

Recognising and assessing pain, suffering and distress in laboratory animals

A survey of current practice in the UK with recommendations



Penny Hawkins
Research Animals Department
RSPCA, Wilberforce Way, Southwater,
West Sussex, RH13 9RS, UK
Research_Animals@rspca.org.uk

July 2002

Contents

	Page
1 Introduction	4
2 Objectives of the survey	5
3 Initial assumptions	5
4 Current techniques for assessing animals and recording observations	6
4.1 Clinical observation sheets	6
4.2 'Score sheets'	6
4.3 Data management systems	7
4.4 Phenotype assessment protocols, e.g. SHIRPA	7
4.5 Visual Analogue Scales	7
5 Method	7
6 Results	8
6.1 Acknowledging animal suffering	8
6.2 Recognising 'normal' animals and the presence of clinical signs	9
6.2.1 <i>How relevant clinical signs are decided for each project</i>	9
6.2.2 <i>Clinical signs used in practice</i>	9
6.3 Methods for aiding animal monitoring and record keeping	10
6.3.1 <i>Score sheets in practice</i>	12
6.4 Avoiding and alleviating adverse effects	14
6.5 Policies on analgesia	17
6.6 Reviewing records and practice	18
6.7 Communicating good practice within and between establishments	19
6.8 People and training	20
6.8.1 <i>A team approach</i>	20
6.8.2 <i>Ultimate responsibility</i>	22
6.8.3 <i>The status of animal technicians</i>	22
6.8.4 <i>Training to recognise pain, suffering and distress</i>	22
6.8.5 <i>Areas where more training is required</i>	24
6.9 Problems in practice	25
6.9.1 <i>Animals concealing discomfort, pain and distress</i>	25
6.9.2 <i>Detecting incipient pain and distress</i>	26
6.9.3 <i>Human subjectivity</i>	26
6.9.4 <i>Human habituation</i>	27
6.9.5 <i>Different consideration for different species</i>	27
6.9.6 <i>Time constraints</i>	27

6.9.7	<i>Genetically modified animals</i>	28
6.10	Miscellaneous good ideas/practices	29
7	Summary of results	29
7.1	Awareness of the potential for suffering	29
7.2	Recognising, assessing and monitoring pain, suffering and distress	30
7.3	Preventing, alleviating and controlling adverse effects	30
7.4	Reviewing the management of pain, suffering and distress	31
7.5	Training issues	31
7.6	Communicating good practice	31
8	Conclusions and recommendations	31
8.1	Awareness of the potential for suffering	32
8.2	Recognising, assessing and monitoring pain, suffering and distress	33
8.3	Preventing, alleviating and controlling adverse effects	34
8.4	Reviewing the management of pain, suffering and distress	35
8.5	Training issues	36
8.6	Communicating good practice	37
8.7	Areas where more research is needed	37
	References	39
	Table 1	43
	Table 2	44
	Table 3	45
	Tables 4a and 4b	46
	Table 5	47
	Tables 6a and 6b	48
	Table 7	49
	Table 8	50
	Appendix 1 Recommended reading	52
	Appendix 2 Example clinical observation sheet	56
	Appendix 3 Examples of score sheets (i to iv)	57
	Appendix 4 Questionnaire used to carry out the survey	61
	Glossary of relevant UK terminology	62

Summary

A survey was undertaken to evaluate how animal pain, suffering and distress are recognised and assessed in UK scientific procedure establishments designated under the Animals (Scientific Procedures) Act 1986. A total of 28 institutions were visited between June 1999 and April 2001, within which 137 people were interviewed including scientists, veterinarians and animal technicians. All 28 establishments use clinical observation sheets to assist the recognition of adverse effects, 9 use score sheets and 7 use computerised data management systems. Clinical signs used as indicators of potential pain, suffering or distress are largely subjective.

The survey also addressed protocols and methods for avoiding and alleviating adverse effects, record keeping, review of policies and protocols and issues relating to teamwork and training. Respondents use a range of techniques for reducing suffering including analgesia, humane endpoints, ensuring competence and refining husbandry. All establishments review projects regularly but few have the time or resources formally to review adverse effects noted in practice and to compare observations with predictions made in licence applications. Training is very consistent between different establishments and most aim to achieve a 'team approach' for monitoring and assessing animals.

Results are set out in full in the present report and summarised in an abridged paper published in *Laboratory Animals*. Both versions of the report, including its recommendations, are intended to provide a source of information, discussion topics and ideas for all establishments that need to monitor animal wellbeing.

Keywords Animal welfare; refinement; pain; suffering; distress; pain assessment; pain scoring; pain management; animal monitoring; humane endpoints; analgesia; environmental enrichment; ethics committee

1 Introduction

A fundamental concern about the use of animals in research and testing is the potential for scientific procedures to cause pain, suffering or distress (Aldhous et al. 1999, Plous 1999). The requirement to reduce suffering to a minimum is widely recognised and is central to many laws that regulate animal experimentation, such as the UK Animals (Scientific Procedures) Act 1986 (A(SP)A) and the US Animal Welfare Act (National Research Council 1996). Discomfort, pain or suffering obviously cannot be minimised unless it is effectively recognised in the first instance, yet recognising suffering is unfortunately not as straightforward as it may seem (Wallace et al. 1990, ILAR 1992, FELASA 1994, Flecknell 1994, 1996, Scharmann 1999, Dobromylskyj et al. 2000).

A number of techniques have been devised to assist with animal monitoring and the recognition of discomfort, pain and distress, some of which are set out in Section 4 below. Nevertheless, their effectiveness has not been widely evaluated in practice (see Flecknell 1996) and the extent to which they are used is not known. Lloyd et al. (2000) conducted an email survey of the use of different assessment techniques to which there was a poor response (over 800 people were contacted and 21 replied). Of those who did reply, most (75 %) used purely subjective measures of welfare such as

appearance and natural and provoked behaviour (Lloyd et al. 2000).

In addition to the methods chosen to monitor and assess animals, there are a number of other important factors that have a direct bearing on the ability of individuals and establishments effectively to minimise animal suffering. These relate to the training of animal users and/or carers, the role of institutional committees, and dissemination of information about good practice. The current project therefore aimed to conduct a broader survey that would examine these issues, establish current practice for monitoring animals in the UK and identify areas in which practice could be improved.

2 Objectives of the survey

The objectives of the survey were to evaluate:

- how animal well-being and departures from well-being (i.e. pain, suffering, distress) are recognised and assessed in a range of UK designated research and testing establishments;
- how records of adverse effects and clinical observations are made;
- how observations and records are integrated into pain/distress management protocols and policies;
- how staff are trained to recognise, assess and alleviate animal distress, discomfort and pain;
- how effectively pain, suffering and distress are controlled;
- how pain management is perceived at the project planning stage, and how the system is applied;
- what role the UK Ethical Review Process (ERP) plays in minimising the impact of adverse effects;
- how good practice with respect to all of the above is disseminated.

The present report aims to provide a ‘snapshot’ of current practice for assessing and monitoring the wellbeing of research animals in the UK, so that other individuals and establishments both in the UK and elsewhere can use it as a source of information and ideas and as a measure of their own standards. Some of the issues within it will be relevant to the work of animal use and welfare committees such as the UK ERP and US IACUC (Institutional Animal Care and Use Committee). It is hoped that other aspects will be useful to those responsible for training people who will then go on to use and/or monitor laboratory animals. The present report necessarily refers to UK legislation and uses UK terminology, but the issues it raises are relevant wherever laboratory animals are used (UK legislation and terms are printed in **bold** within the text and defined in the Glossary on p 62).

3 Initial assumptions

Many publications have examined the nature of nociception, pain and suffering and whether animals are capable of experiencing any or all of them (see Appendix 1 for recent examples). The current consensus is that all vertebrates, and probably many invertebrates, are capable of experiencing pain; such animals are generally given the ‘benefit of the doubt’ and considered also to be capable of suffering. The present study therefore makes the following assumptions:

- animals experience (i.e. suffer) distress, discomfort and pain;

- analgesics or other effective interventions are available and should always be administered if animals are experiencing discomfort or pain, unless there is compelling veterinary justification not to (NB details of analgesic regimes are covered in the veterinary literature and fall outside the scope of this project);
- humane endpoints should always be employed and re-evaluated regularly;
- prevention of pain, suffering or distress is the ideal; if this is not possible then prompt recognition and treatment are essential.

4 Current techniques for assessing animal well-being and recording observations

Before beginning the survey, laboratory animal veterinarians were consulted and a literature search conducted, to see which methods currently exist to record observations and assist with judgements on animal wellbeing. A list of reference material is set out in Appendix 1. The following techniques were found. They vary in complexity and objectivity but are all based on recording clinical signs.

4.1 Clinical observation sheets

Clinical observation sheets are widely used for noting simple, objective measures such as body weight and for logging inspection times and any observed adverse effects. Sheets used to record clinical observations generally have a relatively simple format which permits the entry of 'free text', i.e. written descriptions of any changes or clinical signs (see example clinical observation sheet in Appendix 2).

4.2 'Score sheets'

The principle of 'score sheets' for noting and assessing clinical observations was originally suggested by Morton & Griffiths (1985). A system was proposed where behaviours associated with discomfort, pain and distress and other clinical signs were assigned numerical scores according to their severity so that a total 'score' from an overall assessment could be interpreted and acted upon accordingly. The authors acknowledged the inherent difficulties associated with assessing animal pain and suffering and presented the initial scheme as a prototype that required validation over a wide variety of experimental conditions and assessors (Morton & Griffiths 1985).

The concept of the 'score sheet' has subsequently evolved and become more flexible (Wolfensohn & Lloyd 1998, Lloyd & Wolfensohn 1999, Scharmann 1999, van der Meer et al. 2001). In particular, binary score sheets have been introduced, where clinical signs are marked simply as present or absent (Morton 1990, 1995, 1997, 1998a, 1998b, Morton & Townsend 1995). The term 'score sheet' is thus a misnomer when applied to binary sheets, as numerical scores are frequently not required. This revised approach to setting out and implementing score sheets involves listing likely clinical signs on an observation sheet for checking, with a free text box for writing down other, significant signs that have not been listed (Morton & Townsend 1995, Morton 1997, 1998a, 1998b, 1999). The sheets need to be regularly reviewed so that signs frequently noted in the text boxes can be added to the list and those that are infrequently observed can be removed. Objective measures of health and/or development such as body weight are also included within most sheets (Morton 1990, 1995, 1997, 1998a, 1998b, 1999). For examples of score sheets collected during the

present survey, see Appendix 3. These can be downloaded and edited to suit different studies.

4.3 Data management systems

Preclinical data management systems are commonly used in toxicology and safety testing, e.g. Path/Tox System™ (Xybion Medical Systems) or Datatox™ (Instem). These systems operate in a similar manner to score sheets and include lexicons with lists of terms for observations of environmental conditions and clinical signs, entries for dose routes and levels, and boxes for free text. Master lexicons can generally be adapted for specific studies. Most lexicons use simple, descriptive language as opposed to diagnostic terms. Observations are recorded using a computerised system, where data are either entered directly or from observation sheets.

4.4 Phenotype assessment protocols, e.g. SHIRPA

There are a number of protocols for assessing the phenotype of genetically modified or mutant animals, which may indicate that animals are experiencing pain, suffering or distress although this is not their primary function. Perhaps the most frequently cited is SHIRPA (SmithKline Beecham Pharmaceuticals; Harwell, MRC Mouse Genome Centre and Mammalian Genetics Unit; Imperial College School of Medicine at St Mary's; Royal London Hospital, St Bartholomew's and the Royal London School of Medicine Phenotype Assessment). SHIRPA is a three-stage assessment protocol, where the first two stages are a general phenotype assessment and the third stage is a more specialised screen that is primarily tailored to neurological deficits. For more details see

http://www.mgu.har.mrc.ac.uk/mutabase/shirpa_summary.html

4.5 Visual Analogue Scales

Visual Analogue Scales (VAS) generally take the form of lines between two points usually defined using terms such as 'no pain' and 'worst pain possible'. The observer marks the line at the point that s/he believes to be analogous to the pain that the animal is experiencing (e.g. Conzemius et al. 1997, Cambridge et al. 2000, Slingsby & Waterman-Pearson 2000, 2001). VAS are also used to record pain levels in humans, where they may be marked by the patient, or a carer if the patient is unable to do so.

5 Method

The present survey was carried out by visiting a range of UK research and testing establishments and discussing issues relating to the management of adverse effects, using a questionnaire as a basis for discussion (Appendix 4). The survey did not include any laboratory animal breeding or supplying facilities. A total of 28 institutions were visited between June 1999 and April 2001, which comprised almost 11 % of all establishments designated under **A(SP)A** in the UK at that time (Home Office 2001). The sample is therefore believed to represent a significant proportion of UK user establishments (Table 1, p. 43). It was not possible to select a random sample of establishments as there is no publicly available central list, so the survey was conducted among those where the RSPCA already had direct or indirect contacts.

The aim was to interview six people at each institution; one **NVS** or Deputy NVS, one **NACWO** and four **project** or **personal licensees**, comprising two scientists and two animal technicians or senior animal technicians. A total of 137 people took part (see Table 2, p. 44). Most were interviewed on their own, although some facilities provided two or more people together or asked the NVS to sit in throughout.

All interviews were based on the sheet in Appendix 4 and the respondents had not seen the questions beforehand, although they were aware of the purpose of the project. Written questions were kept relatively short and explained in greater depth verbally at the time of the interviews, because the survey addressed broad topics and it was difficult to word questions so that they always meant the same thing to different people. For example, the first question “Is it routinely assumed that pain is present in animals during or following procedures?” applied to the individual respondent’s assumption, not the establishment as a whole and included discomfort and distress as well as acute pain, all of which was made clear. The scope of the survey included the monitoring of stock animals, procedures involving potential surgical pain, adverse effects due to infection, toxicity and administering substances, tumour growth and genetic modification. Responses were analysed by first extracting quantitative results (e.g. the number of establishments using clinical observation sheets) and then reviewing all interviewees’ answers and statements to see whether common themes emerged. All participants were given the opportunity to comment on the present report before it was submitted for publication.

6 Results

Although numerical results are included wherever possible, many of the results of the present survey are descriptive. This is because every establishment had its own individual culture and employed different policies and protocols to implement the minimum legal requirements of the **A(SP)A**. As a consequence, people explained their practices and concerns in a variety of ways and often wanted to include other issues, opinions and thoughts that were important to them. Despite this, there were many common practices and themes; these are set out below using examples to illustrate them where appropriate.

6.1 Acknowledging animal suffering

The overwhelming majority of respondents (97 %) assumed that animals did or may experience adverse effects¹ to some extent, either during the procedures that they conducted as part of their projects or as a result of those procedures. When asked the question “do you *routinely* assume that adverse effects are present in animals during or following procedures?”

- 112 people (82 %) answered “yes”;
- 4 (3 %) answered “no”;
- 21 people (15 %) felt that pain, suffering or distress were possible but usually prevented in the projects on which they were currently working (see Section 6.4).

¹ This includes pain, suffering and distress, from mild discomfort, stress (e.g. due to handling for administration of substances) or ‘feeling sick’ through to higher levels of suffering such as post-surgical pain.

Many people spoke of different types of pain and included discomfort as a cause of distress, acknowledging the emotional component of animals' experience of pain. This could include animals not understanding why their bodies no longer worked as they once had, for example following an artificially induced stroke. Most respondents also acknowledged that adverse effects may be caused by both administering substances (e.g. distress caused by oral gavage, momentary pain due to an injection) and by the effects of the substances themselves (e.g. toxic effects of a test compound).

