

June 19, 2012

Mr. Devon Wm. Hill
Keller and Heckman, LLP
1001 G Street NW
Washington, DC 20001

Re: GRAS Notice No. AGRN 000-0010

Dear Mr. Hill:

The Food and Drug Administration (FDA) is responding to the notice, dated July 29, 2011 that you submitted, on behalf of Gevo, Inc. under FDA's Center for Veterinary Medicine (CVM) Pilot Program for substances generally recognized as safe (GRAS) added to food for animals (See 75 FR 31800; June 4, 2010). FDA's Center for Veterinary Medicine received the notice on August 1, 2011, filed it on August 25, 2011, and designated it as GRAS Notice No. AGRN 000-010.

The subject of your notice is inactivated modified *Saccharomyces cerevisiae* ("inactivated modified *S. cerevisiae*"). The notice informs FDA of the view of Gevo, Inc., that inactivated modified *S. cerevisiae* is GRAS, through scientific procedures, for use as a nutritional component in distillers grains products at levels up to 20% on a dry-weight basis for food-producing animals (beef cattle, dairy cows, broiler chickens, egg-laying chickens, swine and sheep) and pets (dogs, cats, rabbits, guinea pigs, and horses).

FDA has evaluated the information that Gevo, Inc. discusses in its GRAS notice as well as other data and information that are available to the agency. As discussed more fully below, the notice does not provide a sufficient basis for a determination that inactivated modified *S. cerevisiae* is GRAS under the conditions of its intended use in animal food.

Data and information that Gevo Inc. presents to support its GRAS determination

Gevo, Inc. provided information regarding the common name of the organism, an overview of the isobutanol fermentation process including temperature conditions and heat inactivation data. Gevo, Inc. states that a modified *S. cerevisiae* is used to produce isobutanol for use as a biofuel, with the resultant distillers products intended for use in food-producing animals (beef cattle, dairy cattle, sheep, swine, and poultry) and companion animal (dogs, cats, guinea pigs, rabbits, and horses) foods. The inactivated modified yeast is present in the distillers grains and serves as a source of nutrients for animals. Isobutanol distillers grains products are estimated to contain no more than 20% inactivated modified *S. cerevisiae* on a dry-weight basis, and complete diets will contain no more than 12% inactivated modified *S. cerevisiae*.

Gevo Inc. provides information on a *S. cerevisiae* strain that was bioengineered to produce isobutanol from pyruvate precursors, primarily glucose. The notifier states that the donor organisms for the genes that were inserted into the source organism were from *Escherichia*

coli, *Bacillus subtilis*, and *Lactococcus lactis*. The firm submitted phenotypic analysis data to demonstrate that antibiotic resistance genes were removed from the source strain.

To address target animal safety, Gevo, Inc. discussed the modified *S. cerevisiae* that Gevo, Inc. stated is substantially equivalent to the unmodified *S. cerevisiae*, which (1) has a long history of use in human foods, and (2) is also currently used in the production of ethanol and distillers products that are currently fed to food-producing animals and companion animals.

To address human food safety, Gevo, Inc. discussed the safety of the unmodified *S. cerevisiae* and the substantial equivalence of inactivated modified *S. cerevisiae* to inactivated unmodified *S. cerevisiae*. References to published toxicology studies pertaining to the safety evaluation of the notified substance were included in the notice.

FDA's evaluation of the data and information in Gevo, Inc.'s notice

FDA has the following comments regarding manufacturing chemistry:

1. Your notice did not describe the ingredients or processing aids, including appropriate specifications, used in the production of the yeast and isobutanol fermentation, nor indicate whether they are acceptable for use in animal feeds.
2. Your notice did not include analytical data and methodology to identify the fermentation products that modified *S. cerevisiae* produces, including isobutanol, nor analytical data and a discussion about which fermentation products and their levels that remain in the final distillers grain products.
3. You state that your determination of the GRAS status is on the basis of scientific procedures. The notice contained a vague description of the bioengineering process used to create the source organism. The notice does not describe the number of genes that were inserted into the host organism genome or the precise location of the insertions. The notice does not state whether the genes used in the bioengineering process were obtained directly from the donor organisms or whether the genes were synthesized. Data and information were not identified in the notice that the genes obtained from *Escherichia coli* were obtained from a nonpathogenic, non toxigenic strain. The phenotypic analysis data alone does not provide sufficient information to conclude that the source organism does not contain the antimicrobial resistance genes. The notice does not provide any information on the metabolic pathways that were disrupted, with the possible exception of ethanol pathway, to enhance isobutanol production.

