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Venom: the sharp end of pain therapeutics

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Abstract

Adequate pain control is still a significant challenge and largely unmet medical need in the 21st century. With many small molecules failing to reach required levels of potency and selectivity, drug discovery is once again turning to nature to replenish pain therapeutic pipelines. Venomous animals are frequently stereotyped as inflictors of pain and distress and have historically been vilified by mankind. Yet, ironically, the very venoms that cause pain when directly injected by the host animal may actually turn out to contain the next generation of analgesics when injected by the clinician. The last 12 months have seen dramatic discoveries of analgesic tools within venoms. Spiders, snakes and even centipedes are yielding peptides with immense therapeutic potential. Significant advances are also taking place in delivery methods that can improve bioavailability and pharmacokinetics of these exciting natural resources. Turning proteinaceous venom into pharmaceutical liquid gold is the goal of venomics and the focus of this article.

Keywords

Nociception, pain, venom, toxin, ion channel, analgesic

Introduction

The evolution of animal defence mechanisms against predators has evoked many painful experiences. These can be classified into three types: physical – such as claws, spines and quills, chemical – where toxins are either synthesised or sequestered, and both – where toxins are coupled with active delivery mechanisms such as hollow or grooved spines and fangs. Toxin mixtures that have coevolved with an active delivery mechanism (gland and injection system) are termed 'venoms'.

Nociception is a powerful tool for initiating learned avoidance behaviours, which over time develop into collective knowledge and eventually instinctive behaviours. The net result is increased survival of the species and thus generation of a significant selection pressure. Physical means of defence, such as spines and quills, make the possessor appear more imposing but also produce short-term nociceptive responses in those animals that get too close. The lion cub that gets porcupine quills impaled in its nose quickly learns to avoid porcupines. Many fish species possess spines that, when erected, prevent them being swallowed by their predators. But it appears that this was not enough to improve the survival rates of many species, and venom glands connected to such hollow or grooved spines evolved in multiple fish species. The selection pressure for venom apparatus is so strong that it has evolved many times independently and in many species. This is exemplified in the catfish where there are understood to be over 1250 venomous species and multiple convergent evolution events that have produced venoms that cause pain and distress in predatory fish.[1] Defensive venoms have evolved in many other fish, such as lion fish (*Pterosis* sp.), weaver fish (*Trachinidae* sp.) and stingrays (*Dasyatidae* sp.). In terrestrial

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animals, however, venom is most often associated with the feeding apparatus (fangs, stings and harpoons) where dual selection pressures exist – predator defence and prey capture. Venom as a method of efficient prey capture has evolved multiple times in distinct classes and phyla (not just in terrestrial species) and can be grossly divided into two mechanisms: cytotoxic and neurotoxic. Both mechanisms cause pain, but it is the latter which is of most interest to pain research as the toxins directly interact with neuronal signalling. Neurotoxins are effective tools, enabling predators to capture prey where the habitat makes chasing injured prey difficult (underwater and in trees) or where the predator is pursuing prey much stronger than itself such as cnidarians and cone snails. These are both soft-bodied aquatic invertebrates that prey on fish, which if not subdued quickly could easily break free and damage the predator. Such potent and specific venoms are incredibly valuable to neuroscientists and many other researchers. Drug discovery programmes can learn a lot from natural selection.

Pain target classes

Most pharmacologically useful proteins are represented as target classes for pain therapeutics such as G-protein-coupled receptors (GPCRs), enzymes, ion channels and even growth factors. GPCRs are the most common target for existing therapies with ion channels as a close second.[2] This review will focus on the pain target classes most relevant to venom research. One of the first and most used analgesics, morphine, has long been known to act on opioid receptor GPCR, but significant side effects still blight its use, and pharmacological challenges are still being overcome to improve these most prescribed analgesics. Enzyme inhibitors such as those inhibiting cyclooxygenase (COX) enzymes also have a long history of clinical use. Continual work in this field has refined these non-steroidal anti-inflammatory drugs (NSAIDs) by reducing COX-1 activity and focusing on the pain-relevant COX-2 enzyme pathway.

The rise of biological drugs such as humanised monoclonal antibodies has enabled targeting of non-classical drug targets such as growth factors. Tanezumab is one such antibody that has shown great promise both pre-clinically and clinically, although the programme has recently hit unexpected safety concerns. The US Food and Drug Administration (FDA) has halted all anti-nerve growth factor (NGF) antibody trials except those for cancer pain. This is due to apparent worsening of osteoarthritis in knee joints of phase III patients, and is potentially due to overuse of the joint through significant reduction in pain.[3] This demonstrates the importance of pain as a protective mechanism and the unexpected danger of analgesics reducing pain's protective effect.

But despite this range of target classes to choose from, it is the ion channels that receive the most interest despite being potentially the most difficult to modulate. Voltage-gated sodium channels (VGSCs) and transient receptor potential (TRP) channels are crucial therapeutic targets in the treatment of pain,[2] which form critical components of the nociceptive sensory pathway.

