toxin from *Grammostola spatulata* spider venom that blocks cation-selective stretch-activated channels. *J. Gen. Physiol*., **115**: 583–598.

Sutti, R., Rosa B. B., Wunderlich, B., Junior, P. I. & Silva, T. A., 2015. Antimicrobial activity of the toxin VdTX-I from the spider *Vitalius dubius* (Araneae, Theraphosidae). *Biochem.* and *Biophysics Reports*, **4**: 324–328.

Sutton, K. G., Siok, C. & Stea, A., 1998. Inhibition of neuronal calcium channels by a novel peptide spider toxin, DW13.3. *Mol. Pharmacol*., **54**: 407–418.

Tauxe, R. V., Puhr, N. D., Wells, J. G., Hargrett-Bean, N. & Blake, P. A., 1990. Antimicrobial resistance of Shigella isolates in the USA: the importance of international travelers. *J. Infect. Dis*., **162**: 1107–1111.

Tomalski, M. D., Bruce, W. A., Travis, J. & Blum, M. S., 1988. Preliminary characterization of toxins from the straw itch mite, *Pyemotes tritici*, which induce paralysis in the larvae of a moth. *Toxicon*, **26**: 127–132.

Vassilevski, A. A., Kozlov, S. A. & Grishin E. V., 2009. Molecular diversity of spider venom. *Biochemistry (Mosc.)*, **74**: 1505–1534.

Vassilevski, A. A., Kozlov, S. A., Samsonova, O. V., Egorova, N. S., Karpunin, D. V., Pluzhnikov, K. A., Feofanov, A. V. & Grishin, E. V., 2008. Cyto-insectotoxins, a novel class of cytolytic and insecticidal peptides from spider venom. *Biochem. J*., **411**: 687–696.

Vassilevski, A. A., Sachkova, M. Y., Ignatova, A. A., Kozlov, S. A., Feofanov, A. V. & Grishin, E. V., 2013. Spider toxins comprising disulfide-rich and linear amphipathic domains: a new class of molecules identified in the lynx spider *Oxyopes takobius*. *FEBS J*., **280**(**23**): 6247–6261.

Vassili, N. L., Nadezhda, F. P., Marina, M. S., Elena, S. K., Alexander, A. V., Sergey, A. K., Eugene, V. G. & Vadim, M. G., 2011. Spider Venom Peptides for Gene Therapy of Chlamydia Infection. *Antimicrob Agents Chemother.*, **55**(**11**) : 5367–5369.

Venkatesan, P., Perfect, J. R. & Myers, S. A., 2005. Evaluation and management of fungal infections in immunocompromised patients. *Dermatol. Ther*., **18**(**1**) : 44–57.

Vetter, I., Davis, J. L., Rash, L. D., Anangi, R., Mobli, M., Alewood, P. F., Lewis, R. J. & King, G. F., 2011. Venomics: a new paradigm for natural products-based drug discovery. *Amino Acids*, **40**: 15–28.

Vieira, L. B., Kushmerick, C., Hildebrand, M. E., Garcia, E., Stea, A., Cordeiro, M. N., Richardson, M., Gomez, M. V. & Snutch T. P., 2005. Inhibition of high voltage-activated calcium channels by spider toxin pntx3-6. *J. Pharmacol. Exp. Ther*., **314**: 1370–1377.

Vizioli, J. & Salzet, M., 2002. Antimicrobial peptides from animals: focus on invertebrates. *Trends Pharmacol. Sci.*, **23**: 494-496.

Vooturi, S. K. & Firestine, S. M., 2010. Synthetic membrane-targeted antibiotics. *Curr. Med. Chem*., **17**: 2292–2300.

Wallace, M. S., Rauck, R. L. & Deer, T., 2010. Ziconotide combination intrathecal therapy: Rationale and evidence. *Clin. J. Pain*., **26**: 635–644.

Wang, X. H., Smith, R., Fletcher, J. I., Wilson, H., Wood, C. J., Howden, M. E. & King, G. F., 1999. Structure-function studies of ω-atracotoxin, a potent antagonist of insect voltage-gated calcium . *Eur. J. Biochem*., **264**: 488–494.

