Contains Nonbinding Recommendations

General Considerations for Animal Studies Intended to Evaluate Medical Devices

---

Guidance for Industry and Food and Drug Administration Staff

Document issued on March 28, 2023.

The draft of this document was issued on October 14, 2015.

This document supersedes "Guidance for Industry and FDA Staff: General Considerations for Animal Studies for Cardiovascular Devices" issued July 29, 2010.

For questions about this document, contact CDRHanimalstudies@fda.hhs.gov.



U.S. Department of Health and Human Services
Food and Drug Administration
Center for Devices and Radiological Health

GRZAN

Preface

Public Comment

You may submit electronic comments and suggestions at any time for Agency consideration to https://www.regulations.gov. Submit written comments to the Dockets Management Staff, Food and Drug Administration, 5630 Fishers Lane, Room 1061, (HFA-305), Rockville, MD 20852. Identify all comments with the docket number FDA-2015-D-3419. Comments may not be acted upon by the Agency until the document is next revised or updated.

Additional Copies

Additional copies are available from the Internet. You may also send an email request to CDRH-Guidance@fda.hhs.gov to receive a copy of the guidance. Please include the document number 1802 and complete title of the guidance in the request.

GRZAN

Table of Contents

I. Introduction 1
II. Overview 1
III. Scope 3
IV. Considerations When Determining the Need to Conduct an Animal Safety Study 4
V. Personnel 5
VI. Study Planning and Conduct 6
A. Study Design 6
(1) Study Objectives 7
(2) Selecting an Animal Model 8
(3) Sample Size and Groupings 8
B. Test System Monitoring 9
(1) Intra-Procedural Monitoring 10
(2) Post-Procedural Recovery and Monitoring 10
(3) In-Life Monitoring 10
(4) Unexpected Morbidity and Mortality 11
C. Post-Mortem Assessment Methods 11
(1) Post Mortem Imaging 12
(2) Necropsy and Gross Tissue Evaluation 12
(3) Histologic Evaluation 12
D. Test and Control Article Characterization 13
VII. Testing Facility Selection 13
VIII. Animal Housing 14
A. Animal Quarantine and Conditioning 15
B. Food, Water, and Basic Husbandry 15
IX. Records and Reports 15
X. Preparation of Animal Study Reports for Medical Device Premarket Submissions 16
A. Executive Summary 17
B. Final Study Report 17
(1) In-Life Veterinarian Report 18
(2) Pathology Report 18
(3) Other Reports/Forms 19
Appendix A - Example Organization of Animal Study Final Report 20

GRZAN

General Considerations for Animal Studies Intended to Evaluate Medical Devices

---

Guidance for Industry and Food and Drug Administration Staff

This guidance represents the current thinking of the Food and Drug Administration (FDA or Agency) on this topic. It does not establish any rights for any person and is not binding on FDA or the public. You can use an alternative approach if it satisfies the requirements of the applicable statutes and regulations. To discuss an alternative approach, contact the FDA staff or Office responsible for this guidance as listed on the title page.

I. Introduction

FDA developed this guidance document to assist medical device sponsors, testing facilities, and other persons involved in designing, conducting, and reporting the results of animal studies intended to assess the safety of medical devices to support premarket submissions. These animal studies typically provide initial evidence of device safety, which may include device performance and handling, and the biological effects of the device when used in a living system. The recommendations in this guidance reflect current review practices and are intended to promote consistency and facilitate efficient review of medical device submissions that include animal study data.

In general, FDA's guidance documents do not establish legally enforceable responsibilities. Instead, guidances describe the Agency's current thinking on a topic and should be viewed only as recommendations, unless specific regulatory or statutory requirements are cited. The use of the word *should* in Agency guidances means that something is suggested or recommended, but not required.

II. Overview

The *in vivo* setting generally provides an initial assessment of how a medical device interacts with biological systems, including physiological, pathological, and toxicological effects of the device, and how the biological system may affect the device. Animal studies are often conducted to support medical device premarket submissions. Nonclinical laboratory studies, including

animal studies,[1] intended to support premarket submissions must comply with the Good Laboratory Practice for Nonclinical Laboratory Studies (GLP) Regulations.[2] The GLP Regulations are intended to ensure the quality and integrity of the safety data submitted as part of a premarket submission to FDA.[3]

A primary purpose of an animal study submitted in support of a premarket submission is for the submitter to provide evidence of safety, which could include performance and handling[4], of a medical device. Note that in many cases, the performance and handling of a particular device is intricately linked to its safety, including but not limited to implantable devices or interventional devices. FDA acknowledges that an animal study can have multiple purposes beyond the assessment of safety. For example, a secondary objective for conducting an animal study can be to evaluate the effectiveness of the device or to demonstrate proof of concept and principle of operation. Manufacturers could also conduct an animal study to help meet design verification and validation requirements.[5]

Animal studies submitted to the Agency to support the safety of a medical device must comply with the GLP Regulations. However, the intent of this guidance is not to comprehensively describe when compliance with the GLP Regulations is required for an animal study. The applicability of the GLP Regulations to a specific animal study may be dependent on individual facts and circumstances. If you have specific questions on aspects of your animal study or your study's compliance with the GLP Regulations, we recommend that you seek feedback from FDA through the Q-Submission process.[6]

This guidance outlines the general considerations for certain animal studies used to support device premarket submissions, when a suitable alternative to an animal study is not available. This guidance provides recommendations for various elements of animal studies, including the credentials for personnel conducting an animal study, and the study planning and conduct process, including but not limited to selecting an appropriate animal model, study monitoring, and study evaluation. This guidance also provides recommendations on testing facility[7] selection, animal housing, records and reports, and how to prepare an animal study report for premarket submissions to FDA.

---

[1] This guidance contemplates animal studies that meet the definition of nonclinical laboratory studies, and does not include field trials in animals. See 21 CFR 58.3(d).
[2] See 21 CFR Part 58 which outlines the requirements for Good Laboratory Practice for Nonclinical Laboratory Studies. For the purposes of this guidance document, 21 CFR Part 58 will be referenced as the "GLP Regulation."
[3] See 21 CFR 58.1(a).
[4] For the purposes of this guidance document, "handling" is defined as the way the device or device system responds to the demands of the operator, such as ease of insertion/deployment, radiopacity, tracking, maneuvering, and retrieval.
[5] 21 CFR 820.30 outlines the requirements for design verification and validation.
[6] For more information, see "Requests for Feedback and Meetings for Medical Device Submissions: The Q-Submission Program, Guidance for Industry and Food and Drug Administration Staff," available at https://www.fda.gov/regulatory-information/search-fda-guidance-documents/requests-feedback-and-meetings-medical-device-submissions-q-submission-program.
[7] See 21 CFR 58.3(g) for definition of a testing facility.

