#187B

Heritable Intentional Genomic Alterations in Animals: The Approval Process

Guidance for Industry

This guidance, together with companion final Guidance #187A, "Heritable Intentional Genomic Alterations in Animals: Risk-Based Approach," is a revision of Guidance #187, "Regulation of Genetically Engineered Animals Containing Heritable Recombinant DNA Constructs. It 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 guidance at any time. Submit electronic comments to https://www.regulations.gov. Submit written comments to the Dockets Management Staff (HFA-305), Food and Drug Administration, 5630 Fishers Lane, Rm. 1061, Rockville, MD 20852. All comments should be identified with the docket number FDA-2019-D-2648.

For further information regarding this document, contact ASKCVM@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, 7500 Standish Place, Rockville MD 20855, and may be viewed on the Internet at https://www.fda.gov/animal-veterinary, https://www.fda.gov/regulatory-information/search-fda-guidance-documents, or https://www.regulations.gov.

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Contains Nonbinding Recommendations

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Guidance for Industry

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 responsible for this guidance as listed on the title page.

I. Introduction and Background

FDA (the agency, we) is issuing this Guidance for Industry (GFI) to explain how its approval process applies in the context of products related to heritable intentional genomic alterations (IGA) in animals.[1] We also cover the import tolerance process at the end of this guidance document. In order to make our guidance clearer and more streamlined, and to allow for a process to update the more technical portion of the guidance, we are issuing this guidance document in two parts: this guidance, GFI #187B, to provide technical guidance for those IGAs in animals that go through the approval process, and a previously issued companion final guidance document, GFI #187A,[2] which articulates our risk-based approach to IGAs in animals, including when the Agency would consider it appropriate to exercise enforcement discretion and not take action against a sponsor for the marketing and distribution of an IGA in animals without prior FDA approval. This guidance, GFI #187B, is applicable once a sponsor is submitting an application for approval for IGAs described as Category 3 in GFI #187A. In general, among the products that may be appropriate for Category 3 are those that are intended for human or animal health-related uses, are intended for release into the environment, or that may be less familiar (i.e., we do not have an understanding of the risks) or may be more complex (note that even seemingly "simple" genetic changes may in fact have complex considerations and be appropriate for Category 3, for example, if the alteration results in the production of a novel substance).

IGAs in animals are intentional genomic alterations made 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

[1] Non-heritable intentional genomic alterations in animals are outside the scope of this guidance document.

[2] GFI #187A, "Heritable Intentional Genomic Alterations in Animals: Risk-Based Approach" (https://www.fda.gov/media/74614/download) (May 2024).

the animal.[3,4,5] "Intended" or "intentional" as used here references a developer/sponsor's purposeful action to achieve a particular end result. In other words, the sponsor makes a genomic alteration in order to alter the structure or function of an animal in a particular way and/or to cure, mitigate, treat, or prevent disease.

This guidance is intended to clarify our requirements and recommendations for developers ("sponsors," "you") of IGAs in animals. We note that what we are regulating (i.e., the regulated article) is the IGA in the animal, not the animal itself.

In general, IGAs in animals require premarket approval, unless otherwise excluded (21 U.S.C. 321(g)(1); 321(v); 360b(a)). However, FDA does not expect submission of an application for approval where the IGA is in a nonfood-producing animal that is regulated by another Federal government agency or entity, as explained in GFI #187A.[6, 7] Furthermore, based on FDA's understanding of the risks they pose and whether the risks are appropriately mitigated, on a case-by-case basis, we may decide to exercise enforcement discretion over approval requirements for the introduction or delivery for introduction into interstate commerce of an IGA in an animal, as described in GFI #187A. This guidance document is intended to explain the approval process and requirements for IGAs in animals not subject to the enforcement discretion policies outlined in GFI #187A based on our risk-based approach and their risk profile. If you believe that your product[8] may be eligible for enforcement discretion, you should refer to GFI #187A and contact the Center for Veterinary Medicine (CVM) to discuss whether enforcement discretion may be appropriate.

In addition to consulting this guidance document, we also encourage sponsors to consult CVM's Veterinary Innovation Program (VIP) webpage. This program is intended to assist developers of innovative veterinary products, including certain IGAs in animals, by providing intensive

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[3] FDA used the term "genetically engineered" (GE) to describe the animals within the scope of the previous version Guidance for Industry #187, "Regulation of Genetically Engineered Animals Containing Heritable Recombinant DNA Constructs." We have decided not to use that term in this revised guidance to avoid any confusion as to whether that term encompasses IGAs in animals developed using new technologies. Except for citation of earlier documents, we also do not use the term "transgenic" because that term is generally used to refer to organisms containing foreign DNA.

[4] In Guidance for Industry #236, "Clarification of FDA and EPA Jurisdiction Over Mosquito-Related Products," FDA clarified that articles intended to function as pesticides by preventing, destroying, repelling, or mitigating mosquitoes for population control purposes are not regulated under FDA's authority. Rather, the Environmental Protection Agency (EPA) regulates products intended to function as pesticides under the Federal Insecticide, Fungicide, and Rodenticide Act (7 U.S.C. § 136 et seq.).

[5] The term "modern molecular technologies" does not include induction of polyploidy by heat, pressure, or chemical treatment or selective breeding or other assisted reproductive technologies, including random mutagenesis followed by phenotypic selection.

[6] FDA does not intend to regulate IGAs in animals that meet the definition of a veterinary biologic and are regulated by the Animal and Plant Health Inspection Service (APHIS) of the United States Department of Agriculture (USDA). 21 CFR 510.4.

[7] As noted in GFI #187A, this includes nonfood-producing insects with intentionally altered genomes that are regulated by APHIS under its Plant Protection Act authority, 7 U.S.C. § 7701 *et seq.*

[8] The product is the IGA and the marketed item(s) containing the IGA (e.g., eggs, semen, embryos, live animals, etc.).

technical and programmatic assistance throughout the approval process in order to make it as efficient as possible.

Some IGAs in animals are intended to produce medical and other products, such as human drugs, biologics, or medical devices that are subject to regulation by other FDA centers (i.e., "biopharm animals"). Where sponsors have developed IGAs in animals that are intended to produce human medical products not subject to FDA's enforcement discretion policy 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), sponsors should have discussions with CVM early during the development process. CVM will then work closely with the other FDA Centers to coordinate efficient Center reviews.

