

BL401/CH401 -- Biochemistry --AMINO ACID (AA) SEQUENCE PROBLEMS

1. Acid hydrolysis and AA analysis of a peptide 1 yielded: Arg, Glu, 2 Val, Gly, Lys, Tyr, Thr & Phe.

A) Dansylation gave Dansyl-Glu, while Thr was released first by carboxypeptidase.

B) Cleavage of peptide 1 with trypsin gave 3 peptides: T-1, T-2 & T-3.

- T-1 was a tripeptide; T-2 a dipeptide; T-3 a tetrapeptide.
- T-1 had AA composition = Arg, Tyr, Glu.
- N-terminal of T-2 was Val.
- Edman degradation of T-3 gave Phe, while its C-terminal was Thr.

C) Cleavage of peptide 1 with chymotrypsin gave 3 peptides: C-1, C-2 & C-3.

- C-1 was a tripeptide; C-2 a dipeptide; C-3 a tetrapeptide.
- N-terminal of C-1 was Gly, while its C-terminal was Thr.
- AA composition of C-2 was Tyr, Glu.
- N-terminal of C-3 was Arg.

What is the AA sequence of peptide 1?

Draw the structure of peptide 1 at pH 7.

What is the net charge of peptide 1 at pH 7

What is its pI?

2. After cleavage of a protein X with cyanogen bromide (CNBr), peptide 2 was isolated and it was not the C- terminal peptide.

A) Treatment of peptide 2 with trypsin proved negative.

B) Chymotrypsin cleavage of peptide 2 gave 2 peptides: C-1 & C-2.

- C-1 had AA composition of His, Asp, Tyr.
- C-2 had AA composition of 2 Ala, Lys, Pro, Val and an unknown AA.

C) C-1 was digested by Edman method yielding His first then Asn.

- C-2 treated by Edman method gave Ala first, then Ala, then Val, then Lys.

What is the AA sequence of peptide 2?

Draw its structure at pH 7.

3. During physiological research on a mouse system, peptide Z was isolated with hormone activity.

Deduce the AA sequence of peptide Z from the following information:

- A) Peptide Z had no N- or C-terminal residues detected by conventional methods.
- B) Acid hydrolysis and AA analysis gave: 4 Gly, Asp, His, Val & Phe.
- C) Trypsin had no effect but chymotrypsin cleaved peptide Z into 2 peptides: C-1 & C-2.
 - C-1 had AA composition of Gly, His, Phe.
 - C-2 had AA composition of 3 Gly, Asp, Val.
 - Peptide C-2 had strong UV absorbance at 280 nm.
 - Edman degradation of C-1 gave Gly, then His.
 - Edman degradation of C-2 gave Gly, then Asp, then Gly, then Val.

What are the AA sequences of C-1 and C-2 and the overall structure of peptide Z?

Draw structure of peptide Z.

What is the net charge on peptide Z at pH 3, 5, 7, 9, 11?

What is its pI?

4. Peptide Q was hydrolyzed with acid and gave AA composition:

2 Pro, Ser, Thr, 2 Lys, 2 Arg, Phe, Tyr, Met, Gly, Asp, 2 Glu.

Dansylation of peptide Q gave Dansyl-Asp.

A) Cleavage with CNBr yielded 2 peptides: CB-1 and CB-2.

- i) CB-1 had AA composition of Tyr, Lys, Gly, Asp, Ser, HSL (homo-serine lactone).
 - When CB-1 was cleaved with trypsin, 2 peptides were obtained
 - One of them had AA composition of Gly, Asp, Lys, Ser

- The other had AA composition of HSL, Tyr.

ii) CB-2 was cleaved by trypsin into 3 peptides and by chymotrypsin into 2 peptides.

- One tryptic peptide of CB-2 was degraded by Edman giving Gln, then Pro, then Glu.
- AA composition of peptides of CB-2 were #1 = Pro, Arg, Lys, Phe; #2 = Arg, Thr, Pro, 2 Glu.

B) Peptide Q was cleaved by chymotrypsin into 3 peptides.

- CH-1 yielded by Edman Met, then Lys.
- CH-2 had AA composition of 2 Glu, Thr, Pro, Arg.
- Tryptic digestion of CH-2 yielded 2 peptides, one of which had AA composition of Thr, Arg; while the other was a tripeptide.
- CH-3 had AA composition of Lys, Asp, Tyr, Gly, Ser
- Edman degradation of CH-3 gave Asp, then Ser, then Gly.

C) Peptide Q was degraded by trypsin into 4 peptides.

- T-1 had AA composition of Pro, 2 Glu.
- T-2 had AA composition of Phe, Thr, Arg.
- T-3 had AA composition of Met, Tyr, Arg, Pro, Lys - Edman on T-3 gave Tyr, then Met, then Lys.
- T-4 had AA composition of Asp, Ser, Gly, Lys.

What is AA sequence of peptide Q?

5. Amino acid sequence problem from 1993 BL401/CH401 Exam I.

Peptide P had AA composition after acid hydrolysis: Arg, Ser, Met, 2 Glu, Pro, Phe, Leu, Lys & Tyr.

A) It had no N- or C-terminal found by dansylation and carboxypeptidase.

B) Tryptic cleavage of peptide P gave one peptide (Tryptic peptide P) with same AA composition as peptide P.

- Edman degradation of Tryptic peptide P gave: 1st cycle Gln, 2nd Tyr, 3rd Ser.

C) Chymotryptic cleavage of peptide P gave two peptides.

- Ch-1 had AA composition: Arg, Phe, Ser, Pro.

- Edman on Ch-1 gave: 1st cycle Ser, 2nd Arg.
- Ch-2 had AA composition: Leu, Lys, Tyr, Met, 2 Glu.
- Edman on Ch-2 gave: 1st cycle Leu, 2nd Met, 3rd Glu.

D) CNBr cleavage of peptide P gave one peptide with AA composition: 2 Glu, Tyr, Pro, Phe, Leu, HSL, Lys, Ser, Arg.

- Edman on CNBr-1 gave 1st cycle Glu, 2nd Lys, 3rd Gln.

What is the amino acid sequence of peptide P?

Draw the structure of the Tryptic peptide P at pH 8.

What is the net charge on the Tryptic peptide P at pH 8?

What is the pI of the Tryptic peptide P?

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