# Physiological and ethological aspects of the assessment of pain, distress and suffering

**Wolfgang Scharmann**
Institut für gesundheitlichen Verbraucherschutz und Veterinärmedizin, Diedersdorfer Weg 1, D-12277 Berlin, Germany

## Summary

In this paper some of the important factors in the recognition and assessment of pain are described, and the strategies which may be employed to do so.

In animal experiments, pain, distress and suffering may be the unwelcome accompaniment of a disease being studied, as well as in other types of research. However, intense pain (FELASA 1994) and suffering (Spinelli & Markowitz 1987, Dawkins 1990) are at present also unavoidably linked to the process of the risk assessment of drugs and other chemicals as required by various regulatory authorities. The public views suffering in animal experiments as the major argument against research using animals. Many scientists share this concern and acknowledge that there must be a balance between the legitimate demands of research and the need to protect animals from avoidable suffering. Thus, the main purpose of animal welfare legislation is to prevent or at least minimize pain and distress. The provisions are based on the assumption that pain and suffering are identifiable, predictable and avoidable and can even be graded according to the degree of sensation experienced by the animal subjects. So, using category systems, data are collected on the degree of severity of laboratory animals in Switzerland and The Netherlands. Irrespective of the imprecision of pain assessment and the incomparability of the two systems, I shall quote statistical data from Switzerland and The Netherlands about the degree of discomfort, to give an approximate idea about the number of animal experiments associated with pain and distress in these and, probably also, in comparable countries (Table 1). The differing figures for the two countries, especially in category 3, illustrate not only the incompatibility of different grading systems, but also the difficulty of assessing pain and distress in a reliable and comparable way.

## Recognition of pain and distress

As is well known, pain and discomfort are internal mental states and highly subjective experiences which are influenced and modified by psychic emotions. This is not only true for humans but also for animals since pain perception may be altered by various factors such as anxiety, memories of previous discomfort, the social ranking order within a group, fear of predators in wild species, and other environmental stimuli. Thus, an objective and acceptable way to ascertain or even to measure pain and distress is not possible. When trying to find out what an animal feels, we have to use rather crude measures.

The recognition of pain and distress is based on behavioural changes and physiological responses exhibited by the experimental subject. Therefore, deviations in the behavioural pattern and the physiological function can be realized only when an animal's normal and abnormal behaviour and appearance before it is exposed to a potentially painful procedure are well known. Naturally, species vary widely in their typical behaviour, but there are also considerable intraspecies behavioural differences in certain strains or breeds. Moreover, even within a

**Table 1 Estimated degree of discomfort in laboratory animals**

| | Switzerland (1997)[a] | | The Netherlands (1996 and 1997)[b] | | |
|---|---|---|---|---|---|
| Category | No. of animals | % of total | No. of animals | % of total (1996) | % of total (1997) |
| 1 Minor | 343 385 | 69.8 | 419 576 | 56.6 | 52.0 |
| 2 Moderate | 112 351 | 22.8 | 186 114 | 25.1 | 29.2 |
| 3 Severe | 36 480 | 7.4 | 135 484 | 18.3[c] | 18.8[d] |

[a] Tierversuche in der Schweiz, Statistik 1997
[b] Animal Experimentation in The Netherlands, Statistics 1996/1997
[c] Without pain prevention: 9.2%
[d] Without pain prevention: 9.4%

group of cage mates the normal behaviour of one individual may be different from another (e.g. there may be inquisitive and timid group members). The animal should be observed for its appearance and behaviour before it is disturbed, if possible from a distance (ILAR 1992). Crepuscular animals such as rats and mice, which rest during normal human working hours, should be observed in the evening.

An animal in a state of well-being will move normally, exhibit normal curiosity through exploration, keep itself well groomed, will eat, drink and grow normally, will perform normal social contacts with cage mates, and show other species-typical actions. If there is a change in an animal's behaviour and appearance after a procedure that is likely to have caused pain, then it is possible that pain is the cause of that change (Spinelli & Markowitz 1987). A great number of species-specific behavioural responses to pain and clinical signs have been described elsewhere (LASA 1990, FELASA 1994, Bundesamt für Veterinärwesen 1994). However, many of these observations and manifestations (e.g. decreased activity and reactivity, ungroomed appearance, segregation from cage mates, withdrawal to a corner) do not necessarily indicate pain or distress, but may also be indicators of illness or indisposition. Nevertheless, there are some behavioural signs which are usually associated with acute pain and suffering (Table 2).