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. What We are Regulating

As explained in GFI #187A, FDA's CVM regulates the specific intentional DNA alteration at each site in the genome where the alteration (insertion, substitution, or deletion) occurs. In general, each specific IGA at a specific site is considered to be a separate article subject to approval requirements. However, CVM may consider a specific combination of multiple IGAs under a single application. For example, CVM may consider three IGAs in swine under a single application if all three of the IGAs are present in the commercialized animals and they contribute to the same intended use(s) (e.g., to improve organ compatibility for xenotransplantation in humans). CVM may also consider IGAs in multiple lines or breeds of animals under a single application. For example, an application could be for an IGA in multiple breeds of turkeys generally, rather than one single breed of turkey. In general, IGAs in lines of animals that are significantly genetically different, such as unrelated species, would not be appropriate for a single application even if the intended use(s) is the same. [9]

If a sponsor wishes to include multiple IGAs or include multiple lines of animals containing IGAs under a single application for approval, we recommend that the sponsor contact CVM to discuss this regulatory option and the scientific questions that would need to be addressed in an application. In general, the sponsor would need to demonstrate that it can make the IGA(s) consistently and that the safety and effectiveness of the IGA(s) does not change. For example, the sponsor could provide data from a subset of animals with a particular IGA to demonstrate that the alteration can be made consistently in different founder animals with the same safety and effectiveness and that the outcome in this subset is representative of alterations that would be made in commercial production.

[9] Application user fees, where applicable, would apply once to the one application for the multiple IGAs or lines of animals in these examples. 21 U.S.C. § 379j-12(a)(1)(A).

During the investigational phase, the sponsor may establish one investigational file or veterinary master file (VMF) for multiple IGAs, and the file may contain information on investigational IGAs in animals that contain different numbers or types of IGAs, including those occurring at different locations of the genome, prior to selecting the lineage of animals containing the specific IGA or combination of IGAs that are intended for commercialization.

The data and information in the investigational file can serve to support an application for approval. CVM may review all animals used to generate the commercialized animal with the IGA(s) under a single application.

During the post-marketing phase, if you breed an animal containing an IGA that FDA has approved with another animal containing an IGA that FDA has approved under a separate application or FDA has determined to fit Category 2 as described in GFI #187A and FDA has stated it intends to exercise enforcement discretion over it *or* with an animal that does not contain an IGA, and you are making no claims for a new intended use of the IGA, then CVM would not impose any new requirements, including a new approval, for the resulting progeny.

III. Investigational Use of IGAs in Animals

FDA regulations concerning investigational use, codified at section 511.1 in Title 21 of the Code of Federal Regulations (21 CFR 511.1), apply to investigational IGAs in animals. The development of such IGAs in animals constitutes clinical investigation within the meaning of 21 CFR 511.1(b) because it involves studying the effectiveness of the IGA in the target species, including effects of its expression product(s), if any.

In general, FDA regulations specify labeling and recordkeeping requirements, animal disposition, and conditions under which food[10] from animals used for clinical investigations under 21 CFR 511.1(b) can be introduced into the food supply. 21 CFR 511.1(b) also requires that prior to shipping an investigational product for clinical tests, a sponsor must submit a Notice of Claimed Investigational Exemption containing specified information. In addition, before shipping, you should submit a claim of categorical exclusion or draft environmental assessment (EA) as described in section III.D. *Environmental Considerations* below.

We strongly recommend that you contact CVM early in the development process to determine what information you should submit to CVM and the appropriate file type to utilize based on your intended activities and stage of development. We also encourage you to participate in the VIP, which offers assistance to sponsors of IGAs that provide a benefit to human health, animal health, animal well-being, or enhanced food production. Benefits include intensive interactions with CVM during the development process, pre- and post-review feedback on application submissions, and hands-on assistance, such as technical advice for the development and validation of identification and assay methods. The goals of the VIP include providing greater certainty in the regulatory process and supporting a more efficient and predictable pathway to approval.

[10] The term "food" includes food for humans and animals. 21 U.S.C. § 321(f).

You can submit certain types of early information without the establishment of an investigational file and FDA will consider such information to be confidential (21 CFR 20.61). You should note that establishment of an investigational file will mean you are a "sponsor" under section 739 of the Federal Food, Drug, and Cosmetic Act (FD&C Act) and are responsible for payment of an annual sponsor fee unless you are eligible for a fee waiver. You can find information concerning user fees and fee waivers and reductions here. CVM will work with you to determine the appropriate timeframe to open an investigational file, strategies for generating the data and information that will be needed for an application, 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 IGA in the animal, including the species of animal to be under study, the altered gene(s) or region of the genome, and the intended effect of the IGA, including any gene product(s) that may be produced.

Except where CVM has indicated otherwise,[11] you will need to submit a notice prior to shipping animals with IGAs (21 CFR 511.1(b)(4)). Also, if you wish to introduce any food derived from investigational animals into the food supply, you must get prior CVM authorization to do so through the investigational 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 if your investigational animals may be suitable for consideration for food use. To authorize food use, CVM reviews the provided scientific data and information to determine if authorization of food use is consistent with the public health (21 CFR 511.1(b)(5)(i)).

We encourage you to contact CVM if you have questions about submitting a request to establish an investigational file or requesting participation in the VIP. Once we have established an investigational file, you will receive a letter assigning a unique 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 investigational file. As previously stated, an investigational file can encompass animals derived from multiple alterations. If you are interested in pursuing this type of approach, we encourage you to discuss your plans with us as early in the development process as possible.

We recommend that you schedule a meeting with us before you establish a file with us or immediately thereafter. In that meeting, you can acquaint us with the nature of the IGA and animal under development and the intended use. We can then provide you with more specific information on the kinds of responsibilities you have, 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 an IGA in an animal lineage, the animals may need to be moved from the initial laboratory or barn to other sponsor facilities, or to other investigators. If the investigational IGA in animals or products derived from them are shipped to other investigators, it is important to ensure that those individuals/entities receiving the

[11] GFI #187A describes FDA's intent to exercise enforcement discretion over investigational and approval requirements for certain IGAs in animals. Those products are not within the scope of this guidance, GFI #187B, because this guidance applies only to products for which a sponsor is seeking application approval.

investigational IGA in 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. USDA's general requirements for movement of both animals and animal products that do and do not contain IGAs are found at USDA APHIS | Imports and USDA APHIS Exports. These USDA requirements do not distinguish between requirements for animals and animal products that contain IGAs and those that do not.

