Dietrich Conze, Ph.D.
Spherix Consulting Group, Inc.
751 Rockville Pike, Unit 30-B
Rockville, MD 20852

Re: GRAS Notice No. GRN 001016

Dear Dr. Conze:

The Food and Drug Administration (FDA, we) completed our evaluation of GRN 001016. We received the notice that you submitted on behalf of Chr. Hansen A/S (Chr. Hansen) on June 8, 2021, and filed it on September 24, 2021. Chr. Hansen submitted amendments to the notice on January 28, 2022, April 26, 2022, and May 31, 2022, revising intended uses; clarifying food categories, dietary exposure estimates, and specifications; correcting dietary exposure calculations and intended use levels; and providing additional information on the narrative.

The subject of the notice is 6′-sialyllactose sodium salt (6′-SL) for use as an ingredient at 0.4 g/L in formulas for young children (> 12 months);[1] 2.28 g/L in milk-based meal replacement beverages for children and meal replacements drinks for adults; 1.5 g/kg in instant and ready-to-eat cereals for infants and young children under 3 years of age, snack bars, breakfast bars, and meal replacement bars; 0.7 g/L in non-carbonated sports drinks and flavored waters; and 3.8 g/L in food for enteral feeding. The notice informs us of Chr. Hansen's view that these uses of 6′-SL are GRAS through scientific procedures.

Chr. Hansen states that 6′-SL is an oligosaccharide that is found in human milk. 6′-SL is a trisaccharide with lactose present at the reducing terminus and sialic acid at the non-reducing terminus, linked by an α-2,6 bond. Chr. Hansen's 6′-SL is a white- to ivory-colored powder containing ≥90% 6′-SL on a dry matter basis and small amounts of carbohydrate by-products, ash, and moisture, and is identified by the CAS Registry Number 35890-39-2.

Chr. Hansen states that 6′-SL is produced by fermentation using a genetically engineered strain of *Escherichia coli* BL21(DE3) following the same process as described in GRN 000922,[2] except cobalt is no longer used in the fermentation medium. The descriptions of the production strain, *E. coli* BL21(DE3) strain DSM 33493, and

[1] GRN 001016 does not include the intended use in infant formula.

[2] 6′-SL was the subject of GRN 000922. We evaluated this notice and responded in a letter dated April 23, 2021, stating that we had no questions at that time regarding the notifier's GRAS conclusion.

manufacturing process in GRN 000922 are incorporated into GRN 001016.[3] As described in GRN 000922, and incorporated into this notice, *E. coli* BL21(DE3) strain DSM 33493 is non-pathogenic and non-toxigenic, and is deposited in the Deutsche Sammlung von Mikroorganismen und Zellkulturen (DSMZ) strain collection in Braunschweig, Germany.

Chr. Hansen provides specifications for 6′-SL as follows: 6′-SL (≥90%, dry weight), sum of other carbohydrates (≤ 10%[4]), D-lactose (≤ 5%), sialic acid (≤ 10%), N-acetylglucosamine (≤ 5%), protein (≤ 100 μg/g), ash (≤ 8.5%), moisture (≤ 9.0%), sodium (≤ 4.2%), heavy metals including lead (≤ 0.02 mg/kg), and limits for microorganisms, including *Salmonella* serovars (absent in 25 g) and *Cronobacter* spp.[5] (absent in 10 g). Chr. Hansen provides results from the analyses of five non-consecutive batches to demonstrate that 6′-SL can be manufactured to meet these specifications. Chr. Hansen states that all methods used for the analyses are validated methods that are fit for purpose. Chr. Hansen states that 6′-SL has been shown to be stable for 1 year when stored under ambient conditions.

Chr. Hansen estimates the eaters-only dietary exposure to 6′-SL from its intended uses using food consumption data from the 2015-2016 National Health and Nutrition Examination Survey. Chr. Hansen estimates dietary exposure to 6′-SL to be 0.09 g/person (p)/day (d) at the mean and 0.14 g/p/d at the 90th percentile for young children aged 13 months to 2 years. Chr. Hansen estimates dietary exposure to 6′-SL to be 0.3 g/p/d at the mean and 0.7 g/p/d at the 90th percentile for the U.S. population aged 2 years and older. Chr. Hansen applied the maximum use level for all food uses of 6′-SL, including uses in GRN 001016 as well as uses in earlier GRAS notices for 6′-SL (GRNs 000881[6] and 000922), to estimate a cumulative dietary exposure to 6′-SL. For the U.S. population aged 2 years and older, the cumulative dietary exposure to 6′-SL is estimated to be 208 mg/p/d (3 mg/kg body weight (bw)/d) at the mean, and 420 mg/p/d (6 mg/kg bw/d) at the 90th percentile. Chr. Hansen concludes that the intended uses do not increase the dietary exposure to 6′-SL.

