

lowered from the top of a steep high cliff to within eight feet of the nest.

On Saturday morning papers were read and discussed by members of the academy, and a visit was made to the Libbey Museum. In the afternoon the papers remaining on the program were read, and the retiring president, Professor J. W. Goldthwait, delivered the presidential address, "The Destruction and Preservation of Nature in New Hampshire."

At the business meeting the officers elected for 1932-33 were:

President, Professor Norman E. Gilbert, physics department, Dartmouth College.

Vice-president, Mr. Samuel P. Hunt, New Hampshire Public Service Company, Manchester.

Secretary-Treasurer, Professor Thomas G. Phillips, department of agricultural and biological chemistry, University of New Hampshire.

Member of Executive Council, Professor J. W. Goldthwait, geology department, Dartmouth College.

GEO. W. WHITE,
Retiring Secretary

SCIENTIFIC APPARATUS AND LABORATORY METHODS

A MODIFIED MEDIUM FOR PLATING *L. ACIDOPHILUS*

In a previous communication,¹ a special agar medium was reported for the routine plating of *L. acidophilus*. Within the past year a modification of this medium has been developed which has proved more efficient.

In preparing this modified medium, 1 per cent. Difco peptonized milk is added to the original formula. Since the original medium was first reported, several details of preparation have been changed for reasons not evident at the time of the previous report. Because these have a pertinent bearing on the quality of the medium, the detailed method for the preparation of the new modified medium follows.

Mixture A: Add ten grams Difco peptone and 10 grams Difco peptonized milk to 400 cc of juice filtered from a good quality of canned tomatoes. Heat this mixture gently to dissolve the peptone and peptonized milk. Unnecessary heating of the tomato juice should be avoided. The reaction of the solution is changed to pH 6.0-6.2. There should be little deviation from this suggested reaction.

Mixture B: Add 11 grams dried agar to 600 cc distilled water and autoclave this mixture to dissolve the agar.

Just previous to the removal of Mixture B from the autoclave, bring Mixture A to the boiling point. Then mix A and B while both are hot and filter through a thin layer of absorbent cotton. Distribute the filtered medium in containers (test-tubes preferred) and sterilize by heating in the autoclave at 120° C. for eight minutes.

The properly prepared medium at this point is a clear agar, of a light brown color, having a final reaction of pH 6.0-6.2.

The reaction of the finished medium must not deviate very much from pH 6.0. When this medium was first developed, the reaction before autoclaving was adjusted to pH 7.2-7.4. During the sterilizing process, there was a decided drop in the pH, the final reaction sometimes being as low as 6.0. LaMer,

¹ SCIENCE, lxvi, 1927, pp. 512-513.

Campbell and Sherman,² quoting the work of Nef, have reported that during the heating of an alkaline tomato juice the acidity increases considerably, due to the decomposing action of the hydroxyl ions on the sugars, resulting in the formation of organic acids. In an acid medium this analysis does not take place when this material is heated and, hence, the reaction remains quite stable. In our experimental work the pH of an acid tomato agar (pH 6.0) did not change after being held at 120° C. for 20 minutes.

Recently we made the interesting discovery that some strains of the X-type *L. acidophilus* develop very poorly in the tomato medium if the finished product has a reaction of pH. 6.8-7.0, especially when only a small amount (5 per cent.) of CO₂ is added to the incubation atmosphere. A method for incubation of agar plating of *L. acidophilus* in an atmosphere containing from 5 to 20 per cent. of CO₂ has been previously described by the senior author.³ Further observation brought to light the fact that a tomato medium, which because of its reaction before sterilization (pH 7.2-7.4) had dropped to pH 6.0-6.2, would support a very good development of *L. acidophilus* colonies. Continued study indicated that most strains of X-type *L. acidophilus* will not develop well in a tomato medium with a reaction near the neutral point. When this same medium is acidified to pH 6.0-6.2, it becomes very satisfactory. It is possible that the chief function of added CO₂ in the incubation atmosphere is to lower the pH. This deduction is strengthened by the observation that a tomato medium of pH 6.0 reaction will support a very good growth of *L. acidophilus* without the addition of CO₂ to the incubation atmosphere. However, the development without added CO₂ is not quite as great as when this gas is used. The exact relationship of these two factors (CO₂ and reaction) in this particular case is under further investigation.

