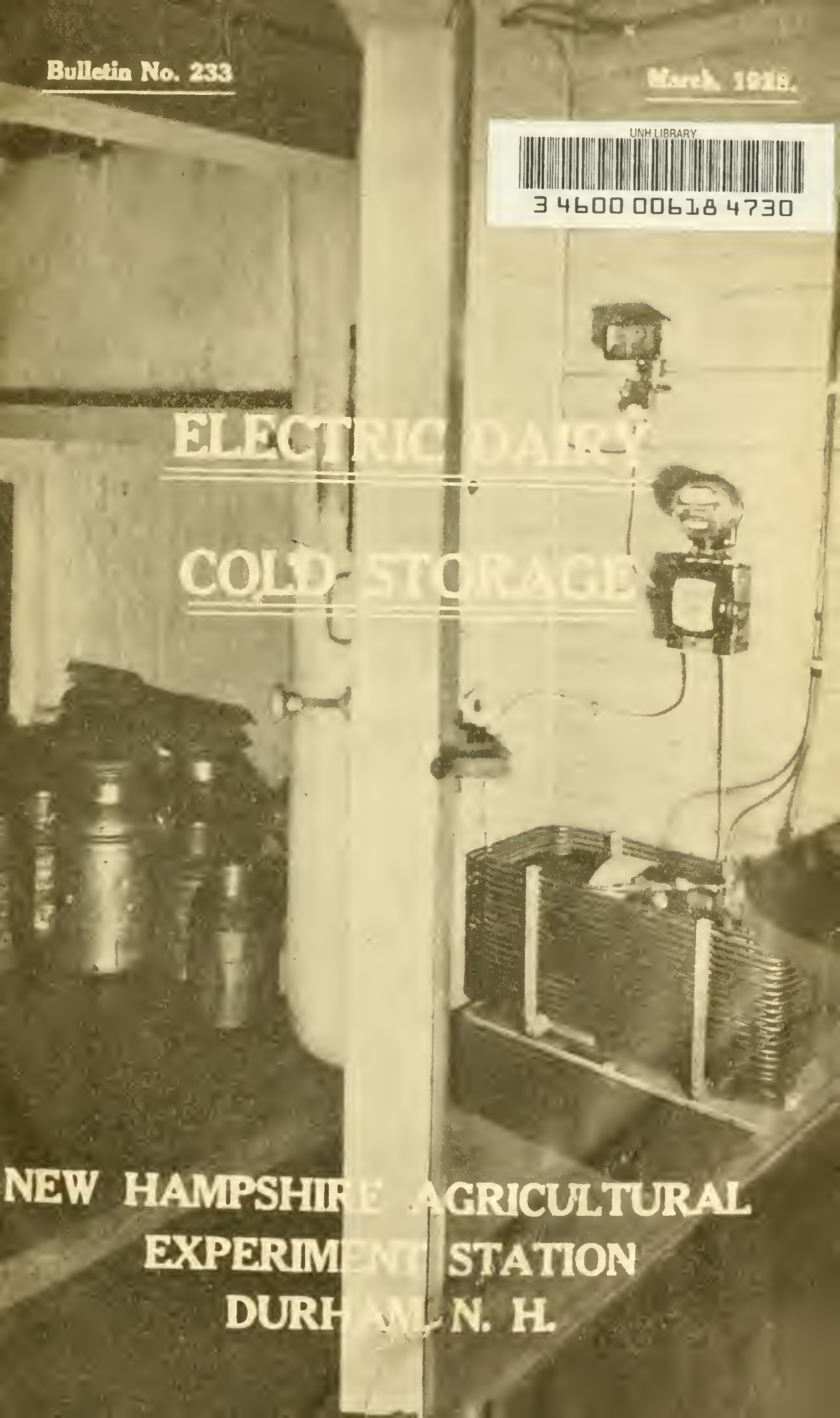
Bulletin No. 233

March, 1928.



ELECTRIC DAIRY COLD STORAGE

NEW HAMPSHIRE AGRICULTURAL EXPERIMENT STATION
DURHAM, N. H.

Bulletin 240 May, 1929

SIMPLIFIED TECHNIQUE AND APPARATUS FOR MEASURING ENERGY REQUIREMENTS OF CATTLE

By ERNEST G. RITZMAN *and*
FRANCIS G. BENEDICT

The Laboratory for Animal Nutrition Studies at Durham

AGRICULTURAL EXPERIMENT STATION
UNIVERSITY OF NEW HAMPSHIRE
DURHAM, N. H.

SIMPLIFIED TECHNIQUE AND APPARATUS FOR MEASURING ENERGY REQUIREMENTS OF CATTLE

By Ernest G. Ritzman, Research Professor in Animal Nutrition, New Hampshire Agricultural Experiment Station, and Francis G. Benedict, Director, Nutrition Laboratory, Carnegie Institution of Washington

INTRODUCTION

The development of the Laboratory for Animal Nutrition at the New Hampshire Agricultural Experiment Station came about as an essential part of an investigation on the physiological effects of under-nutrition on cattle. This work was begun in 1918 and has since been carried on co-operatively with the Nutrition Laboratory of the Carnegie Institution of Washington. The co-operation has been of a singularly satisfactory nature because of the distinct contribution each institution has been able to make to the development of the laboratory and thus to its function as a whole. The activities of this laboratory lie primarily in the field of energy metabolism. Its general aim has so far been twofold: (1) the investigation of the physiological requirements of farm live stock for net energy, and (2) the design of equipment and technique to simplify and improve methods for the study of energy transformations in farm live stock.

Since Lavoisier demonstrated the fundamental relationship between oxygen consumption, carbon-dioxide production, and heat production, further research has conclusively established the relative constancy of this phenomenon under the great variety of conditions in which heat is produced. The applicability of this knowledge in measuring in terms of a single unit the heat produced has revolutionized the potentialities of measuring the energy requirements of animal life, and of measuring the extent to which different feed stuffs may supply these requirements.

The complicated and time-consuming measurements of heat production by direct calorimetry are now no longer necessary, either with humans or with animals, but the energy transformations may be determined indirectly by measurement of the respiratory exchange. Numerous descriptions have been put on record stating the object of indirect calorimetry and explaining the principle on which it depends. A brief summary of the principles involved may, however, be given here.

Energy is the activating force which gives motion, heat, and thus active life to the otherwise inert body protoplasm. Energy is produced by oxidation, which in the human or animal body is in essence a decomposition of organic substances containing primarily the elements carbon, hydrogen, and oxygen in various combinations, but often including additional elements, such as nitrogen in the body cells. In this oxidation, which is either of body tissue or of food material, a definite amount of additional oxygen must be supplied by the outside air. This is delivered through the lungs to the blood stream and thus to the mass of body cells.

In the splitting up of the oxidized material that follows, a definite amount of heat is produced, and a definite amount of carbon dioxide is set free. This latter forms a waste product, which in turn is largely discharged through the lungs. The details of this process will not be explained here, but it is a fact that oxidation is a chemical reaction and is thus characterized by a degree of exactness which may be considered absolute. The rate of production of active energy accordingly has a definite dependence on the rate of lung ventilation, *i.e.*, on the amount of oxygen consumed and on the amount of carbon dioxide expelled. Measurements of oxygen intake and carbon-dioxide output, therefore, give an accurate picture of the rate of energy metabolism taking place within the body at any given time. Hence the terms "respiration chamber" and "respiration apparatus" have been accepted to designate the equipment used to measure this respiratory exchange and thus indirectly to measure the energy transformed.

By indirect calorimetry it is possible not only to measure the amount of material oxidized or burned in the body, but also to determine the character of the material so used. Energy-yielding material, as it is ultimately usable for oxidation, occurs in three forms, viz., carbohydrate, fat, and protein. These differ in the amount of oxygen they require for oxidation and also in the amount of resulting carbon dioxide, as is shown in Table I.

TABLE I—*Oxygen consumed, carbon dioxide and heat produced per gram of food substance as oxidized in the body, and the respiratory quotient, or ratio of carbon dioxide to oxygen* [1]

	Oxygen consumed	Carbon dioxide produced	Heat liberated	Respiratory quotient
	c.c.	*c.c.*	*Cal.*	*Ratio*
Carbohydrate	829.3	829.3	4.20	1.000
Animal fat	2013.2	1431.1	9.50	.711
Protein	956.9	773.8	4.40	.809

[1] Biological tables have been compiled on the basis of these values, whereby the energy metabolized by the animal can be readily computed from the amount of gaseous exchange and from the volume ratio of the gases. See Carpenter, T. M., Carnegie Inst. Wash. Pub. No. 303A, 1924, Tables 13 and 14, pp. 104 and 105.

The determination of the energy metabolism of non-ruminant animals, particularly humans, by methods based on measurement of the exchange of these two gases, oxygen and carbon dioxide, has now attained universal acceptance, and the technique has been highly standardized—so much so that other methods would now be considered impractical, inefficient, and unscientific.

Unfortunately the practical application of indirect calorimetry in the measurement of the energy values of feeds and the energy requirements of cattle has not received an equally unqualified acceptance. The progress made in studying energy transformations has been more rapid with humans than with animals, due in part to the differences in experimental objective and in part to the fact that the use of complex apparatus heretofore necessary for this purpose has been deemed justified in the case of humans but has not been considered justified in the case of large farm animals.

In metabolism studies with humans the needs of the individual and the efficiency of the diet to meet these needs are important primarily as they contribute to health and normality and only secondarily because of their economic significance. For this reason the research on the physiological phases of human energy metabolism has been directed towards supplying essential information on the fundamental relationship between the food of man and his physiological responses to it. In metabolism studies with cattle the urge to determine the utility value of different food stuffs to the animal organism as an economic expediency has kept the more fundamental, physiological investigations in the background. As a result, perfection of the technique necessary in measuring the energy balance of farm live stock has been delayed. Even in the classic researches of Armsby the chief objective, as was pointed out by Magee,[1] was the establishment of net energy values of different food stuffs rather than the determination of the physiological results of their ingestion. Although a great step toward achievement of this latter end was made by Armsby in demonstrating the logic of net energy values obtained by means of short balance experiments in direct or indirect calorimetry, his results have obtained theoretical approval without having so far achieved a corresponding measure of practical application, certainly in so far as this pertains to any general public use in their application of feeding standards to cattle.

