Experimental Drugs for Neuropathic Pain

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Abstract: Background: Neuropathic pain (NP) is an important public health problem and despite recent progress in understanding, diagnosis, pathophysiological mechanisms and the treatment of NP, many patients remain refractory to pharmacotherapy.

Objective: Currently used drugs have limited efficacy and dose-limiting adverse effects, and thus there is a substantial need for further development of novel medications for its treatment. Alternatively, drugs approved for use in diseases other than NP can be applied as experimental for NP conditions. This paper covers advances in the field of NP treatment.

Results: The prime focus of this paper is on drugs with well-established pharmacological activity whose current therapeutic applications are distinct from NP. These drugs could be a potential novel treatment of NP. Data from preclinical studies and clinical trials on these experimental drugs are presented. The development of advanced methods of genomics enabled to propose new targets for drugs which could be effective in the NP treatment.

Conclusion: Experimental drugs for NP can be a treatment option which should be tailor-made for each individual on the basis of pain features, previous therapies, associated clinical conditions, recurrence of pain, adverse effects, contraindications and patients’ preferences. At present, there are only some agents which may have potential as novel treatments. Increasing knowledge about mechanisms underlying NP, mechanisms of drug action, as well as available data from preclinical and clinical studies make botulinum toxin A, minocycline, ambroxol, statins and PPAR agonists (ATx086001) promising potential future treatment options.

Keywords: Neuropathic pain, adrenergic β1 receptors agonists, histamine H1 receptors, TRPA1 channels, T-type calcium channels, p38 mitogen-activated protein kinase, 3-hydroxy-3-methyl glutaryl coenzyme A reductase, reactive oxygen species.

1. INTRODUCTION

As defined by the International Association for the Study of Pain (IASP), neuropathic pain (NP) is the pain caused by a lesion or disease affecting the somatosensory nervous system [1]. This pain type is a debilitating condition which is a significant burden to individuals and society worldwide [2], being a substantial cause for patients’ disability, decreased quality of life and reduced productivity [3, 4]. The estimated prevalence of NP in general population ranges between 1.5% and 6.9%, and is mainly associated with middle age (50-64 years), manual professions, and living in rural areas [5].

Available data indicate that 40-50% of patients with NP do not receive appropriate treatment, which is due to low diagnostic efficacy, ineffectiveness of available analgesics and insufficient knowledge about the pathophysiology of NP [3-5]. Bearing that in mind, not only new recommendations for treatment protocols [3], but also exploration of novel drug targets for agents relieving NP are a strong medical demand [6, 7].

Impaired sensation which is typical for NP reflects and is a consequence of pathological phenomena that occur within the nervous system – both in the periphery (nerves, plexus, roots and sensitive lymph nodes) and at the central nervous system (CNS: spinal cord and brain) level. As such, NP can be triggered by a variety of pathological situations which are able to alter physiological somatosensory functions of the nervous system. These pathologies are responsible for tissue-specific symptoms of NP and they comprise: viral infections (Herpes simplex, Varicella zoster, HIV), metabolic impairments (e.g. diabetes) accompanied by mitochondrial dysfunctions, stroke, mechanical injuries to the CNS, or peripheral nerves [8-10] and adverse (toxic) effects of drugs, in
particular some anti-cancer drugs (e.g. oxaliplatin, vincristine, bortezomib, taxanes) [10-12].

Recently published results from preclinical research have provided insights into NP mechanisms and they offered detailed knowledge about the molecular basis of this pain type [13, 14]. These mechanisms are strictly correlated with maladaptive alterations within the central and peripheral nervous system (Fig. 1) which comprise: (i) nociceptor sensitization and hyperexcitability caused by the release of pronociceptive neurotransmitters within afferent nerve fibers, (ii) increased ectopic excitability/firing of affected neurons, (iii) facilitation of pronociceptive stimuli at the dorsal horn of the spinal cord level, (iv) nociceptive disinhibition of the spinal inhibitory network, and (v) central sensitization of pain defined as increased responsiveness of nociceptive neurons in the CNS to normal or subthreshold afferent inputs which results from repeated and persistent painful stimulation [9, 15, 16].

These abovementioned phenomena are thought to be strongly associated with molecular plastic changes in the somatosensory nervous system – in NP overexpression of voltage-gated ion channels, algogen-sensitive receptors and altered synthesis of selected neurotransmitters are observed. In NP, nociceptor activation is one of the most relevant peripheral mechanisms underlying the development of pain sensitization and subsequent fiber density changes, neuronal hyperexcitability, hyperalgesia and allodynia. These receptors are located at free nerve endings of unmyelinated C fibers and myelinated Aδ fibers and they can be activated or sensitized by metabolic damage, various mechanical, thermal and chemical stimuli, being sensitive to a variety of endogenous substances, such as: substance P (SP), bradykinin, serotonin, calcitonin gene-related peptide (CGRP), prostaglandins, excitatory amino acids (glutamate), histamine, growth factors and proinflammatory cytokines [13, 14, 16]. In addition, a pivotal role of abnormal ectopic excitability of afferent neurons has been demonstrated in NP. This phenomenon is mainly responsible for the development of positive symptoms in NP - paraesthesias and dysesthesias, and is thought to be mediated by voltage-gated sodium channels (mainly Na$_v$1.7, 1.8 and 1.9) and voltage-gated calcium channels (Ca$_v$) overexpressed on the sensory nerves (C, Aδ, Aβ fibers) and overactivated in NP conditions [17].

Sensory C and Aδ fibers which are strongly implicated in NP pathophysiology terminate at two distinct types of spinal dorsal column neurons, i.e. spinal projection neurons which innervate higher neuronal structures, and interneurons which modify synaptic transmission at the spinal cord level. Here, pronociceptive facilitation is mainly mediated by glutamate – a main excitatory neurotransmitter within the CNS and pain pathways [14], and spinal glial cell activation [18]. Glutamate is an endogenous ligand of three types of glutamate receptors that are implicated in the transmission of pain signals from the periphery to the brain: ionotropic AMPA and NMDA receptors and metabotropic (mGluR) receptors. These proteins are responsible for basic response to painful stimuli (AMPA receptors), amplification and prolongation of painful input signals in the spinal dorsal horn (NMDA receptors), as well as enhancement of synaptic transmission and

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**Fig. (1).** Maladaptive alterations in NP within the central and peripheral nervous system. (The color version of the figure is available in the electronic copy of the article).
neuronal discharge via the activation of intracellular signaling cascades, thus contributing to synaptic plasticity and long-term potentiation [16]. Taken together, these mechanisms result in the increased excitability of CNS neurons and phenotypic and functional switch of Aβ fibers (‘Aβ sprout’). The latter phenomenon is due to extended receptive fields of peripheral sensory fibers, which alters the function of Aβ fibers from previously mediating innocuous stimuli to fibers transmitting mechanical stimuli and participating in the development of allodynia and hyperalgesia [8, 16].

