

Helminthes and insects: maladies or therapies

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Received: 13 October 2014 / Accepted: 15 December 2014
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Abstract By definition, parasites cause harm to their hosts. But, considerable evidence from ancient traditional medicine has supported the theory of using parasites and their products in treating many diseases. Maggots have been used successfully to treat chronic, long-standing, infected wounds which failed to respond to conventional treatment by many beneficial effects on the wound including debridement, disinfection, and healing enhancement. Maggots are also applied in forensic medicine to estimate time between the death and discovery of a corpse and in entomotoxicology involving the potential use of insects as alternative samples for detecting drugs and toxins in death investigations. Leeches are segmented invertebrates, famous by their blood-feeding habits and used in phlebotomy to treat various ailments since ancient times. Leech therapy is experiencing resurgence nowadays in health care principally in plastic and reconstructive surgery. Earthworms provide a source of medicinally useful products with potential antimicrobial, antiviral, and anticancer properties. Lumbrokinases are a group of fibrinolytic enzymes isolated and purified from earthworms capable of degrading plasminogen-rich and plasminogen-free fibrin and so can be used to treat various conditions associated with thrombotic diseases. Helminth infection has been proved to have therapeutic effects in both animal and human clinical trials with promising evidence in treating many allergic diseases and can block the induction of or reduce the severity of some autoimmune disorders as Crohn's disease or ulcerative colitis. What is more, venomous arthropods such as scorpions, bees, wasps, spiders, ants, centipedes, snail, beetles, and caterpillars. The venoms and toxins from these arthropods provide a promising source of natural bioactive compounds which can be employed in the

development of new drugs to treat diseases as cancer. The possibility of using these active molecules in biotechnological processes can make these venoms and toxins a valuable and promising source of natural bioactive compounds. The therapeutic use of helminthes and insects will be of great value in biomedicine and further studies on insect toxins will contribute extensively to the development of Biomedical Sciences.

Keywords Helminthes · Insects · Maggot · Alternative medicine

Introduction

Insects and helminthes have been used globally in traditional medicine for centuries in East Asia, Africa, and South America and still used in parts of the world where conventional medical care is scarce. This folk medicine provides a cheap method in economically challenged world (Pechter and Sherman 1983; Cooper et al. 2012). The research on maggots, helminthes, and insect therapy is highly successful promising aspects in biomedicine (Bonn 2000; Amendt et al. 2011). Furthermore, insect's ingredients have been evaluated experimentally and preliminary reports proved their beneficial properties (Marsh and Williams 2005; Kini 2006). Insect-based products that purify arthropod-derived constituents have been developed recently and will yield further benefits (Chaim et al. 2011; Oršolić 2012).

Maggot

Maggot therapy is an inexpensive natural way combating wound infection, and it is under research to obtain more benefits from its implementation in the new alternative medicine. William Baer pioneered the use of sterile maggots as a

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biosurgical method of wound therapy in the late 1930s, after observing its value in traumatic wounds on the battlefield in France during World War I (Baer 1931). By the mid-1940s maggot therapy was neglected after the era of antibiotics therapy and improved surgical techniques. Later on, maggot therapy made its second resurgence with the emergence of antibiotic-resistant bacteria strains in the 1980s (Sherman et al. 2000). Nowadays, larvae of the green bottle blowfly *Lucilia sericata* are used worldwide as a debridement agent for chronic and infected wounds such as for the treatment of venous stasis leg ulcers (Wayman et al. 2000), diabetic foot ulcers, especially neuropathic ulcers (Marineau et al. 2011; Wilasrusmee et al. 2014), wound bed preparation prior to surgical closure (Sherman and Shimoda 2004), traumatic and post-surgical wounds (Namias et al. 2000), osteomyelitis (van der Plas et al. 2008), and burns (Robinson and Norwood 1933). Moreover, the larval excreta/secretions are used as potent agents against methicillin-resistant *Staphylococcus aureus* infection (Bexfield et al. 2004). In 2004, the US Food and Drug Administration (FDA) allowed production and marketing of maggots as a medical tool. Many types of larvae were used as a debridement therapy like *Lucilia caesar* (Baer 1931), the large blue bottle flies (*Calliphora vomitans* and *Calliphora erythrocephala*) (Weil et al. 1933), and *Lucilia cuprina* (Fine and Alexander 1934). But maggots of *L. sericata* are the most commonly used probably because of its starvation on clean granulation tissue (Weil et al. 1933). The most common adverse effects associated with maggot therapy are discomfort which is reported in 5–30 % of treated wounds (Sherman 2003), delivery problems, and escaping maggots (Sherman 2009).

The outstanding effects of medicinal maggot therapy attributed to the synergistic actions of these main activities: (1) Debridement (Fleischmann et al. 1999; Wayman et al. 2000; Sherman et al. 2007), which means the removal of foreign material and dead contaminated tissue from wound bed to expose healthy tissue, and this is the most known and accepted mechanism of action by maggots. It occurs by secreting a rich cocktail of digestive enzymes during its feeding like leucine aminopeptidase, carboxypeptidases A and B (Vistnes et al. 1981), serine proteases (trypsin-like and chymotrypsin-like enzymes) (Chambers et al. 2003), and collagenase (Ziffren et al. 1953). Moreover, four proteolytic enzymes, containing two serine proteases, a metalloproteinase and an aspartyl proteinase, were detected in its secretions with molecular weights ranging from 20 to 40 kDa, with activity across a wide pH range (Chambers et al. 2003). The significant role of a chymotrypsin-like serine proteinase in the larva of *Lucilia* in degradation of wound matrix components laminin, fibronectin, and collagen types I and III has been defined. Moreover, the wriggling of maggots have mechanical effect which promotes debridement as these possess mouth hooks which probes and macerate the necrotic tissue (Barnard 1977). (2)

Disinfection (Sherman and Shimoda 2004; Armstrong et al. 2005; Bowling et al. 2007), the application of maggots to an infected wound results in rapid elimination of many anaerobic and aerobic bacteria (Bowler and Davies 1999) including Gram-negative bacteria like *Escherichia coli*, *Pseudomonas aeruginosa*, and *Salmonella* spp, as well Gram-positive bacteria such as *S. aureus*, *Staphylococcus epidermis*, *Listeria monocytogenes*, and clinical isolates of methicillin-resistant *S. aureus* (Bexfield et al. 2004; Daeschlein et al. 2007; Kerridge et al. 2005). Moreover, maggots produce a large quantity of ammonia which alters wound pH from acidic to neutral or slightly alkaline at pH 7 or 8 which discourage bacterial growth (Mumcuoglu 2001); (3) Stimulation of healing (Sherman et al. 1995; Horobin et al. 2005), maggots have ability to physically stimulate granulation tissue by its crawling motions (Buchman and Blair 1932). Also, it may be due to the nitrogenous waste of *Lucilia* containing 10 % allantoin (2,5-Dioxo-4-imadazolidinyl urea) (Robinson 1935) and 90 % ammonia biocarbonate (Robinson 1940) which are responsible for the abundant growth of granulation tissue; and (4) Eradication and inhibition of *Staphylococcus epidermidis* (Harris et al. 2009) *S. aureus* and, to a lesser extent, *P. aeruginosa* and biofilms (van der Plas et al. 2008).

