

Analgesic Best Practice for the Use of Animals in Research and Teaching - An Interpretative International Literature Review

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Introduction

Anaesthetic agents developed for humans have been routinely used in a broad range of animal species and there is a substantial literature on anaesthetic drug combinations for use in animals. However, the management of pain following surgical procedures performed on animals has received less attention. Although the clinical use of analgesics has increased substantially in veterinary practice, pain alleviation in experimental animals has lagged behind.

Justifications given for withholding analgesia

There are a number of reasons why research scientists do not routinely use analgesics in their surgical patients. These include the following:

- Most scientists are inherently conservative. They tend to follow the literature and established animal models. Their concerns regarding extraneous factors that might alter or compromise their test results, commonly referred to as ‘non-experimental variables’, are well understood. A frequently expressed traditional scientific view would be *‘morphine or other opioids cannot be given to these animals because the drug may interfere with the measurement of X in the study’*. While we acknowledge that reduction or removal of non-experimental variables is good science, we believe that this traditional scientific view has, all too often, taken priority over animal welfare.
- In general, scientists lack appropriate training and experience in the detection of pain in experimental animals. It does not occur to untrained observers that animals might need pain control. The often heard statement *‘they are walking around, eating and drinking - they run away from me when I try to catch them - they are doing fine’* indicates the level of understanding that many people using laboratory animals have of pain control. We choose to call this attitude the ‘evidence-threshold’ approach. There is insufficient evidence, as determined by these untrained observers, to conclude that the animals are in pain. Put another way, the evidence before them has not reached the threshold level that they expect

- to observe, to be convinced that the animal is in pain. This ‘evidence-threshold’ approach is clearly based on the assumption that the observer can detect signs of pain in the species of animal they work with.
- The withholding of pain medication from experimental animals is frequently perpetuated by the scientific literature. When surgical procedure X is consistently reported in the literature to be performed without any appropriate pain control, scientists who decide to incorporate procedure X into their research study may truly believe that no pain medication is necessary. Many manipulative procedures have been passed down from one generation of scientists to the next, often without any consideration for pain control.
 - Historical practice within a research laboratory is commonly used as a justification to avoid any change in current practice. This is best expressed by the phrase ‘*we have always done it this way and the animals appeared fine, so why should we change things now ?...*’
 - A lack of information and access to drugs is sometimes cited as a reason why preemptive or postoperative analgesics are not used. However, most scientists are well-informed and have a mastery of the current scientific literature in their chosen field. A lack of information should not be considered a satisfactory reason to deny animals the benefit of pain control.
 - The final reason we believe research laboratories do not implement appropriate pain control strategies involves the Animal Ethics Committees (AECs). We believe the AECs have been too accepting of scientists’ justifications for a lack of progress on implementation of standard veterinary pain management.

This issue of pain control can be compared to the current use of antibiotics by scientists. Frequently when scientists administer antibiotics, they are given post-operatively, whereas best practice procedures recommend that antibiotics be given pre-operatively in the first instance. This is done to ensure that therapeutic blood levels are produced at the time the surgical wound is open, because contamination of the wound is most likely during the surgery. Additional antibiotic may be required during the post-operative period.

In both situations, the use of pain control and that of antibiotics, the scientist is not familiar with laboratory animal best practice procedures. The fact that animal welfare may be seriously compromised by a lack of knowledge regarding these practices places increased responsibility on the AEC to ensure that all animals are given the benefit of current drug therapy using best practice.

Despite the historical justifications for the lack of analgesia in experimental animals, it is now widely accepted that an animal’s ability to perceive pain is equivalent to that of humans. A cornerstone of any concern for animal welfare is the administration of drugs to control pain. It should be noted that the National Animal Ethics Advisory Committee (NAEAC) has a policy: ‘*that analgesic use in research, testing and teaching should mirror that use in clinical veterinary practice and human medicine*’.

Anaesthetics as analgesics

In the research setting at the present time, anaesthetics are the most common class of drugs used to control pain, principally the pain associated with performing surgical procedures. A basic Medline literature search of the common animal models that involve routine surgical procedures will show that the most common anaesthetic used in rodents is still the barbiturate pentobarbitone sodium, also often known as Nembutal. This drug is generally administered by the intraperitoneal (IP) route in rodents. Inhalation anaesthesia of rodents with diethyl ether would probably be a close second, frequently administered by the open drop method using a nose cone or bell jar. The exceptions to this broad generalisation would include studies that investigate pain mechanisms, or studies on large animals that undergo sophisticated surgical manipulations such as organ transplantation.

Most published research papers these days include a statement that confirms that the work was first approved by an Animal Ethics Committee (AEC), an Institutional Animal Care and Use Committee (IACUC) or equivalent review committee. Such statements of approval can be interpreted in two different ways. Firstly, the reader could rejoice in the knowledge that the animals subjected to the stated surgical procedure, e.g. laparotomy or femoral fracture repair, were unconscious under general anaesthesia during the procedure, as would be considered humane standard practice throughout the world's research institutions. However, the second and alternative interpretation would be to criticise the AEC and investigator involved for inhumane practice in those studies where post-operative pain was not controlled by the use of analgesic agents, either before, during or after surgery. The simple fact is that most of the single drug anaesthetic regimes used, such as Nembutal or ether, do not provide post-operative pain control. These anaesthetic agents are not analgesics.

This paper explores some strategies to improve the welfare of animals that undergo surgical procedures through the use of alternative analgesic and anaesthetic drugs and combinations of drugs.

Gold standard for analgesia

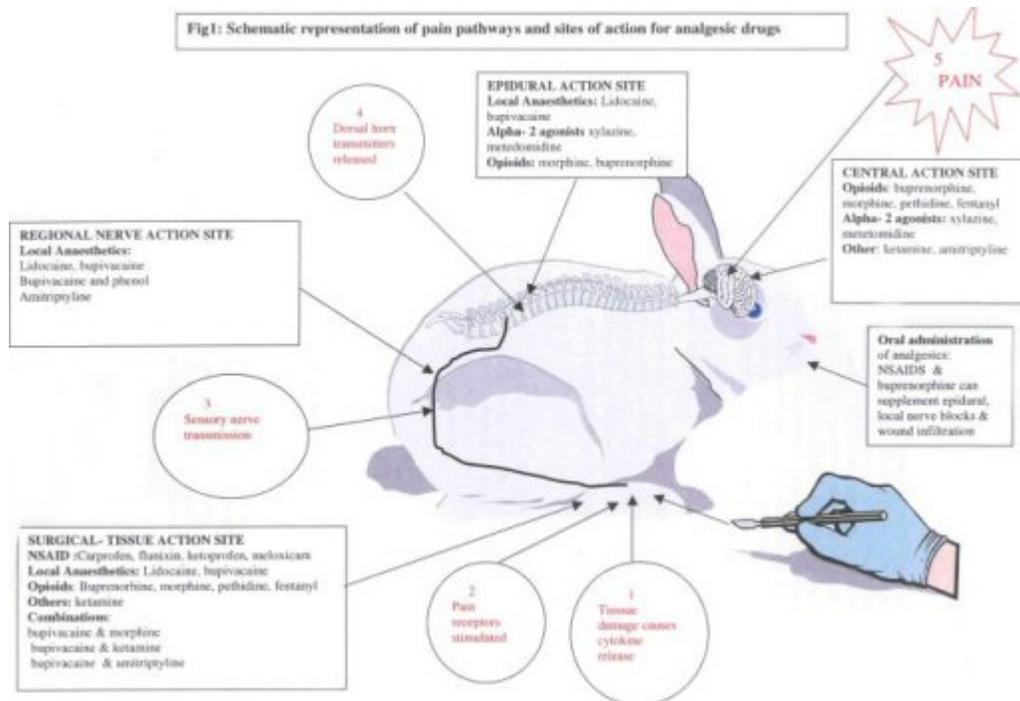
The authors unashamedly hold anthropomorphic views regarding pain control in animals. We reiterate the 'pain equivalence' concept. That is, if the proposed experimental procedure (to be performed on the test animal) were to be performed on a human instead, then the same level of pain control that would be required for that person to comfortably survive and recover from the procedure should be automatically provided to the animal. We would regard this as a gold standard for pain control in animals. When this gold standard is used to judge some of the research papers mentioned above, many experimental manipulations would be condemned as unsatisfactory and some might be considered inhumane. We acknowledge that the reader may consider this as severe and unjust criticism. However, we believe every AEC should review experimental manipulations from the viewpoint of 'pain equivalence'.