The activity of the dorsal horn neurons projecting towards the brain plays an important role in pain modulation and pain perception. This activity is strongly influenced by the descending inhibitory noradrenergic and serotonergic, as well as an opioid, GABAergic and cannabinoid pathways that stem from the periaqueductal gray, raphe nuclei, locus coeruleus and rostral ventral medulla [19]. Both serotonergic and noradrenergic projection neurons from these regions can modulate (i.e. inhibit or facilitate) pain, whereas spinal dorsal horn GABA and glycine neurotransmitting systems are responsible for inhibitory effects. This inhibitory synaptic transmission of GABA and glycine is reduced or completely lost in NP [16] and its restoration might provide analgesia [20, 21], while the disinhibition of nociceptive input at the spinal level contributes to pain signal facilitation and increased pain sensitivity [22].

Taken together, a better understanding of the contribution of the abovementioned mechanisms and elucidation of their role in NP remains an important scientific challenge as it may provide clues to more efficacious treatments for pharmacoresistant NP. So far, several novel drug targets have been discovered for antiallodynic and antihyperalgesic agents. This review is focused on several investigational drugs that have been recently tested in preclinical models of NP and in clinical trials in patients suffering from NP. The prime focus of this paper is on drugs approved for use in diseases other than NP for which they are still considered experimental.

2. DRUG TARGETS FOR EXPERIMENTAL ANTIALLODYNIC AND ANTIHYPERALGESIC AGENTS

Many pathologies involved in the etiology of NP make this pain type complex and resistant to available pharmacotherapy. On the other hand, these mechanisms involved in NP allow for a therapeutic interference in various neurotransmitter, ion channel and receptor systems to alleviate NP symptoms. These interventions involve interdisciplinary multimodal strategies to obtain pain relief, including pharmacotherapy, complementary alternative medicine, cognitive-behavioral therapy and surgical procedures [7, 16]. Despite this, a significant proportion of patients is not treated optimally and there is an unmet clinical need to search for novel efficacious and safe antiallodynic and antihyperalgesic agents. In the recent years, potential new targets for experimental analgesics have been explored. These include:

- Adrenergic system: β2 adrenoceptors
- Angiotensin AT2 receptors
- Histaminergic system: H3 receptors
- Cholinergic system
- Transient Receptor Potential A1 channels (TRPA1)
- T-type voltage-gated calcium channels
- Voltage-gated sodium channels
- Endocrine system
- Peroxisome proliferator-activated receptors and cytokine/chemokine receptors
- Reactive oxygen species
- p38 mitogen-activated protein kinase
- 3-hydroxy-3-methyl glutaryl coenzyme A reductase

2.1. Adrenergic β2 Adrenoceptors

Apart from their wide distribution in the cardiovascular and respiratory systems, β2 adrenoceptors are expressed within pain pathways. Although these receptors do not participate in the regulation of mechanical nociceptive threshold in physiological conditions, recently it has been shown that they are important for the antiallodynic action of antidepressant drugs (tricyclic antidepressants, serotonin-noradrenaline and selective noradrenaline re-uptake inhibitors) used in NP [23-25]. It was suggested that noradrenaline increased by the action of antidepressant drugs on reuptake transporters acts through β2 adrenoceptors. Potential mechanism of the antiallodynic action of β2 receptor stimulation seems to be related to their expression by non-neuronal satellite cells within the dorsal horn of the spinal cord. This location of β2 receptors in pain pathways can be a potential molecular and neuro-anatomical target for antidepressants in NP.

Antidepressants (nortriptyline, venlafaxine) and β2 adrenoceptor agonist (terbutaline) were effective in a mouse model of neuropathic pain induced by cuffing the sciatic nerve. This antiallodynic effect involved a peripheral nervous system and was due to β2 adrenoceptor stimulation which resulted in the inhibition of neuropathy-induced tumor necrosis factor α (TNFα) production. TNFα is a pro-inflammatory cytokine that participates in the development of allodynia in NP by influencing nociceptor sensitivity via intercellular signaling [26].

Noteworthy, the action of tricyclic antidepressants in NP is faster as compared to their effect on mood [27]. Chronic but not acute treatment with the antidepressant drug - nortriptyline and the β2 adrenoceptor agonist – terbutaline, alleviated tactile allodynia in obese leptin-deficient mice (ob/ob) which are regarded a genetic model of human type 2 diabetes useful for studies on diabetes and diabetic polyneuropathy [28]. It has been demonstrated that β2 adrenoceptors and δ opioid receptors seem important to these drugs’ antiallodynic action and a possible use of β2 adrenoceptor agonists to reduce NP in diabetic patients without contraindication for glycemic control was suggested. At present, the mechanism of the antiallodynic effect is not fully understood, as yet there is no evidence that both β2 and δ receptors may be co-expressed by the same cells in the pain pathways, or such
Chronic treatment with $\beta_2$-agonists (clenbuterol, formoterol, metaproterenol and procaterol) (Fig. 2) suppressed alldynia in neuropathic mice with the therapeutic onset similar to that reported for antidepressant drugs [23, 25]. Similar results were obtained by Choucair-Jaafar et al. [29] for terbutaline (Fig. 3) used chronically at clinically effective doses against asthma and no serious change in cardiac parameters was observed at antiallodynic doses. Taken together these results indicate that $\beta_2$-agonists may offer an alternative to currently available drugs for the treatment of NP [29].

This potential application of $\beta_2$-agonists has been recently supported by a case study with salbutamol (Fig. 3) used chronically in 6 patients with pharmacoresistant NP but these results require further confirmation in randomized, controlled and blind studies [23]. A clinical trial evaluating the analgesic effect of the $\beta_2$-agonist, i.e. terbutaline sustained release 5 mg in post-thoracoscopy NP (Betapain), has already started but the results are not available, yet [30]. $\beta_2$-agonists were also effective in patients with NP secondary to thoracotomy. In addition, this study revealed a pain-type specificity of $\beta_2$ receptors showing that non-NP syndromes, such as for example musculoskeletal pain can be alleviated by low doses of non-selective $\beta$ receptor antagonists [24]. A positive effect of $\beta_2$-agonists on post-thoracotomy NP seems to be related to their inhibitory effect on the glial production of TNF$\alpha$ shown previously [26].