FDA has the following comments regarding intended use:

4. We note that the nutritional composition of yeast cannot be easily differentiated from the nutritional composition of other components of distillers grains. Since the yeast strain was specifically bioengineered to produce isobutanol, it is unclear why this notice states that the bioengineered strain is GRAS for its provision of nutrients. The utility of this bioengineered yeast may be best described by its ability to produce isobutanol.
5. Although Gevo does indicate that the metabolic pathway for ethanol production was altered in the notified strain by inserting genes encoding for isobutanol production, no

data were provided to demonstrate that the bioengineered strain produces isobutanol. In addition, the notice does not contain a discussion of the common constituents of yeast and how the bioengineering of the source organism would be expected to affect these constituents and thus, yeast composition. The notice fails to address which of this information is available in the public domain and how the analyses of the distillers grains substantiate the firm's conclusions about the intended use of the inactivated modified *S. cerevisiae* as a source of nutrients.

6. In addition, adequate information was not provided discussing carbon flow and the differences between what would be expected between conventional yeast and the modified *S. cerevisiae*, nor was a discussion present about the impact of these metabolic changes on the composition of the modified *S. cerevisiae*. The GRAS notice also lacks information on how the bioengineering process affected yeast growth and proliferation, which directly impacts the modified *S. cerevisiae* content of resulting distillers grains.

FDA has the following comments regarding human food safety:

7. As mentioned elsewhere, adequate information to substantiate the equivalence to the unmodified *S. cerevisiae* was not provided; therefore, the notice did not provide sufficient information to demonstrate that the use of modified *S. cerevisiae* during the fermentation process would not pose issues of toxicological concern. A residue exposure assessment to address any remaining residue concerns may be appropriate if any human food safety hazards are identified.

FDA has the following comments regarding target animal safety:

8. Your GRAS determination is based on a substantial equivalence argument. You failed to support your conclusion using data or information to explain the similarities and differences between the bioengineered and host strains of *S. cerevisiae*, and how these support a conclusion about safe use of the microorganism. In addition, you did not include in your notice any assessment of safety impacts on the proposed animal species based on differences between the modified *S. cerevisiae* and conventional strains, particularly those related to the metabolic alterations and associated changes in products and their amounts.

FDA has the following administrative recommendations:

9. We note that the notice did not include consecutive page numbers throughout the entire document.
10. The notice should contain a separate bibliography listing all references.
11. Only the odd page numbers from Chapter 16 of “The Alcohol Textbook” were included in Appendix 3.

Conclusions

FDA has evaluated the data and information in AGRN 000-010 as well as other available information. For reasons discussed above, the notice does not provide a sufficient basis for a

determination that inactivated modified *S. cerevisiae* is GRAS under the conditions of its intended use in animal food.

Please be advised that the questions raised in this letter also may impact our assessment of your notice AGRN 000-013 for isobutanol distillers grains.

In accordance with the Federal Register notice announcing the CVM Pilot Program, a copy of the text of this letter responding to AGRN 000-010, and a copy of the information in this notice that conforms to the information described in your GRAS exemption claim is available for public review and copying via the FDA home page at <http://www.fda.gov>. To view or obtain an electronic copy of this information, follow the hyperlinks from the “Animal & Veterinary” topic to the “Products” section to the “Animal Food & Feeds” to the “Generally Recognized as Safe (GRAS) Notifications” page where the Animal Food GRAS Inventory is listed.

If you have any questions about this letter, please contact Dr. Andrea Krause at 240-276-9768 or by email at andrea.krause@fda.hhs.gov. Please reference AGRN 000-010 in any future correspondence regarding this submission. If Gevo, Inc. wishes to have FDA consider any new information regarding inactivated modified *S. cerevisiae*, the appropriate mechanism would be for the notifier to submit, in accordance with proposed 21 CFR 570.36, a complete GRAS notice. FDA would assign a new file number to a new notice regarding inactivated modified *S. cerevisiae*.

Sincerely,

/s/

Sharon A. Benz, Ph.D., PAS
Director
Division of Animal Feeds
Center for Veterinary Medicine