6.2 Recognising 'normal' animals and the presence of clinical signs

Almost everyone believed that the ability to recognise a 'normal' animal was a fundamental skill that had to be present or learned in order effectively to recognise an 'abnormal' animal. In practical terms, given that the health and welfare of stock animals is believed to be acceptable, any changes in behaviour or physiology that indicate a departure from this baseline standard of wellbeing could signify that animals are suffering and warrant further attention or intervention. The recognition of both 'normal' and suffering animals was largely subjective in the first instance, but was then assisted using a range of techniques (listed in Section 4).

6.2.1 How relevant clinical signs are decided for each project

For each individual study, appropriate clinical signs to monitor animals and define humane endpoints were decided by a number of different methods including:

- previous experience of scientists, veterinarians and technicians;
- results of *in vitro* and computer screens;
- pilot studies;
- information supplied on a compound to be tested;
- contacting other laboratory animal veterinarians for advice;
- thinking about how one would assess the wellbeing of a companion animal;
- predicting adverse effects from the literature (for example by using Medline to search for similar studies);
- for human disease models, considering the clinical signs in humans;
- referring to UK **project licences**, which must describe potential adverse effects;
- using criteria on existing score sheets;
- using a publication on pain and distress in laboratory rodents and lagomorphs by the FELASA² Working Group on Pain and Distress (1994);
- consulting online discussion groups such as Compmed and VOLE (see Table 6a, p. 48).

One pharmaceutical company had its own literature database that it used to check predicted adverse effects. Use of this database was made at the project planning stage so that more comprehensive information could be presented in the relevant sections of the project licence.

6.2.2 Clinical signs used in practice

Both subjective and objective clinical signs were commonly noted (Table 3, p. 45). The most important core criteria were considered to be simple, objective and non-

² Federation of European Laboratory Animal Science Associations

invasive, i.e. body mass and food and water consumption. Some animal technicians felt strongly that animals should be initially assessed in the simplest possible way, so that a decrease in food and water consumption and body mass at an early stage in a procedure would indicate that something was wrong and that the animal needed to be carefully assessed and judgement made on subsequent endpoints. This ideal was dependent on the numbers of animals to be monitored and the time available to do this.

There was no universally agreed measure of wellbeing, however, as the relevance of clinical signs was said to vary between species and procedures. Body mass was regarded as a useful, objective measure by more respondents than any other parameter, although it was not always believed to be definitive. For example, animal technicians and scientists stated that in some procedures, e.g. stroke research, the brain's feeding centres can be damaged so that the animal does not feed adequately, but wellbeing may not necessarily have been adversely affected. In some facilities, staff believed that group-housing animals rendered food and water uptake data useless and considered that the benefits of group-housing social animals were more important, so food and water consumption were not monitored.

Clinical signs that were regarded as more conclusive indicators of adverse effects by the majority of respondents were piloerection ('starey' coat) and previously social animals becoming withdrawn. Telemetered body temperature, where available, was occasionally cited as an important aid for monitoring animals and setting humane endpoints to reduce suffering in disease studies and toxicology. Vocalisation in response to pain was taken seriously by all participants and was said by most to occur only rarely. In the case of rodents, audible vocalisation (as opposed to ultrasound) during or following procedures was taken as an indicator of potential acute, severe pain and was generally regarded as an endpoint. One pharmaceutical establishment used bat detectors to listen to ultrasonic vocalisation in rats (see Sandells in Hawkins et al. 2001a).

Indicators present in the cage were also used to monitor animals by some facilities, e.g. abnormal faeces, vomit, blood on bedding, reduced use of cage additions such as nesting material, cardboard tubes and chew sticks were all causes for concern. In these cases, the cages had to be cleaned out by those responsible for monitoring the animals' wellbeing so that important indicators were noted and not discarded.

Various signs were put on the front of cages or pens when there were concerns about an animal inside it, including paper clips, 'Post-it'TM notes, coloured clothes pegs and yellow or red laminated cards for slight or more serious concerns respectively. At some establishments, 'shoe box' cages were pulled slightly forward if there were concerns about any of the animals inside. These indicators could be placed by anyone (but were usually placed by animal technicians in practice) and their presence required action from the project licence holder and/or named persons.

6.3 Methods for aiding animal monitoring and record keeping

A range of different techniques were used to aid the recognition and monitoring of adverse effects; for an explanation of each see Section 4. Table 4a (p. 46) lists those routinely used at establishments (i.e. they were mentioned by most people as part of

the regular monitoring protocol and their use was demonstrated to the author). Table 4b (p. 46) lists *all* methods mentioned by everyone (i.e. techniques that they were aware of and/or had used before) whether they were regularly used, used in a limited number of studies or not currently used within an institution.

Clinical observation sheets (see Table 3, p. 45) for examples of commonly used clinical observations and Appendix 2 for an example sheet) were used at all establishments.

‘Score sheets’ were mentioned by 40 people (29 % of respondents) but were used at 9 institutions, and then not universally. One university and two pharmaceutical establishments used binary score sheets; all the other establishments using score systems assigned numerical scores to clinical signs. There is thus a background awareness of score sheets but their use is not widespread (see Section 6.3.1 below).

Data management systems were used at all Contract Research Organisations (CROs) and within the toxicology departments of two of the pharmaceutical establishments. Subjective terms such as ‘subdued’ were usually avoided, as were terms that required veterinary or histological diagnosis. For example, respondents explained how ‘red stains’ on a rat’s nose could be due to either bleeding or porphyrin secretion, and terms such as ‘necrosis’ can only be diagnosed by performing histological studies. The present study found that more free text was generally used to describe clinical signs in larger animals (e.g. dogs, primates) than rodents.

Specific clinical signs In some types of procedure, experimental objectives could also be used to infer animal wellbeing and these were used as prime indicators that animals were experiencing adverse effects. For example, one scientist studying enzyme deficiency diseases at a university used the ability of mice to cross a bar without falling to infer wellbeing as well as to assess disease progression. An establishment conducting orthopaedic research used force plates to analyse gait and thus indicate whether animals could be in pain (NB changes in gait may also be due to purely mechanical causes rather than discomfort or pain (K Clarke pers. comm.)). Three respondents citing specific clinical signs were members of groups developing candidate analgesics and used the animals’ responses to pain (e.g. tail flicks) elicited as part of the study. Of the 7 people who used specific clinical signs, 6 were scientists and one was a senior animal technician.

A **‘Blip book’** was used at one university to record unpredicted phenotypes in transgenic animals and to undertake monthly reviews of relevant projects. The book had columns as set out below and technicians were instructed that if they observed an unpredicted phenotype they should (i) inform the team leader (who should then inform the site supervisor), (ii) inform the vet or vet nurse, (iii) fill in a health sheet, (iv) fill in a ‘blip’ form. A behavioural or health problem that occurred three times was regarded as a new adverse effect associated with the project such that both the project and personal licensees had to be informed.

Occurrence	Date	Adverse reaction	Source	Genotype	Sex
1					
2					
3					
4					
5					

Video stills were used in one fundamental research establishment to monitor rodents undergoing vaccine trials.

SHIRPA was cited by one person at an academic establishment using genetically modified and mutant mice. There were some doubts among those who used or cared for transgenic animals as to the utility of SHIRPA for assessing phenotypes, but SHIRPA was regarded as a useful tool for the routine screening of mutant mice.

Health is by no means synonymous with welfare, but some species have little capacity to express their state of welfare. Alternatively, humans' interpretation of changes in their behaviour is especially poor. In such cases, the animals' health was taken as the primary indicator of their welfare. Measures of health were used in this way by two scientists at separate academic establishments, using *Xenopus* and zebrafish (*Danio rerio*).

Specific behaviours such as exploration, "inquisitiveness" and (in the case of primates and dogs) desire to interact with humans were cited by some people as their initial and primary criteria for assessing wellbeing, rather than more objective clinical signs. Specific behaviours were mentioned by 5 people (4 %); one from a CRO and 4 from pharmaceutical establishments.

Ultrasonic vocalisation by rats (made audible using a bat detector) was routinely used at one pharmaceutical company that evaluates the effectiveness of candidate analgesics. Note that while such vocalisation undoubtedly occurs and often changes following procedures (Kaltwasser 1991, Miczek et al. 1992, Calvino et al. 1996), it is very difficult to interpret and relationships have not been comprehensively established between vocalisation patterns and possible suffering or distress.

Visual Analogue Scales were not used by any establishments visited during the survey.

In general, a wider variety of methods were cited in academia than industry, which is probably due to the broader range of studies undertaken at universities and other fundamental research establishments. The importance of establishing baseline data using objective criteria (body mass, food and water consumption at least) by observing and/or scoring animals 2 to 3 days before all procedures was frequently stressed within all types of establishment. It was also made very clear that any system is only as good as those using it, so effective training and teamwork are essential (see Section 6.8 below). One CRO described its approach as "holistic", in that each animal was 'owned' by a technician who knew the whole animal and her/his history and was responsible for her/his wellbeing.

6.3.1 Score sheets in practice

Score sheets were described positively as flexible and permitting input from both technicians and scientists – "different people notice different things". Some establishments involved technicians when initially setting out score sheets, whereas other institutions used technicians' input when modifying sheets that were already in use. Score sheets were also regarded as helping people to learn about their animals

and encouraging animal technicians, who might have large numbers of animals to monitor, to look critically at them all. Establishments that used score sheets successfully all stressed that they could and should always be updated and improved: “they are never static; they must evolve”.

Other people at institutions that did not use score sheets appeared not to realise this, with some criticising them on the grounds that those using them would only look for criteria listed on the sheets and overlook other signs that may have been important. Score sheets were also described (largely by those who did not use them) as “inflexible” or “rigid”, but effective where criteria were fairly specific. Many people felt that, with very complex score sheets, their primary purpose can become lost within all the data generated, so that simple, objective criteria were best. One scientist expressed this as “a balance between having lots of boxes to tick and having a positive reporting procedure that will pick up details quickly”.

In an ideal situation, clinical signs would be recognised and scored in the same way by everyone, but this is unlikely to be achieved in practice. Everyone using score sheets recognised this and many stated that pain must always be assessed (at least initially) in the simplest possible way, i.e. using body mass and food and water consumption. A decrease in any of these parameters was taken to signify that something was wrong and that an animal needed immediate and careful assessment with further guidance provided by the score sheets. New score sheets for any type of procedure were therefore based on the objective criteria listed in Table 3 (p. 45), with additional clinical signs obtained from the project licence and with input from technicians and scientists. Establishments using score sheets explained that it did take time to train people to use them correctly and effectively, so animals were initially scored by the team as a whole. Technicians and scientists would also regularly check one another’s scores following the training period.

The inherent variability between observers was cited as the reason for the move from numerical to binary score sheets. One university updated its binary score sheets constantly, removing irrelevant observations and adding new ones. Blank spaces were left in score sheets and room diaries for technicians (including trainees) and scientists to add new observations as they arose. The **NVS** and **NACWO** actively motivated all licensees to do this, for example by encouraging them to think how they would know if a companion animal became unwell. Technicians and named persons at this particular establishment strongly believed that such detailed observations contributed positively to the science and maximised the scientific benefit of the research conducted there.

Where score sheets were routinely used, they were believed to help to:

- ensure close observation of all animals;
- ensure consistency of monitoring;
- train new staff, raising their awareness and confidence;
- identify and refine humane endpoints more effectively;
- indicate specific husbandry requirements;
- record specific effects of drugs such as analgesics;
- record the effectiveness of refinements;
- compare the effects of old vs. new models of disease (see also Morton et al. 2000).

6.4 Avoiding and alleviating adverse effects

Question 1 about adverse effects and the ways in which they are alleviated was deliberately left open (see Appendix 4), to take into account the diverse ways in which pain, suffering and distress can be caused and, conversely, reduced. Most respondents began by discussing post-surgical pain but many expanded their replies to include a wider range of issues that are summarised below. The broad range of responses to this question indicate that effective management of adverse effects requires knowledge and competence with respect to: administering analgesia, refining techniques and husbandry, ensuring staff are competent and empathetic and ensuring that the ‘culture’ within the establishment as a whole is appropriate.

Analgesia

This section sets out analgesia *practices* as opposed to policies. The pain relief administered to animals undergoing surgery in practice reflects the way in which institutions comply with the minimum requirements of the **A(SP)A**. Analgesia *policies* are not a legal requirement in the UK, but some establishments had chosen to put such policies in place to help ensure that pain relief was fairly and properly implemented. This should not be taken to suggest that animals are necessarily at risk of experiencing avoidable suffering at institutions without policies on pain relief. Policies on analgesia are explained and discussed in Section 6.5.

Of the 28 establishments visited, 25 carried out surgery with recovery. Analgesic agents commonly used were Buprenorphine, the first choice at most establishments, and Carprofen (especially for dogs and primates; one CRO also preferred Carprofen for rodents). Flunixin was frequently used for larger animals and was also cited in epilepsy research where Buprenorphine would have interfered with results. Local anaesthetics such as EMLA cream (AstraZeneca) were also often used for blood sampling where practicable, e.g. when taking blood by venepuncture from the rabbit ear vein.

Twelve establishments administered pain relief pre-emptively at least some of the time, and all also gave analgesia post-operatively, with some exceptions:

- Rats were not given any pain relief when fitted with headpieces at 3 establishments, on the grounds that the brain does not feel pain (it is true that brain tissue does not have sensory receptors, but the periosteum and skin have plenty of nociceptors). The rats were not weighed over a period of time pre- and postoperatively to substantiate the decision not to administer pain relief.
- Mice were not given analgesia before or after embryo transfer at one establishment. Hypothermia and slow recovery following dosing with Buprenorphine was cited as the reason for this (NB another establishment did administer Buprenorphine to mice undergoing embryo transfer).
- *Xenopus* were not given any analgesia for surgical oocyte removal; the researcher would have liked to provide pain relief but had not been able to find a dosing regime in the literature.
- One pharmaceutical establishment gave both rodents and larger animals pre-emptive analgesia, but only the larger animals received post-operative analgesia.

Dosing regimes also varied in practice. For example, protocols at 3 different establishments were: (i) a minimum of 3 doses at 8 hour intervals for all species; (ii)

once on the day of surgery and once the day after for rodents; (iii) as required for 24 hours followed by reassessment for all species. Three academic establishments and 2 CROs specified a minimum number of doses or durations of post-operative pain relief, while the other establishments relied on clinical judgement and consultation with the NVS.

- For further guidance on anaesthesia and analgesia, see Flecknell (1996), Flecknell & Waterman-Pearson (2000) Hellebrekers (2001).

Humane endpoints

Where there was no scope for using analgesia to alleviate pain (e.g. many safety tests and toxicology studies) or where there was believed to be no justification in keeping an animal who was suffering alive, humane endpoints were implemented at all establishments. Under the UK licensing process, endpoints must be defined and set out in the relevant section of the **project licence** application form before a study will be considered by the **Home Office Inspectorate**.

- For more information on recent developments in judging humane endpoints, see Hendriksen & Morton (1999), ILAR (2000), OECD (2000).

Refining the administration of substances

Many respondents believed that, when administering substances to animals, it is extremely important to consider the nature of the substance and how its impact on animals can be reduced. Inappropriate pH of a material, incorrect grade of needles, excessive dosing volumes and high or low substance and/or vehicle temperature were all cited as potential causes of suffering that ought to be avoided. Considering the nature of the apparatus carefully was also mentioned by one respondent, for example by using flexible catheters for oral gavage to reduce the risk of mis-dosing or injury.

- For further guidance on refining the administration of substances including routes, substances, species, techniques and staff competence, see Morton et al. (2001). LASA has also produced guidance on administering substances (1998a); for blood sampling see Morton et al. (1993a), LASA (1998b).