Of the nine voltage-gated sodium (Na_V) channel isoforms found in humans, three ($Na_V1.7$, 1.8 and 1.9) are of particular interest to pain researchers. VGSCs are large integral membrane proteins made up of alpha and beta sub-units. The alpha unit forms a pore and consists of four domains (DI–DIV) each with six transmembrane segments (S1–S6) (Figure 1). Segments S1–S4 form voltage sensor modules and S5–S6 form the channel pore. The S4 segments are rich in positive arginine residues that sense membrane depolarisation and move outward to induce channel gating. A combination of studies conclude that the S4 segments in DI–DIII are determinants of channel activation, and DIV is principally involved in channel inactivation.[4]

$Na_V1.7$ is involved in nociception in humans and rodents[5] and is a major contributor to pain signalling (and therefore an important target for the development of specific sodium channel inhibitors).[6] In humans, gain-of-function mutations in sodium channel neuroendocrine type nine alpha (SCN9A), which encodes $Na_V1.7$, leads to severe neuropathic pain, whereas loss-of-function mutations in this gene lead to indifference to pain.[7] Individuals with this mutation have been known to place knives through their arms and be able to walk on burning coal without feeling pain. Without the pain mechanism, there is no warning of actual or potential injury.

$Na_V1.8$ is expressed in dorsal root ganglion (DRG) neurons and peripheral nerve axons and is therefore a useful target in pain patients. Two gain-of-function mutations in SCN10A, which encode $Na_V1.8$ (found in patients with painful neuropathy) enhance the channel's response to depolarisation and produce hyperexcitability in DRG neurons.[8] $Na_V1.9$ is also a desirable drug target (as shown using mouse models) and is also expressed in DRG neurons but has proved difficult to functionally express in heterologous systems and is therefore challenging to study.[9]

TRPV1 (a member of the TRP superfamily of excitatory ion channels that bind vanilloids) is predominantly expressed by nociceptors and is therefore also a popular pain relief target,[10] but in clinical trials, drugs inhibiting the TRPV1 channel cause hyperthermia and decreased sensitivity to painful levels of heat. To overcome these life-threatening side effects, Fischer et al.,[11]

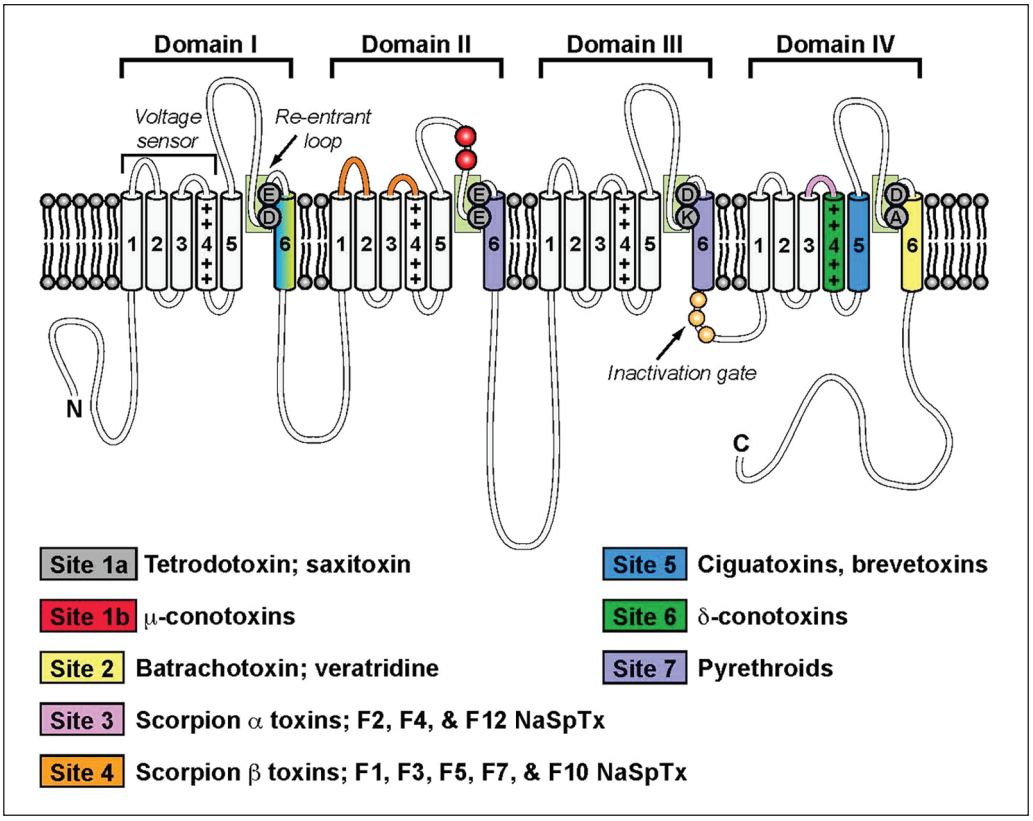

Figure 1. Schematic binding sites for sodium channel toxins. The Na_v channel architecture shown here demonstrates the four domains, their six transmembrane domains and intracellular (bottom) and extracellular (top) structures. One of the key intracellular structures (indicated by orange spheres) is the inactivation gate. Superimposed on this diagram are the binding sites of key toxin tools.
Figure reproduced from Klint JK, Senff S, Rupasinghe DB, et al. Spider-venom peptides that target voltage-gated sodium channels: Pharmacological tools and potential therapeutic leads. Toxicon 2012; 60:478–491 with permission from Elsevier.

