

Physiological and Behavioural Assessment of Pain in Ruminants: Principles and Caveats

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Summary — Pain elicits a range of physiological and behavioural responses. These are commonly used to assess the impact of pain-inducing stimuli on animals, to determine whether or not significant pain is experienced and to devise strategies for alleviating pain. This paper outlines a range of principles and caveats to guide the evaluation of physiological and behavioural responses to painful stimuli, so that they can be better used to minimise pain in the experimental context. Although this advice is based on studies of farm animals responding to painful husbandry practices, it is more generally applicable.

Key words: *behaviour, pain assessment, physiology, validation.*

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Introduction

The subjective experiences of pain cannot be quantified physiologically or behaviourally. Nevertheless, putative indices of pain and distress can be derived by observing physiological and behavioural responses to aversive, painful or otherwise noxious stimuli and, when interpreted with care (1, 2), they can be informative. Substantial progress in the recognition and alleviation of pain in clinical settings has been made on this basis (3). Likewise, using farm livestock, the pain and distress caused by routine husbandry practices that cause injury by cautery, cryocautery, cutting, crushing, constriction and corrosion of ears, skin, bone, horn, scrotum, testes and/or tail have been evaluated and alleviation methods developed (2, 4–6). As before (2, 4–6), the response to these husbandry practices described here will be referred to as “pain-induced distress”. This paper will outline a range of principles and caveats to guide the evaluation of physiological and behavioural responses to painful stimuli, so that they can be better used to minimise pain in the experimental context. Although this advice is based on studies of farm livestock responding to painful husbandry practices, it is more generally applicable.

General Guidelines

Stafford & Mellor (1) outlined five general guidelines to aid the meaningful interpretation of pain-induced distress responses. They are as follows.

1. All parameters chosen to indicate the presence of pain-induced distress must, by definition, also

show variations or frequencies of occurrence that indicate its absence. The values in distress-free and distressed states both have to be determined.

2. It is important to determine whether or not deviations of chosen indices from control values to levels indicating pain-induced distress actually do reflect the expected type of distress or, indeed, reflect distress at all. For instance, in anaesthetised animals, cortisol responses can be elicited by hypoxaemia and/or hypotension, as well as by surgery-induced pain, so that the first two need to be prevented before cortisol responses can be interpreted in terms of stimulation of pain receptors.
3. The significance of different distress-specific behaviours should be checked, where possible, against physiological indices in order to assess the validity of conclusions drawn using behaviour alone. For example, cortisol responses question conclusions based on cursory observation of behaviour regarding the relative intensities and durations of distress caused by castration with a rubber ring or a knife (7).
4. Making recommendations based on unchecked or poorly checked assumptions must be avoided, especially when extrapolating from one species to another, as unexpected differences can arise between the distress responses of different species to the same treatment.
5. Studying the distress caused by husbandry practices is complex, time-consuming and expensive. To be done rigorously, allowance

must be made for possible effects of the species, breed, sex and age of the animals, the different methods used, and even factors such as the rearing methods of young animals.

Physiological Assessment of Pain-induced Distress

Principles

There are ten other points that need to be borne in mind when interpreting physiological responses to noxious stimulation (2).

1. The specificity, or otherwise, of the responding system needs to be considered. For instance, the hypothalamic–pituitary–adrenal system, which controls cortisol release, responds to a wide range of physically, physiologically and emotionally challenging situations. Although some argue that this is an impediment to the use of cortisol as an index of distress, others consider that this non-specificity adds credibility to its use to assess distress. Indeed, it has been demonstrated that provided the stimuli used are obviously noxious, and that appropriate control groups are used, cortisol responses can be informative (2, 4, 5).
2. The parameter of interest must be measured repeatedly and at sufficient frequency to define the response as it manifests and recedes. Only in this way can the magnitude and speed of change, and the duration and pattern of the whole response or each part of it, be determined. Differences between groups in initial or later changes, peak values and the times taken to reach them, and the time of return to pre-treatment values are informative, but only if they can be related to the whole response. Only comparing values before treatment with those at one or two arbitrary times after it provides little valuable information and can be misleading.
3. Physiological responses to different treatments vary in complexity. They may be simple (e.g. rising to a peak and then returning to pre-treatment values), or they may be more complex (e.g. first rising to a peak, then declining to a plateau and finally returning to pre-treatment values). Further complexity arises when responses include two (or more) peaks.
4. Quantitative tools for characterising distress responses include numerical representation of individual facets of the response (e.g. peak height, response duration, area under the response curve) and statistical evaluation of the patterns of response to detect within-group deviations from pre-treatment values and between-group differences after treatment. There is no single numerical factor that adequately defines distress responses, even simple ones, and it is obvious that the more complex a response is the less likely it is that a single number could represent it effectively. Quantitative definition of physiological responses may best be achieved by using a range of numerical approaches, with the chosen combination depending on the characteristics of the particular response.
5. A particular parameter will usually not be useful unless its response magnitude, as indicated by peak height, response duration and/or area under the response curve, accords with the predicted noxiousness of different procedures (for examples, see [2]).
6. When local anaesthetics and/or systemic or other analgesics virtually eliminate a physiological response to a painful treatment, this provides strong evidence that, in the absence of pain relief, post-treatment deviations in the measured parameters reflect the presence of pain.
7. With each parameter, care needs to be taken when interpreting responses at the lower and upper extremes of the response range, because different noxious stimuli applied simultaneously may not have additive effects on the responses. At the upper end of the range, for instance, this may manifest as a “ceiling effect” where the overall noxiousness of two undoubtedly painful stimuli applied simultaneously may be underestimated, because each one alone would elicit a maximum response.
8. Some parameters show wide between-animal variation in their levels and patterns of change after treatment. It is important to distinguish between variable effects of pre-treatment stressors on different animals and animal-specific differences (some animals show consistently high and others consistently low responses to the same stimulus). This is accomplished easily by assessing how close the physiological variable is to its non-stressed levels before treatment and whether or not it subsequently returns to those levels.
9. Assessment of the acute pain-induced distress responses of animals by using a single physiological parameter as the sole index allows conclusions to be drawn only about those features of the acute responses that are reflected by changes in that parameter. Thus, the dynamics and characteristics of the physiological system which incorporates the chosen parameter, and

the part played by that system in responses to painful and distressing stimuli, must be critically evaluated.