B. Animal Disposition

An important goal during the investigational phase of development of animals with IGAs 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 investigational animals and animal products. We recommend that all surplus investigational animals be maintained in a contained environment where appropriate or humanely euthanized; that they and their biological products be disposed of by incineration, burial, or composting or that the sponsor contact CVM regarding investigational food-use authorization or alternative disposition; and that you keep appropriate records of animal identification and disposition.

C. Investigational Food-Use Authorizations

Introducing treated investigational animals or animal products into the food supply requires an Investigational Food-Use Authorization (21 CFR 511.1(b)(5)). FDA may grant authorization for food use, rendering authorization solely for animal food use, or alternative disposition provided that the criteria in 21 CFR 511.1(b)(5) are met.

By "treated investigational animals," we mean animals involved in the study of the IGA, i.e., animals containing an IGA, including their offspring and potentially other animals used in the investigation (we recommend you contact us to discuss each specific case). "Treated investigational animals" as used in this guidance does not include surrogate dams (embryo recipient animals) in certain species (cattle, swine, sheep, and goats) that gestate embryos/fetuses/offspring with IGAs resulting from a variety of assisted reproductive technologies (e.g., embryo transfer, cloning, artificial insemination, in vitro fertilization) and do not themselves contain an IGA. FDA has determined that these animals are not "treated" within the meaning of the regulation because they are extremely unlikely to contain the IGA, through placental transfer or otherwise. We reached this conclusion based on the placental anatomy of these species as well as available scientific research[12] assessing fetal microchimerism for which the risk of transfer of genomic material from fetus to dam was found to be extremely low.

[12] See e.g., Garrels, W. et al. Assessment of fetal cell chimerism in transgenic pig lines generated by Sleeping beauty transposition. *PloS one* **9**, e96673 (2014); Steinkraus, H.B. et al. The absence of detectable fetal microchimerism in nontransgenic goats (Capra aegagrus hircus) bearing transgenic offspring. *Journal of animal science* **90**, 481-488 (2012) (https://journals.plos.org/plosone/article?id=10.1371/journal.pone.0096673).

Therefore, edible products from these animals may enter the food supply without prior FDA authorization. By contrast, fetuses and offspring from any species that do contain IGAs are considered "treated" and must not enter the food supply without prior FDA authorization. Please contact CVM if you have questions about such authorizations, or if you are seeking to introduce into the food supply surrogate dams from species other than cattle, swine, sheep, and goats.

For those animals subject to slaughter inspection by the U.S. Department of Agriculture's Food Safety and Inspection Service (FSIS), we will inform FSIS if the regulatory criteria 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 (which includes dried, frozen, and liquid eggs), and tests for animal drug residues to determine if the residue is above the tolerance concentration (maximum allowable amounts).

We recommend that prior to making an investigational food-use authorization request, you schedule a teleconference or in-person meeting with us to determine if your 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. FDA's food safety evaluation focuses on hazard identification, hazard characterization and exposure assessment, and any mitigations of human exposure. The data and information that should be provided in order to be considered for an investigational food-use authorization are determined on a case-by-case basis, based on the specific IGA in the food animal. The evaluation focuses on any intended and unintended effects of the IGA that may result in a hazard to the consumer who ingests the edible tissues derived from these animals, based on the health status of the animals, and the potential impact on the composition of foods produced from the animals.

D. Environmental Considerations

Actions on investigational files are considered major Federal actions under the National Environmental Policy Act (NEPA), and as such, may require preparation of an 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) unless a categorical exclusion from the requirement to prepare an EA applies. For example, 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 information for us to conclude that extraordinary circumstances will not exist (21 CFR 25.21). This should include information on animals with an IGA and the animals' containment to allow us to conclude that use and disposal of any investigational animals and their products would not have a significant impact on the human environment (42 U.S.C. 4332(2)(C); 21 CFR 25.15).

The amount and type of information needed will vary from animal to animal depending on the potential impacts they could have on the environment. At a minimum, we will need information to evaluate the amount and effectiveness of containment measures in place. By "amount," we mean the number of containment measures and their redundancy. For animals for which the risks of escape and establishment in the environment are high, we would expect to see redundant means of containment so that if one fails, other measures are in place to prevent escape. For example, animals that are highly mobile (e.g., fish, insects) may require more levels of containment and redundancy than less mobile animals (e.g., cows). The redundancy of measures

will contribute to the overall effectiveness of containment; however, "effectiveness" also refers to the reliability of the measures and their ability to prevent escape. In addition, the amount and effectiveness of the containment measures we expect to see will depend on the environmental hazards associated with the alteration and the risk for the alteration to result in environmental impacts.

We recommend that you contact us early in the development process to discuss whether your investigational animal may be eligible for a categorical exclusion under 21 CFR 25.33(e), or whether extraordinary circumstances may exist that would require preparation of at least an EA (21 CFR 25.21) and, if so, the scope of this document.

If the preparation of an EA by the sponsor is needed, FDA will use the EA to examine the potential for environmental impacts, including the potential for inadvertent release or escape of an animal with an IGA and/or the animal's products into the environment, and whether certain measures may mitigate any potential significant impacts that would adversely affect the human environment. Based on the analysis in the EA, FDA will determine whether to prepare a FONSI or an EIS.

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.), administered by the Environmental Protection Agency, and other statutes.

IV. FDA Approval of IGAs in Animals

A. Overview

CVM encourages sponsors to submit data for review at the most appropriate and productive times in the IGA product development process. Rather than submitting all data for review as part of a complete application, we have found that the submission of data supporting discrete technical sections during the investigational phase of the IGA is most productive. This "phased review" of data submissions has created efficiencies for CVM and sponsors.

The phased review of data submissions is a voluntary process a sponsor may use to complete any or all of the technical sections[13] required for approval of an IGA before submitting an application. In a phased-review process, a sponsor submits data and information in support of the various technical sections to their investigational file for CVM review and acceptance. The sponsor submits the application after CVM has reviewed all technical sections necessary to fulfill the requirements for the approval of the IGA under 21 CFR 514.1 and issued technical section complete letters for each of the required technical sections.