Another interesting use of maggots was in forensic entomology, which is the science of collecting and analyzing insect evidence for forensic and legal purposes (Amendt et al. 2007). Most important task in forensic medicine is estimating the minimum post-mortem interval (PMI) that refers to the time between the death and discovery of a corpse (Catts 1992). Insects are the most powerful method to estimate this time quickly and perfectly (Amendt et al. 2011). Depending on environmental conditions, necrophagous insects will colonize a fresh corpse quickly. Usually, the body will be attacked firstly by flies (Diptera), mainly the blow fly of family Calliphoridae (Sukontason et al. 2007), which can locate an odor source with great spatial precision and deposit their eggs on a corpse within minutes of death. Larvae hatch from the eggs and feed on the underlying tissues. They grow and moult twice, and when third instar larvae finish feeding, they enter the post-feeding stage, puparium, and migrate away from the body to find shelter. Decomposition occurs in corpse as a result of continuous process of insect activity in and on the corpse. The degree of this decomposition can be measured, allowing accurate minimum PMI estimates to be made up to several months after death depending on the circumstances and temperature profile of the region (Campobasso et al. 2001; Reibe et al. 2010). The hypothesis behind these estimates is that by calculating the age of developing insects on a body, it is possible to calculate the time of colonization, which infers a minimum PMI (PMI_{min}), and because blowflies are the usual first group to colonize a body, the focus of PMI_{min} estimates is often on them when using entomological evidence (Catts 1992). There is a different rate of development

Table 1 Studies of helminth therapy in experimental animals

Parasite	Mechanism of action	Reference
<i>Trichinella spiralis</i>	Inhibit dinitrobenzene sulphonic acid (DNBS) induced colitis in mice by cytokines skewing towards Th2 and reduction of IFN- γ production Improve the clinical course of induced EAE in rats	(Khan et al. 2002) (Gruden-Movsesijan et al. 2008)
<i>Hymenolepis diminuta</i>	Inhibit ion transport abnormalities in the colon of dextran sodium sulphate -treated mice Block DNBS-induced colitis in mice by IL-10 production	(Wohlleben et al. 2004) (Hunter et al. 2005)
<i>Ascaris sum</i>	Its chronic infection reduces ragweed-triggered allergic eye disease in mice by mobilization of CD4+CD25+ T cells Decrease the severity of CIA in mice by reducing of IL-10	(Schopf et al. 2005) (Rocha et al. 2008)
<i>Litomosoides sigmodontis</i>	Inhibits allergic airway inflammation and hyper-reactivity in mice by mobilization of regulatory T cells and TGF β	(Dittrich et al. 2008)
<i>Schistosoma mansoni</i>	Inhibit DSS-induced colitis through mobilization of an immunosuppressive or immunoregulatory macrophage phenotype Inhibit TNBS-induced colitis in rats by modulation of colonic cytokine levels Protect against anaphylaxis by IL-10-producing (IL-4 deficient) β cells Reduce severity of experimental allergic encephalitis in mice by suppressing IL-12p40 production Prevent diabetes in the nNon-obese diabetic (NOD) mouse by generation of Th2 response suppress collagen-induced arthritis in mice by downregulation of Th1 cytokines, IL-1b, and NF- κ B in the paw Its protein extract ameliorate motility disturbances in TNBS-induced murine colitis	(Smith et al. 2007) (Moreels et al. 2004) (Mangan et al. 2004) (Sewell et al. 2003) (Cooke et al. 1999) (Osada et al. 2009) (Ruyssers et al. 2010)
<i>Schistosoma japonicum</i>	Male cercaria protects mice from allergen-induced airway hyper-responsiveness by reduced IL-5, increased IL-10 and evidence for B cell involvement Its soluble egg antigen ameliorate the severity and progression of EAE by increasing Th2 and regulatory type cytokines (IL-4, IL-10, TGF- β)	(Mangan et al. 2006) (Zheng et al. 2008)
<i>Acanthoecilonema viteae</i>	Ameliorate the course of collagen-induced arthritis (CIA) in mice by production of glycoprotein, termed ES-62	(McInnes et al. 2003)
<i>Fasciola hepatica</i>	Suppression of autoantigen-specific Th1 and Th17 responses that mediate autoimmune diseases by induction of TGF- β in infected mice	(Walsh et al. 2009)
<i>Trichuris suis</i>	Soluble products of <i>T. suis</i> , <i>S. mansoni</i> , and <i>T. spiralis</i> suppress TNF- α and IL-12 secretion by TLR-activated human dendritic cells in murine experimental autoimmune encephalomyelitis	(Kuijk et al. 2012)

DNBS dinitrobenzene sulphonic acid, EAE experimental autoimmune encephalomyelitis, TNBS trinitrobenzene sulfate, CIA collagen-induced arthritis, MLDS multiple low-dose streptozotocin-induced Diabetes, TLR Toll-like receptors

regarding each Calliphoridae species according to its specific energy requirements and the nutritional composition of each type of food (Thyssen et al. 2014).

Entomotoxicology is another relatively new branch of forensic entomology. It involves the potential use of insects as alternative samples for detecting drugs and toxins (Kintz et al. 1990). In death investigations, Diptera and other arthropods can be reliable alternate specimens for toxicological analyses in the absence of tissues and fluids normally taken for such purposes. Entomotoxicology also investigates the effects caused by drugs and toxins on arthropod development in order to assist the forensic postmortem interval estimates (Nolte et al. 1992; Introna et al. 2001). Most of the substances implicated in drug-related deaths are detectable through analyses of maggots: opiates such as morphine and codeine, cocaine and benzoylecognine, amphetamines, tricyclic antidepressants, phenothiazines and benzodiazepines, steroids, barbiturates, and several salicylates (Introna et al. 2001; Carvalho 2010). Drugs and toxins have also been detected by analyzing of

puparial cases (Pien et al. 2004), and moreover, it is founded in beetle exuviae and fecal material (Miller et al. 1994). Bourel et al. (2001) have demonstrated the accumulation of morphine inside the cuticle of *Calliphora vicina* maggots.

