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Biologics: the next generation of analgesic drugs?

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For many decades, there have been few novel therapies for pain, and the number of promising targets that have been genuinely validated in the clinic is small. Discovery and development of biologic therapies for analgesia provides a better opportunity to test such targets, potentially providing new and effective therapies. Biologics have revolutionised the treatment of many diseases, with the greatest advances seen in oncology and inflammatory disorders. Across a broad spectrum of severe, chronic pain disorders – including inflammatory pain, neuropathic pain and cancer pain – biologics could offer patients safer and more-effective alternatives to currently available treatments. As such, progression of large-molecule therapies is becoming a strategic priority for companies as they look to advance their portfolios.

Introduction

Traditionally, the treatment of chronic pain has been dominated by small-molecule therapies. The use of non-steroidal anti-inflammatories (NSAIDS), cyclooxygenase (COX)-2 inhibitors, opioids, gabapentinoids and selective or non-selective monoamine reuptake inhibitors is widespread and brings relief to millions of patients. However, these therapies have significant limitations across the broad spectrum of mechanisms that underlie chronic pain. Firstly, efficacy is limited: responder rates in patients suffering from neuropathic pain is typically in the 30–40% range, and limited to 50–60% in patients with nociceptive or inflammatory pain [1]. This leaves a huge burden of need, with patients and physicians demanding greater efficacy from new treatments. Secondly, tolerability and safety are often limiting: factors including cardiovascular risk, central side effects, tolerance to effect and potential dependence limits the use of commonly available analgesics.

Considerable research efforts have, therefore, been directed towards discovering new approaches to pain relief. A huge investment in understanding the basic mechanisms of pain transduction and perception, as well as outcomes from genomic and genetic research, has yielded a wealth of new targets. However, the search for new small molecules that engage with these targets has proved frustrating with significant attrition in early clinical development [2]. The most frequently cited reason for failure to demonstrate efficacy centres on an inability to engage the target effectively in a pharmacologic manner in humans; and in the majority of cases this is because of off-target interactions that limit dose escalation. The industry has not been successful, therefore, in testing these new targets effectively and, indeed, clinically predictive animal models of pain can also create a shortfall in selecting the right targets and providing preclinical to clinical translational confidence. During the past two or more decades, the number of targets that have been genuinely invalidated for pain are sparse; many opinion leaders cite the neurokinin-1 receptor as the only example [3]. Other small-molecule trials have failed owing to the inability of the trial molecule to penetrate the central nervous system (CNS) in humans [4]. Some prospective molecules have fallen by the wayside owing to poorly designed trials. Adding to these challenges, pain relief research is conducted in an increasingly demanding regulatory environment which applies to large- and small-molecule approaches alike. Since the withdrawal of the COX-2 inhibitor rofecoxib (VIOXX®) in 2004 following safety concerns, there has been an acute awareness that new approaches to pain relief must be able to show robustly favourable benefit:risk ratios in long-term outcome studies. A different approach is needed, and the rise of biologic-based interventions is a model that many pharmaceutical companies are now testing in the

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BOX 1

Conditions associated with chronic pain

- Osteoarthritis
- Neuropathic pain
- Rheumatoid arthritis
- Herpes zoster (shingles) and post-herpetic neuralgia
- Low back, shoulder and neck pain
- Trigeminal neuralgia
- Headache, including migraine
- Diabetic neuropathy
- Cancer pain
- Temporomandibular joint disorder
- Myofascial pain syndromes
- Post-mastectomy pain
- Post-thoracotomy pain
- Angina pectoris
- Chronic regional pain syndromes
- Chronic visceral pain syndromes
- Stump and phantom limb pain

context of bringing forward new, safe and effective therapies for pain.

The need for a new approach

The demand for new treatments to combat chronic pain has never been greater. Chronic pain of moderate-to-severe intensity occurs in ~19% of adults worldwide [5,6], seriously affecting the quality of their social and working lives. Approximately one in five people with chronic pain has to give up work [6]. The causes of this pain are numerous and diverse (Box 1). Most instances are caused by muscle, bone and joint pain, although the cause remains unknown in ~5% of cases (http://www.dh.gov.uk/en/MediaCentre/Media/DH_096271).

