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GROWTH AND DEVELOPMENT

With Special Reference to Domestic Animals

L. The Influence of Fasting and Refeeding on Milk Production, Heat Production, and Respiratory Quotient.

L. E. WASHBURN, S. BRODY, and A. C. RAGSDALE

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FOREWORD

The special investigation on growth and development is a cooperative enterprise in which the departments of Animal Husbandry, Dairy Husbandry, Agricultural Chemistry, and Poultry Husbandry have each contributed a substantial part. The parts for the investigation in the beginning were inaugurated by a committee including A. C. Ragsdale, E. A. Trowbridge, H. L. Kempster, A. G. Hogan, and F. B. Mumford. Samuel Brody served as Chairman of this committee and has been chiefly responsible for the execution of the plans, interpretation of results and the preparation of the publications resulting from this enterprise.

The investigation has been made possible through a grant by the Herman Frasch Foundation, now represented by Dr. F. J. Sievers.

M. F. MILLER
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ABSTRACT

Time curves are presented for absolute and relative milk production, milk composition, respiratory quotients, heat production, and methane production, with advancing fasting and refeeding in dairy cows and goats, with comparisons to similar time changes in rats. The major conclusion of this research on ruminants confirm those on rats[4], namely that the percentage values of milk production, heat production, and respiratory quotient (above 0.7) fall and rise together with fast and refeeding. Hence the inference that it is not possible to determine the basal (post-absorptive) heat production during *normal* lactation because *normal* lactation appears to be incompatible with the post-absorptive state.

PAPER 187 IN THE HERMAN FRASCH FOUNDATION SERIES.

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L. The Influence of Fasting and Refeeding on Milk Production, Heat Production, and Respiratory Quotient*.

L. E. Washburn, S. Brody, and A. C. Ragsdale

I. INTRODUCTION

Does lactation increase heat production?

After reviewing the literature Lusk[1] concluded: "It appears that lactation does not increase the heat production. This is not strange since the arrangement of food materials in the preparation of milk depends upon the hydrolytic cleavages and syntheses which involve hardly any thermal action."

It is difficult to believe that the manufacture of milk from the feed constituents—or even out of milk precursors in blood—does not require some energy[2]; therefore, Lusk's conclusion does not seem reasonable.

Even if there were no energy price attached to the chemical transformation of milk precursors to milk, there should be an incidental higher heat production level during lactation as result of: 1) greater nutrient turnover with a consequent "specific dynamic action;" 2) increased level of endocrine activity[3]; 3) higher maintenance cost of the hypertrophied mammary gland and associated organs.

The apparent fact inferred by Lusk, that the basal metabolism in lactating women is no higher than in non-lactating may simply be due to the fact that milk production is markedly depressed under conditions of fast (required for basal metabolism measurements), rather than that there is no energy cost of milk production. The fact that milk production is profoundly depressed as a result of fasting required for attainment of post-absorptive condition has been shown in

*This manuscript was prepared by S. Brody with the assistance of A. C. Ragsdale after L. E. Washburn left Columbia to join the staff of the Colorado State Agricultural College, at Fort Collins. Grateful acknowledgments for assistance are made to Margaret Sappington, Hudson Kibler, and Paul Whitson.

[1]Lusk, G., The Science of Nutrition, Philadelphia, 1928.

[2]For an extensive discussion of the energetics of milk production see Univ. Mo. Agric. Exp. Station Res. Bulls. 222 and 238 (1935-6), and Graham, W. R., Jr., Houchin, O. B., Peterson, V. E., and Turner, C. W., The efficiency of the mammary gland in the production of milk. Am. J. Physiol. *122*, 150, 1938.

[3]Reece, R. P., and Turner, C. W., The lactogenic and thyrotropic hormone content of the pituitary gland. Mo. Agr. Exp. Sta. Res. Bul. 266, 1937, reported greater concentration of pituitary thyrotropic hormone during lactation. Papers, among others, by Schwarz, O. H., Drabkin, C., Basal rates in late pregnancy and the puerperium, Am. J. Obst. and Gyn. *22*, 3, 1931, point in a similar direction. For quantitative evidence of increase of pituitary thyrotropic hormone concentration during lactation, see Turner, C. W., & Cupps, P. T., "The thyrotropic hormone in the pituitary of the albino rat during growth, pregnancy and lactation." In press in Endocrinology, 1939.

