

**APPLICATION TO AMEND THE
*AUSTRALIA AND NEW ZEALAND
FOOD STANDARDS CODE* TO ALLOW
FOR THE USE OF HONEY TRUFFLE SWEET
PROTEIN AS A FOOD ADDITIVE
(STANDARD 1.3.1)**

EXECUTIVE SUMMARY

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MycoTechnology, Inc. (MycoTechnology) is seeking to amend Schedules 3, 15, and 29 of the *Australia New Zealand Food Standards Code* to include honey truffle sweet protein (HTSP) for use as a food additive. The HTSP additive contains $\geq 60\%$ total protein and $\geq 40\%$ honey truffle active component (HT-AC; a high-intensity sweet protein that is the active constituent of HTSP). Based on HT-AC's estimated sweetness potency of 500 to 2,500 times sweeter than sucrose, and applying the minimum HT-AC content of 40%, HTSP is conservatively estimated to be approximately 200 to 1,000 times sweeter than sucrose, and is intended to be used as a sweetener in a variety of food and beverage products."

HTSP is produced by fermentation using a strain of *Komagataella phaffii* that has been modified to express the HT-AC protein, which is native to the honey truffle (*Mattirolomyces terfezioides*). Specifications are in place for all raw materials, processing aids, purification equipment, and the final HTSP additive to ensure a consistent final product. The final product specifications also ensure product safety by placing limits on microbial contamination, water activity, heavy metals, and potential microbial contaminants. HTSP is manufactured in accordance with the principles of Hazard Analysis and Critical Control Points (HACCP) and current Good Manufacturing Practice (cGMP).

HTSP is intended for use as a replacement for other sweetening additives currently included in Schedule 15 (Substances that may be used as food additives). The dietary intakes of HTSP were estimated based on its sweetness potency (*i.e.*, 200 times sweeter than sucrose at 10 °Brix with 40% HT-AC purity). Intakes of HTSP in non-diabetic and diabetic adults were estimated to be up to 3.38 and 4.49 mg/kg body weight/day, respectively, while intakes in non-diabetic and diabetic children were calculated to be up to 4.95 and 4.54 mg/kg body weight/day, respectively.

The safety of MycoTechnology's HTSP is supported by the long history of consumption of the honey truffle (*M. terfezioides*), the well-established safety of the production organism (*K. phaffii*), and the well characterised genetic modification used to obtain the production strain. The results of simulated gastric and simulated upper gastrointestinal tract studies demonstrate that HT-AC is virtually completely digested. The results of a bioinformatics assessment of HT-AC demonstrate that HT-AC is unlikely to have allergenic or toxigenic potential, and the results of *in vitro* genotoxicity studies demonstrate that HTSP is not genotoxic. Based on a no-observed-adverse-effect level of 5,400 mg HTSP/kg body weight/day (*i.e.*, 1,500 mg HT-AC/kg body weight/day; the highest dose tested) reported in a 13-week oral study in rats, an acceptable daily intake (ADI) of at least 54 mg HTSP/kg body weight/day was calculated. The highest estimated dietary exposure to HTSP is 4.95 mg/kg body weight/day, which is well below the calculated ADI and would therefore not pose any safety concerns under the intended conditions of use of HTSP. Overall, the available evidence and long history of human consumption of the honey truffle support the safety and suitability of the use of HTSP as a food additive.