British Journal of Anaesthesia **101 (1):** 87–94 (2008)
doi:10.1093/bja/aen100 Advance Access publication May 19, 2008

BJA

Translational medicine: cancer pain mechanisms and management

A. Delaney[1], S. M. Fleetwood-Walker[1], L. A. Colvin[2] and M. Fallon[3]*

[1]Centre for Neuroscience Research, College of Medicine and Veterinary Medicine, University of Edinburgh, Scotland, UK. [2]Department of Anaesthesia, Critical Care and Pain Medicine, University of Edinburgh, Western General Hospital, Crewe Road, Edinburgh EH4 2XU, UK. [3]Institute of Genetics and Molecular Medicine, Edinburgh Cancer Research Centre, Scotland, UK

**Corresponding author. E-mail: marie.fallon@ed.ac.uk*

Cancer-induced bone pain (CIBP) is a major clinical problem with up to 85% of patients with bony metastases having pain, often associated with anxiety and depression, reduced performance status, and a poor quality of life. Malignant bone disease creates a chronic pain state through sensitization and synaptic plasticity within the spinal cord that amplifies nociceptive signals and their transmission to the brain. Fifty per cent of patients are expected to gain adequate analgesia from palliative radiotherapy within 4–6 weeks of treatment. Opioid analgesia does make a useful contribution to the management of CIBP, especially in terms of suppressing tonic background pain. However, CIBP remains a clinical challenge because the spontaneous and movement-related components are more difficult to treat with opioids and commonly used analgesic drugs, without unacceptable side-effects. Recently developed laboratory models of CIBP, which show congruency with the clinical syndrome, are contributing to an improved understanding of the neurobiology of CIBP. This chronic pain syndrome appears to be unique and distinct from other chronic pain states, such as inflammatory or neuropathic pain. This has clear implications for treatment and development of future therapies. A translational medicine approach, using a highly iterative process between the clinic and the laboratory, may allow improved understanding of the underlying mechanisms of CIBP to be rapidly translated into real clinical benefits in terms of improved pain management.

Br J Anaesth 2008; **101**: 87–94

Keywords: analgesics, non-opioid; analgesics, opioid; cancer; pain, mechanism

Metastatic cancer-induced bone pain (CIBP) is a severe clinical problem that is often inadequately treated by current analgesics. Indeed, 85% of patients with bony metastases have significant pain associated with increased morbidity, reduced performance status, increased anxiety and depression, and a reduced quality of life.[5 6 56] Tumours that most often result in metastatic bone pain originate from breast, lung, and prostate cancers.[7 8] CIBP is a complex pain syndrome involving background pain (typically opioid responsive), which can be described as a dull ache that increases in intensity with progression of the disease.[43] Additionally, CIBP involves spontaneous breakthrough pain and movement-related pain, which are generally difficult to treat with opioids without intolerable side-effects.[44]

CIBP remains a clinically challenging problem to treat rapidly and effectively. In order to properly evaluate our current therapies and logically direct the development of new therapies, it is important to understand the underlying mechanisms of CIBP and the evidence base for our current standard treatments.

Clinical aspects of CIBP

Assessment and management

Careful clinical observation has demonstrated that CIBP has several components: a tonic background pain at rest, spontaneous pain at rest, and pain associated with movement.[88] Our work has shown that:

(i) pain on movement or spontaneous pain at rest has a mean VAS score of 7/10 compared with pain at rest, which has a mean score of 3/10;
(ii) 83% of patients have pain, which is significantly worse on movement;
(iii) 50% of patients with movement-related or spontaneous pain reported that the duration was <30 min and 25% reported a duration of <15 min;
(iv) 52% of patients felt that both movement-related and spontaneous pain were unpredictable.[88]

Although the tonic background pain is usually controlled with opioid analgesia, the other two components are problematic because of three factors.

(1) Temporal onset of pain in relation to temporal onset of analgesia from opioids. Onset of analgesic effects of immediate release morphine is usually 30 min.
(2) Resolution of pain in relation to duration of opioid analgesia. Although immediate release morphine may have analgesic effects for up to 4 h, spontaneous and movement-related pain can be resolved in <30 min in 50% of patients, rendering the patient more susceptible to opioid side-effects.
(3) Evidence of poor opioid-responsiveness of some aspects of the underlying neurophysiology of the spontaneous and movement-related pain components.[39 88 91]

The combination of the above factors indicates that opioid adverse effects, especially sedation, are more likely to dominate over analgesia. In any pain syndrome where there are sudden, short-lived, peaks of pain over and above a stable background pain, the opioid adverse effects are more likely to be problematic.

Opioid toxicity

Opioid toxicity is a spectrum which at one extreme may involve sleepiness, poor concentration, vivid dreams, or both, whereas at the other extreme may involve hallucinations, confusion, agitation, or hypoactive delirium. A survey of inpatients in a cancer centre showed 50% of patients to have some symptoms of opioid toxicity, most of which were not identified by staff.[70] Significant opioid toxicity is associated with a 50% mortality and it is known that in those patients who recover from significant toxicity, the distress experienced is profound. It is therefore very important to avoid opioid toxicity and to develop treatments that minimize this risk.

Currently, non-steroidal anti-inflammatory drugs (NSAIDs) and palliative radiotherapy form the key strategies which complement simple or opioid analgesia in CIBP relief. The evidence base for these strategies along with emerging newer strategies is discussed below; however, a major problem with the evidence base is the methodology of CIBP assessment. There is almost no information on which components of CIBP are helped by which treatments.

