



Enzymatic analyses demonstrate thermal adaptation of α -glucosidase activity in starch amended gully waste

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HIGHLIGHTS

- ▶ Starch degradation was confirmed as temperature independent.
- ▶ α -glucosidase responded positively to starch addition.
- ▶ Evidence for α -glucosidase adaptation to *in situ* temperature.

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ABSTRACT

In this study we investigated the effect of starch amendment on α -glucosidase activity in an organic waste environment, treated under both mesophilic and thermophilic conditions. The relative effects of temperature on α -glucosidase activity with regard to *in situ* and assay conditions were investigated under a reciprocal design. The decline in dry matter under the different thermal regimes was consistent with the temperature independent degradation of starch. The results of extra-cellular enzyme analysis showed a significant relationship between starch addition and α -glucosidase activity, with evidence of thermal adaptation to the *in situ* temperature. A weaker, but significant, effect of starch addition on β -glucosidase activity was observed, with no evidence for thermal adaptation. Thus, our data is consistent with a substrate adaptive response to temperature, albeit under potentially a high selective pressure.

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1. Introduction

Sustainable organic waste management practises rely ultimately on understanding the mechanisms that control organic matter decay. As a methodology used to elucidate the fate of organic matter in relevant environments, extracellular enzyme analysis (EEA) is a potentially useful qualitative and quantitative approach to investigate these dynamics. Elevated temperature profiles are a key feature of industrial organic waste processing and there has been a long standing fascination with the interaction of this parameter with regard to the degradation process (Finstein and Morris, 1977). Various EEA approaches have been used to investigate aerobic organic waste processing (aka composting) and numerous authors have used this approach to monitor the progression of organic matter stability and maturity (e.g., Cayuela et al., 2008; Komilis et al., 2011; Mondini et al., 2004). However, detailed controlled study with regard to the effect of temperature

on these dynamics is limited. For example, to our knowledge, reciprocal approaches to investigate temperature effects, with regard to adaptive responses of EEA, have only been reported twice (Adams et al., 2008; Adams and Umaphy, 2011).

A key focus of our research has been to understand how enzymes potentially adapt to different temperature regimes. In our initial attempt to investigate the mechanistic behaviour of enzymes related to starch degradation, we found not only little evidence to support α -glucosidase as a predictor for starch degradation, but that also empirical validation of extracellular enzyme analysis specificity towards their putative substrate was limited (Adams and Umaphy, 2011). Due to their applied relevance, it is important to understand the factors that regulate enzymatic processes naturally. There is now considerable interest in identifying glycoside hydrolases of potential commercial interest (e.g., Allgaier et al., 2010; Gladden et al., 2011), particularly in relation to the global interest in bioethanol (Sánchez and Cardona, 2008). A potential application of these, of local interest to this study, is a proposed starch to bio-ethanol production plant that will potentially consume 60% of the local annual wheat yield (Martindale, 2009). The primary aim of this study was to assess the effect of substrate addition (starch) on a putatively linked enzyme substrate

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