Good practice for humane killing

Considering humane killing is part of ERP Function 5: “Considering the care and accommodation standards applied to all animals in the establishment, including breeding stock, and the humane killing of protected animals” [1,2]. The topic may come up within the ERP either:

- as a general issue for the establishment, for example with respect to the number of staff registered as competent to kill animals and the training provided for them, the potential for refinement of methods, and the provision of alternative options such as rehoming schemes; or
- as a specific issue during the review of individual projects when methods of humane killing or, in some cases, outcomes other than euthanasia may be discussed.

As an ERP lay member, you are not expected to be an expert on euthanasia. However, it is helpful to know the establishment policies on the issue, to be aware of the sort of points that can arise in discussion, and to understand the background to these. This resource therefore provides an introduction to the topic, briefly summarising the legal and scientific issues, and highlighting some of the questions and discussion points that commonly arise.

These notes are linked to ‘A resource book for lay members of Local Ethical Review Processes’ [1]. Their production is an integral part of the RSPCA’s Research Animals Department work on ERPs and refinement, and we hope that they will prove useful for these and other types of animal care and use committees.

**Legal requirements**

Methods of killing animals that are considered to be acceptable and humane, with the proviso that they are carried out by a competent person and death is subsequently confirmed, are set out in Schedule 1 to the Animals (Scientific Procedures) Act 1986 (ASPA) [3]. Any other method of killing is counted as a regulated procedure and has to be justified and authorised in a project licence. Schedule 1 was last revised in 1997, so it does not include information from subsequent research or methods that have been developed since that time*

There is a Home Office Code of Practice on Humane Killing of Animals linked to Schedule 1 of ASPA [6] which provides details of appropriate methods and explains how to ensure that they are carried out competently. The Code is not long, and is well worth reading as additional background information.

The responsibilities of staff under the ASPA are as follows:

- The **Certificate Holder** must maintain a register of staff competent to humanely kill animals and ensure that someone competent in these methods is always available.
- The **NACWO** must be knowledgeable about relevant methods of humane killing and either be competent in these or be able to contact someone from the register who is.
- The **NVS** must be knowledgeable about relevant methods of humane killing.

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* The Animal Procedures Committee (APC; the statutory body that advises the Secretary of State on the implementation of the ASPA) reviewed Schedule 1 in 2006 and made a number of recommendations, but this did not lead to any amendments to the text [4,5].
The role of the ERP:
The RSPCA/LASA Guiding Principles on Good Practice for Ethical Review Processes [2], developed by discussion with experienced ERP participants, suggests that the ERP’s role with respect to humane killing is to ensure there are systems in place for:
- the review of site procedures for Schedule 1 euthanasia at appropriate intervals, so that the most appropriate methods for each species are being used and refined as far as possible;
- incorporation of new knowledge about techniques such as the use of carbon dioxide;
- assessing the justification for and refinement of non Schedule 1 methods; and
- maintaining a Schedule 1 staff register with sufficient, well-trained staff, whose emotional wellbeing is supported.

Principles of humane killing
The ideal when humanely killing an animal should be to ensure a death without suffering, treating the animal with respect and minimising any anxiety, discomfort, pain or distress. This usually means inducing unconsciousness as rapidly as possible, although this is not always the case (see the discussion of carbon dioxide on page 3).

There is controversy over the relative humaneness of some methods of killing, so it is not always easy to decide which is the best to use. When making the decision, it is important to take the entire experience of the animals into account, so the issues to consider include:
- any need for capture, handling or restraint;
- any need for removal of the animal from the ‘home’ environment and/or mixing with unfamiliar animals;
- the practical application of the technique itself;
- confirmation of death.

Methods listed in Schedule 1
There are three broad categories of method in Schedule 1:
- Overdose of anaesthetic: Anaesthesia can be given by inhalation or by injection and is considered to be humane provided the inhalation anaesthetic used is not aversive* and the injected anaesthetic is not irritant. Different species and strains can differ widely in how aversive they find inhalational anaesthetics, so this has to be checked in each case. Note that once animals lose consciousness, some gaseous anaesthetics can then take a long time to finally kill them.
- Rising concentration of carbon dioxide (CO₂): This is a commonly used technique, but its humaneness is seriously questioned and there is ongoing debate about the best way of using it (see box: The problem with carbon dioxide).
- Physical methods - dislocation of the neck, concussion of the brain: These methods cause minimal stress if carried out competently by a handler with whom the animals are familiar; loss of consciousness can be rapid and there should be no pain. However, there is a serious risk of suffering if mistakes are made. In addition, some people find chemical methods more acceptable than physical techniques, which they find distressing to carry out [7]. This may affect their competence, which could consequently cause unnecessary suffering. The Home Office Code of Practice recognises this and says that “No person should be expected to kill an animal unless they are willing and feel confident to do so in the prescribed manner” [6].

