Roles of multiple surface sites, long substrate binding clefts, and carbohydrate binding modules in the action of amylolytic enzymes on polysaccharide substrates - DTU Orbit (19/01/2019)

Roles of multiple surface sites, long substrate binding clefts, and carbohydrate binding modules in the action of amylolytic enzymes on polysaccharide substrates

Germinating barley seeds contain multiple forms of alpha-amylase, which are subject to both differential gene expression and differential degradation as part of the repertoire of starch-degrading enzymes. The alpha-amylases are endo-acting and possess a long substrate binding cleft with a characteristic subsite binding energy profile around the catalytic site. Furthermore, several amylolytic enzymes that facilitate attack on the natural substrate, i.e. the endosperm starch granules, have secondary sugar binding sites either situated on the surface of the protein domain or structural unit that contains the catalytic site or belonging to a separate starch binding domain. The role of surface sites in the function of barley alpha-amylase 1 has been investigated by using mutational analysis in conjunction with carbohydrate binding analyses and crystallography. The ability to bind starch depends on the surface sites and varies for starch granules of different genotypes and botanical origin. The surface sites, moreover, are candidates for being involved in degradation of polysaccharides by a multiple attack mechanism. Future studies of the molecular nature of the multivalent enzyme-substrate interactions will address surface sites in both barley alpha-amylase 1 and in the related isozyme 2.

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