U.S. FOOD & DRUG ADMINISTRATION

September 30, 2025

Intralytix, Inc.
Attention: Alexander Sulakvelidze, Ph.D.
President & CEO
8681 Robert Fulton Drive
Columbia, MD 21046

Re: GRAS Notice AGRN 74 – Lytic Bacteriophage Preparation

Dear Dr. Sulakvelidze:

The Food and Drug Administration's (FDA) Center for Veterinary Medicine (CVM or we) refers to a generally recognized as safe (GRAS) notice, dated December 4, 2024, submitted by Intralytix, Inc., (Intralytix). The subject of the notice is a lytic bacteriophage preparation, (hereafter referred to as lytic bacteriophage preparation or the notified substance) to be used as a processing aid to help control *Salmonella enterica* in food for companion animals and livestock animals at a use level up to 1×10^8 plaque-forming units/gram (PFU/g). On January 9, 2025, CVM received an amendment containing additional information. The submission informs us of your conclusion that the subject of the submission is GRAS through scientific procedures. You were notified in a letter, dated January 17, 2025, that the GRAS notice, as amended, was acceptable for filing, and the notice was designated as animal GRAS notice number (AGRN) 74. On May 28, 2025, and June 10, 2025, CVM received amendments from the notifier containing additional information. We have completed our evaluation of AGRN 74 and have no questions at this time regarding the conclusion for use of the notified substance in pet food and ingredients for use in pet food.

To address the identity, method of manufacture, and specifications of the notified substance, Intralytix describes the identity, components, manufacturing process and controls, specifications and potential contaminants/impurities, and analytical methods used to establish the specifications of the notified substance. The notified substance consists of equal titers of up to six individually purified, lytic, double-stranded DNA bacteriophages specific to *S. enterica*. Each bacteriophage in the preparation is prepared using an aerobic fermentation process in which the bacteriophage and the appropriate host *Salmonella* strain are grown in a culture medium followed by filtration, concentration, wash, and final sterile filtration. The final preparation consists of up to six bacteriophages diluted with 0.1M sodium chloride, and with an equal and minimum lytic titer of (10.0 ± 0.33) $\log_{10}$ PFU/mL (i.e., $(0.5 \text{ to } 2.1)\times10^{10}$ PFU/mL).

However, we have questions regarding Intralytix's conclusion that the use of the notified substance is safe for use as a processing aid to help control *Salmonella enterica* in food for livestock animals at a use level up to 1×10^8 PFU/g for the following reason:

> The notice lacks data and information to demonstrate the notified substance controls the pathogenic *Salmonella* serotypes for livestock and poultry species and its ability

U.S. Food and Drug Administration
Center for Veterinary Medicine
CPK1, Room 1B068
5001 Campus Drive
College Park, MD 20740-3835
www.fda.gov

to achieve its intended effect in typical feed ingredients and matrices for livestock and poultry.

To address target animal safety of the intended use of the notified substance, Intralytix provides data and information describing safety and ubiquity of bacteriophages, the natural occurrence of phages in animal microbiota, environments, and in foods and drinking water. Intralytix describes the identity and specificity of the six bacteriophages to be used and provides specifications for the individual bacteriophages and for the notified substance. Intralytix provides a discussion of publicly available scientific literature to summarize that bacteriophages are ubiquitous and non-infectious to healthy humans and animals. Intralytix describes results of a publicly available scientific article which includes data from an in vivo feeding study in dogs and cats (Soffer et al., 2016). In the feeding trial, dogs and cats consumed kibble treated with the notified substance, at the use rate of approximately 2×10^7 PFU/g food, for 14 and 15 days respectively. No adverse events were reported, and Intralytix concludes that the intended use of the notified substance does not impact the gastrointestinal health of pets. Intralytix also discusses the ubiquitous nature and safety of bacteriophages consumed in food and drinking water in support of target animal safety. The level of bacteriophage added in the trial is minimal compared to that which is naturally present in the food of dogs and cats.

