

Structure and Function of Fructose 6-phosphate aldolase

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Background

Fructose 6-phosphate aldolase (FSA) catalyzes formation of fructose 6-phosphate from dihydroxyacetone and D-glyceraldehyde 3-phosphate via an aldolization reaction (1).

Methods

Cryo-EM structures of FSA from *E. coli* were obtained of the wild type enzyme and a mutant, L107C/A129G/R134V/L163C/S166G (FSAm), that had been identified as having substantially improved activity(2).

Results

Both proteins form D5 symmetrical decamers and X-ray structures (pdb: 7qxf, 1l6w) could be used for model building(2,3). FSAm samples contained the substrate 2-hydroxyacetophenone and the map had density corresponding to the crucial iminium reaction intermediate captured in the active site. The refined model showed a modification of a lysine residue (Lys85), 2-imino-2-phenylethanol-lysine not previously described in the Chemical Component Dictionary of the pdb database. Moreover, the side chain of the critical residue Y131 was rotated 100 degrees shifting the position of the phenolic group 3 Å. A water molecule participating in a hydrogen bonding network at the active site and thought to be involved in proton relay was shifted similarly.

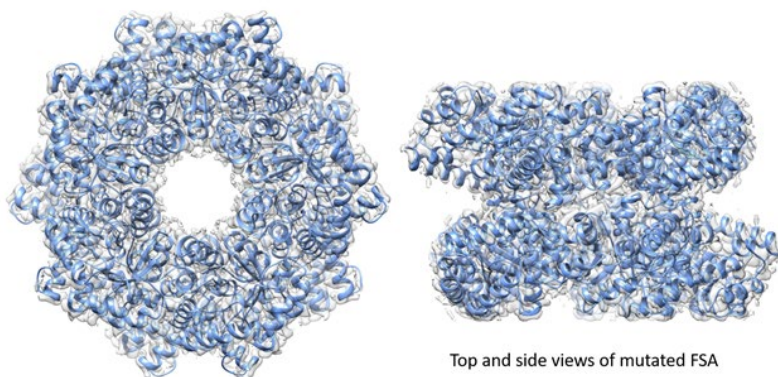
Conclusion

The new structure can guide identification of additional FSA variants that display improved carbonylation activities with 2-hydroxyacetophenone and phenylacetaldehyde.

Keywords

Catalysis, Fructose 6-phosphate aldolase, cryo-EM

Graphic:



Top and side views of mutated FSA

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Reference:

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