#187

Guidance for Industry

Regulation of Intentionally Altered Genomic DNA in Animals

Draft Guidance

(This guidance is a revision of Guidance #187, "Regulation of Genetically Engineered Animals," which has been revised to update information concerning the products of different technologies used to produce such animals, and to provide new weblinks.)

Submit comments on this draft guidance by the date provided in the *Federal Register* notice announcing the availability of the draft guidance. Submit electronic comments to http://www.regulations.gov. Submit written comments to the Division of Dockets Management (HFA-305), Food and Drug Administration, 5630 Fishers Lane, Room 1061, Rockville, MD 20852. All comments should be identified with the Docket No. FDA-2008-D-0394.

For further information regarding this document, contact Laura R. Epstein, Center for Veterinary Medicine (HFV-1), Food and Drug Administration, 7519 Standish Place, Rockville, MD 20855, 301-796-8558, email: Laura.Epstein@fda.hhs.gov.

Additional copies of this guidance document may be requested from the Policy and Regulations Staff (HFV-6), Center for Veterinary Medicine, Food and Drug Administration, 7519 Standish Place, Rockville, MD 20855, and may be viewed on the Internet at either http://www.fda.gov/AnimalVeterinary/default.htm or http://www.regulations.gov.

U.S. Department of Health and Human Services
Food and Drug Administration
Center for Veterinary Medicine
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Table of Contents

Guidance for Industry

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This draft guidance, when finalized, will represent the current thinking of the Food and Drug Administration (FDA or Agency) on this topic.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 responsible for this guidance as listed on the title page.

I. Introduction and Background

FDA is issuing this draft revised Guidance for Industry to clarify its approach to the regulation of intentionally altered genomic DNA in animals. This guidance addresses animals whose genomes have been intentionally altered using modern molecular technologies, which may include random or targeted DNA sequence changes including nucleotide insertions, substitutions, or deletions, or other technologies that introduce specific changes to the genome of the animal.[1,2,3] This guidance applies to the intentionally altered genomic DNA in both the founder animal in which the initial alteration event occurred and the entire subsequent lineage of animals that contains the genomic alteration.

Recombinant DNA (rDNA) technology has been used for the past 40 years to intentionally alter traits in microorganisms, plants, and animals (Cohen and Boyer 1973). Various agencies across the US government (USG) have provided guidance and regulation to affected stakeholders

[1] FDA used the term "genetically engineered" (GE) to describe the animals within the scope of current Guidance for Industry #187. The term "GE" does not suit the discussion in this revised draft guidance because this draft guidance's scope includes animals whose genomes have been intentionally altered with new technologies. The term "transgenic" is also not used for the same reason, except for citation of earlier documents.

[2] In Draft Guidance for Industry #236, "Regulation of Mosquito-Related Products," FDA has proposed to clarify that the phrase "articles (other than food) intended to affect the structure or any function of the body of man or other animals" does not include articles intended to prevent, destroy, repel, or mitigate mosquitoes for population control purposes. Instead, such products are pesticides regulated by the Environmental Protection Agency (EPA) (http://www.fda.gov/downloads/AnimalVeterinary/GuidanceComplianceEnforcement/GuidanceforIndustry/UCM533600.pdf).

[3] The term "modern molecular technologies" does not include selective breeding or other assisted reproductive technologies, including random mutagenesis followed by phenotypic selection.

describing the regulation of these altered organisms and the products of those alterations. Historically, recombinant DNA (rDNA) techniques involved splicing DNA sequences from various sources and introducing them into animals[4] via techniques that resulted in random integration events. Animals whose genomes have been intentionally altered by rDNA technology have been produced since the early 1980s when Brinster et al. (1981)[5] and Palmiter et al. (1982)[6] reported on the development of mice altered in this manner. Not long thereafter, Hammer et al. (1985)[7] demonstrated that rDNA techniques could be used to intentionally alter the genomes of rabbits and pigs.

More recently, new technologies have emerged that are intended to alter the genomes of various organisms, including animals. Some of these include the use of "nucleases" or "genome editing technologies" including engineered nuclease/nucleotide complexes such as zinc finger nucleases (ZFN), transcription activator-like effector nucleases (TALENs), and the clustered regulatory interspersed short palindromic repeats (CRISPR) associated systems.[8] These nucleases are intended to introduce alterations at specific sites in the genome, rather than the more random changes associated with rDNA technology. The process of producing these targeted DNA sequence alterations is often referred to as "genome editing." We anticipate that other technologies intended to alter genomic DNA will arise over time.

Intentional genomic alterations may be heritable or non-heritable (e.g., those alterations intended to be used as gene therapy). Although much of this guidance will be relevant to non-heritable intentionally altered genomic DNA, this guidance primarily addresses heritable intentionally altered genomic DNA. For non-heritable genomic alterations, we recommend that sponsors contact the agency directly for further information.

This guidance is intended to clarify our requirements and recommendations for producers and developers ("sponsors," "you") of animals with intentionally altered genomic DNA. We note that the regulated article for such animals is the intentionally altered genomic DNA of the

[4] For purposes of this guidance, "animals" refers to non-human animals.
[5] Brinster, R.L., et al . (1981) Somatic expression of herpes thymidine kinase in mice following injection of a fusion gene into eggs. *Cell* 27: 223-231
[6] Palmiter, R.D., et al. (1982) Dramatic growth of mice that develop from eggs microinjected with metallothionein-growth hormone fusion genes. *Nature* 300: 611-615.
[7] Hammer, R. E. et al. (1985) Production of transgenic rabbits, sheep, and pigs by microinjection. *Nature* 315: 680-683.
[8] For a more complete description of genome editing techniques, see *Nature* Special Supplement Vol. 528, 2 December 2015.

animal. As a short hand in this guidance document, we sometimes refer to regulation of the article (i.e., the altered genomic DNA) in such animals as regulation of the altered animal.

Some animals with intentional alterations to their genomes are intended to produce medical and other products, such as human drugs or medical devices that are subject to regulation by other FDA centers. Where sponsors have developed animals that are intended to produce human medical products that are separately regulated by FDA's Center for Drug Evaluation and Research (CDER), Center for Biologics Evaluation and Research (CBER), or Center for Devices and Radiological Health (CDRH), or a human food additive regulated by FDA's Center for Food Safety and Applied Nutrition (CFSAN), CVM will work closely with the other FDA Centers that regulate those products derived from these animals to ensure that our oversight is complementary and not unnecessarily duplicative.

