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CCS B46



**NATIONAL STANDARD OF THE
PEOPLE'S REPUBLIC OF CHINA**

GB/T 18634-2009

Replacing GB/T 18634-2002

**Determination of feed phytase activity - Spectrophotometric
method**

饲用植酸酶活性的测定 分光光度法

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Foreword

This standard replaces GB/T 18634-2002 "Feeding phytase activity was measured by spectrophotometry." This standard compared with GB/T 18634-2002 main changes are as follows:

- Narrowed the scope of application of the method;
- Change the minimum limit of quantification;
- Changed the method of preparation of buffer solution;
- Refine the determination of process steps;
- Removed relatively law;
- Expanded with the addition of phytase feed samples allowed two runs difference, relative deviation $\leq 15\%$. Appendix A of this standard is an informative annex. This standard by the National Standardization Technical Committee Feed Industry and focal points. This standard is drafted by: Agricultural Quality Standards and Testing Technology Research Institute, Chinese Academy of Agricultural Sciences [National Feed Quality Supervision and Inspection Heart (Beijing)], Guangdong overflow Dolly Biotechnology Co., Ltd., Wuhan Xinhua Yang Biotechnology Limited. The main drafters of this standard. Ma Dongxia, Zhanzhi Chun, Shi Baojun, Suxiao Ou, Zhan Liansheng, Liangxue Xia, Zhang Su. This standard replaces the standards previously issued as follows:
- GB/T 18634-2002. Determination of feed phytase activity Spectrophotometry

1 Scope

GB/T 18634-2009 is the Chinese national standard on determination of feed phytase activity - spectrophotometric method, in the field of agriculture. The /T suffix marks it as a recommended standard: it is not compulsory by itself, but it becomes binding as soon as a contract, a tender or a customer specification calls it up - which in practice is how most

foreign buyers meet it. It was issued on 26 May 2009 by the General Administration of Quality Supervision, Inspection and Quarantine of the People's Republic of China; Standardization Administration of China, and has been in force since 1 October 2009. 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 standard specifies the spectrophotometric determination of feed with phytase activity. This standard applies to products phytase as a feed additive for use, but also to added phytase feed. Methods lowest The limit of quantification of 130U/kg.

2 Normative references

The following documents contain provisions which, through reference in this standard and become the standard terms. For dated references, subsequent Amendments (not including errata content) or revisions do not apply to this standard, however, encourage the parties to the agreement are based on research Whether the latest versions of these documents. For undated reference documents, the latest versions apply to this standard.

GB/T 14699.1 feed samples

3 Terms and Definitions

The following terms and definitions apply to this standard.

3.1 At a temperature of 37 °C, under pH

5.50 conditions, L solution per minute from sodium phytate concentration of 5.0mmol/release 1μmol inorganic phosphorus, Is a unit of phytase activity, expressed in U. Principle

4 Phytase at a certain temperature and pH conditions, the hydrolysis of the substrate phytate, and inositol phosphate derivatives being generated. In acidic solution, Can form a yellow complex with ammonium molybdate vanadium, can be measured colorimetrically at 415nm wavelength.

5 Reagents and materials

Unless otherwise indicated in the analysis using only recognized as analytical grade reagents and distilled or deionized water or equivalent purity. Wash Test vessels do not use phosphorus-containing cleaning agents.

5.1 potassium dihydrogen phosphate (KH₂PO₄). reference material.

5.2 acetate buffer (1), Ba (CH₃COONa) = 0.25mol/L. Weigh 20.52g of anhydrous sodium

acetate in 1000mL beaker, add into 900mL water and stir to dissolve, with glacial acetic acid pH adjusted to 5.50 ± 0.01 , and transferred to a 1000mL volumetric flask and filled with distilled water to volume. Stored at room temperature for 2 months effective.

5.3 acetate buffer (2), Ba (CH_3COONa) = 0.25mol/L. Weigh 20.52g of anhydrous sodium acetate, 0.5g Triton X-100 (TritonX-100), 0.5g of bovine serum albumin (BSA) in 1000mL beaker, 900mL water and stir to dissolve, with glacial acetic acid to pH 5.50 ± 0.01 , and transferred to a 1000mL volumetric flask with distilled water to the mark. Store at room temperature for 2 months effective.

5.4 substrate solution, Ba ($\text{C}_6\text{H}_6\text{O}_{24}\text{P}_6\text{Na}_{12}$) = 7.5mmol/L. Weigh 0.69g sodium phytate ($\text{C}_6\text{H}_6\text{O}_{24}\text{P}_6\text{Na}_{12}$, relative molecular mass 923.8, purity 95%), accurate to 0.1mg, placed in 100mL beaker, dissolved in about 80mL acetate buffer (5.2), with ice Acetic acid to adjust the pH to 5.50 ± 0.01 , transferred to a 100mL volumetric flask, and treated with acetate buffer (5.2) to the mark, using now