There is no doubt that chronic pain exerts a significant toll, in terms of individual distress and societal impact. The direct and indirect financial costs of pain are substantial. One study estimates that the cost of back pain alone is equivalent to more than a fifth of a country's total health expenditure [7], which equates to €46 056 million in Germany, €37 053 million in France (http://www. epp.eurostat.ec.europa.eu/statistics_explained/index.php?title=File:Current_health_expenditure_totals_per_country.PNG&filetimestamp=20090430100019) and £22 008 billion in the UK (http://www.ukpublicspending.co.uk/index.php). A recent Swedish study estimated that the socioeconomic costs of chronic pain amounted to a tenth of the country's gross domestic product [8].

The search for new approaches

It is estimated that over a quarter of pharmaceutical R&D pipelines are now dedicated to biologic therapies [9]. Monoclonal antibodies (mAbs), in particular, are the mainstay of new biologic approaches. In the context of potential for meeting unmet need in pain they have many attractive properties; generally they have extremely high affinity to the target, and only rarely do they show activity towards anything other than the target against which they were designed to interact. These two properties bring great advantage because the principle reason for clinical trial failure, that of inadequate dose-escalation caused by off-target unwanted effects, is removed. In addition, mAbs tend to have long terminal half-lives, usually of the order of 7–14 days, owing to their large size, metabolic stability and recycling capabilities [10], which in turn translates to a reduced inter-dose interval in the clinic. In addition, the discovery and development of mAbs is typically more straightforward and there is a higher overall success rate than is the case for small molecules.

There are, however, several disadvantages with mAbs that need to be recognised. The most obvious is the route of administration; despite the potential for infrequent (weekly to monthly) dosing, subcutaneous or intravenous (i.v.) administration is a potential drawback for many patients. In addition, the biophysical characteristics of mAbs and other biologics mean that penetration of the blood–brain barrier is limited. Typically, mAb concentrations within the CNS only reach 0.1–0.5% of those in the systemic circulation. Monoclonal antibodies are also limited in their ability to cross cell membranes. Thus, mAb-based therapies tend to be directed against soluble targets that are accessible in extracellular spaces such as the blood, although emerging technologies and new approaches for antigen presentation mean that G-protein-coupled receptors and ion channels are becoming viable targets, and some progress is being made in these areas. In addition, although significant research is being dedicated to surmounting the blood–brain barrier [11], clinical applicability and implications for overall safety profiles are not yet known. These technology platforms are maturing but the current focus is predominantly on cytokines, growth factors and inflammatory mediators as the key targets for mAb-based pain relief.

The potential of mAbs in neuroscience

There are currently 26 mAb fragments approved for use in the USA and the revenue from these drugs is expected to exceed US$50 billion over the next four years, representing 50% of all therapeutic protein sales [12]. To date, there are over 350 mAbs in preclinical and clinical development, and new mAbs are entering clinical development at a rate of ~40 per year [13].

Potential targets for biologic pain relief

Nerve growth factor

Nerve growth factor (NGF) was discovered more than 50 years ago and is the 'founder member' of the neurotrophin super family. It binds to two receptors: a common neurotrophin super family receptor, P75, which has affinity for all neurotrophins, and a specific receptor, tyrosine kinase receptor (Trk)A. Early work established the importance of NGF for the survival of neurons in the developing nervous system [14]; however, for the past 15 years NGF has also been recognised to be a key pain mediator, thereby attracting a great deal of interest from researchers [15]. NGF levels have been shown to be elevated in several painful conditions in humans including arthritis, cystitis, prostatitis and chronic headaches [16]. Injected NGF causes a rapid thermal hyperalgesia, followed by delayed thermal hyperalgesia and mechanical allodynia [17].

NGF antibodies have been reported to be efficacious in a wide range of animal pain models (e.g. a rodent model of metastatic bone pain) and anti-NGF blocks the behavioural readouts of pain as effectively as the maximum-tolerated dose of morphine [18]. NGF-blocking agents have been demonstrated to reverse thermal

TABLE 1

Clinical-stage anti-NGF mAbsSource: clinicaltrials.gov.