In addition to this guidance, there are other guidelines and laws that may apply to animals with intentionally altered genomes:

- Existing guidances and other documents prepared by other FDA Centers including the Points to Consider in the Manufacture and Testing of Therapeutic Products for Human Use Derived from Transgenic Animals (1995) http://www.fda.gov/downloads/BiologicsBloodVaccines/GuidanceComplianceRegulatoryInformation/OtherRecommendationsforManufacturers/UCM153306.pdf, and

- Source Animal, Product, Preclinical, and Clinical Issues Concerning the Use of Xenotransplantation Products in Humans (2003) http://www.fda.gov/downloads/BiologicsBloodVaccines/GuidanceComplianceRegulatoryInformation/Guidances/Xenotransplantation/ucm092707.pdf.

- Medical Devices Containing Materials Derived from Animal Sources (Except for In Vitro Diagnostic Devices) - Draft Guidance for Industry and Food and Drug Administration Staff (2014) http://www.fda.gov/MedicalDevices/DeviceRegulationandGuidance/GuidanceDocuments/ucm381379.htm

- Federal laws, regulations, and guidelines for the humane care, handling, and slaughter of animals, as well as guidelines in place at your institution or establishment;

- Applicable Federal, State, local and tribal laws, regulations, and guidelines addressing environmental safety, including those NIH guidelines that apply to your institution or establishment;

- Applicable Federal, State, local and tribal laws, regulations, and guidelines pertaining to the importation, interstate movement, or release of wildlife;

- Federal laws, regulations, and guidelines governing the import or export of animals across US boundaries; and

- Other applicable Federal, State, or local laws, regulations and guidelines.

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. Statutory and Regulatory Authority

A. The Regulated Article

FDA's authority over new animal drugs comes from the Federal Food, Drug, and Cosmetic Act (FD&C Act) (21 U.S.C. 321 et seq.). The definition of a drug, in section 201(g) of the FD&C Act, includes "articles intended for use in the diagnosis, cure, mitigation, treatment, or prevention of disease in man or other animals"; and "articles (other than food) intended to affect the structure or any function of the body of man or other animals." The definition of "new animal drug" in section 201(v) of the FD&C Act includes any drug intended for use in animals that is not generally recognized as safe and effective for use under the conditions prescribed, recommended, or suggested in the drug's labeling, or that is so recognized but has not been used to a material extent or for a material time.

Generally under the FD&C Act, a new animal drug is "deemed unsafe" under section 512(a)(1) unless FDA has approved a new animal drug application (NADA) for its intended use, unless the drug is only for investigational use and conforms to specified exemptions for such use under an Investigational New Animal Drug (INAD) exemption (21 U.S.C. 360b(a)(1), (a)(3)), or unless

the drug is used in conformance with regulations promulgated under sections 512(a)(4) or (5) of the FD&C Act (21 U.S.C. 360b(a)(4) or (5)). An unsafe new animal drug is "adulterated" under section 501(a)(5) of the FD&C Act.

For purposes of this guidance, "altered genomic DNA" refers to the portion of an animal's genome that has been intentionally altered. Unless otherwise excluded, e.g., certain mosquito-related products[9], the altered genomic DNA in an animal is a drug within the meaning of section 201(g) of the FD&C Act because such altered DNA is an article intended to affect the structure or function of the body of the animal, and, in some cases, intended for use in the diagnosis, cure, mitigation, treatment, or prevention of disease in the animal.[10] Altered genomic DNA may result from random or targeted DNA sequence changes including nucleotide insertions, substitutions, or deletions, or other technologies that introduce specific changes to the genome of the animal. Non-heritable altered genomic DNA that is intended to affect the structure or function of the resulting animal or to cure, mitigate, or treat a disease in the animal also meets the drug definition. As noted previously, this guidance primarily addresses heritable genomic alterations.

A specific DNA alteration is an article that meets the definition of a new animal drug at each site in the genome where the alteration (insertion, substitution or deletion) occurs. The specific alteration sequence and the site at which the alteration is located can affect both the health of the animals in the lineage and the level and control of expression of the altered sequence, which influences its effectiveness in that lineage.[11] Therefore, in general, each specific genomic alteration is considered to be a separate new animal drug subject to new animal drug approval requirements. If a sponsor wishes to introduce multiple genomic alterations resulting in one final animal lineage, we recommend that the sponsor contact the agency to discuss regulatory options and the kinds of scientific questions that would have to be addressed in an application. During

[9] In Draft Guidance for Industry #236, "Regulation of Mosquito-Related Products," FDA has proposed to clarify that the phrase "articles (other than food) intended to affect the structure or any function of the body of man or other animals" does not include articles intended to prevent, destroy, repel, or mitigate mosquitoes for population control purposes. Instead, such products are pesticides regulated by the Environmental Protection Agency (EPA) (http://www.fda.gov/downloads/AnimalVeterinary/GuidanceComplianceEnforcement/GuidanceforIndustry/UCM533600.pdf)

[10] FDA does not intend to regulate altered genomic DNA that meets the definition of a veterinary biologic and is regulated by the Animal and Plant Health Inspection Service (APHIS) of the United States Department of Agriculture (USDA). 21 CFR 510.4.

[11] Because animals with intentionally altered genomes being used for commercial purposes are likely to be descendants of the initially altered animal, rather than the initially altered animal itself, the NADA safety and effectiveness evaluations should be focused on a generation as close to those animals to be used for commercial purposes as possible. Sponsors will need to demonstrate that following approval and use in commerce, the altered genotype and/or phenotype are stably maintained in a representative sample of animals. 21 CFR 514.1(b)(5).

the investigational phase, one INAD file may be established for multiple genomic alterations, and the file may contain information on investigational animals that contain different numbers or types of intentional genomic alterations, including those occurring at different locations of the genome, prior to selecting the lineage of animals with intentionally altered genomes intended for commercialization.

Each new animal drug approval covers all animals containing the same genomic alteration(s) (the regulated article or new animal drug) derived from the same alteration event(s). Animals containing the genomic alteration as a result of breeding between an intentionally altered animal and its non-intentionally altered counterpart animal are covered by the new animal drug approval.

