Maureen Dunn, ND
AIBMR Life Sciences, Inc.
1425 Broadway, Suite 458
Seattle, WA 98122

Re: GRAS Notice No. GRN 001143

Dear Dr. Dunn:

The Food and Drug Administration (FDA, we) completed our evaluation of GRN 001143. We received the notice that you submitted on behalf of Microbial Discovery Group (MDG) on April 8, 2023, and filed it on September 1, 2023. MDG submitted amendments to the notice on November 6, 2023, and December 18, 2023, that clarified the identity, manufacturing process, specifications, intended use, dietary exposure, and safety information.

The subject of the notice is *Bacillus subtilis* NRRL 68053 for use as an ingredient at a maximum level of 1 x 10^{10} colony forming units (CFU)/serving in conventional foods.[1] The notice informs us of MDG's view that these uses of *B. subtilis* NRRL 68053 are GRAS through scientific procedures.

MDG describes *B. subtilis* NRRL 68053 as a light beige to tan colored powder and states that *B. subtilis* NRRL 68053 is a non-pathogenic, non-toxigenic, Gram-positive, rod-shaped, spore forming bacterium. MDG notes that the strain was isolated from the mammalian gastrointestinal tract and has been deposited in the Agriculture Research Service Culture Collection (NRRL) with the depository number B-68053. MDG describes the taxonomic analysis for the identity of the strain. MDG also discusses the results of genomic analyses to confirm the strain identity and states that the strain is not genetically engineered. MDG discusses the results of phenotypic and genotypic characterization performed on *B. subtilis* NRRL 68053. MDG concludes that the virulent factor genes present are intrinsic to the species, the bioinformatically identified secondary metabolites are also present in other *B. subtilis* strains that have been used in food, and the tetracycline gene is unlikely transferrable.

MDG describes the manufacture of *B. subtilis* NRRL 68053 by fermentation of a pure culture under controlled conditions. After fermentation, the cells are separated from the fermentation medium and concentrated by centrifugation,

[1] MDG states that *B. subtilis* NRRL 68053 is not intended for use in infant formula, alcoholic beverages, products under the jurisdiction of the United States Department of Agriculture, or in foods where standards of identity preclude its use.

freeze-dried and milled to yield the final product, which is stored at ambient temperature. MDG states that *B. subtilis* NRRL 68053 is manufactured under current good manufacturing practices using food-grade raw materials. MDG confirms that all materials used in the manufacture of *B. subtilis* NRRL 68053 are permitted for their respective uses under a current U.S. regulation, are the subject of an effective food contact notification, or are GRAS for their intended use. MDG states that *B. subtilis* NRRL 68053 does not contain any major allergens.

MDG provides specifications for *B. subtilis* NRRL 68053 that include total cell count ($\geq 1.5 \times 10^{11}$ CFU/g), water activity (<0.350) and limits for microorganisms, including *Escherichia coli* (<10 CFU/g), *Salmonella* serovars (negative in 25 g), *Listeria* spp. (negative in 25 g), *Staphylococcus aureus* (<10 CFU/g), and heavy metals, including lead (≤0.1 mg/kg). MDG provides the results from the analyses of three non-consecutive batches to demonstrate that *B. subtilis* NRRL 68053 can be manufactured to meet these specifications.

MDG estimates the dietary exposure to *B. subtilis* NRRL 68053 from the intended uses to be 2×10^{11} CFU/day (d) based on the assumption that the average individual in the U.S. consumes 20 servings of food/d, and that all of these servings would contain *B. subtilis* NRRL 68053 at the maximum use level of 1×10^{10} CFU/serving.

MDG discusses data and information used to support the safety of *B. subtilis* NRRL 68053, including a history of safe use of the *B. subtilis* species in fermented foods. MDG incorporates into their notice and provides summaries of the information pertaining to the safety of the *B. subtilis* strains discussed in GRNs 000831, 000905, 000955, 000956, and 000969.[2] MDG discusses opportunistic infection caused by certain *B. subtilis* strains and states that the infection occurs at very low rates, and generally occurs in hospital settings in immunocompromised patients and/or during medical procedures. MDG summarizes published toxicology studies, human studies and reported adverse events on *B. subtilis*, and concludes that there are no indications of safety concerns.

Based on the totality of the data and information, MDG concludes that *B. subtilis* NRRL 68053 is GRAS for its intended use.

Standards of Identity

In the notice, MDG states its intention to use *B. subtilis* NRRL 68053 in several food categories, including foods for which standards of identity exist, located in Title 21 of the Code of Federal Regulations. We note that an ingredient that is lawfully added to food products may be used in a standardized food only if it is permitted by the applicable standard of identity.

[2] *B. subtilis* strains DE111, DSM32444, BS-MB40 PTA-122264, ATCC SD-7280 and ATCC SD-7780 were the subjects of GRNs 000831, 000905, 000955, 000956, and 000969, respectively. We evaluated these notices and responded in letters dated August 13, 2019, June 8, 2020, March 26, 2021, August 18, 2021, and October 6, 2021, respectively, stating that we had no questions at the time regarding the notifiers' GRAS conclusions.

Potential Labeling Issues

Under section 403(a) of the Federal Food, Drug, & Cosmetic (FD&C) Act, a food is misbranded if its labeling is false or misleading in any way. Section 403(r) of the FD&C Act lays out the statutory framework for labeling claims characterizing a nutrient level in a food or the relationship of a nutrient to a disease or health-related condition (also referred to as nutrient content claims and health claims). If products containing *B. subtilis* NRRL 68053 bear any nutrient content or health claims on the label or in labeling, such claims are subject to the applicable requirements and are under the purview of the Office of Nutrition and Food Labeling (ONFL) in the Center for Food Safety and Applied Nutrition. The Office of Food Additive Safety (OFAS) did not consult with ONFL on this issue or evaluate any information in terms of labeling claims. Questions related to food labeling should be directed to ONFL.

Section 301(ll) of the 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 MDG's notice concluding that *B. subtilis* NRRL 68053 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 *B. subtilis* NRRL 68053. Accordingly, our response should not be construed to be a statement that foods containing *B. subtilis* NRRL 68053, if introduced or delivered for introduction into interstate commerce, would not violate section 301(ll).

Conclusions

Based on the information that MDG provided, as well as other information available to FDA, we have no questions at this time regarding MDG's conclusion that *B. subtilis* NRRL 68053 is GRAS under its intended conditions of use. This letter is not an affirmation that *B. subtilis* NRRL 68053 is GRAS under 21 CFR 170.35. Unless noted above, our review did not address other provisions of the FD&C Act. Food ingredient manufacturers and food producers are responsible for ensuring that marketed products are safe and compliant with all applicable legal and regulatory requirements.

In accordance with 21 CFR 170.275(b)(2), the text of this letter responding to GRN 001143 is accessible to the public at www.fda.gov/grasnoticeinventory.

Sincerely,

Susan J. Carlson -S

Digitally signed by Susan J. Carlson -S
Date: 2024.01.19 17:42:30 -05'00'

Susan J. Carlson, Ph.D.
Director
Division of Food Ingredients
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