When submitting an application, you must include the results of the investigations you have conducted. We will evaluate the application to determine whether you have demonstrated that the IGA is safe and effective for its intended use. To demonstrate safety, generally you would

[13] The major technical sections are molecular characterization, phenotypic characterization (or target animal safety), durability assessment and plan, food safety, environmental impact, and effectiveness. Minor technical sections are labeling and "all other information."

characterize the IGA to identify potential hazards and evaluate safety to animals and humans. Further information on demonstrating safety is described in sections IV.C.1. *Product Characterization* and IV.C.3. *Food Safety and Environmental Impact Assessments*. 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 show that the expression product is in fact expressed in the animal. To demonstrate effectiveness of an IGA intended to alter a characteristic of the resulting animal, in general you would show that the animal had the claimed altered characteristic (e.g., that the investigational animals' rate of growth was as claimed or that it was indeed resistant to a disease). Further information on demonstrating effectiveness is described in section IV.C.4. *Effectiveness*.

FDA's review of applications is subject to specific User Fee Performance Goals timeframes,[14] including a 180-day timeframe for review of a technical section of an application under the phased-review process. FDA is required to report its performance in meeting User Fee Performance goals timeframes. 21 U.S.C. 379j-13. Performance reports are available here.

Note that, ordinarily, if CVM finds a submission incomplete, then when it is resubmitted the 180-day process restarts; however, for products enrolled in VIP, we may stop the clock at the time FDA finds the submission incomplete and then restart it when the sponsor submits the additional information as an amendment to FDA. This shortens the time to approval because if FDA "incompletes" a submission at day 60, for example, in the traditional system when the sponsor resubmits, the clock restarts at day 0, with 180 days remaining, but under VIP the clock restarts at day 60, meaning there are 120 review days remaining. FDA's total review time is 180 days. How long approval takes depends on how quickly sponsors submit complete technical sections. See the VIP webpage for further information on how this works.

With respect to how to ensure your submission is "complete," we encourage you to meet with us early in your development process to ensure that any studies you are conducting and the resulting data and reports submitted to CVM will be adequate to support approval. For those enrolled in VIP, we also encourage you to take advantage of pre-review feedback offered to VIP sponsors, which is described at the VIP webpage cited above.

The Agency is interested in increasing the transparency of its actions. In particular, we intend to make as much information publicly available as possible about our decisions. However, FDA is generally prohibited from disclosing data or information that falls within the definition of a trade secret or confidential commercial or financial information without the express written consent of the sponsor (18 U.S.C. § 1905; 21 U.S.C. § 331(j); 21 CFR 20.61(c)). As is the case for all applications, after CVM approval of an application, CVM will post a summary of the information in the application, 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, and the Agency's EA and FONSI, where applicable (21 CFR 514.11(e), 21 CFR 25.51).

[14] As of the date of this guidance, the performance goals can be found at https://www.fda.gov/media/116001/download.

Note that, for IGAs in animals for food use that are subject to USDA authority as amenable species[15] (21 U.S.C. § 453 et seq., 601 et seq.), developers should work with the slaughter and/or processing establishment(s) to determine if USDA Food Safety and Inspection Service (FSIS) sketch label approval for the finished meat and poultry product is required (9 CFR 412.1). The developer may also obtain a USDA APHIS permit for animals of any species that have the potential to pose a livestock pest or disease risk per the Animal Health Protection Act (AHPA; 7 U.S.C 8301, et seq.) based on a USDA APHIS determination that restrictions are required for interstate movement of such animals where USDA has determined that the IGA poses an animal disease risk. FDA will share information with USDA during the phased application review process[16] in order to facilitate USDA's determinations, but it is the developer's responsibility to ensure that they have obtained the necessary USDA determinations prior to marketing.

B. Application Requirements

Section 512(b)(1) of the FD&C Act describes the information that must be submitted to FDA as part of an application. These statutory requirements are further explained in regulations found at 21 CFR 514.1.

The application of some of the statutory and regulatory requirements to IGAs in animals 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 the product as described in section 512(b)(1)(B) of the FD&C Act and 21 CFR 514.1(b)(4) of the application regulations applies to IGAs in animals. Therefore, this section of the guidance document provides a brief summary of the application requirements in 21 CFR 514.1 and describes how these requirements may be addressed for applications submitted for IGAs in animals. Section IV.C. *Recommended Process for Completing Pre-approval Assessments for IGAs in Animals* describes our recommendations for how to present this information in the structure of an application 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, among other things, 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 name of the product.

The information that should be provided to satisfy this requirement for an application for an IGA in a lineage of animals is similar to that provided for a non-IGA product under section 514.1(b)(1). In the case of such an application, the "trade and/or chemical name" should be described by identifying the animal, its ploidy and zygosity, the name and intended function of the IGA, and the number and characterization of the site(s) of alteration,[17] including unintended

[15] Amenable species include cattle, sheep, swine, goats, fish of the order Siluriformes, and poultry (i.e., chickens, turkeys, ducks, geese, guineas, ratites, and squabs).

[16] https://www.fda.gov/about-fda/domestic-mous/mou-225-24-010

[17] The term "site of alteration" in this document refers to the genomic location of the IGA either within a chromosome or as an extrachromosomal element. In general, we are most interested in characterizations that are performed in animals close to commercialization.

alterations (e.g., alterations at identified potential off-target sites), as well as the intended use of the resulting lineage(s) of animals. For a more complete description of how we recommend you present this information in the application, please see section IV.C.1. *Product Characterization*.

We consider this component to be critical to the structure and content of an application 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 investigational phased-review process.

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

Section 514.1(b)(2) requires that an application include a table of contents that identifies the data and other material submitted and a well-organized summary of information that (1) describes the chemistry of the product, 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 application, please see section IV.C.1. *Product Characterization*.

