INFORMATION ON THE SUBSTANCE TO BE EVALUATED BY JECFA

1. ***Proposal for inclusion submitted by:***

Danish Veterinary and Food Administration.

2. ***Name of substance; trade name(s); chemical name(s):***

Substance: Beta-amylase from *Bacillus flexus* expressed in *Bacillus licheniformis*

Chemical name: Beta-amylase; CAS 9000-91-3, EC 3.2.1.2

3. ***Names and addresses of basic producers:***

Novozymes A/S
Krogshøjvej 36
DK-2880 Bagsværd
Denmark

4. ***Has the manufacturer made a commitment to provide data?***

Novozymes A/S commits to provide data to support the proposal for inclusion of the beta-amylase in the list of substances to be evaluated by JECFA.

5. ***Identification of the manufacturer that will be providing data (Please indicate contact person):***

Novozymes A/S
Krogshøjvej 36
DK-2880 Bagsværd
Denmark

Attn.: Peter Hvass
phva@novozymes.com
+45 4446 3610

6. ***Justification for use:***

The beta-amylase enzyme preparation is used as a processing aid during food manufacture for hydrolysis of starch, e.g. in order to obtain more consistent and efficient production of maltose syrups during processing of starch-containing foods.

7. ***Food products and food categories within the GSFA in which the compound is used as a food additive or as an ingredient, including use level(s):***

The enzyme preparation is not added to final foodstuffs but used as a processing aid during food manufacturing. The beta-amylase is used in processing of starch-containing foods.

The beta-amylase is used at the minimum dosage necessary to achieve the desired enzymatic reaction. The range of dosage recommended for the beta-amylase is up to 10000 BAMU per kg of starch dry matter.

8. ***Is the compound currently used in food that is legally traded in more than one country? (please identify the countries); or, has the compound been approved for use in food in one or more country? (please identify the country(ies))***

The enzyme is marketed under the trade name of Secura which was approved in Denmark in 2015. Novozymes has also applied for approval of the enzyme in France, Mexico and Brazil. The approvals are expected in 2016.

9. ***List of data available (please check, if available)***

Toxicological data

(i) Metabolic and pharmacokinetic studies
(ii) Short-term toxicity, long-term toxicity/carcinogenicity, reproductive toxicity, and developmental toxicity studies in animals and genotoxicity studies
(iii) Epidemiological and/or clinical studies and special considerations
(iv) Other data

The following food toxicity program according to the EFSA Guideline[i] has been performed:

- Test for mutagenic activity (Ames Test)
- *In vitro* micronucleus
- 13 weeks oral toxicity study in rats

The main conclusions of the safety studies can be summarized as follows:
The beta-amylase preparation showed no mutagenic activity by testing in a bacterial reverse mutation assay (Ames Test) and did not induce micronuclei in cultured human peripheral blood lymphocytes *in vitro*.

The beta-amylase preparation did not result in treatment-related adverse effects when administered to rats for 13 weeks, and the overall No Observed Adverse Effect Level (NOAEL) is considered to be the highest administered dose, corresponding to 1199 mg TOS/kg body weight (bw)/day.

The safety studies described above were all performed on liquid beta-amylase enzyme concentrate produced in accordance with ordinary production procedure, omitting stabilization and standardization.

Bacillus licheniformis is generally considered to be a safe production organism with a long history of safe use for food ingredients.

Technological data

(i) Specifications for the identity and purity of the listed compounds (specifications applied during development and toxicological studies; proposed specifications for commerce)

(ii) Technological and nutritional considerations relating to the manufacture and use of the listed compound

The beta-amylase enzyme preparation complies with the purity criteria recommended for enzyme preparations by Food Chemicals Codex (VIII online edition, 2012). In addition to this, the enzyme preparation also conforms to the General Specifications and Considerations for Enzyme Preparations Used in Food Processing (2006) as proposed by the Joint FAO/WHO Expert Committee on Food Additives in Combined Compendium of Food Additive Specifications.

Intake assessment data

(i) Levels of the listed substance used in food or expected to be used in food based on technological function and the range of foods in which they are used

(ii) Estimation of dietary intakes based on food consumption data for foods in which the substance may be used.

The exposure assessment is performed according to the Budget Method (ILSI, 1997). The Budget Method assumptions represent a "maximum worst case" situation of human consumption, in which the enzyme would be used at its maximum recommended dosages in all processed food and all processed beverages.

Overall, the human exposure to the beta-amylase will be negligible because the enzyme preparation is used as a processing aid and in low dosages. It is also supposed that the totality of the food enzyme will end up in the final food. This assumption is exaggerated since the enzyme protein and the other substances resulting from the fermentation are diluted or removed in certain processing steps.

Therefore the safety margin calculation derived from this method is highly conservative.

Assumptions in the Budget Method

Solid food	The maximum energy intake over the course of a lifetime is 50 kcal/kg bw/day. 50 kcal corresponds to 25 g foods. Therefore, adults ingest 25 g foods per kg bw per day. Assuming that 50% of the food is processed food, the daily consumption will be 12.5 g processed foods per kg bw. It is further assumed that, in average, all processed food contains 25% starch (or starch-derived) dry matter = 3.12 g starch derived dry matter per kg bw per day.

Liquids	The maximum intake of liquids (other than milk) is 100 ml/kg bw/day. Assuming that 25% of the non-milk beverages is processed, the daily consumption will be 25 ml processed beverages per kg bw. It is further assumed that all processed beverages contain 10% starch hydrolysates = 2.50 g starch derived dry matter per kg bw per day. It is assumed that the densities of the beverages are ~ 1.

Theoretical Maximum Daily Intake (TMDI) calculation

Solid Food:
The highest dosage is 10000 BAMU per kilogram starch based raw material. 10000 BAMU correspond to 99.1 mg TOS. Based on this, 3.12 gram starch-derived dry matter in solid food will maximally contain 0.31 mg TOS.

Liquid Food:
The highest dosage is 10000 BAMU per kilogram starch based raw material. 10000 BAMU correspond to 99.1 mg TOS. Based on this, 2.50 gram starch-derived dry matter in liquids will maximally contain 0.25 mg TOS.

The theoretical maximum daily intake (TMDI) of the enzyme by consumers is therefore:
0.31 + 0.25 = 0.56 mg TOS/kg body weight/day.

Other information as necessary

10. ***Date on which data could be submitted to JECFA:***

August 2016

[i] Guidance of EFSA prepared by the Scientific Panel of Food Contact Material, Enzymes, Flavourings and Processing Aids on the Submission of a Dossier on Food Enzymes. The EFSA Journal (2009) 1305, 1-26