



Shahrzad Tafazoli, MASC, MSc, PhD
Intertek Health Sciences Inc.
2233 Argentia Rd., Suite 201W
Mississauga, ON L5N 2X7
CANADA

Re: GRAS Notice No. GRN 001269

Dear Dr. Tafazoli:

This letter corrects our response letter to GRN 001269 signed on February 4, 2026. The purpose of this revised letter is to correct a typographical error in the first paragraph of the original response letter. The original response letter incorrectly referred to the notice as GRN 001169.

The Food and Drug Administration (FDA, we) completed our evaluation of GRN 001269. We received the notice that you submitted on behalf of Amai Proteins (Amai) on May 6, 2025, and filed it on August 7, 2025. Amai submitted an amendment to the notice on November 17, 2025, providing clarifying information regarding the manufacturing process, specifications, production organism, and safety narrative.

The subject of the notice is monellin preparation produced by *Komagataella phaffii* CBS 150005 expressing a gene encoding for a modified monellin (monellin preparation) for use as a general-purpose sweetener in food at levels consistent with good manufacturing practices. Amai states that monellin preparation is not intended for use in infant formula, infant food products, or in products under the jurisdiction of the United States Department of Agriculture. The notice informs us of Amai's view that this use of monellin preparation is GRAS through scientific procedures.

Our use of the terms, "monellin preparation produced by *Komagataella phaffii* CBS 150005 expressing a gene encoding for a modified monellin" or "monellin preparation" in this letter is not our recommendation of that term as an appropriate common or usual name for declaring the substance in accordance with FDA's labeling requirements. Under 21 CFR 101.4, each ingredient must be declared by its common or usual name. In addition, 21 CFR 102.5 outlines general principles to use when establishing common or usual names for nonstandardized foods. Issues associated with labeling and the common or usual name of a food ingredient are under the purview of the Office of Nutrition and Food Labeling (ONFL) in the Nutrition Center of Excellence (NCE). The Office of Pre-Market Additive Safety (OPMAS) did not consult with ONFL regarding the appropriate common or usual name for "monellin preparation."

Amai describes monellin preparation as a white-to-yellowish powder containing $\geq 70\%$

(w/w) protein, of which $\geq 85\%$ is a monellin protein possessing sweetening properties. The monellin protein is a single-chain protein, composed of 93 amino acids, with a molecular weight of approximately 10.8 kDa. Amai explains that the primary sequence of the monellin protein was designed through specific substitutions and deletions of the amino acids in another single-chain form of monellin (MNEI)¹ to improve biophysical and biochemical properties (thermostability, digestibility, sensory profile, and pH stability). Amai states that monellin protein shares 92% sequence identity with MNEI and 94% sequence identity with wild-type monellin naturally present in the fruit of the West African *Dioscoreophyllum cumminsii* plant.

Amai reports that the production organism, *K. phaffii* CBS 150005, is non-pathogenic and non-toxicogenic. Amai states that the production organism is constructed from *K. phaffii* CBS 7435 mut^s Δ ARG4 Δ HIS4, which is derived from *K. phaffii* NRRL Y-11430. *K. phaffii* NRRL Y-11430 is a well-characterized yeast strain with a history of safe use in the food industry. Amai states that the gene encoding for the monellin protein was synthesized *de novo* and optimized for its biosynthesis and secretion. Amai states that the genomic stability was confirmed using Sanger sequencing and polymerase chain reaction amplification. Amai states that the production organism does not contain any antibiotic resistance genes or allergenic elements. Amai states that the production organism has been deposited in an international microbial culture collection bank, the Westerdijk Fungal Biodiversity Institute, with a designation number of CBS 150005.

Amai states that monellin preparation is manufactured through a fermentation process under sterile and controlled conditions. Amai states that monellin protein expressed by the production organism is secreted into the fermentation medium and separated from the biomass, followed by ion-exchange chromatography and ultrafiltration to remove fermentation media residues, host cells debris, DNA, antifoam, and mineral ions. The concentrated, purified solution is then spray-dried to yield the final monellin preparation. Amai states that none of the raw materials used in the manufacturing process are, or are derived from, major food allergens. Amai states that all raw materials, processing aids, filtration aids, and pH adjusters used are food-grade or pharmaceutical-grade and are either approved under appropriate U.S. regulations or GRAS for their intended use.