Ungulates (hoofed animals such as cows, pigs, sheep and horses) may be killed using recognised slaughter methods such as captive bolt stunning followed by destruction of the brain or exsanguination.

* Aversive: noxious or unpleasant, so that the animal seeks to avoid it and becomes distressed if escape is not possible. Some anaesthetic agents can be highly aversive to some species and strains.
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Supplementary resources for members of local ethical review processes

The problem with carbon dioxide

Carbon dioxide has historically been accepted as a humane way of killing animals, but there is a growing body of research suggesting that it can be aversive or painful at certain concentrations [8,9]. In particular:

- If animals are placed into a chamber which has already been filled with a high concentration of CO\textsubscript{2} (above 50 %), they will experience 10 to 15 seconds of pain in the mucosa of the upper airways, corneas and mouth before they lose consciousness. This is a serious welfare problem, and this administration protocol is therefore not permitted under Schedule 1.
- If animals are placed into a chamber with a rising concentration of CO\textsubscript{2}, they will find it aversive at a certain level and may experience ‘air hunger’. This is highly distressing in humans and may also be a serious welfare problem in animals.

Current research suggests that filling the chamber with 100 % CO\textsubscript{2} at a flow rate of 20 % of the chamber volume per minute will produce a more gradual loss of consciousness without evidence of pain, but air hunger or aversion may still occur [9]. Most people believe this to be preferable to the experience of being placed into a high concentration of CO\textsubscript{2}. Some establishments have changed their CO\textsubscript{2} euthanasia protocols to the above concentration and flow rate on this basis, increasing the flow once animals have lost consciousness.

A possible alternative method has been suggested, where animals are anaesthetised first using a (non- or minimally-aversive) gaseous agent and then killed with CO\textsubscript{2}. This is currently not a Schedule 1 method, so permission for this must be granted by the Home Office in the project licence. Some project licence holders have done this.

In 2006, researchers working in this field concluded that there is no “ideal” way of killing animals using CO\textsubscript{2} [9]. Given that some gaseous anaesthetic agents (such as isoflurane) can also cause aversive responses that suggest significant distress, the choice between using CO\textsubscript{2} or such an agent is not a straightforward one. Research into the humaneness – or otherwise – of CO\textsubscript{2} is ongoing in 2011 and it is important for the establishment’s Named Persons or other relevant staff to keep up with scientific developments in the field.

Use of methods not listed in Schedule 1

There may be a scientific requirement in a project licence to use a technique that is not listed under Schedule 1. The reason for this should always be clearly explained and justified when the licence is being considered by the ERP, especially if the method could cause greater suffering than a Schedule 1 method.

When selecting the best method of killing, the first priority should be the welfare of the animals concerned. The scientific requirements must obviously be addressed, otherwise the animals may be wasted, but the harms and benefits of using a method that may be less humane from the animals’ point of view should always be weighed very critically. The feelings of the people (usually animal technologists and care staff) required to kill animals also need to be taken into account; they may find some methods difficult to accept.

Some non-Schedule 1 methods do not necessarily cause more suffering, but they may be controversial for other reasons. An example is decapitation for animals post-birth or hatch*. This

* Decapitation is unacceptable for birds, reptiles, amphibia and fish; Schedule 1 currently only permits it for embryonic mammals or birds up to 50 g [3].
technique is used where there is a scientific need not to disrupt brain biochemistry, or in the belief that it is a more humane method for neonatal rodents. It is controversial because many people find it aesthetically unpleasant, and because there are uncertainties over the length of time that consciousness persists in decapitated animals and whether they experience pain. The technique may also require longer handling and restraint times than other physical methods, which could be stressful. It can cause severe suffering if done badly, so if it is deemed necessary, it should only be carried out by well trained and experienced staff.

Refinement of methods
There is much ongoing research into humane methods of killing animals and it is important for the establishment to keep its techniques under review and to regularly check whether new or refined methods are available. Refinement of stages in the process, such as catching and handling animals, may be developed during other procedures and this should be applied to humane killing as well.