In the case of chronic pain, the behavioural signs are likely to be subtle, and include decreased appetite, weight loss, reduced activity, sleep loss, irritability and decreased mating and reproductive performance (Soma 1987).

**Table 2 Signs of acute pain in animals**

| | |
|---|---|
| Guarding | Attempting to protect, move away, or bite |
| Crying | Movement or palpation |
| Mutilation | Licking, biting, scratching, shaking |
| Restlessness | Pacing, lying down and getting up, shifting weight |
| Recumbency | Unusual length of time |
| Ambulation | Reluctance to move, difficulty in rising |
| Abnormal positions | Head down, tucked abdomen |
| Increased respiration rate | |

Modified from Soma (1987)

Another approach to detecting pain is based on a classification of the sensitivity of tissues and organs (Table 3). As indicated in Table 3, surgery on the eyes, ears, dental pulp, nerves and testes may be quite painful. Abdominal procedures do not seem to be as painful in the immediate post-surgical period in dogs and cats (and presumably in other

**Table 3 Sensitivity of tissues and organs to pain**

| | |
|---|---|
| Eyes, ears, teeth | +++ |
| Nerves | +++ |
| Testes | +++ |
| Spinal cord | ++ to +++ |
| Skin | ++ to +++ |
| Serous membranes | ++ to +++ |
| Periosteum | ++ to +++ |
| Blood vessels | ++ to +++ |
| Viscera | + to +++ |
| Muscles | + to ++ |
| Joints and bones | + to ++ |
| Brain tissue | − |

Modified from FELASA (1994)

quadrupeds) as in humans (Spinelli & Markowitz 1987), and there are no nociceptors in the brain (LASA 1990).

Biochemical markers, e.g. an increase in catecholamines, corticosteroids and other hormones in the plasma can reinforce the diagnosis that an animal is in pain (ILAR 1992). However, blood-sampling can itself be stressful and should therefore be avoided.

An easier confirmation of the 'pain' diagnosis can be achieved by giving analgesic drugs. If an animal's behaviour can be restored to normal after the administration of analgesics, one can be fairly sure that the animal has been in pain.

## Grading of pain intensity

Although the recognition of pain and distress experienced by an animal may often be difficult, the assessment of the degree of discomfort is even more difficult and problematic. To a lesser extent, this applies to cases of locomotor disorders such as lameness or arthritis, where the grading of the severity of pain can be assessed by measuring the impairment of movement and by using palpation. Evaluation of the severity of experimental arthritic inflammation has been carried out by measuring the swelling of the paws or joints and locomotor activity (Coligan *et al.* 1994). However, in most cases the quantification of pain is much more precarious. Nevertheless, in several countries, an assessment of the severity of pain and distress is performed before starting an experiment, as a tool for balancing the prospective animal harm against the likely scientific benefits—a procedure used to determine whether the experiment using animals can be morally justified.

In order to assess pain in an objective manner, established scales have been published ranking animal pain and distress as either minor, moderate, or severe e.g. the Swiss invasiveness scale. The Swiss system consists of two lists. The first one is used for the assessment of pain likely to be inflicted on the animals before starting the experiment, and is composed of a number of areas of medical research. Procedures commonly performed in the respective area are assessed according to the impairment of the animals and are graded between 0 and 3 (Fig 1). Having finished the animal experiment, the scientist has then to enter in a second list whether the predicted level of pain was confirmed or not. These data are used to develop the statistics describing the degree of discomfort as shown in Table 1. Obviously the value of these grading schemes is substantiated less by the attempt to determine the level of pain likely to be inflicted on the animal; rather the intention is to sensitize the scientists and make them aware of what they are going to do. If it turns out in the retrospective analysis that the impairment of the animal was severe, the experimenter has a moral obligation to avoid suffering by modifying the hypothesis to be tested in such a way that other criteria for the successful conclusion of the experiment can be applied, or by foregoing the anticipated gain of knowledge (Swiss Academy 1997).

