

Category	Description of Severity & Examples
0	None , e.g., use of dead vertebrate or higher invertebrate animals (already euthanized legally and ethically for another purpose and not for this study) or tissue or biological fluids thereof, or using any live lower invertebrate. Also, archived animal specimens from the laboratory, museum, private or any other collections. Non-invasive behavioural studies, e.g., observation of wild animals <i>in situ</i> (natural environment) without interference, including minimal disturbance to other biota. Domestic or farm animals on their home property, where procedures are occurring as normally part of the routine management or professional (e.g., veterinary) care.
1	Mild , e.g., keeping of animals in captive conditions (including all animals housed in registered animal holding facilities), or animals collected domestically, from the farm, in the field or other environment, confined and/or transported for subsequent observation. Handling animals or applying minimally invasive examinations, depriving animals of food/water for a few hours (no longer than experienced in nature) or anaesthetising animals that will not regain consciousness (non-recovery).
2	Medium , defined as induced stress of short duration, not requiring anaesthesia or tranquilisation, e.g., parenteral administrations, small subcutaneous implants, exercise or low-stress behavioural testing. Collection of tissue from animals after non-anaesthetic euthanasia for research purposes. Open field testing. Depriving animals of food or water for periods longer than experienced in nature, but without becoming distressful or causing permanent harm.
3	Severe , defined as induced stress of short duration, requiring anaesthesia or tranquilisation, e.g., surgical procedures (including larger implants), multiple injection sites or high-stress fluid/tissue sampling, e.g., cardiac puncture. Procedures that include known disease or severe stress-inducing agents/interventions (e.g., known genetic manipulations, pathogens, toxicity studies or toxic agents, nutrition and trauma, such as perceived unescapable predator exposure), provided the study is terminated (i.e., endpoint) at the onset of relevant symptoms (e.g., lowest observed effect concentration). Restriction of movement over a prolonged periods of time, e.g., metabolic cages or keeping large animals in bomas.
4	Very severe , defined as induced stress of long duration, e.g., prolonged stress (>30 minutes) without anaesthesia or tranquilisation, anaesthesia of long duration (>30 minutes), procedures producing defects or long-term feeding (> 2 days) of deficient or special diets. Forced swim or exercise tests with exhaustion as the end-point. Cancer models and new genetic manipulation.
5	Excessively stressful , where exceptional motivations and justification are needed for approval, e.g., application of noxious stimuli to conscious animals from which the animal cannot escape or any experiments in which total distress is the likely outcome. Toxicity/virulent studies where the endpoint is the moribund state or death. Extended social isolation of sociable animals (e.g., social isolation rearing of rats). Xenograft studies. Real, staged predator exposure which may lead to attack with harm or kill.