B. Enforcement Discretion for INAD or NADA Requirements for Certain Animals With Intentionally Altered Genomic DNA

Although, unless otherwise excluded,[12] animals with intentionally altered genomes are subject to premarket approval requirements, in certain circumstances, based on the risk(s) they pose, we intend to exercise enforcement discretion with regard to INAD and NADA requirements for certain of these animals (that is, in specified circumstances, we do not intend to enforce the INAD and NADA requirements, including those described in this guidance). For example, FDA has not and does not intend to enforce INAD and NADA requirements for: (1) animals of nonfood-producing species whose genomes have been intentionally altered that are regulated by other government agencies or entities, such as insects whose genomes have been intentionally altered that are under APHIS oversight; and (2) animals of nonfood-producing species whose genomes have been intentionally altered that are raised and used in contained and controlled conditions such as laboratory animals with intentionally altered genomes used in research institutions. Although we generally intend to exercise enforcement discretion with regard to INAD and NADA requirements for such animals, we retain the discretion to take enforcement action if we learn of safety concerns associated with them.

Based on evaluation of risk factors, we may exercise enforcement discretion over INAD and NADA requirements for additional kinds or uses of nonfood-producing species of such animals, as we did after reviewing information about *Zebra danio* aquarium fish genetically engineered to

[12] See footnote 9 regarding mosquito-related products.

fluoresce in the dark (GloFish) (http://www.fda.gov/animalveterinary/developmentapprovalprocess/geneticengineering/geneticallyengineeredanimals/ucm413959.htm) and *Int'l Ctr. for Tech. Assessment v. Thompso*n, 421 F. Supp. 2d 1 (D.D.C. 2006)). We may also modify our approach with respect to INAD and NADA requirements for other kinds or uses of animals based on our evaluation of risk factors.

When FDA reviews and approves an INAD or NADA, it complies with the requirements of the National Environmental Policy Act (NEPA), including a review of environmental risks, if any, where required. When FDA exercises its enforcement discretion over the INAD or NADA requirements, no NEPA review would take place. As a result, the potential for environmental risks are among the factors we intend to consider in determining whether to exercise enforcement discretion.

Among the issues we intend to consider when determining whether to exercise enforcement discretion are whether:

- There is anything about the article itself that poses a human, animal, or environmental risk. For example, does the altered genomic DNA contain sequences that can cause human or animal disease either intrinsically or by recombination?

- For environmental releases, does the animal with intentionally altered genomic DNA pose any more of an environmental risk than its counterpart?

- There are concerns over the disposition of animals with intentionally altered genomic DNA that could pose human, animal, or environmental risks.

- There are any other safety questions that have not been adequately addressed by the sponsor.

You may contact CVM for further information on whether some form of enforcement discretion might be warranted for the intentionally altered genomic DNA in your animal. Although we may decide to exercise enforcement discretion with respect to regulatory requirements for certain animals with intentionally altered genomic DNA after reviewing information about potential risks, this decision may be reevaluated if we become aware of any changes in the animals' risk profiles. Such reevaluation could lead us to conclude that the intentionally altered genomic

DNA in these animals should be subject to FDA enforcement action until a full NADA has been approved.

III. Investigational Use of Animals With Intentionally Altered Genomic DNA

As noted earlier, under the FD&C Act, in general a new animal drug is "deemed unsafe" unless the FDA has approved an application for that particular use (21 USC 360b), unless it is for investigational use and is subject to an exemption from the approval requirement that conforms to FDA regulations or unless it is used consistent with regulations promulgated under sections 512(a)(4) and (5) of the FD&C Act. 21 USC 360b(a)(3), (4), (5), (j).

FDA regulations concerning investigational use of new animal drugs are codified at section 511.1 in Title 21 of the Code of Federal Regulations (21 CFR 511.1). These regulations cover shipments in interstate commerce of new animal drugs for tests *in vitro* and in laboratory research animals (21 CFR 511.1(a)) and for clinical investigation in animals (21 CFR 511.1(b)). The INAD requirements in 21 CFR 511.1(b) apply to investigational animals whose genomes have been intentionally altered. Further, the development of such animals constitutes clinical investigation because it involves studying the effectiveness of the drug in the target species and the effects of the intentionally altered genomic DNA, including those of its expression product(s), if any, on the animal containing it.

In general, the INAD regulations specify labeling and record-keeping requirements, animal disposition, and conditions under which food[13] from animals used for clinical investigations under section 511.1(b) can be introduced into the food supply. Section 511.1(b) also requires that prior to shipping a new animal drug for clinical tests, a sponsor must submit a Notice of Claimed Investigational Exemption for a New Animal Drug (INAD Notice) containing specified information.

We strongly recommend that you contact CVM early in the development process to determine what information should be submitted to CVM and the appropriate file type to utilize. Certain types of early information can be submitted without the establishment of an INAD file and FDA would consider such information to be confidential. You should note that establishment of an INAD file will result in an annual sponsor user fee unless you are eligible for a waiver. See

[13] The term "food" includes food for humans and animals.

Draft Revised Guidance for Industry #170, "Animal Drug User Fees and Fee Waivers and Reductions," http://www.fda.gov/downloads/AnimalVeterinary/GuidanceComplianceEnforcement/GuidanceforIndustry/UCM052494.pdf. CVM will work with you to determine the appropriate timeframe to open an INAD file, how best to develop the data and information that will be needed for an NADA, and how to provide such data and information to CVM for evaluation and comment. We recommend that the early information you provide to CVM include how you intend to develop the animal whose genome has been intentionally altered including the species of animal to be under study, the altered gene(s) or region of the genome, and the intention of the genomic alteration, including any gene product(s) that may be produced.

In most cases, you will need to submit an INAD Notice prior to shipping any animals with intentionally altered genomic DNA. Also, if you wish to introduce any food derived from investigational animals into the food supply, you must get prior FDA authorization to do so through the INAD process. 21 CFR 511.1(b)(5). We recommend that prior to making a request for such authorization, you schedule a teleconference or in-person meeting with us to determine which classes of investigational animals may be suitable for consideration for food use.

We encourage you to contact CVM if you have questions about submitting a request to establish an INAD file. Once we have established an INAD file, you will receive a letter assigning a unique INAD number to that file. This unique identifier (which we refer to as a file number) should be used for all subsequent communications with us regarding that INAD file. As previously stated, an INAD file can encompass animals derived from multiple alteration events, even though an NADA would generally only cover animals derived from a single alteration event.