Handling

Many people working within CROs explained that they tried to calm animals during the administration of a substance (e.g. during inhalation) by stroking them, speaking to them and trying to make them feel secure.

- Training resources that encourage sympathetic animal handling include a video, *Handle with care* (IAT 1986) and a British Small Animal Veterinary Association CD-ROM on *Practical animal handling: small mammals* (<http://www.lal.org.uk/digital.htm>); see also LASA (1998c).

Habituating and training animals to dosing

Acclimatising animals to the facility before procedures begin is likely to reduce distress when procedures begin (see Laule 1999) and many interviewees believed that this also enabled animals to build a relationship with technicians. All CROs insisted on this and also allowed extra time for training animals to accept procedure rooms, restraint devices such as dog slings or primate chairs and stressful dosing procedures

such as inhalation (by habituation to the mask) and oral gavage (by sham dosing for a fixed period). The toxicology department of one pharmaceutical company allowed 7 to 14 days for animals to become acclimatised to their cages and then introduce three periods of sham dosing, so that animals did not have to undergo “huge insults” on day one of the study. One CRO also stated that habituation could not be done if staff were too busy, however.

All CROs also made efforts to fit individual animals to studies; for example, those who become stressed during restraint could be put into groups undergoing shorter dose durations. The CROs also promoted group housing for social species to clients wherever possible and allowed animals to become acclimatised to their groups before going onto procedures.

It is sometimes possible to train animals to cooperate with procedures (Laule 1999), but the present study found that emphasis was almost entirely on rewarding animals such as dogs and primates once procedures were complete. For example, one establishment gave macaques yogurt covered peanuts following manual restraint for dosing. However, in one establishment sheep were trained to cooperate with procedures using sheep nuts.

- For more on training animals (mainly primates), see Heath (1989), Luttrell et al. (1994), Laule (1999), Reinhardt (1991, 1997a).

Husbandry and environmental stimulation

Some establishments group housed social animals before and after surgery, forming stable groups in advance and reforming them as soon as possible (unless there was a risk of trauma to individuals). Besides the presence of conspecifics, a stimulating environment was also cited by some as shifting animals’ attention away from the surgery and (they hoped) reducing suffering. One pharmaceutical establishment had a policy not to remove food and water from rodents before surgery without scientific justification.

Following surgery, it was common practice to try to make animals more physically comfortable by giving soft diets, such as mash, baby food or Complian™, subcutaneous fluid, more bedding or heat pads as appropriate. All of these were supplied to elderly rodents at establishments where projects required aged animals. Other pathologies also received special husbandry considerations as appropriate. For example, one establishment using diabetic rats tilted their cages slightly so that they could still have solid floors and nesting material despite the presence of polyuria; another that used transgenic ‘waltzer’ mice provided them with circular guards that they could jump in and out of but that were believed to provided them with a feeling of security.

- Some useful references on husbandry refinements are Morton et al. 1993b (rabbits), AWIC 1995 & Reinhardt 1997b (both cover a range of species), Jennings et al. 1998 (mice), Hawkins et al. 2001b (birds), Reinhardt & Reinhardt 2001 (rhesus macaques), RSPCA/UFAW Rodent Welfare Group reports (contact RSPCA Research Animals Department).

Competence

Many senior technicians and **named persons** believed that it was fundamentally important to ensure that everyone carrying out procedures was appropriately trained and competent, and that animals were handled proficiently in the hope that they would feel secure (NB this is also a legal requirement under the A(SP)A). A pharmaceutical company paid special attention to rats on long term studies that required repeated dosing, since fast dosing followed by dropping the animals back into their cages can lead to foot lesions and ankle damage over a 2-year period. It therefore ensured that rats were dosed at an appropriate speed and gently put back into their cages. A scientist at another pharmaceutical company believed very strongly that stress caused by incompetent handling and dosing could compromise the validity of the science, in which case “you have ruined animals’ lives for nothing”.

Restricting surgery times

Some establishments only conducted surgical procedures in the morning (and some not at all on Fridays) so that animals could be monitored during the rest of the day and given additional pain relief if necessary. Although this was generally easier to enforce in pharmaceutical establishments and CROs than academic establishments, one university department also restricted the type of procedures that could be performed outside the normal working day. Scientists and technicians at one pharmaceutical and two academic establishments would occasionally stay with animals all night to monitor them if necessary, but this was by no means common practice.

6.5 Policies on analgesia

Six of the 25 establishments conducting surgery with recovery had clearly set out policies on the provision of analgesia (Table 5, p. 47). Ten others insisted that, where appropriate, post-operative analgesia had to be set out in **project licence** applications.

This, of course, does *not* mean that analgesia is withheld unless there is a policy instructing people to provide it. For example, 12 establishments administered pre-emptive analgesia at least some of the time, although only 4 had policies instructing animal users to do this. Appropriate pain relief is a requirement of UK legislation (see **A(SP)A** entry in Glossary) and the legal and moral imperative to administer analgesia wherever needed was widely recognised – inconsistencies generally lay within the recognition of the presence of pain that required alleviation and the duration of analgesia provided.

Five establishments (all academic) had a written policy that analgesia must be administered post-operatively. Two of these policies applied to all species, including rodents, and also stipulated that if commonly used agents such as Buprenorphine were not suitable, another had to be found at the project licence application stage. Emphasis was very much on post-operative pain relief, although one CRO had an explicit, written policy that post-operative and pre-emptive analgesia should be given to all species. Another pharmaceutical company had a written policy stating that post-operative or pre-emptive analgesia should be given.

Policies varied according to their prescriptiveness, especially with respect to the duration of post-operative analgesia. The CRO with the policy on pre-emptive

analgesia also stipulated a minimum of 3 doses of Buprenorphine following surgery, depending on the clinical signs observed. Many establishments required pain relief to be administered ‘as appropriate’ and relied on the NVS and ERP to ensure that adequate analgesia was given. All establishments with policies in place had found that scientists did not express resistance, although some of them had not thought about analgesia for rodents before encountering the policies.

There were varying views on the necessity of an institutional policy on the alleviation of adverse effects. Some respondents felt that policies are a good way to “make a statement” about animal care. For example, people at one academic establishment believed that policies were vital to ensure that everyone understood why pain relief and humane endpoints were in place and that everyone was implementing them in the same way, so that all animals received the same consideration and treatment. Policies were also said to depersonalise issues, and, if devised with input from management, scientists and technicians they were found to be practical and easy to employ. Respondents at other institutions felt that, with a good culture of care in place, a policy on pain management and humane endpoints was not necessary – refined protocols were set out within the project licence which was seen as more of a “living document”.

6.6 Reviewing records and practice

All establishments reviewed aspects of their research projects regularly, using the ERP or separate committees that fed into it such as the ‘Animal Health and Welfare Committee’, ‘Animal Care and Use Committee’ or ‘Procedure Review Panel’. These committees met regularly, e.g. every month or 4 weeks, but all establishments stressed that any adverse effects or welfare problems would always be dealt with immediately they were detected, without waiting for the committee(s) to meet.

Establishments differed, however, in the implementation of formal processes to review observed adverse effects and to compare them with those predicted in the project licence applications. Many establishments held regular meetings to discuss and review animal health and welfare, but few used clinical observation or score sheets to assist in this process. Others did not have a formal process for retrospective review, but carried out post-study ‘round ups’ or (more frequently) relied on individuals to highlight problems if and when they arose. Within industry, regular review meetings were generally more likely for new or especially complex models but there were no overall systems to review observed adverse effects for every project.

Where score sheets were used, most establishments would refer to them reactively in the event of a problem with an animal or study, but not proactively to assess whether there were chronic problems that were not being detected. Two pharmaceutical establishments and one university did use score sheets proactively to review expected and unexpected adverse effects and pain management protocols. In the case of adverse effects, review was seen as a two-way process in that filled-in sheets were used to (i) revise the list of expected effects of a project and update the sheets and (ii) revise experimental protocols and endpoints. This in-depth revision was conducted either at the end of projects or during regular committee meetings. Score sheets were used as tools for review most extensively at one of the two pharmaceutical establishments that used them. At these establishments, study directors reviewed

score sheets weekly and wrote them up as a ‘flowing log’ so that the **Home Office Inspector** could see exactly what had happened to the animals and whether there were any new concerns, such as a greater incidence of a particular clinical sign. The logs were also used to develop the score sheets, in addition to *ad hoc* alterations.

In the majority of cases, however, there was no formal review of observed adverse effects and observations were not compared with licence applications to see how effectively they were predicted; nor were clinical observation or score sheets routinely used to review training or pain management protocols. The fundamental reasons cited for this were lack of time and resources, particularly within academic establishments that had large numbers of diverse sites and projects. Respondents at one academic establishment also felt that scientists would not be prepared to co-operate.

6.7 Communicating good practice within and between establishments

It is obviously extremely important that information on best practice and good ideas is disseminated both within and between establishments. Table 6 (p. 48) lists the media used for (a) external and (b) internal communication mentioned at each establishment. The roles of professional organisations such as the IAT, LASA and LAVA³ were mentioned by people at most establishments, who used either meetings or (less frequently) publications by these bodies to pass on information about refinements in procedures and the management of adverse effects. All 5 CROs and 5 of the pharmaceutical establishments mentioned industrial discussion groups and visits to one another’s facilities to exchange information on issues such as humane endpoints (NB two of the pharmaceutical companies merged during the course of the present study). Two of the CROs also extended this to formulate an ‘open’ policy whereby any external person (within reason) was allowed to visit and ask about their work. Pharmaceutical establishments in the south of England were able to visit one another most easily; one in the north felt geographically isolated from them and would have liked more opportunities to liaise with others.

Three ongoing UK initiatives that aim to make recent refinement innovations widely available are the BVA(AWF)/FRAME/RSPCA/UFAW Joint Working Group on Refinement Workshops⁴, the RSPCA/UFAW Rodent and Rabbit Groups⁵ and the LASA Refinement Meetings⁶. These were mentioned at one academic and two pharmaceutical establishments respectively, by people who were members of the groups and viewed them as forums for discussing refinement and pain management. NACWOs and scientists mentioned external meetings and publications more than technicians, who generally attached greater importance to passing on information within their establishments. This varied from place to place; at some, technicians were more confident about presenting information and more keen to participate in

³ Institute of Animal Technology, Laboratory Animal Science Association, Laboratory Animal Veterinary Association

⁴ The British Veterinary Association (Animal Welfare Foundation)/Fund for the Replacement of Animals in Medical Experiments/RSPCA/UFAW workshops have produced reports on refinements in (i) blood sampling, (ii) rabbit husbandry, (iii) mouse husbandry, (iv) administration of substances and (v) bird husbandry and procedures, all of which have been published in *Laboratory Animals*.

⁵ These groups hold annual, informal one-day meetings that focus mainly on husbandry refinements; reports are published in *Animal Technology* (now *Animal Technology and Welfare*).

⁶ LASA is running an ongoing series of workshops addressing the refinement of particular techniques, e.g. sciatic nerve crush, rodent stroke models.

conferences than others. Technicians tended to regard the veterinarians as important sources of new information on refinement (mainly of procedures), but this was also seen very much as a two-way flow of information.

Eleven scientists said that they included analgesia protocols when publishing papers, and one also mentioned that a scoring system was used to monitor adverse effects. Including refinements such as analgesic regimes was variously described as “an intrinsic part of publications” and a means of “minimising the learning curve for others”. This attitude was not common to all, however, as other scientists did not mention the fact that they had administered post-surgical analgesia to rodents, either because they believed it would be unwelcome or because it had not occurred to them. Others said that the depth with which they would communicate refinement depended on the journal to which they were submitting their work. **Named persons** at three academic establishments made a point of encouraging scientists to include refinements such as analgesia and animal monitoring schemes in their publications.

Within establishments, the importance of good verbal communication was universally stressed. This tended to be informal, with fewer structured meetings for NACWOs to discuss the management of adverse effects. The **ERP** was mentioned at a range of establishments, but this was more in connection with refining protocols than actively disseminating information.

6.8 People and training

Obviously, adverse effects must be detected and acted upon as rapidly as possible, for the benefit of the animals concerned and those people who care about them. It was frequently stressed to the author that any system for recognising and monitoring pain, suffering and distress would only be as good as the people who implemented it in practice, and that appropriate motivation, attitude and competence are absolutely essential. Having or attaining these qualities was regarded as extremely important, and people also placed great emphasis on the need for ‘team work’, in particular on team members knowing how to work together and progress concerns through the proper channels should they arise.

6.8.1 A team approach

All establishments visited held the view that a ‘team’ approach is the best way to ensure consistency and effectiveness in the management of adverse effects. It was considered essential that the team involved technicians and scientists on equal and complementary terms, together with the **NACWO** and **NVS**. A constant message across all types of establishment was that scientists’ competence and attitude could vary widely, so **named persons** and technicians saw a very important part of their role as providing a consistency of care and attention. The study director was also a vital team member in a CRO, as it was necessary to agree on humane endpoints and this could involve some negotiation between clients, regulators and CRO staff.

This ideal seemed to be achieved less convincingly in some academic establishments as opposed to industry ones, although it should be stressed that those institutions with problems in this area were the minority. Within academic establishments, animal technicians are not always recognised as part of a research ‘team’, but are sometimes

regarded as the people who look after the animals for the scientists. This view was increasingly and justifiably seen as an outmoded one given the current level of technician training, expertise and involvement in projects, but was still encountered. People working within the pharmaceutical establishments and CROs visited during the present study were more likely to view technicians as *full* team members (to the extent of including them as co-authors in published papers) and as a valuable resource.

Most scientists in academia were happy for the technicians to be responsible for routinely monitoring animals as they recognised their own limitations in this respect and felt both legally and ethically 'covered'. One project licence holder especially liked the fact that technicians were looking at his animals 'blind' in the sense that they did not always know how much time the researcher has put into the experiment or how many expensive reagents had been used. He believed that they therefore made decisions based solely on the animals' well-being. The idea that technicians have different priorities and objectives from scientists was expressed in all types of establishment.

However, there was occasionally frustration and resentment where scientists believed that the level of monitoring and intervention by technicians and named persons represented unacceptable interference with their work. In extreme cases there was a perception that there were 'good' technicians and 'bad' scientists, such that some technicians referred to themselves as "carers" and the scientists as "users", as though the two were mutually exclusive. Within some academic establishments, technicians also felt that researchers had a very different idea of what constituted pain and distress from them, did not like being questioned and were overly lenient when assessing animals. In these cases, it could be difficult for non-senior technicians to insist that steps were taken to alleviate adverse effects.

All animal technicians insisted that they preferred to retain primary responsibility for animal monitoring and care, but many also wished that scientists "took more of an interest" and would inspect their animals more often and learn more about them (i.e. their behaviour and ecology). Most believed that the scientists they worked with had a good attitude to the animals and acceptable levels of knowledge, although several technicians expressed the view that technicians empathised more with animals than scientists, who (they believed) regarded animals as "tools".

Animal technicians at every level (junior and senior) knew which steps to take if they believed that an animal was suffering excessively and most that believed the NVS would support any decision that they made. This support was perceived as very important in ensuring that technician status and morale remained high. While several scientists from a range of establishment types recalled that technicians had euthanased animals where it had not been warranted in their opinion, most ultimately respected the technicians' judgement.

6.8.2 Ultimate responsibility for assessing animals

Under the **A(SP)A**, the **personal licence** holder is primarily responsible for detecting adverse effects and acting upon them appropriately. Most respondents named animal technicians (whether or not they were the personal licence holder) or a combination of technicians and scientists as being routinely responsible for monitoring adverse effects in practice. The overwhelming consensus among all types of establishment was that animal technicians were most competent at detecting adverse effects early because they had the most experience and knew animals best, both at a species and sometimes an individual level. The importance of recognising a ‘normal’ animal was frequently stressed, and it was widely felt that technicians were most familiar with the animals on a daily basis and therefore best at this.

