THOUGHT STARTER

Second Meeting of Expert Working Group (EWG) on Retrospective Severity Assessment

Brussels, 3 – 4 May 2012

Introduction

Directive 2010/63/EU on the protection of animals used for scientific purposes requires that a prospective assessment is made on the severity of each procedure in a Project (Article 15) and that a severity classification is assigned, either "non-recovery", "mild", "moderate" or "severe".

Article 54 on reporting requires that for statistical information, the actual severity of the pain, suffering, distress or lasting harm experienced by the animal must be taken into account (in contrast to the prediction on severity made at the time of the project evaluation).

These measures provide opportunities to improve the quality of science and welfare through prospective review of project proposals and, by inclusion of the actual suffering experienced by the animal, provide greater transparency and understanding on the impact on the welfare of animals used in scientific procedures.

In addition, the actual severity of any previous procedures will be a key consideration in determining whether or not an animal can be re-used in further procedures (Article 16).

Annex VIII provides guidance on the factors to be given in the consideration of prospective severity and provides some examples in each severity category.

The development of guidance on the assessment of the **actual** severity experienced by animals which have completed scientific procedures will be undertaken by an Expert Working Group (EWG). The first meeting of the EWG held in December 2011 considered the specific severity issues related to the production and maintenance of Genetically Altered (GA) animals.

The meeting in May 2012 will continue the development of guiding principles for the severity classification for animals used in scientific procedures.

We would like to thank the participants of the EWG for providing contributions ahead of the meeting. These were used to inform the content of this document.

Remit and objectives of the EWG

The objective of this meeting is to develop a common severity assessment framework that will be presented to the National Contact Points competent for this Directive for endorsement and published on the Commission web-site to facilitate consistent interpretation and application of Directive 2010/63/EU.

The EWG will

i. **Define and develop a retrospective severity assessment framework including factors which impact on the severity of procedures and how these are considered through different phases of the study to contribute to a consistent retrospective severity assessment.**
ii. **Develop some worked examples to illustrate how severity can be determined.**
iii. **Finalise the welfare assessment framework for GA animals which was initiated at the first meeting of the EWG in December 2011.**

Additional Background Material

In Europe, the Netherlands and Switzerland have had in place for a number of years a requirement to report actual severity.

In the Dutch system, there are six categories for reporting – minor, minor-moderate, moderate, moderate-severe, severe and very severe. Assignation is made by research workers with advice from the local Animal Welfare Officers.

In Switzerland, Severity classification of procedures was introduced in 1995. Two key papers were issued on **prospective** and **retrospective degrees of severity** (DS; respectively OVF 800.116-1-04 and OVF800.116-1.05;
see: http://www.bvet.admin.ch/themen/tierschutz/00777/00778/index.html?lang=fr).

These have been used to assist research workers, ethical review committees and the regulatory authorities to classify the severity of animals used in scientific procedures. These papers are currently under revision because of changes to the Swiss legislation.

Four categories are used in Switzerland to report retrospective severity: No stress: Severity Grade 0, Mild Stress: Severity Grade 1, Moderate Stress: Severity Grade 2, Severe stress: Severity Grade 3.

In the UK, the Laboratory Animals Science Association (LASA) and the Animals Procedures Committee (APC) published a report on the feasibility of reporting data on the severity of scientific procedures on animals
http://www.lasa.co.uk/LASA%20APC%20severity%20project.pdf.

Short presentations on different severity assessment frameworks will be given at the start of the EWG.

Task 1

Consider the factors which contribute to the severity of procedures, how the relative contribution of each can be merged to give an appropriate classification and how consistency in outcomes can be promoted.

1. 1. What factors?

What has been done to the animal?
What effect did this have on the animal?
As evidenced by clinical signs, what type of pain, suffering distress or lasting harm was experienced by the animal and how long did these last for?
What was done to reduce the impact on the animals and were these effective?

1.2 Welfare assessment schemes

What should be monitored / taken into consideration?

The determination of <u>actual</u> severity should be based on the clinical signs observed in the animals, <u>not</u> just by estimating the severity on the basis of the procedure that was conducted. Example lists of procedures are useful in guiding retrospective assessments of severity, but the individual circumstances under which each procedure is conducted can have a significant bearing on the actual severity experienced by the animal, for example the training experience of the animal with respect to the procedure and methods used to eliminate pain, suffering and distress, including refinements of housing, husbandry and care. All of these can contribute to the overall severity of the procedure; either by increasing it (e.g. if prolonged or repeated restraint is required), or reducing it (e.g. if refining husbandry can reduce anxiety). The time that elapses between the steps in a procedure will also have a bearing on assessing the cumulative severity within a procedure.