Current management of CIBP (Fig. 1)

The standard approach to the management of CIBP is a combination of analgesia and radiotherapy. Although it is accepted that radiotherapy is the gold standard treatment for pain relief in CIBP, there are a significant number of patients who fail to receive adequate analgesia. External beam radiotherapy, whether single or multiple fractions, produced 50% pain relief in 41% of patients and complete pain relief at 1 month in 24% of patients.[41] However, many patients are too frail to attend for palliative radiotherapy or it is too late to reasonably expect pain relief before death.

We know that current therapeutic regimens leave up to 45% of patients with inadequate and under-managed pain control.[10 45]

For such reasons, an improved understanding of the pathophysiological mechanisms underlying, in particular, spontaneous pain at rest and movement-related pain are important. The development of effective pharmacological interventions to act as adjuvants or synergists to palliative radiotherapy to improve the degree of pain relief, in addition to providing analgesia to those too unwell to benefit from palliative radiotherapy, is an important area of research.

Current analgesic therapies for CIBP have not altered significantly in over a decade, since the introduction of bisphosphonates. Opioid-based therapy does remain the basis for most analgesia in CIBP. In theory, there are two aspects to the optimization of opioid analgesia: first, establishing the best opioid for an individual based on the pharmacogenomic principle of interindividual variation in balance between analgesia and side-effects and, secondly, assessing the optimal pharmacokinetic:pharmacodynamic profile from the opioids available. There is more weight given to the first aspect in relation to chronic background pain, but more weight given to the second in relation to spontaneous pain at rest and movement-related pain.

A subgroup of patients get uncontrolled CIBP only after considerable and predictable movement, such as a long walk. This group is often satisfied to use a standard opioid such as normal release morphine, 30 min in advance of such activity. Conversely, in those patients whose pain is unpredictable and of fast onset, unacceptable side-effects are likely to occur with standard opioids. In such cases, a faster, short-acting opioid is more appropriate. Most of the evidence for opioids with faster onset of analgesia and faster peak plasma analgesic levels is limited to fentanyl.[94]

It is not uncommon for patients to use a sustained release opioid preparation such as morphine for background bone pain and either immediate release morphine or a fentanyl preparation for spontaneous pain and movement-related pain.

Non-steroidal anti-inflammatory drugs

The use of NSAIDs in CIBP has been questioned due to the lack of robust, clinical evidence. The three randomized trials of NSAIDs in cancer pain do not separate out bone metastases, and six non-randomized trials mention bone metastases but do not record incident pain.[14 16 46 47 72 73 75 83] The newer cyclooxygenase (COX)-2 (an inducible isoform of COX enzyme involved in prostaglandin synthesis) specific inhibitors may in theory be of greater therapeutic potential due to their anti-tumour/antiangiogenic properties.[69 74] In an animal model of CIBP, acute treatment

with a highly selective COX-2 inhibitor attenuated both background and movement-induced pain, whereas chronic treatment additionally reduced tumour burden and osteoclast destruction.[63] Clearly, the use and availability of COX-2 inhibitors has fluctuated because of concerns with some drugs within this class. Most clinicians regard NSAIDs as an important part of CIBP management unless contraindications exist.

Bisphosphonates

There is some evidence for the use of bisphosphonates in bone metastases, with one review showing that regular use of bisphosphonates reduced the number of skeletal-related events in numerous cancers.[61] A Cochrane review of the clinical efficacy of bisphosphonates for pain relief in metastatic bone disease suggested that there was some evidence for their use as analgesics, although the effect was delayed. The number needed to treat to achieve 50% pain relief at 4 weeks was 11, falling to 7 at 12 weeks.[89]

Although bisphosphonates form part of standard therapy for the prevention of skeletal events in some cancers, the role of bisphosphonates in immediate pain relief is less well defined. This highlights the problems in extracting evidence from studies where there are wide variations in quantifying quality of life and bone pain. Multiple different methods have been used to assess quality of life and pain, making it difficult to compare results between different studies. This has prevented the effective collation of evidence.

Analysis of the publications available to date has not revealed any great differences between individual bisphosphonates given at standard doses, but objective assessment is complicated by differences in study design, measurement methods, and statistical analyses. Comparative studies will be needed to help resolve this.

A Cochrane review in 2000 concluded that there is evidence to support the effectiveness of bisphosphonates in providing some pain relief for CIBP but that there is insufficient evidence to recommend bisphosphonates for immediate effect as first line therapy. Evidence defining the most effective bisphosphonates or their relative effectiveness for different primary neoplasms is also limited.[89] However, although a clearer evidence base for these areas is eagerly awaited, it is reasonable to consider bisphosphonates for pain relief in CIBP where analgesics, radiotherapy, or both are inadequate.

Glutamate inhibitors and N-methyl-D-aspartate antagonists

On the basis that CIBP has neuropathic and inflammatory components and clinical and laboratory evidence of central wind-up, it is not surprising that inhibitors of glutamate release have been considered and investigated. It is known that in animal studies, gabapentin reverses dorsal horn changes associated with CIBP resulting in relief of spontaneous and movement-related pain.[12] Clinical studies are currently underway with pregabalin, a more potent inhibitor of glutamate release and the hypothesis is that this class of drug may provide very useful adjuvant analgesia to standard care.

Inhibitors of the *N*-methyl-D-aspartate (NMDA) glutamate receptor complex may also be of interest, especially as NMDA subtype inhibitors are developed. At present the non-specific NMDA antagonist, ketamine, is used in some difficult to manage cases.