2.2. Angiotensin AT$_2$ Receptors

Angiotensin II (Ang II) is an octapeptide that regulates blood pressure and fluid balance via two G protein-coupled receptors, namely AT$_1$ and AT$_2$. Contraction of smooth muscles leading to vasoconstriction and increase in blood pressure, increase in water and sodium intake, renal sodium reten- tion and secretion of vasopressin and aldosterone are mediated by the stimulation of AT$_1$ receptors, while the role of AT$_2$ receptors is not so well understood. It is suggested that the stimulation of AT$_2$ receptors may oppose the biological effects of AT$_1$ receptors – in particular their effects on cell growth, blood pressure and fluid intake.

Apart from the role within the circulatory renin-angiotensin system (RAS), Ang II is a neurohormone and a neuromodulator [31] in the local RAS in various body tissues, including the brain [31, 32] and the spinal cord [32, 33]. Recent evidence showing the co-localization of Ang II with substance P, nerve growth factor and CGRP in neurons of the dorsal root ganglion points out for its role in nociception [34, 35].

Preclinical studies using cultured human and rat dorsal root ganglia cells demonstrated the expression of AT$_2$ receptors in sensory neurons. In addition, abundant distribution of AT$_2$ receptors was demonstrated in the brain and viscera of adult mice and on small- to medium-sized dorsal root ganglia neurons in adult rats and humans. In humans, AT$_1$ receptors are expressed on peripheral nerve fibers, in the skin, urinary bladder and bowel [36]. In vitro studies showed that Ang II and AT$_2$ receptors regulate neuronal membrane excitability and induce neurite outgrowth. This latter effect is caused by a persistent activation of p44/p42 mitogen-activated protein kinase (MAPK). The stimulation of MAPK activates multiple receptors and ion channels that participate in the development of NP [37].

Available literature indicates that AT$_2$ receptor antagonists are efficacious in rodent models of NP [38]. Several AT$_2$ receptor antagonists with >1,000-fold selectivity over AT$_1$ receptor have been developed as potential novel analgesics for alleviation of NP: EMA200 (also referred to as PD 123319), EMA300 (PD 121981), EMA400 (PD 126055) and EMA401 ([S]-enantiomer of EMA400) [38, 39] (Fig. 4).

Their analgesic efficacy was proven in several preclinical models of NP [31, 35, 38] and inflammatory pain [18, 31]. The high analgesic potency without significant CNS distribution or serious CNS-related adverse effects in rats selected.

Fig. (2). Structures of clenbuterol, formoterol, metaproterenol and procaterol.

Fig. (3). Structures of terbutaline and salbutamol.
EMA401 as a lead compound for further clinical investigations on its antinociceptive efficacy in NP [40]. Several mechanisms underlying this antiallodynic efficacy of EMA401 have been hypothesized, including the blockade of Ang II/AT$_2$ receptor signaling pathway in the peripheral somatosensory system and EMA401-induced decrease in p38 MAPK and ERK activation in human dorsal root ganglia sensory neurons [37]. AT$_2$ receptor antagonists including EMA401 may thus act on paracrine/autocrine mechanisms at peripheral nerve terminals, or intracrine mechanisms, to reduce neuropathic pain signalling in Ang II/NGF/TRPV1-convergent pathways.

EMA401 has been tested in a phase II programme in humans as a novel analgesic drug for the treatment of NP-related to postherpetic neuralgia (PHN) and diabetic NP [38-40], but these studies have been withdrawn, recently [41].

2.3. Histamine H$_3$ Receptors

G protein-coupled histamine H$_3$ receptors are involved in numerous biological functions such as sleep-wake rhythms. They have been identified as mainly presynaptic autoreceptors in the histaminergic neurons of the CNS which regulate the release of histamine, as well as heteroreceptors on non-histaminergic neurons regulating the release of other neurotransmitters. H$_3$ receptors are thought to play a role in several CNS disorders, i.e., attention-deficient hyperactivity disorder, epilepsy, cognitive deficits and obesity. Being localized in many regions of the CNS which are involved in nociception (e.g. dorsal horn of the spinal cord, locus coeruleus), recently H$_3$ receptors have been linked to pain [42].

H$_3$ receptors are involved in central sensitization of pain. H$_3$ receptor blockade reduces spontaneous neuronal firing and decreases responses of spinal nociceptive neurons to mechanical stimulation. Previously, it was reported that this effect is mainly mediated via supraspinal sites, including locus coeruleus whose activity modulates the functions of spinal neurons [43].

The potential role of H$_3$ receptors in nociception has been studied in preclinical models of NP using various H$_3$ receptor agonists and antagonists [43, 44]. The available results suggest that although these agents reduce allodynia and hyperalgesia [43], their efficacy strongly depends on the site of administration. Hence, the clinical role of H$_3$ antagonists [44] and inverse agonists [43] is questionable and conflicting. On the other hand, inverse agonists of H$_3$ receptors: GSK189254 and GSK334429 (Fig. 5) showed efficacy in animal models of surgically- and virally-induced NP [43-45] pointing out a possible application for these short-acting and poorly brain-penetrating ligands.

In a study by Hsieh et al. [42], intraperitoneally administered GSK189254 attenuated central sensitization of pain and reduced tactile allodynia in the rat spinal nerve ligation model of NP. This effect was strongly associated with the activation of the noradrenergic system at the spinal cord level. Safety and efficacy of GSK189254 were investigated and compared to duloxetine in a phase I study to demonstrate that the electrical hyperalgesia model can be extrapolated from animals to patients. This trial has been completed but the results are not available, yet [46].
2.4. Cholinergic System

The idea to use cholinergic drugs to achieve pain relief is not novel and is supported by numerous preclinical and clinical studies which demonstrated that acetylcholinesterase inhibitors (AChEIs) administered intrathecally provided analgesia in response to noxious cold in healthy volunteers and in postoperative pain. Moreover, several cholinesterase inhibitors were efficacious in animal models of NP [47].