Legal requirements

However, quite apart from the moral justifications for use of analgesia, there are legal requirements as well. Many readers will be aware that no animals may be ‘manipulated’ for research, testing or teaching purposes in New Zealand without prior approval by an AEC. The relevant New Zealand legislation, the Animal Welfare Act 1999 provides for the humane care and use of research animals in general, and the need to manage their pain in particular, through the actions and decisions of the AEC. The AEC has the authority to require that appropriate pain control be used as detailed in section 100(d) of the Act. The concept of ‘withholding pain medication’ is useful here. The authors consider that all surgical procedures should be managed with appropriate post-operative pain medication and that a failure to provide such pain relief should be regarded as ‘withholding pain medication’.

Pain Physiology

A basic understanding of the physiology of the pain is an essential part of decision-making in the use of analgesics. Fig 1 shows a schematic representation of the pain pathway with sites of action for analgesic drugs.



Mechanisms of pain

Tissue damage leads to pain. Surgery causes tissue damage and in general, the more the tissue damage, the greater the pain produced. It is important to remember that even in the unconscious patient under general anaesthesia, the spinal cord receives a massive barrage of nerve impulses from the surgical site. These impulses are known as ‘afferent impulses’ as they travel from the peripheral tissues towards the spinal cord. These afferent nerve transmissions are exacerbated when peripheral nerves are cut. The afferent impulses

travel in two basic types of nerve fibres: (1) large diameter A-fibres with a myelin coat that transmit impulses rapidly. A-fibre transmission produces a sharp pricking or stinging pain, and (2) thin diameter C-fibres without myelin that transmit impulses slowly. C-fibre transmission produces a burning long lasting pain.

Peripheral sensitisation

After tissue damage that may be caused by surgery, inflammation or lack of blood supply (ischaemia), a 'biological soup of molecules' is produced. The ingredients of the soup include substance P, calcitonin gene-related peptide, histamine, hydrogen ions, bradykinin, nitric oxide, inflammatory cells, platelets and so on.... The soup causes the afferent nerves to discharge, sending impulses to the spinal cord. This is perceived as pain by the patient. The soup, spreading locally, causes areas adjacent to the site of tissue damage to become involved. As a result, the nerve fibres become more sensitive (i.e. their pain threshold is lowered) and spontaneous firing of afferent impulses occurs. The total effect on the patient is an expanding area of pain, an increase in pain and greater sensitivity to a light touch that would normally not be painful (called allodynia). The soup produces 'peripheral sensitisation.'

Dorsal horn wind-up and central sensitisation

The repeated afferent impulses to the spinal cord, as a result of the sensitising soup at the site of tissue damage, cause the dorsal horn neurones within the spinal cord to become hyperexcitable. This state of hyperexcitability is called central sensitisation. The whole concept of spinal cord nerve cells undergoing repeated stimulation and activation has been termed 'wind up'. Once established, central sensitisation requires high doses of narcotic to suppress it.

It should be remembered that the above sequence of events takes place in the patient during general anaesthesia while the patient is unconscious and unaware of the 'molecular sensitising soup' that is cooking in the peripheral tissues at the site of surgery. But once the anaesthetic has worn off, the patient begins to feel the consequences of the soup-mix and wind-up and experiences the pain of the surgical procedure.

Controlling Pain in Laboratory Animals

The pain felt by a laboratory animal, and therefore the appropriate analgesic regime, will depend on the amount of surgical trauma or tissue damage it has undergone as part of the experimental process, as well as on subsequent environmental influences. The degree of trauma per unit body mass has been suggested as a useful measure to determine the analgesic requirements of the patient. A drug with limited analgesic potency may provide sufficient pain control for ovariohysterectomy in the rat, but would be inadequate for the same procedure in a dog, because the procedure is more invasive and causes greater tissue trauma in this species (59).

The degree of enforced movement may have an effect on the pain animals experience after surgery. Most experimental animals do not enjoy the total post-operative bed-rest afforded to human patients. Caged animals must generally move in order to access food and water. Humans (and presumably animals) can be pain-free at rest, but may experience severe pain upon movement or locomotion. Many readers may be aware of the discomfort and pain experienced by some human patients when nursing staff initiate enforced activity as part of post-operative physiotherapy. In general, animal husbandry practices and rodent cage design do not take this into account. Food and water is frequently placed overhead in the lid of the cage. This requires the animal to stretch up, or in the case of many cages designed for mice, animals have to stand on both hind feet to reach food pellets or water.

Methods used to measure analgesic effectiveness

The methods used to assess the effectiveness of analgesics can influence the interpretation of the results of experimental pain studies. When comparisons are made, the reader should take into account the methods used to determine the level of pain experienced by the animal.

Thermal methods include the hot-plate test and the tail flick test. Rodents are placed on a heated surface in the range 50-55°C and their response is noted. The tail flick test uses radiant heat or immersion in hot water. Mechanical methods involve the application of pressure to a digit, ear or tail and the response is noted. These tests measure analgesic activity of test compounds but do not measure anti-inflammatory activity. Local inflammatory reactions can be produced by the injection of irritant substances such as carrageenan or formalin into the footpad of rodents. The ameliorative effects of anti-inflammatory or analgesic drugs can then be tested.

The mouse writhing test is the most common chemical method of analgesic assessment. Intraperitoneal injections of chemicals such as acetic acid produce writhing and spasm of the abdomen. Electrical methods are also used to produce pain. Rodents may be exposed to an electrical shock through the wire grid cage floor, or applied directly to the tail. Electrical stimulation of the tooth pulp has also been used.

In summary, the thermal methods generally require higher analgesic dose rates to prevent a pain response than other methods, and the duration of analgesia is shorter than for mechanical methods. Therefore it is suggested that drug dosages based on thermal assessment may be more clinically relevant than dosages based on other techniques.

In a research setting, where anaesthetics and analgesics are only used to control pain, the above assessment methods are not relevant to the practical concerns of improving animal welfare by a reduction in animal suffering.

Observation of pain in animals is a challenge and there have been many systems devised to record pain behaviour. These include the verbal rating score (VRS), the simple descriptive scales (SDS), the numerical rating scale (NRS) and the visual analogue scale

(VAS). All such methods are subject to significant variability between observers. The NRS system has been reported to be the most suitable for recording pain assessment in dogs (74).

One of the most useful strategies to judge the effectiveness of analgesic activity, administered either preemptively or post-operatively, is a daily record of food and water consumption and body weight, a straightforward procedure for rodents. These simple measurements can readily demonstrate that surgery has some adverse effect on animals. The use of analgesic drugs can be demonstrated to minimise or reverse these adverse effects. The reasonable conclusion therefore is that pain was responsible for the adverse effects, whether or not the animals were observed to experience pain (40).