FDA supports the principles of the "3Rs," to replace, reduce, and/or refine animal testing [8,9,10,] animal use in testing when feasible. FDA encourages sponsors to consult with the Agency through the Q-Submission process if you wish to use a non-animal testing method that is suitable, adequate, validated, and feasible. Through the Q-Submission process, FDA will consider if a proposed alternative testing method could be assessed for equivalency to an animal test method.[11] FDA also encourages the use of the Q-Submission process to obtain feedback on the design of an animal study if an animal study is warranted. Additionally, it is important to consider the adequacy of controls, timing and route of intervention, and methods to minimize bias (i.e., blinding, randomization, use of controls, sample size based on expected magnitude of the biological response, reporting missing data, and clearly stated statistical considerations) when designing an animal study.[12] FDA recommends that sponsors submit a Pre-Submission (a type of Q-Submission) for clarification regarding elements of the animal study to support a premarket submission.

FDA acknowledges that there are other relevant statutes, regulations, and policies that govern laboratory animal care.[13,14,15,16,17,18] FDA maintains a memorandum of understanding (MOU) with the Animal and Plant Health Inspection Service (APHIS) in the United States (U.S.) Department of Agriculture (USDA) and the National Institutes of Health (NIH) that sets forth a framework for reciprocal cooperation, which will assist each agency in meeting its responsibilities in promoting proper laboratory animal care and welfare in the U.S.[19]

III. Scope

The recommendations in this guidance refer to devices, as defined in section 201(h) of the Federal Food, Drug, and Cosmetic Act (FD&C Act), intended for human use. This guidance is applicable to animal studies intended to provide evidence of device safety, which may include

---

[8] "Advancing New Alternative Methodologies at FDA," available at https://www.fda.gov/media/144891/download.
[9] Russell WMS, Burch, RL. The Principles of Humane Experimental Technique. London: Methuen & Co.; 1959. Special edition published by Universities Federation for Animal Welfare, 1992, available at http://altweb.jhsph.edu/pubs/books/humane_exp/het-toc.
[10] "Animal Use Alternatives (3Rs)", available at https://www.nal.usda.gov/animal-health-and-welfare/animal-use-alternatives#:~:text=The%20%E2%80%9C3Rs%20alternatives%E2%80%9D%20refers%20to,Principles%20of%20Humane%20Experimental%20Technique%22.
[11] See FDA webpage for Medical Device Development Tools (MDDT) (https://www.fda.gov/medical-devices/science-and-research-medical-devices/medical-device-development-tools-mddt) for potential alternatives to animal studies.
[12] See 21 CFR 58.120.
[13] See 7 U.S.C. § 2131–59.
[14] See 9 CFR Parts 1–4.
[15] Health Research Extension Act of 1985, Public Law 99–158, 99 Stat. 820 (Nov. 20, 1985).
[16] U.S. Department of Health and Human Services, National Institutes of Health, Office of Laboratory Animal Welfare (2015). *Public health service policy on humane care and use of laboratory animals*. Retrieved from https://grants.nih.gov/grants/olaw/references/phspolicylabanimals.pdf.
[17] National Research Council of the National Academies (2011). *Guide for the care and use of laboratory animals* (8th ed.). Retrieved from https://grants.nih.gov/grants/olaw/guide-for-the-care-and-use-of-laboratory-animals.pdf.
[18] FDA Guidance for Industry (2007). Good Laboratory Practices - Questions and Answers. Available at https://www.fda.gov/media/75866/download
[19] FDA MOU 225-16-010. Available at https://www.fda.gov/about-fda/domestic-mous/mou-225-16-010.

performance and handling, in premarket submissions to FDA.[20] These types of submissions include investigational device exemption (IDE) applications, premarket approval applications (PMA), premarket notification (510(k)), humanitarian device exemption (HDE) applications, or De Novo classification requests. This guidance is not intended to alter or supersede any device-specific final guidance but is intended to augment the recommendations in device-specific guidance.

This guidance also does not apply to basic exploratory studies conducted to determine whether a device has any potential utility or preliminary assessments of physical or chemical characteristics of a device. In addition, this guidance does not apply to, nor changes any regulatory requirements relevant to, animal studies conducted under the Animal Rule.[21] Finally, this guidance is not intended to address the regulations and policies of other agencies.

IV. Considerations When Determining the Need to Conduct an Animal Safety Study

Below outlines general considerations a sponsor may find helpful when determining whether an animal safety study should be conducted to support a premarket submission.

- A risk analysis[22] that considers all sources of relevant information, including known failure modes and previously identified risks for the device, risks commonly attributed to this general device type, and postmarket information for similar marketed devices. Postmarket information can be searched using the published literature and the Manufacturer and User Facility Device Experience (MAUDE) database.[23]
- Testing the evaluable risks on the benchtop to the extent feasible using the device in its final finished form (a device in its final finished form includes all manufacturing processes for the "to be marketed" device including packaging and sterilization, if applicable). Before considering an animal study to support a premarket submission, FDA recommends completion of nonclinical benchtop performance testing with the device in its final finished form to evaluate potential harm(s) identified during the risk analysis.
- Consider conducting an animal study when the risk analysis suggests that such a study is necessary to further assess potential safety concerns that cannot be adequately addressed through alternative methods. When sponsors are uncertain about whether they should conduct an animal study, FDA recommends utilizing the Q-Submission process to request FDA feedback on the overall nonclinical testing approach.

[20] For ease of reference, this guidance will refer to these studies as "animal safety studies."
[21] See 67 FR 37988 (May 31, 2002). Those animal studies submitted for premarket review of drug and biologic products are also specifically outside the scope of this guidance.
[22] For example, ISO 14971: *Medical devices – Application of risk management to medical devices*.
[23] FDA MAUDE Database. Available online at: https://www.accessdata.fda.gov/scripts/cdrh/cfdocs/cfMAUDE/search.CFM.

