



Mary M. Murphy, MS, RD
Exponent, Inc.
1150 Connecticut Ave., NW
Suite 1100
Washington, DC 20036

Re: GRAS Notice No. GRN 001183

Dear Ms. Murphy:

The Food and Drug Administration (FDA, we) completed our evaluation of GRN 001183. We received the notice that you submitted on behalf of Oobli, Inc. (Oobli) on February 28, 2024, and filed it on May 30, 2024. Oobli submitted an amendment to the notice on September 18, 2024, clarifying information about the identity, manufacturing process, specifications, and mechanism of the technical effect.

The subject of the notice is monellin preparation produced by *Komagataella phaffii* P-MON-040¹ expressing a gene encoding for a modified monellin (monellin preparation), for use as a general-purpose sweetener in foods at levels consistent with good manufacturing practices.² The notice informs us of Oobli's view that this use of monellin preparation is GRAS through scientific procedures.

Our use of the terms, "monellin preparation produced by *Komagataella phaffii* P-MON-040 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."

Oobli provides information about the identity and composition of monellin preparation. Monellin preparation is produced using a genetically engineered strain of *K. phaffii* that expresses a gene encoding for monellin, the component responsible for monellin

¹ FDA notes that *Pichia pastoris* was reclassified as *K. phaffii* as reported in Kurtzman (Ref. 1).

² Oobli states that monellin preparation is not intended for use in products under the jurisdiction of the United States Department of Agriculture or in infant formula.

preparation's sweet-tasting properties. Oobli states that monellin preparation is an off-white powder containing primarily protein (minimum of 70%), of which more than 60% of the total protein is monellin, and the remaining protein consists of *K. phaffii* proteins carried over from the fermentation process. The ingredient also contains carbohydrates, moisture, ash, and fat. Oobli states that monellin preparation is synthesized *de novo* and consists of two monellin peptide chains, A and B, linked together by an additional glycine amino acid to form a single chain. Oobli states that peptide chains A and B are identical to peptide chains naturally present in the fruit of *Dioscoreophyllum cumminsii*. Oobli states that the primary sequence of the single-chain monellin is 96 amino acids with a molecular weight of 11.264 kDa.

Oobli reports that the production organism, *K. phaffii* P-MON-040, is a non-pathogenic, non-toxicogenic, and well-characterized yeast strain with a history of safe use in the food industry. Oobli states that the production organism is derived from *K. phaffii* BG10, which is derived from *K. phaffii* NRRL Y-11430. Oobli states that the production organism is constructed through a series of transformations with different expression constructs to enable the biosynthesis of monellin. Oobli states that the production organism does not contain any antibiotic resistance genes or plasmids and is not capable of transferring plasmids or antibiotic resistance to other organisms.

Oobli states that monellin preparation is manufactured through the controlled fermentation of the production organism. Monellin is secreted into the fermentation medium and then separated from the biomass by centrifugation. The fermentation supernatant containing monellin is concentrated, pH-adjusted, and subjected to a sequence of microfiltration and ultrafiltration steps. The resulting solution is spray-dried to obtain the final powder form of monellin preparation. Oobli states that none of the raw materials used during the manufacturing process are, or are derived from, major food allergens. Oobli states that all raw materials, processing aids, filtration aids, and pH adjusters used in the production of monellin preparation are food grade or pharmaceutical grade and are approved for their respective uses in accordance with an appropriate U.S. regulation or are GRAS for their intended use.

Oobli provides specifications for monellin preparation that include total protein content (>70% w/w), monellin as percent of total mass (>50% w/w), and limits for moisture (<10% w/w), ash (<5% w/w), fat (<1% w/w), carbohydrates (<15% w/w), heavy metals, including lead (<0.1 mg/kg), and microorganisms, including *Salmonella* serovars (absent in 10 g). Oobli provides the results from the analyses of three non-consecutive batches to demonstrate that monellin preparation can be manufactured to meet the stated specifications. Oobli evaluates the shelf-life stability of monellin preparation at accelerated conditions (35 °C, 65% relative humidity) over a period of 8 months.

Oobli provides estimates of dietary exposure to monellin preparation based on the relative sweetness intensity of the notified substance and the methodology presented in Renwick, 2008 (Ref. 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.

Based on the methodology described in Renwick, 2008, the estimated relative sweetness intensity of monellin preparation (2700 times sweeter than sucrose), and on the content of 63% monellin as the total mass of monellin preparation, Oobli estimates the average and upper percentile dietary exposures to monellin preparation for non-diabetic adults (0.15 and 0.40 mg/kg body weight (bw)/day (d), respectively), diabetic adults (0.16 and 0.53 mg/kg bw/d, respectively), non-diabetic children (0.25 and 0.58 mg/kg bw/d, respectively), and diabetic children (0.40 and 0.53 mg/kg bw/d, respectively).

Oobli discusses publicly available data and information supporting the safety of monellin preparation and its production organism, *K. phaffii*. Oobli describes monellin as a sweet protein found in the fruit of the West African *D. cumminsii* plant. Oobli 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. Oobli describes the biochemical mechanism of monellin's effects on sweet taste perception and notes that the biochemical mechanism is similar to that of other known sweet proteins.

Oobli summarizes the results of a comprehensive literature search to identify available safety information relevant to monellin produced by *K. phaffii*. Oobli does not identify any safety concerns or information that would contradict its GRAS conclusion. Oobli provides a summary of a published 90-day subchronic oral toxicity study with a test material similar to their article of commerce to support the safety of the intended use of monellin preparation. Based on published studies, Oobli concludes that monellin preparation is non-mutagenic and non-genotoxic. Oobli acknowledges the well characterized metabolic fate of dietary proteins. Based on the weight of evidence, including results of published *in vitro* digestibility assays and *in silico* sequence-alignment-based approaches, Oobli concludes that monellin preparation and copurified *K. phaffii* proteins do not pose an allergenic or toxicogenic risk to consumers. Oobli states that the safety of *K. phaffii* and any derived proteins is further supported by other GRAS conclusions for protein ingredients produced by *K. phaffii* strains (see GRNs 000737, 000967, 001001, 001056, and 001104).³

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

Standards of Identity

In the notice, Oobli 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

³ The subjects of GRNs 000737, 000967, 001001, 001056, and 001104 are soy leghemoglobin preparation from a strain of *Pichia pastoris*, soluble egg-white protein produced by *K. phaffii* strain GSD-1209, myoglobin preparation from a strain of *P. pastoris* expressing the myoglobin gene from *Bos taurus*, β -lactoglobulin produced by *K. phaffii* strain yRMK-66, and egg-white protein produced by *K. phaffii* ATCC GSD-1235, respectively. We evaluated these notices and responded in letters dated July 23, 2018, September 9, 2021, December 3, 2021, February 15, 2023, and October 17, 2023, respectively, stating that we had no questions at that time regarding the notifiers' GRAS conclusions.

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 Act (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 the 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(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 Oobli's notice concluding that monellin 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 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(ll).

Conclusions

Based on the information that Oobli provided, as well as other information available to FDA, we have no questions at this time regarding Oobli'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 001183 is accessible to the public at www.fda.gov/grasnoticeinventory.

Sincerely,

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Mical E. Honigfort -S
Date: 2024.11.22
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for 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. Kurtzman, C. (2005). Description of *Komagataella phaffii* sp. nov. and the transfer of *Pichia pseudopastoris* to the methylotrophic yeast genus *Komagataella*. *International Journal of Systematic and Evolutionary Microbiology*, 55, 973-976. doi: 10.1099/ijs.0.63491-0
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.