10. It is valuable to use several indices of pain-induced distress, as these are likely to cover different facets of responses to painful and distressing stimuli.

Specific physiological parameters

There are two complementary approaches to assessing physiological responses to painful stimuli. The function of the body's pain perception apparatus can be explored by studying the physiological mechanisms that underlie pain experiences — i.e. stimulation of pain receptors, transmission of nerve impulses in pain pathways and electrical activity in those areas of the somatosensory cortex, which apparently translate impulse traffic in pain pathways into perceived pain (2). It is also possible to explore wider physiological changes that accompany, or that are induced by, heightened activity in the body's pain apparatus — i.e. different features of physiological stress responses (2, 8). The latter approach has been the most common in studies of painful husbandry practices.

To date, the vast majority of husbandry distress studies have used changes in plasma cortisol as the primary physiological index (2, 4, 5), but more recently other parameters have been evaluated for their usefulness (9, 10). It has been shown, for instance, that with regard to the durations of acute (short-term) hormonal responses to painful stimuli (2, 4, 5, 9, 10), increased release of adrenaline is the most short-lived (2–5 minutes), noradrenaline is intermediate (0.75–1.0 hour) and adrenocorticotrophic hormone (ACTH) and cortisol are the longest (2.0–3.5 hours to 6–8 hours, depending on the noxious treatment). In addition, heart rate and arterial blood pressure responses peak sooner and last somewhat longer than cortisol responses (10). It has been suggested that observation of adrenaline responses is mainly of value when assessing the immediate effects of a painful treatment; that noradrenaline responses may be useful as an index of the extent of tissue damage; that cortisol is of limited value early in the response; that from about 15 minutes after treatment, cortisol is a useful index of significant pain-induced distress; and that heart rate and arterial blood pressure may be more sensitive than cortisol as indices of low-level pain (2, 4, 5, 9, 10).

Behavioural Assessment of Pain-induced Distress

The behaviour and demeanour of an animal, including normal or abnormal activity, posture, respon-

siveness and changes in temperament, are usually considered to be immediately available and reliable indicators of pain. However, this may not always be the case (11). When an injury or disease physically incapacitates an animal such that its normal functions are not easily performed, its behaviour may reflect the degree of incapacitation and not necessarily the pain being experienced. Also, incapacity may cause distress by limiting an animal's ability to escape from potential predators, but this psychological state is not pain. Likewise, if specific behaviours are beneficial to recovery and convalescence, then they may indicate a recovery process and not pain.

Although behaviours thought to indicate pain have been enumerated (12, 13), correlations between pain and behaviour are poorly defined, and people tend to use of a combination of empathetic projection and behavioural observation as the method of evaluation. It is, perhaps, not surprising that evaluations of pain by simple descriptions of behaviour, numerical rating scales (NRS) or visual analogue scales (VAS), attempted in various species, have been of limited value (14–19).

Nevertheless, behaviour will remain the most immediately accessible tool for assessing pain, but it is of value only when pain-specific behaviours can be identified. Reported below are ten key principles, derived from studies of farm animals (11, 20–28), which are applicable during attempts to establish which particular behaviours are pain-specific.

1. A behaviour may be pain-specific if it is seen during and after a specific tissue-damaging injury, disease or procedure, but is not seen in healthy or non-damaged, i.e. control, animals.
2. A behaviour may identify nociception, and by inference pain, if it is seen during and after a specific tissue-damaging procedure, but not seen when local anaesthesia is delivered.
3. A behaviour may be pain-specific if it is present after a painful experience, but is not present when effective analgesics are used.
4. A behaviour may be injury-specific. Different noxious treatments or injuries may elicit unique behavioural responses, because the sensations experienced by an animal may differ when different tissues are injured or when similar tissues are damaged in different ways.
5. A behaviour may occur only during particular phases of a response to a noxious treatment and not throughout the response, as assessed using physiological parameters.
6. Behaviour can be used to identify the duration of pain if it is not present before or after a

treatment and if it lasts throughout the response to treatment as assessed physiologically.

7. When carefully and rigorously assessed (22, 28), behaviour can be used to evaluate the severity of pain.
8. Given the choice (29, 30), animals will choose the lesser of two noxious treatments, and this indicates what they prefer.
9. Self-administration of analgesics can be used to monitor pain (31).
10. Change in nociceptive thresholds, assessed by behavioural responses to noxious thermal or mechanical stimulation (19, 32, 33), may be used to monitor pain.

Conclusion

Numerous techniques are being used to develop an understanding of the behavioural and physiological responses of animals to painful stimuli, disease, injury and surgical intervention. Attempts are also being made to combine behavioural, physiological, immunological and production parameters in order to improve our ability to quantify the pain experience of animals. Although interpretation of the results from such studies may be difficult, and it remains an issue whether the pain experience of animals will ever be accurately defined, the principles outlined above do provide guidance designed to maximise the value of this important work.

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