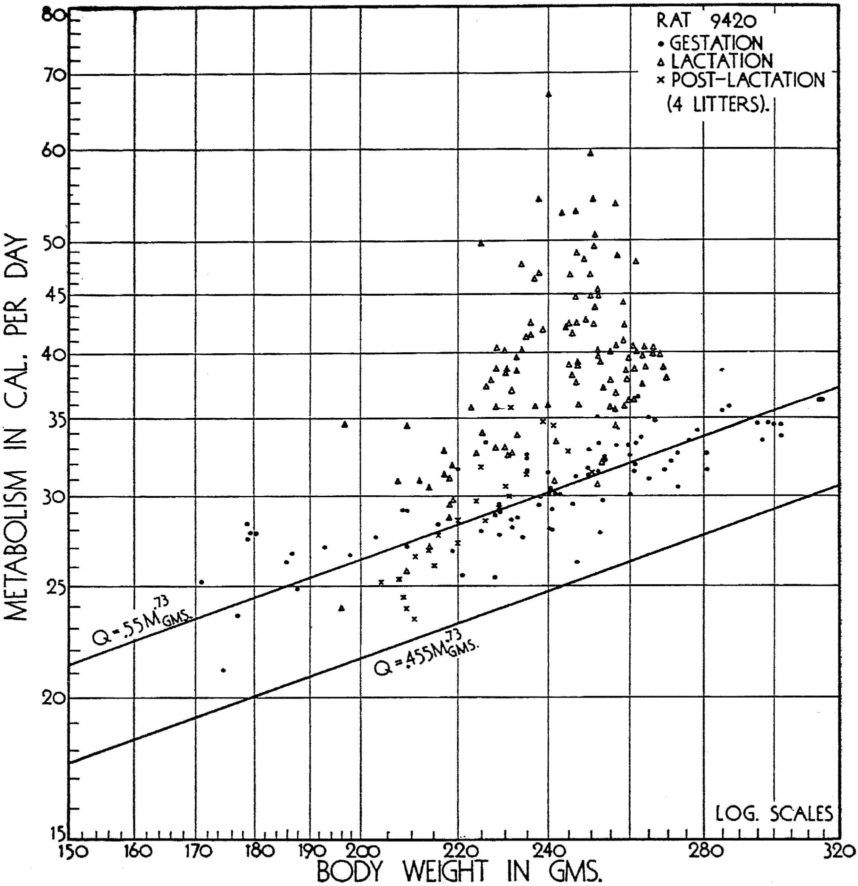

Fig. 1a.—Relative positions of heat-production levels in the same rat including four reproductive cycles during: lactation (triangles); gestation (circles); and post lactation rest (crosses) under conditions of normal food supply. Note that the lactation metabolism level is far above—in some cases about 100% above—the non-lactation level. This curve contradicts the belief that milk production does not increase heat production.

a comparison of the time curves of milk production, heat production, and respiratory quotient in the rat[4]. A summary of this comparison between heat production and milk production in the rat is presented in Figs. 1a and 1b. The present investigation is an extension of the previous one to include dairy cattle and goats; to find whether or not the parallel decline in milk production and heat production observed in rats is also found in ruminants.

[4]For further review of literature see Brody, S., Riggs, J., Kaufman, K., and Herring, V., Energy-metabolism levels during gestation, lactation, and post-lactation rest. Mo. Agr. Exp. Sta. Res. Bul. 281, 1938.