Amai provides specifications for monellin preparation that include total protein content ($\geq 70\%$ w/w), monellin protein as percent of total protein ($\geq 85\%$), and limits for moisture ($\leq 10\%$), ash ($\leq 10\%$), carbohydrates ($\leq 20\%$), heavy metals, including lead (≤ 0.1 mg/kg), and microorganisms, including *Salmonella* spp. (absent in 10 g). Amai provides the results from the analyses of three non-consecutive batches to demonstrate that monellin preparation can be manufactured to meet these specifications. Amai provides the results from the stability studies indicating that monellin preparation is stable for 18 months under ambient conditions (30 °C, 60% relative humidity).

Amai provides dietary exposure estimates for monellin preparation based on the relative

¹ MNEI is composed of monellin chains A and B linked together by a glycine-phenylalanine (Gly-Phe) dipeptide linker.

sweetness intensity of the notified substance (adjusted for the content of monellin protein) and the methodology described in Renwick (2008).² This study reported average and upper percentile (i.e., 90th percentile and higher) estimates of dietary exposure to intense sweeteners among children and adults with and without diabetes and estimated the dietary exposure to a sweetener based on its relative sweetness and an assumption of its substitutional use. Amai states that monellin preparation is approximately 3,500 times sweeter than a 6% sucrose solution and consists of approximately 83% monellin protein. Based on these assumptions, Amai estimates the average and upper percentile dietary exposures to monellin preparation for non-diabetic adults (0.09 and 0.23 mg/kg body weight (bw)/d, respectively), diabetic adults (0.1 and 0.31 mg/kg bw/d, respectively), non-diabetic children (0.15 and 0.34 mg/kg bw/d, respectively), and diabetic children (0.23 and 0.31 mg/kg bw/d, respectively).

Amai discusses publicly available data and information supporting the safety of monellin preparation. Amai characterizes the monellin protein as a sweet protein similar to monellin proteins present in the fruit of the West African *D. cumminsii* plant. Amai notes previous human consumption of *D. cumminsii* fruit as part of the diet in endemic regions of Africa, suggesting previous consumption of monellin as a component of human food. Amai describes the biochemical mechanism of the monellin protein effects on sweet taste perception and notes that the biochemical mechanism is similar to that of other known sweet proteins. Amai thoroughly characterizes the sensory profile of monellin preparation and notes the sweetening effects are transient and unlikely to cause downregulation or desensitization of sweet taste receptors.

Amai summarizes the results of a comprehensive literature search through March 2025 to identify available safety information relevant to monellin preparation. Based on the results of published studies, Amai concludes that monellin preparation is non-mutagenic and non-genotoxic. Amai provides a summary of a published 90-day oral toxicity study with monellin preparation to support the safety of its intended use. Additionally, Amai summarizes additional published genotoxicity and oral toxicity studies of similar single-chain monellin test articles to provide further corroborative evidence to support the safety of monellin preparation. Amai further describes the findings of an unpublished double-blind randomized crossover study in which no adverse effects on assessed clinical chemistry parameters were reported in healthy adults (n=18) administered 51 mg of monellin preparation. Based on the weight of evidence, including results of *in vitro* digestibility assays and *in silico* sequence-alignment-based approaches to assessing allergenicity, Amai concludes that monellin preparation does not pose an allergenic or toxicogenic risk to consumers. Amai does not identify any safety concerns or information that would contradict its GRAS conclusion.

Based on the totality of information, Amai concludes that monellin preparation is GRAS for its intended use.

² Renwick, A.G. 2008. The use of a sweetener substitution method to predict dietary exposures for the intense sweetener rebaudioside A. *Food and Chemical Toxicology* 46:S61–S69.

Standards of Identity

In the notice, Amai states its intention to use monellin preparation in several food categories, including foods for which standards of identity exist, located in Title 21 of the CFR. 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, and 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 monellin 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 ONFL in NCE. 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(l) of the FD&C Act

Section 301(l) 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(l)(1)-(4) applies. In our evaluation of Amai's notice concluding that monellin preparation is GRAS under its intended conditions of use, we did not consider whether section 301(l) or any of its exemptions apply to foods containing monellin preparation. Accordingly, our response should not be construed to be a statement that foods containing monellin preparation, if introduced or delivered for introduction into interstate commerce, would not violate section 301(l).

Conclusions

Based on the information that Amai provided, as well as other information available to FDA, we have no questions at this time regarding Amai's conclusion that monellin preparation is GRAS under its intended conditions of use. This letter is not an affirmation that monellin 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 001269 is accessible to the public at www.fda.gov/grasnoticeinventory.

Sincerely,

Susan J.

Carlson -S

Digitally signed by Susan J.
Carlson -S
Date: 2026.02.05 15:06:04 -05'00'

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