Helminthes

Although parasites are harmful to their hosts, there is a growing evidence from experimental studies using animal models of human disease supporting that helminthes infection play a role in suppressing the progression and incidence of other diseases (Garg et al. 2014) (Table 1). Moreover, it has demonstrated that helminth infections have reduced the incidences of autoimmune and allergic diseases (McKay 2008). It is known that, in helminths infection, Th2 is the predominant immunological response. This Th2 stimulation can ameliorate disorders in which Th1 is the dominant event (McKay 2009). The dependable theory for most researchers to use helminth

infection in blocking the induction of or reducing the severity of some diseases is predicting the immunological Th2 events evoked by helminth infection which can ameliorate disorders in which Th1 events predominated (McKay 2009). This is supported by observing the strong correlation between reduction in parasite exposure and the dramatic increase in atopic, autoimmune, and autoinflammatory disorders in developed countries (Rook 2009). Moreover, clinical evidence support the theory of helminth therapy for unrelated diseases; such as using *Trichuris suis* therapy in Crohn's disease (Summers et al. 2005; Reddy and Fried 2007, 2009) and ulcerative colitis since its colonization promotes goblet cell hyperplasia and mucus production through Th2 cytokines and IL-22 (Broadhurst et al. 2010), *Necator americanus* in the treatment of Crohn's disease (Croese et al. 2006) and protection from asthma (Mortimer et al. 2006), and *Schistosoma haematobium* in decreasing atopy (van den Biggelaar et al. 2000). However, studies oppose this trend not only for psychological reasons, but also for safety measures against risk of zoonosis or tissue damage by accidental worm migration to unwanted sites in the human body (Van Kruiningen and West 2005). On the other hand, studies favor the use of helminth infection as a therapy but considering other factors that determine the relationship between helminth and allergic disease as (1) the duration of infection as early and/or chronic infections are more efficient in down-modulation of allergic disease, but sporadic or transient infections may enhance allergic clinical symptoms (Cooper et al. 2006); (2) infection intensity since light infections may promote allergic disease whereas a heavy one may induce immune suppression (Smits et al. 2010); (3) host Genetics, which is responsible for induction of the specific host immune regulatory mechanisms; therefore, individuals genetically susceptible to atopic disease may be more liable to develop allergic responses to helminth and allergens and will be genetically more resistant to infection (Moller et al. 2007); and (4) concurrent infections and environmental differences affect the outcome of analyses of natural infections in humans (McKay 2009).

Leech

Medicinal leech therapy is an earliest skill that dates back to ancient Egypt. Leeches was one of the most common therapeutic for human illness in Greek, Greco-Roman, and Byzantine medicine (Whitaker et al. 2004). The popularity of the use of leeches reached the peak in the early nineteenth century in Europe (Eldor et al. 1996).

Leeches are sanguivorous or blood-sucking annelid worms, usually black or brown in color and about 10 cm in length. Of the more than 650 known species in the *Hirudo* genus, *Hirudo medicinalis* is the commonly used in medical therapy. The mouth of the leech has three jaws; each has about

100 tiny teeth, in addition to a posterior suction device, which is used for stability. Leech therapy involves an initial painless bite, then sucking about 5 to 15 mL of blood by dissecting the skin with its sharp teeth (Godekmerdan et al. 2011) and simultaneously injecting of a cocktail of several medicinally useful bioactive molecules in their saliva (Zaidi et al. 2011). Once they are detached, the site of bite continues to bleed for several hours (Ikizceli et al. 2005). The most important substance in their saliva is a pure anti-coagulating called 'Hirudine' which is discovered by Haycraft 1884. In 1955, Markwardt have isolated and characterized Hirudin from leech pharyngeal glands. Subsequently, this effective anticoagulant was produced by genetic engineering (Fields 1991). Moreover, the leech also secretes hyaluronidase, which allows the anticoagulant to spread throughout the wound, and antihistamines, which vasodilate and contribute to prolongation of bleeding following a bite (Whitaker et al. 2004).

The most serious complication of leech therapy is infection, which occur in 2.4–20 % of plastic surgical repairs (de Chalaín 1996). The leech's digestive system contains a symbiote, *Aeromonas hydrophila*, which is the most common organism found in leech infections (Whitaker et al. 2003; Ardehali et al. 2006). Infections can arise from 2 to 11 days after therapy begins and can result in abscesses and cellulitis, which can progress in some cases to sepsis (Abdelgabar and Bhowmick 2003). von Rheinbaben et al. (2014) recommended making a quarantine period of 4–6 months after the lasting feeding of leech which supports microbial degradation inside leech gut and so decrease risk of viral infection for treated patients. Also, excessive bleeding can occur with leech therapy which may necessitate blood transfusion (Whitaker et al. 2003).

Leeching is applied to head for treatment of headache or mania and in cases of inflammation of the throat, with their application to the neck (Adams 1988). Currently, leech is extensively used in surgery, such as plastic surgery to aid wound healing (Whitaker et al. 2004), in the treatment of ring avulsion injuries in plastic surgery (Tuncali et al. 2004), after breast surgery to relieve the possible complication of venous congestion (Gross and Apesos 1992), treat venous congestion and hematoma after plastic reconstructive surgery (Riede et al. 2010), alleviate post-phlebitis syndrome, in which venous valves are obliterated by a deep vein thrombosis (Eldor et al. 1996), reduce symptoms caused by osteoarthritis (Michalsen et al. 2003; Andereya et al. 2008), symptomatic relief of refractory cancer pain (Kalender et al. 2010), with replanted digits (Foucher et al. 1981), ears (Cho and Ahn 1999), lips (Walton et al. 1998), and nasal tips (Mortenson et al. 1998), and for treatment coronary ischemia (van de Loo and Bode 2002).

Recently, specific substances from leeches have been refined and utilized to prevent thrombosis (Markwardt 2002). Also, a topical cream has been manufactured from hirudin and used to shrink hematomas caused by musculoskeletal injuries (Stamenova et al. 2001). Recombinant hirudins are

undergoing clinical trials to treat deep venous thrombosis and acute coronary syndromes and also as an adjuvant to thrombolysis in myocardial infarction (Markwardt 1956).

The most serious complication of leech therapy is infection that occurs in 2.4–20 % of plastic surgical repairs (de Chailain 1996) due to the presence of symbiote, *A. hydrophila* in the leech's digestive system (Ardehali et al. 2006). Infections can arise within 11 days after therapy begins and can result in abscesses and cellulitis, which may progress to a life-threatening sepsis (Abdelgabar and Bhowmick 2003). Other reported complications include allergic reactions, anemia, scarring, and bleeding that may necessitate blood transfusion (Whitaker et al. 2003).

Earthworms

Earthworms have been widely used in traditional medicine for thousands of years (Sun et al. 1997) since they contain substances with potential antimicrobial, antiviral, and anticancer properties (Cooper et al. 2004). Earthworms have been used to treat inflammatory, hematological, oxidative, and nerve diseases (Chen et al. 2010; Liu et al. 2013). Baking earthworms with bread have been used to treat kidney stones; in addition, ingestion of dried or burnt ashes of earthworms has been used to improve alopecia and relieve jaundice. Moreover, topical application of dead earthworms alleviates arthralgia and treat injuries (Cooper et al. 2004). Furthermore, treatment of some blood diseases and supporting circulatory health was recorded with oral ingestion of earthworm as powder (Pan et al. 2011).

