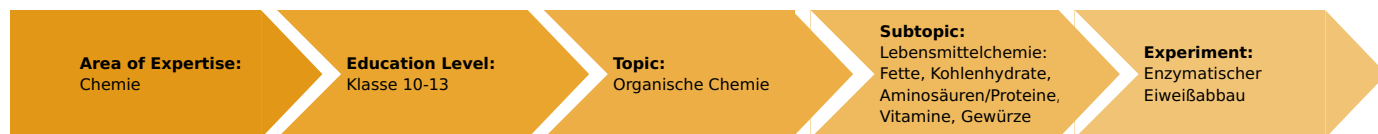


# Enzymatic cleavage of proteins (Item No.: P7188800)

## Curricular Relevance



### Difficulty



Intermediate

### Preparation Time



10 Minutes

### Execution Time



20 Minutes

### Recommended Group Size



2 Students

### Additional Requirements:

### Experiment Variations:

### Keywords:

proteins, enzymatic cleavage

## Task and equipment

## Information for teachers

## Additional Information

Protein is one of our most important nutrients. To maintain life, only certain proteins are required. These cannot be won directly from food. The proteins must be prepared and transformed in our digestive system. This is particularly true for animal proteins.

## Notes on content and learning objectives

- Protein is decomposed in the stomach by the protein-splitting (=proteolytic) enzyme pepsin.
- The digestive enzyme pepsin, which occurs in the stomach of man and animals, is most active at pH values between 1 and 3.
- Protein-splitting enzymes are used, e.g. in the production of quark, as meat tenderizers and to prevent protein precipitation in beverages.

## Notes on the method

The experiment "coagulation of proteins" can be carried out to answer question 2.

The significance of enzymes as bio-catalysts should be gone deeper into in a classroom discussion.

## Fundamentals and remarks

Proteins can be split into their components, oligopeptides and amino acids, by proteolytic enzymes.

Pepsin is the most important enzyme in gastric juice. It is a digestive enzyme which has its optimal action at pH values of between 1 and 3, which is appropriately adjusted by the gastric acid.

Papain in meat tenderizer is extracted from the sap of the tropical melon tree "Carica papaya". It is sensitive to oxidation and is therefore frequently brought to market with added reducing agent. Other protein-splitting enzymes of vegetable origin are bromelain (from pineapples) and ficin (from figs).

## Hints on going deeper

- Production of quark or cheese from full-cream milk using rennet tablets.
- The action of enzymes in the conversion of starch to grape sugar.

## Notes on the set-up and procedure

## Preparation:

The following solutions should be prepared centrally:

### Egg white solution:

Carefully separate the egg white of a hen's egg from the yolk. Stir the egg white into 100 ml of 1 % common salt solution (1 g Sodium chloride solved in 100 ml of distilled water). Subsequently filter the solution through glass wool.

### 1 % Pepsin solution:

Dissolve 1 g of pepsin in 100 ml of distilled water.

### 5 % Aqueous solution of a meat tenderizer:

Stir 5 g of meat tenderizer in 100 ml of distilled water. Should the mixture be turbid, filter through glass wool.

### Notes on the students experiment:

The protein-splitting effect of pepsin and papain (in meat tenderizers) can also be investigated using other proteins, such as meat juice or soybean protein.

The protein-splitting activity of pepsin is particularly high at the body temperature of mammals, i.e. 30-40 °C.



## Hazard and Precautionary statements

### Pepsin

- H315: Causes skin irritation.  
 H319: Causes serious eye irritation.  
 H334: May cause allergy or asthma symptoms or breathing difficulties if inhaled.  
 H335: May cause respiratory irritation.  
 P302+P352: IF ON SKIN: Wash with soap and water.  
 P304+P341: IF INHALED: If breathing is difficult, remove victim to fresh air and keep at rest in a position comfortable for breathing.  
 P305+P351+P338: IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses if present and easy to do – continue rinsing.  
 P342+P311: If experiencing respiratory symptoms: Call a POISON CENTER or doctor/physician.

### Hydrochloric acid

- H290: May be corrosive to metals.  
 H314: Causes severe skin burns and eye damage.  
 H335: May cause respiratory irritation.  
 P234: Keep only in original container.  
 P260: Do not breathe dust/fume/gas/mist/vapours/spray.  
 P303+P361+P353: IF ON SKIN (or hair): Remove/Take off immediately all contaminated clothing. Rinse skin with water/shower.  
 P304+P340: IF INHALED: Remove victim to fresh air and keep at rest in a position comfortable for breathing.  
 P305+P351+P338: IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses if present and easy to do – continue rinsing.  
 P309+P311: IF exposed or you feel unwell: Call a POISON CENTER or doctor/physician.

## Hazards

- Hydrochloric acid is corrosive. Avoid skin-contact with this chemical.
- Wear protective glasses and protective gloves.

## Waste disposal

Pour the solutions in the test tubes to drain.

