

Figure 2.5.26.-1. – Chemiluminescence analyser

01/2005:20527

## 2.5.27. OXYGEN IN GASES

Oxygen in gases is determined using a paramagnetic analyser.

The principle of the method is based on the high paramagnetic sensitivity of the oxygen molecule. Oxygen exerts a strong interaction on magnetic fields, which is measured electronically, amplified and converted to a reading of oxygen concentration. The measurement of oxygen concentration is dependent upon the pressure and temperature and, if the analyser is not automatically compensated for variations in temperature and pressure, it must be calibrated immediately prior to use. As the paramagnetic effect of oxygen is linear the instrument must have a suitable range with a readability of 0.1 per cent or better.

*Calibration of the instrument.* Make the setting in the following manner:

- set the zero by passing *nitrogen R1* through the instrument at a suitable flow rate until a constant reading is obtained. It should be set to zero according to the manufacturer's instructions;
- set the appropriate limit by passing air (20.9 per cent  $V/V$   $O_2$ ) through the instrument at a suitable flow rate until a constant reading is obtained. The limit should be set to 20.9 per cent  $V/V$  in accordance with the manufacturer's instructions.

*Assay.* Pass the gas to be examined through the instrument at a constant flow rate until a suitable reading is obtained.

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## 2.5.28. WATER IN GASES

Water in gases is determined using an electrolytic hygrometer, described below.

The measuring cell consists of a thin film of diphosphorus pentoxide, between 2 coiled platinum wires which act as electrodes. The water vapour in the gas to be examined

is absorbed by the diphosphorus pentoxide, which is transformed to phosphoric acid, an electrical conductor. A continuous voltage applied across the electrodes produces electrolysis of the water and the regeneration of the diphosphorus pentoxide. The resulting electric current, which is proportional to the water content in the gas to be examined, is measured. This system is self-calibrating since it obeys Faraday's law.

Take a sample of the gas to be examined. Allow the gas to stabilise at room temperature. Purge the cell continuously until a stable reading is obtained. Measure the water content in the gas to be examined, making sure that the temperature is constant throughout the device used to introduce the gas into the apparatus.

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## 2.5.29. SULPHUR DIOXIDE

Introduce 150 ml of *water R* into the flask (A) (see Figure 2.5.29.-1) and pass *carbon dioxide R* through the whole system for 15 min at a rate of 100 ml/min. To 10 ml of *dilute hydrogen peroxide solution R* add 0.15 ml of a 1 g/l solution of *bromophenol blue R* in *alcohol (20 per cent V/V) R*. Add 0.1 M *sodium hydroxide* until a violet-blue colour is obtained, without exceeding the end-point. Place the solution in the test-tube (D). Without interrupting the stream of carbon dioxide, remove the funnel (B) and introduce through the opening into the flask (A) 25.0 g of the substance to be examined (*m g*) with the aid of 100 ml of *water R*. Add through the funnel 80 ml of *dilute hydrochloric acid R* and boil for 1 h. Open the tap of the funnel and stop the flow of carbon dioxide and also the heating and the cooling water. Transfer the contents of the test-tube with the aid of a little *water R* to a 200 ml wide-necked, conical flask. Heat on a water-bath for 15 min and allow to cool. Add 0.1 ml of a 1 g/l solution of *bromophenol blue R* in *alcohol (20 per cent V/V) R* and titrate with 0.1 M *sodium hydroxide* until the colour changes from yellow to violet-blue ( $V_1$  ml). Carry out a blank titration ( $V_2$  ml).

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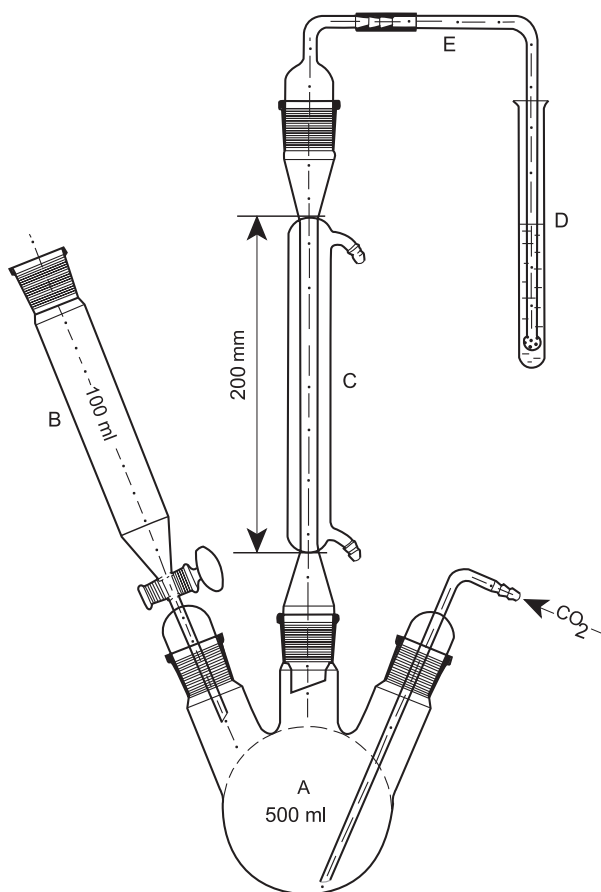