A strong fibrinolytic enzyme from the earthworm, *Lumbricus rubellus* has been purified and is capable of degrading plasminogen-rich and plasminogen-free fibrin. This result suggests that earthworm powder represents a possible oral thrombolytic agent and may thus be applicable for treating patients with thalassemia (Mihara et al. 1991). It was also reported that oral administration of a saline suspension of *L. rubellus* earthworm powder (EWP) to rats could reduce the formation of venous thrombus, suggesting that EWP may be valuable for the prevention and/or treatment of thrombotic diseases (Hahn et al. 1997). Glycolipoprotein mixture (G-90), prepared from the tissue of the earthworm *Eisenia foetida*, was examined in vivo, in an experimental study on Wistar rats, to determine its fibrinolytic and anticoagulation activity. The results have shown that G-90 have a similar effect like heparin, a known anticoagulant, on blood coagulation. Thus, G-90 considered as a new thrombolytic agent of use in veterinary and human medicine (Matausic-Pisl et al. 2011). Additionally, the weight of thrombus has been reduced upon feeding a rat with an arterio-venous shunt on dried powder from *L. rubellus* earth worm (Kim et al. 1998).

Prakash and Gunasekaran (2010) have studied the effect of various doses of earthworm paste (*Lampito mauritii*, Kinberg)

on aspirin-induced gastric ulcer and compared with ranitidine. The earthworm paste showed significant ulcer protective effects due to its anti-secretory action and its effects on mucosal glycoprotein. It is proven to be more effective in gastric cytoprotection than ranitidine in preventing lesion formation. Balamurugan et al. (2009) have reported the anti-inflammatory and anti-pyretic properties of earthworm extract which is similar to glycoprotein complex (G-90). An earthworm-derived Factor Xa (FXa) inhibitor, eisenstasin-derived small peptide (ESP), has been isolated from the midgut of the *Eisenia andrei* worm and found to reduce the expressions of pro-inflammatory cytokines (IL-1 α , IL-1 β , IL-8, IL-16, MCP-1, MIP-1 α , and MIP-1 β) by cultured cells treated with both ESP and FXa. So, ESP might interrupt coagulation cascades by inhibiting FXa and thus may be effective control the bidirectional alternation between coagulation and inflammation (Joo et al. 2009).

Lumbricin-PG, a novel lumbricin-like antimicrobial peptide, has been isolated from skin secretions of the earthworm, *Pheretima guillelmi* (Michaelsen) and has exerted potential antimicrobial activities against bacteria and fungi (Li et al. 2011). The Earthworm decoction can lighten the airway inflammation in asthmatic guinea pigs. It acts by inhibition of eosinophil infiltration, acceleration of eosinophil apoptosis, and improvement of the bronchial tube and the lung tectology changes (Li et al. 2007). Earthworm paste (EP) derived from an indigenous species *Lampito mauritii* (Kinberg) reduce inflammation, restore the levels of antioxidants-reduced glutathione, glutathione peroxidase, superoxide dismutase, catalase, and thiobarbituric acid reactive substances, normalize the values of erythrocyte, leukocyte, differential levels of neutrophils, lymphocytes, eosinophils, hemoglobin, and serum biochemical contents when applied to inflamed Wistar albino rat. It seems to be powerful than standard anti-inflammatory drug, aspirin (Balamurugan et al. 2007).

Regarding its anti-cancer activity, the polysaccharide–protein complex isolated from metabolites of bacteria inhibiting gut of *Raoulitella ornithinolytica* earthworms have anti-fungal and anticancer activity against breast ductal carcinoma and endometrioid ovarian cancer in vitro (Fiołka et al. 2013). Furthermore, earthworm aqueous extracts have been utilized in the green synthesis of gold nanoparticles (AuNPs). Earthworm extracts act as reducing agents to convert Au³⁺ to AuNPs. The earthworm–AuNPs is capable of reinforcing the anticoagulant activities of heparin. So, the newly prepared AuNPs are promising candidates for novel gold nanomedicines (Kim et al. 2013).

Venom

Insect venoms constituents have anti-inflammatory properties that may make them valuable in the treatment of several

Table 2 Peptides isolated from insect venoms

Peptide molecule	Insect	Mechanism	References
Chlorotoxin	Scorpion venom (<i>Leiurus quinquestriatus</i>)	Blocks small conductance chloride channels and selectively interacts with MMP-2 isoforms which are specifically up-regulated in gliomas	(Soroceanu et al. 1999; Deshane et al. 2003; Veisheh et al. 2007)
Bengalin	Indian black scorpion (<i>Heterometrus bengalensis</i>)	Cause human leukemic cells to undergo apoptosis <i>in vitro</i> through mitochondrial pathway against the human histiocytic lymphoma cell line U937 and the human chronic myelogenous leukemia cell line K562 and not affect normal human lymphocytes	(Gupta et al. 2010)
Ziconotide	Snail (<i>Conus magus</i>)	Pain management with a powerful anesthetic effect	(Strigenz 2014)
Conotoxin TVIIA	Snail (<i>Conus magus</i>)	Treatment of intractable pain	(Miljanich 2004; Staats et al. 2004; Stix 2005; Bingham et al. 2010)
		Reduce the size of the myocardial infarct in an ischemia/reperfusion model in rabbits, rats, and dogs <i>in vivo</i>	(Zhang et al. 2003; Lubbers et al. 2005)
TCTP protein	Spider venom gland (<i>Loxosceles intermedia</i>)	Potential cancer biomarker	(Slaby et al. 2009; Kim et al. 2008; van de Sande et al. 2006)
		Is the target of various anticancer drugs and so it could be used as a potential target for therapy	(Tuynder et al. 2004; Efferth 2005; Zhu et al. 2008)
Astacins	Spider (<i>L. intermedia</i>)	Thrombolytic agents	(Chaim et al. 2011)
Tx2–6 toxin	Brazilian wandering spider (<i>Phoneutria nigriventer</i>)	Cause an improvement in the level of nitric oxide in penile tissue in rats	(Andrade et al. 2008; Villanova et al. 2009)
Psalmotoxin 1	Spider (<i>Psalmopoeus cambridgei</i>)	Inhibits Na currents in high-grade human astrocytoma cells	(Escoubas et al. 2000; Bubien et al. 2004)
GsMTx4 peptide	Chinese rose tarantula spider (<i>Grammostola spatulata</i>)	Proved to block pain reaction in rat models	(Park et al. 2008)
Pancratistatin	Spider venom (<i>Pancratium littorale</i>)	Effective against lymphoma, neuroblastoma and breast cancer cell lines, not affecting normal cell lines; induces apoptosis in cancer cells	(McLachlan et al. 2005; Kekre et al. 2005; Siedlakowski et al. 2008)
Gomesin	Spider (<i>Acanthoscurria gomesiana</i>)	Effective topical treatment of subcutaneous murine B16F10-Nex2 melanoma	(Rodrigues et al. 2008)
Tx2–6 toxin	<i>Phoneutria nigriventer</i>	Improvement in the level of nitric oxide in penile tissue in rats and so it can be investigated as a tool in the treatment of erectile dysfunction	(Villanova et al. 2009; Andrade et al. 2008)
Solenopsin A	Ant (<i>Solenopsis invicta</i>)	Antiangiogenic activity	(Arbiser et al. 2007)
μ-SLPTX-Ssm6a peptide	Centipede	Have a promising effect as a novel analgesic treating a broad range of pain conditions exceeding morphine in rodent pain models	(Yang et al. 2013)
Mastoparan	Wasp venom (<i>Polybia paulista</i>)	Mast cell degranulation	(Hirai et al. 1979)
		Activation of G-protein-mediated mechanisms	(Higashijima et al. 1988; Perianin and Snyderman 1989; Ozaki et al. 1990)
		Inhibition of calmodulin-mediated mechanisms	(Wheeler-Jones et al. 1992; Joyce-Brady et al. 1991)
		Stimulation of phospholipases A ₂ and C	(Wallace and Carter 1989; Perianin and Snyderman 1989)
		Stimulation or inhibition of cation-specific channels	(Glavinovic et al. 1992)
		Hemolytic activity	(Souza et al. 2009)
Melittin	Bee venom (<i>Apis mellifera</i>)	Have anti-tumor activity	(Gevod and Birdi 1984)
		Effective against several tumor cell lines, may act as: calmodulin inhibitor; induces cell membrane lysis and apoptosis; potent pore-forming agent; hyperactivates PLA2 in ras oncogene-transformed cells; acts in different cell signaling pathways; effective against tumors <i>in vivo</i>	(Son et al. 2007; Hait et al. 1983; Duke et al. 1994; Shaposhnikova et al. 1997; Sharma 1992; Holle et al. 2003)
Cecropins	Cecropia moth (<i>Hyalophora cecropia</i>)	Have antimicrobial activity	(Andreu et al. 1985)
		Exert strong antibiotic activity against both Gram-positive and Gram-negative bacteria in micromolar concentrations	(Boman et al. 1991)