developed tools to block phosphorylation of TRPV1 instead of blocking the channel: this is effective through the disruption of TRPV1 interaction with A-kinase anchoring protein 79 (AKAP79). The inhibition of phosphorylation reduces inflammatory hyperalgesia and could be a promising new therapeutic route for targeting pain through TRPV1.

Topical capsaicin is also marketed as an analgesic patch, that acts through desensitisation of the TRPV1 expressing nociceptors. This novel mechanism utilises the pharmacological activity of ligand-dependant receptor internalisation as well as other related effects, the result is defunctionalisation of the nociceptors and thus reduced pain perception.[12] However, 8% capsaicin patches need to be applied after administration of local anaesthetics to reduce the burning pain of TRPV1 activation prior to desensitisation.

Also located in DRGs are the acid-sensing ion channels (ASICs), which are also involved in nociception, are voltage independent and are sodium selective. ASICs 1–4 form homo- or heterotrimers, and different ASIC subtypes exist, which have roles in the central and peripheral pain pathways making them potential targets for therapeutic intervention with toxin tools.[13–15]

Pain and venoms

At the time of writing, there were 4356 articles deposited in PubMed[16] containing the search term 'pain' and the terms 'venom' or 'toxin' since 1947. Performing a search in this way does not differentiate the description of envenomation as a painful experience from venoms being used to treat pain. However, the first nine articles from 1947 onwards all depict the use of venom as an analgesic. These early articles focus on cobra venoms with a few mentions of bee venom. Both of these venoms are held in high regard in Asian traditional medicine. Believed to hold mystic powers (due to the profound effect it has on the human body), venom was also historically understood to contain cobric acid as its toxic component. The presence of cobric acid was disproved in 1886, and the toxic component correctly identified as a protein. It is the proteins and peptides in venoms that are of the most

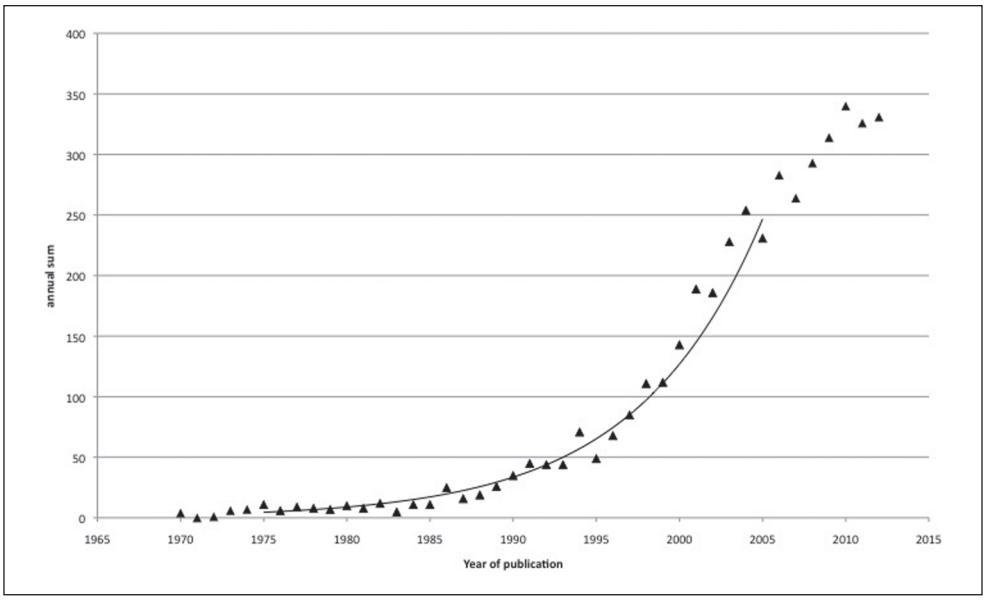

Figure 2. Venom and pain publication rate. Number of articles deposited in PubMed per year from 1970 to 2012 collated by the search term 'pain AND (venom OR toxin)'.

