Later Bessey and Wolbach[2] described other ocular changes due to Vitamin G deficiency but reported that cataract occurred in only a small percentage of their animals. El Sadr[3] observed both the corneal opacity, vascularization and cataract reported by the above workers. Day, Darby and Langston[4] subsequently identified riboflavin as the cataract preventive factor.

Since these workers had used different rations and since their results were varied, a series of experiments was undertaken to explain the inconsistency of cataract formation in rats fed on riboflavin deficient diets.

Sixty rats were placed on the diet of Bourquin and Sherman.[5] A microbiological assay by the method of Snell and Strong[6] showed this ration to contain riboflavin in such amounts that our animals received 0.57 micrograms per day on the basis of average food consumption. Another group of fifty animals was placed on a modified ration of Bourquin and Sherman in which the B-complex was supplied by adding sufficient amounts of crystalline thiamin, pyridoxin, nicotinic acid, pantothenic acid and choline, instead of the 80 per cent. alcoholic extract of wheat. Some of these animals were supplemented with varied amounts of riboflavin while others received only the basal ration.

A third group of twenty animals was placed on a riboflavin free diet in which 14 per cent. Crisco was used as a source of fat instead of the 8 per cent. filtered butter fat used in the Bourquin-Sherman diet. The animals on this ration were supplemented with 50 gamma thiamin, 20 gamma pyridoxin, 200 gamma nicotinic acid, 100 gamma pantothenic acid and 5 milligrams choline per rat per day.

The results of these experiments variously corroborate all the work done on riboflavin deficiency as it affects the eyes of rats. Corneal opacity and vascularization occurred in all animals except those receiving adequate amounts of riboflavin. Cataract occurred in 90 per cent. of the animals on the Bourquin-Sherman diet within 9 weeks.

On the modified ration, 85 per cent. of the animals receiving between 1 and 3 micrograms of riboflavin daily developed cataract in 10 weeks. On the modified ration without the riboflavin, only 14 per cent. of the animals exhibited cataract formation. The time periods in each case were comparable, *i.e.*, cataract had developed in the animals receiving small amounts of riboflavin at a period when the negative controls were alive and exhibiting some growth but without the development of cataract.

All the animals on the third ration failed to develop cataract within twelve weeks after which they were discarded.

These results indicate that rats on a ration devoid of riboflavin do not exhibit cataract. Minute amounts of riboflavin induce cataract formation and rations containing more nearly adequate amounts are non-cataractogenic. This would explain the inconsistency reported by various workers of cataract production on rations which were deficient but not entirely riboflavin free. Our results are also in agreement with the observations of Stokstad and Manning[7] on the incidence of the curled toe paralysis syndrome in chicks. These findings demonstrated a lack of curled toe paralysis except when small amounts of riboflavin were present. A more complete report of these findings will appear elsewhere.

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SCIENTIFIC APPARATUS AND LABORATORY METHODS

A TEST-TUBE SPIRAL ABSORPTION VESSEL

A SIMPLE, efficient and inexpensive vessel for carbon dioxide absorption has been constructed and extensively used for plant respiration measurements. As seen in Fig. 1, this absorption cell has the added advantage of compactness. This feature is obtained by the use of a test-tube for the shell of the apparatus.

The nucleus around which the absorption cell is built consists of a glass spiral through which a chain of gas bubbles moves in contact with the absorbing solution. This spiral tube lengthens the path of the bubbles and thus prolongs the time of contact between the gas and the solution. By this means, increased efficiency of absorption is obtained. Apparatus built upon this principle have been described, heretofore, by Harvey and Regeimbal,[1] and also by many other designers of similar apparatus.