Antibody	Sponsor company	Phase of clinical testing
Tanezumab	Pfizer	Phase III
Fulranumab	Johnson & Johnson R&D	Phase II
RGN475	Regeneron	Phase II
PG110	Abbott	Phase I
MEDI578	MedImmune/AstraZeneca	Phase I

This table details current anti-NGF mAbs in clinical development; further activity in research phases is not shown.

and mechanical hyperalgesia in several models of inflammatory hypersensitivity [15], an anti-NGF blocking molecule inhibits traumatic neuroma formation in the rat [19] and, in animal models of visceral pain, systemic administration of NGF-blocking agents inhibits the behavioural responses elicited by irritants instilled into the colon, stomach and bladder [20].

In addition to preclinical work, five antibodies targeting NGF have progressed into clinical trials (Table 1), seeking to demonstrate efficacy in a range of pain states. In 2008, Pfizer released initial data from a Phase II dose-ranging study in patients with moderate-to-severe osteoarthritis pain using their humanised anti-NGF mAb tanezumab, the most advanced of the clinical-stage antibodies targeting NGF. This study demonstrated levels of analgesic efficacy could be obtained with eight weekly i.v. doses of this mAb, approximately twice that seen with COX-2 inhibitors [21]. Tanezumab has also been reported to have superior efficacy to naproxen in a subgroup of chronic lower back pain patients [22]. Subsequently, Phase III data reported for tanezumab have shown significant improvement in the Western Ontario and McMaster Universities OA Index (WOMAC) pain scores, confirming that therapeutic benefit is reproducible in large studies with tanezumab (http://www.ampainsoc.org/abstract/view/4761/). Therefore, for the first time in decades, a truly novel, efficacious mechanism has been described with the advent of this class of drugs.

Although anti-NGF efficacy is encouraging, the safety profile of treatment with these agents is currently under review. In Phase II studies for tanezumab and fulranumab in a range of painful conditions, these antibodies were reported to be generally well tolerated as monotherapy and adjunctive therapy. The most common adverse events reported with these mAbs included headache, upper respiratory tract infections and also some abnormalities of peripheral sensation (e.g. paresthesia and arthralgia) which were generally mild and transient in nature [21–23]. However, in 2010 clinical studies with tanezumab were suspended in all patients except those with cancer-related pain, owing to a small number of subjects with osteoarthritis requiring total joint replacements (http://www.prnewswire.com/news-releases/pfizer-suspends-tanezumab-osteoarthritis-clinical-trial-program-97012899.html). At the time of writing only tanezumab and fulranumab are currently under active clinical investigation, both in cancer pain patients only, as the FDA has placed this class under 'clinical hold' due to safety concerns. With several anti-NGF mAbs currently at the stage of clinical testing, there is much desire to understand the complete profile of this innovative class of drugs so that they can be used appropriately in the clinic as well as in the laboratory. Because joint replacement is an anticipated outcome for at least some of the patients in these clinical trials, understanding the true event rate seen with these agents in the moderate-to-severe OA pain population is crucial. It is good news, therefore, that the FDA has announced that on March 12, 2012, an Advisory Committee meeting will be held with the express remit to assess if: 'reports of joint destruction represent a safety signal related to the anti-NGF class of drugs, and whether the risk benefit balance for these drugs favours continued development of the drugs as analgesics' (http://www.fda.gov/AdvisoryCommittees/Calendar/ucm286556.htm). Continued clinical development of the anti-NGFs in a broad range of patient groups hinges on this discussion.

Interleukin 6

Interleukin 6 (IL-6) is a pleiotropic cytokine with a wide range of biological effects that has an important role in the regulation of inflammation and the immune response. Preclinical studies have shown that IL-6 levels are increased in inflammatory pain [24] and neuropathic pain models [25,26]. In addition, administration of exogenous IL-6 has been shown to cause alterations in thermal and mechanical hypersensitivity in the rat [25], and sensitize joint afferent nerve fibres [27]. IL-6 knockout mice show reduced mechanical allodynia following spinal nerve ligation [28] and a lack of thermal hyperalgesia and mechanical allodynia following chronic constriction nerve injury [26]. Neutralisation of IL-6 with a mAb reduces mechanical allodynia in a rat model of chemotherapy-induced neuropathy [29], and also reduced thermal hyperalgesia following partial sciatic nerve ligation [30]. Therefore, the evidence across a range of models and approaches to manipulate IL-6 levels suggests that preclinically there is clearly a role for this cytokine in pain generation and maintenance.