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

Section 514.1(b)(3) requires that an application include each piece of labeling to be used for the product.[18]

In the context of IGAs in animals, the sponsor must include labeling paperwork with shipped animals; this includes labels and other written, printed information (i.e., labeling) that will accompany the animals. Labeling paperwork 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(s), and the intended use of the animals containing the article. Where the labeling paperwork for an IGA in animals contains animal care or safety information (e.g., husbandry or containment instructions relating to the IGA such as feeding instructions to accommodate enhanced growth), the labeling paperwork should accompany the animal when distributed from the sponsor or developer to farmers, growers, or producers. However, when farmers, growers, or producers ship such animals of food producing species, FDA expects the labeling to accompany the shipped animals only if it includes care or safety information relevant to the specific IGA and the animals go to a different grower or producer that will need the care or safety information, not when the farmers, growers, or producers send the animals to a slaughter facility. We recommend that you contact CVM for further information regarding the labeling for such animals.

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

Section 514.1(b)(4) requires that an application include (1) a list of all articles used as components of the product; (2) a statement of composition of the product; and (3) a third

[18] This discussion does not pertain to labeling requirements for food derived from animals containing IGAs. Note that such food products may be subject to labeling requirements under the National Bioengineered Food Disclosure Standard, 7 U.S.C. § 1639b, under the authority of the USDA Agricultural Marketing Service.

subsection concerning fermentation that does not apply to you because it is not relevant to IGAs in animals.

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 application, please see section IV.C.1. *Product Characterization* ((b) and (c)).

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

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

For IGAs in animals, this information should encompass:

- the method by which the alteration was introduced into the initial animal(s), including whether the initial animal(s) containing the IGA was chimeric;
- the breeding strategy used to produce the lineage progenitor(s) (a lineage progenitor(s) is the animal(s) containing an IGA from which subsequent animals used for commercial purposes are derived); and
- full characterization of the site of intentional alteration and any unintended alterations (e.g., alterations at identified potential off-target sites, unanticipated insertions, substitutions, or deletions) in 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 any coding or regulatory regions. For more information, see section IV.C.1. *Product Characterization* ((b) and (c)).

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 IGA is stably inherited. For the phenotypic durability portion of the plan, we recommend that you submit data on the consistency of the introduced trait (based on the intended use) over multiple generations or, in the event that animals are not propagated via breeding (e.g., somatic cell nuclear transfer), consistency of the introduced trait over alternative groupings of animals that you have discussed with CVM in advance. We recommend that, where feasible, you gather data on inheritance from at least two generations and that at least two of the sampling points be

from non-contiguous generations (e.g., F_1 and F_3). However, where you believe it is not feasible to gather data on multiple generations, we recommend you contact us to discuss potential alternative methods of demonstrating durability.

Your plan should include a method of identity (i.e., a method to detect the IGA in your animal or product), where applicable and feasible, with sufficient discrimination to determine (1) whether a given animal contains the IGA, and (2) whether the IGA has significantly changed from that which was evaluated in the application. For a more complete description of how we recommend you present this information in the application, please see sections *IV.C.1. Product Characterization* ((b) and (c)) and IV.C.2. *Durability Assessment and Plan*. We recommend that you consult with us on developing these plans.

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

Under section 514.1(b)(6), FDA may request that samples of the product and articles used as components and information concerning them be submitted to CVM.

Sponsors are encouraged to contact CVM to determine what samples (such as a genomic sample containing the article) may be appropriate.

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

Section 514.1(b)(7) requires that an application include a description of practicable method(s) and data to enable determination of the quantity of the article in food-producing animals (i.e., an analytical method), except when data or other adequate information establish that it is not reasonable to expect the article, or any substance formed in or on food, because of its use, to become a component of food at concentrations considered unsafe.

If FDA has determined that an IGA is safe for consumption, which includes a determination that anything formed in or on the food as a result of the IGA is safe (21 U.S.C. § 360b(b)), and no tolerance is required, then an analytical method for residues is not required because it is not reasonable to expect the IGA to be a component of food at concentrations considered unsafe (although, as described above in the section about 21 CFR 514.1(b)(5), a method of identity (i.e., a detection method for the IGA itself) would be required where applicable). However, FDA may in some circumstances find that an IGA is safe for consumption only with a required tolerance, e.g., where there may be excess production of a mineral or protein that could cause adverse reactions at a high concentration in some populations. In these circumstances, a method may be required for "any substance formed in or on food" because of the IGA (21 CFR 514.1(b)(7)).

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

Section 514.1(b)(8) requires that an application include data and information to permit evaluation of the safety and effectiveness of the 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 of the product, favorable and unfavorable.

Information relevant to the (1) target animal safety component of the application is described further in section IV.C.1.d. *Phenotypic Characterization*; (2) food safety component of the

application is addressed further in section IV.C.3.a. *Food Safety*; and (3) establishing effectiveness is described further in section IV.C.4. *Effectiveness*.

We recommend that you contact CVM 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 applications for Veterinary Feed Directive (VFD) products, the application must include the VFD in the format described in 21 CFR 558.6(b)(3)-(b)(4).

This requirement is not applicable to applications for IGAs in animals.

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

Section 514.1(b)(10) requires that if an application 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 applications for IGAs in animals as it does to non-IGA product applications. Sponsors seeking supplemental applications 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 application include a commitment by the applicant that any labeling and advertising for the product is consistent with the conditions stated in the labeling which is part of the application.

This requirement applies to applications for IGAs in animals as it does to non-IGA products. 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 application for an IGA in animals include commitments by the applicants that:

i) the methods, facilities and controls described in section 514.1(b)(5) conform with current good manufacturing practice (CGMP) (section 501(a)(2)(B) of the FD&C Act), and

ii) any nonclinical laboratory studies included in the application are conducted in compliance with good laboratory practice (GLP) regulations (21 CFR part 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 applications for IGAs in animals as it does to non-IGA product applications.

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

Section 514.1(b)(14) requires that an application include either a claim for categorical exclusion or an EA. An EA must be prepared for each major federal action except when the action is categorically excluded by 21 CFR 25.30 – 35 and no extraordinary circumstances exist (21 CFR 25.21). The EA is a public document that provides sufficient information to allow FDA to determine whether to prepare and issue a FONSI or prepare an EIS. The specific information required for an EA is outlined in 21 CFR 25.40. This requirement applies to applications for IGAs in animals as it does to non-IGA product applications.

An EA that demonstrates the IGA in animals will not significantly affect the quality of the human environment leads to a 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 your preparation of the EA, which is described in more detail in section IV.C.3.b. *Environmental Impact*. You can also find more information on environmental review here.