Two establishments differed, both of which carried out fundamental research. At these, it was felt that the skills of the technicians, scientists and veterinarians were all complementary to one another and their expertise was of equal value. A typical comment was that technicians are most observant, scientists are more aware of the predicted experimental outcome (and therefore the likely adverse effects) and vets have the broadest range of experience. At one of these two establishments, it was made clear to everyone that the culture was such that everyone was involved in monitoring animals and it was not expected to be left wholly to technicians.

6.8.3 The role of animal technicians

It was widely perceived among interviewees that animal technicians’ responsibilities had increased and that they had become more involved in carrying out procedures, and so their status had become elevated and scientists respected their skills and judgement more. This had happened largely since the **A(SP)A** was enacted in 1986, although scientists at one academic establishment believed that it also occurred within the career of individual technicians. The same researchers also believed that the greater involvement of technicians in protocols and procedures had led to significant welfare improvements. Good technician status was viewed as essential with respect to ensuring that appropriate interventions took place when animals were suffering, because animal technicians were frequently the first to detect adverse effects and so their expertise and judgement had to be respected.

Some establishments had programmes in place designed to improve the integration of technicians and scientists into the ‘team’ on an equal footing. For example, a university initiated a programme of lunch-time talks by scientists to enable them to explain their research to technicians in more depth. One CRO also trained technicians, study directors and **project licence** holders to communicate with one another so that they could articulate any welfare concerns more effectively.

6.8.4 Training to recognise pain, suffering and distress

The quality of the training that scientists, technicians and veterinarians receive in recognising pain, suffering and distress has a direct bearing on the welfare of the animals whom they will subsequently be responsible for monitoring. Good training is essential in helping to foster the motivation, attitude and knowledge that are needed to assess animals effectively, and so this section reviews how the recognition and

monitoring of pain, suffering or distress is taught. There are two aspects to training in the UK; (i) there is a mandatory requirement for those conducting procedures licensed by the **A(SP)A** to undergo formal training (Home Office 2000), and (ii) establishments have their own in-house training processes.

Accredited training courses

All new personal and project licensees working under the UK **A(SP)A** must successfully complete training programmes accredited by either of two independent bodies, the Universities' Group for the Accreditation of Training (UGAT) or the Institute of Biology (IoB). The training programmes consist of five modules that cover legal, ethical and practical aspects of animal research and testing. The **Home Office** requirements for each module are set out in Table 7 (p. 49), together with the IoB's stated aims (the UGAT's aims are broadly similar).

The minimum syllabus laid out by the Home Office requires that recognition and management of pain, suffering and distress are covered in elements of modules 2, 3 and 4; they may also be included in parts of module 5 (Home Office 2000). The number of modules taken depends on the trainee's role within an establishment. For example, project licence applicants must complete at least modules 1, 2 and 5; personal licence applicants must complete 1 to 3 (and 4 if appropriate, e.g. if they propose to perform major surgery). The present survey covered modules 1 to 3 because these were conducted in-house more frequently than modules 4 and 5.

Training courses for modules 1 to 3 were conducted in-house at 14 establishments (7 academic, 3 pharmaceutical and 4 CROs). Otherwise, trainees were sent to other establishments or to courses run by an external training company. Within those establishments that ran their own in-house training courses, the content, training aids and time allocated to recognising pain, suffering and distress were very consistent (Table 8, pp. 50-51), due to the narrow subject area and also lack of suitable training material. A major problem engendered by the paucity of training aids was that trainees were taught about recognising 'normal' animals, but then had to go on to identify suffering animals in practice. It is difficult to obtain material depicting animals in pain or distress for both ethical and practical reasons. It could be unethical to prolong suffering so that an animal could be photographed or filmed; justification for doing so would need to be assessed on a case-by-case basis depending on a number of factors. These include the individual animals' likely level of suffering, how long it would take to obtain the required material and how much pain or distress the training material would directly or indirectly prevent. Also, people are reluctant to produce and release such material. For these reasons, good quality in-house training is especially important when teaching people to recognise suffering (see below).

The general consensus among named persons and licensees who have worked under the **A(SP)A** both before and after modular training courses were introduced was that the courses have had a significant, positive impact on licensees' awareness, competence and attitude with respect to monitoring animals. This change has occurred in conjunction with underlying improvements in attitudes to and concern for animals in general (i.e. within society as a whole) and increased recognition of the status and expertise of animal technicians. It was also felt that scientists' views on what it is acceptable to do to animals were constantly changing, e.g. tumours were once permitted to attain sizes that would not be acceptable today.

It is extremely important to maintain this momentum by regularly reviewing training syllabuses, materials and courses. The RSPCA is hoping to develop UK initiatives in these areas in association with the accreditation bodies, individual trainers, LASA, the APC Education and Training Sub-committee and other relevant bodies.

In-house training

In addition to the Home Office accredited modular training courses, all scientists and technicians who were involved in carrying out procedures or monitoring animals were trained to recognise pain, suffering and distress in-house. This was always done by the trainee ‘shadowing’ an experienced colleague (usually an animal technician) who ran through the assessment procedure with them, encouraged them to make their own judgements, then discussed how they had arrived at their decisions. Trainers judged when new licensees or animal carers were sufficiently competent and empathetic to assess animals on their own. The widely-recognised abilities of technicians to detect adverse effects first was thus ascribed to a combination of both experience and tuition.

All establishments saw the in-house training period as a chance to help establish their own culture, provide training that was better tailored to each person’s projects where possible and assess trainees’ abilities to empathise with animals (and their attitude to animals in general). Training people to handle animals was viewed as especially important with respect to assessing attitude. Almost all establishments had official or unofficial procedures in place to cater for trainees who proved to be incapable of empathising effectively with animals or had an unsatisfactory attitude towards them. Depending on the intended eventual role of the trainee and the depth of the problem, these included retraining, assigning them to basic husbandry duties only or assigning them to duties that did not involve any contact with animals.

6.8.5 Areas where more training is required

Most respondents were satisfied with the training in recognising pain, suffering or distress that they had received, valuing their in-house training in particular. Any concerns that people had about training standards or content were focused on perceived gaps in the knowledge possessed by other groups. For example, many technicians and **named persons** felt that scientists’ knowledge of signs of pain, suffering and distress and also animal behaviour and basic requirements was deficient (see section 6.8.1 above). This was not, in general, regarded as an immediate problem provided that animals received adequate care and supervision from others.

Concern was also expressed by senior technicians and **NACWOs** at two academic institutions that technicians did not always know that they had the “law on their side” or how much power they possessed. Many participants believed that technicians should ideally be able to address issues such as whether an experiment should be terminated from an animal welfare and from a legal aspect. The ability to do this *and* the knowledge that they would be supported by named persons (especially the veterinarians) were regarded as essential requirements for truly empowered and effective technicians.

6.9 Problems in practice

Although most people felt confident that pain, suffering and distress were being detected effectively within their establishments, the majority also believed that recognising ‘normal’ animals and suffering at any level was neither easy nor quick to learn. An ability to empathise with animals and a positive attitude towards them were both essential prerequisites, but sufficient experience and the *time to monitor animals properly* were also needed. Some factors were regarded as difficult to overcome even then, such as the inherent difficulty in interpreting animal behaviour, especially in rodents. The practical problems most frequently raised by respondents are set out in Sections 6.9.1 to 7 below.

6.9.1 Animals concealing discomfort, pain and distress

The fundamental problem encountered when devising or using any assessment system is that many animals do not readily exhibit clinical signs, and this is extremely hard to overcome. All respondents stressed that it could be very difficult to tell when animals were suffering – indeed, to predict how they felt at all. In general, the wellbeing of ‘prey’ species including rabbits, rodents, horses and sheep was regarded as much more difficult to assess than that of ‘companion’ (predator) species such as dogs and cats. In rabbits and rodents, this was not only due to the instinctive concealment of evidence of physical stress or injury (rodents were sometimes described as “tough”, “resilient” or having a “high pain threshold”), but also to the comparatively larger numbers in which they were used and kept. People at establishments using sheep, ponies and horses also believed that herd/prey animals *appeared* to be stoical and did not always show when they were suffering. Those using sheep felt confident that they knew their animals as individuals and could pick up adverse effects, but an establishment using horses was less confident and stressed the importance of noticing small changes in behaviour, which came with experience.

Many people reported that after undergoing procedures that one would expect to be painful, animals did not show recognisable (behavioural) signs of pain and this made them concerned that it was not possible to tell how animals are feeling. Conversely, people found that procedures that they would not have expected to be painful to humans appeared to cause some other species pain. These observations were very commonly made and were generally used to validate the assumption that animals should be given the ‘benefit of the doubt’. That is, most people assumed that something that would be painful to humans will also hurt animals, but retained an open mind and were prepared to accept that animals may find some procedures more painful than humans would.

Furthermore, as one senior technician pointed out; the definition of a ‘mild procedure’ is ours, not the animals’. Many people cited types of study where they believed it likely that animals were suffering before clinical signs became apparent, such as mice on *Salmonella* vaccine studies. Despite these statements, almost everyone interviewed believed that adverse effects were being recognised and alleviated effectively within their establishments.

6.9.2 Detecting incipient pain and distress

Many respondents emphasised that it could be very difficult to assess the gradual onset of discomfort, pain or distress, for example due to tumour growth or the toxic effects of a substance. This was regarded as very different from predictable and acute adverse effects such as post-surgical pain. People frequently described a continuum from stress to distress to discomfort to pain; whereas severe pain could easily be diagnosed (e.g. by audible vocalisation in rats), moderate pain was much more difficult. Some establishments weighed animals regularly in an attempt to use body mass as an indicator of incipient pain or distress, but even this objective measure could not be guaranteed to reflect the presence of pain.

These observations are borne out by the clinical signs listed in Table 3 (p. 45) – many of these are indicators of rather more substantial adverse effects than mild or moderate pain, suffering or distress (FELASA 1994, Jones et al. 1999). When asked to describe the appearance of a sick rodent, most people described an animal who was hunched in the corner of the cage, with a ‘staring’ coat. It was frequently recognised that a rodent behaving in this way was likely to be feeling very sick or in pain, however, so that s/he was no longer able to suppress pain-coping behaviour. The recognition of discomfort and incipient pain was thus seen as very important, as effectively managing mild pain could potentially prevent more severe suffering. Also, in accordance with the principle that animals should be given the benefit of the doubt and treated with the same concern that humans would expect, many people believed that mild pain should be taken seriously and alleviated in its own right.

6.9.3 Human subjectivity

There was almost universal agreement that a well-trained and empathetic animal technician was the most effective means of detecting discomfort. Facilities that used score sheet or data management systems made it very clear that these were used as adjuncts to technician judgement. However, inconsistency between observers was also frequently cited as an inevitable problem.

Most establishments attempted to keep teams of technicians monitoring each cohort of animals constant wherever possible. There were some exceptions; the toxicology department of one pharmaceutical establishment assigned pairs of technicians to monitor for adverse effects (so that there was always a second opinion) and regularly changed the pairs for greater cross over of ideas and communication. Another toxicology department appointed separate ‘room technicians’ and ‘study technicians’, changing the study technicians every 6 months to obtain different views.

Temporary technicians were employed in significant and increasing numbers at some establishments and this was sometimes regarded as a concern; one scientist at an academic institution felt that ‘temps’ did not always know what to look for. Another veterinarian at an academic establishment valued the temporary workforce but felt that it was more difficult for ‘temps’ to become assimilated into the establishment’s ‘culture of care’. He believed that it would become more difficult to employ permanent technicians because technicians are increasingly made to feel like social outcasts, such that it is not regarded as a desirable career.

There was an extremely widespread belief that a good animal technician would always know when an animal was suffering and be able to detect an animal in distress very quickly. Although this belief is strongly held by technicians, **named persons** and scientists, there does not appear to be any data to substantiate this. Statements were frequently made to the effect that experienced technicians could tell by eye or feel whether animals had lost weight, or tell a sick animal “at a glance”. One **NACWO** strongly believed that a technician could pick up pain, suffering or distress before it showed up in any “measurable” way – this type of statement is rarely challenged or evaluated. For example, a study on subjective and objective measures of pain in dogs in a clinical situation found poor correlation between them, suggesting that clinicians should not rely too heavily on subjective measures such as Visual Analogue Scores when deciding on pain management protocols (Conzemius et al. 1997).

6.9.4 Human habituation

While the importance of knowing a ‘normal’ animal was frequently stressed to the author, some respondents made the point that ‘normal’ can be what is normally seen. Where this was recognised, human habituation was reduced by a variety of measures including regular review of projects, periodically retraining staff, initiating cross-overs of staff (above), exchanging ideas with respect to monitoring and refinements of husbandry and procedures, and by listening to any concerns that new technicians might have. Establishments that used score sheets for recording observations and reviewed them regularly found that this helped to prevent this type of habituation and acceptance of clinical signs as ‘normal’.

6.9.5 Different consideration for different species

Some technicians found it easier to relate to and empathise with larger animals kept for longer periods, such as dogs and primates, than small species kept for shorter periods and where individuals had fewer easily distinguishable features (cf. LASA 1990). This was reflected by some differences in monitoring and alleviating adverse effects. For example:

- one CRO used a lexicon of clinical signs for rodents and free text for larger animals, because there were fewer large animals and technicians regarded them more as individuals and were more aware of their characters
- another CRO made a strong case to clients for rewarding primates with bananas but did not make an equivalent case for mice
- an academic establishment gave rats analgesia before embryo transfer but not mice, on the grounds that analgesia made no difference to the behaviour of the mice (the mice were not weighed to evaluate this objectively).

6.9.6 Time constraints

The effectiveness of any system for monitoring and assessing animals was said to be ultimately dependent on the number of animals and the time available to assess them. If there was only time for a cursory observation, it was not possible to evaluate clinical signs properly and incipient adverse effects might not be detected. This was unfortunately seen as a possibility in a busy animal unit and is a particular problem for establishments using genetically modified animals (see below). Technicians under

pressure tended to prioritise the monitoring of animals on procedures, which meant that stock animals might be temporarily less effectively monitored. This could be a welfare issue; for example, one respondent explained that 0.01 % of BALB/c mice suffer from splenomegaly such that humane killing may be necessary even if an animal has never been subjected to a procedure. Many establishments experienced a chronic shortage of permanent technicians and had to use staff from agencies, which could worsen problems relating to time and resources available for animal welfare. For example, one NVS felt that score sheets would be an ideal way of monitoring animals, but in the absence of sufficient staff his establishment had to rely on “common sense and clinical observations”.

6.9.7 Genetically modified animals

The creation and use of genetically modified (GM) animals presents additional problems for effective monitoring of pain, suffering and distress, mainly because of the large numbers of animals involved and also because of the potential for unexpected adverse phenotypes. An academic facility that used large numbers of GM animals found that most (over 90 %) appeared clinically normal, but some abnormal phenotypes did occur. In this event, judgement was made by the **project licence holder, Home Office Inspector, named persons and ERP** on whether the project had exceeded its severity limit and whether re-breeding from the same founders would be ethical. At this facility, each technician was responsible for checking around 500 cages of 4 to 5 mice every day. If 6 hours a day is spent monitoring animals, this allowed some 43 seconds at most to check each cage or 9 - 11 seconds for each mouse. Objective measures such as body mass were rarely taken at this establishment due to lack of time.

Another academic facility using large numbers of GM animals carried out a welfare assessment programme for all neonatal mice, in which they were compared with wild-type litter mates (see Appendix 3 for score sheet), and carried out SHIRPA tests for all mice used in its mutagenesis programme. Adult animals were monitored using clinical observation sheets, however, and were not routinely weighed.