There have been two predominant reasons for delay in the more general adoption of the net energy principle for guidance in feeding cattle. One of these, which is of a purely physiological nature, has been the immense food storage capacity of the ruminant, particularly the bovine. This has complicated a clear determination of that fraction of energy metabolism representing the basal or maintenance requirements incident to any given plane of activity or nutrition. As long as this essential element of the net energy theory remained problematical, the application of the principle was of course bound to be of questionable value. This deterrent is, however, being largely eliminated by recent researches in physiology. The other impediment has been the lack of apparatus with which energy transformation in cattle could be determined more rapidly and economically, and a technique of operation which could readily be attained by average experiment station staffs. This lack of suitable apparatus has probably been the most potent hindrance to the development of this modern and efficient method of studying the manifold problems of energy requirements and uses by cattle.

Practically all calorimeters and respiration apparatus which have been built heretofore were designed primarily to obtain specific information contributory to the researches of individual investigators rather than to standardize this more scientific method for general adoption. As a consequence most of these apparatus have been complicated, both in design and in operation, and for the most part have also been too costly for general use. In the design of the original respiration apparatus in use at the Laboratory for Animal Nutrition at Durham, this complexity and costliness of apparatus were recognized as the chief deterrents to a more rapid displacement of the early methods employed with cattle (based on the cumulative effect of food on live weight and particularly on live weight alone as the criterion of values) by the more scientific method of measuring the actual progress of energy conversion. As an outcome of the extensive experience of the Nutrition Laboratory at Boston in designing simple

[1] H. E. Magee, *Journ. Agric. Sci.*, Vol. XIV, Part IV, 1924.

forms of apparatus which have now become standard equipment for the study of problems in human nutrition, an attempt to achieve a similar objective in nutrition studies with live stock appeared to merit concentrated effort. It is the purpose of this bulletin to report on the progress made towards the achievement of this objective at the Laboratory for Animal Nutrition at Durham.

THE LABORATORY BUILDING

The laboratory building is a single story, wooden structure with a basement. The development to its present stage has been a gradual process extending over a period of nine years. Originally it contained only the center portion of the present building. This was composed (as shown in Fig. 1) of a room fitted with two metabolism stalls for steers, a room containing the respiration chamber and a platform scale for weighing the animals, and a room containing the necessary electrical and physical accessories. When the Purnell Fund became available to permit extension of this work to dairy cows, a wing providing a feed storage room and space for two metabolism stalls for dairy cows was added. This was followed two years later by the addition of a basement wing to provide necessary laboratory facilities to handle the samples of feed and excreta incident to digestion balances, and to provide space for several small respiration chambers which are being used for metabolism work with sheep. This basement also provides a small laboratory room for experimental study of human dietaries.

The last addition to the building, made during 1928, provides more floor space by the construction of a second floor on top of the basement wing just mentioned. This furnishes a room to be fitted up for a milk laboratory, and two small offices with desks and files.

The building is heated by steam, the radiators having sufficient radiation surface to keep the various rooms comfortable even during the coldest days of winter. It is thus not difficult to maintain a relatively uniform temperature, although experimentally constant conditions cannot be maintained during great changes in outdoor temperature.

The arrangement of the apparatus, stalls and other appliances is shown in the floor plans, Figs. 1 and 2.

Objectives in Laboratory Facilities and Technique and Requirements in Apparatus

The technical objectives of this laboratory have been to develop forms of apparatus and of technique which combine speed and economy with precision, and thus make available more economical and more expeditious processes with which to determine energy balances of farm live stock under varied conditions of nutrition. The apparatus and the technique of procedure are, therefore, largely such as are based on principles and methods of physics and of physical chemistry rather than of pure chemistry.

Chemical determinations are involved only to a minor extent, that is, to carry out nitrogen balances. Separate analyses of the nitrogen in urine and in feces are essential, first, for the determination of the digestibility of the various food constituents; second, to differentiate the character of the

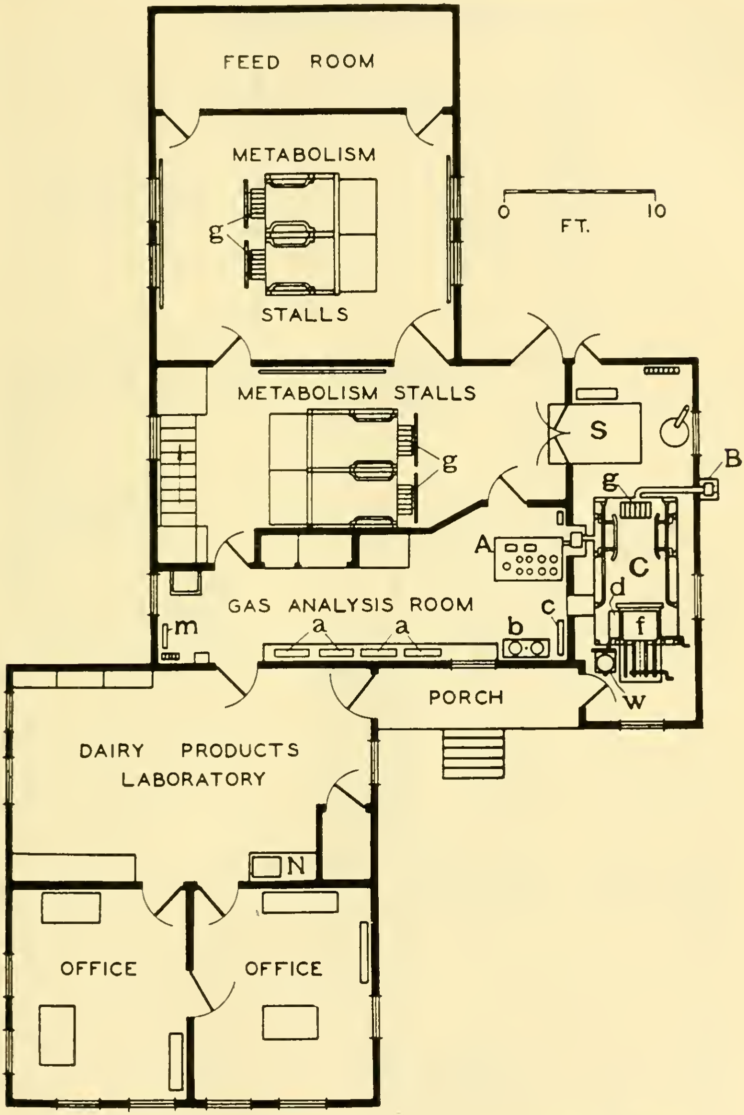


FIGURE 1—*Plan of main floor of Laboratory for Animal Nutrition*

C, respiration chamber for steers and cows, with feed box, f, water trough, d, small water tank set on scales outside the chamber wall to weigh the daily amount given, W; Blower, B, conducts fresh, outdoor air into respiration chamber; A, absorber table for aliquoting of air from chamber and for removal of carbon dioxide from air; S, scales for weighing animals; a, a, Carpenter gas-analysis apparatus; b, large balance to weigh carbon dioxide absorbers; c, control board with switches to operate electric motors, lights in chamber; m, control board with voltmeter, ammeter, and switch to operate motor-generator for direct current electric supply; N, Sink, cupboards for dairy utensils, etc.; g, g, g, feces grids.

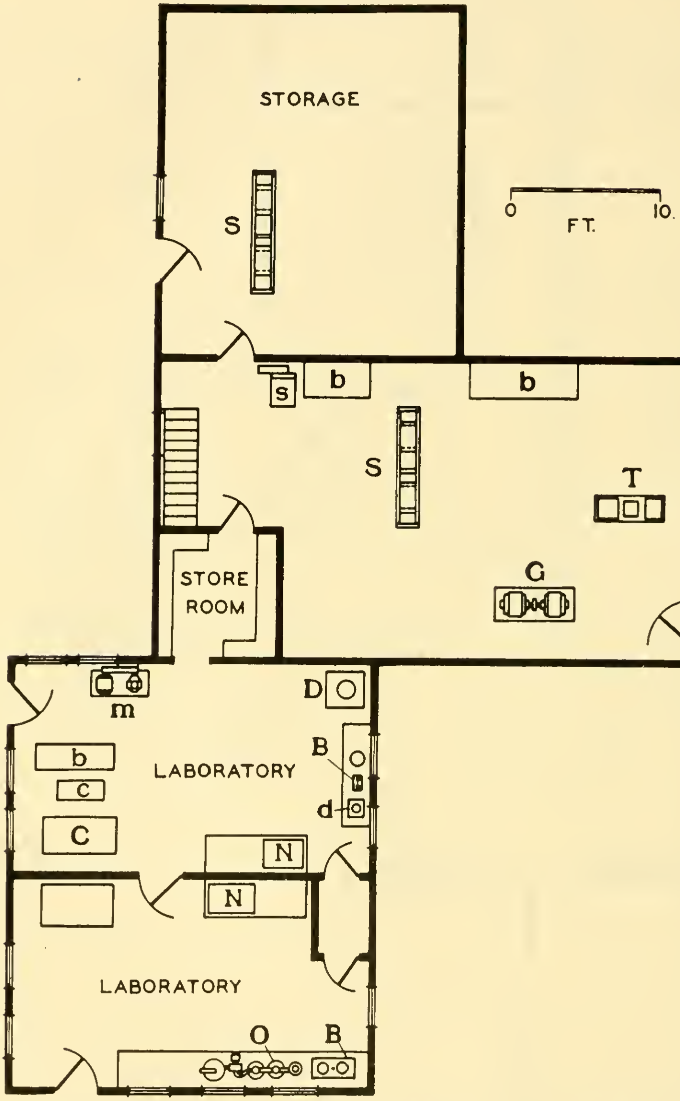


FIGURE 2—*Plan of basement floor of Laboratory for Animal Nutrition*

O, oxy-calorimeter; b, table with apparatus to ventilate respiration chambers and to collect aliquot air samples from the outgoing air current; C, respiration chamber for adult sheep and (c) same for lambs; s, small scale weighing excreta; S, apparatus for separate collection of feces and urine under metabolism stalls; T, same as S (slightly modified) for use under respiration chamber; G, motor-generator set to supply direct current electricity from alternating current; b, tables; m, Wiley mill with motor for grinding feed samples; D, large oven for drying samples of feed and excreta preparatory to grinding and preservation; d, small Freas oven to determine moisture content of feed and excreta; B, balances, N, sinks.

material that is being metabolized, and third, to obtain the heat equivalent of nitrogen. To avoid duplication of chemical laboratory equipment, the determinations of nitrogen have in the past been taken care of by the Nutrition Laboratory at Boston. These analyses will in the future be handled in the chemical laboratory of the New Hampshire Agricultural Experiment Station. The work required of the Laboratory for Animal Nutrition in this connection, therefore, is only a matter of collecting and properly preserving representative samples of the feed and excreta and the determination of their moisture content.

The essential requirements in apparatus and their use may be grouped as follows:

(1) A respiration chamber and apparatus to determine the daily energy exchange of the animal.