Table 2 (continued)

Peptide molecule	Insect	Mechanism	References
		Have anti-cancer activity against multiple tumor cell lines including mammalian leukemia, lymphoma and colon carcinoma cell lines	(Chen et al. 1997; Moore et al. 1994)

medical disorders. Research in venom has increased over recent years, and the challenges and opportunities are still enormous. Studies on insect toxins have contributed extensively to the development of Biomedical Sciences. Many bioactive molecules found in insect's venoms can exert important pharmacological activities in human physiology. So, it is of great value to characterize these active components for development of new drugs capable of directly and specifically acting upon cell physiology. Some of major molecules isolated from in these venoms are introduced in Table 2.

Scorpion

Scorpions are an ancient terrestrial venomous lineage. Scorpion's venom exerts toxic activities on a wide range of biological targets (Gantenbein and Largiadèr 2003; Bryson et al. 2013). Venoms from scorpion is a complex mixture of a large variety of molecules rich in proteins and peptides (Kuhn-Nentwig 2003). Over 1,500 scorpion species have been identified, each producing a different type of venom; each venom is estimated to be composed of 50–100 different toxic polypeptides (Possani et al. 2000). In contrast to other venoms, scorpion venom displays low levels of enzymatic activity (Gwee et al. 2002). Among several toxin types present in scorpion venoms are the CS α / β scaffold cysteine-stabilized α / β which include sodium ion channel (NaV) modulators (Quintero-Hernández et al. 2013).

Scorpion venom has been used in traditional Chinese medicine to relieve pain and treat meningitis, epilepsy, stroke, and rheumatic diseases (Wang et al. 2009). Scorpion toxins are a promising approach to fight cancer. The most studied peptides from scorpion venom are the long-chain toxins composed of 60–70 amino acid residues cross-linked by four disulfide bridges and act mostly by activating Na channels (Goudet et al. 2002). A protein called BmBKTx1 created from Chinese scorpion venom prevents the breakdown of the tumor suppressor protein, and another called “AGAP” retards the growth of intra-peritoneal tumors in mice (Liu et al. 2003). Chinese scorpion venom contains a hyaluronidase, which was used to suppress hyaluronan, a cell adhesion factor that promoted metastasis in an in vitro human breast cancer cell line (Feng et al. 2008).

Chlorotoxin (Cltx), a peptide isolated from the species *Leiurus quinquestriatus*, is one of most active components present in scorpion venom (Lippens 1995). Chlorotoxin

inhibit the activity of human glioma cells in vitro by inhibiting chloride influx in glioma cells membrane (Soroceanu et al. 1999) and binding matrix metalloproteinase II (MMP-2) which is involved in tumor invasion (Veisèh et al. 2007). Gao et al. (2008) isolated a serine proteinase-like protein (BMK-CBP) from BmK venom, which could bind to the breast cancer cell line MCF-7 in a dose-dependent manner, showing the potential of BMKCBP as a delivering drug for cancer treatment. A protein from the Indian black scorpion *Heterometrus bengalensis*, bengalin, caused human leukemic cells to undergo apoptosis in vitro through mitochondrial pathway against the human histiocytic lymphoma cell line U937 and the human chronic myelogenous leukemia cell line K562 (chronic myelogenous leukemia), not affecting normal human lymphocytes (Gupta et al. 2010). Moreover, venom from the Venezuelan scorpion, *Tityus discrepans*, has antimicrobial and antiparasitic activity and is found to suppress the growth of *Leishmania* species in vitro (Borges et al. 2006). In addition to another promising toxin in the treatment of sleep apnea, α -toxin retards inactivation of sodium channels at the neuromuscular junction that might increase neuromuscular reflexes and airway contraction (Conduit et al. 2007).

Spider

Spiders are the most diverse group of arthropods, but relatively few toxins have been studied so far (Escoubas 2006). Spider venom contains a collection of peptides and other compounds that include substances altering neuronal sodium, calcium and potassium channels, and glutamate and acetylcholine receptors (Ori and Ikeda 1998). Regarding the biochemistry of spider venoms, they present a variety of ion channel toxins, novel non-neurotoxins, enzymes, and low-molecular-weight compounds (Rash and Hodgson 2002).

The venom of the brown *Loxosceles* spiders contains a complex mixture of protein and peptide toxins with a molecular mass profile ranging from 1 to 40 kDa (Futrell 1992). Several molecules from the *Loxosceles* spider crude venoms have been identified, including alkaline phosphatase (Sales and Santoro 2008), 5'-ribonucleotide phosphohydrolase (Futrell 1992), sulfated nucleosides (Schroeder et al. 2008), hyaluronidase (da Silveira et al. 2007), fosfolipases-D (Kalapothakis et al. 2007), metalloproteases, serine proteases (Barbaro et al. 2005), and insecticide toxins (de Castro et al. 2004). Brown spider hyaluronidase could be used

therapeutically in many fields (Senff-Ribeiro et al. 2008). It can be used to promote absorption of excess fluids, to increase the efficiency of local anesthesia, and to decrease tissue destruction by subcutaneous and intramuscular injection of fluids (Girish and Kemparaju 2007). Hyaluronidase has also been used to reduce the extent of tissue damage following extravasation of parental nutrition solution, electrolyte infusions, antibiotics, aminophylline, mannitol, and chemotherapeutic agents, including Vinca alkaloids (Goolsby and Lombardo 2006). Additionally, technological advancement has led to synthesis of recombinant human hyaluronidase (rHuPH20) which has been used in the management of chronic pain, improving systemic absorption and bioavailability of drugs (Etesse et al. 2009; Muchmore and Vaughn 2010). Moreover, testicular hyaluronidase (HAase) has been added to drug regimens of cancer therapy to improve drug penetration and to enhance the efficacy of vinblastin in the treatment of malignant melanoma and Kaposi's sarcoma, among other cancers (Lokeshwar and Selzer 2008). Furthermore, it has been found that, when the level of HA decreases under conditions in which hyaluronidase activity increases, the moisture and tension of the skin are reduced, and histamine is released from mast cells (Barla et al. 2009), so, the future recognition and characterization of hyaluronidase inhibitors will be relevant to the development of contraceptives, anti-tumor, anti-microbial, anti-venom, anti-wrinkle, anti-aging agents, and allergy and inflammation suppressors (da Silveira et al. 2007; Barla et al. 2009). From this context, it is obvious that *Loxosceles* recombinant hyaluronidases are associated with numerous potential applications (Gremski et al. 2010).