NB In the report of its consultation on emerging biotechnologies and the **A(SP)A**, the UK **APC** noted that there was little information on how to assess the welfare of GM animals due to a lack of published information. Example approaches and URLs for downloading welfare assessment sheets have been published by van der Meer et al. (1999, 2001) and Mertens & Rühlicke (1999). The APC report recommended that “the APC, possibly with others, should consider the commissioning of a project to examine how to assess the welfare of transgenic animals, especially mice” (APC Biotechnology Sub-group, 2001). Welfare assessment of GM animals is also included in a BVA(AWF)/FRAME/RSPCA/UFAW report on applying the principles of reduction and refinement to the generation, management and care of GM rodents (Robinson et al. in press).

6.10 Miscellaneous good ideas/practices

The following are examples of initiatives set up by establishments that have directly or indirectly helped to prevent or alleviate pain, suffering or distress. Note that all of these initiatives are in addition to the requirements of the **A(SP)A** and **Home Office Inspectorate**.

- Awarding welfare prizes to anyone who furthers the implementation of the Three Rs in a novel and practical way.
- Providing module 1 to 3 training for all undergraduates (the theory component contributed towards their final degree results).
- Videoing unexpected adverse effects – without causing additional suffering – (i) to show the **ERP**, (ii) for technician training, (iii) to show to clients.
- Ensuring that all ERP members visit the animal house and undertake appropriate training courses.
- Training technicians and scientists to communicate effectively with one another.
- Inviting licensees to give lunchtime talks about their work so that technicians and everyone else understands what they do, to help improve technician status and confidence.
- Regularly reappraising and training all staff; conducting ‘Verification of Competencies’ every 2 years.
- Creating a ‘blip book’ to log unpredicted adverse effects in genetically modified animals (see Section 6.3).
- Establishing surgical teams to undertake routine surgery, e.g. fitting jugular catheters or EMG electrodes.
- Contracting an external training consultant to address a serious ‘them and us’ situation between technicians and scientists. The consultant found that technicians felt undervalued and not respected, and resented the lack of post-study communication. This was resolved by designating a ‘study contact’ technician to enable better communication between the study director, **NACWO**, **NVS** and client, and to make decisions on euthanasing animals.
- Instigating a ‘workplace audit programme’, where 2 independent auditors continually observe procedures of their own choosing and check welfare, reporting to the **Certificate Holder**.
- Holding regular and frequent health and welfare meetings for everyone to discuss projects and any concerns.

7 Summary of results

The key points from Section 6 are summarised below.

7.1 Awareness of the potential for suffering

1. There is broad recognition that procedures may cause animals discomfort, pain, suffering or distress; 97 % of respondents assume that this would occur to some extent during or as a result of the procedures that they carry out.
2. Many respondents also include other potential causes of emotional stress or distress that need to be taken account of and minimised, such as handling and the administration of substances.

7.2 Recognising, assessing and monitoring pain, suffering and distress

1. The clinical signs used as indicators of potential pain, suffering or distress are largely subjective (Table 3, p. 45).
2. Major problems with the detection of adverse effects in practice are: animals concealing discomfort, pain and distress; problems with detecting incipient suffering; human subjectivity and habituation; and time constraints.
3. Animal technicians are almost universally believed by all types of respondent to be best at detecting signs of suffering or distress.
4. A range of different techniques is used to assist the recognition and monitoring of adverse effects (Table 4, p. 46). The most commonly used are clinical observation sheets (28 establishments; 100 %), 'score sheets' (9; 32 %) and data management systems (7; 25 %).
5. Less frequently employed techniques include the use of specific clinical signs (at 4 establishments), e.g. in one university the ability of mice with enzyme deficiency diseases to cross a bar without falling is used to infer wellbeing as well as to assess disease progression. Other methods include a 'Blip book' to record unpredicted phenotypes in transgenic animals; video stills, SHIRPA, and ultrasonic vocalisation by rats made audible using a bat detector (all used at 1 establishment respectively). Visual Analogue Scales are not used by any establishments visited during the survey. A wider variety of methods is employed within academia than industry (Table 4, p. 46).
6. There is a good background awareness of 'score sheets', i.e. many people have heard of them, but they are not widely used.
7. Respondents agree that a 'team approach' is the best way to ensure consistency and effectiveness in managing adverse effects, and the team should include technicians and scientists on equal and complementary terms, together with **named persons**. This is generally (but not always) said to be successfully achieved.

7.3 Preventing, alleviating and controlling adverse effects

1. Respondents state that they avoid and alleviate adverse effects by the use of analgesia, humane endpoints, refining the administration of substances, ensuring competent handling, refining husbandry and providing environmental stimulation, restricting permitted surgery times and habituating and training animals.
2. All 25 establishments that conduct recovery surgery administer analgesia postoperatively. Twelve of these administer analgesia pre-emptively at least some of the time.
3. Six of the 25 establishments have written policies on the provision of analgesia (Table 5, p. 47). These vary in prescriptiveness, especially with respect to the duration of postoperative analgesia.
4. Rodents are given anaesthesia but not pain relief for some surgical procedures (headpiece fitting in rats and embryo transfer in mice) at 4 establishments.

7.4 Reviewing the management of pain, suffering and distress

1. All establishments review projects regularly, but in most cases there is no formal review of adverse effects and observations are not compared with the predictions made in **project licence** applications. This is largely due to a lack of time and resources.
2. Most respondents believe that the **ERP** does or should play an important role in setting out how adverse effects should be monitored and managed. Most also feel that the ERP has actively improved the implementation of the Three Rs, especially refinement.

7.5 Training issues

1. Mandatory modular training courses were introduced in the UK in 1994-5. Some establishments organise their own training courses in-house; others place trainees on courses run by commercial companies or by other establishments.
2. The content, training materials and time allocated to recognising pain, suffering and distress are very consistent between establishments with their own training courses (Table 8, pp. 50-51). More training resources are needed.
3. The mandatory training courses have had a significant, positive impact on licensees' awareness, competence and attitude with respect to monitoring animals and alleviating adverse effects.
4. More training is especially required in: the legal requirements for pain relief and humane endpoints (so that everyone is aware that technicians are empowered to implement the law); and, largely for scientists, animals' basic biology, ecology and behaviour including species and procedure-specific signs of pain, suffering and distress.

7.6 Communicating good practice

1. The most commonly used media for disseminating good practice in monitoring and alleviating adverse effects between establishments are presentations at external meetings, informal conversations, published papers, industrial discussion groups and visits to other establishments (Table 6a, p. 48).
2. The most frequently used media within establishments are 'hands on' training in the form of guidance and supervision in working situations (in addition to the modular training courses required by the **A(SP)A**), verbal instructions, mainly *via* veterinarians, and the ERP and associated committees (Table 6b, p. 48).

8 Conclusions and recommendations

The survey shows that people are very concerned about the potential for animal suffering and want to be able to detect and alleviate it effectively. Unfortunately, the principal difficulties outlined by Morton and Griffiths (1985) are still as problematic as ever – there are few if any specific behavioural indicators of pain, suffering or distress and the systems currently in use for assessing animals are heavily reliant on subjective criteria (Morton & Griffiths 1985, Flecknell 1994, Lloyd et al. 2000). Although these problems are likely to remain for some considerable time, those involved with animal care and use are aware of the importance of reducing suffering and the existence of new techniques such as binary score sheets (e.g. Morton 1997, 1998a, 1998b, Morton & Townsend 1995). There appears to be, however, poor

motivation to use them, which respondents to the present survey largely attributed to a lack of resources (i.e. time) to do so.

The recommendations listed in Sections 8.1 to 8.7 below aim to help progress the development and implementation of better ways of assessing and monitoring animals, and to contribute towards more effective management of pain, suffering and distress. They include (i) many basic statements of principle relating to each issue that frequently arose and were reinforced by the respondents to the survey, and (ii) other recommendations that will help to progress the implementation of these principles.

8.1 Awareness of the potential for suffering

Acknowledging that animals experience suffering is a fundamental requirement if suffering is to be recognised and minimised. In general, those who used and/or cared for laboratory animals were concerned about the potential for suffering and very aware of their moral and legal responsibility to prevent or reduce it. A potential unfortunate consequence of the level of concern for animals was that people did not want to think of them suffering and wanted to be convinced that all adverse effects were reliably detected and alleviated. This could result in methods for detecting suffering not being questioned and evaluated as stringently as they ought to be.

There was a divide between scientists and technicians at some establishments, where each group felt that the other's attitude was inappropriate. There was also an element of sizeism or speciesism in practice, as rodents sometimes received less consideration than larger animals (see Section 6.9.5). Participants in general felt that everyone involved with research animal use (e.g. scientists, animal technicians and veterinarians) should:

1. be aware that animals can experience suffering and take this seriously.
2. be aware that the experiments and procedures *that they are involved with* can cause suffering.
3. recognise that interpreting animal behaviour can be very difficult and that peoples' observations can be subjective.
4. have and retain an open mind about the subjective mental states of *all* animals and how their behaviour ought to be interpreted.
5. disseminate as widely as possible the message that *subjective impressions are not necessarily correct*, beginning during training for scientists and technicians and continuing to reinforce it regularly in practice.
6. work within a legal framework which requires that suffering is minimised, but regard national legislation as a point of reference that sets out minimum requirements only.
7. regard the effective management of pain, suffering and distress as a continually evolving discipline that requires ongoing development and continuous retrospective review.

8.2 Recognising, assessing and monitoring pain, suffering and distress

The results of the present study are consistent with those obtained by Lloyd et al. (2000) in that assessment of animals is largely subjective and observations are frequently not recorded comprehensively. Also in accordance with Lloyd et al. (2000), those who do use score sheets regularly find them to be valuable. The present survey has found that few establishments are using score sheets, although their uptake is increasing and many people have heard of them. Reluctance to use score sheets is generally due to a lack of time to implement them and a lack of awareness that score sheets can be continually adapted, tailored to projects and binary rather than numerical. Many respondents feel that increased use of score sheets may be a useful tool for improving objectivity and consistency in many situations, but there appears to be little motivation to change monitoring systems at an establishment level – this is likely to be a critical limiting factor.

Reliance on subjective methods for assessing animal wellbeing is of concern, because current research indicates that this is not always justified (Roughan & Flecknell 2001). It is an article of faith that people who are very familiar with animals (usually technicians) can detect a suffering animal very quickly, but this may not be the case (cf Conzemius et al. 1997). For example, establishments evaluating candidate drugs for arthritis routinely assessed animals for gait disturbance by eye. Objective forms of gait analysis (Clarke et al. 1997, Clarke & Still 1999, 2001), however, are more sensitive and can detect levels of limb favouring earlier and/or with milder versions of arthritic changes that are not apparent to the naked eye (K Clarke, pers. comm). Similarly, comprehensive video analysis of rat behaviour postlaparotomy has identified behaviours that are not commonly used as criteria for assessing postoperative pain and has also suggested that animals may be masking pain-related behaviours (Roughan & Flecknell 2001).

The practical application of such objective techniques is unfortunately currently limited because they are often highly specific, require specialised equipment to develop or employ and may also require time and resources that are not available in most facilities. Objective techniques can help to have an immediate positive impact on welfare, however, if they are used to demonstrate that entirely subjective impressions of animal wellbeing are not always reliable.

In the absence of techniques that could feasibly be used to assess animals objectively in practical situations, binary score sheets appear to be the most effective way of assessing animals and recording observations (Richmond 1999, Lloyd et al. 2000, Morton 2000, Morton et al. 2000). With appropriate revision and critical application, score sheets can yield consistent results and help to refine endpoints (Lloyd & Wolfensohn 1999), although it should be noted that in some studies score sheets are less diagnostic and levels of between-observer variation can reduce their usefulness (Beynen et al. 1987, 1988).

While it can be difficult to recognise subtle behavioural signs of discomfort, pain or distress, making observations at the wrong time of day can result in missing them altogether. Pain-associated behaviours in nocturnal animals such as rats and mice may not occur during the day or evening (e.g. Wallace et al. 1990, ILAR 1992) but

very few establishments monitor animals throughout the night when such behaviours are far more likely to be apparent. Techniques that can assist with night-time monitoring include automated open fields with infrared monitors (van't Land & Hendriksen 1995) or video (J Kelly pers. comm.) but these are not frequently used.

Scientists, animal technicians and veterinarians need to:

1. be able to recognise 'normal' animals, at a species, strain and ideally individual level.
2. be familiar with clinical signs that indicate suffering in general and specifically to each project, species and (where possible) individual.
3. make observations at appropriate times for each species.
4. always raise any concerns they may have about any animal or protocol; ensure that they and everyone else have the confidence to do so.
5. be open to the use of a broad range of techniques for assessing and monitoring animals, and always be prepared to try new methods.
6. use binary score sheets more widely to help assess animals and to record observations more effectively; regularly review score sheets (i) to refine protocols and training for animal users and/or carers, and (ii) to assist more accurate predictions of adverse effects at the project planning stage.
7. approach all techniques for assessing animals critically and evaluate their efficacy in practice (see Section 8.4 below).
8. comprehensively predict potential adverse effects using input from a broad range of resources.
9. work as part of an integrated team to monitor animals effectively and make decisions quickly should problems arise.
10. make sure that the status of animal technicians is high and that everyone respects them as a valued resource – they are frequently the first to detect changes in animal behaviour including signs of suffering, so their expertise and judgement must be respected (see Section 6.8.3).

8.3 Preventing, alleviating and controlling adverse effects

People take the need to treat pain, suffering and distress seriously and many establishments have clear policies on postoperative pain relief. These policies vary within and between establishments, however, particularly with respect to the duration of postoperative pain relief and the provision of analgesia for different species. The latter appears to be due to 'speciesism' or a perception that analgesics are not safe to use in rodents. It is interesting to note by way of comparison that a survey of UK veterinarians in practice found the administration of analgesia perioperatively in companion animals to vary widely (Capner et al. 1999, Lascelles et al. 1999). For example, 52 % of male and 36 % of female veterinarians did not provide pain relief for ovariohysterectomy in dogs (Capner et al. 1999), and 74 % of veterinarians did not administer analgesia for ovaiohysterectomy in cats (Lascelles et al. 1999). The survey of veterinarians in practice also found that analgesics were seldom given to small mammals undergoing surgery, and the authors suggested that this was due to the difficulty in recognising pain in small animals and lack of knowledge of suitable drugs (Lascelles et al. 1999). Although the former problem is universal, awareness of and expertise in providing appropriate analgesics ought to be less of a problem for

those using and caring for laboratory animals, not least because all analgesic agents are tested on animals.

All those involved with animal use should:

1. ensure that everyone responsible for using and monitoring animals is empathetic, competent and confident, both in conducting procedures and monitoring animals before, during and afterwards.
2. administer analgesia to all species whenever necessary.
3. commit themselves to continually researching the literature for new analgesic agents and administration protocols.
4. take on and welcome the responsibility of continually researching ways of improving their implementation of the Three Rs, including humane endpoints and husbandry refinements.
5. think about procedures from the point of view of each individual animal – is this particular animal suitable for this protocol? How could s/he be trained or habituated? Would it be possible to maintain group-housing and provide environmental stimulation?

8.4 Reviewing the management of pain, suffering and distress

All establishments have regular meetings to review protocols and endpoints, but little is done to correlate predicted adverse effects with actual clinical observations. Where score sheets are used, they are generally referred to reactively, in the event of unexpected adverse effects, rather than proactively to assess whether there are chronic problems that are not being detected.