(2) Metabolism stalls to permit of an exact measurement of the food and water intake and the visible waste products excreted by an animal.

(3) An oxy-calorimeter [1] to determine the heat of combustion of food and excreta.

(4) Drying ovens, balances, etc., to determine the percentage of dry material in food and in excreted products.

(5) Dairy laboratory facilities to determine the amount and character of the solids in milk, in order to arrive at their heat equivalent.

(6) No modern laboratory should be without metabolism stalls, sensitive bullock scales, and dependable smaller scales for weighing feed, water and excreta in order to render determination of insensible perspiration as a daily routine.

The Respiration Apparatus for Cattle

The respiration chamber for cattle, as originally installed at the New Hampshire Agricultural Experiment Station, was designed primarily with a view to simplification of construction and technique of operation. In fact, its design was based on the principle of a large respiration chamber which had been built and successfully used by the Nutrition Laboratory at Boston for the study of problems in human nutrition.[2]

Since the specific object of this method of studying the energy conversion by animals is to measure accurately the oxygen absorbed and the carbon dioxide given off, the apparatus necessarily comprises three distinct units. The first consists of a chamber of sufficient size to accommodate the animal and which can be air-sealed against air entering into or out of it except by specially provided openings where both ingoing and outgoing air can be controlled. (See Plate 1 and Fig. 3.) The second unit consists of apparatus for ventilating the chamber, for measuring the amount of the ventilating current, for collecting the carbon dioxide and the water of the outgoing air current, or (as in our latest device) for collecting an aliquot sample of the outgoing air representative of the period over which the experiment lasts. (See Plates 2 and 5.) The third unit consists of apparatus for an exact analysis of the percentage of carbon dioxide and of oxygen in an air sample. (See Plates 3 and 4.)

[1] Benedict, F. G., and E. L. Fox, *Indus. and Eng. Chem.*, 1925, *17*, p. 912; *ibid.*, *Journ. Biol. Chem.*, 1925, *66*, p. 783; Benedict, F. G., and A. G. Farr, New Hampshire Agric. Expt. Sta., Bulletin No. 242, 1929.

[2] Benedict, F. G., W. R. Miles, P. Roth and H. M. Smith, Carnegie Inst. Wash. Pub. No. 280, 1919, pp. 92 et seq.

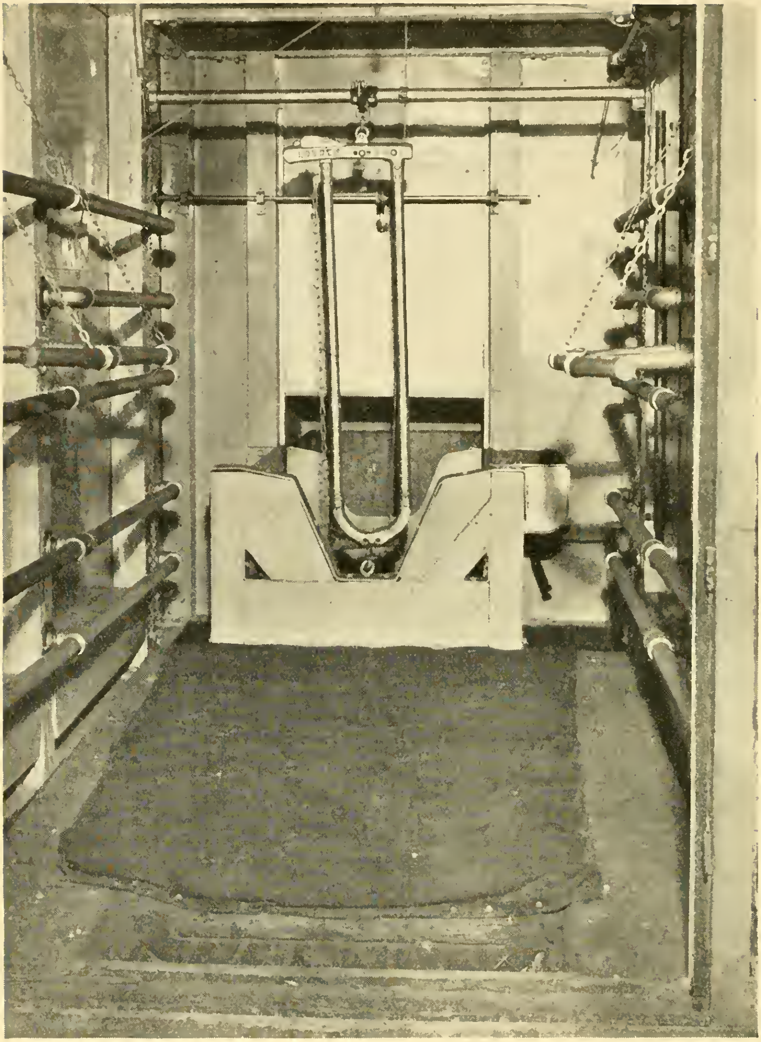


PLATE 1—*Interior of respiration chamber for steers and cows*

The shutter between the chamber and the small compartment attached to the front of it is raised, and the feed box has been moved into the latter. The water trough is attached to the right of the frame enclosing the feed box. The iron pipes protecting the side wall of the chamber are made from stock material used by plumbers. The chain attached to the left of the stanchion is used to force the animal to remain in a standing position. The free end of the chain is attached to a wire cable which can be rolled up by a pulley (manipulated outside the chamber wall), thus raising the loop formed at the bottom until it reaches the throat of the animal. The stanchion is of the adjustable form so that animals of different body length will be forced to stand with their hind feet near the grid, which is seen in the foreground. The tube attached vertically to the post on the right is the device used to signal when the animal lies down or rises. A weight sliding up and down within this tube is connected by means of a heavy cord to a belt around the chest of the animal. As the weight thus slides up or down it connects momentary contact and thus forms a circuit for a small dry cell battery which rings a bell. The two chains, one on each side, attached to the front of the floor platform operate the pneumograph which records the activity of the animal.

Since the respiration chamber in use at Durham was first described,[1] it has been materially improved by the addition of facilities for feeding, watering, and for the separate collection of feces and urine during an experiment. A simple device has been added to indicate when the animal

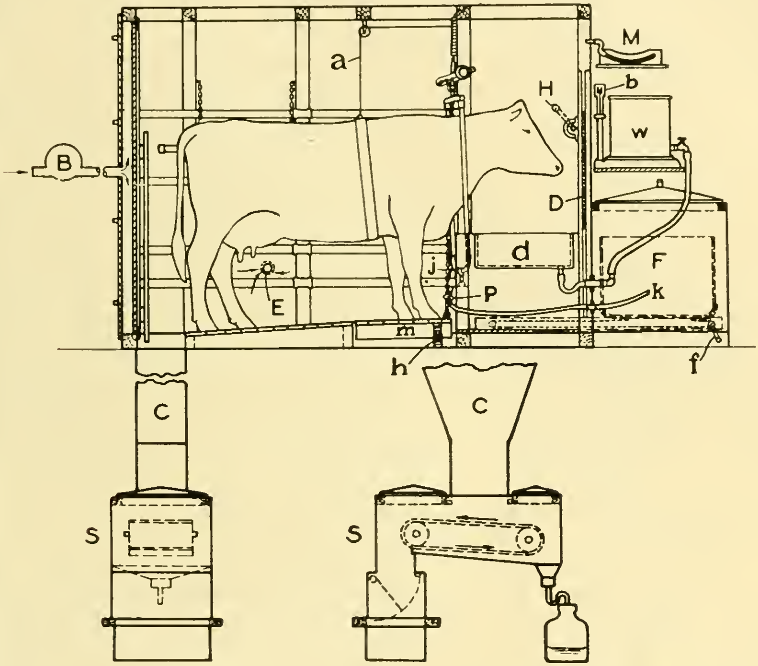


FIGURE 3—*Schematic outline of respiration chamber for cows and steers, and device for separate collection of urine and feces*

B, blower conducting fresh, outdoor air into space between inner and outer doors of chamber; a, line and pulley arrangement for recording when animal changes body position; e, position of blower, on the outside of chamber wall, which removes air from the chamber (see also B, Fig. 5); m, movable platform supported by spring, h, and chain, j, and attached to pneumograph, p, which communicates changes in air pressure due to movement of animal through tubing, k, to a tambour pointer and kymograph drum outside chamber; d, water trough connecting with outside water tank, W; b, balance for weighing of water; F, feed box; D, movable or sliding shutter for use in sealing off chamber from outside feed chute; H, hand crank controlling movement of D and F; M, manometer to indicate air pressure in respiration chamber; f, crank handle for moving feed box; S, S, air sealed sheet metal container (housing device) for separate collection of feces and urine from cows; C, chute leading down from grid. For further details of frame construction and details of apparatus for ventilating the chamber, for measuring the amount of ventilation, and for absorption of carbon dioxide and water, see Bull. 16 of this Station.

changes position from standing to lying, or vice versa, so that separate determinations can be made of energy expended for these body positions. A few minor improvements have also been made in the interior fitting and finish of stall accommodations to make the conditions for the animal

[1] Benedict, F. G., W. E. Collins, Mary F. Hendry and Alice Johnson. N. H. Sta. Tech. Bull. No. 16, 1920; Benedict, F. G. and E. G. Ritzman, Carnegie Inst. Wash. Pub. Nos. 324 (1923) and 377 (1927).

when in the respiration chamber as nearly as possible the same as those which prevail in the metabolism stalls. No change has, however, been made affecting the principle of ventilation or its theory. The chamber was originally designed for the purpose of carrying out experiments of two to four hours' duration, without regard to measurement of the visible excreta; but with the above changes an experiment may now be extended to any desirable length of time.

The respiration chamber consists of a rugged framework of 4″ x 4″ spruce enclosed by a sheet metal shell open at the rear end for entry of the animal, this opening being closed by a double set of doors when the chamber is in use. The details of this frame and door construction have been described in Bulletin 16 of this Station. A four-inch space provided between the double set of doors serves as a reservoir for fresh outdoor air supplied by a blower outside of the building. The inside door is pressed against a felt surface which is somewhat porous; hence when air is removed from the chamber to supply the absorbing apparatus there will be a proportional seepage into the chamber from this fresh air reservoir, as the contact between door and felt is the only part of the chamber that is not absolutely sealed.

