the grain or by mouth. Muscular dystrophy can be produced equally well when petroleum ether extracted No. 1 alfalfa hay replaces the No. 3 alfalfa hay.

The muscular dystrophy producing property of this diet is dependent upon the cod liver oil. This is in agreement with the report of Madsen, McCay and Maynard[5] that cod liver oil added to a synthetic diet increased the rate of development and early severity of the muscular dystrophy. The water-soluble factor associated with nutritional muscular dystrophy as indicated by Morgulis, Wilder and Eppstein[6] is not lacking in this diet.

In this laboratory feeding of alpha-tocopherol,[3] prepared from wheat germ oil[7] has cured muscular dystrophy of rabbits, confirming the findings of Mackenzie and McCollum. Recently 300 milligrams of synthetic alpha-tocopherol[8] were obtained. Six rabbits suffering from muscular dystrophy but still able to walk with difficulty were given doses of this synthetic material. Experience would indicate that each of these rabbits, if left untreated, would within 24 hours have been unable to stand or consume food.

The individual doses of the synthetic alpha-tocopherol in milligrams were as follows: 17, 18, 20, 26, 51 and 65. All animals were cured except the one receiving the 17 milligram dose. A seventh animal which received 30 milligrams also died, but it was in a much more advanced stage of collapse at the time of treatment.

It is apparent that 20 milligrams is near the lower limit as a single curative dose. An animal is considered as cured when it loses its stiffness and begins to gain in weight within 48 hours and continues to gain for 10 days or more. Although no attempt is made to establish the minimum curative dose with such limited amounts of material, it is definitely shown that synthetic alpha-tocopherol will cure muscular dystrophy in rabbits as produced experimentally under the conditions stated. The 5 cures become more significant when one considers that from several hundred dystrophic animals we have never observed spontaneous cure of an untreated animal.

STEWARD G. MORRIS
U. S. DEPARTMENT OF AGRICULTURE

[5] L. L. Madsen, C. M. McCay and L. A. Maynard, *Proc. Soc. Exp. Biol. and Med.*, 30: 1434, 1933.

[6] S. Morgulis, V. M. Wilder and S. H. Eppstein, *Jour. of Nutrition*, 16: 219, 1938.

[7] The wheat germ oil from which the tocopherol was prepared was kindly supplied by the Archer-Daniel-Midland Company, Minneapolis, Minn.

[8] The synthetic alpha-tocopherol was furnished by Merck and Company through the courtesy of Dr. J. M. Carlisle.

SCIENTIFIC APPARATUS AND LABORATORY METHODS

STUDENT APPARATUS FOR ANALYSIS OF RESPIRATORY GASES

THE gas burette to be described was devised especially for medical students for whose instruction analyses of expired air are required of just sufficient accuracy to illustrate the general principles of respiratory exchange and metabolism. The usual complicated apparatus (*e.g.*, the Haldane-Henderson) is, we believe, poorly adapted to this type of instruction, for several reasons: (1) too much time is required to acquire proficiency with the method; (2) due to the excessive cost per unit insufficient apparatus is available; (3) the size of the equipment makes storage a problem; and (4) the difficulty of cleaning the apparatus when it becomes fouled, as so frequently happens, imposes a severe strain on the time and patience of the instructor.

By the simple method, here reported, an inexperienced operator may determine the CO_2 and O_2 of expired air with an error not often exceeding 0.1 volume per cent., corresponding to an error of about 5 per cent. in the calculated value of the metabolic rate. The time required for an analysis (30 to 60 minutes) may seem excessive, but students can actually make more determinations, during the limited period available for the work in respiration, than they can with the ordinary methods. It is suggested, in view of the low cost of the equipment, that each unit consist of two burettes, mounted together for simultaneous determinations. In this way the average time of an analysis will compare favorably with that required by the Haldane-Henderson method.

APPARATUS

The burette[1] is of Pyrex glass, with replaceable stopcock of the type which requires little or no lubricant. The graduations, which extend from the cock to 10 cc by 0.05 cc intervals, should be calibrated to within 0.005 cc for total and partial volumes. The burette should be carefully cleaned to allow complete drainage of the reagents. The leveling bulb should contain approximately 35 cc, and it may be conveniently suspended by a cord wrapped once over a horizontal rod, and a large rubber stopper may be used as counterweight. A thermometer reading to 0.2° C. should be available to each group of students.