In healthy humans, levels of IL-6 are low but can become elevated during painful inflammatory conditions [31]. Raised levels of this cytokine have been associated with known 'painful' conditions such as rheumatoid arthritis (RA) and chronic lower back pain, leading to the development of a range of IL-6 inhibitors. The first such inhibitor was the IL-6-receptor-targeting mAb tocilizumab, which was first approved for use in Europe (as RoACTEMRA®) for RA in 2008. However, as with tumour necrosis factor (TNF) inhibitors, despite the preclinical evidence for a role in pain, there are surprisingly few studies in which a direct effect of IL-6 inhibition of pain has been investigated clinically. Because tocilizumab is more widely studied in patients with RA, more data are becoming available suggesting that administration of an anti-IL-6-receptor antibody results in a significant reduction in the visual analogue scale (VAS) score for pain when compared with placebo at doses of 4 or 8 mg/kg after 24 weeks of treatment [32,33]. The qualification of analgesic effect as a result of direct suppression of afferent activation and firing, or as a result of an ongoing disease modification, is, at this stage, impossible to complete. Whether or not pain and disease modification endpoints are mutually exclusive requires further study.

The only currently marketed therapeutic targeting IL-6 is the mAb tocilizumab, which is currently approved for use in the lymphatic disorder Castleman's disease, RA and recently for systemic juvenile idiopathic arthritis (Still's disease). Interestingly, and perhaps uniquely, a study was recently published in which an orally active small-molecule approach has been shown to be active

in an animal model of pancreatitis pain [34] suggesting that, at least for approaches targeting IL-6, small-molecule and biologic approaches might be plausible.

Tumour necrosis factor

TNF-α is a potent pro-inflammatory cytokine. Anti-TNF treatments have already transformed the management of RA where the TNF inhibitors etanercept and infliximab have been approved for more than ten years. Although studies in RA have demonstrated reduced pain scores for both drugs, there have been relatively few studies specifically looking at their analgesic effects. Indeed, the potential use of TNF inhibitors for the widespread treatment of chronic pain might be limited by the long-term side-effect profile of the drugs, including increased infection rate and heart failure [35].

A wide range of preclinical evidence supports a role for TNF as a potential analgesic therapeutic target. Levels of TNF increase rapidly following an inflammatory stimulus. For example, TNF R1 knockout mice show reductions in the level of hypernociception following injection of TNF-α into the paw [36]. Further, injection of TNF-α has been shown to increase pain sensitivity following intra-plantar injection [37]. Etanercept and infliximab have recently been shown to be effective in reversing the mechanical hyperalgesia in a rat joint pain model [38]. Furthermore, etanercept has shown activity in murine models of neuropathy [39].

Despite all the preclinical evidence supporting a role for TNF as a target in pain, studies investigating effects of TNF inhibitors specifically on clinical pain are not that common in the literature, probably owing to the fact that these drugs were primarily developed as disease-modifying treatments for the painful condition RA where increased levels of TNF are found. The first anti-TNF inhibitor to be launched was the fusion protein etanercept which was approved for the treatment of RA in 1998. This was followed by the mAbs infliximab and adalimumab, which were approved for RA treatment in 1999 and 2002, respectively. However, etanercept has been shown to produce a highly significant improvement in the pain associated with RA using a VAS scoring system in a double-blind trial with a dose of 16 mg/m^2 [40]. In addition, infliximab significantly reduced pain scores in RA patients at 3 and 10 mg/kg when administered at four weekly intervals for a period of 14 weeks [41]. Again, the mutual exclusivity of pain and disease modification endpoints requires further study, because the timecourses of human disease and rodent pain models are different. One of the main drawbacks of TNF inhibitors is the incidence of observed side effects which act as a major limiting factor in use of these treatments in the clinical setting. For example, Maini *et al.* [41] reported side effects in 82% of patients treated with infliximab ranging from headache through to a higher incidence of respiratory problems, compared with 57% for the placebo group. The risk of serious infections such as tuberculosis is also increased with this class of drug and all current anti-TNF drugs carry black-box warnings regarding this.

