Christoph H. Röhrig, Ph.D.
Glycom A/S
Kogle Allé 4
2970 Hørsholm
DENMARK

Re: GRAS Notice No. GRN 000895

Dear Dr. Röhrig:

This letter revises our response letter to GRN 000895 signed on August 28, 2020. The purpose of the revised letter is to correct the typographical error in the name "lactose-N-triose II" to "lacto-N-triose II" in paragraph 1 on page 2, revise the specifications to include a minimum for total saccharides in paragraph 3 on page 2, and correct errors in footnote 3 to include lacto-N-neotetraose (LNnT) fructose isomer in the sum of specified saccharides. We are also revising the language regarding the drying manufacturing step in paragraph 2 on page 2, as it is not restricted to freeze- or spray-drying.

The Food and Drug Administration (FDA, we) completed our evaluation of GRN 000895. We received Glycom A/S (Glycom)'s notice on December 9, 2019 and filed it on January 15, 2020. Glycom submitted amendments to the notice on April 2 and April 28, 2020 that clarified the intended uses, allergen information and analytical methods used to verify specifications, and corrected typographical errors.

The subject of the notice is LNnT for use as an ingredient at levels up to 0.6 g/L in non-exempt infant formulas for term infants;[1] 0.58 g/L in drinks for young children; 0.6 g/L in toddler formula (> 12 months of age); 3 g/kg in foods for infants and young children; 2.67 g/kg in yogurt and non-dairy yogurt; 0.58 g/L in buttermilk, milk (flavored and unflavored), and imitation milk; 2.5 g/L in meal replacement drinks; 20 g/kg in meal replacement bars, and 0.58 g/L in fruit juices, nectars, and sports, energy, and isotonic drinks. The notice informs us of Glycom's view that these uses of LNnT are GRAS through scientific procedures.

Glycom provides information on the identity and composition of LNnT. Glycom describes LNnT as a white to off-white powder that consists of ≥ 80 % LNnT, which is a tetrasaccharide consisting of D-galactose, N-acetyl-D-glucosamine, D-galactose, and D-glucose. The chemical name for LNnT (CAS Registry 13007-32-4) is β-D-galactopyranosyl-(1→4)-2-acetamido-2-deoxy-β-D-glucopyranosyl-(1→3)-β-D-

[1] Glycom states that LNnT may be used individually or in combination with other human milk oligosaccharide (HMO) ingredients that were the subjects of previous GRAS notices. Glycom notes that infant formula manufacturers may use different HMO combinations and are ultimately responsible for meeting the requirements of section 412 of the Federal Food, Drug, & Cosmetic (FD&C) Act.

galactopyranosyl-(1→4)-D-glucopyranose. Glycom notes that LNnT may contain other carbohydrates that principally include lactose, lacto-N-triose II, *para*-lacto-N-neohexaose, and LNnT fructose isomer.

Glycom describes a two-stage manufacturing process for LNnT. The first stage consists of the production of LNnT by the cellular enzymes of a modified strain of *Escherichia coli* K-12 DH1, in a fermentation medium containing lactose derived from cow's milk. Glycom incorporates by reference information on the production organism and fermentation steps that are described in GRN 000659.[2] LNnT is secreted into the fermentation medium, from which the microbial biomass is separated by filtration. In the second stage, the filtered permeate is subjected to additional filtration, deionization, and decolorization to remove water, minerals, and other small molecules. Glycom notes that the resulting solution may be subjected to an optional chromatography step and that the final LNnT product is obtained by drying. Glycom states that LNnT is manufactured in compliance with current good manufacturing practices.

Glycom provides specifications for LNnT that include minimum levels of LNnT (≥ 80 % on a dry matter (DM) basis), total saccharides[3] (≥ 92 % DM), and limits on D-lactose (≤ 10 %), lacto-N-triose II (≤ 3 %), *para*-lacto-N-neohexaose (≤ 5 %), LNnT fructose isomer (≤ 1 %), moisture (≤ 9 %), lead (≤ 0.1 mg/kg), residual proteins (≤ 0.01 %), and microorganisms[4] including *Salmonella* (absent in a 25 g sample) and *Cronobacter sakazakii* (absent in a 10 g sample). Glycom provides the results of five, non-consecutive batch analyses of LNnT to demonstrate that their product meets specifications. Glycom predicts the LNnT to be stable for up to 5 years under ambient storage conditions and for up to 2 years under accelerated conditions based on the similarity to the production method and the stability of the ingredient described in GRN 000659.

