A new method based on structural signatures to propose mutations for enzymes β-glucosidase used in biofuel production

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Abstract

β-glucosidases (E.C. 3.2.1.21) are key enzymes in the second-generation biofuel production process. They act synergically with endoglucanases and exoglucanases to convert cellulose of biomass in fermentable glucose used in biofuel production. However, it has been reported in the literature that the majority of known β-glucosidases is inhibited by high concentrations of glucose. Hence, it has increased the search for mutations that improve the activity and glucose tolerance. In this study, we present a method to propose mutations for enzymes β-glucosidase that may improve the activity and tolerance to glucose inhibition. Our method is based on structural signatures: numerical representations of proteins extracted from the number of pairwise residues. We hypothesized that proteins with similar structural signatures of catalytic pockets present similar characteristics. Hence, mutations that approximate non-tolerant β-glucosidase structural signatures of other enzymes classified in the literature as glucose-tolerant may improve the activity of these enzymes. We used Euclidian distance to calculate signature variations. If the signature variation was negative, the distance between signatures reduced, so we consider as a beneficial mutation. If the signature variation was positive, the distance between signatures increased, so we consider as a not beneficial mutation. We collected 27 mutations in β-glucosidases from literature and classified them in beneficial or not beneficial based on the experimental effects reported. Then, we calculated the signature variation for every mutation and compared the predicted result with the real result. We obtained a precision value of 0.89. In addition, we proposed 15 mutations for Bgl1B, a non-tolerant β-glucosidase extracted from marine metagenome. We detected experimental data in the literature for three of these mutations: H228C, H228T e H228V. The experimental data demonstrate that these mutations improve the activity even in high glucose concentrations. These results show that our method is efficient to detect mutations that increase the activity of β-glucosidases and it can help to produce new mutant enzymes that may improve the second-generation biofuel production.

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