In research facilities, the majority of surgical manipulations and post-operative care are performed and provided by technicians or senior students who do not have veterinary or medical qualifications. Often the detection of post-procedural pain is left to these personnel and, without training or guidance, such personnel simply do not recognise the signs of pain in their patients, and may erroneously conclude that animals are pain-free.

Analgesia

The analgesic regime used, then, should be appropriate for the level of tissue damage or surgical trauma involved, should take into account those environmental factors that may add to postoperative or post traumatic pain, and should be based on a measurable assessment of both the adverse effects of pain and the effectiveness of the analgesia.

This paper, which looks at recent advances in analgesia for laboratory animals, has been written for the guidance of investigators using animals in research and teaching, veterinarians, Animal Ethics Committee members, research technicians and animal care staff. The authors believe that the AECs can and should play a key pivotal role and initiate strategies of pain management.

The subject matter is a challenge, in our opinion, for all except those with a sound working knowledge of pharmacology. Like all scientific disciplines, it has its own unique language. The body of anaesthetic and analgesic knowledge is vast, ever changing and complicated. It deals with an increasing array of new molecules, receptors, drugs, neurological pathways and a plethora of theories regarding the mode of drug actions.

Research study results are often conflicting, frequently confusing and a challenge to interpret in a useful way. We have chosen to avoid the scientific jargon and detailed pharmacological considerations. Instead we present some key guidelines and principles that we believe should be considered in any 'best practice' regime for animals used in research and testing.

Our intention is to challenge the AECs that approve animal experiments and the investigators and technicians who perform them to reconsider their current practices in the use of anaesthetics and analgesics. We will discuss a new concept we call

‘vaccination against pain’. We will use the acronym "PREEVAP", or pre-emptive vaccination against pain to describe this new concept. The principle goal should be to ensure that every single animal used in research and testing is routinely given the benefit of appropriate pain medication. The lack of appropriate pain medication should be regarded as ‘deliberate withholding’ of pain relief, and should be fully justified to the AEC.

The literature review involved a keyword search of Medline limited to the last ten years. The search was mainly limited to those species which are most commonly used in laboratory research, testing and teaching in New Zealand - namely sheep, pigs, rabbits, guinea pigs, rats and mice. These are species about which there is less information in the general veterinary literature because surgical procedures of the complexity as are performed in some research institutes are uncommon in members of these species that are privately owned. Other references identified as relevant from the search were also included. Over 300 references were initially identified, with a final total of 122 being considered directly relevant to the subject.

Key areas identified

From the review, we have determined that the recent advances in pain control are covered by four major headings:

1. pre-emptive analgesia;
2. multimodal drug therapy;
3. local or regional analgesia; and
4. new molecules.

Pre-Emptive Analgesia

Introduction

The notion that post-operative pain can be forestalled or even prevented derived from the simple observation of human patients who underwent orthopaedic surgery. It was noticed that patients who received opiate premedication prior to general anaesthesia took four times longer to request pain relief after surgery than those who received a general anaesthetic without any premedication; these unpremedicated patients all requested pain relief within 2 hrs. The author, P D Wall comments wryly that such an observation could have been made at any time in the past 40 years (107).

Wall also cites the example of elderly patients who had elective surgical leg amputations for ischaemic and diabetic problems. Patients who had 3 days of lumbar epidural block with bupivacaine and/or morphine prior to amputation, reported no pain at 6 months, whereas 50% of patients who received no premedication reported pain at 6 months. Similar results occurred at 7 days post surgery and 12 months later. This second example is noteworthy because of the extended duration of preoperative pain medication used. Many of the papers that report little or no preemptive effect administer preoperative

analgesics over a shorter duration. Wall was the first to suggest that analgesics will be more effective when used before rather than after tissue damage

Aims of preemptive treatment

The goal of preemptive analgesia is to prevent the initiation of wind-up so that central sensitisation does not occur. Under ideal conditions, the preemptive medication has prolonged analgesic effects that outlast the presence of the analgesic drugs.

Pain management models

The preemptive literature contains much conflicting evidence and considerable debate regarding the value of preoperative medication for pain control. Some variation must certainly be due to differences in study design between experimental models.

A problem with experimental investigations into preemptive medication appears to be the lack of a reliable animal model. Several have been used, with variable results. Gonzalez et al (29) have reported that ovariectomy in the rat is a useful model which demonstrates a preemptive analgesic effect of morphine given 30 minutes before surgery. Furthermore, when morphine was administered before and twice after surgery, pain was controlled for up to two days post-operatively.

In contrast, Giamberardino et al (28) failed to show any preemptive benefit of analgesics for the induction of ureteric calculi in the rat, created by the injection of dental cement into the ureter. In this study morphine was administered either 45 minutes before or after surgery and daily thereafter for 4 days. Ketoprofen was also tested in this study and similar negative results were obtained. The authors concluded that morphine or ketoprofen demonstrated no preemptive effect.

However, given the severe pain experienced by human patients with ureteric calculi, we question whether the dose, frequency and duration of analgesia used in these rats was sufficient to make valid judgements regarding the preemptive effects of these drugs.

These two examples highlight the need for a clinically relevant approach to studies investigating the potential preemptive effects of drugs. It is not surprising therefore to learn that preemptive morphine controlled the pain associated with ovariectomy in the rat, but not the pain associated with calculi in the ureter of the rat.

We suggest that a simplistic general conclusion can be drawn from these studies. In experiments where a preemptive drug effect was demonstrated, the drug administration regime was clearly appropriate, whereas, when no preemptive effect was demonstrated, the drug administration regime was inadequate in some way. The reasons for the inadequacy are frequently not well defined.

There are several possible explanations for failure of preemptive drug action. The route of administration may result in variable drug absorption. For example, intramuscular

pethidine can produce variable plasma levels in man. The drug may be injected into fat or fascial planes or the injected muscle masses may have different perfusion. In humans, pethidine is more rapidly absorbed from the deltoid muscles than the gluteal muscles (59). This variation may occur in animal patients, although the authors are not aware of any studies that have examined this possibility.

The analgesic drug must be bound to the relevant receptors prior to the first incision and remain effective throughout the surgical procedure until the end of the wound closure (59).

For experimental rodents housed on softwood bedding materials, the possibility of altered drug metabolism through the induction of liver enzymes by aromatic hydrocarbons in the bedding material should be considered. Enzyme induction can lead to an increased rate of drug breakdown, as was described for the barbiturate pentobarbitone in rats (106, 116). Many research papers that report on analgesic action in pain studies regrettably do not include details of the type of cage bedding materials used.

Studies that report the preemptive effects of drugs based on only one method of pain assessment, may not provide robust information, particularly when the method does not measure the animal's response to thermal stimulation.

The benefits of preemptive medication for animals in research and testing

Even if the preemptive action of an analgesic is doubtful, the preoperative administration of analgesics can be beneficial for other reasons. Premedication will reduce the total dose of anaesthetics required. This can reduce or minimise the unwanted side effects of the drugs used for general anaesthesia. In addition, with analgesics on-board before the animal recovers, the patient will not experience pain between the return of consciousness and the administration of the first post-operative dose of analgesic. The overall benefit is a more rapid return to normal function without suffering a painful recovery (40).

From an anthropomorphic view, who would deny their child (or experimental animal) the possible benefits of preemptive analgesia when that child (or experimental animal) was to undergo a painful procedure? There are at least two approaches to pain management. The first approach embraces the preemptive concept, by which patients are given medication in anticipation of pain so as to provide the best possible pain control. The second approach is the more conservative and does not give preemptive analgesia, but instead employs the '*wait and see if the patient calls for pain medication after surgery*' approach. Regrettably this is all too common in the research setting using experimental animals. Few parents would be comfortable with this '*wait and see....*' approach, particularly if they were aware of the preemptive option. As we have discussed earlier, all surgery is painful because it causes tissue damage.