Osteoprotegerin

Osteoprotegerin (OPG) is a promising target, which may act through reduction of osteoclast function to diminish tumour-induced bone destruction. Indeed, early clinical work with OPG is interesting and may hold future promise.[33] A single SC dose of AMGN-0007 (a recombinant construct of OPG) suppressed bone resorption as indicated by a rapid, sustained, and profound decrease of urinary *N*-telopeptide of collagen (NTX)/creatinine in multiple myeloma and breast carcinoma patients. Changes were comparable with those with pamidronate.

Endothelin-1 antagonists

Androgen refractory prostate cancer continues to evade effective treatment. The potent vasoconstrictor endothelin-1 is produced by prostate cancer and appears to have a role in prostate cancer progression and morbidity. On the basis of preclinical and clinical trial data, the endothelin axis is emerging as potentially important in this response.[19] Drugs targeting the endothelin axis, such as the potent ET(A) receptor antagonists atrasentan, have been studied in large clinical trials and appears to have an impact on disease progression and morbidity. The role of the endothelin axis in prostate cancer deserves further investigation in the laboratory and clinic. Laboratory studies of ET(A) in CIBP have demonstrated analgesic effects on both background and movement-evoked pain.[53]

Future concepts

Alpha-v-beta3 ($\alpha v\beta 3$) integrin blocker

The integrin $\alpha v\beta 3$ mediates cell–matrix interactions.[49] Vitaxin®, a humanized monoclonal antibody that blocks human and rabbit $\alpha v\beta 3$ integrins, is in clinical trials for metastatic melanoma and prostate cancer. Vitaxin® decreases bone resorption by impairing osteoclast attachment, without affecting osteoclast multinucleation. Data also show that the inhibitory effects of Vitaxin® on osteoclasts can be modulated by factors known to alter the conformation of $\alpha v\beta 3$. The effects of Vitaxin® on reducing osteoclast activity may have future clinical utility in the management of CIBP.

The fundamental clinical issues in moving CIBP management forwards are:

(i) the temporal characteristics of spontaneous and movement-related pain;
(ii) the onset of action of opioid analgesia;
(iii) opioid adverse effects;
(iv) emerging evidence of intrinsically poorer opioid responsiveness of spontaneous and movement-related bone pain.

Animal models of CIBP

Until relatively recently, it was difficult to study the pain associated with bone metastases as the systemic models of cancer have much more widespread effects making the underlying CIBP difficult to evaluate. The emergence of focal bone metastasis models, displaying behavioural signs compatible with the clinical syndrome, has meant that our understanding of the underlying mechanism of this chronic pain syndrome has advanced significantly in the last decade.

Earlier models of CIBP were reliant upon the systemic[2 64] or i.m.[32] injection of carcinoma cells, which can result in more than one randomly sited bone metastasis, with the associated effects of disseminated malignancy. These systemic CIBP models made investigation of the mechanisms of CIBP difficult to assess. A model more closely linked to the human clinical condition in terms of both pain development and bone destruction was required. Schwei and colleagues first described a murine model of CIBP, whereby the injected tumour cells are confined to the marrow of the femur without invading adjacent soft tissues. After injection, the tumour cells proliferate resulting in extensive tumour-induced bone destruction and development of pain-related behaviours,[65] in parallel with the clinical condition.[27 33] This model has subsequently been developed for use with different carcinoma cell lines, bones and for use in rats.[42 62 86]

Numerous factors involving peripheral tissues and the central nervous system have been shown to be involved in the development and maintenance of CIBP. Recent studies have shown that mineralized bone, marrow, and the periosteum are all innervated by sensory neurons and post-ganglionic sympathetic neurons.[25 36] Although the tumour itself may not be highly innervated by sensory neurons,[50 66 80] it is thought that rapid tumour growth in the marrow may lead to injury of these nerves. Indeed, a recent study has shown that as tumour cells grow within the bone, they come into contact, injure, and then destroy the distal processes of sensory fibres that innervate the bone marrow and mineralized bone.[53] To identify the sensory fibres that innervate the marrow, mineralized bone, and periosteum, several markers were employed including calcitonin gene-related peptide (CGRP) to label peptidergic sensory neurons and isolectin B4 (IB4), which labels non-peptidergic sensory neurons.[36] The sensory neurons that innervate the marrow and mineralized bone appear to be largely CGRP-containing (and IB4-negative), although a subpopulation may also co-express the tachykinins, substance P (SP) and neurokinin A.[36] Recent proteomic analysis of spinal cord showed increased CGRP, but not SP levels ipsilateral to CIBP injury.[52] This contrasts with models of surgical peripheral nerve injury, where a down-regulation of CGRP and SP expression in the injured sensory neurons is observed[38 48] and in models of inflammatory pain, where both of these neurotransmitters increase.[17]

Tumour-induced proliferation and hypertrophy of osteoclasts in bone may also contribute to development of CIBP. Osteoclasts destroy bone by maintaining a highly acidic microenvironment surrounding themselves and mineralized bone.[4] The use of OPG, a decoy receptor which reduces osteoclast function through binding to the OPG ligand (known as the receptor for activator of NF-kB ligand, RANKL),[59 71 92] has resulted in a reduction in CIBP-induced pain behaviours in the mouse.[26 33]

Transient receptor potential channel, TRPV1 (or vanilloid receptor 1), and the acid-sensing ion channels (ASICs) are activated by a decrease in pH[87] and are expressed by the sensory neurons that innervate bone,[22 51] suggesting that inactivation of TRPV1 or ASIC may reduce CIBP by blocking the excitation of sensory neurons. Following inflammation or tissue injury, sensory neurons have a higher expression of TRPV1 induced by nerve growth factor (NGF).[90] Osteoclast-induced bone resorption can induce the release of growth factors,[21 93] which can directly activate nociceptors.[55 60] Reducing osteoclast activity and thereby inhibiting bone resorption with bisphosphonates has been demonstrated to reduce CIBP-induced pain behaviours.[68]