Chr. Hansen states that 6′-SL will be partially substitutional to other forms of 6′-SL currently in use in conventional foods.

Chr. Hansen states that its 6′-SL is structurally identical to the 6′-SL in human milk and that the intended use level of 6′-SL is within the published range of 6′-SL that occurs in

[3] In GRN 000922, the notifier states that all raw materials, processing aids and medium ingredients used in the manufacture of 6′-SL are food grade and used in accordance with U.S. regulations or determined to be GRAS for their intended use.

[4] In the amendment dated January 28, 2022, Chr. Hansen described "other carbohydrates" as residual carbohydrates that are structurally related to 6′-SL.

[5] In an amendment dated May 31, 2022, Chr. Hansen clarified that their specification is for *Cronobacter* spp. using the test method ISO 22964. Chr. Hansen also stated that if any *Cronobacter* spp. are detected the batch would not be released.

[6] 6′-SL was the subject of GRN 000881. We evaluated this notice and responded in a letter dated April 13, 2020, stating that we had no questions at that time regarding the notifier's GRAS conclusion.

human milk. Chr. Hansen states that 6′-SL is resistant to digestion, poorly absorbed, and it passes through the gastrointestinal tract where it is either fermented by the gut microbiota or excreted unchanged.

Chr. Hansen incorporates safety data and information discussed in GRN 000922 and GRN 000881 into the current notice. Chr. Hansen provides a summary of published pivotal toxicology studies including genotoxicity studies and 90-day repeat dose feeding studies in rats conducted using 6′-SL. The study authors concluded that the feeding studies demonstrate the lack of toxicologically relevant effects even at the highest dose tested (5 g/kg bw/d). The study authors concluded that the mutagenicity and genotoxicity studies demonstrate that 6′-SL is not mutagenic or genotoxic. Chr. Hansen discusses additional toxicology studies in which 6′-SL was tested alone or in combination with other human milk oligosaccharides (HMOs), and the study authors concluded that there were no toxicologically relevant effects.[7] Chr. Hansen discusses published 21-day tolerance studies in neonatal piglets conducted using either 6′-SL (highest dose: about 1160 mg/L) or a mixture containing 6′-SL and other HMOs, and the study authors concluded that 6′-SL was well-tolerated alone or in combination at all doses over the 21-day treatment. Chr. Hansen also cites multiple human studies that include studies in both infants and adults. The study authors concluded that these studies demonstrate that the ingestion of 6′-SL and other related HMOs with similar metabolic fate, such as 3′-SL, 2′-FL, and LNnT, is well-tolerated in infants, children, and adults. At high doses in adults, such as around 20 g or more/day, the most common effects observed were occasional flatulence, abdominal distress, diarrhea, and loose stool.

Chr. Hansen includes the statement of a panel of individuals (Chr. Hansen's GRAS panel). Based on its review, Chr. Hansen's GRAS panel concluded that 6′-SL is safe under the conditions of its intended use.

Based on the results of the toxicology studies and human tolerance studies, Chr. Hansen concludes that the consumption of 6′-SL is GRAS for its intended use.

Standards of Identity

In the notice, Chr. Hansen states its intention to use 6′-SL 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 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

[7] FDA did not evaluate the use of 6′-SL in combination with other HMOs during our evaluation of GRN 001016.

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 6′-SL 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). The Office of Food Additive Safety 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.

Allergen Labeling

The FD&C Act requires that the label of a food that is or contains an ingredient that contains a "major food allergen" declare the allergen's presence (section 403(w)). The FD&C Act defines a "major food allergen" as one of nine foods or food groups (i.e., milk, eggs, fish, Crustacean shellfish, tree nuts, peanuts, wheat, soybeans, and sesame (effective January 1, 2023)) or a food ingredient that contains protein derived from one of those foods. 6′-SL requires labeling under the FD&C Act because it contains protein derived from milk.

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 Chr. Hansen's notice concluding that 6′-SL 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 6′-SL. Accordingly, our response should not be construed to be a statement that foods containing 6′-SL, if introduced or delivered for introduction into interstate commerce, would not violate section 301(ll).

Conclusions

Based on the information that Chr. Hansen provided, as well as other information available to FDA, we have no questions at this time regarding Chr. Hansen's conclusion that 6′-SL is GRAS under its intended conditions of use. This letter is not an affirmation that 6′-SL 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 001016 is accessible to the public at www.fda.gov/grasnoticeinventory.

Sincerely,

Susan J. Carlson -S

Digitally signed by Susan J. Carlson -S
Date: 2022.07.15 17:15:28 -04'00'

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