The use of preemptive analgesia in standard veterinary practice

A change in practice trends has occurred over the years. New veterinary graduates are more likely to use preemptive medication than more senior members of the profession. Preemptive use is now the norm in all aspects of veterinary practice. This shift in pain management is similar to changes that have occurred in human analgesia. A report on a teaching study designed to make emergency physicians more aware of their inadequate use of analgesics, confirms the need for continuing education on pain control. After training, the physicians prescribed more analgesics using more appropriate dosage schedules (74).

The concept of PREEVAP

The use of drugs to prevent pain, when given before the painful procedure is performed, can be compared to the administration of a vaccine, given in advance, to immunise the patient (human or animal) against one or more pathogenic agents. We describe this concept as '*Preemptive Vaccination Against Pain*'. Most personnel working in research with experimental animals have an understanding and appreciation of the value of vaccination against disease. We believe that the use of preemptive analgesics should be considered in the same way, so that animals are protected in advance from the pain they might otherwise suffer upon recovery from anaesthetic. Table 1 outlines the comparative aspects of this new concept.

Table 1: A comparative review of vaccination and preemptive analgesia

Vaccination/immunisation	Preemptive use of analgesic drugs
Prophylactic administration in advance, in anticipation of disease	Prophylactic administration in advance, in anticipation of pain
Prolonged period between immunisation and protection (wks)	Short period between administration and pain prevention (hours)
Targets whole body systems	Targets central and peripheral pain receptors
Facilitates management of disease outbreaks	Facilitates management of pain
Provides group animal health benefits	Provides individual animal benefits
Clinically proven beneficial effects in reducing disease	Clinically proven effects in minimising post operative pain

Summary and recommendations

- AECs should promote the PREEVAP concept by proactively requiring preemptive medication on a routine basis for all surgical procedures in experimental animals. We believe that this requirement is justified by sufficient scientific evidence to warrant such a broad generalisation. The only exceptions should be: (a) when acute non-survival procedures are performed and (b) when experimental evidence is provided to confirm that the analgesic drug in question

will present a significant non-experimental variable effect on the parameters to be studied.

- In order to prevent wind-up and central sensitization, the administration should be pre-operatively or during general anaesthesia.

Multimodal Analgesia

Introduction

Multimodal analgesia is the combined use of analgesic drugs that act on different regions of the pain pathway. The rationale behind multimodal analgesia is to improve the level of pain control by the use of drugs that target more than one level of the system.

Furthermore, synergism between the drugs will allow lower doses of individual drugs to be administered thus reducing untoward side effects. Many adult readers will probably have personal experience of what we choose to call the "multimodal martini". The clinical intoxication that results from mixing alcoholic drinks is well known. Just as the gin and vermouth martini is more potent than gin and tonic, the combination of ketamine and xylazine is more potent than either drug administered alone. We should note here that the toxic side effects of martinis appear to be a greater problem than the use of multimodal analgesic drugs, particularly when the martini is consumed in anaesthetic proportions.

Sites of action

Figure 1 shows the pain pathway, giving a schematic representation of both sites of administration and sites of action of various analgesic drugs. Sites of action can be summarised as follows:

1. Local sites - These can be targeted by use of local infiltration of analgesics or local anaesthetics injected into the tissues, or by using anti-inflammatory drugs such as the NSAIDs. These anti-inflammatory drugs act peripherally to decrease inflammation during and after surgery, and thus limit the nociceptive information entering the central nervous system as a result of the tissue damage.
2. Peripheral nerve blocks -Locally active analgesics used at this level include local anaesthetics and amitriptyline
3. Central/spinal nerve blockade - epidurals and intrathecal injections of local anaesthetics, opioids, alpha-2-adrenoreceptor agonists.
4. Cortical effects – opioids and NSAIDS act centrally to limit the input of nociceptive information into the central nervous system.

While some drugs do act at different sites, the NSAIDs for example, which have an effect on both a peripheral and a central level, combinations of drugs that are more selective in their points of action can be used to provide a more comprehensive, enhanced analgesic regime than can usually be obtained by using one drug on its own.

Clinical studies

Clinical studies in humans over the last decade have supported the validity of improved analgesia using combinations of drugs. However, in the medical field, there is still a need for large efficacy studies, to document the optimal drug dosages and combinations for specific surgical procedures. (51).

Although there is an increasing usage of multimodal analgesia in clinical veterinary practice (74), most of the animal studies reported in the literature have used rats (as models for human pain) to compare unimodal with multimodal analgesia. Regrettably the rat-based data is not directly applicable to standard veterinary practice. These have compared combinations used together at the one site (local, regional, epidural, intrathecal or parenteral) as well as combinations with one drug applied at one site, spinally for example, and another applied parenterally perhaps.

Although the conclusions drawn in many studies indicate an improvement in analgesia, in an additive or synergistic manner, we query how well the types of assessment used (tail withdrawal, reduced allodynia etc) correlate with surgical pain. There are relatively few studies in animals that directly compare unimodal with multimodal analgesia, for specific surgical procedures in particular species.

It should be noted that the assessment of the effectiveness of multimodal analgesia in animals is confounded by the multiplicity of possible drug combinations, dose rates and mode of use. The type of surgical intervention and the use of different species further complicates interpretation of multimodal drug regimes .

Research Literature

In summary the literature shows that the multimodal effect can be brought about in three different ways: (1) by combining drugs with different pharmacological mechanisms, (2) by using different routes of administration for drugs with similar modes of action and (3) by using drugs which not only produce the 'martini effect' but may also counteract the potential side effects of each drug.

1. Combining drugs with different pharmacological mechanisms

Opioids with NSAIDs - when morphine and ketorolac are administered together by spinal injection they produce anti-allodynic effects in rats at doses where the drugs were ineffective individually (58). While another study (73) showed no significant interaction between these same two drugs administered intravenously in sheep. However, pain was measured using the response to a mechanical stimulus. The literature recommends that that an animal's response to thermal stimuli is more relevant to surgical pain in this species.

Opioids with alpha-2- adrenoceptor agonists - Several studies have reported at least an additive and more often a synergistic effect between a variety of opioids (morphine,

fentanyl and meperidine) with a variety of alpha-2- adrenoceptor agonists (medetomidine, dexmedetomidine, xylazine, clonidine, detomidine) in both parenteral and spinal routes of administration (75, 83, 84, 85, 90)

Local anaesthetics with various analgesics - In studies that investigated spinal, regional (sciatic nerve block) and local administration, local anaesthetics have shown synergistic effects with a number of other drugs. For instance, intrathecal administration of morphine with lidocaine resulted in a synergistic analgesic effect in rats (95), while phenol potentiated sensory blockade of the sciatic nerve in rats with bupivacaine (56).

With local use, while morphine and bupivacaine showed only additivity only in wound infiltration in rats (22), coinjection of amitriptyline and bupivacaine with epinephrine enhanced the analgesic duration of both drugs in cutaneous analgesia in the rat (52). In a more unusual combination, ketamine has been shown to profoundly enhance the local anaesthetic and analgesic actions of bupivacaine in wound infiltration in humans. (103)

Other - The NMDA receptor antagonist ketamine was shown to potentiate the effect of opioid (morphine) and the alpha-2-adrenoceptor agonist (dexmedetomidine), when administered intrathecally (48), while a non-NMDA antagonist (CNQX) was shown to be synergistic with gabapentin in producing anti-allodynic effect in rats (6).