The modified medium, prepared as described, has

² *Journal of American Chem. Society*, 44, 1922, pp. 172-181.

³ SCIENCE, lxiv, 1926, pp. 304-306.

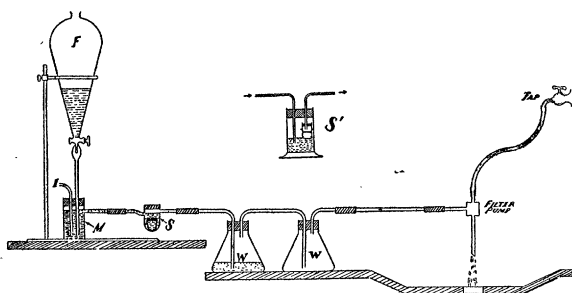
proved more satisfactory because *L. acidophilus* develops larger colonies than are produced in the original medium. Quantitative plate counts are also usually higher with this new medium. Extensive comparative tests have indicated that this medium is as good or better than any of the more complicated digest mediums previously advocated by the senior author and others for plating *L. acidophilus*.

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APPARATUS FOR VERY GRADUAL CHANGE OF FLUIDS

THIS apparatus, as set up in the text figure, is recommended in the treatment of tissues used for cytological studies. It also can be used for specimens which do not require a gradual change and can be run through the alcohols more rapidly. The tissues to be treated are placed in a perforated crucible and the crucible is stoppered with a finely perforated cork.



This crucible is placed in the specimen bottle (S). The replacing fluid in the Walter's special separatory funnel for single drops (F) is started dropping at the desired rate, the flow being regulated by the stopcock. The suction from the filter pump draws air from the inlet (I) through all the containers and

carries the excess fluid from the mixing bottle (M) and the specimen bottle (S) into the waste bottle (W). The air bubbling into the mixing bottle (M) will insure a quick and thorough mixing of the fluids in both the mixing bottle (M) and the specimen bottle (S).

For smaller and more delicate specimens the specimen bottle (S'), shown in the inserted diagram, may be used. This is constructed by cutting the bottom from a small homeopathic vial, tying a piece of bolting silk to the stopper-end of the vial to prevent loss of specimens in the outlet tube, and attaching this vial to the outlet tube by means of a cork.

For higher alcohols or clearing agents the air passing into the inlet (I) should be dried by passing it through a calcium chloride tube or through a bottle containing sulphuric acid or absolute alcohol. Very little water can be absorbed from the waste bottle (W) since the air currents will pass away from the specimen bottle (S) and toward the waste bottle (W). It is necessary to introduce a second waste bottle in order to prevent a back-flow of the water from the filter pump in case the water pressure from the tap is reduced.

Large specimens, such as termatodes, have been successfully transferred from 95 per cent. alcohol to water in three hours. The success is probably due to the constant mixing of the fluids before coming into contact with the specimens instead of the usual abrupt change from one grade of alcohol to the other. This apparatus facilitates washing since it can be done in the same container in which the specimens can be stained and run up through the alcohols.

(This apparatus was constructed through the courtesy of the Empire Laboratory Supply Company of New York City.)

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SPECIAL ARTICLES

FERROUS IODIDE AS A SUBSTITUTE FOR VITAMIN A IN RATS¹

IN view of the observations of Chidester, Eaton and Thompson^{2, 3} that small doses of syrup of ferrous iodide can substitute for vitamin A in the cure of xerophthalmia and promotion of growth in rats on vitamin-A deficient diets, the author has reinvesti-

¹ Published with the approval of the Director, West Virginia Agricultural Experiment Station, as Scientific Paper No. 107.

² F. E. Chidester, A. G. Eaton and G. P. Thompson, *SCIENCE* 68, 1766, 432, 1928.

³ F. E. Chidester, A. G. Eaton and N. K. Speicher, *Proc. Soc. Exper. Biol. & Med.* 28, 187, 1930.

gated this subject, using the same dosage of ferrous iodide, and supplying irradiated ergosterol as a source of vitamin D throughout the experiment. Observations include the effect of ferrous iodide on (1) xerophthalmia, (2) terminal infections of the glands about the mouth, (3) age at which xerophthalmia appears, (4) age at which constant weight is reached, (5) age at death, (6) weight at death, and (7) food consumption.

The animals used were albino rats from a stock raised for generations on Sherman Diet 13, and were placed at 4 weeks of age on Sherman vitamin-A free