Donepezil is an orally available AChEI with improved tolerability and safety profile as compared to older AChEIs [47]. Recently, it has been investigated as adjunctive therapy with gabapentin in the management of NP. The results obtained suggest that donepezil may provide benefits when added to patients with NP who do not receive sufficient pain relief from gabapentin used as monotherapy. Of note, this combination may provide additional benefits for patients who experience troublesome adverse effects from gabapentin treatment, enabling gabapentin dose reduction [47]. At present, donepezil (Fig. 6) 5 mg once daily for 6 weeks is being investigated in a randomized phase 4 clinical trial for its efficacy in NP (diabetic neuropathy, failed back syndrome with neuropathic symptoms). The estimated completion date for this trial was December 2016 [48].

Botulinum toxin A is a potent neurotoxin derived from the anaerobic bacterium Clostridium botulinum. There are several subtypes of botulinum toxin, but the most commonly used is the subtype A. It acts primarily as a muscle paralytic agent by inhibiting the presynaptic release of acetylcholine from motor neurons at the neuromuscular junction. Botulinum toxin also has effects on the afferent nerves by decreasing sensory response of the afferent C-fibers [49].

This potent neurotoxin cleaves the 25-kDa synaptosomal-associated protein (SNAP-25), which is anchored to the cell membrane. This cleavage results in the formation of a non-functional soluble N-ethylmaleimide-sensitive factor attachment receptor (SNARE) complex which blocks synaptic transmission and inhibits neurotransmitter and neuropeptide release. A retrograde transport and transcytosis of botulinum toxin A partially responsible for the analgesic effect of this agent has been also suggested. The latter mechanism enables the action of botulinum toxin A not only at the injection site but also at distant areas that project to the infused region [50].

Analgesic properties of botulinum toxin A both in animal models and humans are well known. The administration of botulinum toxin A attenuated NP in chronic constriction injury (CCI)-exposed rats and restored the neuroimmune balance. It reduced the levels of upregulated after injury IL-1β and IL-18 in the spinal cord and dorsal root ganglia and increased the levels of the antinoceptive cytokines, such as IL-10 and IL-1RA. The changes observed at the dorsal root ganglia level suppressed microglial activation within the spinal cord. Therefore, it has been suggested that botulinum toxin A, similarly to minocycline, can influence microglia and monocytes. Intraplantar injection of botulinum toxin A diminished the CCI-mediated increase in the levels of IL-18 in dorsal root ganglia and IL-1β in both the spinal cord and dorsal root ganglia. Additionally, botulinum toxin A injection simultaneously increased the levels of the antinoceptive interleukins IL-1RA and IL-10 in the dorsal root ganglia of CCI-exposed rats compared to control animals [50].

Mechanisms underlying a beneficial effect of botulinum toxin A in NP are still not fully explored. It has been suggested that this neurotoxin: (i) cleaves neuronal SNARE, (ii) inhibits the release of pain mediators (CGRP) from the peripheral nerve terminals, dorsal root ganglia, and spinal cord neurons, (iii) reduces TRPV1 protein expression, (iv) reduces inflammation by decreasing the release of peripheral neurotransmitters and pro-inflammatory mediators, (v) deactivates sodium channels [51]. Botulinum toxin A showed efficacy in pain related to nociceptor sensitization, ectopic firing, and central sensitization and these mechanisms might be important for botulinum toxin A-mediated analgesia in NP [52].

Clinical applications of botulinum toxin A are still increasing. There is level A evidence for established efficacy of botulinum toxin A injection in PHN; level B evidence for probable efficacy of botulinum toxin A injection in trigeminal neuralgia and post-traumatic neuralgia; level C evidence for possible efficacy of botulinum toxin A injection in diabetic polyneuropathy; and level U (insufficient) evidence for botulinum toxin A injection in complex regional pain syndrome (CRPS), phantom limb and stump pain, and occipital neuralgia. There is also some concern regarding unwanted neuromuscular adverse effects, especially weakness [53, 54].

The impact of botulinum toxin A on trigeminal neuralgia has been reviewed thoroughly in [51]. It has been also suggested that botulinum toxin A might be a promising alterna-
tive treatment option for refractory cases of trigeminal neuralgia [55].

The efficacy of 50 U botulinum toxin A (intramuscular injection) in trigeminal neuralgia accompanied by severe, episodic pain in the lower left gingival area was also confirmed. Interestingly, a good therapeutic effect lasted for about 5 months [56]. Botulinum toxin A reduced pain severity and attack frequency in trigeminal neuralgia when injected to the maxillary and mandibular roots [57]. In trigeminal neuralgia, it was efficacious and well-tolerated. It should be however noted that the mechanism underlying its analgesic effect in this type of NP may not be strictly related to the cholinergic system [58], but it rather involves the inhibition of the release of pronociceptive neurotransmitters (e.g. SP, CGRP, glutamate) [59].

In PHN botulinum toxin A-treated group exhibited a significant reduction of pain intensity scores and improved sleep quality compared with control groups [51].

In post-surgical neuralgia, 20–190 U of botulinum toxin A was injected intradermally. A significant improvement in postoperative pain, and a reduction of postoperative analgesic use was observed [51].

Two randomized, double-blind, placebo-controlled studies used botulinum toxin A for pain control in diabetic neuropathy. In this type of NP, botulinum toxin A provided pain relief [51].

Botulinum toxin A (50 U) was also tested in the occipital neuralgia. Although pain scores were significantly reduced, it was concluded that these studies were insufficiently reliable to prove its full effectiveness in this pain type [51].

There is also a small clinical report regarding two patients with CRPS who experienced a reduction of pain after intramuscular administration of botulinum toxin A. However, it should be emphasized that the effect of botulinum toxin A in CRPS patients has not been reliably proven [51].

Patients with phantom limb pain were also treated with botulinum toxin A. At present, the effect of botulinum toxin A on phantom limb pain cannot be verified as the results were inconclusive and therefore not reliable [51].

In patients with the spinal cord injury-induced NP, the use of botulinum toxin A provided promising results [51]. In these subjects, a single dose of subcutaneous botulinum toxin A provided analgesia. Two administrations of botulinum toxin A 12 weeks apart significantly reduced pain intensity over 24 weeks compared with placebo [52]. It was suggested that botulinum toxin A may reduce intractable chronic NP in patients with the spinal cord injury [60]. Subcutaneous injection of type A botulinum toxin was effective and relatively well-tolerated in NP related to traumatic spinal cord injury [60]. Botulinum toxin A may be also effective in patients with central post-stroke pain [51].