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

Section 514.1(b)(15) describes certain administrative requirements for submitting an application to FDA. These requirements apply to applications pertaining to IGAs in animals as they do to non-IGA product applications. We recommend that you contact CVM for further information on assembling your application.

C. Recommended Process for Completing Pre-approval Assessments for IGAs in Animals

To facilitate the evaluation of IGAs in animals under the existing application regulatory framework, we have developed the following approach for submitting data for an application. It fulfills the regulatory requirements described in the preceding section and helps guide sponsors in developing their regulatory submission strategies.

This approach is cumulative, in that each component of the assessment forms the basis on which the next section is evaluated. The approach is also risk-based because it examines both the *potential hazards* (that is, components that may cause an adverse outcome) identified at each section along the pathway and the *likelihood of harm* among the receptor populations (the IGAs in animals themselves as well as those individuals or populations exposed to these animals, e.g., increased risk of zoonotic disease). It is also conducted on a case-by-case basis because the potential 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.

1. Product Characterization

a. 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(s) of animals containing an IGA (that is, the IGA as well as the animals containing it) and the purpose (i.e., intended use) of the IGA that is the subject of the application. We believe that the concept of product identification is so important to the structure and content of the application that we encourage you to consult with us on this topic as early as possible in the development process.

A product identification characterizes the IGA in an animal. Therefore, as indicated in section IV.B. *Application Requirements*, as appropriate to the submission, we recommend that the product identification include the following information:

- Characterization of the IGA including site(s) of alteration, nature of the alteration (deletion, substitution, addition, and if so, number of copies, etc.) and the sponsor's name for the IGA. For example, an IGA might be characterized as "gamma virus receptor nonsense mutation, inserted [new gene] e.g., the fatty acid desaturase *n*;"
- Ploidy;
- Zygosity;
- Description of the animal (e.g., common name/breed/line; genus and species);
- Name of resulting animal line; and
- The intended use or claim being made for the lineage(s) of animals with an IGA.

b. Molecular Characterization of the IGA

This section of the process serves to describe the components and composition of the article (21 CFR 514.1(b)(4)). For this section, we recommend that you provide information for identifying and characterizing the IGA that will be introduced into the progenitor(s) of the animal to be marketed. This and the next section in the process are part of the hazard identification component of the safety review of the application (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 IGA (e.g., inserted DNA sequences; replaced DNA/nucleotide(s); or deletion of nucleotides or sequences):

- details of how the IGA(s) was achieved;
- a description of the source(s) of the various functional components of the IGA, as appropriate;

- the sequence of the IGA 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 IGA;
- the intended function(s) of the IGA; and
- the purity of the preparation containing the materials used to affect the IGA 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, among other safety considerations, whether the IGA contains any potentially mobilizable DNA sequences, the presence of sequences that may be predicted to result in altered disease susceptibility or encode pathogens, the production of novel substances not ordinarily produced by the animal (including allergens), the addition or deletion of substances likely to dysregulate the growth control of cells, tissues, or organs, except by explicit design, and any other changes to the physiology and health of the animal.

c. Molecular Characterization of the Lineage(s) of Animals with IGAs

This section continues the analysis of the IGA and the location of the IGA 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 section addresses the identity and some manufacturing requirements of your application (21 CFR 514.1(b)(1) and (b)(5)).

To characterize the lineage(s) of animals with IGAs, we recommend that you provide a detailed description of how the IGA was introduced into the founder animal(s)'s genome. This methodology information should include whether the IGA was introduced into cells or embryos as well as the source and type of critical reagents used (e.g., source of recombinant DNA constructs or guide RNA molecules, form or type of nuclease used, etc.). This information will be case-specific and help to inform the subsequent analytical steps (e.g., expected chimerism or mosaicism in founder animals should be considered when designing the approach to characterize the IGA in the animal's genome). For example, the appropriate type of sequencing data will be highly dependent on the methods used to generate the IGA and the complexity of the IGA(s). In some cases, whole genome sequencing may be the most appropriate approach to characterize the intended and unintended alterations that may result from the introduction of the IGA, while in other cases targeted sequencing of specific genomic loci may be appropriate. Additionally, if mosaic alterations may be present at low levels, the depth of sequencing coverage[19] may need to be increased.

Below are two general examples of when a given approach may be more appropriate:

- Example 1: CRISPR-Cas genome editing using multiple guide RNAs and templates to introduce large transgene insertions into cells from which founder animals are cloned.

[19] Sequencing coverage refers to the average number of times a DNA segment, in its given position in the genome, is sequenced during analysis and is reported as "(average number of reads)X" (e.g., 30X).

Each guide RNA increases the number of potential off-target sites to screen, which may produce a very large number of empirically-determined sites to target. The large transgene insertions may be better assessed with longer reads compared to shorter reads. However, given the editing of cells to be used in cloning (not an embryo), mosaic alterations are not expected. Thus, long-read whole genome sequencing of the cell line to an average sequencing coverage (e.g., 30X) may be most appropriate.

- Example 2: CRISPR-Cas genome editing using one guide RNA introduced into embryos to produce founder animals that may be mosaic. Higher sequencing coverage is necessary to detect low-level unintended alterations (mosaicism). However, complementary methods may be used to empirically determine a list of off-target sites for the one guide RNA that can be targeted for screening of specific loci. Thus, the combination of an *in silico* prediction, empirical prediction assay, and hybridization-based short read targeted sequencing to a significantly higher depth of sequencing coverage (e.g., 1000X) may be most appropriate.

In addition, we recommend that you describe the breeding strategy you used to produce the lineage progenitor(s) (the animal(s) that contains the final stabilized version of the initial alteration and from which the animals to be used for commercial purposes are derived). You should fully characterize the final stabilized IGA in the animal using methods appropriate to the specific IGA(s). For example, for randomly inserted rDNA constructs, an evaluation of the site of insertion and confirmation of the expected sequence should be provided. For an IGA targeted to a specific site in the genome, that site should be evaluated to determine if the IGA was achieved as expected without introducing any unintended on-target alterations (e.g., unintended DNA integration or large deletions at the target site). You should also provide a description of the methods used and data evaluating the potential for unintended alteration(s) at the target site or elsewhere in the genome (e.g., mutations at potential off-target sites identified from *in silico* predictions or biochemical or cell-based assays).