Effective systems for monitoring animals, assessing suffering and recording observations should be in place and used throughout the project. This is essential for recognising and alleviating suffering during procedures as well as to facilitate retrospective reporting of severity.

In the assessment of severity classification the FELASA guidelines (1) offers general guidance on clinical signs equating to a severity category. These signs relate primarily to rodents. A similar document for fish has recently been published (Hawkins P, Dennison N, Goodman G et al. (2011) a). There are a number of publications which give examples of different types of severity classification systems (for example JWGR (BVAAWF/FRAME/RSPCA/UFAW) Joint Working Group on Refinement) (2011), Mellor et al (2009)).

An ideal assessment system would include simple objective measurements which could be applied consistently and used to detect the onset and monitor the development of pain, suffering and distress in animals undergoing scientific procedures.

Although there are common monitoring criteria which can be applied in all species, the observed clinical signs, behaviours and behavioural responses are very varied, and as a consequence the severity classification assigned among the different species used in scientific procedures will differ.

However, the use of tailored assessment systems specific to the project using trained and experienced personnel can facilitate consistent assessment of severity and contribute towards subsequent significant refinements to animal models. Thus, it is important to list possible and/or observed behavioural or clinical responses, which can be monitored during the course of the study and can be used to identify humane end-points.

Attached as Appendix 1 is an example of clinical observations commonly used in rodent welfare assessments that will be further developed at the EWG to broaden the application. Appendix 2 provides examples of how different clinical observations can relate to differing severities.

The regular observations and records made during the course of each study will facilitate the monitoring of severity and the identification of humane end-points.

Assessment of severity

FOR DISCUSSION

Assigning relative importance of measures? Use of weighted scoring systems?

How are the clinical findings in a welfare assessment during the course of a study and during the retrospective assessment used to determine severity?

What adverse effects are of most importance to animal welfare?

Duration? Intensity?

The effectiveness of refinement measures, for example use of analgesia or intensive care practices needs to be considered.

How is cumulative severity assessed during the course of the procedure? Species differences?

How should information be recorded?

Records should be maintained in such a manner that the condition of the animal can be assessed throughout the study. This may be a simple system where only "positive" recordings are noted – this does however require that all involved have a clear understanding of "normal" base-lines.

Numerical and binary scoring systems can be used but each has its own strengths and weaknesses (see JWGR p13-14).

Prospective planning for the study should ensure that all relevant personnel have the opportunity to contribute to the assessment system to be used – although a "standard" framework is very helpful, many studies will require some additional study-specific measures, for example gait/locomotor assessment in arthritis studies.

We aim to discuss and agree on the key elements to include within a severity assessment framework, including the information to be collected and subsequent analysis.

How to promote consistency in severity assessment? Who assigns severity?

In order to promote a consistent interpretation within a user facility, in addition to the scientists, other trained, experienced persons involved in the care and use of the animals should be included in the final assessment considerations. As a minimum, the assessment should include input by the care staff i.e. the animal care staff and the veterinarian.

All those involved in the severity assessment should have a detailed knowledge and understanding of the species being used, be able to recognise normal and abnormal behaviours and signs of pain, suffering distress and lasting harm. It is important to note in this context that new developments on the understanding and recognition of pain, suffering and distress are emerging continuously, highlighting the need for effective continuous training in this field.

The determination of, and agreement of, monitoring procedures should be undertaken in advance of the study and reviewed as necessary during the course of the study.

Training in welfare assessment should be part of one of the core modules.

Regular review of decisions on severity assessments should be made by the Animal Welfare Body and designated veterinarian within the establishment to promote consistency.

Task 2

Develop some worked examples to illustrate the process by which severity can be determined in a consistent manner

A main objective of the meeting is to understand and develop the concept of severity assessment as a single continuous process from the initial design, through the course of the study until the final assessment of actual severity is made, and not a series of isolated actions to assign prospective severity classification or report actual severity. A consistent approach to severity assessment from the start until the end of the procedure ensures pro-active consideration and application of the Three Rs at all stages.

Therefore, Task 2 is to bring together the different elements discussed previously. A small number of examples of multi-step procedures will be prepared for the meeting and attendees are encouraged to bring an example of their own.

The intention is to focus on and agree the process on a few detailed examples – not to work through many different cases.

One submitted illustrative example of the severity process is attached as Appendix III.

Task 3

Finalise the welfare assessment framework for GA animals which was initiated at the first meeting of the EWG in December 2011

Although good progress was made at the first EWG meeting in December, agreement on the principles for a standardised approach to GA welfare assessment has yet to be finalised. Meeting notes are attached.

Sub-task 3a:

The following two tables are extracted from the meeting notes taking into account all comments received following the meeting in December 2011. These should be finalised to facilitate a common approach to the welfare assessment of a new GA line.