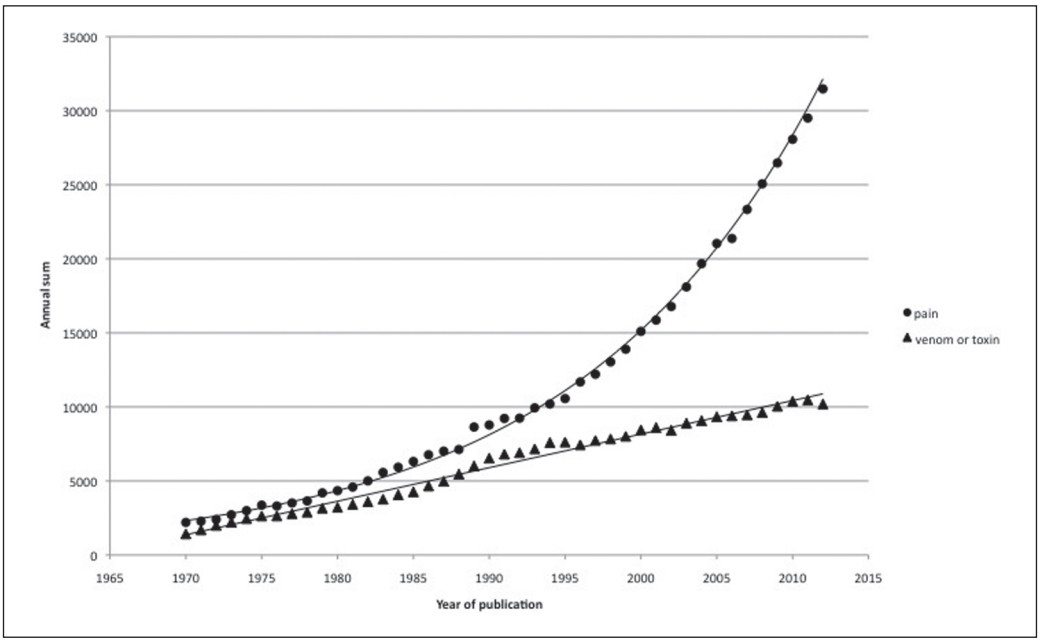

Figure 3. Venom and pain publication rate displayed separately. Number of articles deposited in PubMed per year from 1970 to 2012 showing separate search terms 'pain' (black circles) and 'venom OR toxin' (black triangles).

interest as biological tools and potential therapies. From 1970 to 2004, there was an exponential increase in publication rate of articles focusing on both venom and/or toxin, and pain (Figure 2). Post 2004, the publication trend has moved away from the exponential rate seen for all pain articles and taken on the linear increase seen in the venom and toxin field (Figure 3). This early exponential publication rate was not seen in other fields such as oncology and antimicrobial research (data not shown) where the publication rate has displayed a linear increase in line with the general publication rate for venoms and toxins. Therefore, it is reasonable to suggest that the rate of discovery and research of the use of venoms in the pain field is limited by the publication rate in this field. This in turn is potentially limited by the discovery and supply of new

Figure 4. The Trinidad chevron theraphosid (*Psalmopoeus cambridgei*) resting on a banana leaf, demonstrating the phenotype of these pharmacologically important spiders. Source: Photo by S Trim.

venoms and methods to investigate their utility. Although the rate of deposition of new toxins in the Tox-Prot database appears to be exponential for scorpions and cone snails between 1967 and 2005,[17] other species have not featured quite so significantly.

Another significant issue with the rapid increase in discovery of therapeutically useful toxins was the lack of standard nomenclature. This has, however, been addressed by King et al.[17] and is now being widely taken up. Essentially, the system consists of three parts: a broad activity descriptor with subscript to confer target identity, a generic toxin family name suffixed by single letter genus and species (upper and lower case letters, respectively) with numeral and letter to differentiate toxins with similar pharmacology.[17] Thus, psalmotoxin-1, an ASIC blocking toxin from the Trinidad chevron spider[18] (Figure 4), is correctly referred to as π-theraphotoxin-Pc1a, where π (pi) denotes ASIC activity and theraphotoxin-Pc1a identifies it as the first theraphosid toxin from *Psalmopoeus cambridgei* (Figure 4) with this activity as stated on ArachnoServer.[19] This system has already been adopted for conotoxins and integrated for other venomous species. Many venoms act on the vertebrate nervous system pre- and post-synaptically primarily to disable prey. These toxins are key to our understanding of pain and neuronal signalling as well as providing potential therapeutic leads. Figure 5 displays these synaptic sites of action.[20]