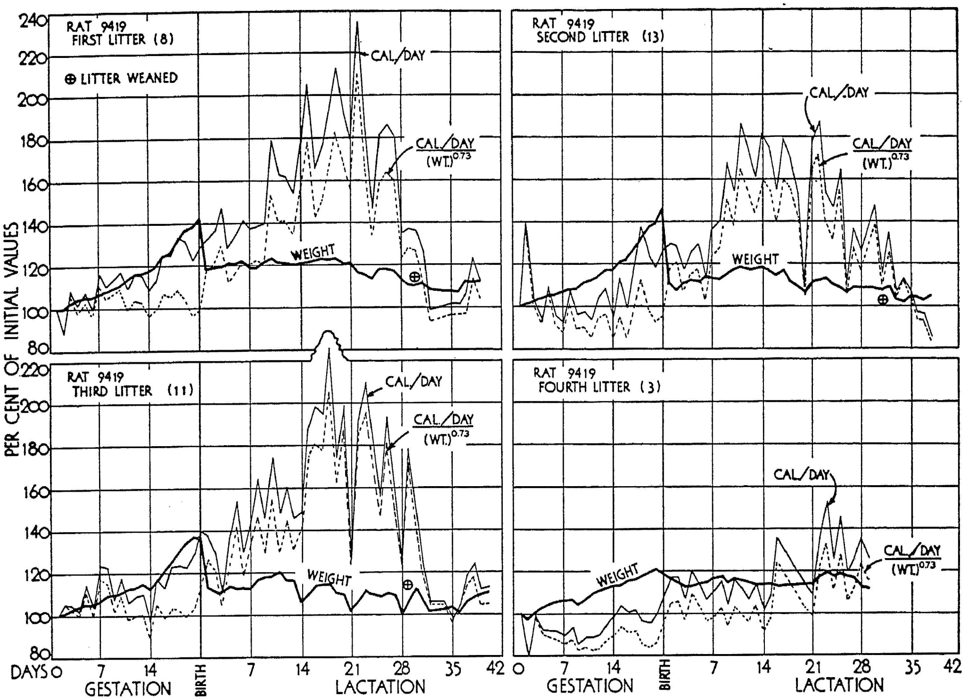

Fig. 1b.—The influence of milk-production level (as measured by number of young in litter) on heat production in the rat.

On *a priori* considerations it would be expected that the results on rats would be substantiated on cows and goats for the following reasons: 1) It is generally known that the heat production and the R. Q. (respiratory quotient) decline with fasting, reaching the basal heat production level in ruminants about 3 days after the last feeding. 2) It is also known that milk production in dairy cattle declines, reaching a very low level after three days of fast. Hence the conclusion that the attainment of a condition of fast is associated not only with decline in heat production and decline in R. Q. but also with decline in milk production.

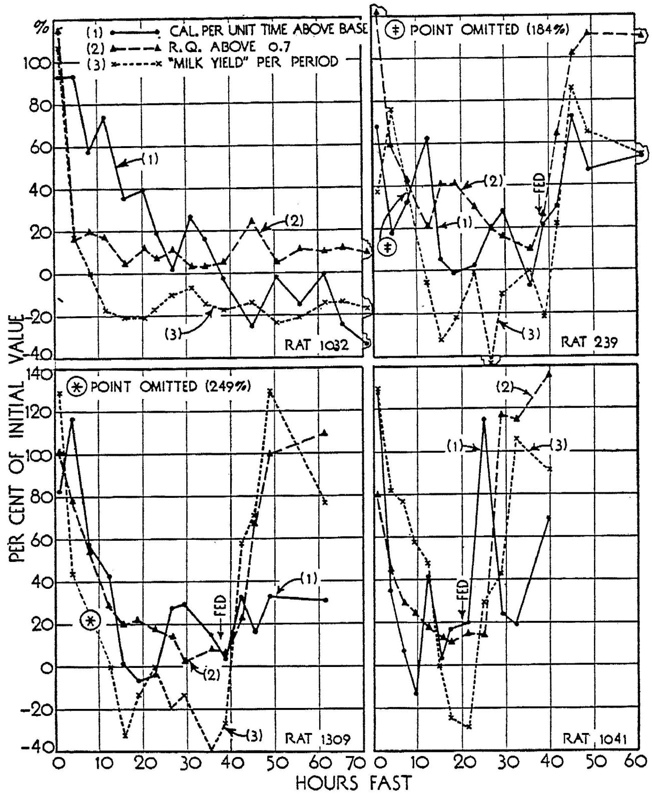

Fig. 1c.—Comparison of the time curves of heat production above basal level (curves 1), R. Q. above 0.7 (curves 2), and "milk yield" (curves 3 measured by gains in weight of litter during suckling) in rats. All data are represented in terms of percentage of the initial (normal) values. Note the striking parallelism of curves 1, 2, and 3 indicating that it is not possible to measure the *basal* metabolism of rats yielding the *normal* amount of milk. Fasting which eliminates the heat increment of feeding in the lactating rat also eliminates milk production.

Excellent numerical data on the decline of production of milk and its constituents with advancing fast have been published by Overman and Wright[5]. We have recomputed their data in terms of percentages of the prefasting (control) periods, and are presenting the resultant percentage changes in Fig. 2.

The lower curves in Fig. 2 show quite clearly that on approach to the post-absorptive (fast) condition, the milk yield (curve 6) declines very rapidly, reaching 5% to 20% of the original level within 3 days.

[5]Overman, O. R., and Wright, K. E., The effect of inanition upon the yield and composition of cow's milk. J. Agric. Res. *35*, 637, 1927. See also Gowen, J. W., and Tobey, E. R. Studies of milk secretion: The influence of inanition. J. Gen. Physiol. *15*, 45, 1931, and The influence of insulin and phloridzin, Id., p. 67.