- When there is no established animal model for the type of device (i.e., one that has been described in the literature or used to support the clearance or approval of a similar device), as applicable, we recommend that sponsors:
    - Assess the anatomy and physiology (e.g., angiographic, radiographic, computed tomography (CT) screening) of commonly used laboratory animal species for intended use, allometric scaling, and procedural approach features.
    - Consult an experienced laboratory animal veterinarian to determine the availability and utility of common laboratory species.
    - Engage with the Agency using the Q-Submission process to discuss significant challenges that preclude the use of a reasonable animal model for all or some of the animal studies recommended by the risk analysis and potential alternative approaches for collecting information to support device safety before clinical use in humans. As part of this discussion, sponsors should include any available information that demonstrates that animal studies would not be feasible and propose alternative solutions, including any available simulations, cadaveric studies, and clinical information. Although FDA generally does not consider cost as sufficient justification for not conducting animal studies, we apply least burdensome principles to premarket regulatory activities.[24]

FDA recommends that sponsors submit a Pre-Submission if how to structure or conduct the animal study is unclear. FDA recommends the Pre-Submission include a detailed proposal for the animal study design that includes specific questions for which they would like FDA input.

V. Personnel

FDA believes that a carefully planned and executed animal study is more likely to provide useful data in support of a device premarket submission. In this regard, animal studies should be planned and conducted by a team with appropriate credentials and training. Under the GLP Regulations, a study director[25] with relevant education, training and experience must be designated before initiation of a nonclinical laboratory study[26] to provide direct oversight of the technical conduct of the study, among other responsibilities.[27]

Further, the GLP Regulations also require the use of a Quality Assurance Unit (QAU). QAU personnel are independent from the personnel engaged in the direction and conduct of the study.[28] The QAU for an animal study could be an independent contractor or employed elsewhere within the testing facility organization.

---

[24] See "The Least Burdensome Provisions: Concept and Principles: Guidance for Industry and FDA Staff," available at https://www.fda.gov/regulatory-information/search-fda-guidance-documents/least-burdensome-provisions-concept-and-principles

[25] Consistent with 21 CFR 58.3(m), study director means the individual responsible for the overall conduct of a nonclinical laboratory study.

[26] See 21 CFR 58.31(a).

[27] See 21 CFR 58.33.

[28] See 21 CFR 58.35.

FDA also recommends that the animal study team include skilled clinical veterinary staff to help ensure humane care of study animals.[29] Animal studies may also benefit from input from investigators with expertise in specialty fields, such as engineering, pathology, radiology, surgery, and laboratory animal science. For example, testing for sensorimotor function involves specific neurologic and/or behavioral expertise. Animal models may also involve unique surgical approaches or present anatomical limitations that would benefit from having trained veterinary surgical expertise as part of the research team.

FDA notes that appropriate training and experience of study personnel that may apply to animal studies are also addressed in other relevant guides, regulations, and policies.[30,31, 32, 33, 34]

VI. Study Planning and Conduct

Under the GLP Regulation, animal studies shall be conducted according to a written protocol that clearly indicates the objectives and all methods for the conduct of the study.[35] The protocol should provide enough description to allow FDA to reconstruct the study.

FDA recommends conducting exploratory animal studies before conducting animal safety studies when *in vivo* or *in vitro* information exists that shows a significant learning curve that would significantly increase animal response variability. This could be caused by factors, such as investigator training and familiarity with the device or inherent challenges in the placement or tolerance of the device.

A. Study Design

FDA recommends that the animal study is designed to ensure that the testing environment simulates (to the extent possible) a clinical setting that reflects the intended use and proposed labeling of the device (i.e., instructions for use). Under the GLP Regulation, animal studies must be guided by a study protocol that is approved by the sponsor and signed and dated by the study director.[36] If the study is conducted outside of the United States, an equivalent, signed protocol should be in place.[37]

---

[29] See 21 CFR 58.29 and 9 CFR 2.31 which requires the USDA licensed facilities to appoint an Institutional Animal Care and Use Committee to oversee the animal study program.

[30]Russell WMS, Burch, RL. The Principles of Humane Experimental Technique. London: Methuen & Co.; 1959. Special edition published by Universities Federation for Animal Welfare, 1992, hereafter called Russell. Available online at: http://altweb.jhsph.edu/pubs/books/humane_exp/het-toc.

[31] 9 CFR Parts 1–3.

[32] U.S. Department of Health and Human Services, National Institutes of Health, Office of Laboratory Animal Welfare (2015). *Public health service policy on humane care and use of laboratory animals*. Retrieved from https://grants.nih.gov/grants/olaw/references/phspolicylabanimals.pdf.

[33] FDA MOU 225-16-010. Available at https://www.fda.gov/about-fda/domestic-mous/mou-225-16-010.

[34] See 21 CFR 58.29.

[35] See 21 CFR 58.120(a).

[36] See 21 CFR 58.120(a)–(b).

[37] See 21 CFR 58.120(a).

FDA generally recommends that sponsors use the endpoints, time points, and methods reported for similar devices, if available. Sponsors should consider identifying new endpoints, time points, and methods when there are:

- Features of the device that would suggest the need to evaluate different study time points and objectives from those previously used in studies for similar devices with similar intended use; or
- New indications that suggest the use of different or additional evaluation time points or methods.

The study protocol should not contain statements that limit the collection of data or the reporting of findings so that FDA has a more complete understanding of all data collected in the study, as well as the interpretation of the data.

FDA recommends limiting study bias to help ensure the quality of the data collected during an animal study.[38] FDA recommends incorporating principles of experimental methodology to minimize bias or the perception of bias and to control variation, including but not limited to:

- randomization procedures to control for animal selection and procedural bias;
- blinding/masking procedures to control for assessment bias;
- use of more than one observer to control for perception bias;
- choice of experimental design to increase precision and reduce variation (i.e., blocking, stratification, and/or factorial designs);
- inclusion of positive, negative, and/or sham control arms;
- ensure calibration of instruments and/or equipment to minimize measurement error; and
- minimize contributions from study personnel with financial conflicts.

(1) Study Objectives

a. Device/Procedure Safety Risks

FDA recommends the study director consider the risk analysis (i.e., the identified risks associated with the device through bench testing, device design, exploratory studies, literature review) to determine the specific risks to be evaluated in the study. The study director should also consider the known risks of the device type for the same intended use.