We recommend that you schedule a meeting with us (either in-person or via teleconference) before an INAD file has been established or immediately thereafter. In that meeting, you can acquaint us with the nature of the animal under development and the intended use. We can then provide you with more specific information on the kinds of regulatory responsibilities you have under an INAD, and the nature of the regulatory decisions we can make during the investigational phase of research, including the following:

A. Shipping and Labeling Investigational Animals and Their Products

During the investigational phase of the development of a lineage of animals with intentionally altered genomic DNA, the animals may need to be moved from the initial laboratory or barn to other locations within the sponsor's facilities, or to other investigators at the same facilities or off-site. If the investigational animals or products derived from them are shipped to other investigators, it is important to ensure that those individuals/entities receiving the investigational animals or their products use them only for research purposes. All shipments must bear labeling that clearly identifies that edible products derived from investigational animals are not to be used for food without prior authorization from FDA. 21 CFR 511.1(b)(1)-(5). We recommend that you contact us to determine the appropriate labeling for the particular investigational animal or its products.

B. Animal Disposition

A primary goal during the investigational phase of development of the animal with an intentionally altered genome is to ensure that edible products from these investigational animals do not enter the food supply without prior FDA authorization. Edible products include, but are not limited to milk, honey, eggs, muscle tissue, as well as other tissues such as liver, kidney, skin, and fat. We encourage you to provide a disposition plan for all classes of investigational animals and animal products. We recommend that all surplus investigational animals and their biological products be disposed of by incineration, burial, or composting, and that appropriate records be kept of animal identification and disposition. In some special cases, alternative disposition may be appropriate provided that our safety concerns are met (see Section III.C). 21 CFR 511.1 (b)(5).

C. Investigational Food Use Authorizations

If you wish to introduce investigational animals or animal products into the food supply, you must request an Investigational Food Use Authorization (21 CFR 511.1(b)(5)). For those animals subject to slaughter inspection by the USDA Food Safety and Inspection Service (FSIS), we will inform FSIS if our safety concerns are met and we grant you an Investigational Food Use Authorization.

FSIS has oversight of meat, poultry, fish of the Order Siluriformes, and egg products, and tests for animal drug residues above tolerance levels (maximum allowable amounts). FDA and FSIS have enjoyed longstanding open communications during the drug approval process, and are discussing how to adapt and improve these existing procedures to fully accommodate the needs of both agencies in addressing animals with intentionally altered genomes intended to go into the food supply.

We recommend that prior to making a food-use authorization request, you schedule a teleconference or in-person meeting with us to determine which classes of investigational animals may be suitable for consideration for food use and the nature and extent of data you will need to provide for us to make that determination.

D. Environmental Considerations

Actions on INADs are considered major federal actions under the NEPA, and as such may require preparation of an environmental assessment (EA) and a finding of no significant impact (FONSI) (21 CFR 511.1(b)(10), 21 CFR 25.15) or an environmental impact statement (EIS) (21 CFR 25.22).

Through the preparation of an EA/FONSI or EIS, FDA will examine the potential for environmental impacts, including the potential for inadvertent release or escape of the animal with an intentionally altered genome and/or its products into the environment, and whether certain measures may mitigate any potential significant impacts that would adversely affect the human environment. Additionally, sponsors may be subject to applicable environmental requirements with respect to runoff from animal production facilities and land receiving animal waste under the Clean Water Act. 33 U.S.C. 1251 et. seq. and other statutes.

In order to determine the nature and extent of the potential for environmental risks that your investigational animals may pose, we recommend that you contact us early in the development of your animals with intentionally altered genomes so that we can determine the scope of this environmental assessment. These early discussions can help to focus your environmental assessment under the NADA component as well.

Categorical exclusion from the requirement to prepare an EA may be possible under 21 CFR 25.33(e) for investigational studies on certain animals, if you can provide sufficient information for us to conclude that extraordinary circumstances will not exist (21 CFR 25.21). This should

include sufficient information on the animals whose genomes have been intentionally altered and their containment to allow us to conclude that use and disposal of any investigational animals or their products would not have a significant impact on the human environment. We recommend that you contact us early to discuss whether your INAD may be eligible for a categorical exclusion under 21 CFR 25.33(e), or whether extraordinary circumstances may exist that would require an EA/FONSI or EIS.

IV. FDA Approval of Animals With Intentionally Altered Genomic DNA

A. Overview

Other than for investigational uses, section 512(a)(1) of the FD&C Act (21 U.S.C. 360b(a)(1)) requires that a new animal drug be the subject of an approved NADA based on a demonstration that it is safe and effective for its intended use.

When submitting an NADA, you should include the results of any investigations you conducted under an INAD. We will evaluate the NADA to determine whether you have demonstrated that the new animal drug is safe and effective for its intended use. To demonstrate effectiveness of an article intended to express an extractable protein (e.g., for use as a human biological product), generally you would simply have to show that the expression product is in fact expressed in the animal. To demonstrate effectiveness of an article intended to alter a characteristic of the resulting animal, in general you would have to show that the animal whose genomic DNA had been intentionally altered had the claimed altered characteristic (e.g., that its rate of growth was as claimed or that it was indeed resistant to a disease).

The agency is interested in increasing the transparency of its deliberations and actions. In particular, we intend to seek input from experts and the public where there is significant public interest in an issue, and FDA believes the public may have relevant data or information to contribute.

Additionally, as is the case for all NADAs, after completion of an NADA, the agency will post a summary of the information in the NADA file, including information used to assess safety (to the animal and for food consumption, if appropriate) and in support of the claims made by the sponsor. 21 CFR 514.11(e).

B. New Animal Drug Application Requirements

Section 512(b)(1) of the FD&C Act describes the information that must be submitted to FDA as part of an NADA. These statutory requirements are further explained and expanded upon in the regulations for new animal drug applications, 21 CFR 514.1.