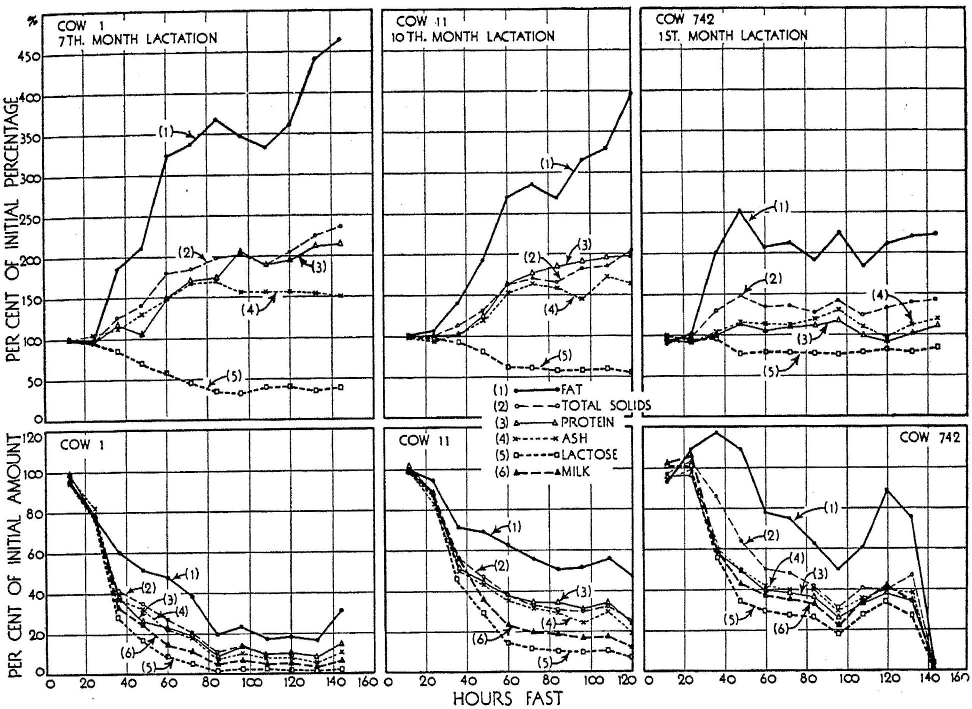

Fig. 2.—Fasting cattle for 3 days (said to be time required to reach the post absorptive level for basal metabolism measurements) reduces milk yield to 5%-20% of the original level (depending on the stage of lactation and individuality of the animal). The fat production is not reduced as rapidly as the milk because the percentage fat in milk rises on fasting due to rise in the lipoids percentage in blood, but the milk sugar declines more rapidly than the milk as a whole because the percentage of sugar in milk declines with advancing fast as a consequence of decline of body glycogen depots and probably decline of blood sugar concentration. This curve illustrates strikingly the fact that it is not possible to measure the *basal* metabolism of animals yielding *normal* amount of milk because the fast required for basal metabolism profoundly depresses milk production.

There are of course differences in the rate of decline of the several constituents of milk with advancing fast, depending on the rate of decline of the several constituents in the blood with advancing fast. It is generally known (as indicated for example by the decline in the R. Q.) that fast lowers first the glycogen stores, hence presumably lowers the blood sugar concentration and consequently lowers milk sugar concentration. Curves numbered (5) in Fig. 2 demonstrate that the milk sugar undergoes the most rapid decline with advancing fast. It is likewise generally known that fast raises the blood-lipoid concentration due to body-fat mobilization, and there is a corresponding rise of fat percentage in milk as the fasting period advances. The curves in Fig. 2 demonstrate that as fast advances there is more rapid rise in fat percentage (upper curve) in the milk, and slower decline in absolute (total) fat yield than of the other milk constituents, or of the milk yield as a whole.

Likewise, the rate of decline in milk production is dependent on the concentration of lactation hormones, so that on fast the decline in milk production is slower in the early stages of lactation (see cow 742 in Fig. 2) than in later stages (cows 1 and 11).

But regardless of the dissimilar changes in milk composition with advancing fast, there is no doubt that there is general similarity between the decline in milk production as a whole (Fig. 2) and decline in heat production (see numerous time curves of fasting in Mo. Res. Bul. 143).