2. Using different routes of administration for drugs with similar modes of action

A synergistic effect on antinociception has been demonstrated between intrathecal morphine and both subcutaneous morphine and subcutaneous buprenorphine a rat model (78), while spinal morphine and intraperitoneal buprenorphine were shown to be synergistic in the same species (80)

3. Using drugs which not only produce the 'martini effect' but may also counteract the potential side effects of each drug

A classic example of this, more commonly used as an anaesthetic, but demonstrated as providing effective analgesia in sheep (43), is the combination of xylazine and ketamine, where the hypotensive effect of the xylazine is counteracted by ketamine.

A number of studies have demonstrated that anticholinergic drugs (carbachol, neostigmine and physostigmine) can act synergistically to produce analgesic effects when used in combination with opioids (4, 44) and alpha-2- adrenoceptor agonists (61). These anticholinergic drugs could also be expected to counteract some of the side effects caused by the other drug - this was successfully demonstrated (121) when neostigmine counteracted the clonidine-induced hypotension.

Summary and Recommendations

- By targeting the pain pathway at multiple levels, the 'multimodal martini' offers an ideal way of producing maximum analgesic effect with minimum side effects.

- Multimodal drug delivery should be used whenever possible, to promote animal welfare by effective control of post-procedural pain.
- Multimodal analgesia allows a reduction in drug dosages. This is of benefit to the animal and to the researcher, because the physiological parameters under investigation are less likely to be perturbed by drug action
 - Further studies are needed to develop optimal drug combinations and dosages for specific surgical procedures in laboratory animals. Clinically relevant analgesic treatment regimes are required, in order to ensure that experimental animals benefit from pain medication that is at least commensurate with standard veterinary practice.

Local and Regional Anaesthesia and Analgesia

Introduction

Almost all readers who are of the 'pre-fluoride' vintage have probably experienced the benefits of modern dentistry as demonstrated by the use of local anaesthetics to prevent the pain associated with trauma to a tooth pulp and its associated nerves. Few people would deny themselves the advantage of such pain prevention. Toothache can be a salutary reminder of the adverse effects of pain and personal experience of dental nerve blocks can reinforce the value of such local anaesthetics, when they are used to control pain in experimental animals.

Local and regional anaesthesia is defined as the use of drugs to prevent or control pain in specific areas of the body. This is generally achieved by blocking conduction of sensory nerves, as in the dental example above, or by local infiltration of drug into the skin for example, in order to suture a skin wound, or by epidural or spinal injections of drug in order to anaesthetize a region of the body. A common example would be the epidural anaesthesia used for women who undergo a caesarian section.

Mode of action

The mode of action of local anaesthetics such as lidocaine or bupivacaine is to stop the passage of sodium ions (Na^+) across the nerve membrane and hence the drug prevents the conduction of nerve impulses. This can result in a complete absence of pain perception and therefore the events known as 'wind up' do not occur. The consequences for patient welfare are considerable because the central nervous system does not become sensitized.

Advantages

The advantages of local or regional anaesthesia do not appear to be as readily understood in the research, testing and teaching arena, when compared with clinical veterinary practice. This may reflect a lack of training in administration techniques.

Local anaesthetics are generally used in conjunction with sedatives or general anaesthetics to provide additional pain relief. Because they can produce a complete block of sensation, the dose of other anaesthetic agents used concurrently, can be significantly reduced.

These drugs can be injected into wound margins, into joints, such as the knee or shoulder joint, or into bone fracture sites, around nerves (e.g. sciatic or peroneal), around ribs as intercostal nerve blocks for thoracotomy chest surgery, as epidural injections to anaesthetize whole regions of the body, or instilled into a body cavity, such as the chest or abdomen. Some of these routes of administration are technique-sensitive and require anatomical knowledge for correct needle placement and some supervised training. However, veterinarians in practice use many of these techniques on a regular basis and could provide assistance with mastery of the skills required.

Factors affecting duration of effect

The factors that affect the clinical outcome of local or regional anaesthesia include timing of administration, age of animal, duration of action of the drug, toxicity of the agent used, isomer-nature of the compound and the dose administered.

The most effective time to administer local anaesthetics appears to be before the surgical incision is made, rather than after the incision. In a well-controlled human trial of patients undergoing inguinal-hernia repair under general anaesthesia, pre-incisional local anaesthesia proved a more effective method of post-operative pain control than when given after the surgical incision was made (10).

The duration of nerve blockade is reported to be longer in neonatal rats than adult animals (57). However, the reason for this difference is not well understood. The degree of lipid solubility can influence the duration of action, ropivacaine being less soluble than bupivacaine (13). The stereoisomer chemistry of these agents can effect their potency. The R-isomer of many local anaesthetics is more potent than the S-isomer (53).

Local anaesthetics can have toxic side effects. Bupivacaine is more toxic than lidocaine, it can cause cardiac arrhythmias and myocardial depression when injected intravenously. Ropivacaine is less cardiotoxic than bupivacaine.

The duration of action of local anaesthetics can be prolonged by adrenaline through its vasoconstrictor activity (53). A range of adrenaline concentrations are available combined with the local anaesthetic solution.

Drug combinations

A number of drug combinations have been used to attempt improvements in local and regional pain control, to extend the usefulness of the common local anaesthetics.

Morphine has been used as a local injection into inflamed tissues and proved effective in neonatal rats. Morphine was more effective in inflamed tissues than normal tissue (3).

Morphine has been injected into the bone marrow cavity of femoral fractures to effectively block hyperalgesia and allodynia in a rat model (41). The evidence suggests that morphine could be used locally during orthopaedic surgery and for bone graft or bone marrow harvesting procedures to reduce post-operative pain.

Morphine combined with bupivacaine has been used for local infiltration of surgical wounds. The anti-inflammatory effect of morphine has an additive effect and can potentially improve patient recovery (22).

Amitriptyline is a tricyclic antidepressant drug generally used to treat chronic pain. It is a potent Na⁺ channel blocker. The addition of a phenylethyl group to amitriptyline has produced a local anaesthetic drug with more potency and a longer duration of action than the clinically used current alternatives (26).

Amitriptyline has also been combined with bupivacaine to produce a prolonged cutaneous analgesic effect (52).

Bupivacaine activity has been enhanced by the addition of phenol and used in the rat sciatic nerve block model. The 0.5% phenol does not appear to have toxic effects. It should be noted that 0.5% phenol is approved for human use for application to mucous membranes for the relief of dental pain. Both bupivacaine and phenol are Na⁺ channel blockers. However phenol is believed to act at an alternative channel site, thus producing an additive effect when used in combination with bupivacaine (56).

The local anaesthetic effect of bupivacaine has also been enhanced by the addition of ketamine. In a trial of human patients the level of post-operative pain was reduced by the injection of ketamine and bupivacaine together into the wound margins of hernia repairs. The addition of ketamine almost doubled the duration of action from 3.5 to 6 hours. This study also tested the analgesic effects of ketamine alone, administered as a 0.3% solution subcutaneously in human volunteers. The local anaesthetic effect lasted 10-20 minutes. The study confirmed that ketamine can act via a peripheral mechanism, in addition to the known central mechanism (103).

The literature, then, supports the value of multiple drug combinations used to improve the level of pain control. As our understanding of pain mechanisms improves, the use of multiple drugs, all acting at different sites of the pain pathway appears to be more scientifically justified and acceptable. Scientists studying pain control mechanisms increasingly use new combinations of old drugs and mix new drugs with well established compounds. Table 2 gives some examples of drug combinations that can be used for local or regional pain control.