REAGENTS

(1) Solution of NaCl (U.S.P.) 23.5 per cent. by weight, having a specific gravity of 1.18, to which is

[1] A suitable burette may be obtained from the Scientific Apparatus Company, Bloomfield, N. J., or from the Fisher Scientific Company, Pittsburgh, Pa.

added 5 cc of 85 per cent. lactic acid per 1,000 grams of salt solution. Phenol red or other indicator may be added if desired. Approximately 150 cc of the acid-saline is required for each analysis. The solution may be dispensed from stock in 500 cc cylinders, for convenient filling of the apparatus.

(2) Solution of NaOH, 16.0 per cent. by weight, having a specific gravity and vapor pressure approximately the same as the salt solution, for absorption

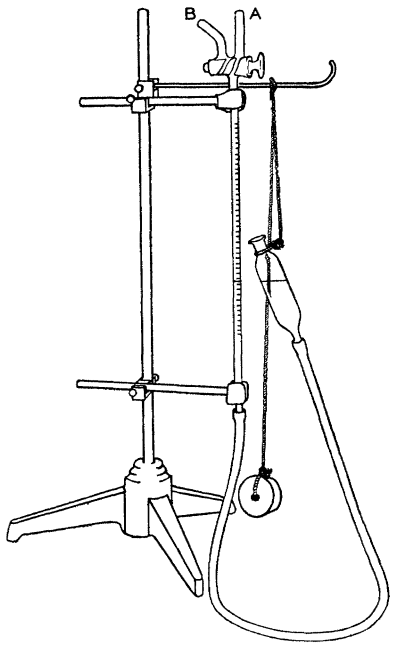

FIG. 1

of CO_2 and rinsing down of oxygen absorber. A single separatory funnel, with pipette-like tip, will allow the reagent to be conveniently accessible to the operators of 3 or 4 sets of apparatus.

(3) Oxygen absorber, consisting of 10 grams sodium hydrosulfite (Baker) and 1 gram sodium anthraquinone β sulfonate (Eastman) dissolved rapidly in 50 cc of 3N KOH, filtered through cotton and stored in a separatory funnel under paraffin oil. A single container should supply 6 to 10 students.

PROCEDURES

Levelling bulb, tubing and burette are filled with salt solution and about 1 cc is run into the reagent cup "A," a gas sample is then introduced through the tube "B" to approximately the 10 cc mark. Most of the saline is run in from the cup, the cock is closed, and the burette tapped to dislodge traces of solution from the upper end. Levels are adjusted and readings recorded at intervals until drainage is complete, which may require 5 to 15 minutes. The room temperature should be noted to the nearest 0.2° C. in vicinity of the burette. It is essential that the gas sample should be protected from sunlight or other causes of sudden variation of temperature. To this end the operator should remain at least 3 feet distant from the burette except during necessary manipulations, for heat radiated by the body may elevate the temperature of the gas without affecting the thermometer. A water jacket will not assist in maintaining constancy of temperature, for the heat of neutralization and of dilution which is liberated during the analysis must be completely dissipated, a process which occurs most quickly by air conduction.

CO_2 is now absorbed by running in 2 cc of the alkali, during about a minute. Time for complete drainage of the alkali rarely exceeds 8 minutes. Levels should be adjusted for the new reading and the temperature should be recorded. The volume of the gas should be corrected in each instance for temperature changes greater than 0.5° C.

O_2 is absorbed by running in 3 to 4 cc of the hydrosulfite reagent during approximately 2 minutes. To facilitate complete drainage and to avoid errors due to vapor pressure differences, the hydrosulfite should be washed down with 4 to 5 cc of the NaOH solution. Complete drainage may require from 10 to 15 minutes. The temperature, corresponding to the final volume, should be recorded.

After each analysis the fluid is run out by lowering the leveling bulb to a container placed on the floor. The burette is then washed through the reagent cup with 5 to 10 cc of NaOH, followed by water, followed by saline. The leveling bulb is then raised and refilled with saline. If sulfur scum appears in the burette it may be necessary to discard the first filling.

Since incomplete drainage is the chief source of error of the method, scrupulous attention should be paid to this factor.

During changeable weather it is advisable to record the barometric pressure and to make appropriate corrections of the gas volume.

J. MAX LITTLE
HERBERT S. WELLS

PHYSIOLOGY DEPARTMENT,
VANDERBILT UNIVERSITY SCHOOL
OF MEDICINE

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