To address identity and microbial safety, Intralytix provides a narrative based on scientific data and literature that addresses genomic analysis, the prevalence of bacteriophages in humans, animals, and foods, bacteriophage lifecycles, pathogenicity, and toxin production to support its conclusion that the six bacteriophage strains included are GRAS to help control *S. enterica* in pet food and ingredients for use in pet food. Since the *Salmonella* host strains are known pathogens, Intralytix also provides the bioinformatics information indicating that these host strains do not produce any known *Salmonella* exotoxins other than lipopolysaccharides (i.e., endotoxins) which are the major components of a Gram-negative bacterial outer membrane. To address the utility of the notified substance in pet food, Intralytix provides two published studies of the notified substance against *S. enterica* isolates on kibble (Heyse et al., 2015), turkey trim, chicken, tuna, cantaloupe, and lettuce (Soffer et al., 2016). The publication by Heyse et al., (2015) also includes the lytic spectrum information of the notified substance. In addition, Intralytix provides three unpublished studies to corroboratively support the utility of the notified substance. These unpublished studies report a reduction of *S. enterica* by one- to two-log colony-forming units per gram on several raw pet foods when the notified substance was applied by several spray methods at the 1×10^8 PFU/g inclusion rate followed by thorough mixing.

Section 301(ll) of the Federal Food, Drug, and Cosmetic Act (FD&C Act)

Section 301(ll) of the FD&C Act prohibits the introduction or delivery for introduction into interstate commerce of any food that contains a drug approved under section 505 of the FD&C Act, a biological product licensed under section 351 of the Public Health Service Act, or a drug or a biological product for which substantial clinical investigations have been instituted and their existence made public, unless one of the exemptions in section 301(ll) (1)-(4) applies. In our evaluation of this notice, concluding that the notified substance is GRAS under its intended conditions of use, we did not consider whether section 301(ll) or any of its exemptions apply to foods containing the notified substance. Accordingly, our response should not be construed to be a statement that foods containing the notified substance, if introduced or delivered for introduction into interstate commerce, would not violate section 301(ll).

Conclusion

Based on the information contained in the notice, as amended, submitted by Intralytix, Inc., and other information available to the FDA, we have no questions at this time regarding your conclusion that the lytic bacteriophage preparation is GRAS as a processing aid to help control *S. enterica* in pet food and ingredients for use in pet food at an inclusion level up to 1×10^8 PFU/g. However, the notice does not provide a sufficient basis for a conclusion that the lytic bacteriophage preparation is GRAS under the condition of its intended use in food for livestock animals. The Agency has not made its own determination regarding the GRAS status of the intended use of the notified substance in animal food under 21 CFR 570.35. Unless noted above, our evaluation did not address other provisions of the FD&C Act. As always, it is the continuing responsibility of Intralytix, Inc., to ensure that animal food ingredients that you market are safe and are otherwise in compliance with all applicable legal and regulatory requirements.

In accordance with 21 CFR 570.275(b)(2), the text of this letter responding to AGRN 74 is accessible to the public on our website for the Current Animal Food GRAS Notices Inventory at https://www.fda.gov/animal-veterinary/generally-recognized-safe-gras-notification-program/current-animal-food-gras-notices-inventory.

If you have any questions or comments, please contact Ms. Lauren Howell at animalfood-premarket@fda.hhs.gov.

Sincerely,

/s/
Jeanette B. Murphy, M.S.
Acting Director
Office of Surveillance and Compliance
Center for Veterinary Medicine

References

Heyse, S., L. F. Hanna, J. Woolston, A. Sulakvelidze, and D. Charbonneau. 2015. Bacteriophage cocktail for biocontrol of *Salmonella* in dried pet food. J. Food Prot. 78:97-103. doi:10.4315/0362-028X.JFP-14-041.

Soffer, N., T. Abuladze, J. Woolston, M. Li, L. F. Hanna, S. Heyse, D. Charbonneau, and A. Sulakvelidze. 2016. Bacteriophages safely reduce *Salmonella* contamination in pet food and raw pet food ingredients. Bacteriophage 6:e1220347. doi:10.1080/21597081.2016.1220347.