TABLE 2: Drug combinations used for local or regional pain control

Drug combinations

Details

Bupivacaine and morphine	Morphine produces peripheral analgesia that is additive with bupivacaine. Morphine has some anti-inflammatory effects(22)
Bupivacaine and ketamine	Ketamine enhances the anaesthetic and analgesic effects of bupivacaine by a 2-fold increase in duration of action (103)
Bupivacaine and phenol	A marked increase in duration of analgesia by the addition of phenol (56)
Bupivacaine and amitriptyline	Enhanced cutaneous analgesia by the addition of amitriptyline (52)

Prolongation of effect

Prolonged nerve block for 12-16 hrs has been achieved by the experimental development of tonicaine. This new compound is a lidocaine derivative with the addition of phenylethyl group (55, 108). Of great clinical significance is that sensory blockade was longer than motor blockade. The new compound was tested in the rat sciatic nerve model. Regrettably this drug is not yet commercially available.

The use of microspheres to encapsulate local anaesthetics, holds great promise for the management of acute and chronic pain. Bupivacaine encapsulated by poly (D,L)-lactide microspheres is a new drug delivery system that leads to slow uptake of the local anaesthetic and a sustained release of drug. In addition to the prolonged action, this novel system reduces the possible toxic side effects of bupivacaine (23).

Liposomes have been used to deliver bupivacaine to provide a prolonged duration of action. Liposome encapsulated drug was used to infiltrate surgical wounds in rats. The slow depot release of local anaesthetic provided an 8-fold increase in duration of action (36). This new delivery technique offers major advantages for safe and effective analgesia as the potential toxic effects of bupivacaine are avoided.

This literature review has highlighted a number of novel drug combinations that have been used experimentally, in both humans and animals for local or regional anaesthesia.

Uses

Table 3 provides a summary of how local and regional anaesthesia can be used for some common surgical and non-surgical procedures in addition to general anaesthesia.

TABLE 3: Manipulations for which local or regional anaesthesia can be used in addition to general anaesthesia

Common manipulations for which local or regional anaesthesia can be used	Comments on drug administration
Surgery on joints, tendon repairs	Intra-articular injections of anaesthetics can

	be given preoperatively and again at wound closure.
Abdominal surgery	Local infiltration at site of proposed surgical incision and/or at wound margins at time closure
Skull electrode implants	Local infiltration at site of proposed skin incision before surgery
Chest surgery-thoracotomy incisions	Local infiltration around intercostal nerves at time of wound closure
Orthopaedic procedures, bone fracture studies	Instillation of analgesic into the fracture site
Bone marrow collection	Infiltration of the collection site with anaesthetic
Subcutaneous implants such as osmotic minipumps and catheters	Local infiltration of the skin incision site pre-operatively
Induced inflammation models by injection of formalin, carrageenan or other noxious agents into feet	Local nerve blocks around major nerves (sciatic N) or injection of anaesthetics into the feet directly
Nerve ligation models of chronic pain-neuropathic pain	Epidural administration of anaesthetic agents

Summary and Recommendations

- Local anaesthetics can provide very effective pain control when used in the skin, in wound margins and around sensory nerves. The effective use of local nerve blocks requires some knowledge of regional anatomy and practice of the administration technique.
- A range of drugs can be used in combination with the common local anaesthetics to enhance analgesia and prolong the duration action of local anaesthetics. These include amitriptyline, morphine, ketamine and phenol.
- Microspheres and liposomes can significantly prolong the analgesic effect of local anaesthetic agents, by an eight-fold increase. However, these products are not yet commercially available, but they will become extremely useful additions for the management of pain.
- Epidural administration of local anaesthetics, opioids and other analgesic drugs can provide effective regional analgesia and anaesthesia. But for routine use in small laboratory animals, the method is of questionable value due to technique-sensitive nature of the procedure.

New Molecules and New Uses of Old Molecules

Introduction

In veterinary medicine, pain relief has traditionally relied on three main groups of drugs: opioids, the non-steroidal anti-inflammatory drugs (NSAIDs) and the local anaesthetics. Although many of these have been in use in a number of species for quite some time, research continues to look at new ways of using them, by varying routes of administration, the use of different delivery formulations or species to which they may be given.

Recent advances in the study of pain physiology have led to a greater understanding of the complex mechanism of the pain pathway. A number of different physiological and neurochemical changes have been described that occur along that pathway, following a noxious stimulus. The possibility exists to counteract or interrupt those changes through targeted antagonism of neurotransmitters. This has resulted in a proliferation of studies that utilise a variety of new molecules which have the theoretical possibility of reducing the perception of pain.

In addition to these new molecules, new variants of the traditional opioids, NSAIDs and local anaesthetics have continued to be developed. These compounds offer improved analgesia, with a reduction in side effects. This section is divided into three main areas: new molecules; new formulations; and new ways of using traditional analgesics.

New Molecules

We define "new molecules" to be those outside the traditionally used opioids, NSAIDs and local anaesthetics. This also includes new variants within those groups. The classes of drugs identified in the literature that are potentially useful as analgesics include:

Drugs traditionally used in the treatment of depression or mental illness:

- Amitriptyline, a tricyclic antidepressant and sodium channel blocker, which has been shown to provide effective analgesia in the rat when used both locally, spinally and parenterally. (12, 26, 52, 62, 97, 111)
- Fenfluramine, which blocks the reuptake of serotonin and has shown analgesic properties in a rat model of neuropathy (111).
- Dopamine D2 receptor blockers such as remoxipride, traditionally used to treat acute schizophrenia, which have shown analgesic effects in sheep and rats (71, 72)

Anti-epileptic drugs:

- Gabapentin and pregabalin are anti-epileptic drugs which exhibit antihyperalgesic activity by their interaction with calcium channels. The potency of this effect as demonstrated by Field (15) suggests the possibility of these drugs being useful in the painful neuropathies that occur in diabetic patients where conventional analgesics are less effective.

Drugs that target the neurotransmitters involved in the pain pathway.

Analgesic or anti-allodynic effects have been demonstrated in experimental models of pain in rats by the following compounds:

- Neurokinin 1 receptor antagonists which reduce the effect of substance P, an excitatory peptide neurotransmitter (21, p46)
- Metabotropic [AMPA (alpha-amino-3-hydroxy-5-methylisoxazole) and NMDA (N-methyl-D-aspartate)] glutamate receptor antagonists, for example ketamine, that is more widely used as an anaesthetic agent, but shows significant analgesic properties when used alone or in combination. (103, 104)
- Cholecystokinin (CCK) appears to have an anti-opioid effect, and CCK receptor blockers have been shown to have anti-allodynic activity in rats.(79)

New opioids:

- Enadoline, a selective kappa-opioid agonist has shown anti-hyperalgesic and anti-allodynic properties in rats, without causing respiratory depression. (14)

New local anaesthetics:

- Some isomers of local anaesthetics may have greater potency or more prolonged effectiveness - for example, isomers of bupivacaine were shown to be more potent for cutaneous analgesia (53), although a study by Dyhre (9) found similar activity of bupivacaine, ropivacaine and levobupivacaine.

Drug availability

While there are a wide range of different molecules that have been found to have analgesic properties in experimental studies, most of these are not yet commercially available. However, they do give an indication of future trends in the field of analgesia, and they offer the possibility of alternatives where traditional analgesics may not be acceptable because, for instance, a particular receptor is one of the experimental variables.

Experimental models in assessing new molecules

One of the problems with drug research and development, from an animal welfare perspective, is that many studies are preliminary investigations which use animals as experimental models of pain. These studies, in general, do not evaluate the effect of analgesics on pain that arises in a clinical setting, whether surgical, inflammatory or neuropathic. Hence the clinical relevance of these drug research and development studies is questionable when applied to animal models used in other disciplines. Even when the research is focused on the animals themselves, rather than using them as models for human pain, there are problems. Studies by Welsh, 1995, for instance, showed that abdominal surgery in sheep induced thermal, but not mechanical hyperalgesia. The inference being that using a mechanical stimulus to assess the effect of an analgesic in sheep, as in (71) and (74) may not provide clinically relevant information.