The lack of review to compare observed and predicted adverse effects means that the ability accurately to predict discomfort, pain and distress is not routinely evaluated. This has consequences for the decision making process when research proposals are assessed by the **Home Office Inspectorate** and **ERP**. The UK **project licence** application process requires the potential welfare ‘costs’ to animals to be set out in full for each project so that they can be considered alongside the possible benefits (Home Office 2000). Costs (pain, suffering, distress and lasting harm) must be predicted reliably if the ethical judgement of the decision-makers is to be sound. Regular review of observation sheets would help to ensure that those responsible for setting out costs and considering how they could be minimised could do so more accurately.

Ideally, establishments should:

1. have a clear, establishment-wide policy on the administration of analgesia pre-emptively and postoperatively as appropriate.
2. ensure that the relevant animal care and use committees receive new information on pain management (see Appendix 1) so that they can regularly reassess policies on the provision of pain relief for all species at all establishments. Within the present survey, analgesia policies and practice vary considerably between establishments. This variation indicates that policies and practice ought to be reviewed at every establishment.

3. regularly review each research project, using records of observed clinical signs to assess: how effectively adverse effects were predicted; whether observation or 'score' sheets need to be updated; and whether the severity of a given procedure has increased or decreased during the course of longer term projects.
4. ensure (in the UK) that the ERP plays a role in managing pain and other adverse effects at the project planning stage. In other countries, ensure that the relevant local committees do the same.
5. review policies and practice on the provision of environmental stimulation, to ensure that optimal environments are provided for recovery from procedures and wellbeing in general. This is important because environmental stimulation is recognised to play an important role in helping to shift animals' attention from any discomfort, pain or distress that procedures may have caused them (e.g. Gentle 2001).

Also;

6. it is unacceptable that some people do not appear to have enough time to monitor animals objectively, e.g. by weighing or using score sheets. In such instances, the reasons should be evaluated and a management strategy set up to address the problem. For example, consideration should be given to addressing staffing levels, allocation of tasks, time management, established priorities and support.

8.5 Training issues

To interpret animal behaviour, people primarily need to be able to empathise effectively with the animals in their care. Appropriate training is also required in information retrieval (and input), technical skills and empathetic skills if animal users and/or carers are to understand animal behaviour to the best of their knowledge and ability. Training programmes for new technicians, scientists and laboratory animal veterinarians are prime opportunities to motivate people, challenge their assumptions and provide them with the expertise to find the resources they need to reduce suffering and disseminate good practice. The results of the present survey indicate that training should include:

1. establishment of a good 'culture of care' to ensure that animal users and/or carers have an appropriate attitude towards, and understanding of, animals.
2. comprehensive training in pain recognition and alleviation directly relevant to the projects that each trainee will be carrying out.
3. demonstration (e.g. video) of one or more objective means of measuring pain (see Section 8.2 above), to show that it may not be as easy to detect animal suffering as some trainees may think. This should be tailored to licensees' projects wherever possible.
4. a thorough understanding of the implementation of relevant sections of the national law regulating animal experimentation, so that everyone is aware of how to ensure that they are familiar with **severity limits** and what to do if they are approached or breached.
5. biology and behaviour of the study species (Hau 1999), where trainees are also made aware that this behaviour is likely to be innate in laboratory animals, although they may not have the opportunity to express it (Berdoy in Hawkins et al. 2001).
6. training in retrieving information on animal assessment, including new developments, and knowing how to feed this into licensees' own projects and

ensure that it is passed through the proper channels within her/his establishment (e.g. the **ERP** or IACUC) (Jennings 1994, Mench 1999).

7. time allowed for discussion so that trainees can explore how they feel about potentially causing animal suffering or caring for suffering animals and compare approaches.

8.6 Communicating good practice

Good practice in pain management and in many other areas of animal care and use is frequently not passed on. New methods for preventing, monitoring and reducing suffering are of great importance to everyone and so they need to be proactively disseminated in every possible way. For example, talks and posters at meetings should include more information on the means used to alleviate adverse effects, working groups on husbandry and protocol refinements should disseminate their findings more widely and actively and materials and methods sections of published papers should include more detail on refinement.

There is a perception that journal editors will discourage what they regard as excessive detail on refinement (but see Sumner-Smith 1998, Cowell 1998) but, if true, this can and should be challenged (Morton 1992, Smith et al. 1997) and authors should at least outline the techniques used so that others conducting similar research can contact them for further details. It may be unrealistic to expect journals to reproduce observation sheets and full monitoring protocols within every paper, but journals should use their websites to provide more detailed information about animal monitoring and other aspects of refinement. Journal websites represent an ideal opportunity to promote refinement, for example by including relevant URLs in published papers (e.g. the present report), but few currently do so.

Everyone responsible for reporting animal use in any forum should:

1. include all techniques used to reduce pain, suffering and distress, such as analgesia, refinement of techniques and husbandry, monitoring techniques etc. – these are essential elements of experimental protocols that ought to be included in all presentations and the methods sections of mainstream life science journals. If this is genuinely not possible, published papers should include URLs of journal websites, where information on refinement can be set out in full.
2. make a strong case for retaining such detail on scientific and ethical grounds if asked to remove it.
3. encourage others to do the same.

8.7 Areas where research is needed

Many of the recommendations in Section 8.2 above would be greatly facilitated if more knowledge and techniques were available to help the objective recognition and assessment of states of wellbeing in animals. The requirement for a small number of easily assessed indices for assessing wellbeing has not been fulfilled (Flecknell 1994, see Roughan & Flecknell 2001), yet this is especially important for recognising incipient suffering (i.e. discomfort and mild to moderate pain, sickness or distress) (Wallace et al. 1990, van't Land & Hendriksen 1995). Even clinical score sheets tailored to specific projects, which appear to be the best current option for objective

monitoring, can be unreliable; new approaches need to be explored. The areas of research outlined below therefore need to receive adequate funding to make good progress *and* their findings need to be implemented once they have been developed and validated.

1. New approaches and indices for assessing wellbeing should be researched; in particular, (i) studies aiming to define behaviours that are indicative of discomfort, pain or distress and that occur frequently and are easy to assess and (ii) research into the development of new automated techniques for recognising behavioural changes. Where it would be necessary for animals to experience discomfort, pain or distress to evaluate new techniques, studies should be done in conjunction with projects licensed for other purposes. Note that the aim should *not* be to fully automate animal monitoring; it should be to assist and augment sound clinical judgement.
2. Research into objective measures of GM animal welfare and into the type of variables that need to be assessed should be undertaken as a matter of urgency. While all of the conclusions and recommendations in the present report apply equally to GM and wild-type animals, GM animals can be regarded as a special case in that there are a number of other parameters that need to be assessed to evaluate whether developmental or phenotypic abnormalities are present that could compromise welfare (e.g. van der Meer et al. 1999, 2001). This requires comprehensive monitoring that can take a relatively long time; for example testing animals after weaning can take 15 to 20 minutes per litter (van der Meer et al. 2001), yet the number of GM animals used in the UK is increasing exponentially (Home Office 2001) and this is likely to be the case elsewhere.
3. Further research should also be conducted to evaluate physiological correlates of suffering that may provide useful adjuncts to other assessment systems, for example where heart rate or body temperature are available from telemetry transmitters that have been implanted for experimental purposes. It is recognised that objective measures such as heart rate and body temperature and levels of humoral factors such as cortisol are of limited use for predicting pain or distress alone (Conzemius et al. 1997, Dobromylskyj et al. 2000). For ethical reasons, such research should be done in conjunction with studies initiated for other purposes that involve measurement of the appropriate parameters.

The lack of current objective techniques for assessing levels of animal discomfort, pain or distress mean that sweeping statements about levels of laboratory animal suffering should be avoided. Even with the (limited and largely subjective) tools currently available, most UK establishments do not have the time and resources to compare observed and predicted adverse effects and there is no available centrally collected information about levels of suffering experienced by animals in practice. These difficulties mean that it is currently just not possible to make any broad-ranging statements about suffering with any degree of confidence.

Some of the recommendations in the present report should be immediately achievable in all establishments and are already being carried out by many people. Others will require careful thought and planning in addition to significant time, commitment and other resources. It is essential that these are provided. The very high level of openness and willingness to participate in the present survey suggests that motivation

on the part of those directly responsible for assessing animal wellbeing is not the limiting factor. Everyone working with laboratory animals who wants to reduce the conflict between animal welfare and science should therefore receive the resources and financial and practical support that they, and the animals, deserve.

Acknowledgements Sincere thanks to everyone who gave up their time to speak to me so openly and honestly and, in many cases, show me their animal accommodation (sometimes including animals undergoing procedures). This survey would not have been possible if people had not been prepared to trust me and offer me so much information, and I hope that they all found it worthwhile. Thanks also to Maggy Jennings and Jane Smith for their very helpful comments on the organisation of this report. This project is dedicated to the memory of Sheila Silcock.

References

- Aldhous P, Coghlan A, Copley J (1999) Animal experiments: where do you draw the line? *New Scientist* **2187**, 26-31
- APC Biotechnology Sub-group (2001) *APC Report on Biotechnology*. London: APC
- Beynen AC, Baumans V, Bertens APMG, Haas JWM, Van Hellemond KK, Van Herck H, Peters MAW, Stafleu FR, Van Tintelen G (1988) Assessment of discomfort in rats with hepatomegaly. *Laboratory Animals* **22**, 320-325
- Beynen AC, Baumans V, Bertens APMG, Havenaar R, Hesp APM, van Zutphen LFM (1987) Assessment of discomfort in gallstone-bearing mice: a practical example of the problems encountered in an attempt to recognize discomfort in laboratory animals. *Laboratory Animals* **21**, 35-42
- Cambridge AJ, Tobias KM, Newberry RC, Sarkar DK (2000) Subjective and objective measurements of postoperative pain in cats. *Journal of the American Veterinary Association* **217**, 685-690
- Capner CA, Lascelles BDX, Waterman-Pearson AE (1999) Current British veterinary attitudes to perioperative analgesia for dogs. *Veterinary Record* **145**, 95-99
- Clarke KA, Heitmeyer SA, Smith AG, Taiwo YO (1997) Gait analysis in a rat model of osteoarthritis. *Physiology and Behaviour* **62**, 951-954
- Clarke KA, Still J (1999) Gait analysis in the mouse. *Physiology and Behaviour* **66**, 723-729
- Clarke KA, Still J (2001) Development and consistency of gait in the mouse. *Physiology and Behaviour* **73**, 159-164
- Conzemius MG, Hill CM, Sammarco JL, Perkowski SZ (1997) Correlation between subjective and objective measures used to determine severity of postoperative pain in dogs. *Journal of the American Veterinary Association* **210**, 1619-1622
- Cowell HR (1999) Editorial: management of postoperative pain in animals used in research. *Journal of Bone and Joint Surgery* **81-A**, 751
- Dobromylskyj P, Flecknell PA, Lascelles BD, Livingston A, Taylor P, Waterman-Pearson A (2000) Pain assessment. In: *Pain Management in Animals* (Flecknell PA, Waterman-Pearson A, eds). London: WB Saunders
- FELASA Working Group on Pain and Distress (1994) Pain and distress in laboratory rodents and lagomorphs. *Laboratory Animals* **28**, 97-112
- Flecknell PA (1994) Refinement of animal use – assessment and alleviation of pain and distress. *Laboratory Animals* **28**, 222-31
- Flecknell PA (1996) *Laboratory Animal Anaesthesia*, 2nd edn. London: Academic Press
- Gentle MJ (2001) Attentional shifts alter pain perception in the chicken. *Animal*

- Welfare* **10**, S187-194
- Hau J (1999) Humane endpoints and the importance of training. In: *Humane Endpoints in Animal Experiments for Biomedical Research* (CFM Hendriksen, DB Morton, eds). London: Royal Society of Medicine Press, pp 71-74
- Hawkins P, Berdoy M, Deacon R, Morrison P, Sandells B, Peters A, Lewis R, Harding K, Jones D, Jennings M, Hubrecht R (2001) Report of the 2000 RSPCA/UFAW Rodent Welfare Group Meeting. *Animal Technology* **52**, 29-42 and erratum *Animal Technology*, **53**
- Home Office (2000) *Guidance on the Operation of the Animals (Scientific Procedures) Act 1986*. London: The Stationery Office
- Home Office (2001) *Statistics of Scientific Procedures on Living Animals: Great Britain 2000*. London: The Stationery Office
- ILAR (1992) *Recognition and Alleviation of Pain and Distress in Laboratory Animals*. Washington, DC: National Academy Press
- Jennings M (1994) *Ethics Committees for Laboratory Animals: A Basis for their Composition and Function*. Horsham, UK: RSPCA
- van't Land CJ, Hendriksen CFM (1995) Change in locomotor pattern in mice: a model for recognition of distress? *Laboratory Animals* **29**, 286-293
- Lascelles BDX, Capner CA, Waterman-Pearson AE (1999) Current British veterinary attitudes to perioperative analgesia for cats and small mammals. *Veterinary Record* **145**, 601-4
- Lloyd MH, Wolfensohn SE (1999) Practical use of distress scoring systems in the application of humane endpoints. In: *Humane Endpoints in Animal Experiments for Biomedical Research* (CFM Hendriksen, DB Morton, eds). London: Royal Society of Medicine Press, pp 48-53
- Lloyd M, Wolfensohn S, Thornton P (2000) Quantitative assessment of welfare in experimental animals: the development and use of scoring systems. In: *Progress in the Reduction, Refinement and Replacement of Animal Experimentation* (Balls M, van Zeller A-M, Halder M, eds). Amsterdam: Elsevier, pp 1107-1117
- van der Meer M, Costa P, Baumans V, Olivier B, van Zutphen B (1999) Welfare assessment of transgenic animals: Behavioural responses and morphological development of newborn mice. *ATLA* **27 suppl 1**, 857-868
- van der Meer M, Rolls A, Baumans V, Olivier B, van Zutphen LFM (2001) Use of score sheets for welfare assessment of transgenic mice. *Laboratory Animals* **35**, 379-389
- Mench J (1999) Defining endpoints: the role of the animal care committee. In: *Humane Endpoints in Animal Experiments for Biomedical Research* (CFM Hendriksen, DB Morton, eds). London: Royal Society of Medicine Press, pp 133-138
- Mertens C, Rüllicke T (1999) Score sheets for the monitoring of transgenic mice. *Animal Welfare* **8**, 433-438
- Morton DB (1990) Adverse effects in animals and their relevance to refining scientific procedures. *ATLA* **18**, 29-39
- Morton DB (1992) A fair press for animals. *New Scientist* **1816**, 28-30
- Morton DB (1995) Recognition and assessment of adverse effects in animals. In: *Proceedings of Animals in Science Conference: Perspectives in their Use, Care and Welfare* (Johnston NE, ed). Clayton, Victoria: Monash University Research Ethics Unit, pp 131-148