The purpose of this bulletin is to report on the relative courses of decline of milk production, heat production, respiratory quotients, and cardio-respiratory activities in dairy cattle and goats with the object of throwing further light on the question as to whether or not milk production is associated with increased heat production. As previously noted, Lusk[1] concluded that lactation does not increase heat production because the basal (i.e., post-absorptive fast) heat production is no higher in lactating than non-lactating women; whereas our paper on the metabolism of the rat[4] reported that milk production in the rat is profoundly depressed after a day's fast, and concluded the reason that the basal heat production as reported by Lusk was no greater in lactating than non-lactating individuals was that on reaching the condition of fast required for measuring basal heat production the lactating animal practically ceases to be a milk-producing animal.

Washburn[6] suggested that lactation is "an endocrine-stimulated energy-converting system, not necessarily measured by quantity of milk produced." In this sense of the meaning of lactation as "not necessarily measured by quantity of milk produced," Lusk's conclusion that "lactation does not increase milk production," may be correct if it is understood that Lusk's conclusion applied to lactation during fast (post-absorptive state), when *milk yield* is greatly depressed. However, Ritzman and Benedict[7] reported in a recent extensive summary of their work that the difference even in *basal metabolism* of two cows when lactating and when dry and not pregnant amounted to about 30%. There thus appears to be contradictions between the results summarized by Lusk[1] on humans, our[4] results on rats, Ritzman and Benedict's[7] results on cows, and Washburn's[6] interpretation. This bulletin attempts to throw further light on these

[6]Washburn, L. E., Fasting energy metabolism during lactation. J. Dairy Sc., *21*, 697-703, 1938.

[7]Ritzman, E. R., and Benedict, F. G., Nutritional physiology of the adult ruminant. Carnegie Institution of Washington Publication 494, 1938 (see p. 136).

contraditions, and incidentally present other cogent data accumulated in this research.

The tentative conclusions that we infer from the data presented in this bulletin with reference to rats, cattle and goats are: 1) normal lactation, as contrasted to lactation under conditions of fast or under conditions of *basal* energy metabolism, is associated with a high increment of heat production. 2) It is not possible to say how much of this extra heat production represents the heat increment of feeding (SDA), and how much it represents other expenses of lactation such as cost of synthesis of milk from its blood precursors, maintenance of the hypertrophied gland and associated organs, accelerated metabolism stimulated by the endocrine system associated with lactation. 3) Attainment of the condition of fast, referred to as post-absorptive condition, required for basal heat measurements, profoundly depresses milk production, as well as heat production.

The data are presented in tabular form in the appendix. The following text presents these data in graphic form with explanatory comments. Some of the material collected incidentally for control purposes are also presented and discussed for their general or incidental interest.

II. DETAILED PRESENTATION AND DISCUSSION OF DATA

1. Methods and Materials

The heat production and respiratory quotients (R. Q.) of the cows and goats were computed from analyses of directly expired air. The methods of collecting and analyzing the expired air, and also the methods of computing the heat production, methane, and respiratory quotients, have been previously described[8].

As previously explained[8], our method differs from those previously employed (chamber methods), in that: 1) the animals are not in chambers but trained to lie quietly under customary barn conditions, under complete control of the operator, with a rubber sleeve (mask) over the muzzle; 2) the combined pulmonary and esophageal air is collected directly, uncontaminated by chamber air, or by gases excreted by way of the anus, or by gases formed by fermentation of the feces in the chamber; 3) the expired air is measured and analyzed at short intervals (about 30 minutes), in contrast to the longer chamber-method intervals.

[8]Washburn, L. E., and Brody, S., Methane, hydrogen, and carbon dioxide production in the digestive tract of ruminants in relation to the respiratory exchange. Univ. Missouri Agric. Exp. Sta. Res. Bul. 263, 1937.

The experimental animals consisted of an 8-year old 373 Kg. (822-pound) lactating Guernsey cow, number 428, in the 4th to 7th months of lactation, and an 8-year old 504 Kg. (1100-pound) dry (dry over a year) Jersey cow, number 831, which served as control.

It is unfortunate that only two animals were investigated, and that the two cows, 428 and 831, differed in body weight and breed, so the data could not be subjected to statistical analysis. The limitation and differences were made necessary by economic and other exigencies of the situation. Because of this situation the conclusions are necessarily tentative.

To compensate for the limited number of dairy cows used in this investigation, similar experiments were conducted on two lactating and two dry goats each weighing about 100 pounds, and also on rats, the results of which have already been reported[4].