Despite these issues, the biologic approach has been highly successful for TNF inhibitors. In 2008, the three leading products at the time (etanercept, remicade and adalimumab) generated sales of US$16 billion and this market is still growing in size. Because of this success, several new biologics are currently being investigated at the preclinical stage. To decrease the side-effect potential these candidates are targeted towards the soluble form of TNF and, recently, Zalevsky *et al.* reported positive effects of such a molecule (XPro1595) in a mouse arthritis model [42]. Furthermore, a TNF RI receptor antagonist has been reported to be as effective as etanercept in a mouse arthritis model but to have fewer side effects [43]. As more anti-TNF biologic drugs enter the marketplace, further studies could emerge in pain patient populations, beyond RA, where there is an appropriate benefit:risk profile.

Other biologic approaches

Although mAbs clearly represent the biologic molecules with the most potential to achieve a novel approach to pain control, there are several other avenues under investigation. These are beyond the scope of detailed review here but a brief overview is appropriate given some reports of progress. The first of these is the use of antibodies and peptides to inhibit ion channel function. Elucidation of the structural motifs involved in ion channel gating has led to an increase in efforts to target extracellular domains with mAbs and peptides but, so far, other than the launched product ziconotide, a peptide that blocks function of the N-type calcium channel [44], there are few reports of clinical development for functional ion channel blocking biologics. As technologies mature for discovery and manufacture of peptide and mAb-based approaches, this looks probably to change over the next few years because there is a wealth of emerging publications on the use of biologics in the research arena for modulation of ion channel function.

The second emerging area is the therapeutic targeting of G-protein-coupled receptors using biologic approaches, and here research efforts have started to identify methods for generating functional antibodies that might be suitable for progression [45]. Successful examples are emerging in other therapeutic areas, showing that the technology is sufficiently mature to be delivered into the clinic, and significant progress should be anticipated here.

Finally, the discovery of technologies to facilitate delivery of biologics across the blood–brain barrier provides a potentially transformational platform for biologic-selective manipulation of CNS-based targets, and provides a step change opportunity over the numerous small-molecule approaches in this area, most of which are beset by selectivity challenges. The blood–brain barrier platform has yet to mature into a clinically applicable approach but recent progress suggests that this is only a matter of time [11].

Concluding remarks

Although the pipeline of small-molecule analgesics has yet to deliver a genuinely novel and transformational therapy for pain, there is reason for optimism that biologic research could soon be able to replenish the supply of novel therapies in pain control. Clearly, there are many challenges still to overcome. However, the exquisite selectivity offered by mAbs is already enabling researchers to focus specifically on therapeutic targets that have previously proved to have low tractability to small-molecule approaches and those identified by genomic research.

As a result of this research, we have already observed proof-of-concept with biologic-based therapies (e.g. ziconotide and the anti-NGF mAbs) and several exciting molecules are progressing well through preclinical and clinical testing. Moreover, technologies that enable the transport of active macromolecules, including mAbs

across the blood–brain barrier, are extending the limits of biologic therapy beyond the soluble mediators such as cytokines, growth factors and inflammatory mediators. During the past ten years, biologic therapy has transformed the management of inflammatory conditions such as RA. In the coming decade, it is possible that it will do the same for pain control.