Pain-relevant species

Theraphosids. Commonly called tarantulas even though the original tarantula is a wolf spider (*Lycosa sp.* tarantula). The authors, therefore, propose to adopt the less ambiguous term 'theraphosids' for large hirsute spiders of the family *Theraphosidae*. Figure 4 illustrates the typical phenotype of theraphosid spiders. This term was proposed at the British and Irish Association of Zoos and Aquariums Terrestrial Invertebrate Working Group (BIAZA TIWG) by Mark Bushell, Assistant Curator of Invertebrates, Bristol Zoo Gardens (UK) (July 2012, personal communication). Although not formally adopted by the committee, it was largely agreed as the accurate term to use. This nomenclature has already been taken up in the ArachnoServer database[19] with the adoption of the theraphotoxin nomenclature. Venoms from theraphosids were first studied in the early 1970s with a trickle of articles published annually until 2002. Indeed, 2002 saw publication of several articles on theraphosid venoms acting on ion channels, but it was 2004 with the publication of 'Tarantulas: eight-legged pharmacists and combinatorial chemists'[21] that really heralded a surge in research on theraphosid venoms with submissions rising to nine articles per year on average. In 2004, Escoubas and Rash[21] described tarantula (now theraphosid) venom peptides as having a bimodal size distribution with peaks at 4–4.5 kDa and 6.5–7 kDa and tabulated a list of 31 published toxins. In just under 10 years, there are now nearly 500 published theraphosid toxins in Tox-Prot,[22] and the distribution of published masses is quaternary with many larger peptides now identified (Figure 6). The majority of these larger peptides are described from two Asian theraphosid species, *Chilobrachys jingzhao*[23] and *Haplopelma hainanum*.[24] The excitement is focused around one class of protein, the disulphide directed β-hairpin (DDH), which is thought to have been the evolutionary precursor to the inhibitor cystine knot (ICK) peptide.[21] These ICK peptides comprise six cystine residues in three disulphide pairs resulting in the knotted loop motifs with the length of these loops being a key variable between species.

ICK toxins ProTx-II (β/ω-theraphotoxin-Tp2a) and huwentoxin-IV (HWTX-IV, μ-theraphotoxin-Hh2a) bind multiple sodium channel types but are more than 100-fold selective for human $Na_V1.7$.[25] For example, ProTx-II (β/ω-theraphotoxin-Tp2a) is ~50-fold more selective for $Na_V1.7$ than $Na_V1.5$.[26]

Huwentoxin-I (μ/ω-theraphotoxin-Hh1a) and huwentoxin-IV (μ-theraphotoxin-Hh2a) from the tarantula *Ornithoctonus huwena* are potent inhibitors of $Na_V1.7$, IC_{50} ~26 nM[25] and other neuronal tetrodotoxin (TTX)-sensitive channels. These toxins bind to segments three and four of transmembrane domain two (DII) demonstrated by the exchange of two residues in the DII S3–S4 linker of $Na_V1.7$, and $Na_V1.4$ reverses the affinity of huwentoxins to these channels.[4]

Apart from action on voltage-gated sodium channels, theraphosid toxins also exhibit activity at ligand-gated ion channels such as TRPV1. TRPV1 is stably