Numerous other cells within the tumour site, for example, immune cells, including macrophages, neutrophils, and T-cells, can secrete various factors that can directly act on sensory neurons, such as prostaglandins, cytokines, endothelins, and growth factors.[1 37 40] In addition to the tumour, immune, and inflammatory cells activated by CIBP, there is a prominent activation of glial cells, such as astrocytes, in the spinal cord, which is only observed ipsilateral to injury in the segments of the spinal cord that receive sensory innervation from the affected bone.[65] Growth factors released as a result of CIBP, such as NGF, may play numerous roles, such as directly activating sensory neurons and modulating the expression for example, of various neurotransmitters (SP and CGRP), ion channels (TRPV1, ASIC3, the purinergic receptors, such as P2X or sodium channels), and stress-activated transcription factors, such as activating transcription factor 3.[11 30 58 84] Anti-NGF therapy in a CIBP model was found to reverse both early- and late-stage CIBP pain-related behaviours and to be more efficacious than the

acute administration of morphine.[23 67] Although several cytokines can be released, such as interleukin-1, tumour necrosis factor-alpha from both microglia and astrocytes,[79] and immune cells, the effects of their inhibition in CIBP has yet to be assessed. Cancer cells and macrophages associated with tumours appear to express high levels of COX-2. Prostaglandins can directly activate sensory nociceptor neurons by binding to several prostanoid receptors.[82] Drugs that target prostaglandins and endothelins have been evaluated in CIBP. The use of COX-2-specific inhibitors appears to alleviate some aspects of CIBP and may also slow down the cancer progression.[15] A recent study has shown that administration of an endothelin A receptor antagonist attenuated both ongoing and movement-evoked CIBP without affecting either tumour growth or bone destruction.[54]

The destruction of bone observed in CIBP may also add to the development of pain. As the disease progresses, the mechanical strength of the bone is compromised and may lead to fractures.[34] Previous studies have illustrated that mechanical distortion of the periosteum may be a major source of pain, perhaps contributed to by the dense meshwork of CRGP containing sensory neurons on the periosteum.[36 39 43]

CIBP appears to be mechanistically distinct compared with neuropathic or inflammatory pain states, where major differences occur in the cellular and neurochemical changes in the nervous system. There is a prominent up-regulation of glial cells in the spinal cord ipsilateral to CIBP.[65] Whilst no change in sensory neurotransmitters, SP or CGRP expression was observed ipsilateral to CIBP[52] (compared with the contralateral [uninjured] CIBP spinal cord).[28] Notably, there was no reported change in the sensory neurotransmitters, galanin, or neuropeptide-Y after CIBP, whereas they are markedly up-regulated after peripheral nerve-injury,[28 29] although a more recent study has shown an increase in galanin in the sensory neurons ipsilateral to CIBP.[53] Collectively, these data indicate that CIBP is clearly a distinct entity not a chronic pain state due entirely to either inflammation or neuropathy.

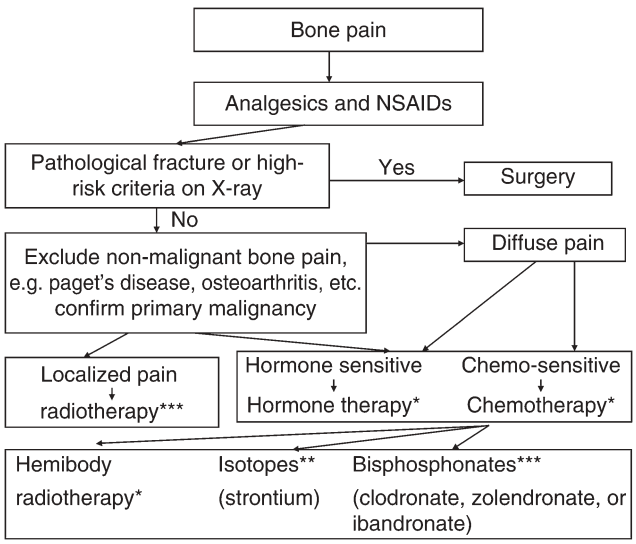

Fig 1 Overview of the management of metastatic bone pain. *Non-randomized controlled trials, cohort study etc.; **one or more well-designed randomized controlled trials; ***systematic review or mega-analysis. This figure is attributed to P. Hoskin.

More than 50% of patients with malignant bone pain suffer unacceptable opioid side-effects, making pharmacological management difficult.[70] A recent study has shown that pain-related behaviours, such as movement-evoked pain, are actually relatively resistant to opioid treatment, with 10-fold higher doses of morphine required when compared with those for chronic inflammatory pain states in animal models.[35] Another animal study showed worsening pain with continued opioid use.[31] Indeed, a recent report illustrates that in CIBP patients who become tolerant to their current opioid therapy with a resultant escalation of their cancer-induced pain,[24] increasing the opioid dose can worsen, rather than ease the pain.[9]

Radiotherapy (XRT), as the current standard treatment for CIBP, has been studied in a mouse model with a single fraction (6 Gy) resulting in functional improvement.[18] This was enhanced by combined treatment with the NSAID, ketorolac, suggesting that such low doses of irradiation may not give maximal analgesia.[85] How XRT results in analgesia is poorly understood, although it is likely to be due to a combination of peripheral and central effects, including alterations in nociceptive processing in the central nervous system. After irradiation, no differences were observed in peripheral tissues, such as tumour size, inflammatory cytokines, or osteoclast activity, whereas spinal changes were evident, with a reduction in spinal glial activity, dynorphin, COX-2, and CGRP expression.[52 85]