A framework of metal steam pipes (Plate 1), allows free circulation of the air in the chamber. A steel swivel stanchion gives the animal a reasonable amount of freedom of movement, and eliminates any irksome restraint that causes nervousness. The floor is heavily padded and thus restful in the standing position and favors longer lying periods. Feed box and water are conveniently placed so that during experiments of from one to five days' duration, as now conducted, the conditions of existence in the chamber are comparable to those maintained in a modern dairy barn, except for periodical exercise and daylight. The lack of exercise is partly compensated by the floor padding, which relieves muscular strain. Perhaps it may be advisable to install a substitute for sunlight to approximate greater daily normalcy for long experiments. At present the chamber is lighted by electricity, controlled from the outside.

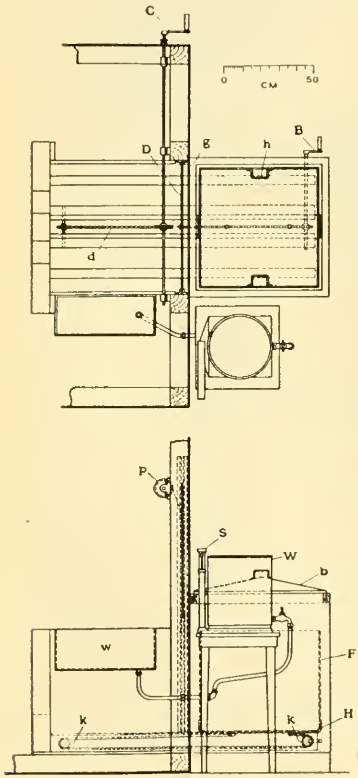


FIGURE 4—*Details of mechanism for feeding and watering animal in respiration chamber*

Above—Floor plan. Below—side elevation. C, crank to raise shutter D; g, groove for water seal for removable cover b; h, handles to feed box; B, crank to move chain d to shift feed box. Water supply tanks W and w; p, spool on shaft operated by crank; S, scales. K, sprockets on chain H which moves feed box, F.

Appliance for Feeding and Watering

In order to feed the animal during a respiration experiment it was necessary to provide facilities to introduce a feed and to remove any feed refused from a previous meal under conditions in which an accurate account could be made for any changes in the composition of the chamber air caused by this operation. This provision was obtained by a movable feed box (Plate 1 and Figs. 3 and 4) which can be slid out of the chamber proper into a small, sheet-metal compartment soldered to the outside front of the chamber wall. By means of a heavy, sheet-iron shutter which can be raised and lowered from the outside, the opening connecting the chamber with the small outside compartment may be closed practically air-tight. The top of the outside compartment is provided with a lid, which sets in a water seal. Hence at feeding time the shutter is raised, the feed box is moved to the outside compartment, and the shutter is lowered again, closing off the respiration chamber. The lid is then removed from the water seal and any left-over feed is taken out and weighed. A fresh feed is then placed in the feed box, the lid is replaced in the water seal, and the feed box is restored to the chamber by the same operation.

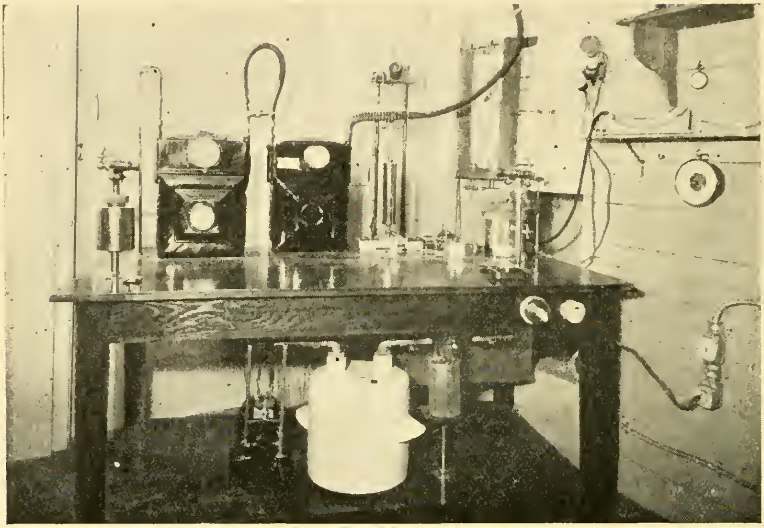
The operation of moving the feed box from one compartment to the other is accomplished by means of a hand crank on the outside of the apparatus, as is also the raising and lowering of the shutter by which the two compartments may be closed off from each other.

The feed box rides on four pieces of 2″ x 4″ material placed edgewise and spaced five inches apart, and extending the entire distance to which the feed box must move. Between the two middle pieces are two small bicycle sprockets, one at each end, carrying a continuous chain. The sprocket in the smaller outside compartment is mounted on a long shaft which projects through a machined bearing to the outside where it is fitted with a crank to move the chain backward, or forward. To facilitate removal of the feed box for cleaning, the chain is not attached directly to it but to a piece of sheet-iron, 5 inches wide and one-eighth inch in thickness, which is bent up at the ends so that the feed box just fits into it. This sheet-iron saddle is of just sufficient width to fit between the two middle pieces of 2″ x 4″ on which the feed box rides. This plate is supported by two cleats which are nailed in to the 2″ x 4″ low enough to leave the top surface of the plate flush with the top surface of the 2″ x 4″.

The water tank is hung on the outside of the wooden framework (Plate 1) which encloses the feed box inside the respiration chamber. The water is introduced through a tube which enters the tank in the bottom so that any water refused can be drawn off for reweighing. The tube is so adjusted as to maintain a constant water seal to prevent exchange of air.

Ventilating Apparatus

The original apparatus for ventilating the chamber, measuring the volume of ventilation, and absorbing the carbon dioxide for weighing is still in use. (See Plate 2, Fig. 5.) It consists of a small electric blower located just outside the wall of the chamber, which removes the air from the chamber (See Fig. 5) and drives it into a large metal can, or wind-chest, hung under the absorber table. Another electric blower, located on the lower shelf of the table, removes an aliquot fraction of this air from the



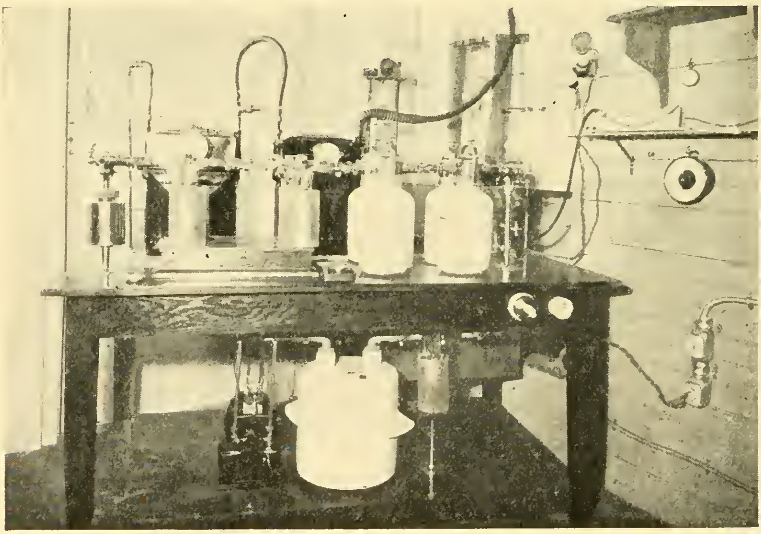


PLATE 2—Shows (below) table with absorbers to collect carbon dioxide and water from ventilating air current and meters for measuring the volume of ventilation. The long glass aspirator tubes hung in a frame attached to the wall behind the right end of the table are used to collect an aliquot sample of carbon dioxide free air for determination of marsh gas. The manometers on the right above barometer indicate air pressure in chamber and in the air sampling can. Above is same table with absorbers removed showing pump operated by electro-magnetic device to collect an aliquot air sample from ventilating current.

windchest and forces it in turn through the driers, absorbers for CO_2, and meters, all of which are connected in series. The air not passed through the absorbers leaves the windchest through a free opening on top and is carried off through a room ventilating hood above the table. The object of using only a fractional part of the total ventilating current is to reduce the size of the absorbers so that they may be conveniently handled

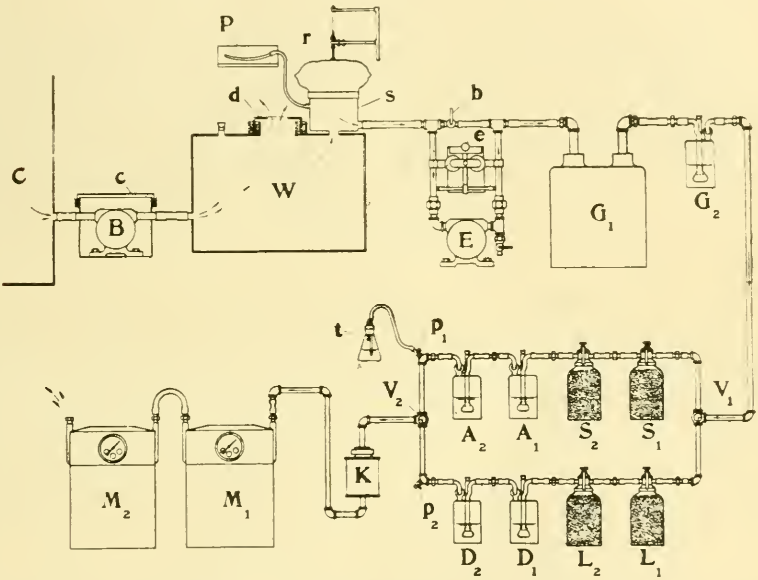


FIGURE 5—*Schematic outline of apparatus for ventilating respiration chamber and for analysis of aliquot sample of air coming from chamber*

C, respiration chamber; B, blower for removal of air from chamber; c, cover in water seal of blower container; W, windchest; d, opening for free discharge of air into laboratory room; s, can with rubber cap expansion top attached to circuit breaker r, which actuates magnetic bypass e thus automatically maintaining control of aliquot air sample from windchest W; b, hand-controlled bypass to aliquot air sample; P, manometer to indicate the air pressure in sampling can; G_1, sulphuric acid to remove moisture from air stream; E, airpump; G_2 serves as check to G_1 to indicate approach of saturation; V_1 and V_2, valves to deflect air stream from one set of carbon dioxide absorbers (S_1 and S_2) to the other (L_1 and L_2); A_1 and A_2, sulphuric acid to retain moisture absorbed from soda lime in S_1 and S_2; D_1 and D_2, sulphuric acid to retain moisture absorbed from L_1 and L_2; P_1 and P_2 petcocks to tap air stream for sampling; t, barium hydroxide to test air stream for loss of carbon dioxide; K, sodium bicarbonate to absorb fumes; M_1 and M_2, meters.

and, secondarily, to economize on absorbent materials and on labor. The size of the aliquot relative to total ventilation may be varied from 4 to 50 per cent by altering the size of the free opening for waste. The principle of the aliquoting mechanism has been repeatedly described.[1]

[1] Benedict, F. G., W. E. Collins, M. F. Hendry, and A. Johnson, New Hampshire Agric. Expt. Station Technical Bulletin No. 16, 1920; Benedict, F. G., and E. G. Ritzman, Carnegie Inst. Wash. Pub. No. 324, 1923, pp. 61 et seq.; *ibid.*, Carnegie Inst. Wash. Pub. No. 377, 1927, pp. 29 et seq.

