

Problem Set 1 **BIOC 425**

Due Oct 6, 2023

1a. The first figure of this problem set identifies some of the biochemical reactions performed by enzymes of the crotonase superfamily. For each reaction, indicate the type of reaction being perform according to the EC. Classification (ie. Hydrolase, oxidoreductase, lysase etc)

1b. Of these reactions, which one is most amenable to biocatalysis. Explain your answer.

- 2.Transglutaminase (EC 2.3.2.13) catalyzes the formation of a peptide bond between polypeptide chains. The acyl donor is the γ -carboxamide group of glutamine and the acyl acceptor is the amino group of lysine. Ammonia is formed as a by-product.
- a. What is the reaction mechanism for transglutaminase.(3 points)
- b. What industrial processes utilizes transglutaminase? Are there any other applications for its use? (3 points)

c. Is the enzyme used in a kinetic or equilibrium controlled process? Explain your answer (hopefully including a rxn mechanism) (3 points)

d. What do the EC numbers refer to in the case of transglutaminase.(2 points)

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3. Below is the fasta sequence for CreD from *Steptomyces cremeus*, which catalyzes the following reaction:

Streptomyces cremeus

>ALA99201.1 CreD [Streptomyces cremeus]
MTRPPAPPPGAPGADELLDCGLLSPVRAGTPVEALVCDSAWLQAMLDAEAALTRAQARTGFLPAAAAEAI
TAAARADRIDLLAVARGARETANPVVGLVAALTAAVRRDDPAAAEYVHRGSTSQDVLDTGAMLVARRALR
LIGDDLDRAADALAALAADHRDTPMAGRTLALHAVPTTFGLKAAGWLELVSEAAGRVARLRDGLPFSLGG
AAGTLAGYFGDRTDRGDPAVLLDRLLDAYAAETGLARPVLPWHVLRTPVADLAAVLAFTAGALGKIAVDV
QSLARTEVAEVAEPAVEGRGASSAMPHKRNPVLSTLIRSAALQVPALATGLTQCLVSEDERSAGAWHAEW
QPLRECLRLTGGAARTAVELAAGLEVDAARMRANLDLTDGRIVSESVAVALTPLLGRQAAKELLTRAAFT
AGHEGRTLGEVLGELPELDGVLPKERWEALLDPARATGVAGALVDGALARRRPPAR

I would like you to Blast this protein against the UniProtKB/SwissProt database. Perform a multiple sequence alignment, download the fasta alignment. Input the alignment file into the Weblogo sequence generator. Find two consensus sequence and use these to design degenerate oligonucleotides.

Websites that you will use:

https://blast.ncbi.nlm.nih.gov/Blast.cgi?PROGRAM=blastp&PAGE_TYPE=BlastSearch&LINK_LOC =blasthome

https://weblogo.berkeley.edu/logo.cgi

hint: select multi-line logo