Urch and colleagues[81] first characterized the magnitude of dorsal horn neuronal responses after CIBP, which was found to be hyperexcitable compared with sham-operated animals. This excitability was driven by a population of wide dynamic range lamina I neurons that respond to both noxious and innocuous stimuli. Gabapentin has been shown to reduce CIBP-induced dorsal horn neuronal responses when administered acutely or chronically, with alterations in CIBP-induced pain behaviours observed with chronic administration, thereby suggesting a possible use for gabapentin in the treatment of CIBP.[12] Changes in the excitability of dorsal horn neurons contributing to central sensitization may be dependent not only on spinal circuits but also control from higher centres.[77] The descending $5HT_3$ system is excitatory and has been shown to be enhanced after some forms of inflammation (formalin) and peripheral nerve injury,[20 57 77] indicating that activation of this descending facilitatory pathway may contribute to spinal excitability.[13] This system is thought to be driven by a population of NK1 positive lamina I neurons, thus creating a loop that enhances nociceptive processing. This also receives input from higher centres involved in the affective and autonomic aspects of pain,[76] which given the

correlation of increased anxiety and depression in the clinical CIBP condition may be a possible mechanism of the emotional condition of the patient affecting their experience of pain.[43 78]

Conclusions and future work

The underlying mechanisms of CIBP appear to be complex and to have both an inflammatory and tumourigenic component.[53] Understanding the role played by inflammatory cells, tumour-secreted factors, and the injury to sensory neurons that innervate the bone, in addition to the changes in spinal glia and neurons after CIBP, may aid the development of novel efficacious mechanism-based therapies.

It is clear that clinical CIBP is complex and that optimum treatment in the future is likely to be multimodal. Clarity of thought about future CIBP management depends on the appropriate design of clinical trials, in particular in relation to patient and pain assessments and outcome measures. In addition, the information from basic science is increasing rapidly, with an excellent animal model of CIBP. Close basic science and clinical collaboration will help to inform the direction of clinical research. It is also possible that optimum management of CIBP will be individualized not just on pain characteristics but on primary tumour site.

Funding

Cancer Research, UK; *British Journal of Anaesthesia*/ Royal College of Anaesthetists; The Melville Trust for the Case and Cure of Cancer.

References

1 Alvarez FJ, Fyffe RE. Nociceptors for the 21st century. *Curr Rev Pain* 2000; **4**: 451–8

2 Arguello F, Baggs RB, Frantz CN. A murine model of experimental metastasis to bone and bone marrow. *Cancer Res* 1988; **48**: 6876–81

3 Body JJ, Greipp P, Coleman RE, *et al*. A phase I study of AMGN-0007, a recombinant osteoprotegerin construct, in patients with multiple myeloma or breast carcinoma related bone metastases. *Cancer* 2003; **97**: 887–92

4 Boyle WJ, Simonet WS, Lacey DL. Osteoclast differentiation and activation. *Nature* 2008; **423**: 337–42

5 Bruera E, Kim HN. Cancer pain. *JAMA* 2003; **290**: 2476–9

6 Coleman RE. Metastatic bone disease: clinical features, pathophysiology and treatment strategies. *Cancer Treat Rev* 2001; **27**: 165–76

7 Coleman RE, Rubens RD. The clinical course of bone metastases from breast cancer. *Br J Cancer* 1987; **55**: 61–6

8 Coleman RE, Rubens RD. Bone metastases and breast cancer. *Cancer Treat Rev* 1985; **12**: 251–70

9 Davis MP, Shaiova LA, Angst MS. When opioids cause pain. *J Clin Oncol* 2007; **25**: 4497–8

10 de Wit R, van Dam F, Loonstra S, *et al*. The Amsterdam Pain Management Index compared to eight frequently used outcome measures to evaluate the adequacy of pain treatment in cancer patients with chronic pain. *Pain* 2001; **91**: 339–49

11 Donnerer J, Schuligoi R, Stein C. Increased content and transport of substance P and calcitonin gene-related peptide in sensory nerves innervating inflamed tissue: evidence for a regulatory function of nerve growth factor in vivo. *Neuroscience* 1992; **49**: 693–8

12 Donovan-Rodriguez T, Dickenson AH, Urch CE. Gabapentin normalizes spinal neuronal responses that correlate with behavior in a rat model of cancer-induced bone pain. *Anesthesiology* 2005; **102**: 132–40

13 Donovan-Rodriguez T, Urch CE, Dickenson AH. Evidence of a role for descending serotonergic facilitation in a rat model of cancer-induced bone pain. *Neurosci Lett* 2006; **393**: 237–42

14 Estape J, Vinolas N, Gonzalez B, *et al*. Ketorolac, a new non-opioid analgesic: a double-blind trial versus pentazocine in cancer pain. *J Int Med Res* 1990; **18**: 298–304

15 Fosslien E. Biochemistry of cyclooxygenase (COX)-2 inhibitors and molecular pathology of COX-2 in neoplasia. *Crit Rev Neurobiol* 2000; **37**: 41–502

16 Fuccella LM, Conti F, Corvi G, Mandelli V, Randelli M, Stefanelli G. Doubleblind study of the analgesic effect of indoprofen (K 4277). *Clin Pharmacol Ther* 1975; **17**: 277–83