Pump for Aliquot Air Sample

In addition to this system of collecting the carbon dioxide for weighing, another and still simpler system for determining the oxygen deficit and the carbon dioxide increase of chamber air is now being tried out on the large respiration apparatus simultaneously with the absorber method. (See Plates 2 and 5.) This consists simply of an air-tight pump which collects an aliquot air sample for the period of the experiment. The pump is actuated by an electro-magnet, which is timed by one of the meters that record the amount of ventilation. The use of this pump eliminates all the absorbers for carbon dioxide and water vapor and thus eliminates much work in weighing. It involves of course several additional gas analyses, but the amount of time and expense saved by this procedure promises a great advance in method. If meters of sufficient size to take the whole ventilating current are used, then the aliquoting of the air current may also be dispensed with on the large chamber for cattle. On the small chambers for sheep this method of measuring the total air is now followed entirely.

Operation of the Respiration Apparatus

The technical object of a respiration experiment is, as has been stated, to measure the amount of oxygen the animal absorbs and the amount of carbon dioxide it eliminates during a given time, generally during 24 hours. The period may, however, be shorter, provided it permits calculating the true 24-hour equivalent. The measurement of the amount of oxygen consumed and of carbon dioxide excreted by the animal during the period of the experiment thus involves determination of the following conditions:

1. Composition of chamber air at start.
2. Volume of outdoor air entering chamber.
3. Composition of outdoor air.[1]
4. Volume of air withdrawn from the chamber.
5. Composition of chamber air at end.
6. Composition of aliquot air sample.[2]
7. Chamber temperature, wet and dry bulb.
8. Barometer pressure.

Ventilation

The volume of ventilation is measured by meters which record the amount in liters, the air removed from the chamber being passed through two meters set in tandem for this purpose. The object of using duplicate meters is to serve as a check, both on the accuracy of meters and of reading the meters. The volume of in-going air must be proportional to the amount withdrawn, as withdrawal would otherwise create a vacuum which is impossible with the conditions supplied by the inside door. (See page 12.)

Since the animal consumes oxygen and excretes carbon dioxide, the air in the chamber is being altered as soon as the doors are closed. The composition of outdoor air is constant, that is, it contains 20 940 per cent of oxygen and 0.030 per cent of carbon dioxide. If the ventilating motors

[1] As the percentage of oxygen and of carbon dioxide in outdoor air is constant these determinations are made only to test efficiency of gas analysis apparatus itself.

[2] Necessary only when no absorbers are used.

are not set in motion, the oxygen content in the chamber air will decrease below that of outdoor air, and the carbon dioxide content will rise above it, so that in case of a full-fed animal the former may decrease to one per cent below and the latter may increase to one per cent above the outdoor conditions within half an hour. It will thus be observed that while the oxygen supply is not materially exhausted, the carbon dioxide content does increase above the outdoor or fresh condition in a relative sense. Contrary to common opinion, such increase in carbon dioxide approaching one per cent is not injurious or disturbing. It is thus an advantage to let the animal build up the carbon dioxide content of the chamber air to from 0.60 to 0.80 per cent before starting ventilation and then to maintain the rate of ventilation at a speed whereby the removal of air from the chamber maintains the percentage of carbon dioxide in the chamber at an even level. This minimizes the probable error in gas analysis, particularly in the calculation of the oxygen deficit.

Gas Analysis

The experiment is thus always begun when the animal has already materially altered the composition of the air which is residual in the chamber. When the experiment is ended there will remain in the chamber some carbon dioxide which the animal has contributed during the experiment but which has not been collected. This makes it necessary to analyze samples of the chamber air taken both at the moment of starting and of ending the experiment, in order to determine the amount of oxygen and of carbon dioxide which was in the chamber when the experiment was begun and the amount left over when it ended. These determinations enable one to strike a balance for the amount of oxygen consumed and carbon dioxide produced during the specific time interval of the experiment.

The great improvements which have recently been made in apparatus for gas analysis have made possible a much broader and more thorough study of metabolism and thus have advanced the potential significance of indirect calorimetry. With open-circuit apparatus gas analysis was formerly carried out on carbon dioxide in order to provide a proper balance for the amount absorbed. With the newer types, such as the Carpenter gas analysis apparatus,[1] with which both carbon dioxide and oxygen can be determined rapidly and with an extraordinary degree of accuracy (one part in 100,000), the respiratory quotients (that is, the ratio of the two gases) may now be obtained at any time. Thus, one may not only determine the amount of energy expended during a given period, but one may also follow the course of digestion and show the character of the material that is being oxidized, or burnt, in the body at any given time.

The apparatus used here (see Plates 3–4) are of the most recent type designed by Carpenter. Two of these, which are duplicates (Plate 4, right), allow both oxygen and carbon dioxide determinations. The raising and lowering of the mercury level is performed mechanically by an electric motor. The arrangement is such that one or both apparatus may be operated at the same time by one person. These apparatus are provided with large reservoirs for the absorbing fluids, and the manner of

[1] Carpenter, T. M., *Journ. Metab. Research*, 1923, *4*, p. 1; Carpenter, T. M., and E. L. Fox, *Journ. Biol. Chem.*, 1926, *70*, p. 115; Carpenter, T. M., *Journ. Biol. Chem.*, 1929, in press.

forcing the air through these absorbing fluids is such as to secure speed as well as thoroughness of absorption. The third apparatus (Plate 4, left) is designed primarily to determine the methane (or marsh gas) in the air sample. Its principle feature in addition to the burette and absorption fluid for CO_2 is a small, electrically heated, combustion pipette in which the methane of the air sample is broken down by high temperature, thus liberating the CO_2 it contains, the latter being then absorbed in the usual

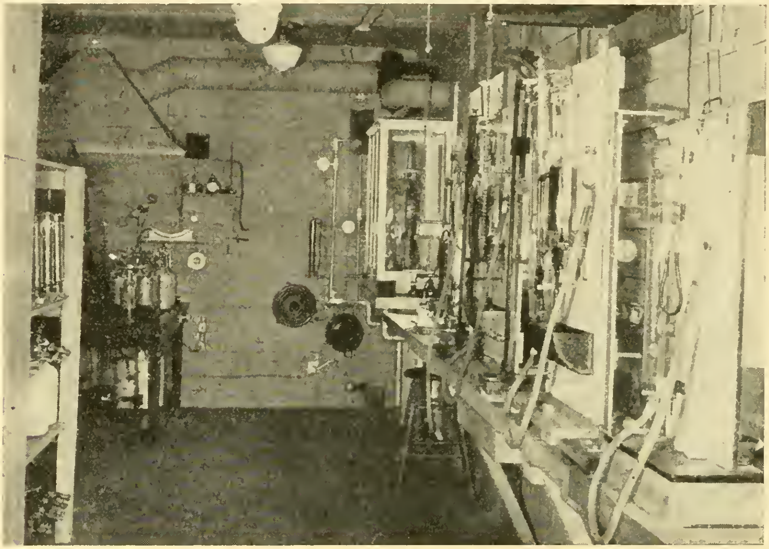


PLATE 3—*General view of gas analysis room*

way. All three of these apparatus have been thoroughly described by Carpenter.[1]

Duration and Technique of Respiration Experiment

The duration of a respiration experiment with the large chamber for cattle depends mainly on the purpose in view. In so far as the mechanical utility of the apparatus is concerned, an experiment may be carried out satisfactorily for any desirable period of time from one and a half hours up to a week. Since a daily, or 24-hour, period forms the logical time unit for expressing food requirements and metabolic activities, it follows that all respiration experiments of less than 24 hours' duration must finally be standardized with reference to their relationship to a whole day. At present all our respiration experiments with cattle are carried out on the 24-hour basis and are continued for two or three consecutive days in order to check the results.

An example of the data obtained in eight different experiments with each of two cows is shown in Table II. All conditions were identical during the full feed days. Thereafter the only difference occurred in food supply.

[1] *Loc. cit.*

The data on page 20 represent the total calories per 24 hours produced by two different cows. If computed on basis of surface area, or to an equal unit of live weight, the results would probably become even more uniform.

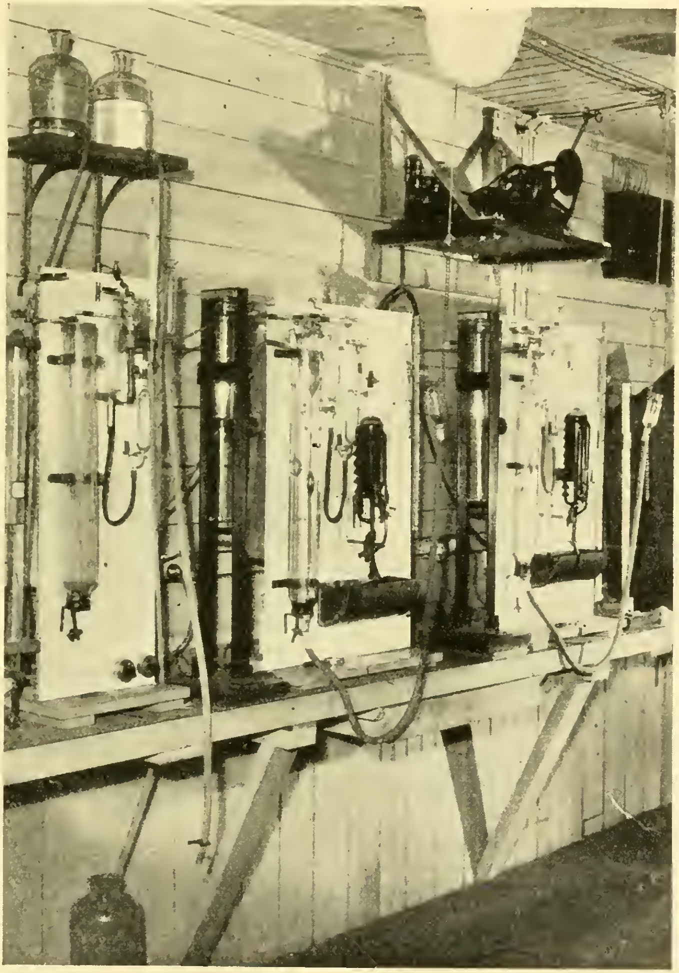


PLATE 4—*Apparatus for determination of percentage of carbon dioxide and oxygen in air*

The apparatus on left is for carbon dioxide only. This apparatus is provided with a combustion tube in which methane gas can be split up by electric heat. The two apparatus on right are duplicates which can be operated simultaneously by the motor-driven rotating arm above. These two apparatus are for determination of carbon dioxide and oxygen. Note the large pryo capacity (black) which is sufficient to analyze 75 samples without replacing absorbent.