Key Elements of a GA Rodent Welfare Assessment Scheme

CRITERIA	WHAT TO LOOK FOR
Overall Appearance	Is the animal morphologically 'normal'? Are there any malformations or any other indicators that the phenotype has been affected? For example skeletal deformity or hydrocephalus.
Size, conformation and growth	Are there any deviations from expected size or growth curve?
Coat condition	Is there any piloerection, areas of fur loss, loss of whiskers, barbering? Is the skin / fur in good condition?
Behaviour – **Posture, gait, activity and interactions with the environment**	Do they exhibit the full repertoire of behaviours appropriate for the strain/species, including social interactions, grooming, walking, running, digging, climbing? Are these normal? Is the animal hunched or reluctant to move? Is movement impaired or is there any difficulty with orientation? Any signs of rigidity or tremors? Any abnormal activity levels? Prolonged inactivity could indicate chronic stress or depression (anhedonia) and/or sickness/pain, particularly if linked with a hunched posture and/or rough or unkempt coat. Unusual activity, such as hyperactivity, could indicate stereotypy or other behavioural abnormality.
Clinical signs	For example - nasal or ocular discharge, swollen or closed eyes; increased respiratory rate; dyspnoea; seizures/twitches/tremors; increased vocalisation with handling; overgrown teeth; presence of tumours, neurological or musculoskeletal abnormalities. Is metabolism impaired, for example, increased or decreased food or water intake, excessive urination? Consistency of faeces.
Relative size	Any unusual changes in size of the animals should be noted, and comparisons made within the litter. It may be helpful to generate a growth curve for the line.
Numbers	Where death occurs, it is important to maintain accurate records such that any pre- or post-weaning losses can be investigated. Where appropriate (e.g. higher than anticipated mortality rate), post mortem examinations should be carried out to help determine the

	cause of death. A review of fertility can also be helpful in assessment of whether or not the modification is having an effect eg conception rates; abortions; stillbirths.

Additional considerations for assessment in Neonatal animals

CRITERIA	WHAT TO LOOK FOR
Colour of pups (for neonate only)	Do any pups show evidence of abnormal skin colour (e.g. anaemia, poor circulation)
Activity of pups (for neonate only)	Any abnormal activity, e.g. reduced wriggling? Righting reflex intact?
Milk spot (for neonate only)	Do any pups fail to show presence of a milk spot? Any evidence of mis-mothering?
Litter	Litter sizes; litter homogeneity; development and growth of pups ;

Sub-task 3b:

Develop some worked examples of prospective welfare assessment of a new GA line.

Sub-task 3c:

The following text was developed from the Thought starter document incorporating comments received subsequent to the December meeting. To consider whether the principles can be agreed and identify any additional opportunities.

"Opportunities for Refinement in GA breeding

Having identified the harmful effects within the line, careful consideration is required to ensure that any suffering is minimised. This is particularly true within breeding colonies where breeding animals may be maintained for many months, and without careful colony management to balance supply and demand there can be potential welfare issues in unused stock animals.

Much can be done in GA breeding colonies to reduce or eliminate entirely the potential welfare compromise to the animals.

For example, barrier maintenance of immuno-compromised animals can significantly reduce the risk of infection.

Consider the design of breeding strategies – maintain as heterozygote breeding colony if adverse effects would adversely affect welfare in a homozygote breeding colony.

If line is not being used, consider cryopreservation to eliminate need to maintain any animals exhibiting harmful phenotypes.

If late onset of phenotypic abnormality, e.g. tumours develop at 16 weeks, ensure that all breeding animals are retired before the onset of clinical problems.

What principles could be drawn for Refinement in GA breeding?"

Sub-task 3d:

If time permits, develop an example of prospective and retrospective (actual) severity assessment within a GA breeding context, to assist in improving the understanding of the two different concepts of severity assessment.

References

1. FELASA (1994) Pain and distress in laboratory rodents and lagomorphs. *Laboratory Animals* 28: 97-112
2. JWGR (BVAAWF/FRAME/RSPCA/UFAW Joint Working Group on Refinement) (2011) A guide to defining and implementing protocols for the welfare assessment of laboratory animals. *Laboratory Animals* 45: 1-13
3. Mellor DJ, Patterson-Kane E & Stafford KJ (2009) Animal Welfare, grading compromise and mitigating suffering. P 71-94 in *The Sciences of Animal Welfare.*Oxford: Wiley-Blackwell
4. Hawkins P, Dennison N, Goodman G et al. (2011) Guidance on the severity classification of scientific procedures involving fish: report of a Working Group appointed by the Norwegian Consensus-platform for the Replacement, Reduction and Refinement of animal experiments (Norecopa). *Laboratory Animals***45:** 219-224