17 Galeazza MT, Garry MG, Yost HJ, Strait KA, Hargreaves KM, Seybold VS. Plasticity in the synthesis and storage of substance P and calcitonin gene-related peptide in primary afferent neurons during peripheral inflammation. *Neuroscience* 1995; **66**: 443–58

18 Goblirsch M, Mathews W, Lynch C, *et al*. Radiation treatment decreases bone cancer pain, osteolysis and tumor size. *Radiat Res* 2004; **161**: 228–34

19 Gramoun A, Shorey S, Bashutski JD, *et al*. Effects of Vitaxin, a novel therapeutic in trial for metastatic bone tumors, on osteoclast functions in vitro. *J Cell Biochem* 2007; **102**: 341–52

20 Green GM, Scarth J, Dickenson A. An excitatory role for 5-HT in spinal inflammatory nociceptive transmission; state-dependent actions via dorsal horn 5-HT(3) receptors in the anaesthetized rat. *Pain* 2000; **89**: 81–8

21 Guise TA. Molecular mechanisms of osteolytic bone metastases. *Cancer* 2000; **88**: 2892–8

22 Guo A, Vulchanova L, Wang J, Li X, Elde R. Immunocytochemical localization of the vanilloid receptor 1 (VR1): relationship to neuropeptides, the P2X(3) purinoceptor and IB4 binding sites. *Eur J Neurosci* 1999; **11**: 946–58

23 Halvorson KG, Kubota K, Sevcik MA, *et al*. A blocking antibody to nerve growth factor attenuates skeletal pain induced by prostate tumor cells growing in bone. *Cancer Res* 2005; **65**: 9426–35

24 Hanks GW, Conno F, Cherny N, *et al*. Expert Working Group of the Research Network of the European Association for Palliative Care. Morphine and alternative opioids in cancer pain: the EAPC recommendations. *Br J Cancer* 2001; **84**: 587–93

25 Hill EL, Elde R. Distribution of CGRP-, VIP-, D beta H-, SP-, and NPY-immunoreactive nerves in the periosteum of the rat. *Cell Tissue Res* 1991; **264**: 469–80

26 Honore P, Luger NM, Sabino MAC, *et al*. Osteoprotegerin blocks bone cancer-induced skeletal destruction, skeletal pain and pain-related neurochemical reorganization of the spinal cord. *Nat Med* 2000; **6**: 521–8

27 Honore P, Mantyh PW. Bone cancer pain: from mechanism to model to therapy. *Pain Med* 2000; **1**: 303–9

28 Honore P, Rogers SD, Schwei MJ, *et al*. Murine models of inflammatory, neuropathic and cancer pain each generates a unique set of neurochemical changes in the spinal cord and sensory neurons. *Neuroscience* 2000; **98**: 585–98

29 Honore P, Schwei J, Rogers SD, *et al*. Cellular and neurochemical remodeling of the spinal cord in bone cancer pain. *Prog Brain Res* 2000; **129**: 389–97

30 Ji RR, Samad TA, Jin SX, Schmoll R, Woolf CJ. p38 MAPK activation by NGF in primary sensory neurons after inflammation increases TRPV1 levels and maintains heat hyperalgesia. *Neuron* 2002; **36**: 57–68

31 King T, Vardanyan A, Majuta L, *et al*. Morphine treatment accelerates sarcoma induced bone pain, bone loss, and spontaneous fracture in a murine model of bone cancer. *Pain* 2007; **132**: 154–68

32 Kostenuik PJ, Orr FW, Suyama K, Singh G. Increased growth rate and tumor burden of spontaneously metastatic Walker 256 cancer cells in the skeleton of bisphosphonate-treated rats. *Cancer Res* 1993; **53**: 5452–7

33 Luger NM, Honore P, Sabino MA, *et al*. Osteoprotegerin diminishes advanced bone cancer pain. *Cancer Res* 2001; **61**: 4038–47

34 Luger NM, Mach DB, Sevcik MA, Mantyh PW. Bone cancer pain: from model to mechanism to therapy. *J Pain Symptom Manage* 2005; **29**: S32–46

35 Luger NM, Sabino MAC, Schwei MJ, *et al*. Efficacy of systemic morphine suggests a fundamental difference in the mechanisms that generate bone cancer vs. inflammatory pain. *Pain* 2002; **99**: 397–406

36 Mach DB, Rogers SD, Sabino MC, *et al*. Origins of skeletal pain: sensory and sympathetic innervation of the mouse femur. *Neuroscience* 2002; **113**: 155–66

37 Mantyh PW, Clohisy DR, Koltzenburg M, Hunt SP. Molecular mechanisms of cancer pain. *Nat Rev Cancer* 2002; **2**: 201–9

38 Marchand JE, Wurm WH, Kato T, Kream RM. Altered tachykinin expression by dorsal-root ganglion neurons in a rat model of neuropathic pain. *Pain* 1994; **58**: 219–31

39 Martin CD, Jimenez-Andrade JM, Ghilardi JR, *et al*. Organization of a unique net-like meshwork of CGRP+ sensory fibers in the mouse periosteum: implications for the generation and maintenance of bone fracture pain. *Neurosci Lett* 2007; **427**: 148–52

40 McMahon SB, Jones NG. Plasticity of pain signaling: role of neurotrophic factors exemplified by acid-induced pain. *J Neurobiol* 2004; **61**: 72–87

41 McQuay HJ, Collins SL, Carroll D, Moore RA. Radiotherapy for the palliation of painful bone metastases. *Cochrane Database Syst Rev* 2000; CD001793