The guiding idea of this research was to follow the time courses of heat production, R. Q., and milk production, with advancing fast in order to discover whether the curves of decline of these three functions are parallel. If they are, we should conclude that milk production is associated with an inseparable heat production increment, that is, with an energy cost of milk production; and the reason for the apparently observed fact[1] that the *basal* heat production of lactating individuals is no greater than of non-lactating, is simply that the attainment of the condition of fast (required for basal heat production measurement) depresses milk production to such a low level (and possibly depresses the heat production of the organs not associated with the lactation system to an especially low level so as to compensate for the extra heat production of the lactation system) that the associated heat production of milk production is likewise depressed.

2. The Time Course of Combustible-Gas Production in Milking and Dry Animals.

About 10% of the gross feed-energy intake is lost in ruminant digestion as result of anaerobic fermentation in the digestive tract[9]. For practical purposes all this gas may be considered to be methane (CH_4) having a heat value of about 9.5 Calories (kilo-calories) per liter. In addition there is a heat production—a waste heat of methane formation which is useless except for keeping animals warm in cold weather—of about 4 Calories per liter of CH_4 formed[10]. From a practical viewpoint the fermentation gas represents an apparently unavoidable waste. From a theoretical viewpoint it is a source of

[9]Cf. Brody, S., and Procter, R. C., Influence of the plane of nutrition on the utilization of feed stuffs. Univ. Mo. Ag. Exp. Sta. Res. Bul. 193, 1933.

[10]See Krogh, A., and Schmit-Jensen, The fermentation of cellulose in the paunch of the ox and its significance in metabolism experiments. Biochem. J. *14*, 686, 1920.

confusion in the interpretation of respiratory-exchange data because in addition to CH_4, at least as much carbon dioxide, CO_2, is produced by this fermentation; with the result that the tissue-metabolism CO_2 is confused by the fermentation CO_2. Hence the importance of evaluation of CH_4 production in the study of ruminant metabolism not only for estimating the feed-energy wastes and the heat of fermentation, but also for interpreting respiratory exchange and energy-metabolism data.

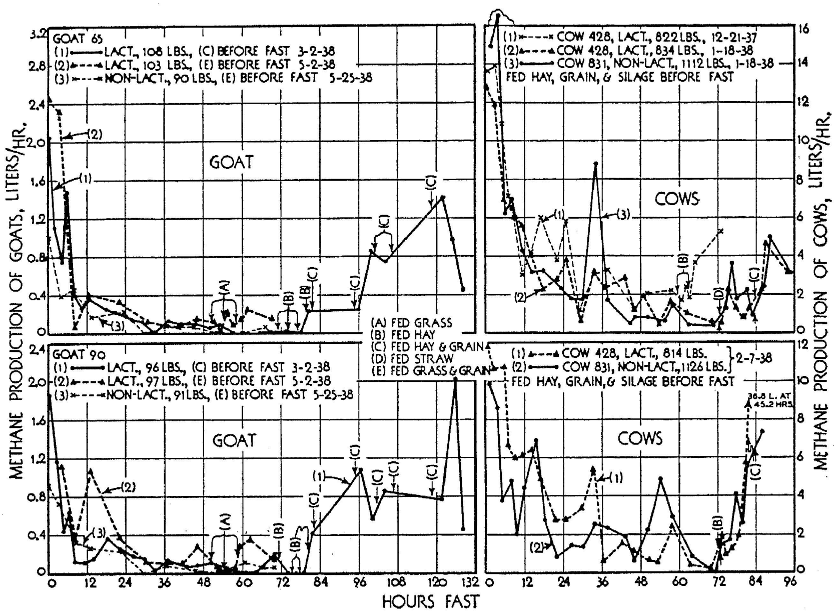

Fig. 3.—Time curves of combustible-gas (methane) production with increasing fast and refeeding in lactating and non-lactating goats (left) and cows (right).

The course of CH_4 production is also of interest in the present connection because it indicates the course of decline of the feed residue with the advance of fast as indicated by Fig. 3. From the curves in Fig. 3 it seems that two days fast reduces the fermentation level, and perhaps feed residue, to about 10% of the original level.

3. The Time Course of the Respiratory Quotient in Milking and Dry Animals.

Fig. 4 represents the time course of the respiratory quotient (R. Q.) corrected for fermentation, as previously described[8], with advancing fast in lactating and non-lactating goats and cows.