FDA recommends study objectives be designed to address the risks that have been identified for the device or for the type of device. Acceptance criteria should be developed with a scientific rationale to address each study objective. Effectiveness could be a secondary objective in these studies.

b. Device Performance and Handling

FDA considers studying relevant performance and handling parameters essential to the assessment of device safety. Therefore, FDA recommends that study protocols include study objectives to address how device performance and device handling contribute to the overall safety profile of the device. For example, a study to evaluate the safety of a device that is intended to ablate tissue by heating, cooling, or mechanical disruption should include an assessment of successful ablation of the target tissue to demonstrate that the procedure can be

[38] See 21 CFR 58.120(a)(6)

performed safely. FDA recommends addressing this by ensuring that all steps to deliver, implant, and/or use the device be identified and assessed. For example, if the device is surgically placed, there should be an evaluation of all steps undertaken from the anatomic point of surgical entry to placement location.

When reporting study findings pertaining to device performance and handling, there should be enough detail to understand the outcomes and potential device safety issues. FDA recommends that a rating scale be created that allows semi-objective scoring of device performance and handling parameters, such as flexibility, pushability, visibility, torquability, kinking, bending, leaking, ease of use, and surgical placement. Rating criteria should encompass steps between the preparation of the device through device placement or use as well as withdrawal and redeployment, if appropriate. We recommend using a semi-quantitative method of assessment for each parameter (e.g., for a scale of 1-4, 1 = poor and 4 = excellent). The score used for acceptability (total score, by parameter, or a combination of both) should be established *a priori* and based on a scientific rationale. This information should be provided in the study protocol.

c. Local and Systemic Effects

Medical devices can have both local and systemic effects on a biologic system. This includes distal physiologic and/or tissue effects that might be caused by the device and/or delivery system. For example, perturbation of the vagus nerve in the cervical region can produce adverse effects, such as gastrointestinal distension and bradycardia. The study should include objectives to evaluate potential adverse effects on the structure and function of tissues locally and systemically, as identified in the risk assessment.

(2) Selecting an Animal Model

The animal and its related environmental and physiologic attributes should provide a testing condition that offers a best attempt at simulating the clinical setting.[39] There should be scientific information to support the appropriateness of the chosen animal model. In some cases, there may not be an established or accepted animal model for a specific device type. The selected animal model should be able to address the identified objectives of the study. The sponsor should consider scientifically accepted animal models of disease when they exist. FDA feedback on the animal model selected can be obtained through the Q-Submission process.[40]

(3) Sample Size and Groupings

The number of animals and experimental groupings should be designed after exploratory testing to provide some information on reliability and outcome if other data are not available to justify the design of the GLP study (e.g., sample size and grouping).[41] The study should use the least number of animals that could provide a predictive outcome and meaningful interpretation; this includes such measures as choosing the appropriate experimental control and consideration of potential experimental confounders and best observation intervals.

---

[39] Michel, K. D., Sorenmo, K., Shofer, F. S. (2004). Evaluation of body condition and weight loss in dogs presented to a veterinary oncology service. *Journal of Veterinary Internal Medicine*, 13(5), 692-5.

[40] For more information, see Q-Submission Guidance, available at https://www.fda.gov/regulatory-information/search-fda-guidance-documents/requests-feedback-and-meetings-medical-device-submissions-q-submission-program.

[41] Exploratory studies provide another means to identify and estimate variability, often providing key information for planning the study design and adequate sample size to obtain valid results.

Recognizing the inherent variability of animal studies, sample sizes should be sufficient to help meet the objectives of the study. Low animal enrollment risks data confounders such as missing data, unexpected animal death, or data outliers which make interpretation of data challenging.

FDA recommends performing a sample size calculation[42,43] or estimating the number of animals based on exploratory studies. Considerations that may assist in the calculation of the sample size include:

- determining the standard deviation from an exploratory study, a previous study using a similar device, or relevant published studies;
- using an adequate number of control animals to minimize experimental variability and error;
- determining the difference between treatment and control that would be clinically significant; and
- using uniform animals (breed, sex, age, weight) to reduce inter- and intra-animal variability.

B. Test System Monitoring

FDA recommends that the animal study protocol follow current veterinary standards of care appropriate for the device and procedure. The study director should work with veterinary staff at the testing facility to develop pre-specified plans to monitor for and manage anticipated adverse events at each phase of the study. Additionally, animal responses may be unpredictable; monitoring should be adequate to address all adverse events that occur. Such monitoring is important not only for humane reasons but also because it can help differentiate spontaneous conditions from device-related events.

Many animal studies involve procedures that call for anesthesia and analgesia. Anesthetic/analgesic protocols are used for any procedures that may cause more than momentary or slight pain or distress to the animals. The study director should consult with the veterinarian when developing anesthetic and analgesic plans,[44] such that appropriate drugs and dosages for the species, age, and weight of the animals, as well as the type of procedure are used. It may also help prevent introducing potential confounders to the study data.

Pain can negatively affect many physiologic systems, including wound healing, behavior, metabolism, and body system functions (e.g., immunity, pulmonary, cardiac, gastrointestinal, and urinary).[45] The type and extent of pain that an animal is likely to experience during or after a specific experimental procedure should be predicted. This can enable the veterinarian to devise

---

[42] ACLAM position statement on reproducibility (2016). *Journal of American Association for Laboratory Animal Science*, 55(6), 826-828824-825.
[43] Snedecor, G. W. & Cochran, W. G. (1990). *Statistical methods* (8th ed.). Ames, IA: Iowa State University Press.
[44] See 9 CFR 2.31(d)(iv).
[45] DeMaraco G. J. & Pascoe, P. J. (2008). Anatomy, physiology, and effects of pain. In R. E. Fish, M. J. Brown, P. J. Danneman, & A. Z. Karas (Eds.), *Anesthesia and analgesia in laboratory animals* (2nd ed., pp. 3-25). Burlington, MA: Academic Press.

plans that include the use of suitable analgesics, supportive therapeutic measures, and criteria for triggering intervention and/or humane euthanasia. Anticipation of the type(s) of pain expected also informs the frequency and type(s) of assessment needed to effectively manage pain.