42 Medhurst S, Bowes M, Kidd BL, *et al*. Antinociceptive effects of the bisphosphonate, zoledronic acid, in a novel rat model of bone cancer pain. *Br J Pharmacol* 2001; **134**: 156P

43 Mercadante S. Malignant bone pain: pathophysiology and treatment. *Pain* 1997; **69**: 1–18

44 Mercadante S, Maddaloni S, Roccella S, Salvaggio L. Predictive factors in advanced cancer pain treated only by analgesics. *Pain* 1992; **50**: 151–5

45 Meuser T, Pietruvk C, Radbruch L, Stute P, Lehmann KA, Grond S. Symptoms during cancer pain treatment following WHO-guidelines: a longitudinal follow-up study of symptom prevalence, severity and etiology. *Pain* 2001; **91**: 247–57

46 Minotti V, de Angelis V, Righetti E, *et al*.. Double-blind evaluation of short-term analgesic efficacy of orally administered diclofenac, diclofenac plus codeine, and diclofenac plus imipramine in chronic cancer pain. *Pain* 1998; **74**: 133–7

47 Minotti V, Patoia L, Roila F, *et al*. Double-blind evaluation of analgesic efficacy of orally administered diclofenac, nefopam, and acetylsalicylic acid (ASA) plus codeine in chronic cancer pain. *Pain* 1989; **36**: 177–83

48 Nahin RL, Ren K, De León M, Ruda M. Primary sensory neurons exhibit altered gene expression in a rat model of neuropathic pain. *Pain* 1994; **58**: 95–108

49 Nelson JB. Endothelin inhibition: novel therapy for prostate cancer. *J Urol* 2003; **170**: S65–7

50 O'Connell JX, Nanthakumar SS, Nielsen GP, Rosenberg AE. Osteoid osteoma: the uniquely innervated bone tumor. *Mod Pathol* 1998; **11**: 175–80

51 Olson TH, Riedl MS, Vulchanova L, Ortiz-Gonzalez XR, Elde R. An acid sensing ion channel (ASIC) localizes to small primary afferent neurons in rats. *Neuroreport* 1998; **9**: 1109–13

52 Park HC, Seong J, An JH, Kim J, Kim UJ, Lee BW. Alteration of cancer pain-related signals by radiation: proteomic analysis in an animal model with cancer bone invasion. *Int J Radiat Oncol Biol Phys* 2005; **61**: 1523–34

53 Peters CM, Ghilardi JR, Keyser CP, *et al*. Tumor-induced injury of primary afferent sensory nerve fibers in bone cancer pain. *Exp Neurol* 2005; **193**: 85–100

54 Peters CM, Lindsay TH, Pomonis JD, *et al*. Endothelin and the tumorigenic component of bone cancer pain. *Neuroscience* 2004; **126**: 1043–52

55 Poon RT, Fan ST, Wong J. Clinical implications of circulating angiogenic factors in cancer patients. *J Clin Oncol* 2001; **19**: 1207–25

56 Portenoy RK, Payne D, Jacobsen P. Breakthrough pain: characteristics and impacts in patients with cancer pain. *Pain* 1999; **82**: 263–74

57 Rahman W, Suzuki R, Webber M, Hunt SP, Dickenson AH. Depletion of endogenous spinal 5-HT attenuates the behavioural hypersensitivity to mechanical and cooling stimuli induced by spinal nerve ligation. *Pain* 2006; **123**: 264–74

58 Ramer MS, Bradbury EJ, McMahon SB. Nerve growth factor induces P2X(3) expression in sensory neurons. *J Neurochem* 2001; **77**: 864–75

59 Rodan GA, Martin TJ. Therapeutic approaches to bone diseases. *Science* 2000; **289**: 1508–14

60 Roman C, Saha D, Beauchamp R. TGF-beta and colorectal carcinogenesis. *Microsc Res Tech* 2001; **52**: 450–7

61 Ross JR, Saunders Y, Edmonds PM, Patel S, Broadley KE, Johnston SR. Systematic review of role of bisphosphonates on skeletal morbidity in metastatic cancer. *Br Med J* 2003; **327**: 469

62 Sabino MA, Luger NM, Mach DB, Rogers SD, Schwei MJ, Mantyh PW. Different tumors in bone each give rise to a distinct pattern of skeletal destruction, bone cancer-related pain behaviors and neurochemical changes in the central nervous system. *Int J Cancer* 2003; **104**: 550–8

63 Sabino MAC, Ghilardi JR, Jongen JLM, *et al*. Simultaneous reduction in cancer pain, bone destruction, and tumor growth by selective inhibition of cyclooxygenase-2. *Cancer Res* 2002; **62**: 7343–9

64 Sasaki A, Boyce BF, Story B, *et al*. Bisphosphonate risedronate reduces metastatic human breast cancer burden in bone in nude mice. *Cancer Res* 1995; **55**: 3551–7

65 Schwei MJ, Honore P, Rogers SD, *et al*. Neurochemical and cellular reorganization of the spinal cord in a murine model of bone cancer pain. *J Neurosci* 1999; **19**: 10886–97

66 Seifert P, Spitznas M. Tumours may be innervated. *Virchows Arch* 2001; **438**: 228–31

67 Sevcik MA, Ghilardi JR, Peters CM, *et al*. Anti-NGF therapy profoundly reduces bone cancer pain and the accompanying increase in markers of peripheral and central sensitization. *Pain* 2005; **115**: 128–41

68 Sevcik MA, Luger NM, Mach DB, *et al*. Bone cancer pain: the effects of the bisphosphonate alendronate on pain, skeletal remodeling, tumor growth and tumor necrosis. *Pain* 2004; **111**: 169–80

