

## **Cumulative Use and Reuse of Research Animals**

Many experiments performed in support of JHU Animal Care and Use Committee (ACUC) approved protocols do not require euthanasia once study endpoints are reached. Animals are occasionally reused in multiple ACUC approved studies provided those studies are first approved by the ACUC and that reuse can be achieved without unnecessary suffering (Nunamaker et al., 2021). Reuse and cumulative use are responsive to federal guidance related to the 3 Rs (refinement, replacement, reduction), but must necessarily give precedence to humane care and use. JHU has already established policies and guidelines for animal adoption and use of animals designated for euthanasia. This guideline will focus on best practices for reuse of animals in ACUC approved protocols.

For the purposes of this Guideline, we will define reuse as the use of animals in sequentially performed experiments that may be supported by a single ACUC approved protocol or, after transfer, endpoints described in separate ACUC approved protocols. Cumulative use will be defined as repeated use for studies with essentially the same experimental endpoint (often in support of a single protocol). Both reuse and cumulative use may imply a single repetition or multiple repetitions of the same experiment or different experiments. The Principal Investigator(s) for these sequentially performed studies do not have to be the same person but the transfer of animals from one protocol to another must follow established Guidelines. Both the transfer and reuse of animals must be reviewed and vetted by the Attending Veterinarian and approved by the ACUC before subsequent experiments or procedures can be performed. Health status assessments should be performed prior to each subsequent experiment or procedure.

Examples of initial uses of animals where reuse would be appropriate include:

- Offspring from breeding colonies either not carrying the genetic composition of interest or from litters that exceed the immediate needs of the lab supporting their breeding colonies.
- Animals in studies fulfilling an untreated control endpoint that did not require surgical interventions or painful or stressful assessments of physiology or behavior.
- Animals used in drug distribution studies (e.g. PET imaging, pharmacokinetics)
- Retired breeding animals

Examples of initial uses/ circumstances where reuse of animals would likely NOT be appropriate include:

- Animals that have undergone major survival surgeries
- Animals exposed to infectious agents or toxic chemicals
- Animals that have had encounters with physically or psychologically demanding interventions or environments.
- Animals that have, either through happenstance or experimental intervention, experienced any physiological, neurological, psychological or immunological disorder.

The ACUC will consider all proposed plans for reuse of animals. Assessing the merits of these proposals will always prioritize animal welfare. Cumulative use, perhaps even throughout lifespans, may be permissible if investigators can document no decrement to the well-being of the animals as they accrue ACUC approved uses. Investigators should carefully consider humane endpoints associated with reuse prior to seeking ACUC approval while also considering efforts to reduce the total number of animals used in any of their studies.

Nunamaker EA, Davis S, O'Malley CI, Turner PV. Developing Recommendations for Cumulative Endpoints and Lifetime Use for Research Animals. Animals (Basel). 2021; 11(7):2031.