Summary and Recommendations for New Molecules

- Because many of these new molecules are still in the experimental stages and not yet commercially available, the traditional analgesics will continue to be the mainstay of laboratory animal pain relief, for the foreseeable future. However, when appropriately used, they provide a comprehensive and effective treatment strategy in the management of pain.
- Nevertheless, there are cases where the classic analgesics may be inappropriate for a particular study, because of interference with physiological parameters or because opioid receptors are the experimental variable under study. In these cases, the new molecules, particularly those that are very specifically targeted, may provide a suitable alternative.
- At least some new molecules have been shown to provide analgesia without the potential side effects of opioids and NSAIDs, or the loss of motor function that can result from the use of local anaesthetics.
- A watch on the potential commercial release of new analgesics is recommended.

New Formulations

The new formulations of analgesic drugs reported in the literature are aimed either at providing sustained release of drugs, or at simplifying their application. Sustained release systems include:

Liposomes - Liposomes are very small spherical vesicles produced from natural phospholipids and cholesterol. The vesicles are extremely versatile and can be loaded with a great variety of molecules. They can be used as carriers for lipophilic drugs and the antiviral derivatives. As well as local administration, liposomes can be injected intravenously and when they are modified with lipids, to make their surface more hydrophilic, their circulation time in the bloodstream can be significantly increased.

Studies in mice (36, 37) have shown that encapsulation of both morphine and bupivacaine in liposomes significantly prolonged the analgesic action compared to the unencapsulated drug; approximately six-fold for morphine, and three-fold for bupivacaine. In addition, the sustained release of both these drugs was shown to significantly reduce systemic toxicity.

Microspheres - Like liposomes, microspheres can be used as vehicles for drugs. They are manufactured from biocompatible, biodegradable polymers. Their formulation allows control of the rate at which a drug is released to the body.

Studies in rats (23) have shown a 50% prolongation of analgesic effect for bupivacaine, and the safe administration of prolonged dosages that would cause systemic toxicity if used in the plain rather than the encapsulated form.

Other - Studies in dogs (8, 101) of the pharmacokinetics of morphine in a sustained-release gel matrix that provided considerable prolongation of bioavailability when injected

subcutaneously. However the oral formulation resulted in a large individual variability in drug absorption.

Formulations that allow for easy application include:

Oral Formulations - An oral formulation of buprenorphine in a flavoured gelatin base has been shown to provide analgesia in rats undergoing laparotomy (20).

Transdermal Applications - Fentanyl patches are increasingly used in clinical veterinary practice for cats and dogs, although individual variation in reaching therapeutic drug concentrations has been reported (74). Additional analgesia may be required until effective analgesia has been reached. Because of relatively slow absorption, it is also recommended in dogs that the patches are applied 20 hours before the desired effect is required (21, p34). Problems can also occur with failure of the patches to stay in place, but the advantages of a system that provides sustained release, without the need for constant intervention makes this an area worthy of further work.

Transdermal fentanyl has been studied for its analgesic effect after experimental surgery in pigs (39, 120), with animals showing less pain on a behavioural scoring system than those treated with parenteral buprenorphine, although this was dependent on the drug dosage.

Summary and Recommendations for New Formulations

- These new formulations have the potential to improve the welfare of laboratory animals by reducing the invasiveness of analgesia administration. Encapsulating drugs for slow release provides a prolongation of analgesia without the need for repeated handling and manipulation, and also reduces the potential for toxic side-effects from drugs in unencapsulated form. Oral and transdermal formulations are both easier and less invasive to administer.
- Regrettably these slow-release formulations are still under development and not yet commercially available in New Zealand. The practicality of applying transdermal patches to small rodents does not appear to have been studied to date. . However, because of the promise of animal welfare benefits in this area, close attention to market place developments is advised.

New applications of traditional drugs - routes, uses, species

This literature review highlights the broad range of possible analgesic treatment options that are available to control pain in laboratory animals. There are many variables to consider, these include variation in dose rates and routes of administration, species and age of animal, combinations of new and currently used drugs, different levels of pain severity, and the techniques used to monitor the painful stimulus.

There appear to be significant differences between species in the distribution and function of opioid receptors. This physiological fact complicates analgesic treatments because it prevents the extrapolation of drug dosages from one species to another.

Our literature review has identified three main drug groups: (1)- those where drugs traditionally used as anaesthetics or sedatives are trialled as analgesics, (2) those where drugs are delivered by novel methods and (3) those where analgesics of proven efficacy are assessed in a species not previously tested.

Anaesthetics and sedatives used as analgesics

In the first group, a number of studies have investigated the use of the alpha-2-adrenoceptor agonists xylazine, as an intravenous infusion that successfully provided analgesia in sheep. (33,34,81). Other drugs within this group, more usually used as anaesthetics or sedatives, include clonidine (also an antihypertensive drug) and medetomidine, which, when administered in sub-anaesthetic doses, have also displayed analgesic properties in this species. (77). Sheep given subanaesthetic doses of xylazine with ketamine (an NMDA receptor antagonist) a combination commonly used for anaesthesia, have also shown behavioural indications of an analgesic effect after surgery. (43)

Novel methods of administration

Although buprenorphine given intranasally to sheep results in effective plasma concentrations, this method of application is probably more clinically relevant to humans than animals. (69) However, the use of the transdermal (fentanyl) and oral (buprenorphine) preparations, as mentioned in the previous section, are potentially very useful for laboratory animals.

Studies also report the use ketamine administered locally, rather than its more usual parenteral form. Tverskoy (103) found that, when combined with bupivacaine for wound infiltration after surgery in humans, ketamine enhanced the effect of the local anaesthetic both in terms of the quality of the analgesia and its duration.

Analgesics of proven efficacy assessed in a species not previously tested

As mentioned previously, analgesic drugs which are routinely used in some species have not been accurately assessed in others. Recent studies in the literature in which such drugs have shown an analgesic effect include fentanyl in sheep (112); ketorolac in pigs (2) and rats (30); ketoprofen in rats (70, 93, 94); meloxicam in mice (96); medetomidine in sheep (77).

Summary and Recommendations for New Uses

- There is an urgent need for further research in the use of clinically relevant anaesthesia and analgesia, for the common laboratory animal models that involve

standard surgical modifications or experimental surgical procedures. The current studies generally focus on induced pain in animal models that are designed to evaluate analgesic use for humans. The relevance of some of these investigations is of questionable value and can seldom be directly applied to the management of clinical pain in animals.

- The potential for alpha-2 adrenoceptor agonists as analgesics in sheep provides an alternative to the use of opioids. The effectiveness of opioids in this species is controversial. However, there are some significant side-effects of alpha-2 agonists that can compromise their routine use in ruminants. There are relatively few studies in which different analgesics are compared for their effectiveness in a particular surgical procedure. Liles and Flecknell (91,93,94,98) are among the few who have published studies on comparative surgical analgesia.