(1) Intra-Procedural Monitoring

Adequate physiologic monitoring during anesthesia with collection (or recording) of physiological parameters monitored is an important aspect of a well-conducted animal study. FDA recommends that tracked vital signs, such as heart rate, respiratory rate, oxygen saturation, end-tidal CO2, body temperature, and blood pressure, be recorded at regular intervals throughout an anesthetic procedure. Consider whether additional monitoring, such as imaging, measurement of blood gases, or assessment(s) of specific clinical pathology(ies) (e.g., lactate, glucose, hematocrit/hemoglobin, or activated clotting time), is warranted. All monitoring information should be correlated with the timing of insertion, implantation, deployment, or use of the device, as well as the use of contrast agents or other device-associated materials and noted on the anesthetic and/or operative records.

(2) Post-Procedural Recovery and Monitoring

Sponsor should follow the current standard of care for laboratory animals during and immediately following an anesthetic procedure.[46] Human clinical care should be replicated whenever possible. Animals should be monitored for pain, hypo/hyperthermia, hypovolemia, and changes in mentation. Preemptive standard operating procedures should be established with the veterinarian to address any potential complications, including thermal and/or fluid interventions and rescue analgesic protocols that are appropriate to the device and procedure. Multimodal analgesic plans are often most effective. These plans should be in place before initiating the study to avoid confusion during the conduct of the study.

(3) In-Life Monitoring

During periods when animals have fully recovered from study procedures but are to be monitored for device-associated risks, FDA recommends observation at least twice daily at feeding times when they are active. The study veterinarian should be notified promptly when abnormal behaviors or clinical signs are noted. A pre-specified plan should be in place to address any abnormal clinical finding during sub-acute and chronic periods of the study.

Important parameters to consider for daily evaluation include but are not limited to:

- general appearance;
- appetite;
- mentation;
- respiratory rate, pattern, and effort;
- posture, gait; and
- urination and defecation.

[46] See 9 CFR 2.31(d)(ix)

FDA also recommends consulting the veterinarian and developing a weight monitoring plan. Consider inclusion of body scoring as an adjunct to periodic observations of the animals.[47,48]

If the study involves in-life monitoring procedures, such as wound dressing changes, imaging, collection of clinical pathology data, or more advanced diagnostics, the sponsor should plan the protocol for maximum efficiency. Grouping data collection activities requiring chemical restraint together may also minimize handling stress. In these cases, FDA recommends developing a chemical restraint protocol that does not interfere with the data. Some animals, such as dogs and sheep, may be conditioned to be compliant for these activities without the need for chemical restraint, while other animals, such as pigs, may be more difficult to condition.

Imaging methods, such as radiography, computed tomography, magnetic resonance imaging, and/or fluoroscopy, can be helpful in evaluating device safety (e.g., migration and structural integrity). *In vivo* device visualization data can also provide information of device stability (e.g., spatial positioning).

To best collect in-life study monitoring data, FDA recommends following current standards of record-keeping in veterinary medicine, such as the problem-oriented veterinary medical record (POVMR) or equivalent.[49] Additionally, these records should be readily available to all key support personnel to optimize data entry.

(4) Unexpected Morbidity and Mortality

FDA recommends that all animal illness and unexpected or early deaths be assessed and documented with regards to causation. Assessments should include any post-operative, interim, and terminal clinical pathology, gross pathology, and histopathology as well as any other indicated diagnostic methods. The supporting rationale for statements made regarding whether adverse events are related, directly or indirectly, to the device should be thoroughly described. Retrospective testimonials and statements made by study directors, their designees, or their consultants that explain veterinary clinical outcomes should be supported by appropriate records and reports. In the event of an unscheduled death, FDA recommends that a timely (for example, <24 hours to minimize tissue degradation), complete necropsy be performed on the animal to provide information to support the cause of death.

C. Post-Mortem Assessment Methods

Post-mortem assessment methods are important to characterize test and control tissue responses, as well as to visualize the device in situ and/or after the device is explanted. The assessment methods described below may not be appropriate or adequate for all studies. Planned assessments, including tests, analyses, and measurements, must be documented in the protocol.[50]

---

[47] ACLAM position statement on reproducibility (2016). *Journal of American Association for Laboratory Animal Science*, 55(6), 826-828824-825.
[48] Snedecor, G. W. & Cochran, W. G. (1990). *Statistical methods* (8th ed.). Ames, IA: Iowa State University Press
[49] Saidla, J. F. (1983). Problem-oriented veterinary medical record. In S. J. Ettinger & E. C. Feldman (Eds.), *Textbook of veterinary internal medicine: Diseases of the dog and cat* (Vol. 1, 2nd ed.). Philadelphia, PA: W.B. Saunders Co.
[50] See 21 CFR 58.120(a)(9)

(1) Post Mortem Imaging

Imaging methods are helpful in evaluating device safety parameters, such as migration, stability (e.g., spatial positioning), and/or structural integrity. Before necropsy, an *in situ* evaluation of the device could assist in the evaluation of device safety and should be considered when developing the protocol. Imaging may also be useful to assess the structural integrity of the explanted device. Evaluation of surface characteristics of some medical devices (e.g., some implanted devices) may be an important method to assess safety. When warranted, FDA recommends the use of scanning electron microscopy to characterize an implant device surface after it has been explanted from the animal.

(2) Necropsy and Gross Tissue Evaluation

FDA recommends that a comprehensive, systematic necropsy[51] be performed by a board-certified veterinary pathologist on all animals enrolled in the study to identify and describe any abnormal findings that may be a result of the device or the procedure. This can help maximize the information gained from each animal and helps identify confounding outcomes and attribute causes to certain findings. FDA recommends that a timely, complete necropsy be performed on the animal to provide information to support the cause of death. Deceased animals awaiting necropsy should be refrigerated and not frozen, to avoid changes caused by ice crystal formation.

Gross observations are necessary for appropriate tissue sampling and trimming orientation. FDA recommends that you describe the process and rationale for the sectioning of tissues to help ensure unbiased, representative tissue sampling from each animal in the protocol. The study prosector should have sufficient training and experience to assure an accurate, objective process in the sampling of gross tissue for microscopic examination.[52] High-quality color photographs should be taken of gross tissues, including all abnormal findings and representative corresponding normal tissue. All gross images should be labeled with an animal identifier (ID), study group ID, size marker, and date for ease of identification. It is helpful if images include arrows, or other identifying mark, to highlight specific observations.

