



Jessica Edward
Microbial Discovery Group, LLC.
7420 S Howell Ave, Suite 300
Oak Creek, WI 53154

Re: GRAS Notice No. GRN 001221

Dear Ms. Edward:

The Food and Drug Administration (FDA, we) completed our evaluation of GRN 001221. We received Microbial Discovery Group, LLC (MDG)'s GRAS notice on September 24, 2024, and filed it on January 10, 2025. MDG submitted amendments to the notice on March 27, 2025, April 17, 2025, April 30, 2025, May 7, 2025, and May 13, 2025, that clarified the manufacturing process, specifications, intended uses, dietary exposure and safety information.

The subject of the notice is *Heyndrickxia coagulans*¹ NRRL B-67744 (*H. coagulans* B-67744) spore preparation for use as an ingredient at a maximum level of 2×10^9 colony forming units (CFU)/serving in baked goods and baking mixes; non-alcoholic beverages and beverage bases; breakfast cereals; chewing gum; coffee and tea; condiments and relishes; confections and frosting; dairy product analogs; fresh fruit juices; frozen dairy desserts and mixes; fruit and water ices; gelatins, puddings and fillings; grain products and pastas; hard candy and cough drops; herbs, seeds, spices, seasonings, blends, extracts, and flavorings; jams and jellies; milk (whole and skim); milk products; nuts and nut products; plant protein products; processed fruits; processed vegetables and vegetable juices; snack foods; soft candy; soups and soup mixes; sugar; and sweet sauces, toppings and syrups.² The notice informs us of MDG's view that these uses of *H. coagulans* B-67744 spore preparation are GRAS through scientific procedures.

MDG describes *H. coagulans* B-67744 spore preparation as a light beige to tan colored powder and states that *H. coagulans* B-67744 is a non-pathogenic, non-toxicogenic, Gram-positive, rod-shaped, endospore forming bacterium. MDG notes that the strain was isolated from the mammalian gastrointestinal tract and has been deposited in the Agricultural Research Service Culture Collection with the depository number B-67744. MDG describes the taxonomic analysis for the identity of the strain. MDG also discusses the results of genomic analyses to confirm the

¹ We note that *Bacillus coagulans* was reclassified as *Weizmannia coagulans* in 2020 and *Heyndrickxia coagulans* in 2023 as reported in Gupta, et al. (Ref. 1) and Narsing Rao, et al. (Ref. 2), respectively.

² MDG states that *H. coagulans* B-67744 is not intended for use in infant formula, foods formulated for infants, products under the jurisdiction of the United States Department of Agriculture, alcoholic beverages, or in foods where standards of identity preclude its use.

strain identity and states that the strain is not genetically engineered. MDG discusses the results of phenotypic and genotypic characterization performed on *H. coagulans* B-67744 and states that *H. coagulans* B-67744 has no genes for pathogenicity and virulence, is lack of antibiotic resistance characteristics, and that the putative virulence factors found in the genome were involved in housekeeping functions.

MDG describes the manufacture of *H. coagulans* B-67744 spore preparation by fermentation of a pure culture under controlled conditions. After fermentation, the spore biomass is subjected to centrifugation, freeze-dried and milled to yield the final product that can be optionally mixed with a food-grade carrier to obtain a desired final concentration. MDG states that *H. coagulans* B-67744 spore preparation is manufactured under current good manufacturing practices and that all raw materials are food grade and are used in accordance with an applicable U.S. regulation, are GRAS for their intended use, or are the subject of an effective food contact notification. MDG states that *H. coagulans* B-67744 spore preparation does not contain any major allergens.

MDG provides specifications for *H. coagulans* B-67744 spore preparation that include total cell count ($\geq 1.50 \times 10^{10}$ CFU/g), water activity (< 0.350), limits for heavy metals, including lead (≤ 0.1 mg/kg) and microorganisms, including *Escherichia coli* (< 10 CFU/g), *Salmonella* spp. (not detected in 25 g), *Listeria* spp. (not detected in 25 g), and *Staphylococcus aureus* (< 10 CFU/g). MDG provides the results from the analyses of three non-consecutive batches to demonstrate that *H. coagulans* B-67744 spore preparation can be manufactured to meet these specifications.