- Morton DB (1997) A scheme for the recognition and assessment of adverse effects in animals. In: *Animal Alternatives, Welfare and Ethics* (van Zutphen LFM, Bamms M, eds). Amsterdam: Elsevier Science BV, pp 235-240
- Morton DB (1998a) The use of score sheets in the implementation of humane end points. In: *Proceedings of the Joint ANZCCART/ NAEAC Conference on Ethical Approaches to Animal-based Science* (Mellor D, Fisher M, Sutherland G, eds). Adelaide, Australia and Wellington, New Zealand: ANZCCART, pp 75-82
- Morton DB (1998b) Implementing assessment techniques for pain management and humane endpoints. In: *Proceedings for 'Pain Management and Humane Endpoints'*:
<http://altweb.jhsph.edu/meetings/pain/morton.htm>
- 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* (CFM Hendriksen, DB Morton, eds). London: Royal Society of Medicine Press, pp 5-12
- Morton DB (2000) A systematic approach for establishing humane endpoints. *ILAR Journal* **41**, 80-86
- Morton DB, Ambrose A, Leach MC, Kelly J, Poirier G (2000) Adverse effects – recognition and assessment, and humane endpoints. In: *Progress in the Reduction, Refinement and Replacement of Animal Experimentation* (Balls M, van Zeller A-M, Halder M, eds). Amsterdam: Elsevier, pp 1083-1093
- 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**, 431-436
- Morton DB, Townsend P (1995) Dealing with adverse effects and suffering during animal research. In: *Laboratory Animals – an Introduction for Experimenters*, 2nd edn (Tuffery AA, ed.). Chichester, UK: John Wiley & Sons Ltd.
- National Research Council (1996) *Guide for the Care and Use of Laboratory Animals*. Oxford: National Academy Press
- Plous S (1999) Opinion research on animal experimentation: Areas of support and concern. In: *Proceedings for 'Pain Management and Humane Endpoints'*:
<http://altweb.jhsph.edu/meetings/pain/plous.htm>
- Robinson V, Morton DB, Anderson D, Carver R, Franics RJ, Hubrecht R, Jenkins E, Mathers KE, Raymond R, Rosewell I, Wallace J, Wells DJ (in press) Applying the principles of reduction and refinement to the generation, management and care of genetically modified mice. *Laboratory Animals*
- Richmond J (1999) Criteria for humane endpoints. In: *Humane Endpoints in Animal Experiments for Biomedical Research* (Hendriksen CFM, Morton DB, eds). London: Royal Society of Medicine Press, pp 26-32
- Roughan JV, Flecknell PA (2001) Behavioural effects of laparotomy and analgesic effects of ketoprofen and carprofen in rats. *Pain* **90**, 65-74
- Scharmann W (1999) Physiological and ethological aspects of the assessment of pain, distress and suffering. In: *Humane Endpoints in Animal Experiments for Biomedical Research* (Hendriksen CFM, Morton DB, eds). London: Royal Society of Medicine Press, pp 33-39
- Slingsby LS, Waterman-Pearson AE (2000) Postoperative analgesia in the cat after ovariohysterectomy by use of carprofen, ketoprofen, meloxicam or tolfenamic acid. *Journal of Small Animal Practice* **41**, 447-450
- Slingsby LS, Waterman-Pearson AE (2001) Analgesic effects in dogs of carpreofen and pethidine together compared with the effects of either drug alone. *Veterinary*

- Record* **148**, 441-444
- Smith JA, Birke L, Sadler D (1997) Reporting animal use in scientific papers. *Laboratory Animals* **31**, 312-317
- Sumner-Smith G (1998) Editorial. *Veterinary and Comparative Orthopaedic Traumatology* **11**, 5
- Wallace J, Sanford J, Smith MW, Spencer KV (1990) The assessment and control of the severity of scientific procedures on laboratory animals. *Laboratory Animals* **24**, 97-130
- Wolfensohn S, Lloyd M (1998) *Handbook of Laboratory Animal Management and Welfare*, 2nd edn. Oxford: Blackwell Science

Table 1 Types of designated user establishment visited when conducting the survey

Type of establishment	Number visited	Number with Certificate of Designation in force on 31.12.2000 ^a	Percentage of procedures conducted by establishment type in 2000 ^a
University/medical school	7	90	39
Fundamental research/government	7	63 ^b	23
Pharmaceutical company	9	105 ^c	38 ^c
Contract research organisation	5		
Total	28	258	

- a Home Office (2001). A Certificate of Designation must be granted by the UK Home Office before any establishment is permitted to use, breed or supply research animals.
- b These were 9 Government establishments, 31 non-departmental public bodies (NDPBs), 13 non-profit making organisations, 7 National Health Service hospitals and 3 public health laboratories. An NDPB is a national or regional public body that operates independently of Government Ministers, although Ministers are ultimately responsible for it, e.g. the Medical Research Council (MRC) and the Biotechnology and Biological Sciences Research Council (BBSRC).
- c Pharmaceutical companies and CROs are listed together as ‘commercial organisations’ in the Home Office Statistics.

Table 2 Categories of interviewee

Role in establishment	Type of establishment			Total
	Academic	Pharmaceutical	CRO	
Scientist	28	18	7	53
Senior technician	9	5	9	23
Technician	4	3	2	9
NACWO	16	7	3	26
NVS/Deputy NVS	11	6	3	20
Veterinary nurse	2	1		3
Study director		1	1	2
Home Office liaison officer		1		1
Total	70	42	25	137

Table 3 Clinical signs cited by interviewees as indicators of potential pain, suffering or distress

Clinical sign	Examples cited by interviewees
Objective signs	Body mass; food consumption; water consumption; body temperature (telemetered or measured with thermometer); experimental variables (e.g. telemetered heart rate, EEG, analgesimeter readings).
Behaviour	Normal and provoked behaviour; degree of interaction with conspecifics; irritation at injection sites; vocalisation; grinding teeth (especially sheep); chattering teeth (badgers); writhing; tremors; lethargy (especially primates); unusually aggressive behaviour (e.g. in Sprague Dawley rats); 'not bouncy' (dogs); 'wary', staying at back of pen or cage.
Discharges	Nasal discharge; salivation (NB this may occur in anticipation of oral dosing); porphyrin staining (rats).
Movement	Locomotion (staggering, laboured gait, ataxia); movement impeded by tumours.
Physical signs	Altered respiration rate; brightness/dullness of eyes; ulceration of tumours; estimated body temperature (whether 'cool to touch', pale/red extremities); signs of wound infection.
Posture	Hunching; differences in resting posture (to find comfortable position); head down (sheep).
Skin/coat	Piloerection ('starey' coat); not grooming/stained coat; hair loss; colour of skin; 'saggy' skin (dehydration); whether grooming normally.
Other	'General appearance'; changes in food and water consumption; presence of pain on moving; 'just not right'.

Table 4a Techniques *routinely* used for recognising and recording adverse effects

Assessment method	Number of establishments using method			Total
	Academic	Pharmaceutical	CRO	
Clinical observations	14	9	5	28
Score sheets	4	4	1	9
Data management systems		2	5	7
Specific clinical signs	2	2		4
'Blip' book	1			1
Video stills	1			1
SHIRPA	1			1
Ultrasonic vocalisations		1		1

Table 4b Numbers of people citing different techniques for recognising and recording adverse effects

Assessment method	Number of people citing method			Total
	Academic	Pharmaceutical	CRO	
Clinical observations	62	22	23	107
Score sheets	19	17	4	40
Data management systems		3	13	16
Specific clinical signs	4	3		7
'Blip' book	1			1
Video stills	3			3
SHIRPA	1			1
Health	2			2
Behaviour		4	1	5
Ultrasonic vocalisations		1		1

Table 5 Policies on the prevention and alleviation of postoperative pain

	Number of establishments^a
Separate written policy/standard operating procedure stipulating analgesia	6
Postoperative analgesia must be included in Section 19b of licence application	10
Postoperative analgesia routinely given ^b	25
Pre-emptive analgesia routinely given	4
Vets must be consulted for each procedure	2
No policy or specifications ^c	5
Not applicable (no recovery surgery)	3

- a This table refers only to the 25 establishments that conduct recovery surgery.
- b There are some exceptions to this within establishments (see *Analgesia* in section 6.4).
- c NB this does NOT mean that analgesia is not administered to animals undergoing invasive surgery.

Table 6a Methods that individuals use to communicate good practice with respect to monitoring and alleviating adverse effects outside their own establishments

Method	Number of people citing
Presentations at external meetings, e.g. LASA, IAT, LAVA ^a	26
Verbally/informally	16
Within published papers	15
Industrial discussion groups/visits to other establishments	10
‘Open’ policy for all external visitors	6
IAT/LASA publications	5
Courses run at establishment	5
Internet, e.g. VOLE, Compmed ^b	4
Specialist user groups	3
LASA refinement meetings	3
BVA(AWF)/FRAME/RSPCA/UFAW Joint Working Group on Refinement ^c	2
RSPCA/UFAW Rodent and Rabbit Groups ^c	2
Input into European Union Guidelines on maximum dose volumes	1

- a Laboratory Animal Science Association (<http://www.lasa.co.uk/>), Institute of Animal Technology (<http://www.iat.org.uk/>), Laboratory Animal Veterinary Association (<http://www.blava.org.uk/>).
- b Vets On Line Email (<http://www.blava.org.uk/vole.htm>), Comparative Medicine Discussion List (<http://www.aalas.org/association/links/compmed.htm>).
- c Further details in Appendix 1.

Table 6b Methods of communicating good practice with respect to monitoring and alleviating adverse effects within establishments

Method	Number of establishments
‘Hands on’ training	27
Verbally (mainly via vets)	21
The ERP & associated committees	12
Internal discussion groups	6
NACWO/team leader meetings	5
Internal training courses	5
Written protocol sheets	4
Between-site meetings	3
Internal networks for named persons	2

Table 7 Relevant sections of UK modular training courses

Module	Elements of module covering recognition and alleviation of pain, suffering and distress.
2	1. Recognition of wellbeing: signs of pain, suffering and distress in relevant species. ‘The programme shall promote ... the ability to recognise animals that are unwell or distressed.’
3	2. Common diseases and recognition in the relevant species. 4. Introduction to anaesthesia and analgesia in the relevant species. ‘The programme shall promote ... knowledge of the principles of good anaesthetic practice and the provision of adequate analgesia; an appreciation of how those principles are applied to eliminate or minimise discomfort or distress in procedures requiring anaesthesia with or without recovery.’
4	1. Surgical anaesthesia and analgesia 2. Post-surgical care and monitoring. ‘The programme shall promote ... knowledge of the principles and techniques available for postoperative care of animals including the use of analgesia.’
5	1. Ethical aspects of the use of live animals 3. Alternatives (Refinement, Reduction, Replacement) 5. Project licence management (responsibilities, record keeping). ‘The programme shall promote ... a thorough appreciation of the ethical debate relating to the use of animals and an awareness of the legal obligations to minimise discomfort or distress ...’

Table 8 In-house modular training courses – modules 1 to 3

	Time allocated to recognising adverse effects (min)	Training aids (in addition to lectures)	Topics covered in course	Tailored to each trainee's project?
Academic 1	90 to 105	Discussion, LAVA slides ^a	Recognising and assessing pain, suffering and distress (due to procedures and housing); why this is important; pain alleviation and scoring systems	Where possible
Academic 2	60	Discussion, LAVA slides, sample score sheets	Recognising and assessing pain, suffering and distress; ethical, scientific and legal aspects; score sheets; types of analgesic; establishment's policy on pain relief	Where possible
Academic 3	60	Discussion, looking at stock animals, LAVA slides	Recognition and alleviation of pain; legal issues; why assessing and alleviating is important; analgesics – when to administer and pharmacokinetics	Where possible
Academic 4	60	LAVA slides, looking at animals, discussion and individual talks	Recognition of pain; score sheets and tailoring them to procedures; analgesia	No
Academic 5	1 day ^b	LAVA slides, handout from commercial breeder, looking at stock animals, IAT videos ^c	Recognition of pain and distress	Where possible
Academic 6	2 days ^b	Stock animals, LAVA slides, IAT videos, ABPI ^d interactive video	Range of normality – adverse effects at this establishment were highly varied and unpredictable	Slightly
Academic 7	30	LAVA slides, stock animals	Recognising distress; causes of stress; moral responsibility to minimise suffering	No
Pharm 1	60 + ½ day practical	LAVA slides, in-house slides, ABPI CD-ROM, discussion	Recognition and alleviation of pain; perioperative care	No
Pharm 2	60	LAVA slides, interactive discussion, healthy animals, videos ^e	Pain and its alleviation; disease recognition; analgesia; welfare; empathising with animals	Yes
Pharm 3	1 ½ days ^b	Animals – stock and during dosing by experienced technicians, FELASA publication ^f	Recognising adverse effects; why this is important; what a 'normal' animal looks like	Yes
CRO1	120	In-house handouts, LAVA slides, video	Definition and recognition of pain and distress; preventing and	Yes

			alleviating	
CRO2	60	LAVA slides, looking at animals	Recognising adverse effects	Yes
External trainer	180	Discussion, LAVA slides, own slides, own handbook	Recognising 'normal', stressed and sick animals, prevention and alleviation of pain	Where possible

- a A slide set produced by LAVA which includes some pictures of animals experiencing adverse effects.
- b People at these establishments felt that training in recognising pain, suffering and distress ran throughout their Module 1 to 3 courses so that they could not quote a discrete time.
- c *Handle with care* and *Procedures with care*.
- d Association of the British Pharmaceutical Industries
- e Two videos were used to train those working with primates; (i) *Paradise lost*, a television documentary on the wild-caught primate trade and an undercover exposé of a primate supply establishment showing poor handling and attitudes towards the animals and (ii) *Benevolent primate husbandry* by the Wisconsin Regional Primate Center.
- f FELASA Working Group on Pain and Distress (1994).

Appendix 1 Recommended reading

Background

American Veterinary Medical Association (1987) AVMA Colloquium on Recognition and Alleviation of Animal Pain and Distress. *Journal of the American Veterinary Association* **191**, 1184-1296

Moberg GP, Mench JA (eds) (2000) *The Biology of Animal Stress: Basic Principles and Implications for Animal Welfare*. Wallingford, UK: CAB International

Rutherford KMD (2002) Assessing pain in animals. *Animal Welfare* **11**, 31-53

Recognition and assessment

Baker RM, Jenkin G, Mellor DJ (eds) (1994) *Improving the Well-being of Animals in the Research Environment*. Glen Osmond: Australian and New Zealand Council for the Care of Animals in Research and Teaching

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

ILAR Committee on Regulatory Issues in Animal Care and Use (2000) *Definition of Pain and Distress and Reporting Requirements for Laboratory Animals*. Washington, DC: National Academy Press (read online at <http://www.nap.edu/books/0309072913/html/>).

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

Wallace J et al. (1990) The assessment and control of the severity of scientific procedures on laboratory animals. *Laboratory Animals* **24**, 97-130

Pain management

Flecknell PA (1996) *Laboratory Animal Anaesthesia*, 2nd edn. London: Academic Press

Flecknell PA, Waterman-Pearson A (eds) (2000) *Pain Management in Animals*. London: WB Saunders

Hellebrekers LJ (ed) (2001) *Animal Pain: A Practice-oriented Approach to Effective Pain Control in Animals*. Oxford: Blackwell Science

Kohn DF et al. (1997) *Anaesthesia and Analgesia in Laboratory Animals*. New York: Academic Press

National Health and Medical Research Council Animal Welfare Committee (1994) *Ways of Minimising Pain and Distress in Animals in Research*. Canberra: AWC - NHMRC

Refining procedures

Laboratory Animals Ltd. online reprints – see <http://www.lal.org.uk/laband.htm>

Laule G (1999) Training laboratory animals. In: *The UFAW Handbook on the Care and Management of Laboratory Animals*, 7th edn (Poole T, English P, eds). Oxford: Blackwell Science, pp 21-27

Morton DB et al. (2001) Refining procedures for the administration of substances. *Laboratory Animals* **35**, 1-41

Morton DB et al. (1993) Removal of blood from laboratory mammals and birds. *Laboratory Animals* **27**, 1-22

Refining husbandry and handling

Animal Welfare Information Center (1995) *Environmental Enrichment Resources for Laboratory Animals 1965-1995*. Maryland: AWIC

Animal Welfare Institute (2002) *Database on Refinement of Housing and Handling Conditions and Environmental Enrichment for Laboratory Animals: Rodents, Rabbits, Cats, Dogs, Ferrets, Farm Animals, Horses, Birds, Fishes, Amphibians and Reptiles*. http://www.awionline.org/lab_animals/biblio/refine.htm

BVA(AWF) (2001) *Practical Animal Handling 1: Small Mammals*. <http://www.lal.org.uk/>

Hawkins P et al. (2001) Laboratory birds: refinements in husbandry and procedures. *Laboratory Animals* **35 Suppl. 1**, 1-163

IAT (1986) *Handle with care*. Oxford: IAT (Video which is being updated at the time of writing.) <http://www.iat.org.uk/>

Jennings M et al. (1998) Refining rodent husbandry: the mouse. *Laboratory Animals* **32**, 233-259

Morton DB et al. (1993b) Refinements in rabbit husbandry. *Laboratory Animals* **27**, 301-329

Reinhardt V, Reinhardt A (eds) (2002) *Comfortable Quarters for Laboratory Animals*, 9th edn. Washington, DC: Animal Welfare Institute <http://www.awionline.org/pubs/cq02/cqindex.html>

Reinhardt V, Reinhardt A (2001) *Environmental Enrichment for Caged Rhesus Macaques*, 2nd edn. Washington, DC: Animal Welfare Institute

Reinhardt V, Reinhardt A (2002) *Environmental Enrichment for Primates – Annotated Database*. http://www.awionline.org/lab_animals/biblio/enrich.htm

RSPCA/UFAW Rodent Welfare Group Reports (contact RSPCA Research Animals Department)

Humane endpoints

Hendriksen CFM, Morton DB (eds) (1999) *Humane Endpoints in Animal Experiments for Biomedical Research*. London: Royal Society of Medicine Press (download at <http://www.lal.org/labam.htm>).