References

1 Burgess, G. and Williams, D. (2010) The discovery and development of analgesics: new mechanisms, new modalities. *J. Clin. Invest.* 120, 3753–3759
2 Schuster, D. *et al.* (2005) Why drugs fail – a study on side effects in new chemical entities. *Curr. Pharm. Des.* 11, 3545–3559
3 Hill, R. (2000) NK1 (substance P) receptor antagonists – why are they not analgesic in humans? *TIPS* 21, 244–246
4 Sneyd, J.R. *et al.* (2007) Multicentre evaluation of the adenosine agonist GR79236X in patients with dental pain after third molar extraction. *Br. J. Anaesth.* 98, 672–676
5 Gureje, O. *et al.* (1998) Persistent pain and well-being: a World Health Organization study in primary care. *JAMA* 280, 147–151
6 Breivik, H. *et al.* (2006) Survey of chronic pain in Europe: prevalence, impact on daily life, and treatment. *Eur. J. Pain* 10, 287–333
7 Phillips, C.J. (2006) Economic burden of chronic pain. *Expert Rev. Pharmacoecon. Outcomes Res.* 6, 591–601
8 Gustavsson, A. *et al.* (2012) Socio-economic burden of patients with a diagnosis related to chronic pain – register data of 840,000 Swedish patients. *Eur. J. Pain* 16, 289–299
9 Pickering, L. (2009) Lessons learned in the pharmaceutical industry, 2009. *Spectrum: Decision Resources* 17, 1–32
10 Daugherty, A.L. *et al.* (2006) Formulation and delivery issues for monoclonal antibody therapeutics. *Adv. Drug Del. Rev.* 58, 686–706
11 Yu, Y. *et al.* (2011) Boosting brain uptake of a therapeutic antibody by reducing its affinity for a transcytosis target. *Sci. Transl. Med.* 3, 84ra44
12 De Souza, E.B. *et al.* (2009) Novel therapeutic modalities to address nondrugable protein interaction targets. *Neuropsychopharmacology* 34, 142–158
13 Reichert, J.M. (2011) Antibody-based therapeutics to watch in 2011. *mAbs* 3, 76–79
14 Lewin, G.R. and Barde, Y.A. (1996) Physiology of the neurotrophins. *Annu. Rev. Neurosci.* 19, 289–317
15 Pezet, S. and McMahon, S.B. (2006) Neurotrophins: mediators and modulators of pain. *Annu. Rev. Neurosci.* 29, 507–538
16 Hefti, F.F. *et al.* (2006) Novel class of pain drugs based on antagonism of NGF. *Trends Pharmacol. Sci.* 27, 85–91
17 Petty, B.G. *et al.* (1994) The effect of systemically administered recombinant human nerve growth factor in healthy human subjects. *Ann. Neurol.* 36, 244–246
18 Sevcik, M.A. *et al.* (2005) Anti-NGF therapy profoundly reduces bone cancer pain and the accompanying increase in markers of peripheral and central sensitization. *Pain* 115, 128–141
19 Kryger, G.S. *et al.* (2001) Nerve growth factor inhibition prevents traumatic neuroma formation in the rat. *J. Hand Surg. Am.* 26, 635–644
20 Jaggar, S.I. *et al.* (1999) Inflammation of the rat urinary bladder is associated with a referred thermal hyperalgesia which is nerve growth factor dependent. *Br. J. Anaesth.* 83, 442–448
21 Lane, N. *et al.* (2010) Tanezumab for the treatment of pain from osteoarthritis of the knee. *N. Engl. J. Med.* 363, 1521–1531
22 Katz, N. *et al.* (2011) Efficacy and safety of tanezumab in the treatment of chronic low back pain. *Pain* 152, 2248–2258
23 Evans, R. *et al.* (2011) Proof of concept trial of tanezumab for the treatment of symptoms associated with interstitial cystitis. *J. Urol.* 185, 1716–1721
24 Chessell, I.P. *et al.* (2005) Disruption of the P2X7 purinoceptor gene abolishes chronic inflammatory and neuropathic pain. *Pain* 114, 386–396
25 DeLeo, J.A. *et al.* (1996) Interleukin-6-mediated hyperalgesia/allodynia and increased spinal IL-6 expression in a rat mononeuropathy model. *J. Interferon Cytokine Res.* 16, 695–700