The application of some of the statutory and regulatory requirements for new animal drug applications to animals with intentionally altered genomic DNA may not be obvious. For example, it may not be obvious how the requirement to provide a full list of the articles used as components of a drug as described in 512(b)(1)(B) of the FD&C Act and 21 CFR 514.1(b)(4) of the NADA regulations applies to such animals. Therefore, this section of the guidance document provides a brief summary of the NADA requirements in 21 CFR 514.1 and describes how these requirements may be addressed for applications submitted for animals with intentionally altered genomic DNA. Section IV.C describes our recommendations for how to present this information in the structure of an NADA submission to meet these regulatory requirements and the statutory requirements of safety and effectiveness.

1. Identification (21 CFR 514.1(b)(1))

   Section 514.1(b)(1) requires that certain identifying information be provided including the nature of the application (i.e., original or supplemental application), the name and address of the applicant, date of application, and the trade and/or chemical name of the new animal drug.

   The information that should be provided to satisfy this requirement for an application for a lineage of animals with an intentionally altered genome is similar to that provided for a conventional new animal drug. In the case of such an application, the "trade and/or chemical name of the new animal drug" should be described by identifying the animal, its ploidy and zygosity, the name and intended function of the altered genomic DNA, and the number and characterization of the site(s) of alteration[14], including unintended (e.g., off-target) alterations, as well as the intended use of the resulting lineage of animals. For a more

[14] The term "site of alteration" in this document refers to the genomic location in the animal with the altered genomic DNA either chromosomally integrated or maintained as an extrachromosomal element. In general, we are most interested in characterizations that are performed in animals close to commercialization.

complete description of how we recommend you present this information in the NADA submission, please see Section IV.C., Steps 1, 2, and 3.

We consider this component to be critical to the structure and content of an NADA submission and so encourage you to consult with us on this topic as early as possible in the development of these animals, for example, as an early part of the INAD process.

2. Table of Contents and Summary (21 CFR 514.1(b)(2))

Section 514.1(b)(2) requires that an NADA include a table of contents which identifies the data and other material submitted, and a well-organized summary of information that (1) describes the chemistry of the new animal drug, and (2) describes the clinical purpose and provides a summary of laboratory and clinical studies.

For more information on how we recommend you present this information in the NADA submission, please see Section IV.C., Steps 1, 2, and 3.

3. Labeling **(**21 CFR 514.1(b)(3))

Section 514.1(b)(3) requires that an NADA include three copies of each piece of labeling to be used for the new animal drug.[15]

In the context of animals with intentionally altered genomic DNA, this includes labels and other written, printed information (i.e., labeling) that will accompany the animals. Labeling should include a summary description of the article, the animal into which the article is introduced (e.g., common name/breed/line; genus and species), the name of the resulting animal lineage, and the intended use of the animals containing the article. Where the labeling for an animal whose genome has been intentionally altered contains animal care or safety information (e.g., husbandry or containment), we recommend that the labeling accompany the animal throughout all stages of its lifecycle. We recommend that you contact CVM for further information regarding the required labeling for such animals.

[15] This discussion pertains to new animal drug labeling requirements, not labeling requirements for food derived from animals whose genomes have been intentionally altered.

4. Components and Composition **(**21 CFR 514.1(b)(4))

Section 514.1(b)(4) requires that an NADA include (1) a list of all articles used as components of the drug product; (2) a statement of composition of the drug product; and (3) a complete description of the fermentation of antibiotic drug substances.

For animals whose genomes have been intentionally altered, (3) would not be relevant. The information described in (1) and (2) should encompass the molecular characterization of the article. It should enable us to determine whether the article contains any potentially mobilizable DNA sequences, and whether sequences are present that encode pathogens, toxicants, allergens, or substances likely to dysregulate the growth control of cells, tissues, or organs, except by explicit design. We would expect that such information would describe the source, identity, purity, and functionality of the introduced article. For a more complete description of how we recommend you present this information in the NADA submission, please see Section IV.C., Steps 2 and 3.

5. Manufacturing Methods, Facilities, and Controls (21 CFR 514.1(b)(5))

Section 514.1(b)(5) requires that an NADA include a detailed description of the methods used in and the facilities and controls used for the manufacturing, processing, and packing of the new animal drug.

For animals with intentionally altered genomes, this information should encompass:

- the method by which the alteration was introduced into the initial animal, including whether the initial animal whose genome was intentionally altered was chimeric;

- the breeding strategy used to produce the lineage progenitor. (A lineage progenitor is the animal whose genome has been intentionally altered from which subsequent animals used for commercial purposes are derived); and

- full characterization of the site of intentional alteration, any unintended alterations (e.g., off-target alterations, unanticipated insertions, substitutions, or deletions) and the alterations persisting once the genome has been stabilized in animals contributing to the lineage of animals to be commercialized, including the number and orientation of any

introduced DNA sequences, if applicable. In particular, we recommend that you evaluate whether there are any unintended interruptions of a coding or regulatory region.

Information submitted to satisfy the requirements for finished product analytical controls and a stability program should include information demonstrating the durability of the genotype and phenotype—that is, whether the article is stably inherited, and the phenotype is consistent and predictable. This should include developing a sampling plan.

For genotypic durability, we recommend that you use the results of studies demonstrating that the altered genomic DNA is stably inherited. For the phenotypic durability portion of the plan, we recommend that you submit data on the consistency of the expressed trait (based on the intended use) over multiple generations. We recommend that, where feasible, you gather data on inheritance from at least two generations, preferably more, and recommend that at least two of the sampling points be from non-contiguous generations (e.g., F_1 and F_3).

Your plan should include a method of identity with sufficient discrimination to determine (1) whether a given animal contains the altered genomic DNA, and (2) whether the altered genomic DNA has significantly changed from that which was evaluated in the NADA (i.e., a detection method for your animal and regulated article). For a more complete description of how we recommend you present this information in the NADA submission, please see Section IV.C., Steps 2, 3, and 5. We recommend that you consult with us on developing these plans.

6. Samples **(**21 CFR 514.1(b)(6))

Section 514.1(b)(6) requires that samples of the new animal drug and articles used as components and information concerning them be submitted to CVM if requested.

This requirement applies to NADAs for animals whose genomes have been intentionally altered as it does to conventional new animal drug applications. Sponsors are encouraged to contact CVM to determine what samples (such as a genomic sample containing the article) should be provided.