Some examples are:
- When whole cages of rodents have to be killed by gaseous methods such as CO\(_2\), it may be possible to leave them in their home cage and place this in the CO\(_2\) chamber. This avoids the need for handling and mixing of unfamiliar animals, allowing them to remain together in a familiar environment, thus reducing stress. It is probably best of all to kill animals in situ where this is possible, for example by administering a gaseous anaesthetic or CO\(_2\) via the air supply of animals housed in individually ventilated cages.
- An overdose of pentobarbital sodium (PBS) is commonly used to kill animals, but in the case of small rodents it is commonly injected intraperitoneally, where its high pH can cause irritation. This can be a problem if there is a lag, however small, between injection and loss of consciousness. An easy refinement is to dilute or buffer the PBS, or to add a local anaesthetic to the solution.
- Recent research has shown that catching and picking up mice by the tail induces aversion and high anxiety, whereas use of handing tunnels or scooping mice up on the open hand does not [10]. Refining catching and handling techniques will help to reduce the sum total of distress throughout the euthanasia process.

Staff issues
All establishments must keep a register of staff competent to humanely kill animals and ensure that someone competent in these methods is always available. The ERP will want to reassure itself that this is done and that appropriate training is provided.

It is also important to be aware of how staff feel about killing animals. Developing the ‘culture of care’ that should be integral to the philosophy of any establishment breeding or using animals can result in staff becoming very attached to the animals in their care and experiencing emotional distress when it is time to kill them [16]. It is important for establishments to address this issue and ensure their staff feel supported in this particular role.

Alternatives to killing animals
Most experimental animals are killed following procedures, either because their tissues are required as part of the experiment, or because their welfare might be compromised as a result of the procedures that have been carried out. However, this is not always the case and healthy animals may also be killed if they are surplus to requirements.

Many people believe that animals have intrinsic value, are sentient, capable of positive experiences and have an ‘interest’ in remaining alive (see references [11] and [12]). (The revised EU Directive 2010/63 regulating animal experiments specifically states this in its preliminary
recitals [12]). It could therefore be argued that killing animals who are not suffering, whether or not they have been used in a study, is a ‘harm’ and that alternative options should be explored. For example, it may be possible for some animals to be rehomed or released to the wild, a collection or a farm. These options require authorisation in the project licence and all of them have implications for animal welfare that need to be considered.

**Rehoming:** Some establishments have successful rehoming schemes for species including rodents, rabbits [13], dogs, horses and *Xenopus*, albeit in small numbers. Suitable candidates for rehoming include animals used in non-invasive or minimally invasive studies, surplus breeding stock, or animals acquired for a study which was subsequently cancelled. Rehoming may seem the ideal option but there are many factors that need to be considered to ensure that it is truly in the animals’ best interests. This is most effectively dealt with by the establishment having a well thought through, formal rehoming process. With this in mind, the UK Laboratory Animal Science Association (LASA) has produced guidance on rehoming laboratory dogs, which includes practical advice and useful principles that apply to other species as well [14].

**Release:** Release of animals into the wild is only likely to be appropriate for wild animals held in the short term, where it is considered that they have a good chance of being able to survive in the field and reoccupy their original niche. This is achievable with appropriate planning, and guidance is available from bodies such as the British Wildlife Rehabilitation Council [15]. If there is any doubt, rehoming to a well run wildlife collection may be an option.

**ERP member ‘to do’ list**

**Familiarise yourself** with the establishment’s policies and practices regarding euthanasia. You could either discuss this with the NVS or NACWO outside of the ERP meeting, or ask for a summary of establishment practice at a meeting. Examples of specific discussion topics you might like to raise are:

- The number of staff on the Schedule 1 register, and whether people think this is sufficient.
- The training they undergo, and how competence is monitored and maintained.
- How the establishment keeps up to date with finding out about and implementing refinement of humane killing methods, and other relevant refinements such as improved ways of handling animals.
- How the establishment ensures that animals are not wasted (*i.e.* killed just because they are surplus to requirements).
- Whether any animals are rehomed or released. If so, whether there is a formal rehoming or release policy and how this operates. If not, whether there is scope to begin a rehoming or release scheme.
- Whether the ERP could receive information on the fate of animals within individual studies, so that any opportunities to minimise wastage or better implement humane endpoints could be identified.

The establishment should be able to provide the ERP with all of the information needed to discuss these topics.
Guidelines on methods of killing:


Emotional support for staff:


Other background information:

- National Centre for the 3Rs information portal on euthanasia: [http://www.nc3rs.org.uk/euthanasia](http://www.nc3rs.org.uk/euthanasia)

… feedback please!

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