FDA has funded collaborative research with the National Institute of Standards and Technology (NIST) to develop standardized approaches for evaluating genome editing as part of the VIP Sci-Assist program under VIP Plus, an expansion to our VIP program. These approaches are intended to serve as examples of how developers may characterize the IGA(s) in the animal's genome.

d. Phenotypic Characterization of Animals with IGAs

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

The application requires information regarding the health of the animals containing IGAs, including the target animal safety requirements of 21 CFR 514.1(b)(8) (see 21 U.S.C. § 321(u)). While target animal safety is not synonymous with "animal welfare," our review includes an evaluation of animal health to ensure animals containing the IGA maintain a state of well-being such that the animal has normal (or if intentionally altered by the IGA, the intended) body function, productivity, behavior, and absence of disease. We also assess whether the

environmental, behavioral, and nutritional needs of the animal can be met under ordinary conditions of animal care and whether or not specific instructions are needed to appropriately care for the animal.

For this section of the application, we recommend that you submit data regarding whether the IGA or its expression product(s) cause any toxicity as well as any risk to human users or animals other than the target animal. In general, we recommend that you compile and submit data and information addressing the health of these animals, which may include animal health and treatment records, growth rates, reproductive function, and behavior. In addition, where appropriate, we recommend that you submit data on the physiological status of the animals, including clinical chemistry, hematology, histopathology, and any post-mortem results. We recommend that you collect data from generation(s) of animals or, in the event that animals are not propagated via breeding (e.g., somatic cell nuclear transfer), from alternative grouping(s) of animals as close as possible to those intended for use in commerce.

2. Durability Assessment and Plan

As in section IV.C.1. *Product Characterization*, this section 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 IGA in the animal resulting from the specific alteration, and defining (identifying) the animal being evaluated, is durable — that there is a reasonable expectation that the IGA is stably inherited, and the phenotype is consistent and predictable. This would include developing a sampling plan for monitoring durability post-approval, as necessary.

For the genotypic durability assessment, we recommend that you use the results of studies demonstrating that the IGA is stably inherited. For the phenotypic durability portion of the assessment, we recommend that you submit data on the consistency of the introduced trait (based on the intended use) over multiple generations or, in the event that animals are not propagated via breeding (e.g., somatic cell nuclear transfer), through an alternative approach that you have discussed with CVM in advance. 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). However, if you believe it is not practical or feasible to gather data on two or more non-contiguous generations, we recommend you contact us to discuss potential alternative methods of demonstrating durability.

Your durability plan should include validated genotypic and phenotypic durability methods. The genotypic durability method is a method of identity with sufficient discrimination to determine (1) whether a given animal contains the IGA, and (2) whether the IGA has significantly changed from that which was evaluated to be safe and effective (i.e., a detection method for your IGA in its final stabilized genomic location(s) in the animal). The phenotypic durability method is a method that evaluates whether the intended phenotype was achieved (e.g., if an introduced protein is expressed or if a deletion has resulted in the absence or truncation of an expression product as intended). The amount of change considered significant could vary based on the type of IGA but, in general, significance is determined by whether the change impacts the safety or effectiveness of the IGA. One example of significant change would be any change that leads to

an altered phenotype (as identified via the phenotypic durability method). Another example of a significant change would be if an IGA is used to introduce an amino acid substitution into the encoded protein and a mutation occurs that changes which amino acid is encoded by the IGA. An example of a change that would not be significant would be if an IGA is used to completely inactivate a gene and a substitution occurs in close proximity to the deletion, so long as the gene is still completely inactivated.

We recommend that you consult with us on developing these assessments and plans.

3. Food Safety and Environmental Impact Assessments

a. Food Safety

This section addresses the food safety requirements in 21 CFR 514.1(b)(8). It focuses on the applicable risk questions identified above in relation to whether food derived from animals with IGAs is safe for humans or animals consuming edible products from the animals. Note that for biopharm animals of food-producing species that have multiple controls in place to help ensure that they will not enter the food supply, we will expect less information than described below.

The risk questions involved in determining food safety can be divided into two overall categories. The first addresses whether there is any toxicity, including allergenicity, via food consumption of any potential expression products resulting from the IGA, if applicable (assessed partly in section IV.C.1.b. *Molecular Characterization of the IGA*). The second category addresses potential toxicity associated with both the article and any potential expression products resulting from the IGA (e.g., whether location of the IGA or expression of products resulting from the alteration affects physiological processes in the resulting animal such that unintended food consumption hazards are created, or whether existing food consumption risks are increased). Plausible pathways for adverse outcomes via the food exposure pathway should be identified by determining whether there are any expected biologically relevant changes (1) to the physiology of the animal (assessed partly in section IV.C.1.d. *Phenotypic Characterization*), and (2) in the composition of edible tissues from the animals with IGAs that suggest reason for toxicological concern compared with the appropriate comparator.

CVM's evaluation of the product characterization components of the application informs the scope of the food safety evaluation. CVM evaluates the impact of the IGA and any identified unintended alterations that arose from the introduction of the IGA into the animal's genome, as well as the animal's overall health with respect to food safety. For example, if the IGA results in expression of a protein in the edible tissues, information regarding the digestibility and stability of the protein in the human gastrointestinal tract and its potential for causing human allergy may be needed to ensure that there are no food safety concerns. In other circumstances, if the IGA is intended to result in a change in the composition of the edible tissues, then a tissue compositional analysis may be warranted.

The evaluation for food safety is specific to each product and will be based on the integration of information from various components of the application for approval. In order to understand the specific requirements for your product, FDA recommends that you discuss how you intend to address food safety with us prior to conducting studies.

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

FDA participated in the Codex Alimentarius *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.[20] The information needed to establish food safety for food from animals with IGAs under an application is consistent with that described in the Codex Guideline.

b. Environmental Impact

This section addresses the environmental component of your application (21 CFR 514.1(b)(14)). We expect that, at least until we have more experience, applications for animals with IGAs would require preparation of an EA to determine whether such an approval will result in significant environmental impacts. An EA that demonstrates these animals do not significantly affect the quality of the human environment leads to a 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; reared in contained facilities vs. intended for free release). Therefore, we recommend that you contact and work closely with us on these issues before proceeding with preparation of the EA.