2.5. TRPA1 Channels

In the mammalian organisms, TRPA1 are strongly involved in chemonociception, being sensitive to numerous chemical (e.g. reactive oxygen species, formalin, bacterial products, allicin, isothyocyanates) and physical (UV light) stimulators [61]. Since TRPA1 channels are widely expressed in nociceptors and in Schwann cells, they are regarded as important sensors of various pain types, including NP. Recent studies on the role of TRPA1 in NP have shown that TRPA1 silencing in Schwann cells reduces both allodynia and neuroinflammation in neuropathic mice [62].

One of the main functions of TRPA1 within pain pathways is thought to be related to its contribution to mechanotransduction and noxious cold sensation [61]. These activities of TRPA1 make this channel particularly important in NP-related to chemotherapy-induced peripheral neuropathy (CIPN) [11, 12] and diabetes [63, 64]. In diabetic animals, it has been demonstrated that the blockade of TRPA1 channels attenuates the development of mechanical hypersensitivity and in the advanced phase of this disease TRPA1 might mediate the loss of SP-expressing fibers causing alterations in tactile sensitivity, and TRPA1 inhibitors could restore their function [63]. As mentioned, TRPA1 is strongly implicated in CIPN caused by oxaliplatin, paclitaxel, or vincristine [63, 64]. Recently, it has been demonstrated that oxaliplatin-induced neuropathies are related to altered expression of TRPA1 channels which serve as sensors of reactive oxygen species generated during oxaliplatin administration. An increased expression of TRPA1 and other members of TRP channel family has been shown in dorsal root ganglia neurons at an early stage following oxaliplatin administration, and it has been suggested that these changes may contribute to the development of oxaliplatin-induced NP [65, 66]. This increased expression of TRPA1 via the activation of p38 MAPK in small-diameter dorsal root ganglia neurons might at least in part contribute to the development of cold hyperalgesia and cold allodynia induced by oxaliplatin. Hence, TRPA1 antagonists have been widely tested as potential lead compounds and drug candidates for the treatment of CIPN [67].

A novel TRPA1 antagonist and a derivative of lipoic acid, ADM_09 (Fig. 7), reverted oxaliplatin-induced NP in rats without causing adverse effects. Of note, ADM_09 had lower antioxidant capacities than the parent compound [68]. In preclinical research that involved paclitaxel-induced peripheral neuropathy HC-030031, a TRPA1 channel antagonist, attenuated tactile allodynia in the von Frey test in mice [64].

2.6. T-type Calcium Channels

Impaired function of T-type calcium channels is linked to epilepsy but recently, these channels have also emerged as a potential novel target for the treatment of NP [69, 70]. It has been demonstrated that their activation contributes to nociceptive signaling as it facilitates action potential bursting and modulates membrane potentials of hyperexcitable neurons. The role of T-type Ca²⁺ channels in chronic pain is also proven by gene knockdown experiments showing that their decreased expression attenuates neuropathic pain in the CCI model in rats [71].

Currently, an antiepileptic drug, a T-type calcium channel blocker, ethosuximide (Fig. 8), is assessed in phase 2 clinical trial for its efficacy in traumatic NP and the estimated primary completion date of this trial is April 2018 [72].
2.8. Endocrine System

Apart from its key role in the regulation of circadian rhythms, sleep and mood [76], melatonin (Fig. 10), a hormone synthesized in the pineal gland, modulates pain. This effect results from the activation of melatonin MT1 receptors, inhibition of 5-lipoxygenase and cyclooxygenase expression, indirect activation of opioid receptors by opening potassium channels and antioxidant activity [77]. Melatonin attenuated NP in animal models [76-78] by reducing thermal hyperalgesia and this effect involved nitric oxide synthase, as well as μ and δ opioid receptors [77].

Melatonin showed neuroprotective properties in NP induced by oxaliplatin administration. *In vitro*, in oxaliplatin-exposed neuro-2a cells, it prevented the loss of mitochondrial membrane potential and it stimulated neuritogenesis without affecting cytotoxic activity of oxaliplatin in human colon cancer HT-29 cell line. Melatonin significantly attenuated oxaliplatin-induced pain behavior and it maintained physiological density of epidermal nerve fibers in oxaliplatin-treated neuropathic rats. It reduced nitro-oxidative stress caused by oxaliplatin and prevented nitrosylation of proteins. Apart from this, melatonin significantly improved functions of the mitochondrial electron transport chain and maintained optimal ATP levels in cells. These protective effects of melatonin resulted from its autophagy-inducing and mitoprotective properties within peripheral nerves and dorsal root ganglia [79].

Data from human studies indicate that melatonin is effective in patients with fibromyalgia [77]. In patients receiving melatonin during chemotherapy treatment with taxanes and cisplatin a decreased incidence of neuropathy was noted. This effect was attributed to its anti-inflammatory and antioxidative properties [78].

Recently, clinical trials with a non-selective MT1/MT2 receptor agonist, ramelteon (8 mg) in patients with NP have been terminated because recruitment did not accrue as expected [80]. Piromelatine, a novel melatonin MT1/MT2 and serotonin 5-HT1A receptor agonist, with pain-related P2X3, TRPV1 and Na+1.7 channel-inhibition properties, exerted antinociceptive effects in the partial sciatic nerve ligation model in mice via melatonin receptors, opioid and 5-HT1A receptors. It also improved sleep and was devoid of motor-impairing properties. It was concluded that piromelatine may serve as an effective analgesic drug able to improve sleep disturbances in patients with NP accompanied by insomnia [81].

Endocrinopathies could be present in patients treated for NP using various drugs (e.g., opioids, antidepressants and antiepileptics), thus leading to serious impairments of their quality of life. In view of this, the restoration of the physiological endocrine balance seems to play a key role. The status of hypothalamic-pituitary-gonado-adrenal axis with a potential collapse in hormone concentration in patients with NP is now assessed in clinical trials. Additionally, a study evaluating analgesic efficacy of intrathecal oxytocin (100 micrograms) in patients with NP is currently recruiting patients [82].

