

# α-L-ARABINOFURANOSIDASE from Bifidobacterium sp. (Lot 91201e)

### Recombinant

**E-AFAM2** (3.2.1.55) alpha-L-arabinofuranoside arabinofuranohydrolase CAZy Family: GH43 CAS: 9067-74-7

# PROPERTIES

# I. ELECTROPHORETIC PURITY:

- Single band on SDS-gel electrophoresis (MW ~ 59,404)

- Broad diffuse band on isoelectric focusing (pl  $\sim$  4.6)

#### 2. SPECIFIC ACTIVITY AND LEVEL OF OTHER ACTIVITIES: 102 U/mg protein (on wheat arabinoxylan) at pH 6.0 and 40°C.

**One Unit** of  $\alpha$ -L-arabinofuranosidase activity is defined as the amount of enzyme required to release one  $\mu$ mole of arabinose per minute from wheat arabinoxylan (10 mg/mL) in sodium phosphate buffer (100 mM) pH 6.0 at 40°C.

# 3. SPECIFICITY:

Highly specific hydrolysis of  $\alpha$ -1,3-linked L-arabinofuranose residues from doubly substituted D-xylosyl or L-arabinosyl residues of arabinoxylans and branched arabinans, respectively

# 4. RELATIVE RATES OF HYDROLYSIS OF SUBSTRATES:

Substrate	%
Wheat Arabinoxylan	100
Xyalanse-treated Wheat Arabinoxylan	90.4
Sugar Beet Arabinan	4.0
p-Nitrophenyl-α-arabinofuranoside	0.095

Action on pNP-substrates and polysaccharides or oligosaccharides was determined at a final substrate concentration of 2.5 mM and 3.5 mg/mL, respectively, in sodium phosphate buffer (100 mM), pH 6.0 at 40°C.

# 5. PHYSICOCHEMICAL PROPERTIES:

Recommended conditions of use are at pH 6.0 and up to  $50^{\circ}C$ 

# 6. STABILITY:

The enzyme is supplied as an ammonium sulphate suspension in 0.02% (w/v) sodium azide and should be stored at 4°C. For assay, this enzyme should be diluted in sodium phosphate buffer (100 mM), pH 6.0. Swirl to mix the enzyme immediately prior to use.

# 7. **REFERENCES**:

Van Laere, K.M.J., Beldman, G. & Voragen, A.G.J. (1997) A new arabinofuranohydrolase from *Bifidobacterium adolescentis* able to remove arabinofuranosyl residues from double substituted xylose units in arabinoxylan. *Appl Microbiol. Biotech.* **47**: 231-235.

Van den Broek, L.A.M., Lloyd, R.M., Beldman, G., Verdoes, J.C., McCleary, B.V. & Voragen, A.G.J. (2005) Cloning and characterization of arabinoxylan arabinohydrolase D-3 (AXH-D3) from *Bifidobacterium adolescentis* DSM20083. *Appl Microbiol. Biotech.* **67**: 641-647.

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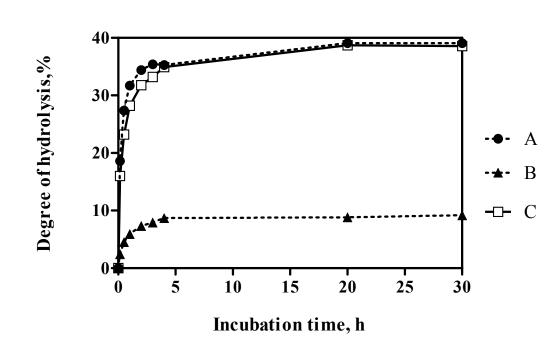


Figure 1. Hydrolysis of xylanase degraded wheat arabinoxylan by A. niger and B. adolescentis  $\alpha$ -L-arabinofuranosidase.

Xylanase degraded wheat arabinoxylan (5 mL, 2 mg/mL) was incubated with A. A. niger  $\alpha$ -L-arabinofuranosidase (500 U on *p*-NP- $\alpha$ -L arabinofuranoside) in 100 mM sodium acetate buffer (pH 4.5), or B. B. adolescentis  $\alpha$ -L-arabinofuranosidase (7 U on wheat arabinoxylan) in 100 mM sodium maleate buffer (pH 6.5), or C. A. niger  $\alpha$ -L-arabinofuranosidase (500 U on *p*-NP- $\alpha$ -L arabinofuranoside) plus B. adolescentis  $\alpha$ -L-arabino-furanosidase (7 U on wheat arabinoxylan) in 100 mM sodium maleate buffer (pH 6.5), or C. A. niger  $\alpha$ -L-arabinofuranosidase (500 U on *p*-NP- $\alpha$ -L arabinofuranoside) plus B. adolescentis  $\alpha$ -L-arabino-furanosidase (7 U on wheat arabinoxylan) in 100 mM sodium acetate buffer (pH 5.0). Aliquots (50 mL) were removed at various time intervals, inactivated by incubation at 100°C for 2 min, and analysed for released L-arabinose with  $\beta$ -galactose dehydrogenase. Degree of hydrolysis was calculated as L-arabinose released as a percentage of total carbohydrate determined with the phenol-sulphuric acid procedure.

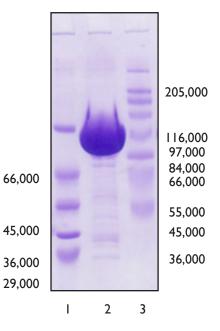


Figure 2. SDS-PAGE analysis of  $\alpha$ -L-arabinofuranosidase (Bifidobacter sp.)

Electrophoresis was performed using a 10% acrylamide gel. Lane I, low molecular weight markers (Sigma cat. no. M-3918); lane 2, 5  $\mu$ g *B. adolescentis*  $\alpha$ -L-arabinofuranosidase; lane 3, high molecular weight markers (Sigma cat. no. M-3788).