If FDA establishes a tolerance for the new animal drug, FDA will notify FSIS and provide it with a summary of the information and evaluation upon which it based the tolerance, and a method of analysis to be used to enforce the tolerance. FDA and FSIS are discussing how to

adapt and improve existing communication procedures so that they will fully accommodate the needs of both agencies in addressing animals whose genomes have been intentionally altered and that are intended to go into the food supply.

7. Analytical Methods for Residues **(**21 CFR 514.1(b)(7))

Section 514.1(b)(7) requires that an NADA include method(s) and data to enable determination of residues of the new animal drug in food-producing animals, except when data or other adequate information establish that it is not reasonable to expect the new animal drug to become a component of food at concentrations considered unsafe.

The information that should be provided to satisfy this requirement for an application for animals whose genomes have been intentionally altered includes a method of detection that can be used to identify the altered genomic DNA in the resulting animals.

8. Evidence to Establish Safety and Effectiveness **(**21 CFR 514.1(b)(8))

Section 514.1(b)(8) requires that an NADA include data and information to permit evaluation of the safety and effectiveness of the new animal drug product for the use as suggested in the proposed labeling. Section 21 CFR 514.1(b)(8)(iv) also requires that sponsors supply all information relevant to safety and effectiveness for a new animal drug, favorable and unfavorable.

Information relevant to the (1) target animal safety component of the NADA is described further in Step 4 of Section IV.C.; (2) food safety component of the NADA is addressed further in Step 6 of Section IV.C.; and (3) establishing effectiveness is described further in Step 7 of Section C.

We recommend that you contact the Center for help in determining the most efficient manner to submit all the above relevant information.

9. Veterinary Feed Directive **(**21 CFR 514.1(b)(9))

Section 514.1(b)(9) requires that in the case of NADAs for Veterinary Feed Directive (VFD) drugs the application must include three copies of the VFD in the format described in 21 CFR 558.6(a)(4).

This requirement is not applicable to NADAs for animals whose genomes have been intentionally altered.

10. Supplemental Applications **(**21 CFR 514.1(b)(10))

Section 514.1(b)(10) requires that if an NADA is a supplemental application, such application must include full information on each proposed change concerning any statement made in the previously approved application.

This requirement applies to NADAs for animals whose genomes have been intentionally altered as it does to conventional new animal drug applications. Sponsors seeking supplemental applications for such animals should contact CVM to determine how to prepare such an application.

11. Applicant's Commitment **(**21 CFR 514.1(b)(11))

Section 514.1(b)(11) requires that an NADA include a commitment by the applicant that any labeling and advertising for the new animal drug is consistent with the conditions stated in the labeling which is part of the application.

This requirement applies to NADAs for animals whose genomes have been intentionally altered as it does to conventional new animal drug applications. Sponsors should refer to 21 CFR 514.1(b)(11) for a complete description of the conditions of this commitment.

12. Additional Commitments **(**21 CFR 514.1(b)(12))

Section 21 CFR 514.1(b)(12) requirements that are relevant to an NADA for animals whose genomes have been intentionally altered include commitments by the applicants that

i) the methods, facilities and controls described in section 514.1(b)(5) conform to the current good manufacturing practice (GMP) regulations in 21 CFR 211, and

ii) any nonclinical laboratory studies included in the application are conducted in compliance with good laboratory practice (GLP) regulations (21 CFR 58), or, if not conducted in compliance with such regulations, a brief statement of the reason for the noncompliance.

The requirement to comply with GLP regulations, including a statement regarding compliance or noncompliance, applies to NADAs for animals whose genomes have been intentionally altered as it does to conventional new animal drug applications.

13. Environmental Assessment **(**21 CFR 514.1(b)(14))

Section 514.1(b)(14) requires that an NADA include either a claim for categorical exclusion or an environmental assessment (EA). An EA must be prepared for each agency action except when the action is categorically excluded by 21 CFR 25.30 – 34 and no extraordinary circumstances exist. 21 CFR 25.21. The EA is a public document that provides sufficient information to allow FDA to either prepare an environmental impact statement (EIS) or issue a finding of no significant impact (FONSI). The specific information required for an EA is outlined in 21 CFR 25.40. This requirement applies to NADAs for animals with intentionally altered genomes as it does to conventional new animal drug applications.

An EA that demonstrates the animals whose genomes have been intentionally altered will not significantly affect the quality of the human environment leads to a finding of no significant impact (FONSI). We recommend that the EA focus on environmental issues and potential impacts related to the use and disposal of the animals and their final products, if relevant. The appropriate scope and content of the EA may vary widely depending on the animal product, claim, and conditions of use. Therefore, we recommend that you contact and work closely with us on these issues before proceeding with preparation of the EA, which is described in more detail in Step 6 of Section C.

14. Assembling and Binding the Application **(**21 CFR 514.1(b)(15))

Section 514.1(b)(15) describes certain administrative requirements for submitting an NADA to FDA. These requirements apply to NADAs pertaining to animals whose genomes have been intentionally altered as they do to conventional new animal drug applications. We recommend that you contact CVM for further information on assembling your NADA.

C. Recommended Process for Completing Pre-approval Assessments for Animals Whose Genomes Have Been Intentionally Altered

To facilitate the evaluation of animals whose genomes have been intentionally altered under the existing regulatory framework for new animal drugs, we have developed the following approach for submitting data for an NADA. It fulfills the regulatory requirements described in the preceding section and helps guide sponsors in developing their regulatory submission strategies.

This approach is cumulative and risk-based. Each component of the assessment forms the basis on which the next step is evaluated. The approach is risk-based because it examines both the *potential hazards* (that is, components that may cause an adverse outcome) identified at each step along the pathway and the *likelihood of harm* among the receptor populations (the animals whose genomes have been intentionally altered themselves as well as those individuals or populations exposed to these animals). It is also conducted on a case-by-case basis, because the potential hazards and risks are likely to be unique to each application.

We encourage you to consult with us as you develop data to satisfy the elements below, to ensure that the process is as efficient as possible and that the data and information you provide is in a format that will facilitate our ability to review it.

Step 1: Product Identification

Product identification (21 CFR 514.1(b)(1)), which many molecular biologists would refer to as product definition, forms the foundation for the evaluation process and drives subsequent data generation and review. It encompasses the specific lineage of animals whose genomes have been intentionally altered (that is, the altered genomic DNA as well as the animals containing it) and the purpose (i.e., intended use) of the altered genomic DNA that is the subject of the NADA. We believe that the concept of product identification is so important to the structure and content of the NADA submission that we encourage you to consult us on

this topic as early as possible in the development process, for example as an early part of the INAD process.