The routine by which such results are obtained is as follows:

The animal is put into the respiration chamber and the doors are closed at 4 P.M. At 4.15 P.M. the blowers are set in motion for a preliminary

TABLE II—*Total calories of energy metabolized by Holstein cows*

Date	Cow	Full feed		Half-feed or fasting[1]	
		1st day	2nd day	2nd day	3rd day
		Cals.	*Cals.*	*Cals.*	*Cals.*
Dec. 2–7	H I	14,790	14,290	9,750[2]	9,630[2]
Dec. 30–Jan. 4		14,530	14,780	11,800[3]	11,350[3]
Jan. 20–25		14,670	15,180	11,100[3]	10,940[3]
Feb. 10–15		14,650	15,200	11,800[4]	11,510[4]
Dec. 16–21	H III	13,580	14,850	8,300[2]	8,440[2]
Jan. 6–11		13,590	14,390	10,660[3]	10,850[3]
Jan. 27–Feb. 1		13,740	14,450	11,250[3]	11,000[3]
Feb. 17–22		14,190	14,370	10,520[4]	10,520[4]

[1] No respiration experiments are made on first day after feed reduction. [2] Represents fasting.
[3] Represents half-feed. [4] Same as (3) less 500 gms. linseed oil meal.

ventilation of the chamber, to stabilize air conditions and rate of ventilation. At 4.30 P.M. the experiment is begun by deflecting the air current through freshly weighed absorbers for the collection of CO_2. At the time when this absorption begins, a sample of air is taken from the chamber to analyze its residual content of carbon dioxide and oxygen. Absorption is continued through one set of absorbers for eight hours, at the end of which another residual sample is taken and the current is deflected through freshly weighed absorbers. This process is continued for 48 hours, during which the animal is fed and watered at regular intervals. Additional observations on general deportment and appearance of the animal, or unusual incidents, are recorded in notes. A typical data sheet showing the nature of the records obtained during an 8-hour period of an experiment is reproduced on page 21. Each day is divided into three such periods. Separate data sheets are also used to record: (1) change of body position from standing to lying and vice versa, (2) temperature of chamber air every half hour, (3) various steps in gas analysis, and (4) computations of the results.

A graphic record showing the relative activity of the animal throughout the experiment is also obtained by a kymograph clock.

Respiration Chambers for Sheep

Plate 5 shows the respiration apparatus which were designed primarily for use with individual sheep and with which about 75 separate respiration experiments have been carried through up to the present time.

This apparatus (as stated on p. 16) is not provided with means to absorb the carbon dioxide and water, the carbon dioxide and oxygen being determined from the aliquot air sample collected by the pump shown on the table. The carbon dioxide and oxygen content of the sample are then determined with the Carpenter gas analysis apparatus. As this is on the volumetric basis it is, of course, necessary to determine the barometric pressure and also the temperature and degree of moisture in the ventilating current. The chamber for adult sheep consists of three parts: (1) a floor platform provided with groove around the outside which is filled with water to serve as a seal against exchange of air, (2) a crate built of a wooden framework enclosed with one-half-inch mesh wire to accommodate the animal, and (3) a sheet-metal shell (Plate 5), open at the bottom,

DATA SHEET FOR ABSORBER METHOD

Respiration Experiment

Subject.................. Weight.................. Date..................
Chamber closed..................................
Preliminary ventilation started..........................

Time		*Temperature Deg. C.* of Chamber Wet	of Chamber Dry	of Ventilating Current Dry	*Barometer*
Start..................M		....	....	....	....
End..................M		....	....	....	....
E. T..................	Avg.	....	....	....	....

Absorber Weight	*Meter Readings Liters* 1st	2nd
End..................gms.	....	....
Start.................. "	....	....
Diff.	....	....

Gas Analysis

	Residual Chamber Air CO_2 %	O_2 %	R. Q.	*Aliquot Air Sample from Ventilating Current* CO_2 %	O_2 %	R. Q.
Start	......	....				
End	....			....		

Body Position	Hours	Minutes
Standing	....	....
Lying	....	....

Activity Record	
Very Active	....
Moderately Active	....
Quiet	....

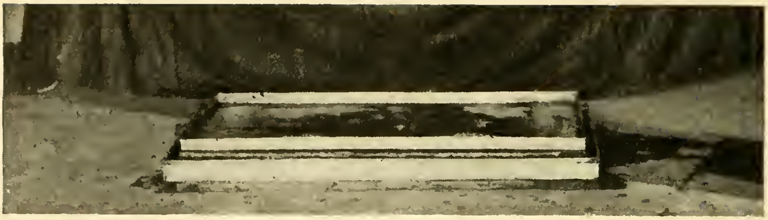
which is placed over this crate and inserted in the water seal of the floor platform.

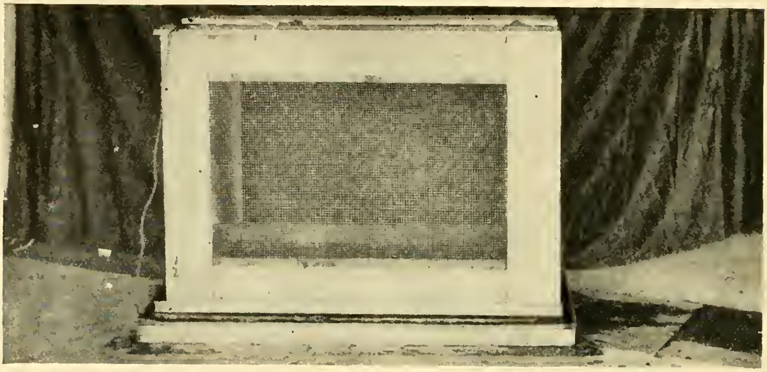
The chamber for small lambs is the same in principle. This consists of a large sheet-metal can, open at the top, and supplied with a cover on top which sets in a water seal. A correspondingly small crate to house the animal is placed inside the can. Both chambers have an air intake opening at the head end which consists of a male nipple to fit the coupling of ordinary garden hose, the air outlet at the opposite end being the same.

Metabolism Stalls

The laboratory contains four metabolism stalls which may be used either for steers or for cows. At present only cows are being used. (See Plate 6.) Two of these, used for dry cows, are located in one room. The other two, used for milking cows, are located in another room so that separate temperature conditions can be maintained, if desirable.

In designing these stalls it was deemed essential to provide conditions similar to those which prevail in a modern dairy in order to meet the just criticism often heard, that during such experimental observations cows





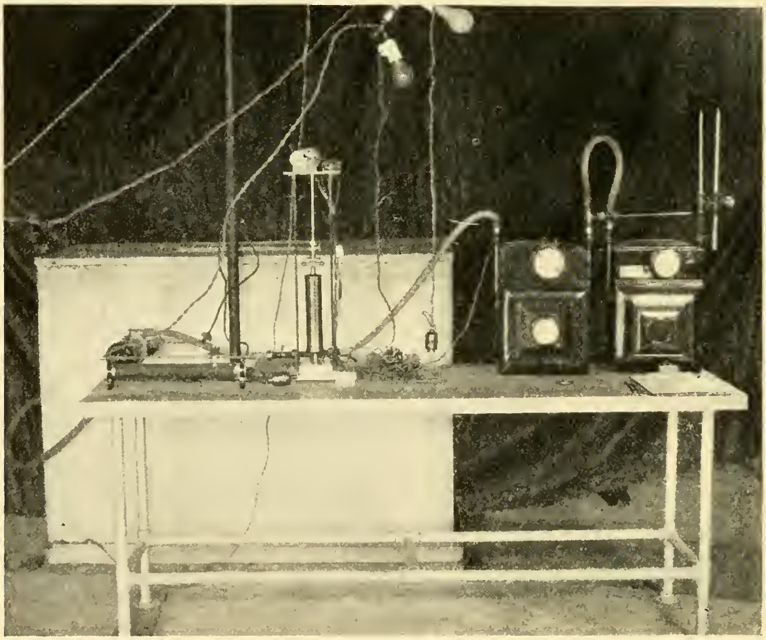


PLATE 5—*Respiration chamber for sheep*

The top view shows the floor platform with arrangement for water seal. The middle view shows the wooden crate with wire mesh sides, resting on the floor platform. The bottom view shows the respiration chamber complete, with metal shell placed over the wooden crate and fitted into the water seal of the base. On the table in front are the accessories (blower, aliquoting pump device, and meter) for ventilating the chamber, securing representative sample of chamber air for subsequent gas analysis, and metering the entire volume of air coming from the chamber.

are usually maintained under conditions quite different from those found in general dairy practise. The stalls are provided with a standard make of steel swivel stanchion which allows some freedom of movement. The floor is heavily padded to provide comfort, and the stalls themselves are entirely open, airy and cheerful.

The stalls, as will be noted, are provided at the rear with an opening in the floor through which excreta is deposited on an apparatus below designed especially for the separate collection of urine and feces with cows. (See Fig. 6 and Plate 7.) This opening is supplied with a grid, spaced so

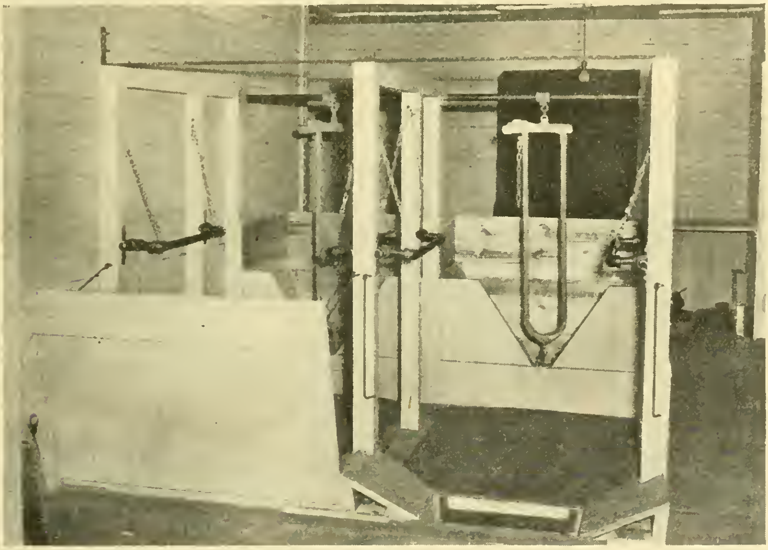


PLATE 6—*The metabolism stalls for cows*

These stalls may be adjusted for animals of different lengths by pushing the two feed boxes which are built as a unit backward or forward so as to keep the hind feet about 6 or 8 inches from the grid. The guards on the sides keep the animal centered over the grid while standing without discomfort, and thus allow a wide spreading of legs while lying down. When a cow is to be taken out the splashboard is removed (as on right stall) and a piece of plank is placed on the grid.

as to prevent the cow's rear feet from stepping down in but providing a sufficient aperture for the urine and feces to pass through readily.