4. Effectiveness

The previous sections of the review process primarily address identity and safety issues. This last section of pre-market review addresses effectiveness, i.e., whether you have validated your claims for the characteristics that the animals with IGAs are intended to exhibit (21 CFR 514.1(b)(8)). For example, in the case of animals with an IGA that is intended to allow the animals to resist disease, you must demonstrate that those animals are indeed resistant to that disease. In the case of animals with an IGA that is intended to cause the animal to produce a non-food product (e.g., a human drug), you must 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 CDER. 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, the Human Foods Program, or CBER as appropriate.

[20] 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 (https://www.who.int/docs/default-source/food-safety/food-genetically-modified/cxg-068e.pdf).

V. Post-Approval Responsibilities

Once an IGA in an animal is approved, sponsors have on-going responsibilities including registration and listing, recordkeeping and reporting, and filing supplements (21 U.S.C. § 360, 21 U.S.C. § 356a, 21 CFR 514.80, 21 CFR 514.8). We recommend that you, the sponsor, use the general approach outlined below to fulfill these requirements, but that you work closely with us during the development of the animals and the durability plan in order to determine the specific data and information to submit. In some cases, FDA may find that certain post-market requirements, such as particular post-market reports, may not be appropriate for a particular product based upon risks and characteristics of the product. In such circumstances, FDA may indicate that it intends to exercise enforcement discretion for particular post-market requirements for that product.

Please note that post-approval responsibilities belong to the sponsor (or "applicant"), as the entity that owns the approved application.[21] If a farmer or producer independent of the sponsor acquires animals with IGAs and raises them at a farm or facility, and the farmer/producer is not the owner of the approved application, does not hold the application on behalf of the sponsor, and is not under the same ownership or control as the sponsor, then the farmer or producer is not the sponsor and the requirements described here do not apply to them. As noted in GFI #187A, if farmers, growers, and other entities just have animals with IGAs on their farms or other premises, including the offspring of those animals, and they are not developing an IGA in an animal or marketing the animals with any new claims, then, as a general matter, they do not have to register or list with FDA and can engage in ordinary activities (e.g., breeding, growing, etc.) without contacting FDA.

A. Statutory Registration and Listing Requirements

As part of the registration requirements under 21 U.S.C. § 360, you are required to register your establishment (i.e., your name and place of business) and identify any facility or facilities engaged in the production or testing of the IGAs in animals (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 approved IGAs in animals that you have produced. In the context of animals with IGAs, which facilities you must register may depend upon specific information about your product, such as the type of animal and your business plan, including where you develop the lineage of animals. We will discuss how the registration requirement applies to your product as part of the approval process. We note, though, that as a general matter we do not intend to require registration of pet stores, farms, or other animal production facilities simply because they have an animal with an IGA on the premises. Additionally, if you/the developer or a contract service provider has facilities that are only engaged in standard breeding practices, such as embryo transfer and cloning, we may not expect you or the contract service provider to register those facilities. Contact us if your development plan includes such facilities.

[21] "Nonapplicants" also have some post-approval reporting responsibilities (21 CFR 514.80). However, a "nonapplicant" is an entity whose name appears in product labeling and who is engaged in manufacturing, packing, distribution, or labeling of the product (21 CFR 514.3). We do not anticipate that a farmer or rancher would be a "nonapplicant" (though, depending upon their relationship with the sponsor, it is possible that they could be).

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 with IGAs that has not been previously submitted as part of the application (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 by the sponsor throughout the year and reported on an annual or semi-annual basis from a subset of marketed approved IGAs in animals in the "Minor Changes and Stability Report" under 21 CFR 514.8(b)(4). You should consult with us during the investigational file phased-review 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 where applicable).

You must submit information on all changes that have been made, or that you propose to make to the IGAs in animals (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 IGAs in animals (21 CFR 514.80). For example, you are required to submit reports of adverse events and product defects as specified in your approval. In addition, you must submit periodic experience reports containing data, studies, and other information related to experience every 6 months for the first 2 years following approval, and annually thereafter (21 CFR 514.80(b)(4)).

VI. Import Tolerances

Section 512(a)(6) of the FD&C Act enables FDA to establish safe residue levels of the IGA and any substance formed in or on food because of its use in edible portions of animals (i.e., food) imported into the United States (an import tolerance) when the IGA has not been approved or conditionally approved for use in the United States (see 21 CFR 510.201 *et seq.*). An IGA that is the subject of an import tolerance request may be one that is approved in another country or its use may otherwise be legal in another country and present in imported animal-derived food and food products. Any entity may request the import tolerance – for example, it may be the sponsor, a government entity, or a private organization. Whether a sponsor seeks approval of an application for an IGA in a lineage(s) of animals or establishment of an import tolerance for food

from such animals, the food safety standard is essentially the same.[22] If FDA determines that food from an animal with an IGA that is the subject of an import tolerance request is safe, then FDA would convey to the entity making the request that there is no need to establish an import tolerance for that IGA because any amount of the IGA would be safe to consume. Information about import tolerances, which enable imports of such food from animals with IGAs 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 Alimentarius *Guideline for the Conduct of Food Safety Assessment of Foods Derived from Recombinant-DNA Animals*. We recommend that you consult with CVM if you are interested in establishing an import tolerance.

VII. Additional Relevant Laws and Guidances

In addition to the ones described in this guidance, there are other laws that may apply to IGAs in animals and other guidances that may provide relevant recommendations. These include:

- Existing guidances and other documents prepared by other FDA Centers, including:
    - Points to Consider in the Manufacture and Testing of Therapeutic Products for Human Use Derived from Transgenic Animals (1995) https://www.fda.gov/media/76253/download;
    - Source Animal, Product, Preclinical, and Clinical Issues Concerning the Use of Xenotransplantation Products in Humans – Guidance for Industry (2016) https://www.fda.gov/media/102126/download;
    - Medical Devices Containing Materials Derived from Animal Sources (Except for *In Vitro* Diagnostic Devices) - Guidance for Industry and Food and Drug Administration Staff (2019) https://www.fda.gov/media/87251/download;
- Federal and State 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 National Institutes of Health 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. See USDA APHIS | Imports and USDA APHIS Exports.

[22] 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 application approval. 21 U.S.C. § 360b(a)(6); 21 CFR 510.206.