A product definition characterizes the animal whose genome has been intentionally altered. Therefore, as indicated in section IV.B.1, as appropriate to the submission, we recommend that the product identification include the following information:

- Ploidy;
- Zygosity;
- Description of the animal (e.g., common name/breed/line; genus and species);
- Characterization of the alteration of the genomic DNA (e.g., site(s) of alteration, nature of the alteration (deletion, substitution, addition, and if so, number of copies, etc., including the sponsor's name for the altered genomic DNA (e.g., gamma virus receptor nonsense mutation, inserted [new gene] e.g., the fatty acid desaturase n))
- Name of resulting animal line; and
- The intended use or claim being made for the lineage of animals whose genomes have been intentionally altered.

Step 2: Molecular Characterization of the Altered Genomic DNA

This step of the process serves to describe the components and composition of the article. (21 CFR 514.1(b)(4).) For this step, we recommend that you provide information for identifying and characterizing the altered genomic DNA that will be introduced into the progenitor of the animal to be marketed. This and the next step in the process are part of the hazard identification component of the safety review of the NADA. (21 CFR 514.1(b)(8)). Typically, the information should include, but not be limited to the following, as applicable to the particular type of altered genomic DNA (e.g., inserted DNA sequences; replaced DNA /nucleotide(s); or deletion of nucleotides or sequences):

- details of how the genomic alteration(s) was achieved;
- a description of the source(s) of the various functional components of the altered genomic DNA, as appropriate;
- the sequence of the altered genomic DNA, or of a sufficient number of nucleotides surrounding it such that the alteration can be uniquely identified (e.g., especially in the case of deletion alterations);
- the purpose of the alteration;

- the intended function(s) of the genomic alteration; and
- the purity of the preparation containing the materials used to effect the genomic alteration prior to introduction into recipient animals or cells.

In order to determine whether any risks exist that would make the product unsafe, we expect to evaluate whether the altered genomic DNA contains any potentially mobilizable DNA sequences, or whether sequences are present that encode pathogens, toxins (including allergens), or the addition or deletion of substances likely to dysregulate the growth control of cells, tissues, or organs, except by explicit design.

Step 3: Molecular Characterization of the Lineage of Animals Whose Genomes Have Been Intentionally Altered

This step continues the analysis of the intentionally altered genomic DNA and the location of the genomic alteration in the resulting animal, as well as the production of the animal(s) intended to be used in commerce and any potential hazards that may be introduced into those animals as part of their production. As such, this step addresses the identity and some manufacturing requirements of your NADA. 21 CFR 514.1(b)(1) and (b)(5). We recommend that you provide data and information describing the method by which you effected the alteration to the genomic DNA in the initial animal, including whether the initial animal was chimeric. In addition, we recommend that you describe the breeding strategy you used to produce the lineage progenitor (the animal that contains the final stabilized version of the initial event and from which the animals to be used for commercial purposes are derived). You should fully characterize the final stabilized altered genomic DNA in the animal.

Step 4: Phenotypic Characterization of Animals Whose Genomes Have Been Intentionally Altered

The previous steps of the review process have concentrated on establishing and characterizing the altered genomic DNA in the resulting animals. Information in this and the following steps helps establish whether the genomic alteration poses any risks to humans, risks to health of the animal, or risks to the environment.

With regard to health of the animals whose genomes have been intentionally altered, including the target animal safety requirements of 21 CFR 514.1(b)(8), we recommend that you submit data regarding whether the genomic alteration or its expression product(s) cause

any direct or indirect toxicity. In general, we recommend that you compile and submit data and information addressing the health of these animals, including veterinary and treatment records, growth rates, reproductive function, and behavior. In addition, we recommend that you submit data on the physiological status of the animals whose genomes have been intentionally altered, including clinical chemistry, hematology, histopathology, and post-mortem results. We recommend that you collect data from a generation of animals as close as possible to those intended for use in commerce.

Step 5: Genotypic and Phenotypic Durability Assessment

As in Step 3, this step also addresses some additional components of the manufacturing requirements codified in 21 CFR 514.1(b)(5). It is intended to provide information to ensure that the altered genomic DNA in the animal resulting from the specific alteration event and defining (identifying) the animal being evaluated is durable — that there is a reasonable expectation that the altered genomic DNA is stably inherited, and the phenotype is consistent and predictable. This would include developing a sampling plan.

For genotypic durability, we recommend that you use the results of studies demonstrating that the altered genomic DNA is stably inherited. For the phenotypic durability portion of the plan, we recommend that you submit data on the consistency of the expressed trait (based on the intended use) over multiple generations. We recommend that, where feasible, you gather data on inheritance from at least two generations, preferably more, and recommend that at least two of the sampling points be from non-contiguous generations (e.g., F_2 and F_4).

Your plan should include a method of identity with sufficient discrimination to determine (1) whether a given animal contains the altered genomic DNA, and (2) whether the altered genomic DNA has significantly changed from that which was evaluated to be safe and effective (i.e., a detection method for your genomic alteration in its final stabilized genomic location(s) in the animal whose genome has been intentionally altered). We recommend that you consult with us on developing these plans.

Step 6: Food Safety and Environmental Safety Assessments

Food Safety

This part of Step 6 addresses the food safety requirements in 21 CFR 514.1(b)(8). It focuses on the issue of whether food derived from animals whose genomes have been intentionally altered is safe for humans or animals consuming edible products from the animals.

The risk issues involved in determining food safety can be divided into two overall categories. The first addresses whether there is any direct toxicity, including allergenicity, via food consumption of the expression product of the article. The second category addresses potential indirect toxicity associated with both the article and its expressed product (e.g., whether location or expression of the article affects physiological processes in the resulting animal such that unintended food consumption hazards are created, or whether existing food consumption risks are increased). Potential adverse outcomes via the food exposure pathway should be identified by determining whether there are any biologically relevant changes (1) to the physiology of the animal (assessed partly in *Step 3: Phenotypic Characterization*), and (2) in the composition of edible tissues from the animals whose genomes have been intentionally altered that suggest reason for toxicological concern compared with the appropriate comparator.