The process of collecting excreta without significant loss represents the critical feature of a metabolism stall. These particular stalls were aimed to provide conditions which would compel the animal to deposit its excreta through as small an aperture in the floor as practicable, in order to prevent scattering which would tend to greater wastage. The dimensions of this opening are 12″ x 24″, *i.e.*, just one-half the width of the stall itself, which necessitates keeping the animal fairly well centered in the stall while standing. This is attained by a pair of automatic bumpers made of 1½″ iron pipe (see Plate 6) which project a sufficient distance into the stall to accomplish the purpose. They are hinged to the posts on the side of the stall so that they can freely rotate up and down within an arc of 90 degrees, a pair of small chains holding them suspended laterally, to which position they drop of their own weight. If the bumpers were of a fixed instead of hinged type, the animal would probably have great difficulty in rising from the lying position, since they would form a rigid obstruction. With the hinged type the animal, on rising, simply pushes one or both of them up without resistance and then they drop down again to the lateral position of their own weight.

An adjustable feed box and stanchion were provided by which the length of the stall could be varied to conform to different sized animals so that the hind feet are always about 6 inches from the floor opening or grid when the animal stands in a normal position. Wastage to the rear is prevented by a removable splashboard.

Excreta Collecting Apparatus

The special feature connected with these metabolism stalls is the apparatus for the separate collection of feces and urine from cows. The primary object in developing this apparatus has been to do away with all the usual harness used for such purpose, in order to relieve cows of encumbrances which tend to produce abnormal conditions. Incidentally the employment of this apparatus has resulted in an improvement in the sanitary conditions of the stalls and in a saving of labor. Furthermore, it enables the separate collection of urine and of feces continuously for the whole experimental year which is, of course, preferable to the short periods of two or three weeks customary when much paraphernalia must be attached to the animal for separate collections.

The apparatus for collecting urine and feces from cows in separate receptacles is located in the basement immediately under the grid of the metabolism stall. It consists of a framework (as shown in Plate 7 and in Fig. 6) made of wood which carries a large metal pan. Within this pan there is a belt 20″ wide, mounted on two wooden rollers, one 8″ in diameter and the other 6 inches in diameter. The roller on the end toward the receptacle which receives the feces is the larger in diameter and it is set somewhat higher so that the top surface of the belt on which the excreta drop is on an incline and moving uphill. The chute leading down from the stalls above it directs the excreta to the center of the belt, which latter moves very slowly, giving the urine time to drain off the sides and back into the pan, from which it drains into the carboy below, while the feces (which remain stationary on the belt) move upgrade and thus are carried over into the receptacle used for this purpose.

The principle of this apparatus is based on the fact that cows do not urinate and defecate at the same time, or even very nearly the same time, and hence there is little chance of mixing of feces and urine while they are on the belt. The rate of movement of the belt must be slow enough to permit urine to drain off before it is carried over into the receptacle for feces. Although small amounts of feces do occasionally work down the sides of the belt into the urine pan, these are recovered by a strainer in the cup-shaped receptacle at the bottom. Any feces collecting in this receptacle are, of course, removed and weighed to obtain the correct weight of total feces, but the material thus collected is discarded after weighing.

The question of contamination of urine by these fragments of feces in the draining pan has been raised. This has not been checked by analytical process as yet, but it is apparent that errors due to such contamination would be exceedingly small for the following reasons: (1) The determination of digestibility of feed would not be affected at all, because the total weight of feces is known and its analysis is based on a sample of the bulk which has not been contaminated (2) Urine is collected separately

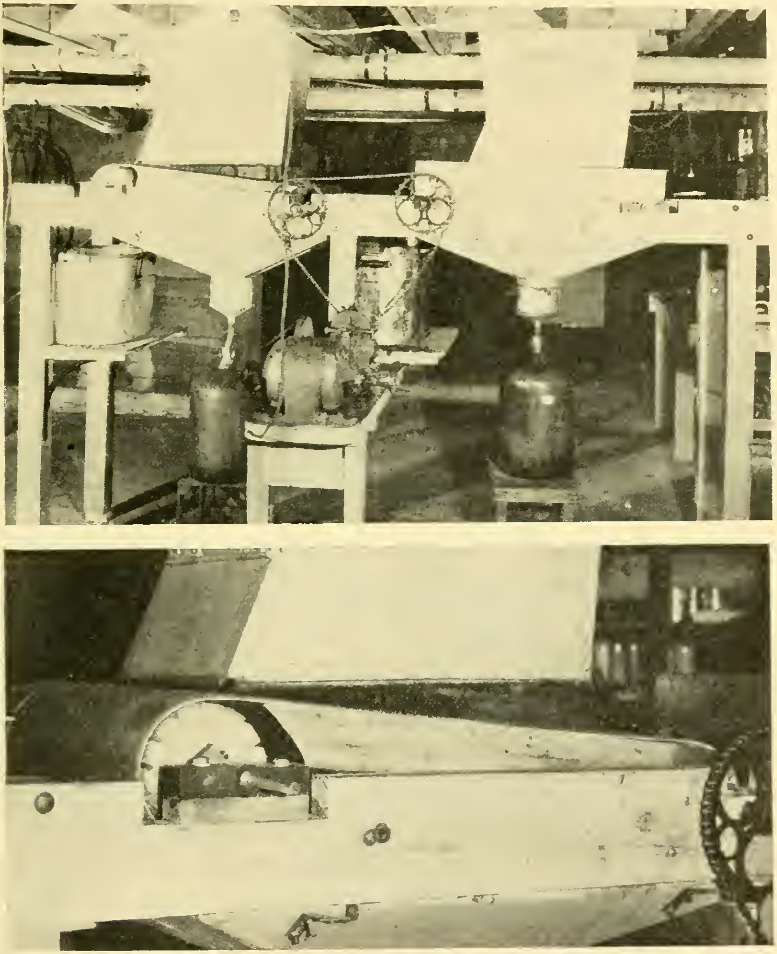


PLATE 7—*Apparatus for collecting separately the feces and urine from cows*

merely for the determination of the total nitrogen output. The chance for contamination of urinary nitrogen from feces is exceedingly small, because whatever readily extractable nitrogen the feed may originally have contained has in all probability been removed in the digestive tract. (3) The amount of feces which escapes into this drain pan is so small a fraction of the total feces that even the total nitrogen of the feed which these feces represent would not be sufficient to cause any significant error in the determination of the urinary nitrogen.

The greatest difficulty in developing this apparatus has been encountered in securing a good type of waterproof belt capable of adjustment to provide sufficient tension without running off the rollers. After several years of experimentation this difficulty has now been satisfactorily overcome by the use of a standard make of conveyor belting, which is composed of rubber with a fabric basis. The use of this heavy, 3-ply belting has, however, necessitated a considerable modification of the original construction of the apparatus. This belt is operated by a one-quarter h.p. motor

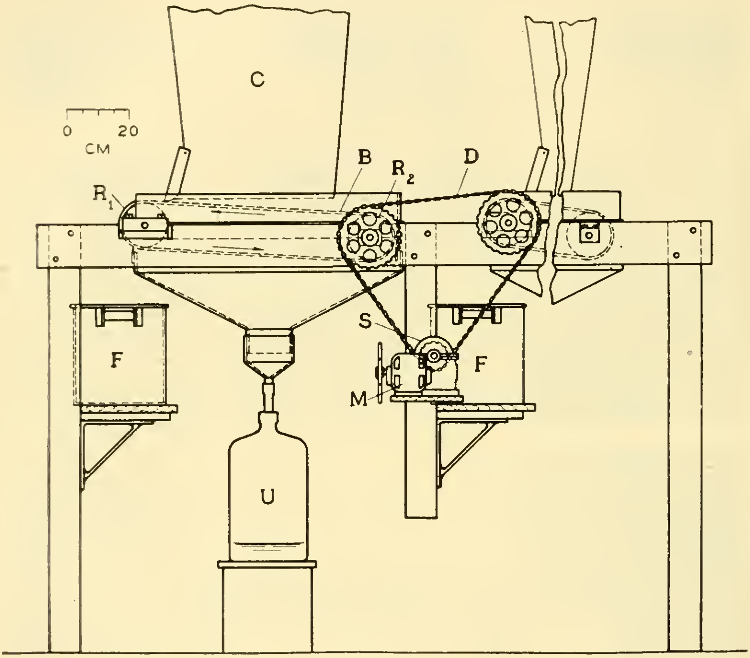


FIGURE 6—*Diagram of apparatus for separate collection of feces and urine of cows*

The legs of 4″ x 4″ fir and the horizontal pieces are 2″ x 6″ and 24 inches apart. The drainage pan, held in place by two clamps, is removable. Removable cups at the bottom have screen strainers to prevent fragments of feces from washing down into the urine bottles.

C, chute from stall above; B, top surface of revolving belt; R_1, belt roller; R_2, driving sprocket; D, driving chain; S, speed reduction gear; M, motor; F, receptacles for feces; U, receptacle for urine.

having 1200 r.p.m. hitched to a speed reduction unit by which the speed of the belt itself is reduced to about one revolution in two minutes.

With this apparatus it is now a very simple matter to collect and weigh urine and feces separately for any desired length of time. At this laboratory collections are made continuously from September until the following May or June. The apparatus beneath the respiration chamber for the separate collection of feces and urine from cows is identical in design and construction to the apparatus used for this purpose under the metabolism stalls. (See Plate 7 and Figs. 3 and 6.) In this case, however, the 2″ x 6″ frame which supports the rollers rests on the inclined floor within the sheet-metal container, which is designed to prevent exchange of air in the respiration chamber. By means of a sheet-iron shutter, hinged on the wall of the chute leading down to the receptacle for feces, the latter can be closed off from the apparatus above in order to permit an exchange of containers for feces without changing the composition of the air in this apparatus and in the respiration chamber. With steers such an apparatus was not necessary, as the urine could be separately collected by means of a very simple rubber funnel adjusted to the animal by means of a belt.