The R. Q. is the ratio of the volume of carbon dioxide exhaled to the oxygen absorbed. When pure carbohydrate is burned (without or

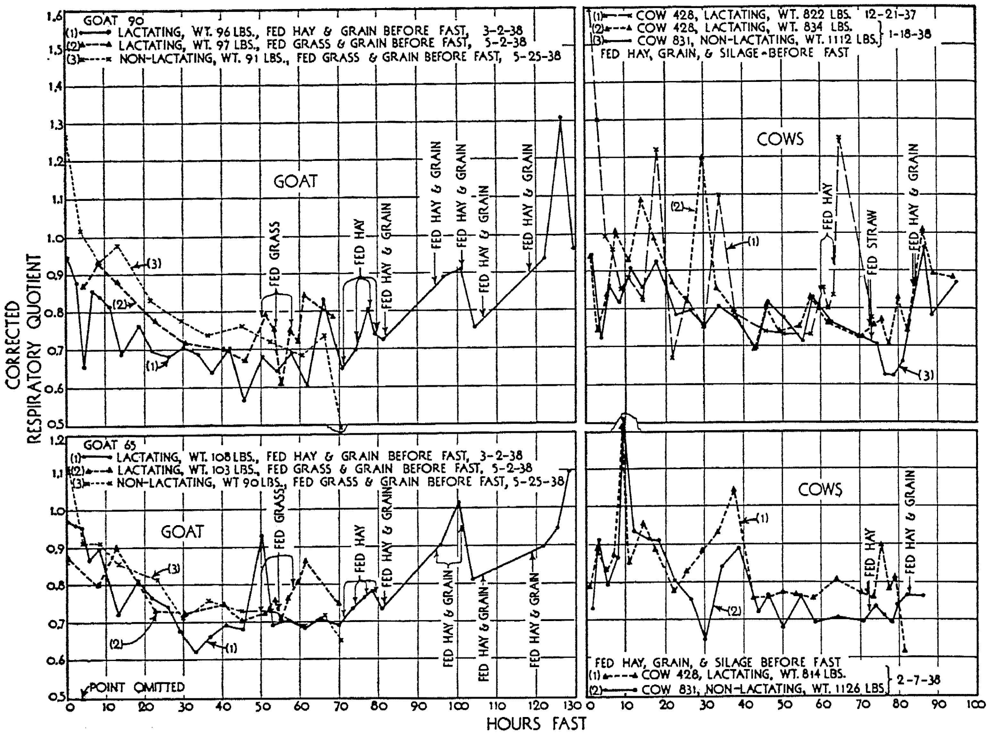

Fig. 4a.—Time curves of respiratory quotients (corrected for fermentation CO_2) with increasing fast and refeeding in lactating and non-lactating goats (left) and cows (right). Note time required to reach the post-absorptive R. Q.

within the body) the R. Q. is 1.0; when fat is burned the R. Q. is of the order of 0.7. An intermediate R. Q. indicates various proportions of fat and carbohydrate oxidation depending on the R. Q. value. An R. Q. over 1.0 normally indicates conversion of carbohydrate into fat. However, it is possible that in ruminants other factors enter into the conditioning of the R. Q. level.

Fig. 4 shows that the R. Q. corrected for the CO_2 fermentation is considerably above 1.0 shortly after feeding, and approaches 0.7 after two to three days of fast. The initial R. Q. is higher if not corrected for the fermentation CO_2. The high initial R. Q. in the lactating cow may be accounted for by the fact that the feed was largely carbohydrate and that considerable amounts of this carbohydrate is transformed to milk fat, and it is known that the R. Q. increases with the increase in fat formation, that is lipogenesis.