(3) Histologic Evaluation

Proper interpretation of tissue responses to a device is critical to FDA's evaluation of safety, especially in the absence of clinical data. Tissues should be adequately sectioned and examined to enable reasonable evaluation of the tissue. FDA recommends performing histology on all tissues potentially affected, directly and indirectly, by the device. For example, if a device may form emboli (i.e., particulates or thrombi), either from the device itself or its action (e.g., ablation devices), then the histologist should also perform full histology on all potentially affected tissue, such as kidney, lung, heart, brain, and coronary bands. The histologist should include high-resolution, labeled color microphotographs of each slide in the final study report.

---

[51] King, J. M., Roth-Johnson, L., Dodd, D. C., & Newsom, M. E. (2013). *The necropsy book: A guide for veterinary students, residents, clinicians, pathologists, and biological researchers.* Retrieved from https://ecommons.cornell.edu/bitstream/handle/1813/37948/King_John_Necropsy_Book_10Mar16.pdf?sequence=5&isAllowed=y.

[52] Crissman, J. W., Goodman, D. G., Hildebrandt, P. K., Maronpot, R. R., Prater, D. A., Riley, J. H., Thake, D. C. (2004). Best practices guideline: Toxicologic histopathology. *Toxicologic Pathology*, 32(1), 126-131.

The testing facility should seek the expertise of an experienced, board-certified veterinarian and/or physician pathologist when developing and executing methods for preparing tissues for histologic analysis. FDA also recommends developing pre-specified objective methods for scoring and analyzing observations of tissue reaction. Specific assessments, such as inflammation, vascularization, calcification, proteoglycan/collagen, and fibrin/thrombus should be considered in the evaluation as appropriate. The use of pathology keys that further detail the histology grading system may be useful in this regard.[53, 54]

D. Test and Control Article Characterization

FDA recognizes that a medical device is often modified multiple times during the development process before an IDE or marketing authorization is requested from the Agency. FDA recommends that the animal study use the device in its final (or finished) form whenever possible. This ensures that study data can be best interpreted for evaluation of safety of the final device. If the final device design was not used in the animal study, the sponsor should identify all differences between the tested device and the final device design and explain why these differences would not be expected to affect study outcomes. Additionally, the sponsor should justify why the final clinical device design presents no new risks as compared to the design evaluated in the animal study.

If the device is intended to be used in conjunction with any accessories, the sponsor should describe these in the study protocol. The protocol should clearly state which, if any, accessories used in the animal study are intended to be provided separate from the medical device under study (i.e., commercially available) versus accessories that are expected to be marketed together with the device (i.e., a device system). FDA recommends the any test accessories used in the animal study also be in their final finished form.

Medical devices should be packaged, sterilized, and shipped to the testing facility in the same manner as the device in its final finished form. The sponsor should develop and follow a method for tracking the test and control devices from their manufacture or procurement to final use.[55]

VII. Testing Facility Selection

Testing facilities can significantly affect the outcome of animal studies. The sponsor should ensure that the selected facility complies with all applicable regulations related to housing, care, and transport of research animals.[56,57,58] FDA recommends that the sponsor consult published guidelines involving the well-being of research animals to minimize experimental confounding

---

[53] *ANSI/AAMI/ISO 10993-6: Biological Evaluation of Medical Devices, Part 6: Tests for Local Effects after Implantation*.
[54] Gibson-Corley, K. N., Olivier, A. K., & Meyerholz, D. K. (2013). Principles for valid histopathologic scoring in research. *Veterinary Pathology*, 50(6), 1007-1015.
[55] See 21 CFR 58.107.
[56] Russell WMS, Burch, RL. The Principles of Humane Experimental Technique. London: Methuen & Co.; 1959. Special edition published by Universities Federation for Animal Welfare, 1992, available athttp://altweb.jhsph.edu/pubs/books/humane_exp/het-toc.
[57] FDA MOU 225-16-010. Available at https://www.fda.gov/about-fda/domestic-mous/mou-225-16-010.
[58] National Research Council Institute of the National Academies (2006). *Guidelines for the humane transportation of laboratory animals*. Washington, DC: National Academies Press. Retrieved from http://www.nap.edu/openbook.php?record_id=11557&page=R1

factors that could adversely affect the study results.[59,60] The referenced guidelines address recommendations for minimum housing, husbandry standards, social and environmental enrichment, and the development of standard operating procedures that address timely and adequate veterinary medical care.

In addition, FDA recommends that the sponsor carefully consider the animal study design when selecting the testing facility. If the proposed animal study design is complex, use of facilities and staffing equivalent to the human clinical setting may be warranted to facilitate high quality study outcomes. Sponsors should ensure that testing facilities have appropriate equipment, supplies, and adequate staff with appropriate training, including training with the medical device, to perform the study. For example, some animal studies may benefit from advanced imaging and intensive care procedures. Sophisticated post-procedural monitoring and care may also be warranted, including 24/7 staffing similar to the clinical care patients receive in an intensive care unit.

FDA does not license or certify testing facilities with respect to compliance with the GLP Regulations. A testing facility conducting a GLP animal study must comply with all applicable requirements under 21 CFR Part 58, and other applicable requirements. Consistent with 21 CFR 58.15, a testing facility must also permit an authorized FDA employee, at reasonable times and in a reasonable manner, to inspect the facility and to inspect all records and specimens required to be maintained for a GLP animal study.

VIII. Animal Housing

Testing facilities are required to have enough animal enclosures to assure proper separation of species, quarantined animals, and test systems.[61] The sponsor may determine that animal study procedure(s) warrant more intensive care during certain predicted sub-acute periods. In these cases, FDA recommends that the testing facility also include access to recovery rooms or enclosures that allow for individual animal monitoring and treatment as appropriate.

Outside of the post-procedural monitoring period, FDA recommends housing social animals in conspecific groups whenever possible to reduce stress. However, the environmental conditions should not interfere with the assessment of the study. Group-housed animals should be monitored for bullying and territorial stress to minimize weight loss that can result from the inadvertent peer-induced decreased access to food and water. All animals should have sufficient access to resources, such as food and water receptacles, species-appropriate resting surfaces, and species-specific enrichment devices (toys and treats). FDA notes that adequate play and exercise are increasingly recognized as important to animal well-being.[62,63]

---

[59] Reinhart, V. & Reinhart A. (2002). *Comfortable quarters for laboratory animals* (10th ed.). Washington, D.C.: Animal Welfare Institute.

[60] National Research Council Institute of the National Academies (2006). *Guidelines for the humane transportation of laboratory animals*. Washington, DC: National Academies Press. Retrieved from http://www.nap.edu/openbook.php?record_id=11557&page=R1.