A more useful and widely accepted method for grading the severity of pain and distress has been presented by Morton and Griffiths (1985) (Fig 2). They proposed a clinical scoring system composed of a number of independent variables: body weight, appearance, measurable clinical signs (e.g. respiratory rate), unprovoked behaviour and behavioural responses to external stimuli. In each of these categories a rating of 0 (normal or mild) to 3 (severe) is made, the total score yielding an assessment of how serious is the change of the animal's condition. Although the scoring system has been regarded as a major support for all who care for laboratory animals, it has been noted that using numerical scoring can give an aura of precision which may not exist and could exclude informed clinical judgement (Mellor & Morton 1997). In recent years, Morton (1997, 1999) has improved and refined the scoring system by developing an assessment sheet recording system. These so-called score sheets also cover behavioural and clinical signs, but they take into consideration that an experiment will not only produce general signs of pain and distress, but also specific signs related to the particular scientific procedure. Therefore, every type of experiment needs a special score sheet specifying general signs such as activity,

**Swiss Federal Veterinary Office**

**Classification of Animal Experiments Prior to Experiments by Degree of Severity**

**8 Models of analgesia and inflammation (excerpt)**

| **Grade 0** (no exposure) | | |
|---|---|---|
| Analgesia | – | |
| Inflammation | – | |
| Arthritis | – | |

| **Grade 1** (minor, short-term exposure) | | |
|---|---|---|
| Experiments causing *short-term, low-degree pain* | | |
| Analgesia | *Examples*: | Hot plate test, tail flick test without restrainer, tail immersion test, writhing test with 0.25 ml aqueous suspension of pheny-p-benzoquinone 0.02 % in tragacanth 0.4 % |
| Inflammation | *Examples*: | Anti-pyresis in the rat using LPS or IL-1; tissue chamber model (mouse); arachidonic acid test in the mouse ear |
| Arthritis | – | |

| **Grade 2** (moderate, short-term *or* minor, intermediate to long-term exposure) | | |
|---|---|---|
| Experiments causing *moderate, short-term pain* or *conditions of chronic, low-degrees pain*, without essential impairment of mobility | | |
| Analgesia | *Examples*: | Writhing test using $< 0.2$ ml acetic acid or 0.4 ml 1% aqueous acetic acid. Tail flick test using restrainer. All models with acute paw oedema and 'withdrawal' as measuring criterion. Writhing test using alcoholic solution of phenyl-p-benzoquinone 0.02 % in tragacanth 0.4 % |
| Inflammation | *Examples*: | Air pouch model in the rat; encephalomyelitis model with killing of animals on first seizure; screening of anti-inflammatory agents in mouse strains with spontaneous autoimmune disease (M RL Ipr/lpr mice). All models with acute paw oedema, 'paw volume' as measuring criterion; duration of experiment $< 6$ h |
| Arthritis | *Examples*: | Randall-Selitto test, collagen II-induced arthritis with early killing; adjuvant arthritis with killing of the animals on day $< 19$ after induction of arthritis |

| **Grade 3** (severe, short-term *or* moderate, intermediate to long-term exposure) | | |
|---|---|---|
| Experiments causing *short-term, high-degree pain* or *conditions of chronic moderate to high-degree pain*, with or without essential impairment of mobility | | |
| Analgesia | *Examples*: | Writhing test using $> 0.2$ ml and $> 2$ % aqueous acetic acid. All models with acute paw oedema and 'localization' as measuring criterion |
| Inflammation | *Examples*: | Recidivating encephalomyelitis model, without killing of the animals during the first seizure; pertussis pleuritis in rats or mice. All models with acute paw oedema, duration of experiment $> 6$ h |
| Arthritis | *Examples*: | Adjuvant arthritis, experimental period $> 18$ days after induction of arthritis; carragheen arthritis model; induction of arthritis in inbred mouse strains using *Borrelia* spirochaetes; autoimmune arthritis (except collagen II arthritis) |