2.9. Peroxisome Proliferator-activated Receptors and Cytokines/Chemokine Receptors

Cytokines and chemokines have immunomodulatory properties, being released in response to tissue injury, or
Chemokines play a key role in diabetic neuropathy. Recently, it has been shown that some chemokines such as CCL3, CCL2, CCL4, and CCL5 are also implicated in the development of neuropathic pain. The role of CCL1/CCR8 neuronal ligand 1 (CCL1)-chemokine and CC motif receptor 8 (CCR8) interaction and the role of CCL1/CCR8 neuronal signaling in the development of diabetic neuropathy. Previously, it has been reported that intrathecal injection of recombinant CCL1 in naïve mice induced hypersensitivity and resulted in the activation of microglia to release pronociceptive cytokines, such as IL-1β and IL-6. Recent studies have also implicated CCL1/CCR8 in the development of remifentanil-induced hyperalgesia. It has been also postulated that chemokines such as CCL3, CCL2, CCL4, and CCL5 are important for opioid analgesia. Hence, it was concluded that the spinal CCL1/CCR8 axis might be not only a potential target for the treatment of NP but also a novel therapeutic target for the development of drugs used in the treatment of opioid-induced hyperalgesia [86].

A number of cytokine and chemokine receptor antagonists have been tested for their potential use in NP [84]. AZD2423 (Fig. 11) is a potent and selective inhibitor of chemokine CCR2 receptors which are overexpressed in the spinal cord after nerve injury. This compound was efficacious in a rat model of NP. It reversed hyperalgesia due to CCI but two phase 2a clinical trials with AZD2423 (20 mg or 150 mg once daily for 28 days) in patients with post-traumatic or diabetic NP failed to show its efficacy against primary endpoints [84].

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Fig. (10). Structures of melatonin, ramelteon and piromelatine.

Fig. (11). Structures of AZD2423, minocycline, rosiglitazone, pioglitazone and ATX08-001.
Minocycline (Fig. 11), the inhibitor of matrix metalloproteinase 3, an enzyme controlling the integrity of blood-brain barrier, myelin protein turnover and phenotypic remodeling of glia and neurons, is currently being investigated for prevention and treatment of CIPN and pain caused by nerve damage [50]. Minocycline prevented paclitaxel-evoked allodynia and was well-tolerated. Hence, it might be a potential agent for preventing paclitaxel-induced NP and improving patients' outcome [81].

The repeated administration of minocycline – a microglial inhibitor and a chemokine CCR2 receptor antagonist (RS504393) attenuated pain symptoms in neuropathic rats subjected to CCI of the sciatic nerve. This activity was associated with a decreased spinal microglia activation and the protein level of CCL2 and CCR2 [87].

In the spinal nerve-injured rats at chronic stages of NP, a role of not only spinal but also brain microglia/macrophages in the anti-inflammatory and analgesic actions of minocycline was demonstrated [88]. In the same model of NP in mice, it has been shown that minocycline alleviated NP through actin binding and this effect was due to its modulatory effect on long non-coding RNAs expression in the spinal cord of these neuropathic animals [89].

In microglia primary cells cultures minocycline was able to downregulate lipopolysaccharide-stimulated expression of both CCL2 and CCR2 proteins. In astroglia, primary cell cultures minocycline, reduced the expression of CCL2. These results showed that modulation of CCL2/CCR2 pathway by minocycline might be an efficacious approach to attenuate the development of NP [87].

The role of activated microglia in NP is well-established and relatively well documented by the beneficial effects of minocycline which attenuated allodynia, hyperalgesia, prevented the up-regulation of pro-inflammatory proteins and increased the levels of anti-inflammatory molecules in diabetic NP. Zychowska et al. [85] investigated the role of chemokine-C-motif ligand (XCL) subfamily in diabetic neuropathy. They demonstrated that the XCL1/XCR1 signaling pathway plays an important role in diabetic neuropathy and the prevention of microglial activation by the use of minocycline strongly affects XCL1/XCR1 levels. These authors showed that minocycline not only caused analgesia in a mouse model of diabetic NP but also prevented microglial activation and influenced immune factors. Minocycline inhibited the up-regulation of XCL1 and XCR1. Its administration in diabetic NP models increased the levels of antiinflammatory interleukins (IL-1α, -2, and -10) and diminished the up-regulation of TNFa, IL-1β, and inducible nitric oxide synthase (iNOS) [85, 90]. These data are in line with clinical findings showing that minocycline is efficacious in diabetic neuropathy, being safe and well tolerated. It has high analgesic potential and seems to be an efficient analgesic agent for the treatment of diabetic NP [85, 91, 92] and possibly for its prevention [90].

In CCI-exposed rats minocycline was also able to potentiate antihyperalgesic and antiallodynic effects of ceftriaxone against thermal: heat [93], cold and mechanical stimuli [94]. Both drugs diminished the levels of TNFa, IL-1β [94].

Available literature suggests that there is a link between chemokines in NP and the activation of peroxisome proliferator-activated receptors (PPARs). PPARs are a family of nuclear receptors which act as lipid-activated transcription factors. This family consists of three different isoforms: PPARα, PPARβ/δ, and PPARγ [84]. It has been demonstrated that ligands that bind PPARs inhibit the expression of inflammatory genes in a process called transrepression which is mediated by targeting specific transcription factors, including NF-κB, AP-1 and STAT [84]. Interestingly, experimental evidence suggests a connection between pain-relieving effects of PPAR agonists and suppression of inflammatory gene expression. Among genes that undergo PPAR-induced transrepression there are those of chemokines linked to NP (MCP-1, RANTES, MIP-1α, fractalkine, SDF-1) and their receptors [84]. In cultured monocytes, PPARγ agonists were also able to decrease levels of pro-inflammatory cytokines, such as: TNFa, IL-6, and IL-1β [95].

The PPARγ subtype is expressed both in neurons and glial cells and its stimulation is correlated to the protection of neuronal tissue under oxidative stress conditions. Rosiglitazone – a PPARγ agonist improved catalase activity in the nervous tissue of oxaliplatin-treated rats and prevented oxidative stress in the spinal cord. The restoration of proper redox balance contributed to analgesia in oxaliplatin-treated rats tested in the cold plate test. This effect was observed at a low dose (3 mg/kg) but not at a high dose (10 mg kg), so it was concluded that a mild PPARγ stimulation is necessary for the attenuation of NP [96].