In the end, if the expression product(s) is shown to be safe, and the composition of edible tissues from the animals whose genomes have been intentionally altered is shown to be as safe as those from animals of the same or comparable type that are commonly and safely consumed, then we expect to view this as evidence that food derived from the animals whose genomes have been intentionally altered is safe (i.e., there is a reasonable certainty of no harm from consumption of the food).

FDA participated in the Codex ad hoc Intergovernmental Task Force on Foods Derived from Biotechnology and its Working Group that developed the guideline for assessing food safety of foods from rDNA animals (Codex Alimentarius Commission: *Guideline for the Conduct of Food Safety Assessment of Foods Derived from Recombinant-DNA Animals*; in ALINORM 08/31/34, Appendix II; (http://www.codexalimentarius.org/download/standards/11023/CXG_068e.pdf). The information needed to establish food safety for food from animals whose genomes have been

intentionally altered under an NADA is consistent with that described in the Codex Guideline.

Environmental Safety

This portion of Step 6 addresses the environmental component of your NADA. 21 CFR 514.1(b)(14). We expect that, at least until we have more experience, most applications for animals whose genomes have been intentionally altered would have to be evaluated to determine whether such an approval will individually or cumulatively result in significant environmental impacts (i.e., whether an extraordinary circumstance exists). 21 CFR 25.21. An EA that demonstrates the animals whose genomes have been intentionally altered do not significantly affect the quality of the human environment leads to a finding of no significant impact (FONSI).

We recommend you contact us early in the development of your animal so that we can focus the EA on the environmental issues and potential significant impacts related to the use and disposal of your animal and its final product, if relevant. The appropriate scope and content of the EA may vary widely depending on the animal product, claim, and conditions of use (e.g., aquatic vs. terrestrial animal species). Therefore, we recommend that you contact and work closely with us on these issues before proceeding with preparation of the EA.

Step 7: Effectiveness/Claim Validation

The previous steps of the review process primarily address identity and safety issues. This last step of pre-market review addresses effectiveness, i.e., whether you have validated your claims for the characteristics that the animals whose genomes have been intentionally altered are intended to exhibit. 21 CFR 514.1(b)(8). For example, in the case of animals whose genomes have been intentionally altered that are intended to resist disease, you should demonstrate that those animals are indeed resistant to that disease. In the case of animals whose genomes have been intentionally altered that are intended to produce a non-food product, you should demonstrate that those animals indeed produce the claimed product. If that product is, for example, a drug or component of a drug intended for use in humans, the safety and effectiveness of that drug would be evaluated separately by Center for Drug Evaluation and Research. We recommend that you work closely with us to determine the nature and extent of data to meet these requirements, and to coordinate with CDER, CDRH, CFSAN, or CBER as appropriate.

V. Post-Approval Responsibilities

Once an animal whose genome has been intentionally altered is approved, sponsors have on-going responsibilities including registration and drug listing, recordkeeping, filing supplements, and periodic reporting. (21 USC 360, 21 USC 356a, 21 CFR 514.80, 21 CFR 514.8). We recommend that you use the following general approach to fulfill these requirements, but that you work closely with us during the development of the animals in order to determine the specific data and information to submit.

A. Statutory Registration and Drug Listing Requirements

As part of the registration requirements under 21 USC 360, you are required to register your name and place of business, and identify any facility(ies) engaged in the production or testing of the animals whose genomes have been intentionally altered. See 21 CFR Part 207. As part of your listing responsibilities, you are required to list all regulated articles, 21 CFR 207.22(a)(1), which should be a list of all lines of animals whose genomes have been intentionally altered that you have produced.

B. Recordkeeping

You must establish and maintain indexed and complete files containing full records of all information relevant to the safety or effectiveness of animals whose genomes have been intentionally altered that has not been previously submitted as part of the NADA. 21 CFR 514.80(a)(1). This would generally consist of Adverse Event Reports or other data or information from domestic or foreign sources, such as published literature.

C. Annual Reports, Supplements, and Other Changes to an Approved Application

We recommend that information demonstrating genotypic and phenotypic durability be collected on an annual basis from a subset of marketed approved animals whose genomes have been intentionally altered. You should consult with us during the INAD process on the nature of the information to be collected, as it will be determined on a case-by-case basis. We recommend that you maintain current standard operating procedures (SOPs) for each test method employed, and that you maintain SOPs for other procedures used in the husbandry of these animals (e.g., those resulting in biological containment).

You must submit information on all changes that have been made, or that you propose to make to the animals whose genomes have been intentionally altered (21 CFR 514.8(b)). Depending on the risk(s) that could be introduced by that change, the nature and timing of the reporting may be different. Information on the types of changes and which type of reporting they require are found in 21 CFR 514.8. We recommend contacting us if you have any questions regarding determining the category in which your changes may fall.

D. Records and Reports Concerning Experience with Approved Products

You are required to submit reports of data, studies, and other information of experience with the animals whose genomes have been intentionally altered. 21 CFR 514.80(a)(2). These experience reports must be submitted to our Division of Surveillance every six (6) months for the first two years following approval, and annually thereafter. 21 CFR 514.80(a)(4).

We remind you that the labeling associated with animals whose genomes have been intentionally altered may only prescribe, recommend, or suggest use under the conditions approved in the labeling that was submitted as part of the approval. 21 USC 360b(a)(1). This labeling must use the same language and emphasis as in the approval, including descriptions of relevant hazards and precautions.

VI. Import Tolerances

Section 512(a)(6) of the FD&C Act enables FDA to establish a safe level of new animal drugs and drug residues in edible portions of animals (i.e., food) imported into the United States (an import tolerance) when those drugs have not been approved for use in the United States. Whether a sponsor seeks approval of an NADA for a lineage of animals whose genomes have been intentionally altered or establishment of an import tolerance for food from such animals, the food safety standard is essentially the same.[16] Information about import tolerances, which enable imports of such food from animals whose genomes have been intentionally altered and that have been developed outside the United States, is found in the sections of this guidance relevant to evaluating food safety and is consistent with the recommendations in the Codex

[16] To establish an import tolerance, FDA must review data showing that the tolerance is safe based on food safety criteria similar to those used for a full NADA approval.

Guideline for the Conduct of Food Safety Assessment of Foods Derived from Recombinant-DNA Animals. We recommend that you consult with us on establishing an import tolerance.