**Fig 1 Swiss invasiveness scale to be used for pre-experimental evaluation of an animal experiment.**
Bundesamt für Veterinärwesen CH-Bern (1994)

| Score | Variable |
|---|---|
| | **Body weight changes** |
| 0 | Normal |
| 1 | Uncertain; $<5\%$ loss of maximum previous weight |
| 2 | 10–15% weight loss; faeces may be altered in amount or consistency |
| 3 | $>20\%$ weight loss; no food or water to be consumed |
| | **Physical appearance** |
| 0 | Normal (coat smooth, eyes clear and bright) |
| 1 | Lack of grooming |
| 2 | Rough coat, nasal/ocular discharge |
| 3 | Very rough coat, abnormal posture (e.g. hunched up), eyes pale, enlarged pupils |
| | **Measurable clinical signs** |
| 0 | Normal (rates within the physiological norm) |
| 1 | Small changes of potential significance |
| 2 | Temperature change of 1–2°C, cardiac and respiratory rates increased up to 30% |
| 3 | Temperature change of $>2$°C, cardiac and respiratory rates increased up to 50%, or markedly reduced |
| | **Unprovoked behaviour** |
| 0 | Normal behaviour pattern |
| 1 | Minor changes |
| 2 | Abnormal behaviour, reduced mobility, decreased alertness, inactive, separation from the group |
| 3 | Unsolicited vocalizations, self mutilation, either very restless or immobile, expiratory grunts |
| | **Behavioural responses to external stimuli** |
| 0 | Normal (behavioural responses normal for the expected conditions) |
| 1 | Minor depression/exaggeration of responses |
| 2 | Moderately abnormal response, moderate change of behaviour |
| 3 | Violent reactions to stimuli, or very weak muscular responses as in a pre-comatose state |
| | **Total** |

**Fig 2 Qualifying pain, distress, suffering** (adapted from Morton & Griffiths 1985)

posture, body weight, body temperature etc. and cardinal signs characteristic of the particular experiment. Using this checklist, a careful clinical examination should take place at least once a day. By these means, changes in the behaviour and clinical symptoms will be recognized early and provisions can be taken to alleviate suffering. The overall evaluation of the animal's condition results from a combination of general and specific signs assessed as positive or negative. Sheets designed for procedures likely to cause severe pain should describe the point at which an animal ought to be humanely killed (Morton 1997).

The score sheets proposed by Morton has been shown to offer a number of noteworthy advantages:

- a regular checking of clearly defined parameters reduces the risk of overlooking important signs;
- if more than one person checks the list the between-observer variation will be reduced, thus decreasing subjective judgement;
- the scoring system ensures that certain actions such as medical or analgesic treatment or humane killing will be performed in time; and

- not only veterinarians, but other adequately trained staff can be involved in the monitoring over the course of an experiment.

Speaking of pain and suffering we have to ask ourselves: What should be the limit of animal suffering? In this connection I was really impressed by the remarks of Sir Andrew Huxley (1983), President of the Royal Society. He said

*'Severe but short-lived pain should be permitted only exceptionally, e.g. perhaps occasionally for the investigation of pain itself; severe and enduring pain, or excruciating pain, never.'*

Also our Animal Welfare Acts and Animal Protection Acts aim at avoiding, or at least minimizing, pain and distress. In reality, however, it is not like this. Hundreds of thousands of animals die every year in painful experiments due to regulatory safety requirements. Some people say: 'Don't worry, most of these animals are just rats and mice'. However, rats and mice are provided with the same capacity for suffering as cats and dogs—nobody will be able to prove to the contrary. Current regulatory guidelines recommend that animals should be humanely killed if they are obviously in pain, or show signs of severe distress, or are found moribund. However, we still hear at this workshop how difficult and often impossible it is to define the 'moribund state'. Therefore, unnecessary suffering cannot be avoided and many animals must still perish in agony. To change this situation we have to think about how studies could be refined without interfering with the predictive value of the test. One way to alleviate any expected pain in animals could be the use of sedatives, analgesics, or appropriate anaesthetics. As an example of a test which should be urgently refined in this regard is the 'acute inhalation toxicity' of chemicals, usually tested in rats. According to the OECD guidelines, animals to be tested are placed in a chamber for 4 h and are exposed to the test substance. The inhalation of irritants can cause serious injuries to the respiratory tract. However, although not prescribed in the guidelines, animals are usually kept in a tube which does not allow any movement, and so even more distress is caused by the confinement as the rats are unable to respond to the irritation by moving away. Another procedure calling for the alleviation of pain is the test method for ocular and dermal toxicity in the use of local and systemic analgesics. Toxicologists are afraid that the use of sedatives in toxicity studies will introduce an unwanted variable into the experiment. But how large is the risk? Can it outweigh our moral obligation to minimize suffering in these laboratory animals?