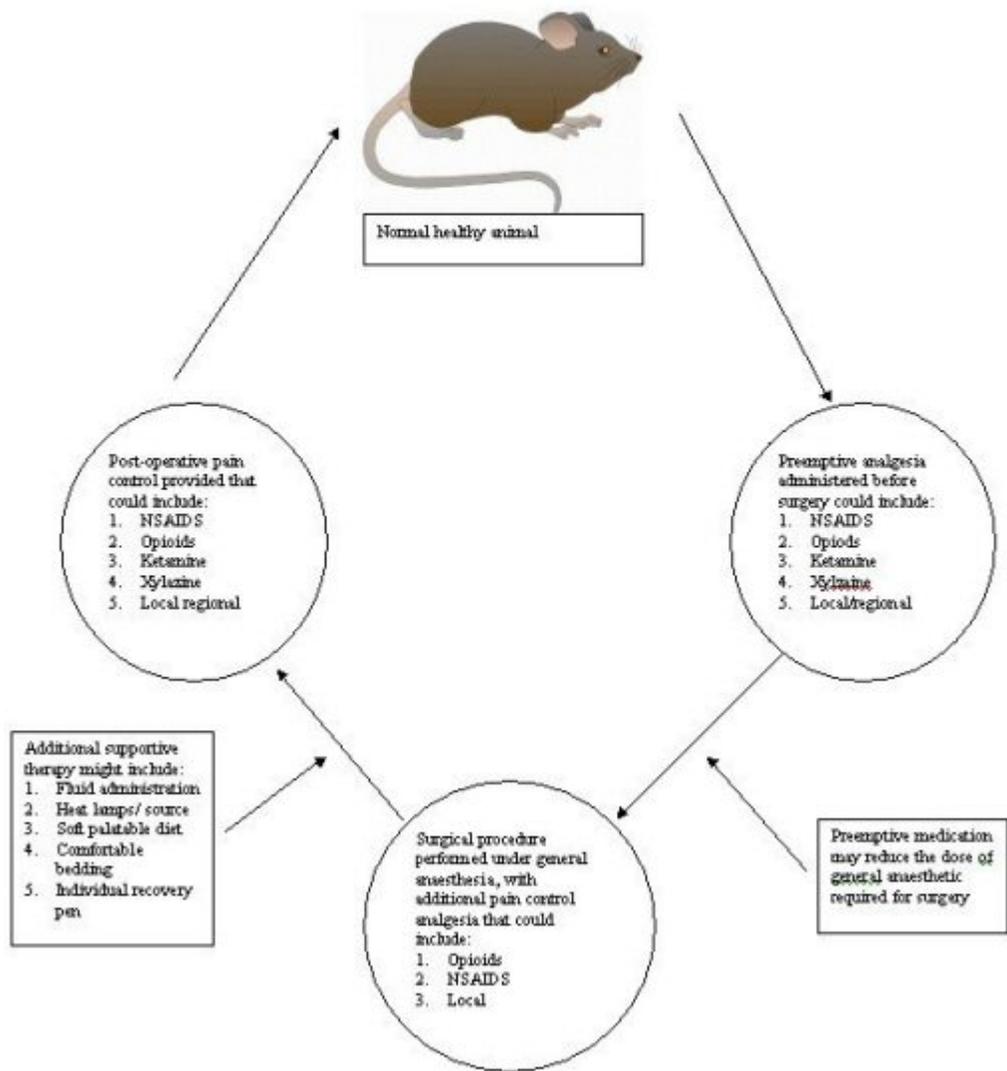
Key Recommendations for Analgesic Best Practice in Animals

1. An anthropomorphic view of animal welfare should be used whenever animals are subjected to painful procedures. If the procedure would cause pain in humans, it must be assumed that the animal would also experience pain and appropriate analgesia should be administered even when signs of pain cannot be readily detected in the animal.
2. The absence of evidence of pain does not constitute evidence for the absence of pain. Most personnel who use animals in research and testing poorly understand pain detection in animals.
3. AECs should challenge the accepted and traditional research methodologies that are used to justify the continued practice of outmoded, outdated or obsolete anaesthetic and analgesic techniques. Pilot studies should routinely be used to test the effects of analgesic drugs whenever the outcome is uncertain.
4. Despite the difficulties in agreement regarding the definition of anaesthesia and the best method to determine the depth of anaesthesia, the use of a deep pain reflex response is still considered the most reliable and practical measure of anaesthetic depth, and therefore of ensuring a lack of sensation during surgical procedures.
5. Most drugs commonly administered to induce general anaesthesia only produce unconsciousness. Despite being unconscious, the nervous system is still able to respond to painful stimuli. This is perceived as pain once the animal recovers consciousness. Therefore additional drugs should always be administered to alleviate post-operative pain.
6. Significant advances in animal welfare can still be achieved by the use of existing drugs used in different ways.
7. There can be significant species variation in response to drugs, both in relation to their efficacy and to effective dose rates, making extrapolation from one species to another unsafe. Reference to the current literature should be made when a drug is proposed and personnel have no previous experience of its use.
8. The beneficial effects of analgesics should be promoted and understood in terms of promoting a rapid return of physiological function, a stress-free animal and

more normal physiology during the recovery period. (See Fig 2 - The Analgesic Circle)

9. Pain intensity varies with the surgical procedure and pain control should be appropriate to the degree of surgical trauma and tissue damage produced. Having said that, there is a paucity of information on the effect of analgesics on surgically induced as opposed to contrived experimental models of pain.
10. Prevention of pain by the use of preemptive drugs provides more effective pain control than alleviation of pain after it has been initiated. Animals can be "vaccinated" against pain.
11. The use of a combination of drugs is more effective in preventing and controlling pain than the use of a single drug administered alone. The synergistic action of many drug combinations allows a net reduction in dosages with a concomitant reduction in side effects.
12. Local anaesthetics can provide very effective pain control when used in the skin, in wound margins and around sensory nerves. While nerve blocks and spinal analgesia are also effective, they are "technique sensitive" in that they require some knowledge of regional anatomy and practice of the administration technique.
13. The development of new analgesic molecules may provide appropriate alternatives to traditionally used pain relief .
14. New formulations of analgesic drugs have the potential to contribute positively to the welfare of laboratory animals by easing the initial application or by prolonging the effect of the drugs, thus allowing for less manipulation of the animal.
15. To achieve the best possible pain alleviation and fully promote animal welfare, multimodal and preemptive use of analgesics at a series of time points should be used wherever possible. Use of analgesics in research, testing and teaching should mirror that use in veterinary practice unless reasons for withholding pain relief can be justified to an AEC.

Fig. 2 The Analgesic Circle
 The purpose of the circle is to restore the experimental surgical patient to its healthy preoperative condition as rapidly as possible through the use of anaesthetics, analgesics and supportive therapy



Glossary

TABLE 4: Definition of terms

Term	Definition
AEC	Animal Ethics Committee
Allodynia	A condition in which stimuli that would normally not evoke pain are perceived as painful
Anthropomorphism	The ascribing of human characteristics to animals
Central sensitisation	State of hyperexcitability of dorsal horn neurones as a result of

	repeated afferent impulses
Dorsal horn	Area in the spinal cord where sensory neurones connect with spinal cord neurones
Epidural analgesia	Injection of analgesics drugs into the epidural space surrounding the nerves within the spinal column
Hyperalgesia	A heightened perception of pain after a period of stimulation
Hyperexcitability	A lowering of the threshold at which nerve impulses are initiated
Intrathecal analgesia	Injection of analgesics into the cerebrospinal fluid
Local anaesthesia	Application of anaesthetic or analgesic drugs directly at the site of surgery or tissue damage
Multimodal analgesia	The combined use of analgesic drugs that act on different regions of the pain pathway
Nociceptors	Pain receptors
Parenteral	Any treatment administered by a route other than the digestive tract
Peripheral sensitisation	Increased sensitivity of nerve fibres due to the release of inflammatory molecules in areas of tissue damage
Preemptive analgesia	Application of analgesia prior to tissue insult so as to prevent the initiation of wind-up
Premedication	Medication given prior to surgery
Regional anaesthesia	Application of anaesthetic or analgesic drugs as nerve blocks or spinal injections to provide analgesia to a region of the body
Synergism	Occurs between drugs when the combined effect of two drugs is greater than the sum of their individual effects

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