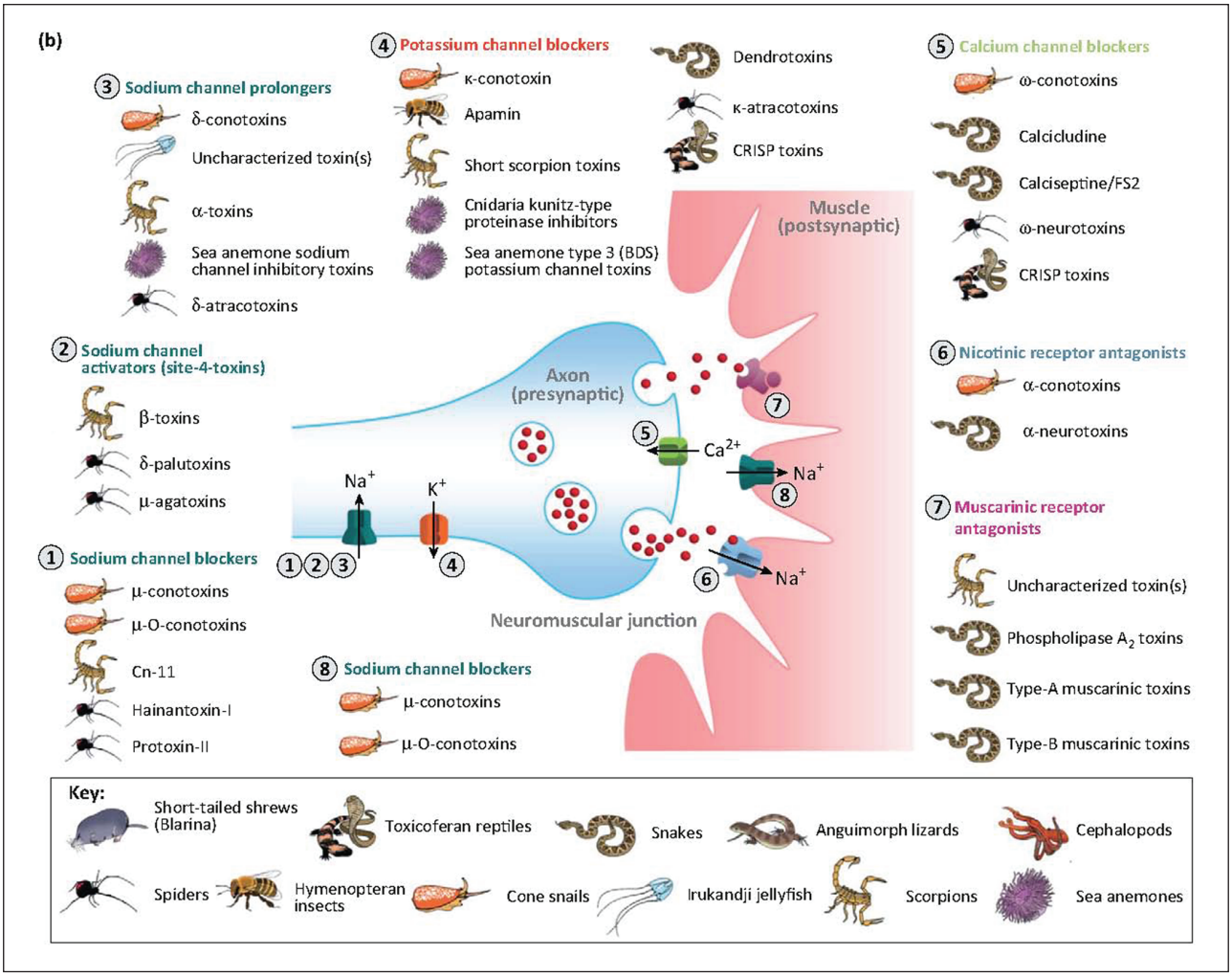

Figure 5. Pre- and post-synaptic neurotoxin binding sites. Diagrammatic representation of the wealth of toxin tools known to bind ion channels and receptors pre- and post-synaptically.
Figure reproduced from Casewell NR, Wüster W, Vonk FJ, et al. Complex cocktails: the evolutionary novelty of venoms. Trends Ecol Evol 2013; 28: 219–229 with permission from Elsevier.

activated by DkTx, (not found in ArachnoServer) a double-knot toxin from *Ornithoctonus huwena*, by targeting the outer pore domain.[27]

The aforementioned π-theraphotoxin-Pc1a is a potent (IC_{50} 0.9 nM) homomeric ASIC1a blocking peptide.[18] At the time, this was the most potent and selective tool for this important class of ion channels involved in pain signalling.

Other arthropods. Many venomous arthropods such as bees, wasps and ants (*Hymenoptera*), true spiders (*Aranaeomorpha*), centipedes (*Chilopoda*) and scorpions (*Scorpionidae*) possess toxins that act on ion channels. Scorpion toxins are voltage sensor modifiers of sodium channels, and alpha-scorpion toxins hinder fast inactivation by interacting with the DIV S3–S4 linker to stabilise DIV S4 in the closed state. Beta-scorpion toxins enhance channel activation by binding to the DII S3–S4 linker, trapping the DII S4 in the activated state[28] (Figure 1). Envenomation by centipedes has long been known to cause significant pain, but, until recently, their pharmacology has been understudied. Recently 26 neurotoxin-like peptides were discovered from a single species.[29] These peptides contain novel representatives from 10 different peptide families and were demonstrated to have in vitro potency at voltage-gated potassium (K_v), sodium (Na_v) and calcium (Ca_v) channels.[29] Although at this stage, the majority of the Na_v activity was at tetrodotoxin-sensitive channels, this was only from one species, and it highlights the untapped potential of venomous arthropods.

In an email from Professor Glenn King,[30] he described discovery of a novel potent (IC_{50} of ~25 nM) and selective (>150 fold, except $Na_v1.2$, 32 fold) human $Na_v1.7$ blocking peptide from centipede venom. This peptide has no significant homology to any previously characterised peptide or protein and

