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DISCOVERED OXIGEN
Indigo Carmine Method

PRINCIPLE

The indigo-carmin method is a simple, rapid, accurate colorimetric procedure for determining small amounts of dissolved oxygen in water (0 - 50 ppm). A solution of reduced indigo-carmin dye added to a water sample is oxidized by the presence of any dissolved oxygen and changes color. The change in color of this dye is directly proportional to the amount of dissolved oxygen present in the sample and can be determined by using a Johnson color comparator.

APPARATUS

1. Glass stoppered B.O.D. bottle (clear) - 300 ml capacity.
2. Hypodermic syringe and needle - 5 ml capacity.
3. A color comparator is available from Mr. R. L. Johnson, 2430 Wallace

Drive, Jackson, Michigan. However, the following table may be used:

<u>Color</u>	<u>Oxygen (ppm)</u>
Yellow	0.000
Orange	0.005
Orange-Pink	0.010
Pink	0.015
Pink-Red	0.025
Red-Purple	0.050
Purple	0.100

REAGENTS

1. Indigo-Carmine Stock Solution: Dissolve 0.25g indigo-carmin (National Aniline Cat. #476) and 2.50g of C.P. dextrose in 220 ml distilled water. Add 280 ml of U.S.P. glycerol and store in a dark bottle, under refrigeration, if available. Use within a week or ten days.

2. Potassium Hydroxide Stock Solution: Dissolve 100 g of KOH in distilled water.
3. Working Solution: Half an hour before use, mix 20 ml of the indigo-carmin stock solution and 5 ml of the potassium hydroxide solution. On standing, the indigo-carmin dye will change from a deep red to lemon yellow and is then ready to use. Draw 4 ml plus into the syringe, invert, rid of air bubbles, and measure to exactly 4 ml.

PROCEDURE

A standard 300 ml B.O.D. bottle is placed in a Pyrex or stainless steel beaker so that the neck and stopper will be below the overflow level and is filled by means of a sample tube extending to the bottom of the bottle. The water samples should be below room temperature and allowed to run for 10 - 15 minutes.

Slowly withdraw the sample tube, leave the bottle in place and insert the tip of the syringe into the bottle as far as possible below the water level. Add 4 ml of the working indigo-carmin reagent. Replace the stopper without trapping air and invert the bottle several times to mix. Place the bottle on a white surface and estimate the oxygen from the color according to the comparator or the above table.

* * * * *

¹ L. S. Bachoff, N. M. Ingber and J. H. Brady, "Colorimetric Determination of Low Concentrations of Dissolved Oxygen in Water," Anal. Chem. 27, 1401 (1955).