ILAR (2000) Humane Endpoints for Animals Used in Biomedical Research and Testing. *ILAR Journal* **41**, 59-123

OECD Environment Directorate (2000) *Guidance Document on the Recognition, Assessment, and Use of Clinical Signs as Humane Endpoints for Experimental Animals Used in Safety Evaluation*. Paris: OECD

UKCCCR (1997) *UKCCCR Guidelines for the Welfare of Animals in Experimental Neoplasia*, 2nd edn. London: United Kingdom Co-ordinating Committee on Cancer Research

Training material

Digital material for trainers produced at the University of Newcastle upon Tyne, UK includes 35mm slide CDs, e.g. (i) handling, husbandry and minor procedures, (ii) animal health and welfare images, (iii) anaesthesia; and video CDs, e.g. (i) pain assessment in the rat, (ii) practical animal handling – small mammals and (iii) small mammal anaesthesia. For further information and ordering see <http://www.lal.org.uk/digital.htm>

Some online resources

Altweb Pain Management Database

Database with information about anaesthesia and analgesia for most commonly used laboratory animals and some exotic species, with information about available drugs and their side effects.

http://www.altwebsearch.org/aadb/aadb_search.cfm

Boschert K (1999) Pain-related Internet Resources for IACUCs. In: *Proceedings for 'Pain Management and Humane Endpoints'* (below).

<http://altweb.jhsph.edu/meetings/pain/boschert.htm>

Center for Management of Animal Pain (C-MAP) at the University of Tennessee College of Veterinary Medicine.

<http://www.vet.utk.edu/cmapi/>

Crawford RL (2000) *A Reference Source for the Recognition and Alleviation of Pain and Distress in Animals*. Beltsville: Animal Welfare Information Center

<http://www.nal.usda.gov/awic/pubs/awic200003.htm>

Humane Society of the United States *Pain and Distress Initiative*

<http://www.hsus.org/ace/11400>

including *Pain and Distress Reports* (digests of recent reports and publications)

<http://www.hsus.org/ace/11401>

Kuiper JD, Allen T (1999) *Compilation of Literature on the Assessment of Animal Welfare and Animal Distress*.

<http://www.vetinfo.demon.nl/aw/index.html>

Proceedings for 'Pain Management and Humane Endpoints' (1999) A workshop of the Johns Hopkins Center for Alternatives to Animal Testing, National Institutes of Health Office for Protection from Research Risks, NIH Office for Animal Care and Use and National Academy of Sciences Institute for Laboratory Animal Research.

<http://altweb.jhsph.edu/meetings/pain/proceedings.htm>

Appendix 2
Clinical observation sheet from a pharmaceutical company

Pre/Post Surgery Observation Sheet
for Small Animals

Animal Identification	
-----------------------	--

Project licence:		Start date:	
Procedure:		Location:	
Severity band:		Species/strain/sex:	
Licensee:		Cage number:	
Protocol number:		Observation frequency:	Daily
Stock/experimental batch:			

Any animal found dead or those which are culled must be recorded in "RED"

Date:		Food		Residue:		Grams eaten:	
		Given:					
Bodyweight:		Water		Residue:		Volume used:	
		Given:					
Observations:							
				Balance GLP number.		Initials.	

Appendix 3
Examples of score sheets

i) Binary score sheet for Transmissible Spongiform Encephalopathy (TSE)

SCRAPIE

HAMSTER NO:		ISSUE No:					
MICROCHIP NO:							
DATE OF INNOCULATION:		PRE-INNOCULATION WEIGHT:					
DATE							
DAY							
TIME							
FROM A DISTANCE							
Inactive							
Not inquisitive or alert							
Isolated							
Hunched posture							
Starey coat							
Not nest building							
Not hoarding							
Not grooming							
Abnormal gait							
Persistent scratching							
Lethargy							
Ataxia							
Rigidity							
ON HANDLING							
Not eating							
Not drinking							
Bodyweight (g)							
% change from start							
Body temperature (°C)							
Discharge eyes/nose							
Coat wet/soiled							
Diarrhoea/faecal pellets +/-							
Wound OK? Open/infected							
Dehydration: skin pinch							
Vocalisation							
Other signs noted							
SIGNATURE:							

Special husbandry requirements:

Monitor animal daily for 1st week until wound healed.

Scoring details to be ascertained

Humane endpoints and actions

If wound infected contact NVS

Must inform user of any neurological signs eg. : rigidity, ataxia, lethargy, lameness, fits, change in behaviour.

Scientific measures: None

ii) Binary score sheet for a neurology study

STRUCTURE OF NEURONS

RABBIT No.	ISSUE No:						
DATE OF OPERATION:	PRE-OPERATION WEIGHT:						
DATE							
DAY							
TIME							
FROM A DISTANCE							
Inactive							
Isolated							
Hunched posture							
Not hopping normally							
Wobbly							
Leg affected: left, right, front, back							
Dragging leg/s: left, right, front, back							
Not grooming							
ON HANDLING							
Vocalisation/pain on handling							
Not inquisitive or alert							
Not eating							
Not drinking							
Faecal impaction							
Urine retained (bladder palpable)							
Stained perineum							
Bodyweight (g)							
% change from start							
Body temperature (°C)							
Pedal withdrawal reflex absent							
Stitches missing							
Other signs noted							
Nothing abnormal detected							
SIGNATURE:							

Special husbandry requirements:

Plenty of straw and hay in pen. Hand feeding may be required. Isolate in cage for 24 hrs post op.

Monitor feed intake.

Humane endpoints and actions

Animals showing signs of abnormal motor and sensory changes following recovery from anaesthesia will be closely monitored under supervision of the named veterinary surgeon or his deputy. If no improvement of the motor or sensory deficits occurs within 24 hrs the animals will be killed. Animals that are improving will be carefully monitored and if they exhibit weight loss (or failure to grow) of greater than 20% (compared with similarly operated cohorts), or show marked signs of distress, e.g. marked piloerection, dehydration, hunched appearance, subdued behaviour, solitary – for more than 1 day, will be killed. Abnormal motor or sensory effects are expected initially in some animals. These effects are not expected to be severe and are expected to be temporary and those affected should show definite improvement over 24 hours and be completely normal within five days. If any animal shows severe motor or sensory defects or pain on recovery then it is to be humanely killed.

Scientific measures: None

iii) Numerical score sheet for Inflammatory Bowel Disease

DISTRESS SCORING SHEET: INFLAMMATORY BOWEL DISEASE RATS

ANIMAL IDENTIFICATION	Score						
APPEARANCE							
Normal	0						
General lack of grooming	1						
Coat staring, ocular/nasal discharge	2						
Pinched features, ridge lines	4						
BODYWEIGHT							
Normal - $\square < 5\%$	0						
Body wt. 5-10%	1						
Body wt. 5-15%	2						
Body wt. 15-22%	4						
CLINICAL SIGNS							
Faeces normal - slightly soft	0						
Diarrhoea	1						
Soft distended gut + no faeces	2						
Hard and hot distended gut	4						
PROVOKED BEHAVIOUR							
Normal	0						
Minor depression or exaggerated	1						
Moderate change	2						
Reacts violently/vocalisation	4						
START BODY	MIN						
SCORE ADJUSTMENT							
If scored 4 more than once score 1							
TOTAL							

JUDGEMENT

0 – 4 Normal

5 – 9 Monitor carefully, consider analgesics.

10-14 Suffering, provide relief, observe regularly. Seek second opinion from NACWO and/or NVS. Consider termination.

15-20 Severe pain or distress; does this procedure need refining?

iv) Numerical score sheet for assessing neonatal genetically modified rodents

WELFARE ASSESSMENT SHEET FOR NEONATAL MICE AND RATS

Parameter (neonates)	Animal ID							
	Age							
Appearance/colour (see colour chart)*	Normal (pink)	0						
	Pink/blue abdomen	1						
	Pink/pale extremities	2						
	Blue/pale	3						
Surface temperature	Warm	0						
	?	1						
	Cold	2						
Natural activity	Wriggling ++	0						
	Wriggling +	1						
	+/-	2						
	Still	3						
Reflexes/respond to touch	+++ righting reflex +	0						
	++, righting reflex +/-	1						
	+	2						
	-	3						
Milk in stomach	++	0						
	+	1						
	-	2						
TOTAL SCORE (Neonate)	0-13							
Parameter (mother)	NB Include Milk Score							
Nest building	Good nest making	0						
	Some nest making	1						
	No nest	2						
Retrieval of young	Always	0						
	Sometimes	1						
	Never	2						
TOTAL SCORE (Mother)	0-6							

Judgement: Neonate

0-4 = Good

5-8 = Fair

9-12 = Poor

Always assess maternal factors as well. Maternal score 5-6 = Will these animals need fostering?

* The sheet is used in conjunction with a laminated 'colour chart' that provides guidance on the range of normal skin colours for neonates on days 0, 1, 2, 3 and 4 following birth.

Appendix 4 Questionnaire used to carry out the survey

Assessing and alleviating adverse effects in animals.

1. Is it routinely assumed that adverse effects are present in animals during or following procedures?
 - If yes, how and when are they alleviated?
 - If no, why would this not be appropriate?
2. If discomfort or pain is possible or likely but not assumed to be immediately present, how is this assessed?
 - Clinical observations?
 - Visual analogue scores?
 - ‘Score sheets’ (e.g. Morton & Griffiths 1985)? If yes, have existing scoring systems been modified, and how?
 - A combination of these/another method?
3. How does the implementation of the systems above vary with the cause and nature of the pain, for example post surgical pain, adverse effects in toxicological studies or chronic pain due to arthritis or cancer?
4. Who is responsible for routinely monitoring adverse effects?
 - Animal technicians?
 - Scientists using animals – personal or project licence holders?
 - Named persons - NVSs, NACWOs?
5. Does the establishment have a policy regarding the alleviation of pain, either by the use of analgesia or humane endpoints? E.g. must project licence applications say that analgesia will be administered or explain why this would not be necessary/appropriate?
6. How much time is allotted within the training courses used by the establishment for training licensees in the recognition of well-being and recognition and alleviation of adverse effects? Is the training tailored to each licensee’s own project(s)?
7. How are the policy and training requirements above reviewed within the establishment? How are retrospective assessments as to whether they are working effectively carried out?
8. Does/will the local ethical review process have a role in developing the policy on pain and other adverse effects? Have members been/will they be trained and expected to question analgesia protocols and endpoints when processing licence applications?
9. How are any effective recognition/record keeping systems or new analgesic regimes that may be developed at the establishment communicated to others, both within the establishment and outside it?

Glossary of relevant UK terminology

Animals (Scientific Procedures) Act 1986 (A(SP)A): Law that regulates research animal breeding, supply and use in the UK (see <http://www.homeoffice.gov.uk/animact/aspileaf.htm>).

The A(SP)A requires that any pain, distress or discomfort must be prevented or reduced to the minimum consistent with the purposes of the authorised procedures. It also includes an inviolable termination condition, i.e. a condition specifying circumstances in which an animal must in every case be immediately killed by an appropriate method.

Schedule 2A to the A(SP)A (transcribed from Article 8 of Council of Europe Directive No 86/609/EEC) requires all experiments to be carried out under general or local anaesthesia unless anaesthesia is judged to be more traumatic to the animal than the experiment itself or would be incompatible with the aims of the experiment. In such cases, appropriate measures must be taken to ensure that no such experiment is carried out unnecessarily and the law requires that anaesthesia “should be used in the case of severe injuries which may cause severe pain”. If anaesthesia is not possible, Schedule 2A requires that analgesics or other appropriate methods should be used to minimise pain, suffering, distress or lasting harm and prevent severe pain, distress or suffering (Home Office 2000).

Animal Procedures Committee (APC): An independent advisory body that advises the Home Secretary on matters relating to regulated procedures and examines other related subjects that it considers worthy of further study.

Ethical Review Process (ERP): Local process that must be set up at every designated establishment to review each project before the application is submitted to the Home Office. One of the key remits of the ERP is to ensure that the Three Rs of replacement, reduction and refinement have been implemented, in addition to addressing wider issues of animal care and use within each establishment.

Home Office Inspectorate: Body that administers the A(SP)A and advises the Secretary of State on whether to grant project licences for research programmes on the basis of a cost-benefit assessment (Home Office 2000). See also project licence, below.

Named persons: NACWO and NVS (below).

Named Animal Care and Welfare Officer (NACWO): Designated person, usually a senior animal technician, who is responsible for the welfare of animals on a day-to-day basis and should take an active role in the ERP.

Named Veterinary Surgeon (NVS): Designated attending veterinarian; should also take an active role in the ERP.

Personal licence: Licence allowing an individual to carry out identified regulated procedures on specified types of animals.

Procedure: A regulated procedure is any experimental or other scientific procedure

that may cause an animal⁷ pain, suffering, distress or lasting harm. This includes death, disease, injury, discomfort, and physiological and psychological stress.

Project licence: Licence specifying a programme of research involving regulated procedures. When deciding whether to grant a project licence for a programme of work, the Secretary of State must consider the welfare 'cost' to each animal (i.e. potential pain, suffering, distress or lasting harm caused to the animal throughout her/his lifetime) against the potential benefits that may accrue from a study (i.e. its potential contribution to its field and the likelihood that it will achieve its aims).

Severity limit: There are four degrees of severity that are used to classify regulated procedures in the UK; mild, moderate, substantial and unclassified. There are no rules for assessing severity, but the Home Office gives examples as follows: a small, superficial biopsy may be mild; a surgical procedure with appropriate anaesthesia and analgesia may be moderate; acute toxicity procedures with significant morbidity as an endpoint are likely to be substantial. Unclassified procedures are conducted under terminal anaesthesia. If it seems likely that the severity limit of a procedure may be exceeded, the A(SP)A requires that the procedure must be terminated or the Home Office Inspector informed (Home Office 2000).

⁷ Under the A(SP)A this includes all vertebrates and *Octopus vulgaris*. Procedures on mammals, birds and reptiles are regulated from half way through gestation or incubation periods; those on fish, amphibia and *O vulgaris* from the time at which they can feed independently.