TCTP protein has been identified in *Loxosceles intermedia* venom gland, and it has been proposed as a potential cancer biomarker (Slaby et al. 2009) as TCTP protein levels are upregulated in cancer cells and in human tumors (Ma et al. 2009) and downregulated in biological models of tumor reversion (Tuynder et al. 2004). Also, this protein is the target of various anticancer drugs (Efferth 2005), and so it could be used as a potential target for therapy (Zhu et al. 2008). Astacins from *L. intermedia* could also be used as starting materials to design new drugs, as agonists and/or inhibitors. The possible therapeutic use of astacins from *L. intermedia* is the vascular diseases (acute myocardial infarction, acute ischemic stroke, thrombosed aortic aneurysms, pulmonary embolism, etc.) and as thrombolytic agents (Chaim et al. 2011). Meprins, which are members of the astacin family, hydrolyze and inactivate several endogenous vasoactive peptides. Recently, it has shown that a meprin inhibitor suppresses the formation of atherosclerotic plaques (Gao et al. 2009). The recombinant astacins could also be used as reagents for laboratorial tests to diagnose Loxoscelism, anti-*loxosceles* serum production, and in the treatment of envenomation (Chaim et al. 2011).

The toxin Tx2–6 from *Phoneutria nigriventer* (Brazilian Wandering spider) venom causes an improvement in the level of nitric oxide in penile tissue in rats, and so it can be investigated as a tool in the treatment of erectile dysfunction (Villanova et al. 2009). Additionally, other peptides were identified in *Cupiennius salei* spider venom with antibacterial properties. These peptides act as channel-forming toxins within the bacteria wall. Therefore, it is expected that analogous synthetic molecules would have great potential, especially in the emergence of multiple-antibiotic-resistant bacteria (Haeberli et al. 2000). The oxyopinins from the wolf spider *Oxyopes kitabensis*, forming pores in lipid membranes (Belokoneva et al. 2003), considered the anti-tumor action of other pore-forming peptides, oxyopinins could also be considered as good candidates for anti-cancer therapy (Shaposhnikova et al. 1997). Gao et al. (2007) have verified the effects of the venom of *Macrothele raven* (Araneae, Hexathelidae) upon the proliferation and cytotoxicity of human cervical carcinoma cells (2005) and on the human breast carcinoma cell line MCF-7. Psalmotoxin 1, a component of the venom of a West Indies tarantula, is a 40-amino acid peptide that inhibits cation currents mediated by acid-sensing ion channels (ASIC) (Escoubas et al. 2000).

Ants and centipedes

Ant venom has also been used to treat diseases especially arthritis. A partially purified extract of the South American tree ant *Pseudomyrmex sp.* venom was tested by Altman et al. (1984) in a case-control study of patients with rheumatoid arthritis. It has been demonstrated that venom-treated patients have an improvement in overall efficacy and a significant reduction in the number of tender/painful joints and swollen joints. Another study showed that solenopsin A, a primary alkaloid from the fire ant *Solenopsis invicta*, exhibits anti-angiogenic activity. Authors investigated the ability solenopsin A for inhibiting a series of kinases involved in the process of angiogenesis. Mouse embryonic fibroblast cell lines (3 T3-L1 and NIH3T3) were used, and solenopsin prevented the activation of PI3K, the phosphorylation of Akt-1 at both Thr308 and Ser473, and the phosphorylation and subsequent subcellular localization of forkhead box O1a (FOXO1a), a physiologic substrate of Akt. The selective inhibition of Akt by solenopsin in vitro is of great interest since few Akt inhibitors have been developed so far, and Akt is a key molecular target in the pharmacological treatment of cancer (Arbiser et al. 2007). An inhibitor of PI3K/Akt, Perifosine is being used in clinical assays to treat patients with advanced stages of cancer (Chee et al. 2007).

Regarding centipede, a study has reported the anti-tumoral action of centipede venom (Sonoda et al. 2008). It was shown that a synthetic compound, Manb β (Fuca(1–3))Glc β 1-Cer, (glycosphingolipid 7), which was identified in the centipede

Parafontaria laminate armigera, had an antiproliferative effect on melanoma cells. The effect of glycosphingolipid 7 on the proliferation of melanoma cells was through suppression of the activation of FAK-Akt and Erk1/2, the phosphorylation of pRb, and the expression of cyclinD1 and CDK4. The authors concluded that Glycosphingolipid 7 might be a candidate as an inhibitor of cell proliferation in melanomas. Moreover, μ -SLPTX-Ssm6a, a unique 46-residue peptide from centipede venom, has a promising effect as a novel analgesic treating a broad range of pain conditions exceeding morphine in rodent pain models (Yang et al. 2013).

Snail

Snail has been used in medicine since ancient times and prepared according to several formulations. The venoms of these snails contain a cocktail of up to 200 pharmacologically active components that mainly target different voltage- and ligand-gated ion channels (Becker and Terlau 2008). The first description of snails as medication was during the Roman Empire when it was used in the mental illness, syncope, vertigo, and infectious diseases (Bonnemain 2005). In the eighteenth century, American investigators have assessed hundreds of neurotoxins derived from sea snails which are utilized by snail to immobilize their prey and induce a neuromuscular blockage. That is why Ziconotide (SNXIII), a synthetic equivalent to a 25-amino-acid peptide derived from snail venom, has been under FDA review since 1999 to be administered intrathecally as an alternative to opioids in cases of severe pain due to its powerful anesthetic effect (Vitale et al. 2008). In twentieth century, a neurotoxin called conotoxin TVIIA, was extracted from *Conus magus*, a fish-eating sea snail and approved for the treatment of intractable pain (Stix 2005; Bingham et al. 2010), and it has been demonstrated to reduce the size of the myocardial infarct in an ischemia/reperfusion model in rabbits, rats, and dogs in vivo (Lubbers et al. 2005).