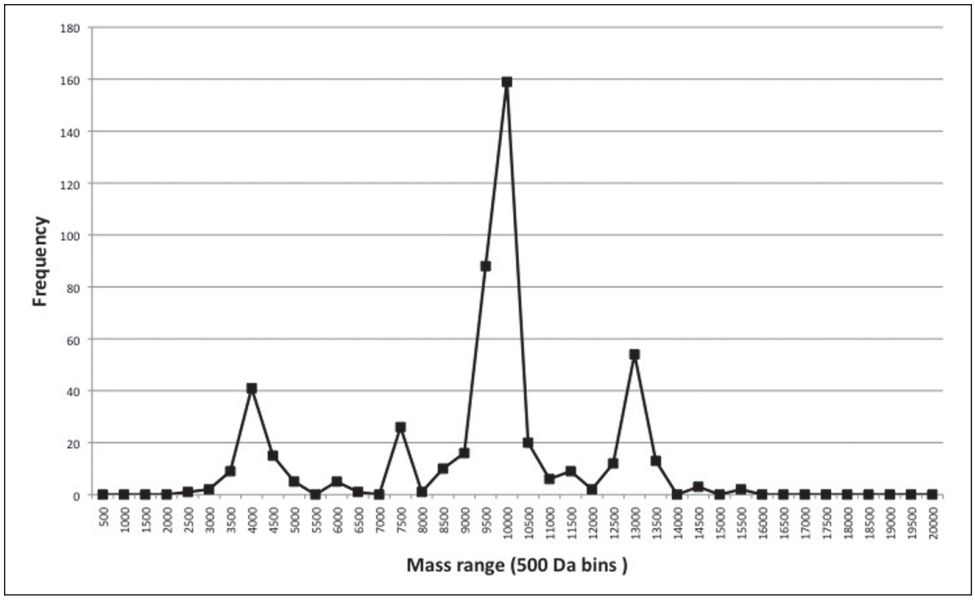

Figure 6. Quaternary distribution of theraphosid venom peptide sizes as deposited in UniProt, under the animal toxin annotation programme Tox-Prot.

displays greater potency than morphine in rodent pain models (publication under review). It is discoveries like these that are paving the way for a new generation of potent analgesics.

Cnidarians. Sea anemones, jellyfish and corals that make up this group have long been of interest principally to understand marine envenomation pathology and, more recently, to look for novel ion channel tools. All cnidarians catch prey through toxin-induced immobilisation. Their venoms are delivered through specialised stinging cells called nematocysts. This mechanism enables soft-bodied predators to prey upon powerful, often armoured arthropods and vertebrates that would otherwise damage their soft bodies. Cnidarian venom has two major functions: blockade of ion channels such as voltage-gated sodium and potassium channels, and ASICs and cell lysis to aid digestion through phospholipase enzymes called cytolysins.[31] To date, approximately 200 toxins have been identified from cnidarians, and the International Society on Toxinology has published guidance on nomenclature protocol, which is still to be fully adopted for the naming of such toxins.[32] Cnidarian venoms are a rich source of novel analgesic tools where only a small percentage of the available species have been characterised. p-AITX-Ael2b, initially called APETx-2, from the anemone *Anthopleura elegantissima* is another tool useful for improving our understanding of ASIC pharmacology as it targets the heterotrimeric ASIC3 channels.[13]

Molluscs. Another group of soft-bodied invertebrates that have evolved complex venom systems to allow predation of vertebrate prey, the cone snails (*Conus* sp.) are also a significant source of valuable ion channel tools for pain research. This review will not cover this area in significant depth due to the wealth of reviews already available on conotoxins and pain.[33–35] However, cone snails have yielded the first toxin-derived analgesic approved for clinical use – Prialt® (Ziconotide, Elan Pharmaceuticals).[36] This 25 amino acid peptide is a synthetic form of the natural snail venom that blocks N-type calcium channels in the dorsal horn of the spinal cord. Due to low blood brain barrier penetration, the peptide needs to be delivered intrathecally. However, novel methods in peptide cyclisation have lead to oral availability of such peptides.[37] Another lead peptide from these animals has been identified by the CONCO Project.[38] The project (lead by Atheris Laboratories, CEO Reto Stöcklin) has identified peptide XEP-018 with in vivo proof of concept success as a novel analgesic and myorelaxant, and is expected to enter clinical trials for pain control and local anaesthesia.[38]

Preliminary investigations into another group of predatory marine snails (*Gemmula* sp.) have also embraced a venomics approach. So new is this field, only three articles relating to *Gemmula* sp. venom have been submitted to PubMed as of April 2013. Currently, the potential is clear from the rapidly diverging toxin superfamily, which is expected to produce several new ion channel tools structurally different from the cone snails.[39] Although no toxin function has been published to date, these soft-bodied snails are using venoms to catch fish thus there is a strong possibility that they will contain novel ion channel tools and analgesic lead material.

Snakes. Cobratoxin – a long chain alpha neurotoxin from the Thailand (sic) cobra (*Naja* sp.) – has had mixed usage in the field of pain from its inclusion in traditional medicine and over-the-counter homeopathic remedies to examining its analgesic effects in rodent models. It has long been recognised that the venom from elapid snakes such as cobras, mambas, kraits and coral snakes contains complex neurotoxins which primarily block nicotinic acetylcholine receptors: this causes the diagnostic flaccid paralysis exhibited by envenomation victims. Although nicotinic acetylcholine receptors are of potential therapeutic benefit in pain and many other neurological conditions, there are comprehensive reviews in this area.[40] Many of these elapid venoms and, perhaps somewhat surprisingly, those from rattlesnakes (*Crotalinae* sp.) also possess analgesic effects through opioid and non-opioid mechanisms.[41] This is in contrast to the previous pathological dogma that vipers, such as rattlesnakes, only produced cytotoxic and haemotoxic effects on envenomation and were not thought to contain neurotoxins. Activities of opioid-like peptides such as crotalphine are of significant interest as they are orally active, although the reason for this resistance to digestion is unclear.[42] Venom from the coral snake *Micrurus lemniscatus* is also an orally active anti-nociceptive where, again, the exact mechanism of action and the pharmacokinetics remain to be determined.[41] Crotoxin, a phospholipase A2 toxin from the rattlesnake *Crotalus durissus terrificus* used in cancer patients as an anti-tumour agent, also reduces analgesic intake through a non-opioid mechanism.[43] The effects of Crotoxin are currently understood to be through central muscarinic receptors and 5-lipoxygenase-derived mediators. Rodent data support this clinical observation as this toxin produces a significant reduction in nociceptive pain post neurectomy.[43] It is clear that snake venoms have not given up all their secrets, and novel analgesics are not far away.