MDG states that the intended uses of *H. coagulans* B-67744 spore preparation are the same as those in GRNs 000399, 000526, 000597, 000601, 000691, and 000949.³ MDG estimates the dietary exposure to *H. coagulans* B-67744 spore preparation from the intended uses to be the same as in GRN 000399, i.e., 36.4×10^9 CFU/person/d. MDG states that because the intended uses of *H. coagulans* B-67744 spore preparation are substitutional for the existing uses of other *H. coagulans* spore preparations, an increase in the dietary exposure to *H. coagulans* from the intended uses is not expected.

MDG discusses data and information used to support the safety of *H. coagulans* B-67744 spore preparation, including a history of safe use of the *H. coagulans* species in fermented foods. MDG incorporates into their notice and provides summaries of the information pertaining to the safety of live *H. coagulans* spore preparations in food discussed in GRNs 000399, 000526, 000597, 000601, 000691, and 000949. MDG also demonstrates genomic similarity of 98% between *H. coagulans* B-67744 and the notified *H. coagulans* strains in GRNs 000399, 000526, and 000949. MDG discusses

³ The subjects of GRNs 000399, 000526, 000597, 000601, 000691, and 000949 are *B. coagulans* GBI-30 6086, Unique IS2, SNZ1969, MTCC 5856, SANK 70258, and DSM 17654 spore preparations, respectively. We evaluated these notices and responded in letters dated July 31, 2012, March 23, 2015, February 29, 2016, August 28, 2017, and January 7, 2021, respectively, stating that we had no questions at that time regarding the notifiers' GRAS conclusions.

opportunistic infection caused by certain *H. coagulans* strains and states that there is no concern for pathogenicity or opportunistic infection from oral consumption of *H. coagulans* because the opportunistic infections were not due to direct ingestion of the bacterium. MDG summarizes published toxicology studies and human studies on *H. coagulans*, and states that no toxicity was reported in animal studies and no adverse effects were noted in human studies. MDG concludes that there are no indications of safety concerns for *H. coagulans* B-67744 spore preparation.

Based on the totality of the data and information, MDG concludes that *H. coagulans* B-67744 spore preparation is GRAS for its intended use.

Standards of Identity

In the notice, MDG states its intention to use *H. coagulans* B-67744 spore preparation 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.

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 *H. coagulans* B-67744 spore preparation 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 Nutrition Center of Excellence. The Office of Pre-Market Additive Safety (OPMAS) 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 *H. coagulans* B-67744 spore preparation 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 *H. coagulans* B-67744 spore preparation. Accordingly, our response should not be construed to be a statement that foods containing *H. coagulans* B-67744 spore preparation, if introduced or delivered for introduction into interstate commerce, would

not violate section 301(l).

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 *H. coagulans* B-67744 spore preparation is GRAS under its intended conditions of use. This letter is not an affirmation that *H. coagulans* B-67744 spore preparation 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 001221 is accessible to the public at www.fda.gov/grasnoticeinventory.

Sincerely,

Susan J. Carlson -S

Digitally signed by Susan J.
Carlson -S
Date: 2025.06.09 11:58:26 -04'00'

Susan J. Carlson, Ph.D.
Director
Division of Food Ingredients
Office of Pre-Market Additive Safety
Office of Food Chemical Safety, Dietary
Supplements, and Innovation
Human Foods Program

References:

- 1) Gupta RS, Patel S, Saini N, Chen S (2020). "Robust demarcation of 17 distinct *Bacillus* species clades, proposed as novel *Bacillaceae* genera, by phylogenomics and comparative genomic analyses: description of *Robertmurraya kyonggiensis* sp. nov. and proposal for an emended genus *Bacillus* limiting it only to the members of the Subtilis and Cereus clades of species". *International Journal of Systematic and Evolutionary Microbiology*. **70** (11): 5753–5798.
- 2) Narsing Rao MP, Banerjee A, Liu GH, Thamchaipenet A (2023). "Genome-based reclassification of *Bacillus acidicola*, *Bacillus pervagus* and the genera *Heyndrickxia*, *Margalitia* and *Weizmannia*". *International Journal of Systematic and Evolutionary Microbiology*. **73** (7):1-7.