Results of clinical trials suggest that PPAR agonists can effectively alleviate NP, even if it is resistant to other therapies. The mechanisms underlying this activity are complex. Data from animal studies suggest that the analgesic effect of PPARα agonists (fibrates) and PPARγ agonists (rosiglitazone, pioglitazone) (Fig. 11) results from their action at targets in the pain pathways via PPAR-dependent mechanisms, their influence on gene transcription and alteration in the expression of pro-inflammatory mediators, including chemokines and their receptors [84]. Recently, positive results for a novel PPAR agonist, ATX08-001 (Fig. 11), have been shown in patients with PHN. ATX08-001 is an orally available, selective PPARγ modulator with a good safety profile. ATX08-001 is being developed as a first-in-class treatment for NP, in particular, pain associated with PHN. ATX08-001 is chemically and functionally distinct from other PPARγ agonists, such as thiazolidinediones, displaying a distinct target gene activation profile and a higher selectivity for PPARγ as compared to rosiglitazone and pioglitazone [97].

2.10. Reactive Oxygen Species

Oxidative stress has been implicated in the pathophysiology of NP and there is considerable evidence that superoxide and peroxynitrite play a key role in the development of chronic pain, central sensitization, the transition of acute to chronic pain, opiate-induced hyperalgesia and antinociceptive tolerance [98, 99]. The increased level of superoxide in
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that are involved in the regulation of synthesis of pro-inflammatory cytokines (IL-1, TNFα) [103]. p38 MAPK has two major isoforms, i.e., α and β, which are expressed in the adult spinal cord, each of them having a distinct role in pain processing [104]. Results from in vivo and in vitro studies show that p38 MAPK inhibitors decrease cytokine production thereby leading to the attenuation of inflammatory pain and NP [103, 104]. p38 MAPK inhibitors reduce heat hyperalgesia and mechanical allodynia in CCI and diabetic rats [103].

Dilmapimod (SB-681323) (Fig. 13) is a potent, selective, reversible inhibitor of p38α MAPK which inhibits cytokine production. Results from a placebo-controlled clinical trial aiming to assess its efficacy after oral 14-day administration in NP showed that dilmapimod significantly reduced average daily pain intensity and was well tolerated in patients with NP following nerve injury with a maximum permissible clinical dosing period of 28 days, which finally precluded further development of this compound in chronic pain conditions [103-105].

Despite its previously established efficacy equal to gabapentin in the attenuation of mechanical allodynia after oral dosing in CCI rats, losmapimod (GW856553) (Fig. 13), a p38αβ MAPK inhibitor (given 7.5 mg twice a day, 28 days) was tested in a double-blind, placebo-controlled study in patients with NP following traumatic peripheral nerve injury. In these patients, it failed to show clinically relevant analgesic properties [104].

Cetuximab, the inhibitor of epidermal growth factor receptor and MAPK-signaling inhibitor was shown to alleviate NP in cancer patients. There is also some evidence from

the dorsal horn neurons was observed in NP conditions [100], whereas heat hyperalgesia in neuropathic rats was reduced by systemic administration of antioxidants [98]. N-acetyl-L-cysteine (NAC) (Fig. 12) is a mucolytic agent used in the diseases of the respiratory system and as a treatment for paracetamol poisoning where it acts to maintain glutathione levels in the liver. In CCI rats, NAC reduced mechanical and heat hyperalgesia, as well as cold allodynia [98]. Recent preclinical studies revealed that its anti-hyperalgesic properties in CCI rats were due to its inhibitory effect on iNOS gene expression, which significantly reduced nitric oxide level. Moreover, NAC suppressed c-Jun N-terminal kinase and p38 proteins which are strongly involved in the central sensitization of pain [101]. A single systemic injection of this drug causes analgesia in NP conditions. This effect is due to the reinforcement of mGlu2 receptor activation [102].

2.11. p38 Mitogen-activated Protein Kinase

p38 MAPK phosphorylates intracellular proteins, including signal transduction molecules and transcription factors

Fig. (12). Structure of N-acetylcysteine.

Fig. (13). Structures of dilmapimod and losmapimod.

Fig. (14). Structures of rosuvastatin and simvastatin.
### Table 1. Experimental drugs for the treatment of neuropathic pain - a summary.

| Pharmacological Class (Example) | Mechanism of Action in NP | Preclinical Efficacy (Animal Models) | Clinical Efficacy |
|--------------------------------|--------------------------|-------------------------------------|-------------------|
| β₂-adrenomimetics (terbutaline, salbutamol) | ↓TNFα | Traumatic nerve injury-related NP; diabetic NP | Post-thoracotomy NP (salbutamol); post-thoracoscopy NP (terbutaline) |
| Angiotensin AT₂ receptor antagonists (EMA-401) | ↓ p38 MAPK; ↓ ERK | Surgically-induced NP | PHN; diabetic NP (withdrawn) |
| H₂ receptor inverse agonists (GSK189254, GSK334429) | ↓ central sensitization; ↓ spontaneous neuronal firing | Surgically- and virally-induced NP models; Spinal nerve ligation NP model | No data available |
| Acetylcholinesterase inhibitors (donepezil) | ↑ acetylcholine – M₄ receptor stimulation | CIPN; spared nerve injury model | Diabetic NP; patients with chronic neuropathic pain who are currently taking gabapentin or pregabalin (phase 4) |
| Neurotoxins (botulinum toxin A) | SNAP-25 degradation; ↓ glutamate; ↓ CGRP; ↓ TRPV1 expression; ↓ IL-1β, ↓ IL-18; ↑ IL-10, ↑ IL-1RA; ↓ central sensitization sodium channel inactivation | Diabetic NP; Surgically-induced NP | Established efficacy in PHN; probable efficacy in trigeminal neuralgia and post-traumatic neuralgia; possible efficacy in diabetic polyneuropathy; insufficient evidence in CRPS, phantom limb and stump pain, and occipital neuralgia |
| Microglial activation inhibitors (minocycline) | Microglia inhibition; matrix metalloproteinase 3 inhibitor; ↓ chemokines; ↓ TNFα; ↓ IL-1β; ↓ iNOS; ↑ IL-1α; ↑ IL-2; ↑ IL-10 | CIPN; diabetic NP; traumatic nerve injury-related NP | Prevention of postoperative intercostal neuralgia |
| PPARγ agonists (rosiglitazone, ATx08-001) | ↓ IL-1β, ↓ IL-6; ↓ TNFα; ↓ chemokines | CIPN | PHN |
| Kinase inhibitors (dilmapimod, losmapimod) | ↓ p38 MAPK | NP following traumatic peripheral nerve injury | No longer in development due to low efficacy |
| T-type calcium channel antagonists (ethosuximide, ABT-639) | T-type calcium channel antagonism | NP following traumatic peripheral nerve injury (ethosuximide, ABT-639); spinal nerve ligation NP model; CIPN (ABT-639) | Diabetic NP – ABT-639 (no longer in development) |
| Sodium channel antagonists (ambroxol) | Na,1.8 sodium channel antagonist | CIPN; Spinal cord injury-related NP | PHN; Post-operative neuralgia; phantom pain; foot neuropathy; deafferentation pain |
| Melatonin MT₁/MT₂ receptor agonists (ramelteon, melatonin, piromelatine) | Neuritogenesis stimulation; neuroprotection; ↓ nitro-oxidative stress; induction of autophagy; mitochondrial protection | CIPN | Ramelteon: terminated (recruitment did not accrue as expected) |
| TRPA1 antagonists (ADM_09; HC-030031) | ↓ oxidative stress; ↓ p38 MAPK | CIPN; diabetic NP | No data available |
| 3-Hydroxy-3-methyl glutaryl coenzyme A reductase inhibitors = statins (simvastatin, rosuvastatin) | ↓ oxidative stress; ↓ iNOS inhibition; decreased translocation of RhoA; reduced microglial and astrocyte activation; ↓ IL-1β secretion; immunomodulation; neuroprotection | CIPN; CRPS; surgically-induced NP | Diabetic NP; phantom limb pain |
Clinical trials that it prevents the development of oxaliplatin-induced neuropathy, but further detailed studies are required to confirm these preliminary observations [106].