[61] See 21 CFR 58.43.

[62] See 9 CFR 3.8.

[63] Menor-Campos, D., Molleda-Carbonell, J., & Lopez-Rodriguez, R. (2011). Effects of exercise and human contact on animal welfare in a dog shelter. *Veterinary Record*, 169(16), 388.

A. Animal Quarantine and Conditioning

Animals should have an adequate amount of time for quarantine and stabilization[64] before study enrollment to allow for exclusion of animals with any disease or condition that might interfere with the purpose or conduct of the study.[65] Background levels of disease can adversely affect study data and should be minimized. For example, animals may have sub-clinical infections that can cause disease caused by the stress of surgery or dense housing. FDA also recommends initiating a program of socialization to minimize stress during the study; familiarity with handlers can reduce background stress, thus potentially minimizing additional confounding factors that could affect study results.

B. Food, Water, and Basic Husbandry

The testing facility must have standard operating procedures for the housing (including pest management), feeding, handling, and care of animals.[66] If contaminants that may affect study results are reasonably expected to be present in the feed and/or water, the limits should be stated in the protocol and an appropriate testing program described.

Sometimes large animals enrolled in chronic studies are transferred to a more typical agricultural setting where animals can graze on open pasture as a portion of their diet. In these cases, the testing facility should ensure that the animal's nutritional needs are met, and the pasture is well-maintained. Depending on the type and duration of the study, animals housed in agricultural settings may benefit from routine health checks, including fecal parasite checks and body condition scoring, to help avoid possible interference with study outcomes stemming from animal health issues.

IX. Records and Reports

A final study report must be prepared for animal studies, and any corrections or additions to the final study report must be in the form of a report amendment by the study director.[67] FDA recommends that the study director prepare the records and reports for the animal studies such that FDA can most efficiently evaluate the animal safety study.

The Agency recommends copies of raw data[68] be included as indexed secondary attachments to the final report (Appendix A - Example Organization of Animal Study Final Report). These should include the following when appropriate:

- any data used for statistical analysis;
- copies of case report forms;
- copies of individual animal medical records;
- full clinical pathology reports with the laboratory's reference ranges for each analyte; and

[64] Russell WMS, Burch, RL. The Principles of Humane Experimental Technique. London: Methuen & Co.; 1959. Special edition published by Universities Federation for Animal Welfare, 1992, available at http://altweb.jhsph.edu/pubs/books/humane_exp/het-toc.

[65] See 21 CFR 58.43, 58.90(c).

[66] See 21 CFR 58.90.

[67] 21 CFR 58.185(a)–(c).

[68] Consistent with 21 CFR 58.3(k), raw data means any laboratory worksheets, records, memoranda, notes, or exact copies thereof, that are the result of original observations and activities of a nonclinical laboratory study and are necessary for the reconstruction and evaluation of the report of that study.

- copies of all images (e.g., gross, microphotographs, angiograms, radiographs, CT, magnetic resonance imaging (MRI)).

FDA recommends that a detailed justification of the animal model be provided with the study report. This justification should include a rationale for selection of the animal model and a brief review of the scientific evidence supporting this model. In cases where an animal model has not been established or where the animal model has limited use, selection of an animal model should include consideration of anatomical and physiological differences and a detailed description of the animal model limitations.

The final report must include the locations where all specimens, raw data, and the final report are to be stored.[69] FDA also recommends dating all study observations. This helps to capture events accurately, which aids in the assessment of the inter-observational differences between study subjects.

Further, all raw data, documentation, protocols, final reports, and specimens (with certain exceptions) generated as a result of the animal study must be retained.[70] FDA may request copies of additional raw data[71] during the premarket review process depending on study results reported in the final report.

X. Preparation of Animal Study Reports for Medical Device Premarket Submissions

When preparing premarket submissions, FDA recommends that the sponsor include a high-level summary of the animal testing that was conducted to demonstrate device safety, which includes performance and handling. When a series of studies has been conducted, the sponsor should explain the effect of the animal studies on device development and the final device design. The sponsor should also describe all device design changes that were implemented during animal testing. Lastly, the sponsor should describe and justify any modifications to the device design that were made to accommodate the animal model. Bookmarked and linked PDF files are recommended for FDA premarket submissions whenever possible.

If your animal study was not conducted in compliance with 21 CFR Part 58, FDA recommends you include a statement in your submission explaining the reasons why the study was not in compliance with the GLP Regulations, as well as a detailed description of all deviations from the regulation. FDA recommends the statement include information that will help FDA reconstruct the study, explain any confounding variables, and demonstrate that authentic and complete test data have been collected and reported.

If multiple animal studies are being submitted, FDA recommends that sponsors present animal studies in chronological order so that the Agency can follow the device design history and *in vivo* safety profile from the first to the last study. Each final study report should include a table of contents that lists each supplementary report (e.g., in-life veterinarian report, pathology report),

[69] See 21 CFR 58.185(a)(13).
[70] 21 CFR 58.190(a).
[71] As defined under 21 CFR 58.3(k).

and if applicable, copies of raw data. An example organizational template for relevant content of an animal study report for regulatory submissions is provided in Appendix A.

The list below is intended as an example of the organization of a final report for recommended and required content.[72]

A. Executive Summary

This summary should also include the following information for each animal study:

- the rationale for the animal model;
- any differences between the animal model and humans that may have affected the study; and
- the specific animal study methodology used.

FDA recommends providing a table for each study that allows FDA to track individual parameters for animals through the study stages. This table should include the following information:

- unique animal ID(s) (e.g., USDA, institutional/testing facility, study cohort);
- allocation to study subgroups;
- endpoints for each study group, if applicable;
- the number of animals in each group;
- study duration;
- the device design iteration used;
- type(s) of procedure(s) performed;
- fate or disposition of each animal; and
- summary of study outcomes.

If animals are purchased with individual USDA IDs but then subsequently identified with an institutional ID and then further described by a group ID, this information should be clearly described and presented in the report so that a chain of custody of any individual test or control animal is possible.

B. Final Study Report

Each animal study must have a final report that is signed and dated by the study director.[73] The final study report for each animal study must include the information specified in the GLP Regulation, including but not limited to a description of all circumstances that may have affected the quality or integrity of the data.[74] Any change to the signed and dated study protocol must be reported as an amendment and attached to the final study report.[75]

[72] This outline does not include all of the requirements of 21 CFR 58.185. For more information on required content necessary to be in compliance with Part 58 reporting requirements, please see 21 CFR 58.120 and 21 CFR 58.185.
[73] See 21 CFR 58.185(b).
[74] See 21 CFR 58.185(a)(9).
[75] See 21 CFR 58.120(b).

