Chymotrypsin is considered a serine protease and requires the amino acids Ser 195 and His 57 in the active site for catalytic activity. This enzyme exhibits several catalytic mechanisms, including acid-base catalysis, covalent catalysis and transition state binding catalysis. Use a diagram to illustrate the catalytic mechanism of chymotrypsin (use the curved arrow convention to trace electron-pair rearrangements).
QUESTION 2

2.1 How do carbamoyl phosphate, aspartate, ATP, and CTP affect the T ⇋ R equilibrium of ATCase (aspartate transcarbamoylase)? (6)

2.2 Covalent modification is an example of how enzyme regulation prevents unnecessary build up of metabolites. Give a common example of an enzyme that is regulated by covalent modification and a brief explanation of how covalent modification regulates its activity. (5)

2.3 Regulatory enzymes in metabolic pathways are often found in the first step that is unique to that pathway. How does regulation at this point improve metabolic efficiency? (3)

[14]

QUESTION 3

When a slice of apple is exposed to air, it quickly turns brown. This is because the enzyme o-diphenol oxidase catalyzes the oxidation of phenols in the apple to dark-coloured products (reaction shown in (A)). An alternative substrate, known as PHBA (shown in (B)) can inhibit the enzyme reaction:

![Reaction Diagram]

a) What type of inhibition will you observe if excess inhibitor PHBA (B), which binds to the same site as catechol, is added to the reaction mix. Briefly motivate your answer. (3)
b) Use a double reciprocal plot and illustrate (hypothetically) the rates of reaction that occurs during reaction (A) and (B). (4)

c) Write a reaction scheme to illustrate the complexes involved in the type of inhibition described in question (a). (3)

**Question 4**

Discuss the mechanism of action of the 20s Proteasome. (6)

TOTAL: 40
SECTION B

QUESTION 1

A: \[ H_4 = 20.8\% \]
\[ H_3M = 36.4\% \]
\[ H_2M_2 = 25.2\% \]
\[ HM_3 = 12.0\% \]
\[ M_4 = 5.6\% \]

B: \[ H_4 = 37.4\% \]
\[ H_3M = 41.2\% \]
\[ H_2M_2 = 8.6\% \]
\[ HM_3 = 12.8\% \]
\[ M_4 = 0\% \]

Figure A represents the LDH isozyme patterns as detected by electrophoresis (pH 8.8) of normal serum, while figure B represents the patterns of an ill patient. The patient complained about shortness of breath and chest/abdominal pain. He has not recently had surgery, but has previously been exposed to viral hepatitis. The patient is not anemic. \([H = \alpha; M = \beta]\)

a) What would your interpretation of this report be? (4)
b) What would you diagnose the patient with? (2)
c) What other enzyme(s) would you analyze to confirm your diagnosis? (2)

[8]

QUESTION 2

Elaborate on the use of enzymes for the treatment of patients with burn wounds. [4]

QUESTION 3

a) Describe the processes involved in the manufacturing of:

i) natural sweeteners (fructose ⇔ glucose) (2)

ii) artificial sweeteners (aspartame). (4)
b) Discuss the contribution genetic engineering has made to the first step (milk clotting) of the cheese-making industry (4)

c) What is/are the function/s of the following enzymes/micro-organisms in the cheese-making industry:

i) *Streptococcus* spp.

ii) *Penicillium roqueforti*

iii) Lipases (3)

**QUESTION 4**
The most widely used biosensor in clinical diagnosis is for the measurement of blood glucose. Describe which enzymes are used to measure glucose (show reactions) and elaborate on the principles of these specific biosensors. (10)

**QUESTION 5**
a) The decision on type of support material for the immobilization of enzymes/whole cells requires careful evaluation. List any four properties that are important when choosing a support material. (4)

b) Discuss the rationale behind using multi-enzyme complexes as immobilized units, and give an example of such a complex. (2)

**QUESTION 6**
‘Biostoning’ is an alternative method to produce stone-washed denim material. Describe and compare the classical method vs. the enzymatic method of stone washing denim. (10)

**QUESTION 7**
‘Enzymological methods have been useful in providing supporting evidence for myocardial infarction and also monitoring the course of the infarct’. Motivate this statement in detail. (9)

**TOTAL: 60**