## **Guidelines for Determining USDA Pain/Distress Levels**

USDA Category B	USDA Category C	USDA Category D	USDA Category E
Breeding or Holding Colony Protocols	No more than momentary or slight pain or distress and no need of pain-relieving drugs.	Pain or distress appropriately relieved with anesthetics, analgesics and/or tranquilizer drugs or other methods for relieving pain or distress.	Pain or distress or potential pain or distress that is <b>not</b> relieved with anesthetics, analgesics and/or tranquilizer drugs or other methods for relieving pain or distress.
	Examples	Examples	Examples
	<ol> <li>Holding or weighing of animals in teaching or research activities.</li> <li>Injections, blood collection or catheter placement (without cut- down) via superficial vessels.</li> <li>Tattooing animals.</li> <li>Ear punching of rodents.</li> <li>Toe clipping in mouse pups before 7 days of age</li> <li>Tail clipping before 21 days of age in mice.</li> <li>Fin clipping for genotyping in fish.</li> <li>Routine physical examinations.</li> <li>Observation of animal behavior.</li> <li>Feeding studies, which do not restrict food amount or result in clinical health problems</li> <li>AVMA approved humane euthanasia procedures</li> <li>Tissue collection after euthanasia</li> <li>Positive reward projects.</li> <li>Physical restraint with appropriate acclimation</li> <li>Induced seizures</li> <li>Decapitation and cervical dislocation in rodents without anesthesia but proper training of personnel</li> </ol>	<ol> <li>Diagnostic procedures, e.g. Laparoscopy or needle biopsies.</li> <li>Non-survival surgical procedures and post-operative pain alleviated.</li> <li>Ocular blood collection in mice.</li> <li>Terminal cardiac blood collection under anesthesia.</li> <li>Exposure of blood vessels for catheter implantation.</li> <li>Exsanguination under anesthesia.</li> <li>Tumor studies with solid tumors which are localized, non-invasive, and metastases do not occur before euthanasia of the animal or early stages without affecting animals</li> <li>Food or water regulation and restriction</li> <li>Toxicological, microbiological, or infectious disease research that requires continuation until moderate clinical signs are evident but death is not the endpoint</li> </ol>	<ol> <li>Death as an endpoint – Toxicological or microbiological testing or cancer research that requires continuation until death occurs. This must be scientifically justified and approved by IACUC.</li> <li>Application of noxious stimuli such as electrical shock if the animal cannot avoid/escape the stimuli and/or it is severe enough to cause injury or more than momentary pain or distress.</li> <li>Any potentially painful/distressful procedures for which needed analgesics, tranquilizers, anxiolytics, sedatives, or anesthetics must be withheld for justifiable study purposes.</li> <li>Exposure to abnormal or extreme environmental conditions, which induce pain/distress.</li> <li>Experimental allergic encephalomyelitis</li> <li>Chemically induced or infectious colitis</li> <li>Hypoxic and hyperoxic (&gt;24 hours) environment</li> <li>CFA, formalin or carrageenan injected in foot pads (only allowed for pain studies)</li> <li>Tumor studies with known early ulceration, invasiveness and/or occurrence of metastases,</li> <li>Induced pancreatitis</li> <li>Parkinson's models</li> <li>Stroke models, both ischemic and hemorrhagic</li> <li>Asthma models with respiratory distress Administration of drugs leading to addiction including a withdrawal period and associated</li> </ol>

(Note: there is no USDA Category A.) Last IACUC review/approval: January 29, 2021