Practical 5.17 Protein materials

Purpose

To describe and carry out experiments, where appropriate, to investigate the characteristic behaviour of amino acids [5.4.2i]

Safety

Wear eye protection. Ninhydrin in propanone is harmful – the spray from an aerosol can is highly flammable. Only spray in a fume cupboard and wear gloves.

Each group of students will need:
Eye protection
Gloves (nitrile)
Test tubes and rack
Dropping pipette
Chromatography paper

Access to:
Fume cupboard
Full-range indicator solution
0.01 M hydrochloric acid
0.01 M L-glutamic acid
0.01 M glycine
0.01 M sodium hydroxide
0.02 M ninhydrin solution in propanone (store in refrigerator); other solvents can sometimes be used HARMFUL, HIGHLY FLAMMABLE
Protein materials, e.g. casein, gelatin, fresh milk, egg, etc.
Oven at 110 °C
Polarimeter (optional)
Sodium glutamate, 50 cm^3 of concentrated solution for polarimetry (or sucrose – see Chirality)

Method

Make aqueous solutions of your samples of protein materials and amino acids, warming if necessary, and use them for the following tests.

1 Acidity and basicity

To 2 cm^3 of 0.01 M hydrochloric acid add a few drops of full-range indicator and note the effect on the pH of adding 0.01 M sodium hydroxide in 0.5 cm^3 portions.

Repeat the experiment, using a solution of 0.01 M glycine in place of the hydrochloric acid.

Then repeat in reverse, adding 0.01 M sodium hydroxide instead of hydrochloric acid.

a Does the pH change gradually or sharply?

b What type of acid–base behaviour is occurring?

c Write an equation for the reactions of glycine with hydrochloric acid and with sodium hydroxide.

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2 Ninhydrin test

Ninhydrin is a reagent used as a specific colour test for amino acids. You do not need to be concerned with the formula of the compound (which is complicated) nor with the details of the chemical reaction which produces the colours.

Put small drops of your solution onto a piece of chromatography paper, and allow to dry. Spray lightly with 0.02 M ninhydrin solution in propanone (take care: *spray in a fume cupboard*) and again allow to dry. Avoid getting the spray on your fingers by wearing gloves. Heat for 10 minutes in an oven at 110 °C. Red to blue coloured spots will develop if proteins or amino acids are present.

d Make a note of any unexpected coloured areas.

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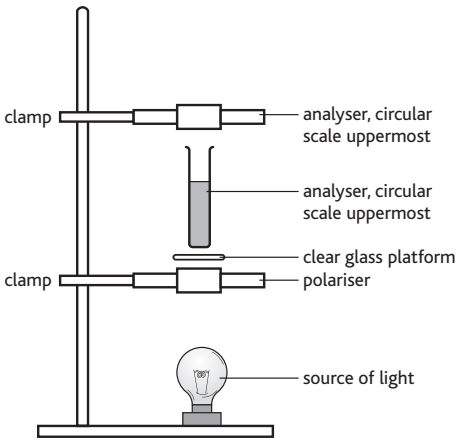
3 Chirality

You will need a polarimeter for this optional experiment.



Compounds which rotate the plane of polarised light are said to be *optically active* and this is the standard property by which chiral compounds can be recognised in the laboratory. The two isomers of a chiral compound will produce the same amount of rotation, but in opposite directions.

The property can be indicated by adding a prefix to their names: for example, (+) glutamic acid and (−) glutamic acid.

You also need to know that:

- the rotation of the plane of polarisation in the clockwise sense, as viewed by an observer looking towards the source of light, is given a (+) sign
- the (+) isomer rotates the plane of polarised light clockwise; the (−) isomer has the opposite effect.

Remove the specimen tube from the polarimeter. Adjust the polarimeter by rotating the centre of the analyser until, on looking through the analyser and polariser, you see that the source of light is extinguished. Note the position of the pointer on the scale.

Half fill the specimen tube with concentrated sodium glutamate solution and put it in position. Look through the instrument once more. Do you have to alter the setting of the analyser to extinguish the light, and if so, by how much?

Now fill the specimen tube so as to double the length of the liquid through which the light passes. Is a further adjustment of the analyser necessary for extinction of the light?

Edexcel practical materials adapted from Nuffield Advanced Chemistry materials