Beetles (Cantharidin)

Mylabris is the dried body of the Chinese blister *Mylabris phalerata* and *Mylabris cichorii* beetles. Its use as a traditional medicine in China traced back more than 2,000 years (Wang 1989). Dried bodies of beetles are ingredients of traditional Chinese medicine used to treat esophageal cancer, hepatoma, and skin diseases (Efferth et al. 2005). The active constituent of mylabris is cantharidin. Cantharidin is a vesicant found in all body fluids of blister beetles, including *Mylabris phalerata* and *M. cichorii* (Chinese blister beetles) and *Lytta vesicatoria* (Spanish fly) (Ellenhorn 1997). In Asia, cantharidin was used in the treatment of furuncles and piles, ulcers, and tuberculous scrofuloderma (Wang 1989). In Europe, Hippocrates prescribed cantharidin as a treatment for dropsy (Cheng et al.

1990). In South Africa, the Tswanas grind cantharidin-producing beetles into a powder and use it in a medicine called *seletsa* as an aphrodisiac, as an abortifacient, and for purifying the blood (Ellenhorn 1997).

Cantharidin may be a useful candidate to develop novel strategies for cancer therapy. Cantharidin has anti-cancer activity inhibiting human leukemia cell lines growth in vitro (Rauh et al. 2007). Unlike other chemotherapeutic agents, cantharidin acts on leukemia progenitor and stem cells (Dorn et al. 2009). Derivatives of cantharidin also inhibit the growth of prostate, colon, oral, cervical, and gall bladder cancer cell lines (Hill et al. 2007; Liu and Chen 2009). Moreover, Cantharidin stopped the production of P-gp, a membrane transport protein that produces chemotherapeutic drug resistance in a hepatoma cell line (Zheng et al. 2008). More than that, cantharidin has anti-metastatic effect in A549 human lung cancer cells (Kim et al. 2013). There is growing evidence that Norcantharidin (NCTD), a water-soluble synthetic small-molecule derivative of naturally occurring cantharidin from the medicinal insect blister beetle (*Mylabris phalerata* Pallas) is a demethylated analog of cantharidin (Wang 1989). Norcantharidin is capable of chemoprevention and tumor inhibition (Hsieh et al. 2013) and also promotes tumor cell adhesion and metastasis (Chen et al. 2005). Lee et al. (2013) suggest that norcantharidin exhibits anticancer effects against non-small cell lung cancer cells in vitro, and his report supports the potential use of Norcantharidin as a chemotherapeutic agent for treating non-small cell lung cancer. Topical cantharidin in a collodion vehicle has been used by dermatologists in the treatment for warts and molluscum contagiosum since the 1950s (Funt 1961). Moye et al. (2014) has reported that Cantharidin is a safe treatment modality for molluscum contagiosum and should be considered when symptomatic infection necessitates treatment (Kadioglu et al. 2014).

Bees

The therapeutic application of bee venom has been used in traditional Oriental medicine to treat many diseases as arthritis, rheumatism, and pain (Jang et al. 2009). Bee venom possesses radioprotective (Varanda and Tavares 1998), antimutagenic (Varanda et al. 1999), anti-inflammatory (Nam et al. 2003), antinociceptive (Son et al. 2007), and anticancer activities (Moon et al. 2006). The anti-inflammatory effect of bee venom is attributable to suppression of phospholipase A2, free radical production, alpha-1 acid glycoprotein gene expression, and activation of nitrous oxide (Son et al. 2007; Yoon et al. 2008). Other anti-inflammatory mechanisms include suppression leukocyte migration, inhibition of inflammatory gene activation, reduction of cyclooxygenase-2 (COX-2) activation, mRNA expression, decrease of inflammatory cytokines tumor necrosis factor- α (TNF- α) and interleukin-1 β (IL-1 β), and superoxide

production and direct inhibition of NF- κ B (Becker and Terlau 2008; Hama and Sagen 2009).

Regarding its role in treatment in rheumatoid arthritis, a study enrolling rheumatoid arthritis patients showed an additive effect when bee sting was used simultaneously with classical oral drugs such as methotrexate, sulfasalazine, and meloxicam (Liu et al. 2008). An in vitro human study showed that bee venom induced apoptosis of synovial fibroblasts in subjects with rheumatoid arthritis (Zhang et al. 2003). Darwish et al. (2013) have concluded that bee venom potentiates the anti-arthritic effects of methotrexate, possibly by increasing its bioavailability. Also, it provides a potent antinociceptive effect. Furthermore, bee venom protects against methotrexate-induced hepatotoxicity mostly due to its inhibitory effect on TNF- α and NF- κ B.

Depending on the fact that people perceive bee stings as being painful or hyperesthetic, bee venom can be used to influence pain regulation as it induces pain through the activation of spinal neurons (Son et al. 2007). In an experimental study in rats, bee venom application by plantar injection increases a marker of neuronal activity, c-fos gene expression, in nociceptive neurons of the dorsal horn of the spinal cord (Luo et al. 1998). On the other hand, other study showed that bee venom promotes the expression of genes in spinal neurons that down-regulate pain. The acupuncture needle coated with bee venom has been utilized to treat pain (Son et al. 2007). A clinical trial in Korea has been performed using apipuncture to treat arthritis. Subjects had significantly fewer tender and swollen joints and less pain and morning stiffness after therapy (Lee et al. 2005). Another study reported the efficacy and safety of bee venom acupuncture in patients with chronic non-specific low back pain (Seo et al. 2013). In a pilot study, both acupuncture and bee venom acupuncture showed promising results as adjuvant therapies for idiopathic Parkinson's disease (Cho et al. 2012).

Moreover, purified bee venom (PBV) has been used in treating acne vulgaris. The anti-microbial activity of PBV were translated in vivo study which showed that cosmetics containing PBV provided a certain degree of efficacy in terms of lesion counts and skin microorganism concentration compared with cosmetics without PBV in subjects with acne vulgaris. This study proved that PBV could be a good candidate compound for developing therapeutic drug for the treatment of acne vulgaris (Han et al. 2013). Also, bee venom has been used in treatment of polycystic ovary by its anti-inflammatory effect. It decreases levels of IL-6, COX-2, VEGF, and serum androgens and increased number of corpora lutea in Wistar rats (Karimzadeh et al. 2013). Furthermore, Honeybee venom (apitoxin) can be used as therapeutic option to lower blood glucose and lipids in diabetic rats (Mousavi et al. 2012). Another use of bee venom was in the treatment of multiple sclerosis. A preliminary human study showed that subjects treated with venom injection have improvement in neurological examinations with no side effects (Castro et al. 2005).

