

Accepted Manuscript

Title: The use of starch azure for measurement of alpha-amylase activity

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PII: S0144-8617(17)31444-3
DOI: <https://doi.org/10.1016/j.carbpol.2017.12.037>
Reference: CARP 13099



To appear in:

Received date: 16-11-2017
Revised date: 21-11-2017
Accepted date: 14-12-2017

Please cite this article as: Lehoczki G, Kandra L, Gyémánt G, The use of starch azure for measurement of alpha-amylase activity, *Carbohydrate Polymers* (2010), <https://doi.org/10.1016/j.carbpol.2017.12.037>

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The use of starch azure for measurement of alpha-amylase activity

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Highlights

- Enzymatic hydrolysis rate of starch azure has increased after boiling pre-treatment.
- ITC experiment was carried out to compare the natural and blue starch hydrolysis.
- Substrate inhibition of mammalian amylases occurred on starch azure.
- Modification of amylase assay protocol was suggested.

Abstract

Despite being widely used, there is no standard protocol for α -amylase activity measurement with starch azure substrate. Boiling pre-treatment of starch azure suspension increased the reaction rate of hydrolysis catalysed by human salivary α -amylase (HSA) or porcine pancreatic α -amylase (PPA) and the sensitivity of spectrophotometric activity measurement has been improved. Kinetic constants, K_M , and v_{max} , obtained from parallel isothermal titration calorimetric (ITC) measurements on natural and starch azure revealed, that the blue starch derivative does not differ significantly from its natural counterpart from kinetic point of view. Interestingly, substrate inhibition was observed in starch azure hydrolysis characterised by

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