Glycom discusses the estimated dietary exposure to LNnT. Glycom states that the intended uses of LNnT are substitutional for those described in GRN 000659 and that dietary exposure to LNnT will not change. In GRN 000659, Glycom provided estimates of dietary exposure to LNnT based on the intended use and food consumption data from the U.S. National Center for Health Statistics' 2009-2010 and 2011-2012 National Health and Nutrition Examination Surveys (NHANES). Glycom reported the mean and 90$^{th}$ percentile dietary exposures to LNnT to be 0.3 and 0.6 g/person (p)/d, respectively, for the total population and to be 0.5 and 0.9 g/ p/d, respectively, for toddlers aged 1 to 3 years. Glycom reported the mean and 90$^{th}$ percentile dietary exposures to LNnT for infants up to 6 months of age to be 0.51 and 0.73 g/p/d (83.2 and 133.9 mg/kg body weight (bw)/d), respectively, and for infants 7 to 12 months of age to be 0.42 and 0.66 g/p/d (48.5 and 79.5 mg/kg bw/d), respectively.

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[2] We evaluated GRN 000659 and responded in a letter dated November 29, 2016, stating that we had no questions at that time regarding Glycom's GRAS conclusion.

[3] Glycom states that the specification for saccharides include the sum of LNnT, D-lactose, lacto-N-triose II, para-lacto-N-neohexaose, and LNnT fructose isomer.

[4] Glycom's *C. sakazakii* specification is intended for use of LNnT in powdered infant formulas. We note that use of LNnT in liquid infant formulas require a manufacturing heat treatment step and the specification is not needed.

Glycom discusses publicly available information relevant to their safety conclusion. An updated literature search since the submission of GRN 000659 was performed on scientific literature through December 2019. Glycom states that they did not identify any new animal safety studies. Glycom incorporates by reference and discusses safety information from GRNs 000547[5] and 000659 to support their conclusion for the current GRN. Glycom states that LNnT is structurally identical to the LNnT in human breast milk and that it has been shown to be non-toxic in a 90-day rat oral toxicity study and non-mutagenic in genotoxicity assays. Glycom states that the absorption, distribution, metabolism and excretion characteristics of this non-crystalline form of LNnT will be the same as their crystalline form. Glycom notes that the specifications for the percentage of content for this ingredient have changed from their previous GRNs. They provide evidence demonstrating why these new specifications do not have an impact on the safety of LNnT. Because the non-crystalline form of LNnT is substitutional for the crystalline form, Glycom notes that dietary exposure to LNnT will not increase for the intended uses. Glycom concludes that there is no new information from investigational studies of LNnT *in vitro* or in animal models to suggest that use of LNnT as an ingredient in infant formula or conventional foods would be unsafe.

Based on the totality of evidence, Glycom concludes that LNnT is GRAS for its intended use.

**Standards of Identity**

In the notice, Glycom states its intention to use LNnT 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 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). The notice raises a potential issue under these labeling provisions. Glycom states that LNnT is intended to be used as an ingredient. If products containing LNnT 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.

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[5] GRN 000547 was submitted by Glycom for food uses of LNnT. We evaluated this notice and responded in a letter dated October 2, 2015, stating that we had no questions at that time regarding Glycom's GRAS conclusion.

## 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 eight foods or food groups (i.e., milk, eggs, fish, Crustacean shellfish, tree nuts, peanuts, wheat, and soybeans) or a food ingredient that contains protein derived from one of those foods. LNnT may require labeling under the FD&C Act because the final product may contain protein derived from cow's milk. Questions about petitions or notifications for exemptions from the food allergen labeling requirements should be directed to the Division of Food Ingredients in OFAS. Questions related to food labeling in general should be directed to the ONFL in the Center for Food Safety and Applied Nutrition.

## Intended Use in Infant Formulas

Under section 412 of the FD&C Act, a manufacturer of a new infant formula must make a submission to FDA providing required assurances about the formula at least 90 days before the formula is marketed. Our response to Glycom's GRAS notice does not alleviate the responsibility of any infant formula manufacturer that intends to market an infant formula containing LNnT to make the submission required by section 412. Infant formulas are the purview of 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 Glycom's notice concluding that LNnT 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 LNnT. Accordingly, our response should not be construed to be a statement that foods containing LNnT, if introduced or delivered for introduction into interstate commerce, would not violate section 301(ll).

## Conclusions

Based on the information that Glycom provided, as well as other information available to FDA, we have no questions at this time regarding Glycom's conclusion that LNnT is GRAS under its intended conditions of use. This letter is not an affirmation that LNnT 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 000895 is accessible to the public at www.fda.gov/grasnoticeinventory.

Sincerely,

Susan J. Carlson -S

Digitally signed by Susan J. Carlson -S
Date: 2020.12.03 13:42:50 -05'00'

Susan Carlson, Ph.D.
Director
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