Figure 2.5.29-1.- Apparatus for the determination of sulphur dioxide

Calculate the content of sulphur dioxide in parts per million from the expression:

$$32\,030 \times (V_1 - V_2) \times \frac{n}{m}$$

$n$  = molarity of the sodium hydroxide solution used as titrant.

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### 2.5.30. OXIDISING SUBSTANCES

Transfer 4.0 g to a glass-stoppered, 125 ml conical flask and add 50.0 ml of *water R*. Insert the stopper and swirl for 5 min. Transfer to a glass-stoppered 50 ml centrifuge tube and centrifuge. Transfer 30.0 ml of the clear supernatant liquid to a glass-stoppered 125 ml conical flask. Add 1 ml of *glacial acetic acid R* and 0.5 g to 1.0 g of *potassium iodide R*. Insert the stopper, swirl, and allow to stand for 25 min to 30 min in the dark. Add 1 ml of *starch solution R* and titrate with 0.002 M *sodium thiosulphate* until the starch-iodine colour disappears. Carry out a blank determination. Not more than 1.4 ml of 0.002 M *sodium thiosulphate* is required (0.002 per cent, calculated as  $H_2O_2$ ).

1 ml of 0.002 M *sodium thiosulphate* is equivalent to 34  $\mu$ g of oxidising substances, calculated as hydrogen peroxide.

### 2.5.31. RIBOSE IN POLYSACCHARIDE VACCINES

**Test solution.** Use a volumetric flask with a suitable volume for preparation of a solution containing about 5 mg per millilitre of dry polysaccharide. Transfer the contents of a container quantitatively to the flask and dilute to volume with *water R*. Dilute the solution so that the volumes used in the test contain 2.5  $\mu$ g to 25  $\mu$ g of ribose. Introduce 0.20 ml and 0.40 ml of the diluted solution into tubes in triplicate.

**Reference solutions.** Dissolve 25 mg of *ribose R* in *water R* and dilute to 100.0 ml with the same solvent (stock solution containing 0.25 g/l of ribose). Immediately before use, dilute 1 ml of the stock solution to 10.0 ml with *water R* (working dilution: 25 mg/l of ribose). Introduce 0.10 ml, 0.20 ml, 0.40 ml, 0.60 ml, 0.80 ml and 1.0 ml of the working dilution into 6 tubes.

Prepare a blank using 2 ml of *water R*.

Make up the volume in each tube to 2 ml with *water R*. Shake. Add 2 ml of a 0.5 g/l solution of *ferric chloride R* in *hydrochloric acid R* to each tube. Shake. Add 0.2 ml of a 100 g/l solution of *orcinol R* in *ethanol R*. Place the tubes in a water-bath for 20 min. Cool in iced water. Measure the absorbance (2.2.25) of each solution at 670 nm using the blank as the compensation liquid. Draw a calibration curve from the absorbance readings for the 6 reference solutions and the corresponding content of ribose and read from the curve the quantity of ribose in the test solution for each volume tested. Calculate the mean of the 3 values.

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### 2.5.32. WATER: MICRO DETERMINATION

#### PRINCIPLE

The coulometric titration of water is based upon the quantitative reaction of water with sulphur dioxide and iodine in an anhydrous medium in the presence of a base with sufficient buffering capacity. In contrast to the volumetric method described under (2.5.12), iodine is produced electrochemically in the reaction cell by oxidation of iodide. The iodine produced at the anode reacts immediately with the water and the sulphur dioxide contained in the reaction cell. The amount of water in the substance is directly proportional to the quantity of electricity up until the titration end-point. When all of the water in the cell has been consumed, the end-point is reached and thus an excess of iodine appears. 1 mole of iodine corresponds to 1 mole of water, a quantity of electricity of 10.71 C corresponds to 1 mg of water.

Moisture is eliminated from the system by pre-electrolysis. Individual determinations can be carried out successively in the same reagent solution, under the following conditions:

- each component of the test mixture is compatible with the other components,
- no other reactions take place,
- the volume and the water capacity of the electrolyte reagent are sufficient.

Coulometric titration is restricted to the quantitative determination of small amounts of water, a range of 10  $\mu$ g up to 10 mg of water is recommended.