A Typical Experiment with Cattle

Energy transformation is readily influenced by the degree of activity and by the amount and the character of the food consumed. In the study of metabolism with humans there are, of course, differences in food level due to differences in age, in vocations, in habits of exercise, etc., but with the exception of occasional special meals the human diet for the individual tends to remain on a fairly even level throughout the year, so that a metabolism measurement after a 12-hour fast yields essentially a normal result for the individual. In the nutrition of farm animals the quantity and character of food consumed by the individual (except during grazing) may present an extraordinarily wide range, owing to the fact that these animals are kept primarily for the purpose of systematic conversion of vegetable matter into more valuable animal products. A metabolism experiment with such animals, therefore, must first be preceded by a preliminary preparation in feeding on a uniform level of supply, so that the measured energy expenditure can be attributed to a definite nutritional level.

By some years of experience in measuring the energy expenditure of animals under different conditions of feeding, we have found that a metabolism measurement should be preceded by from seven to fourteen days of feeding during which the amount and character of the food consumed is kept uniform at the nutritional level which the metabolism is to represent. This is particularly important when a radical change in quantity and quality of food has been made.

Digestion Balance

With cattle it is highly desirable to precede each metabolism measurement with a digestion balance. When the urine and feces can be collected separately, simple combustions (with the oxy-calorimeter) of dried samples of feeds and of feces will indicate the digestibility of the energy yielding constituents of the feed consumed, and Kjeldahl tests of such samples will give a good picture of the digestibility of the total nitrogen of the food.

The nitrogen of the urine has always been regarded as representing mainly the katabolism of body tissue. Carpenter[1] has recently shown that a small fraction of urinary nitrogen in steers (on pasture) may repre-

[1] Benedict, F. G., and E. G. Ritzman, Carnegie Inst. Wash. Pub. No. 377, 1927, p. 117; Carpenter, T. M., *Am. Journ. Physiol.*, 1927, 81 p. 519.

sent unutilized food nitrogen, that is, hippuric acid nitrogen and amino acid nitrogen. The quantities, however, are small so that for general purposes of study the urinary nitrogen may be accepted as a satisfactory measure of metabolized body protein or nitrogen.

The nitrogen balance is essential to the assessment of the energy metabolism either when the animal is being fed or is fasting, since in the former case food nitrogen supplies a material source of energy to the body, and in the latter case the nitrogen output is a direct measure of the protein disintegration of the body.

Insensible Perspiration

During all our experimental work we have aimed to determine the daily insensible perspiration of the experimental animals. The insensible perspiration represents the daily loss in weight due to the invisible carbon and water vapor expired through lungs and skin. It is thus a measure of the rate of metabolism because both carbon and water vapor are the by-products of metabolism. This insensible perspiration represents specific daily losses in weight (as shown in Table III) which vary with feed level, exercise, and other causes that influence the rate of metabolism.

It consists of the weight with which an animal starts the day plus what it eats and drinks minus the weight of the animal at the end of the day plus the visible excreta (*i.e.* urine and feces). Except for the first day, it requires simply one daily weighing of the animal with a careful record of the weight of food and water consumed and of the urine and feces voided. The insensible perspiration forms a good general measure of the rate of metabolism, but its accuracy depends entirely on the accuracy and dependability of the weighings, particularly of the animal. Unless scales for weighing the animal are dependable to within two or three hundred grams (*i.e.* half a pound) the insensible perspiration loses significance. Even then such scales must be tested frequently for their degree of accuracy.

Recent researches on humans[1] indicate a reasonably close correlation between the insensible loss and general metabolism. On basis of our investigations along this line extending over a period of ten years the same may be said with regard to steers and cows.[2]

Careful daily records of the insensible loss are of experimental value quite apart from its relation to metabolism in that they offer a basis for calculation of those changes in body weight from day to day which are due to irregularities in the replenishment and in the losses of the so-called "fill" which must always form part of the live body weight although it does not represent body material.

[1] Benedict, Carnegie Inst. Wash. Pub. No. 203, 1915, p. 84; Benedict and Hendry, Boston Med. and Surg. Journ., 1921, 184, pp. 217, 257, 282, 297, and 329; Benedict, Boston Med. and Surg. Journ., 1923, 188, p. 127; Benedict, Bull. Soc. Sci. d'Hygiène Alimen., 1923, 11, p. 343; Benedict, Schweiz. med. Wochenschr., 1923, 53, p. 1101; Benedict, The correlation between perspiratio insensibilis and total metabolism. Collection of articles dedicated to the seventy-fifth birthday of Professor I. P. Pawlow, published from the Institution of Experimental Medicine in Leningrad, 1924, p. 193; Benedict and Root, Arch. Intern. Med., 1926, 38, p. 1.

[2] Benedict, F. G., and E. G. Ritzman, Carnegie Inst. of Wash. Pub. No. 377, 1927, p. 63.

An ideal experiment thus involves records of the insensible perspiration over a period of 24 days, divided as follows:

7 days preliminary feeding
14 " feeding, with digestion balance and metabolism measurement during last two days and
1 day fasting followed by
2 days fasting with metabolism measurements and determination of urinary nitrogen output

With suitable equipment and organization for conducting an experiment of this order the work involved (excepting of course the periods of metabolism measurements) requires the time of two dependable assistants to weigh and water and to feed the animals, and to collect and to weigh aliquot samples. With four animals at our laboratory this procedure requires the time of one man for a quarter of an hour in the morning merely to check off any left-over feed and to put a fresh feed into the mangers, the rations being weighed out in advance. In the evening it requires the service of two assistants for about one hour, to feed the animals and to perform the other requirements mentioned above. The general results of such an experiment are illustrated in Tables III and IV.

TABLE III—*Record of live weight, water, food, feces and urine, and of the daily insensible loss during the two weeks of the digestion experiment preceding the respiration experiment*

Day	Weight of animal		Feed			Water	Urine	Feces	Insensible perspiration
	Start	End	Timothy hay	Beet pulp	Oil meal				
	Kgs.	*Kgs.*	*Kgs.*	*Kgs.*	*Kgs.*	*Kgs.*	*Kgs.*	*Kgs.*	*Kgs.*
1	597.8	599.7	5.0	2.6	1.3	24.5	3.0	16.4	12.2
2	599.7	603.1	5.0	2.6	1.3	23.8	3.3	13.4	12.6
3	603.1	582.9	5.0	2.6	1.3	0	4.3	13.3	11.5
4	582.9	595.6	5.0	2.0	1.0	31.8	3.9	10.9	12.3
5	595.6	603.5	5.0	2.0	1.0	29.0	4.0	14.0	11.1
6	603.5	585.1	5.0	2.0	1.0	0	3.1	11.8	11.5
7	585.1	598.1	5.0	2.0	1.0	30.8	2.8	11.2	11.8
8	598.1	599.2	5.0	2.0	1.0	21.1	3.2	14.1	10.7
9	599.2	596.5	5.0	2.0	1.0	15.4	3.5	12.1	10.5
10	596.5	598.1	5.0	2.0	1.0	18.8	2.7	11.7	10.8
11	598.1	601.9	5.0	2.0	1.0	22.3	3.4	12.0	11.1
12	601.9	594.7	5.0	2.0	1.0	10.7	2.4	10.9	12.6
13	594.7	601.9	5.0	2.0	1.0	26.1	3.3	9.3	14.3
14	601.9	599.2	5.0	2.0	1.0	16.8	2.7	14.4	10.4
Avg.	597.0	597.1	5.0	2.13	1.07	19.3	3.26	12.54	11.66

TABLE IV—*Average daily dry matter, protein, and heat of combustion in feeds, feces and urine during digestion balance*

	Dry Matter	Protein	Energy
	Gms.	*Gms.*	*Cals.*
Timothy hay	4420	150	19,890
Beet pulp	1950	100	8,770 [1]
Oil meal	970	320	4,950
		570	33,610
Feces	2520 [2]		12,060
Urine		280 [2]	630 from feed N.
		300 [3]	370 " body tissue [4]

Digestibility of Protein $\frac{570-280}{570}=51\%$. Digestibility of Energy $\frac{33610-12690}{33610}=62\%$.

[1] Estimated heat of combustion.
[2] Based on period 13, Table 27, Steer A (Pub. 324).
[3] Calculated on basis of period 13, Steer A, Table 33. Pub. 324, Carn. Inst. of Wash.
[4] Based on Discussion page 159, Pub. 324 (factor 12.63 cals. per gram nitrogen, period 13).

The total energy metabolized (measured by respiration experiments) per 24 hours on the feed level shown in Table IV, all other conditions being constant, was:

	Calories
1st day full feed	14,530
2nd " " "	14,780
1st half feed	
2nd " " "	11,800
3rd " " "	11,350
1st day fast following full feed	
2nd " " " " "	9,750
3rd " " " " "	9,630

It is not our purpose to discuss the physiological relations of these values, but to illustrate the nature of the data obtained and particularly to point out the relative constancy of metabolism measurements when conditions under which they are carried out are not varied.

Dependability of Metabolism Measurements

The degree of usefulness of metabolism measurements obtained by means of respiration experiments is determined by two tests: In the first place, when the conditions of different experiments are identical, the results must be identical. This test shows merely the comparative value of such data. An indication of this test is given in text table above. Secondly, it is equally necessary that the results of such an experiment be capable of interpretation in terms of absolute values. This test is carried out by a so-called gas check in which definitely measured amounts of carbon dioxide are introduced into the ventilating system and recovered by the usual experimental procedure.[1] Such tests are made several times each season in order to insure that the apparatus is functioning with a degree of accuracy which does not permit of a deviation of more than one per cent from 100.

CONCLUSION

It has been the purpose in this report primarily to point out the progress which has been made in design of equipment and in the corresponding technique, whereby it has become possible to make the measurements of animal metabolism a matter of normal daily procedure rather than an occasional event. The gratifying feature of this progress is that it has been accomplished by simplification of equipment and method rather than by augmenting the former complexities that have made the study of animal metabolism all but impossible to investigators in general. There has always been a reluctance among investigators not thoroughly familiar with the principles and practice of calorimetric methods to accord more than a mild physiological interest to the results of this kind. Apparently there still exists some confusion relative to the extent to which the physical objectives in animal feeding may be expressed by physiological manifestations. It has been maintained that the latter have no direct bearing on problems of a purely economic nature. This attitude can only be held by those unfamiliar with the continuous and unfailing response with which animal life registers its energetic activities and the extreme sensitiveness of present-day calorimetric apparatus to measure them.

[1] Pubs. 324 and 377, Carnegie Institution of Washington, and New Hampshire Sta. Bull. No. 16.