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(54) Title: PHYTASES WITH IMPROVED THERMAL STABILITY

(57) Abstract: The invention provides variant phytase enzymes having increased thermal stability relative to their counterpart parent enzymes. The modifications to the enzymes include both single substitutions and various combinations of substitutions that provide improved stability and activity. The invention further provides nucleic acids encoding the variant phytase enzymes, host cells and vectors containing and expressing them, as well as feed compositions useful for providing improved nutrition, particularly with respect to the bioavailability of dietary phosphate, calcium, iron and zinc, among others.



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**INTERNATIONAL SEARCH REPORT**

International application No.

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**A. CLASSIFICATION OF SUBJECT MATTER**  
 IPC: C12N 9/06( 2006.01);C07K 1/00( 2006.01);C07H 21/04( 2006.01);A23J 1/00( 2006.01);A61K 38/46( 2006.01)  
 USPC: 424/94.6;435/195,69.1,196;530/350;435/196,19,174,176  
 According to International Patent Classification (IPC) or to both national classification and IPC

**B. FIELDS SEARCHED**  
 Minimum documentation searched (classification system followed by classification symbols)  
 U.S. : 424/94.6;435/195,69.1,196;530/350;435/196,19,174,176

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)  
 MEDLINE, BIOSIS, BIOTECHDS, CAPLUS, EMBASE, US-PGPUB, USPAT, USOCR, DERWENT, EPO, JPO

**C. DOCUMENTS CONSIDERED TO BE RELEVANT**

| Category * | Citation of document, with indication, where appropriate, of the relevant passages  | Relevant to claim No. |
|------------|---|-----------------------|
| Y          | US 7,078,035 B2 (SHORT et al) 18 July 2006 (18.07.2006), entire document, particularly column 5, lines 50-61, column 9, lines 1-3, column 29 lines 55-65.   | 1-59                  |
| Y          | RODRIGUEZ E. Cloning, sequencing, and expression of an Escherichia coli Acid Phosphatase/Phytase Gene (aapA2). Biochemical and Biophysical Research Communication 1999, Vol. 257, pages 117-123, especially page 120. | 1-59                  |
| Y          | LIM D. Crystal structures of Escherichia coli phytase and its complex with phytate. Nature Structural Biotechnology, 2000, Vol. 7, No. 2, pages 108-113, specially pages 108-109.                                     | 1-59                  |
| A          | US Publication No. 2007/01964449 (LEI et al) 23 August 2007 (23.07.2007), pages 8-9.  | 1-59                  |

Further documents are listed in the continuation of Box C.  See patent family annex.

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| Name and mailing address of the ISA/US<br>Mail Stop PCT, Attn: ISA/US<br>Commissioner for Patents<br>P.O. Box 1450<br>Alexandria, Virginia 22313-1450<br>Facsimile No. (571) 273-1085 | Authorized officer: 2008<br>Maria Leavitt<br>Telephone No. 571-272-1085 |