

## Product Information

### **$\alpha$ -Galactosidase I, Alkaline from *Cucumis melo*, His-tagged recombinant, expressed in *Escherichia coli***

Catalog Number **G7673**  
Storage Temperature  $-20\text{ }^{\circ}\text{C}$

EC 3.2.1.22

Synonyms: AGALI,  $\alpha$ -D-galactoside galactohydrolase

#### **Product Description**

Raffinose and stachyose are ubiquitous galactosyl-sucrose oligosaccharides in the plant kingdom, which play major roles, second only to sucrose, in photo-assimilate translocation and seed carbohydrate storage. These sugars are initially metabolized by  $\alpha$ -galactosidases. Two  $\alpha$ -galactosidases are found in melon fruit (*Cucumis melo*) with alkaline pH optima (AGALI and AGALII). The plant specific  $\alpha$ -gal family of glycosyl hydrolases, with optimal activity at neutral to alkaline pH values, probably functions in key processes of galactosyl-oligosaccharide metabolism, such as seed germination and translocation of RFO photosynthate. These alkaline  $\alpha$ -galactosidases seem to belong to a novel family of proteins named Seed Inhibitory Proteins or SIP.<sup>1</sup>

The reaction sequence of sugar hydrolysis by  $\alpha$ -galactosidase is as follows:



Inhibition of this reaction by increasing concentrations of raffinose or galactose is defined as product inhibition. For most industrial applications, it is important the enzyme used is not inhibited by either. AGALI is not inhibited by raffinose up to a concentration of 120 mM; whereas, AGALII and the acid form of  $\alpha$ -galactosidase are 50% inhibited at this concentration.<sup>2</sup> AGALI has a  $K_M$  similar to AGALII for stachyose ( $\sim 4.0$  mM) but a much higher affinity for the sugar raffinose (AGALI,  $K_M = 1.5$  mM and AGALII,  $K_M = 26.3$  mM). AGALI is only weakly inhibited by the reaction product galactose (AGALI,  $K_i = 13$  mM; AGALII,  $K_i = 1.34$  mM; and acid  $\alpha$ -galactosidase,  $K_i = 0.06$  mM).<sup>2</sup>

AGALI has an apparent molecular mass of 84 kDa (SDS-PAGE).<sup>2</sup>

The product is supplied as a lyophilized powder containing Tris-HCl buffer salts, DTT, EDTA, and NaCl.

Purity:  $\geq 85\%$  (SDS-PAGE)

Specific activity:  $\geq 20$  units/mg-protein

Unit definition: One unit will hydrolyze 1.0  $\mu$ mole of *p*-nitrophenyl  $\alpha$ -D-galactoside to *p*-nitrophenol and D-galactose per minute at pH 7.8 at 30  $^{\circ}\text{C}$ .

#### **Precautions and Disclaimer**

This product is for R&D use only, not for drug, household, or other uses. Please consult the Material Safety Data Sheet for information regarding hazards and safe handling practices.

#### **Preparation Instructions**

Reconstitution with 0.9 ml of 0.05% (v/v) TWEEN® 20 (Catalog Number P7949 or equivalent) aqueous solution will yield a solution containing  $\sim 0.5$  mg-protein/ml, 50 mM Tris-HCl, pH 7.5, 1 mM DTT, 0.1 mM EDTA, 200 mM NaCl, and 0.05% TWEEN 20.

After initial thawing, the enzyme may be kept at 2–8  $^{\circ}\text{C}$  for up to 2 weeks or frozen in aliquots at  $-20\text{ }^{\circ}\text{C}$ .

#### **Storage/Stability**

Store the product at  $-20\text{ }^{\circ}\text{C}$ . The product is stable for at least 2 years as supplied. Avoid repeated freezing and thawing.

## References

1. Carmi, N. et al., Cloning and functional expression of alkaline  $\alpha$ -galactosidase from melon fruit: similarity to plant SIP proteins uncovers a novel family of plant glycosyl hydrolases. *Plant J.*, **33**, 97-106 (2003).
2. Gao, Z., and Schaffer, A.A., A novel alkaline  $\alpha$ -galactosidase from melon fruit with a substrate preference for raffinose. *Plant Physiol.*, **119**, 979-988 (1999).

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