Wattal, C., Goel, N., Oberoi, J. K., Raveendran, R., Datta, S. & Prasad, K. J., 2010. Surveillance of multidrug resistant organisms in tertiary care hospital in Delhi, India. *J. Assoc. Physicians India*., **58**: 32–36.

Wei, A. D., Gutman, G. A., Aldrich, R., Chandy, K. G., Grissmer, S. & Wulff, H., 2005.

International Union of Pharmacology. LII. Nomenclature and molecular relationships of calcium-activated potassium channels. *Pharmacol. Rev*., **57**: 463–472.

Wellems, T. E. & Plowe, C. V., 2001. Chloroquine-Resistant Malaria. *J. Infect. Dis*., **184**(**6**): 770-776.

Wemmie, J. A., Price, M. P. & Welsh, M. J., 2006. Acid-sensing ion channels: Advances, questions and therapeutic opportunities. *Trends Neurosci*., **29**: 578–586.

Williams K., 1993. Effects of *Agelenopsis aperta* toxins on the n-methyl-d-aspartate receptor: polyamine-like and high-affinity antagonist actions. *J Pharmacol Exp Ther.,* **266**(**1**): 231–236.

Windley, M. J., Herzig, V., Dziemborowicz, S. A., Hardy, M. C., King, G. F. & Nicholson, G. M., 2012. Spider-venom peptides as bioinsecticides. *Toxins (Basel)*, **4**(**3**): 191-227.

Wirkner, K., Sperlagh, B. & Illes, P., 2007. P2X3 receptor involvement in pain states. *Mol. Neurobiol*., **36**: 165–183.

World Health Organization, 2006. Neurological disorders: public health challengers. Switzerland. pp:1-232.

World Health Organization, 2012. Dementia: a public health priority. pp: 1-112.

World Health Organization, 2014. Antimicrobial resistance: global report on surveillance. pp: 1-232.

World Malarial Report, 2015. New report signals country progress in the path to malaria pelimination. Brussels. pp: 1-181.

Wright, G. D., 2007. The antibiotic resistome: the nexus of chemical and genetic diversity. *Nat. Rev. Microbiol*., **5**: 175–186.

Xiong, Z. G., Pignataro, G., Li, M., Chang, S. Y. & Simon, R. P., 2008. Acid-sensing ion channels (ASICs) as pharmacological targets for neurodegenerative diseases. *Curr. Opin. Pharmacol*., **8**: 25–32.

Xiong, Z., Zhu, X., Chu, X., Minami, M., Hey, J. & Wei W., 2004. Neuroprotection in ischemia: blocking calcium-permeable acid-sensing ion channels. *Cell*, **118**(**6**): 687–698.

Xu, K., Ji, Y. & Qu, X., 1989. Purification and characterisation of an antibacterial peptide from venom of *Lycosa singoriensis*. *Acta Zoologica Sinica*, **35**: 300–305.

Yan, L. & Adams, M. E., 1998. Lycotoxins, antimicrobial peptides from venom of the wolf spider *Lycosa carolinensis*. *J. Biol. Chem*., **273**: 2059–2066.

Youdim, M. B., Kupershmidt, L., Amit, T. & Weinreb, O., 2014. Promises of novel multi-target neuroprotective and neurorestorative drugs for Parkinson's disease. *Parkinsonism Relat Disord*., **20**(**1**): 132–136.

Zamponi, G. W., 1997. Antagonist sites of voltage dependent calcium channels. *Drug Dev. Res*., **42**: 131–143.

Zasloff, M., 2002. Antimicrobial peptides of multicellular organisms. *Nature*, **415**: 389-395.

Zhao, H., Kong, Y., Wang, H., Yan, T., Feng, F., Bian, J., Yang, Y. & Yub, H., 2011. A defensin-like antimicrobial peptide from the venoms of spider, *Ornithoctonus hainana*. *J. Pept. Sci*., **17**: 540–544.

Pinheiro A. C., da Silva A. J., Prado M. A. et al. (2009) Phoneutria spider toxins block ischemia-induced glutamate release, neuronal death, and loss of neurotransmission in hippocampus. Hippocampus 19, 1123–1129.

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