The refinement of test methods should be only one step. It may be advantageous to go one step further and to question the underlying philosophy of the toxicity testing of chemicals which still is based on 'death as the endpoint'. If there are no exceptional scientific reasons for insisting upon death as the endpoint, a more-humane endpoint such as 'evident and marked signs of toxicity' should be mandated. In my opinion, signs of poisoning are the principal information we need from toxicological testing. I know, of course, that important objections are brought forward against this proposal but the potential loss of 'valuable' data must be weighed against the pain and suffering of so many animals. We have to ask: 'How far would a more-humane endpoint compromise the purposes of testing?' At least we all should agree with the long-term objective of moving away from death as an endpoint in toxicity testing (Mellor & Morton 1997).

If we are serious about our statement that only animal experiments that are morally justified should be performed—this is the official opinion told to the public—then we cannot go on like this without becoming deceitful. So, I wish to call for a working party to be established composed not only of toxicologists, but also of ethologists, pain specialists, ethicists and even consumers. It should be the goal of this group to find a fair balance between the public and scientific demands for safe products and the ethical demand of minimizing the pain and suffering in laboratory animals. In my opinion we are accountable to the public to take the suffering of laboratory animals seriously, in order

to retain the public's consent to the continued use of laboratory animals in experiments.

## References


Bundesamt für Veterinärwesen (1994) *Retrospektive Einteilung von Tierversuchen nach Schweregraden (Belastungskategorien)*. CH-3097 Liebefeld-Bern

Coligan JE, Kruisbeek AM, Margulies D, Shevach EM, Strober W, eds (1994) *Current Protocols in Immunology*, Vol 3, National Institutes of Health. New York: John Wiley and Sons

Dawkins MS (1990) From an animal's point of view: motivation, fitness, and animal welfare. *Behavioural and Brain Sciences* **13**, 1–61

FELASA Working Group (1994) Pain and distress in laboratory rodents. *Laboratory Animals* **28**, 97–112

Huxley A (1983) Anniversary Address by the President. *Supplement to Royal Society News* **2**, No. 6

ILAR (1992) *Recognition and Alleviation of Pain and Distress in Laboratory Animals, Institute of Laboratory Animal Resources*. Washington DC: National Academy Press

LASA Working Party (1990) The assessment and control of the severity of scientific procedures on laboratory animals. *Laboratory Animals* **24**, 97–130

Mellor DJ, Morton DB (1997) Humane endpoints in research and testing. Synopsis of the workshop. In: *Animal Alternatives, Welfare and Ethics* (van Zutphen LFM, Balls M, eds). Amsterdam: Elsevier Science, pp 297–9

Morton DB, Griffiths PHM (1985) Guidelines on the recognition of pain, distress and discomfort in experimental animals and an hypothesis for assessment. *Veterinary Record* **116**, 431–6

Morton DB (1997) A scheme for the recognition and assessment of adverse effects in animals. In: *Animal Alternatives, Welfare and Ethics* (van Zutphen LFM, Balls M, eds). Amsterdam: Elsevier Science, pp 235–40

Morton DB (1999) Humane endpoints in animal experimentation for biomedical research: ethical, legal and practical aspects. In: *Humane Endpoints in Animal Experiments for Biomedical Research* (Hendriksen CFM, Morton DB, eds). London: Royal Society of Medicine Press, pp 5–12

Soma LR (1987) Assessment of animal pain in experimental animals. *Laboratory Animal Science* (Special Issue), 71–4

Spinelli JS, Markowitz H (1987) Clinical recognition and anticipation of situations likely to induce suffering in animals. *Journal of the American Veterinary Association* **191**, 1216–18

Swiss Academy of Medical Sciences and Swiss Academy of Sciences (1997) Ethical principles and guidelines for scientific experiments on animals. *Alternatives to Laboratory Animals* **25**, 379–84