ICS 65.120
CCS B46

NATIONAL STANDARD OF THE PEOPLE'S REPUBLIC OF CHINA

GB 7300.501-2021

Replacing GB/T 22547-2008

Feed additives - Part 5: Live microorganisms - Saccharomyces cerevisiae

饲料添加剂 第5部分：微生物 酿酒酵母

Issued on: October 11, 2021 **Implemented on: November 1, 2022**

Issued by: State Administration for Market Regulation;
Standardization Administration of China

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1 Scope

GB 7300.501-2021 is the Chinese national standard on feed additives - part 5: live microorganisms - saccharomyces cerevisiae, in the field of agriculture. It carries no /T suffix, which in the Chinese system means compliance is mandatory: a product or a practice within its scope has to meet it to be lawfully made, sold or carried out in China. It was issued on 11 October 2021 by the State Administration for Market Regulation; Standardization Administration of China, and has been in force since 1 November 2022. Classification: ICS 65.120, CCS B46. This page is published from the official record of the standard held by the Chinese standards administration: the identification, the dates, the classification and the issuing body are taken from there. The clause text, the tables and the numeric limits are in the document itself, which is delivered complete in English translation.

This document specifies technical requirements, sampling, test methods, inspection rules, labeling, packaging, transportation and storage for Saccharomyces cerevisiae, a feed additive. This document is applicable to the feed additive Saccharomyces cerevisiae prepared by liquid fermentation, dehydration and drying with Saccharomyces cerevisiae as strain.

2 Normative references

The following referenced documents are indispensable for the application of this document. For dated references, only the edition cited applies. For undated references, the latest edition of the referenced document (including any amendments) applies.

GB 4789.15-2016, National food safety standard - Food microbiological examination - Enumeration of moulds and yeasts

GB/T 6435, Determination of moisture in feedstuffs

GB/T 6682, Water for analytical laboratory use - Specification and test methods

GB/T 8170, Rules of rounding off for numerical values and expression and judgement of limiting values GB 10648, Feed label

GB/T 13079, Determination of total arsenic in feeds

GB/T 13080, Determination of lead in feeds - Atomic absorption spectrometry

GB/T 13081, Determination of mercury in feeds

GB/T 13082, Determination of cadmium in feeds

GB/T 13091, Determination of Salmonella in feeds possible foreign contamination.

5.2 Sampling method

5.2.1 Samples shall be collected in the same batch. The sampling amount of each sample shall meet the requirements of microbiological indicator inspection. Generally, it is not less than 500g.

5.2.2 For products with independent packaging less than or equal to 500g, take the complete package.

5.2.3 For products larger than 500g individually packaged, a sterile sampler shall be used to take appropriate samples from different parts of the same package. Place them in the same sterile sampling container as one sample.

5.3 Storage and transportation of collected samples

5.3.1 Samples shall be sent to the laboratory for inspection as soon as possible.

5.3.2 Samples shall be kept intact during transportation.

5.3.3 Samples shall be stored close to their original storage temperature. Or take necessary measures to prevent changes in the number of microorganisms in the sample.

6 Test method

Unless otherwise specified, only the reagents confirmed as analytically pure are used in the analysis, and the water meets the requirements of grade 3 water in GB/T 6682.

6.1 Sensory inspection Take an appropriate amount of specimen. Place on a clean white piece of paper. Observe its shape, color, presence or absence of impurities under natural light. Smell its odor.

6.2 Strain identification Carry out in accordance with Annex A. Use panel method as the arbitration method.

6.3 Yeast viable cell count Carry out in accordance with Annex B.

6.4 Moisture Carry out in accordance with GB/T 6435.

6.5 Total arsenic Carry out in accordance with GB/T 13079.

6.6 Lead Carry out in accordance with graphite furnace atomic absorption spectrometry in GB/T 13080.

6.7 Mercury Carry out in accordance with GB/T 13081.

6.8 Cadmium Carry out in accordance with GB/T 13082.

6.9 Salmonella Carry out in accordance with GB/T 13091.

7 Inspection rules

7.1 Batching A batch of products refer to the products of the same specification produced in the same material and in the same production process through continuous production or in the same shift. But each batch of products shall not exceed 50t.

7.2 Exit-factory inspection Appearance and properties, yeast live cell count, and moisture are the inspection items of the exit-factory inspection.

7.3 Type inspection Type inspection items are all items specified in Chapter 4 of this document. Under normal production conditions, type inspection shall be carried out at least once a year. Type inspection shall also be carried out in one of the following situations:

a) When the product is stereotyped and put into production;

b) When there is a major change in the production process, formula or source of main raw materials, which may affect the quality of the product;

c) When production is stopped for more than 3 months and production is resumed;

d) When there is a significant difference between the exit-factory inspection results and the last type inspection results;

Annex A

(normative) Identification method for Saccharomyces cerevisiae A.1 Morphological identification A.1.1 Culture medium A.1.1.1 Wort liquid medium: The wort is diluted with water to 10°Bx~15°Bx (Bahrain Brix Meter). Sterilize by autoclaving at 115°C for 15min. A.1.1.2 Wort agar medium: The wort is diluted with water to 10°Bx~15°Bx (Bahrain Brix Meter). Add 2% agar powder. Sterilize by autoclaving at 115°C for 15min. A.1.1.3 Sporulation medium: 0.1% of glucose, 0.18% of potassium chloride, 0.25% of yeast juice,

0.82% of sodium acetate, 2% of agar. Use distilled water to prepare. Put in a tube. Sterilize by autoclaving at 115°C for 15min. Shelve bevel. A.1.1.4 Potato dextrose agar: 200g of peeled potato, 20g of glucose, 20g of agar, 1L of tap water. Clean the potatoes. Remove the peel. Cut into small pieces. Immediately put into water to avoid oxidation. Boil 30min. Filter by gauze. Add water to the filtrate to 1L. Add glucose and agar. Subpack into conical flasks or test tubes after they are dissolved. Sterilize by autoclaving at 115°C for 20min. A.1.2 Growth in wort liquid medium After 3 days of static culture at 28°C, bacteria settle tightly at the bottom in liquid medium. The culture medium is clear. No biofilm is formed. Take a small number of bacteria. Observe under a 400x microscope. Cells are oval or round, single or double, occasionally in clusters, cell budding. The ratio of cell length to width is 1~2. Cell size is divided into three types: large, medium and small. The size of large cells is (4.5μm~10μm) x (7.0μm~21μm). The size of medium cells is (3.5μm~8μm) x (5.0μm~17.5μm). The size of small cells is (2.5μm~7μm) x (4.5μm~11μm). A.1.3 Growth on wort agar medium After culturing at 28°C for 3 days, the colonies are large and moist, slightly raised or flat, milky white. The surface is smooth and wrinkle-free. The edges are neat. A.1.4 Growth on potato dextrose agar After culturing at 28°C for 3 days, grow on potato agar medium. There is no pseudohyphae or relatively developed but atypical pseudohyphae.

Annex B

(normative) Determination method for yeast viable cell count B.1 Method One -- Plate Method (Arbitration Method) Test according to Method One of GB 4789.15-2016. When testing, the temperature of "adding 225mL of sterile diluent" in