26 Murphy, P.G. *et al.* (1999) Endogenous interleukin-6 contributes to hypersensitivity to cutaneous stimuli and changes neuropeptides associated with chronic nerve constriction in mice. *Eur. J. Neurosci.* 11, 2243–2253
27 Brenn, D. *et al.* (2007) Sensitization of unmyelinated sensory fibers of the joint nerve to mechanical stimuli by interleukin-6 in the rat: an inflammatory mechanism of joint pain. *Arthritis Rheum.* 56, 351–359
28 Ramer, M.S. *et al.* (1998) Spinal nerve lesion induced mechanoallodynia and adrenergic sprouting in sensory ganglia are attenuated in interleukin-6 knockout mice. *Pain* 78, 115–121
29 Kiguchi, N. *et al.* (2008) The cirtical role of invading peripheral macrophage-derived interleukin-6 in vincristine-induced mechanical allodynia in mice. *Eur. J. Pharmacol.* 592, 87–92
30 Ma, W. and Quirion, R. (2006) Increased calcitonin gene-related peptide in neuroma and invading macrophages is involved in the upregulation of interleukin-6 and thermal hyperalgesia in a rat model of mononeuropathy. *J. Neurochem.* 98, 180–192
31 Kiefer, R. *et al.* (2001) The role of macrophages in immune-mediated damage to the peripheral nervous system. *Prog. Neurobiol.* 64, 109–127
32 Smolen, J.S. *et al.* (2008) Effect of interleukin-6 receptor inhibition with tocilizumab in patients with rheumatoid arthritis (OPTION study): a double-blind, placebo-controlled, randomised trial. *Lancet* 371, 987–997
33 Yazici, Y. *et al.* (2012) Efficacy of tocilizumab in patients with moderate to severe active rheumatoid arthritis and a previous inadequate response to disease-modifying antirheumatic drugs: the ROSE study. *Ann. Rheum. Dis.* 71, 198–205
34 Vardanyan, M. *et al.* (2010) Reversal of pancreatitis-induced pain by an orally available, small molecule interleukin-6 receptor antagonist. *Pain* 151, 257–265
35 Cush, J.J. (2004) Unusual toxicities with TNF inhibition: heart failure and drug-induced lupus. *Clin. Exp. Rheumatol.* 22 (Suppl. 35), 141–147
36 Cunha, T.M. *et al.* (2005) A cascade of cytokines mediates mechanical inflammatory hypernociception in mice. *Proc. Natl. Acad. Sci. U.S.A.* 102, 1755–1760
37 Woolf, C.J. *et al.* (1997) Cytokines, nerve growth factor and inflammatory hyperalgesia: the contribution of tumour necrosis factor alpha. *Br. J. Pharmacol.* 121, 417–424
38 Schaible, H.G. *et al.* (2010) The role of proinflammatory cytokines in the generation and maintenance of joint pain. *Ann. N. Y. Acad. Sci.* 1193, 60–69
39 Sommer, C. *et al.* (2001) Etanercept reduces hyperalgesia in experimental painful neuropathy. *J. Peripher. Nerv. Syst.* 6, 67–72
40 Moreland, L.W. *et al.* (1997) Treatment of rheumatoid arthritis with a recombinant human tumor necrosis factor receptor (p75)-Fc fusion protein. *N. Engl. J. Med.* 337, 141–147
41 Maini, R.N. *et al.* (1998) Therapeutic efficacy of multiple intravenous infusions of anti-tumor necrosis factor alpha monoclonal antibody combined with low-dose weekly methotrexate in rheumatoid arthritis. *Arthritis Rheum.* 41, 1552–1563
42 Zalevsky, J. *et al.* (2007) Dominant-negative inhibitors of soluble TNF attenuate experimental arthritis without suppressing innate immunity to infection. *J. Immunol.* 179, 1872–1883
43 Mukai, Y. *et al.* (2009) Fast binding kinetics and conserved 3D structure underlie the antagonistic activity of mutant TNF: useful information for designing artificial proteo-antagonists. *J. Biochem.* 146, 167–172
44 Schmidtko, A. *et al.* (2010) Ziconotide for the treatment of severe pain. *Lancet* 375, 1569–1577
45 Gupta, A. *et al.* (2008) Antibodies against G-protein coupled receptors: novel uses in screening and drug development. *Comb. Chem. HTS* 11, 463–467