As indicated in Fig. 1, the apparatus includes a test-tube (b) fitted with a two-hole rubber stopper (a) through which pass a long inlet tube and a short outlet tube. The inlet tube is bent just below the rubber stopper so that its longer portion is centered in the

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[4] P. L. Day, W. J. Darby and W. C. Langston, *Jour. Nutrition*, 13: 389, 1937.
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test-tube. The lower end of the inlet tube is drawn out to form a short nozzle (d), which is bent sharply upward. A spiral of glass tubing (c), open at both ends, fits loosely over the upturned nozzle. As the carbon-dioxide laden air stream passes through the nozzle and enters the spiral in the form of bubbles, some of the absorbing liquid also enters the spiral. This results in a continuous circulation of liquid, both through the spiral tube in which the absorption takes place and in the test-tube itself.

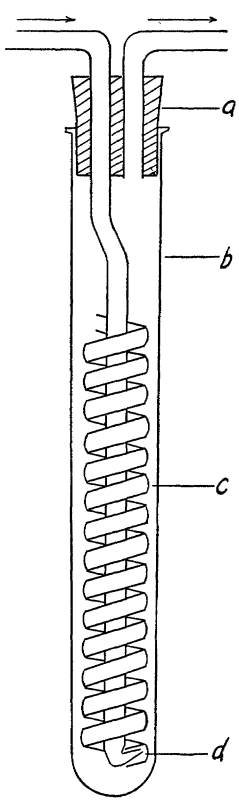

FIG. 1. Details of absorption vessel: (a) two-hole rubber stopper; (b) test-tube; (c) glass spiral; (d) nozzle at end of inlet tube.

The spiral may be formed of glass tubing softened in the flame of a blast lamp and turned around a piece of brass tubing of the proper size. A slight taper on the brass tube makes for ease in the removal of the glass spiral after turning.

Two sizes of these absorption vessels have been constructed and used in respiration studies. The smaller one holds 25 ml of solution and consists of a 22×175 mm test-tube with spiral and inlet tube made of 6 mm tubing. The spiral is 18 mm in diameter and 105 mm in length. The larger vessel utilizes a 25×200 mm test-tube with proportionately larger spiral and inlet tube. This absorption vessel holds 50 ml of solution. In all cases, the volume of the absorbing solution should be sufficient to cover the top of the spiral but should not be so great as to cause liquid to be forced up into the outlet tube when in operation.

Potassium and sodium hydroxide solutions of 0.10 and 0.05 normality have been successfully used as carbon-dioxide absorbents with this apparatus. Solutions of barium hydroxide are not recommended because of a tendency to block the inlet nozzle with precipitated carbonate. At the end of a run, the absorption vessel is disconnected and tilted so as to drain out the alkali solution through the outlet tube into a small flask. This flask should then be stoppered to protect the solution from atmospheric carbon dioxide. Aliquots may be pipetted from this solution and titrated in the presence of excess barium chloride against phenolphthalein with dilute standard hydrochloric acid (0.10 or 0.05 N). The addition of barium chloride in excess results in the precipitation of the absorbed carbon dioxide as barium carbonate previous to the titration. Acid titration values thus obtained give a measure of the unused hydroxide and, in comparison with corresponding values obtained with the original solution, give an accurate index of the carbon dioxide absorption.

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A SIMPLE METHOD OF REMOVING SCALES FROM LARGE LEPIDOPTERA

CONSIDERABLE difficulty is usually experienced in removing scales from the bodies of large thickly clothed Heterocera in preparation for morphological study. The following method has been used successfully by the writer with specimens of the tobacco hornworm (*Protoparce sexta* (Johan.)). This method can be used effectively with either fresh or dried specimens, although fresh specimens are denuded more easily. The wings are removed and the scales are dislodged from the body by brushing with a small toothbrush. A child's brush with moderately stiff bristles, all of approximately equal length, will be found best for this purpose. Stroking is most effective over the soft chitin of the abdomen, while a reciprocating motion produces better results on the head and thorax.

Fine scales on the legs and wings can not be removed readily by brushing. These parts may be cleaned easily by placing them in a 5.25 per cent. solution of sodium hypochlorite for a short period. They should be removed and rinsed in distilled water not over two hours after immersion in this solution.

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