2.12. 3-Hydroxy-3-methyl Glutaryl Coenzyme A Reductase

Statins, the inhibitors of 3-hydroxy-3-methyl glutaryl coenzyme A reductase, are widely used for the treatment of hypercholesterolemia. Because of their cholesterol-independent pleiotropic effects, such as anti-inflammatory, antioxidant, immunomodulatory and neuroprotective activities, they have also been tested in non-cardiovascular disorders, including NP. Data from preclinical studies indicate that atorvastatin reversed the decrease in mechanical and thermal nociceptive threshold in CCI model in rats and this effect was due to its antioxidant [15, 107] and anti-inflammatory [108] properties. It inhibited cytokines, matrix metalloproteases and nerve growth factor in the sciatic nerve and in the spinal cord, which altogether suggested that it could be a treatment option for NP [108].

Rosuvastatin and simvastatin (Fig. 14) attenuated symptoms of painful diabetic neuropathy in streptozotocin-treated diabetic animals and ischemia-reperfusion-induced nerve injury models [15]. In the cold plate test in oxaliplatin-treated neuropathic mice cold allodynia caused by oxaliplatin was not influenced by a single dose of simvastatin, but in contrast to this, a 7-day treatment with this drug partially reversed allodynia induced by cold. It indicated that the repeated administration of simvastatin elevated pain threshold in animals with CIPN symptoms [109].

Similarly, data from clinical studies indicate that statins may protect against neuropathic stump pain, phantom limb pain [107] and peripheral sensory neuropathy in type 2 diabetes [15]. This effect is not related to lipid-lowering action of statins and it may comprise distinct mechanisms, such as: inhibition of iNOS and increase in eNOS expression, decreased translocation of RhoA (a small molecular GTPase) from the cytoplasm to plasma membrane, reduced microglial and astrocyte activation and a marked reduction in IL-1β secretion in human peripheral blood mononuclear cells [108].

CONCLUSION

Most analgesic drugs used in NP not only offer limited efficacy but they also cause dose-limiting side effects, which negatively influences patients’ quality of life. Hence, it is evident that the identification of novel drug targets for analgesic drugs used in NP treatment, as well as using reliable animal models for the prediction of the efficacy of analgesic compounds in humans is of special relevance (Table 1). To achieve this, at the preclinical stage of analgesic drug development process, rodents are widely used. However, as in other areas of experimental pharmacology, one may face translation problems from animals to humans. This issue has been shown by numerous studies in which animal models of NP were poorly predictive of clinical efficacy of analgesic drugs in neuropathic patients (Table 1). Because of this, many promising drug candidates for NP selected in the preclinical phase failed to be effective in clinical trials. It should be however emphasized that this did not preclude their use in the drug discovery process as these agents could remain valuable tools for experimental studies on the role of certain biomolecules (e.g., ion channels, receptors or enzymes) in NP etiology.

Patients’ needs for a more effective pain relief without unwanted side effects are currently being developed by the introduction of new targets for analgesic drugs, or by the search for new analgesic agents with a known mechanism of action. Also, the application of genetic approaches and biochemical assays leading to deeper understanding of molecular mechanisms of NP enables the progress in NP treatment.

In this context, novel experimental drugs can be considered a treatment option which should be tailor-made for each individual on the basis of pain features, previous therapies, associated clinical conditions, recurrence of pain, adverse effects, contraindications and patients’ preferences. Furthermore, physicians wishing to treat patients with these drugs in an off-label fashion should have experience using them for other conditions. Noteworthy, one of the undoubtedly benefits of using well-known drugs as experimental drugs for new therapeutic indications (here in the treatment of NP) is the fact that these drugs have been thoroughly studied in humans, in terms of their tolerability, drug-drug interactions and potential toxic effects.

Importantly, it should be emphasized that there is a strong medical demand to discover drugs that act causally, i.e., by the elimination of mechanisms that trigger NP. As shown in Table I experimental therapies for NP have the ability to interfere with pathomechanisms responsible for the development of NP. Such an approach would provide not only pain relief, but it could also cease the progress of neuropathy.

Moreover, the use of experimental drugs for NP treatment potentially allows multidirectional approach to neuropathic patients using a single drug to treat symptoms of NP and other co-morbidities. This is of particular importance in the case of many lifestyle diseases, such as asthma, diabetes, cardiovascular diseases.

At present, among numerous drug classes tested as experimental therapies for NP, there are only some which may have potential as novel treatments. Increasing knowledge about mechanisms underlying NP, mechanisms of drug action, as well as available efficacy and safety data from preclinical and clinical studies, make some of the abovementioned agents promising potential future treatment options. In that context, botulinum toxin A, minocycline, ambrroxol, statins and PPAR agonists (ATX086001) are particularly interesting. Of note, NAC, statins and β2-agonists may also offer an alternative to currently available analgesic drugs for the treatment of NP, as they may be used in patients suffering not only from chronic pain but also other serious comorbidities. Moreover, their safety profile is well established, which might be of great importance for some groups of patients.

LIST OF ABBREVIATIONS

AChEI = Acetylcholinesterase inhibitors
Ang II = Angiotensin II
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