Design of Trypanosoma rangeli sialidase mutants with improved trans-sialidase activity

Design of Trypanosoma rangeli sialidase mutants with improved trans-sialidase activity

A sialidase (EC 3.2.1.18) from the non-pathogenic Trypanosoma rangeli, TrSA, has been shown to exert trans-sialidase activity after mutation of five specific amino acids in the active site (M96V, A98P, S120Y, G249Y, Q284P) to form the so-called TrSA₅mut enzyme. By computational and hypothesis driven approaches additional mutations enhancing the trans-sialidase activity have been suggested. In the present work, we made a systematic combination of these mutations leading to seven new variants of the T. rangeli sialidase, having 6-16 targeted amino acid mutations. The resulting enzyme variants were analyzed via kinetics for their ability to carry out trans-sialidase reaction using CGMP and D-lactose as substrates. The sialidase variants with 15 and 16 mutations, respectively, exhibited significantly improved trans-sialidase activity for D-lactose sialylation. Our results corroborate, that computational studies of trans-glycosylation can be a valuable input in the design of novel trans-glycosidases, but also highlight the importance of experimental validation in order to assess the performance. In conclusion, two of the seven mutants displayed a dramatic switch in specificity from hydrolysis towards trans-sialylation and constitute the most potent trans-sialidase mutants of TrSA described in literature to date.

General information
State: Published
Organisations: Department of Chemical and Biochemical Engineering, Center for BioProcess Engineering
Contributors: Nyffenegger, C., Nordvang, R. T., Jers, C., Meyer, A. S., Mikkelsen, J. D.
Number of pages: 11
Publication date: 2017
Peer-reviewed: Yes

Publication information
Journal: PLOS ONE
Volume: 12
Issue number: 2
Article number: e0171585
ISSN (Print): 1932-6203
Ratings:
BFI (2018): BFI-level 1
Web of Science (2018): Indexed yes
BFI (2017): BFI-level 1
Scopus rating (2017): CiteScore 3.01 SJR 1.164 SNIP 1.111
Web of Science (2017): Indexed yes
BFI (2016): BFI-level 1
Scopus rating (2016): CiteScore 3.11 SJR 1.236 SNIP 1.101
Web of Science (2016): Indexed yes
BFI (2015): BFI-level 1
Scopus rating (2015): CiteScore 3.32 SJR 1.427 SNIP 1.136
Web of Science (2015): Indexed yes
BFI (2014): BFI-level 1
Scopus rating (2014): CiteScore 3.54 SJR 1.559 SNIP 1.148
Web of Science (2014): Indexed yes
BFI (2013): BFI-level 1
Scopus rating (2013): CiteScore 3.94 SJR 1.772 SNIP 1.153
ISI indexed (2013): ISI indexed yes
Web of Science (2013): Indexed yes
BFI (2012): BFI-level 1
Scopus rating (2012): CiteScore 4.15 SJR 1.982 SNIP 1.156
Web of Science (2012): Impact factor 3.73
ISI indexed (2012): ISI indexed yes
Web of Science (2012): Indexed yes
BFI (2011): BFI-level 1
Scopus rating (2011): CiteScore 4.58 SJR 2.425 SNIP 1.233
Web of Science (2011): Impact factor 4.092
ISI indexed (2011): ISI indexed no