The report must include a quality assurance statement that specifies the dates and phases of study during which inspections were made and reported to management and the study director.[76] This can help provide documentation that the study was conducted per the protocol such that the methods, procedures, recording, and reporting of raw data are truthful and accurate, and that the facilities, equipment, personnel, methods, practices, records, and controls are in conformance with the GLP Regulations.[77]

The final study report must also include the signed and dated reports of each individual scientist or other professional involved in the study.[78] The following section provides recommendations on commonly included reports from other professionals involved in the study.

(1) In-Life Veterinarian Report

One of the advantages of animal studies is the opportunity to assess the clinical response to a device and/or procedure. Study animals are maintained in a controlled environment, which facilitates in-life observations. In contrast, the environment in clinical studies may be more difficult to control, which can lead to loss of follow-up data. Therefore, FDA recommends that the final study report include an in-life report from the study veterinarian that summarizes all key events for each animal enrolled in the study. The report should include copies of the individual animal medical records maintained in standard POVMR format, including any provision of veterinary care.

(2) Pathology Report

The pathology report represents the pathologist's best judgment regarding relevant tissue changes and their interpretation in the context of the study. The pathology report should include a table that correlates the gross pathological findings with the histopathologic findings in each animal/tissue. It should include a narrative that describes and interprets all observed adverse tissue findings. The pathologist should justify any statements regarding whether any adverse outcomes are or are not device-related with appropriate information from gross pathology data, histopathology data, and in-life assessments, including the relevance of any extenuating circumstances (e.g., concurrent disease).

FDA recommends that the pathologist describe any non-standard methods used to collect tissues as well as the methods of fixation, cutting, and staining.[79] The pathologist should describe the sectioning methods, noting the tissue type and device orientation as appropriate. Diagrams may be helpful in illustrating device location and how the device should be sectioned for histology.

The pathology report should include high-resolution, properly illuminated, color, gross, and photomicrograph images of all abnormal findings and corresponding normal tissues from all test and control animals. The gross images and photomicrographs should be labeled with a minimum of the animal ID, study group ID, size marker, and date. All photomicrographs should also indicate tissue section, magnification, scale, stain, and any other important identifiers. The accompanying legend should fully describe the pathological changes in the tissues.

---

[76] See 21 CFR 58.35(b)(7), 58.185(a)(14).
[77] See 21 CFR 58.35(b)(1)–(7).
[78] 21 CFR 58.185(a)(12).
[79] Dettmeyer, R. (2013). *Forensic histopathology: Fundamentals and perspectives*. Heidelberg, Germany: Springer.

(3) Other Reports/Forms

Depending on the nature of the study, the final study report should also include any other reports that are signed and dated by contributing scientists. Examples include but are not limited to clinical pathology, radiology, or other forms of imaging, and behavioral studies. Clinical pathology reports should identify the laboratory performing the tests, the laboratory's reference range values, and the clinical pathology data for each animal. A table, presenting clinical pathology results by animal and time point and including laboratory reference ranges, can help FDA interpret larger data sets. If applicable, FDA recommends sponsors provide copies of completed case report forms, and surgical/anesthesia monitoring forms in the final study report, to aid in the interpretation of study results.

The study director should ensure that the report details the source of the animals.

Additionally, the final study report should include documentation of protocol approval by an Institutional Animal Care and Use Committee (IACUC)[80] before initiation of the study or, if conducted outside the United States, an equivalent approved protocol.

[80] See 9 CFR 2.31.

Appendix A - Example Organization of Animal Study Final Report

The list below is intended as an example of the organization and content of a final report This example does not include all the requirements of 21 CFR 58.185. For more information on required content necessary to be in compliance with 21 CFR Part 58 reporting requirements, see 21 CFR 58.120 and 21 CFR 58.185.

I. A coversheet for each animal study that includes:
   - A. Title of the report
   - B. Report identification numbers (as applicable):
     1. IACUC/ethics committee protocol number
     2. Study director protocol number(s)
     3. Testing facility protocol number(s), if applicable
   - C. Contact information (i.e., mailing address, street address, city, state, country, and ZIP code for each contact) for the following entities:
     1. Sponsor
     2. Sponsor representative, if different than sponsor
   - D. Testing facility name and address
   - E. Study initiation and completion dates
   - F. Final report signatures with date of signature:
     1. Study director
     2. QAU
     3. Key personnel

II. Table of contents (preferably hyperlinked)

III. List of abbreviations and definitions

IV. Copy of the approved protocols, including:
   - A. Final GLP study protocol
   - B. IACUC-approved protocol with documentation of approval (if different from the GLP study protocol)

V. Final test report that includes:
   - A. Overview/executive summary of animal study:
   - B. Body of test report
     1. Study objectives and endpoints with acceptance criteria
     2. Study time point(s)
     3. Animal model and number of animals enrolled
     4. Characterization of test and control articles:
        - a. Design iteration of device used, i.e., how design iteration compares to the final device version intended for clinical use in humans
        - b. Referenced serial or model numbers

5. Description of methods used, including surgical/procedural approach and technique, imaging, animal monitoring, wellness interventions (as applicable), necropsy, and histology as appropriate
6. Study deviations and amendments
7. Statistical analysis plan
8. Study results, organized by objective
    a. Deployment/procedural results, including system/equipment compatibility
    b. Impact of animal on device (e.g., structural integrity)
    c. Biologic (physiological) response to the device
    d. Imaging
    e. Adverse events
9. Conclusions:
    a. Comparison with controls
    b. Success in meeting acceptance criteria
    c. Discussion of study limitations

C. Quality assurance compliance statement

VI. Indexed attachments, if applicable:

A. Copies of raw data and individual test reports used to draw study conclusions
B. Signed and dated contributing scientist(s) reports (i.e., interventionist, surgeon, radiologist, clinical veterinarian, clinical pathologist, pathologist, neuro/behaviorist).
C. Animal medical records, including baseline and interim health examinations
D. Surgery and anesthesia monitoring forms
E. Imaging reports
F. Clinical pathology results, with reference ranges
G. Completed case report forms
H. Animal vendor reports/invoices