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## Task and equipment

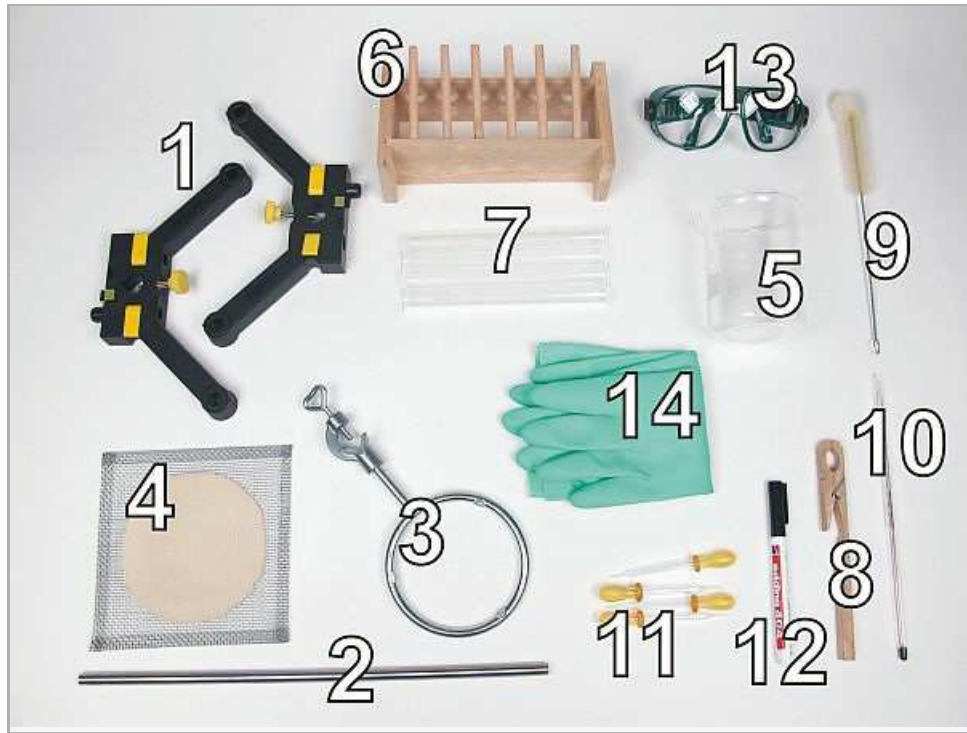
### Task

#### How is protein decomposed in the stomach?

Test the action of different protein-splitting enzymes.



Equipment



Position No.	Material	Order No.	Quantity
1	Support base, variable	02001-00	1
2	Support rod, stainless steel, l=370 mm, d=10 mm	02059-00	1
3	Ring with boss head, i. d. = 10 cm	37701-01	1
4	Wire gauze with ceramic, 160 x 160 mm	33287-01	1
5	Glass beaker DURAN®, short, 400 ml	36014-00	1
6	Test tube rack for 12 tubes, holes d= 22 mm, wood	37686-10	1
7	Test tube, 180x18 mm,100pcs	37658-10	(4)
8	Test tube holder, up to d 22mm	38823-00	1
9	Test tube brush w. wool tip,d25mm	38762-00	1
10	Students thermometer,-10...+110°C, l = 180 mm	38005-02	1
11	Pipette with rubber bulb	64701-00	4
12	Labor pencil, waterproof	38711-00	1
13	Protecting glasses, clear glass	39316-00	1
14	Rubber gloves, size S (7)	39325-00	1
	Butane burner f.cartridge 270+470	47536-00	1
	Butane cartridge CV 300 Plus, 240 g	47538-01	1
	Pepsin powder,soluble 100 g	30181-10	1
	Water, distilled 5 l	31246-81	1
	Hydrochloric acid 25% 1000 ml	31822-70	1
	Boiling beads, 200 g	36937-20	1
Additional material			
	Egg		
	Meat tenderizer		

## Set-up and procedure

### Set-up

### Hazards

- Hydrochloric acid is corrosive. Avoid skin-contact with this chemical.
- Wear protective glasses and protective gloves.



Number four test tubes from 1 to 4 and stand them next to each other in the test tube rack (Fig. 1).



Assemble the stand as shown in figures 2 to 6. Fasten the support ring to the support rod and place the wire gauze on it. Adjust the height of the support ring so that the flame of the burner just reaches the wire gauze.



Fig. 2



Fig. 3



Fig. 4



Fig. 5



Fig. 6

Half-fill a 400 ml beaker with water and add a few boiling stones (Fig. 7). Heat it to boiling, then put it aside. Extinguish the bunsen burner flame!



Fig. 7

## Procedure

Pipette egg white solution to a height of 2 cm in each of the four test tubes (Fig. 8). Place the test tubes containing egg white solution in the prepared water bath until the protein solutions are turbid (Fig. 9).



Fig. 8



Fig. 9

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Pipette pepsin solution (1 %) into test tubes 1 and 2, to a height of 6 cm.

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Add the same amount of a 5 % aqueous solution of a meat tenderizer to test tubes 3 and 4 (Fig. 10).



Fig. 10

Pipette 10 drops of hydrochloric acid (25 %) to the solutions in test tubes 2 and 4 (Fig. 11).



Fig. 11

Using a student thermometer check the temperature of the water bath, it should be 30-40 °C. If the temperature of the water bath is higher, then let it cool for some time until the temperature has reached 30-40 °C; should the temperature of the water bath be cooler, then heat it up again above the Bunsen burner flame.

Place all four test tubes in the beaker containing warm water for approx. 30 minutes (Fig. 12).





Fig. 12

## Waste disposal

Pour the solutions in the test tubes to drain.

## Report: Enzymatic cleavage of proteins

### Result - Table 1

Note your observations in the following table:

Test tube	Protein solution with	Dissolution of the turbidity of the protein
1	Pepsin	1
2	Pepsin and hydrochloric acid	1
3	Meat tenderizer	1
4	Meat tenderizer and hydrochloric acid	1

### Evaluation - Question 1

Draw conclusions from your observations.

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### Evaluation - Question 2

How can enzymes be inhibited?

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### Evaluation - Question 3

For which purpose are protein-cleaving enzymes added during the production of foods?

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### Evaluation - Question 4

Complete the following statements:

1. The digestive enzyme ....., in combination with ....., cleaves proteins.
2. Pepsin is only active in ..... liquids.
3. The digestive enzyme ..... cleaves proteins in ..... solutions.
4. Protein-splitting enzymes are used in the production of ..... and .....