Considering the anti-cancer activity of bee venom, it is the most studied venom due mainly to two substances that have been isolated and characterized: melittin and phospholipase A2(PLA2) which are derived from the venom of species *Apis mellifera* (Ownby et al. 1997). Melittin is a small amphiphilic peptide containing 26 amino acid residues having the greatest antitumor activity (Gevod and Birdi 1984). Many cancer cell lines, including leukemia, renal, lung, liver, prostate, bladder, and mammary cancer cells, can be targets of melittin (Son et al. 2007). Moreover, Chueng (1982) has shown that melittin have the capability of binding calmodulin, which has a role in cellular proliferation. Melittin have a powerful inhibitory effect on calmodulin activity, and so it is considered as an inhibitor of cell growth and clonogenicity of human and murine leukemic cells (Lee and Hait 1985). Another study by Gest and Salomon (1987) showed that melittin inhibits the melanotropin receptor in M2R melanoma cell membranes. Other mechanism of action of melittin is acting as pore-forming agent-killing malignant cells (Shaposhnikova et al. 1997). Arora et al. (1996) also reported that melittin, as a PLA2 activator, increased the calpain activity and cell necrosis in the hepatocellular carcinoma cell lines N1S1 and McA-RH7777. More recent studies have shown that melittin kills tumor cells by apoptosis through many cancer cell death mechanisms, together with the activation of caspase and matrix metalloproteinases (MMP) (Moon et al. 2006). Kollipara et al. (2014) suggested a new approach that can enhance the anti-cancer effect of bee venom on NSCLC cells at a much lower concentration by co-culture of bee venom with NK-92MI cells through inactivation of NF- κ B.

It is reported that bee venom significantly inhibited mammary carcinoma metastasis in mice (Orsolić et al. 2003), exhibited morphological changes typical of apoptotic cells on NCI-H1299 lung cancer cells Jang et al. (2003), inhibits proliferation and induces apoptosis of SMMC-7721 human hepatoma cells Hu et al. (2006), decreased the viability of human lymphoma cell line HL-60 and human lymphocytes after 24 h (Lee et al. 2007), induces apoptosis in human breast cancer MCF-7 cells (Ip et al. 2008a) and human cervical cancer Ca Ski cells (Ip et al. 2008b), potentiates the apoptotic effects of TRAIL (TNF-related apoptosis-inducing ligand) on human hepatocellular carcinoma (HCC) cells and may be a promising therapeutic approach in the treatment of TRAIL-resistant human cancer (Wang et al. 2009).

Caterpillar

Caterpillars are the larval form of members of the order Lepidoptera (the insect order comprising butterflies and moths) (Hossler 2009). The first isolation of caterpillar peptide was from the hemolymph of the giant silk moth, *Hyalophora cecropia*. This peptide named Cecropins and proved to have antimicrobial activity (Andreu et al. 1985).

Cecropin exert strong antibiotic activity against both Gram-positive and Gram-negative bacteria in micromolar concentrations (Boman et al. 1991). Besides their antimicrobial activity, recent studies have demonstrated that Cecropin have anti-cancer activity against multiple tumor cell lines including mammalian leukemia, lymphoma, and colon carcinoma cell lines (Chen et al. 1997), more to the point, small cell lung cancer (Shin et al. 1997) and gastric cancer cells (Chan et al. 1998).

Also, Cecropins A and B (two well characterized members of the Cecropin-family) exert selective cytotoxic and antiproliferative efficacy in bladder cancer cells while sparing targets of benign murine or human fibroblast origin. Both peptides may offer novel therapeutic strategies for the treatment of bladder cancer with limited cytotoxic effects on benign cells, and thus, this is considered as promising novel therapeutic strategies for the treatment of bladder cancer with limited cytotoxic effects on benign cells (Suttmann et al. 2008). In vivo, Cecropin B improves survival of mice with ascitic colon adenocarcinomas (Moore et al. 1994). Transfection of human bladder cancer cells with Cecropin genes reduces their tumorigenicity in nude mouse models (Winder et al. 1998). Both cecropins A and B inhibit the viability and proliferation of bladder cancer cells, but with no effect on fibroblasts (Suttmann et al. 2008). Thus, these novel peptides offer new therapeutic strategies for the treatment of bladder cancer with limited cytotoxic effects on benign cells. Moreover, many active components from the venom of *L. oblique* have been isolated and characterized, including fibrinogenases (Pinto et al. 2006), hyaluronidases (Gouveia et al. 2005), a phospholipase A2 (Seibert et al. 2006), a factor X activator (Alvarez Flores et al. 2006), a prothrombin activator (Reis et al. 2001), and an antiapoptotic protein (Souza et al. 2005). Hyaluronidases, named lonogliases, have been identified from *L. oblique* venom (Gouveia et al. 2005). These hyaluronidases have been reported to affect cancer cell growth as well as tumor invasion; thus, they bear a potential as tools in studies of cancer cell biology (Matsushita and Okabi 2001) and in the pharmaceutical industry (Smith et al. 1997).

Lonomia obliqua caterpillar bristle extract (LOCBE) induces in vitro calcium-dependent pro-coagulant activity both in human and rat plasma (Kelen et al. 1995). It may be due to the activation of prothrombin and factor X, with no effects on platelets (Donato et al. 1998). In vivo, it causes blood incoagulability in laboratory animals (Reis et al. 1999). LOCBE proved also to be effective in preventing experimental venous thrombosis in rats, justifying further studies using purified fractions of the extract to clarify the mechanisms of this effect (Prezoto et al. 2002).

Wasp

Wasp stings cause severe pain and tissue damage and may be fatal (Argiolas and Pisano 1984). Although wasp stings may cause serious health problems, studies have paid attention to

its bioactive compounds such as biogenic amines, peptides, and proteins (Nakajima et al. 1986). Mastoparan is a 14-amino-acid amphipathic peptide obtained from wasp venom (Zimmerberg and Parsegian 1986). It have a wide spectrum of pharmacological activities including mast cell degranulation (Hirai et al. 1979), activation of G-protein-mediated mechanisms (Ozaki et al. 1990), inhibition of calmodulin-mediated mechanisms (Wheeler-Jones et al. 1992), stimulation of phospholipases A₂ and C (Wallace and Carter 1989), and stimulation or inhibition of cation-specific channels (Glavinovic et al. 1992). Mastoparan have been reported to have anti-cancer effect by inducing potent mitochondrial permeability transition in the concentration range between 5 and 100 mM, by forming a permeability transition pore (Pfeiffer et al. 1995). Based on this finding, Yamada et al. (2005) have encapsulated mastoparan with a transferrin-modified liposome with a pH-sensitive fusogenic peptide (GALA) for selective delivery to mitochondria in K562 cells—human chronic myelogenous leukemia. The encapsulated mastoparan was able to release cytochrome C in the cell line studied, indicating its potential use as an anticancer agent. Souza et al. (2009) isolated two novel mastoparan peptides, Polybia-MP-II and Polybia-MP-III, from venom of *Polybia paulista* wasp, which exhibited hemolytic activity on erythrocytes. Also, Wang et al. (2008) have reported that Polybia-MPI have anti-tumor activity. Polybia-MPI is able to target non polar lipid cell membranes, forming ion permeable channels, and leading to depolarization, irreversible cytolysis, and cell death (Matsuzaki et al. 1997).

Conclusion

Insect-based medicine has been long used in traditional medicine and is coming nowadays under increasing interest in biomedicine. Even though research into the science behind using helminthes and insects as therapies is proliferating, challenges and opportunities are still enormous. Moreover, bioactive molecules found in insect's venoms can exert important pharmacological activities and will contribute extensively in Biomedical Sciences.

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