Recently, novel analgesic activity has been discovered in a three-finger toxin from the black mamba (*Dendroaspis polylepis*). These new toxins, mambalgins,[14] bind to ASIC1a homomeric channels and ASIC1a-containing heteromeric channels in an ASIC1a-dependent fashion. These peptides produce analgesic effects centrally through ASIC1a/ASIC2a heteromers and peripherally through ASIC1a/1b heteromers, and unlike π-theraphotoxin-Pc1a, this effect is not attenuated by naloxone (opioid antagonist). Diochot et al.[14] report isolation of two mambalgin peptides differing in a single amino acid at position four from the same venom. Preliminary work at Venomtech Ltd indicates that mambalgin-2 is not present in all animals of this species, *Dendroaspis polylepis* (unpublished).

Other venomous species. Many other venomous species are rapidly being discovered and therefore hold a potential for new therapeutic tools. The refinement of experimental techniques has allowed investigation of these novel tools and improved understanding of the host species. Casewell and colleagues compiled a review of the evolution of venom systems in animals which highlights global diversity of a whole range of species from mammals to lizards and fish to echinoderms and worms.[20] Currently, the main connection with pain and these venoms is that they have evolved not only to immobilise and kill prey but also as a defence mechanism – and it is a mechanism that causes significant pain in humans. Since the discovery that the male platypus (*Ornithorhynchus anatinus*) possesses a venomous spur, it has been known that this venom induces significant and intense pain in human victims and appears to have evolved for intraspecific conflict.[20]

Toxins to drugs

The term 'venomics' has been applied to the wide diversity of techniques used to understand venom components. The Institute of Molecular Bioscience at the University of Queensland (Australia) published a comprehensive review on venomic and drug discovery from natural sources.[44] Their review covers the major steps in using venom-based libraries for drug discovery screening through recombinant expression and synthetic production. Developing toxins from drug discovery in vitro leading into licensed pharmaceuticals has been a tough challenge as demonstrated by the clinical pipeline: the majority of venom-derived drugs approved for use are non-peptide derivatives of the original toxins. These projects have used the venom-derived toxin as a tool to elucidate mechanisms and for proof-of-concept studies where they are replaced with a small molecule pharmaceutical to improve their drug-like characteristics. Peptides generally have extremely poor oral bioavailability due to digestion in the stomach, and when injected, they still have a poor circulating half-life ranging from minutes to a few hours due to high clearance. Significant advances in peptide chemistry are resolving these problems in novel ways such as cyclisation,[45] which offers oral bioavailability. Another method to improve the half-life of peptides from minutes to months is through PEGylation, whereby locking the peptide in non-circulating polyethylene glycol (PEG) scaffold delivers tuneable clearance rates.[46] Overcoming some of the previous hurdles for peptide-derived therapeutic intervention will undoubtedly unlock the full potential of venom-derived toxins.

Ethical sourcing of novel drug leads from natural biodiversity is not mentioned often enough in articles describing potential drug leads. The Convention for Biological Diversity (CBD) outlines guidance for companies wishing to seek genetic resources from another country. However, it states that *ex situ* resources derived before signing of the treaty are exempt.[47] A significant advance in the ethical treatment of venomous animals has come from the Alistair Reid Venom Research Unit (Liverpool, UK) where the isolation of intact RNA transcripts of venom toxins has been demonstrated from crude snake venom.[48] Previously, after identification of novel active compounds in venom, the venom glands were terminally dissected from the venomous animals in order to clone the toxin genes. This technique of RNA isolation from venom will dramatically reduce the supply of venom gland tissue required and have a positive impact on the support of biodiversity and maximisation of drug discovery pipelines.

Conclusion

Since the latter part of the 20th century, many significant advances have deepened our understanding of venoms and pain. With modern '-omics' technologies, and synthetic chemistry, we are in the golden age of biological drug discovery especially for pain research. Millions of years of evolution have honed venoms with exquisite potency and selectivity to make any chemist jealous, and we now have the technology to maximise this potential.

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Conflict of interest

Steven A Trim is Managing Director of Venomtech Ltd a commercial company that produces venoms for research. Carol M Trim has no conflict of interest to declare.

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