69 Sheng H, Shao J, Kirkland SC, *et al*. Inhibition of human colon cancer cell growth by selective inhibition of cyclooxygenase-2. *J Clin Invest* 1997; **99**: 2254–9

70 Sherry K, Fallon M. Opioid toxicity in cancer patients. *Research Congress in Palliative Care* 2000

71 Simonet WS, Lacey DL, Dunstan CR, *et al*. Osteoprotegerin: a novel secreted protein involved in the regulation of bone density. *Cell* 1997; **89**: 309–19

72 Stambaugh J, Drew J. A double-blind parallel evaluation of the efficacy and safety of a single dose of ketoprofen in cancer pain. *J Clin Pharm* 1988; **28**: S34–9

73 Staquet MJ. A double-blind study with placebo control of intramuscular ketorolac tromethamine in the treatment of cancer pain. *J Clin Pharmacol* 1989; **29**: 1031–6

74 Sumitani K, Kamijo R, Toyoshima T, *et al*. Specific inhibition of cyclooxygenase-2 results in inhibition of proliferation of oral cancer cell lines via suppression of prostaglandin E2 production. *J Oral Pathol Med* 2001; **30**: 41–7

75 Sunshine A, Olson NZ. Analgesic efficacy of ketoprofen in postpartum, general surgery, and chronic cancer pain. *J Clin Pharm* 1988; **28**: S47–54

76 Suzuki R, Morcuende S, Webber M, Hunt SP, Dickenson AH. Superficial NK1-expressing neurons control spinal excitability through activation of descending pathways. *Nat Neurosci* 2002; **5**: 1319–26

77 Suzuki R, Rahman W, Hunt SP, Dickenson AH. Descending facilitatory control of mechanically evoked responses is enhanced in deep dorsal horn neurones following peripheral nerve injury. *Brain Res* 2004; **1019**: 68–76

78 Suzuki R, Rahman W, Rygh LJ, Webber M, Hunt SP, Dickenson AH. Spinal-supraspinal serotonergic circuits regulating neuropathic pain and its treatment with gabapentin. *Pain* 2005; **117**: 292–303

79 Suzumura A, Ito A, Yoshikawa M, Sawada M. Ibudilast suppresses TNFalpha production by glial cells functioning mainly as type III phosphodiesterase inhibitor in the CNS. *Brain Res* 1999; **837**: 203–12

80 Terada T, Matsunaga Y. S-100-positive nerve fibers in hepatocellular carcinoma and intrahepatic cholangiocarcinoma: an immunohistochemical study. *Pathol Int* 2001; **51**: 89–93

81 Urch CE, Donovan-Rodriguez T, Dickenson AH. Alterations in dorsal horn neurones in a rat model of cancer-induced bone pain. *Pain* 2003; **106**: 347–56

82 Vasko MR. Prostaglandin-induced neuropeptide release from spinal cord. *Prog Brain Res* 1995; **104**: 367–80

83 Ventafridda V, Martino G, Mandelli V, *et al*. Indoprofen, a new analgesic and antiinflammatory drug in cancer pain. *Clin Pharmacol Ther* 1975; **17**: 284–9

84 Verge VMK, Richardson PM, Wiesenfeld-Hallin Z, Hokfelt T. Differential influence of nerve growth factor on neuropeptide expression in vivo: a novel role in peptide suppression in adult sensory neurons. *J Neurosci* 1995; **15**: 2081–96

85 Vit JP, Ohara PT, Tien DA, *et al*. The analgesic effect of low dose focal irradiation in a mouse model of bone cancer is associated with spinal changes in neuro-mediators of nociception. *Pain* 2006; **120**: 188–201

86 Wacnik PW, Kehl LJ, Trempe TM, Ramnaraine ML, Beitz AJ, Wilcox GL. Tumor implantation in mouse humerus evokes movement-related hyperalgesia exceeding that evoked by intramuscular carrageenan. *Pain* 2003; **101**: 175–86

87 Waldmann R, Champigny G, Bassilana F, Heurteaux C, Lazdunski M. A proton-gated cation channel involved in acid-sensing. *Nature* 1997; **386**: 173–7

88 Walley J, Colvin LA, Fallon MT, Murray G, Clausen E. Characterisation of cancer-induced bone pain. *Proc The British Pain Society* 2006; A45

89 Wong R, Wiffen PJ. Bisphosphonates for the relief of pain secondary to bone metastases. *Cochrane Database Syst Rev* 2002; CD002068

90 Xue Q, Jong B, Chen T, Schumacher MA. Transcription of rat TRPV1 utilizes a dual promoter system that is positively regulated by nerve growth factor. *J Neurochem* 2007; **101**: 212–22

91 Yamamoto J, Kawamata T, Niiyama Y, Omote K, Namiki A. Down-regulation of mu opioid receptor expression within distinct subpopulations of dorsal root ganglion neurons in a murine model of bone cancer pain. *Neuroscience* 2008; **151**: 843–53

92 Yasuda H, Shima N, Nakagawa N, *et al*. Identity of osteoclastogenesis inhibitory factor (OCIF) and osteoprotegerin (OPG): a mechanism by which OPG/OCIF inhibits osteoclastogenesis in vitro. *Endocrinology* 1998; **139**: 1329–37

93 Yoneda T, Hashimoto N, Hiraga T. Bisphosphonate actions on cancer. *Calcif Tissue Int* 2003; **73**: 315–8

94 Zeppetella G, Ribeiro MD. Opioids for the management of breakthrough (episodic) pain in cancer patients. *Cochrane Database Syst Rev* 2006; CD004311