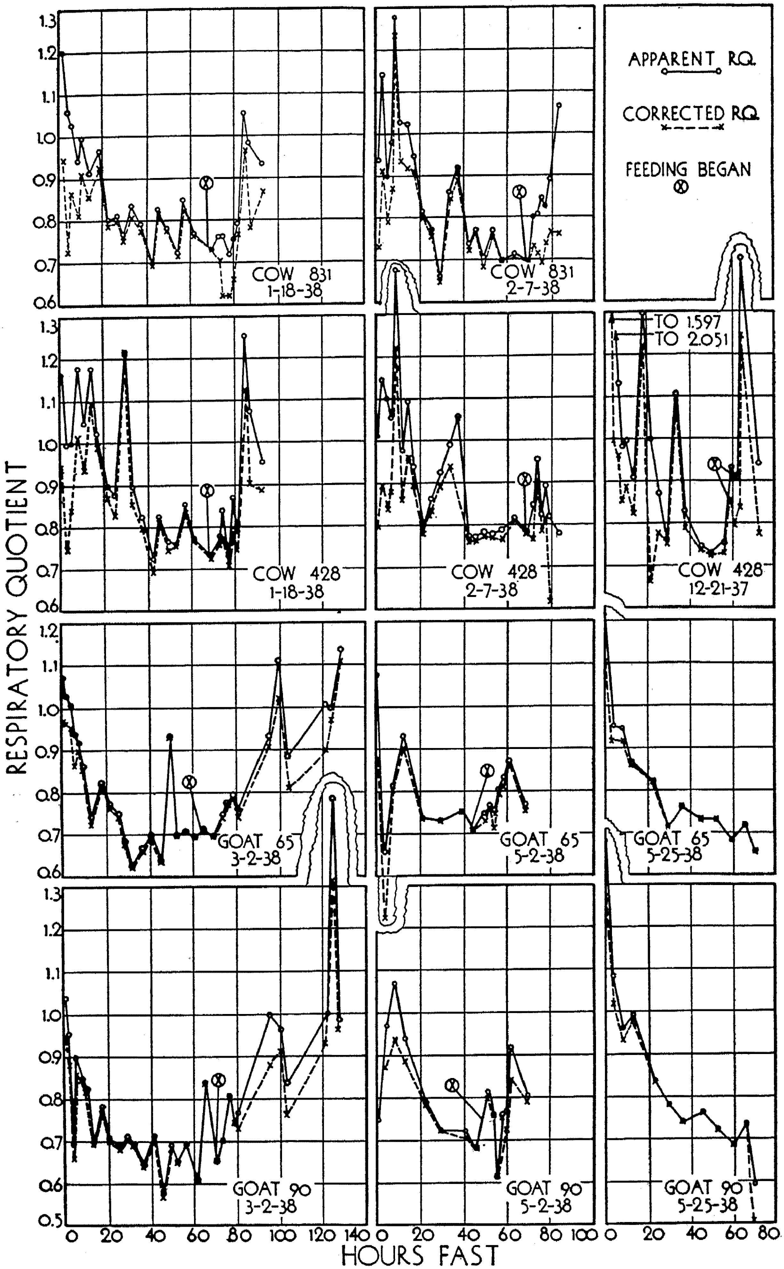

Fig. 4b.—Comparison of the R. Q. time curves corrected and uncorrected for fermentation CO_2.

The R. Q. of lipogenesis, with its extensive literature was recently discussed in detail by Benedict[11], and we need not go into this problem except to say that it bears on our computations of heat production. The heat production is computed from the amount of oxygen consumed and the R. Q. The heat equivalent of oxygen (amount of heat produced per unit oxygen consumed) has a characteristic value for each R. Q. level. The heat values are generally agreed upon for R. Q. levels between 0.7 and 1.0, but not for levels above 1.0. However, Benedict concluded on the basis of his investigation of lipogenesis in the goose (l. c. p. 230) that "in all respiratory-exchange measurements where the quotient is above 1.00, it is believed that the heat production can be directly calculated from the measured oxygen consumption with sufficient accuracy by use of the factor 5.047 Calories per liter of oxygen"—which is of course the heat value of oxygen when the R. Q. is 1.0. The metabolism values in the following sections were therefore computed on the basis of the assumption that the heat values of oxygen when the R. Q. level is above 1.0 is the same as when the R. Q. is 1.0.

4. Time Course of Heat Production, Total and Per Unit Physiologic Weight in Lactating and Dry Animals.

Fig. 5a presents the time course of heat production in cows and Fig. 5b in goats.

The outstanding feature in Fig. 5a is that the total heat production per day of the 820-pound lactating cow 428 is the same as of the 1120-pound dry cow 831, in spite of the 300 pounds live-weight difference. This might indicate that the extra energy expended for milk production in cow 428 is equivalent to the extra energy expended for maintaining the extra 300-pound body weight in dry cow 831. This may be the case, but if this were true, we would expect that after two to three days fasting, when milk production is reduced to a very low level, the heat production in the small lactating cow would fall below that of the large dry cow, which it did not as strikingly as might be expected. As previously noted, Washburn[6] suggested that lactation is "an endocrine-stimulated energy-converting system, not necessarily measured by quantity of milk produced," so that it would not be expected that heat production would be depressed to the same degree as milk production during fasting. A share of the extra heat production during lactation may be attributed to higher maintenance of the hypertrophied active mammary gland and to higher endocrine activity which may continue even during starvation.

[11]Benedict, F. G., and Lee, R. C., Lipogenesis in the animal body, with special reference to the physiology of the goose. Carnegie Institution of Washington, Publication 489, 1937.

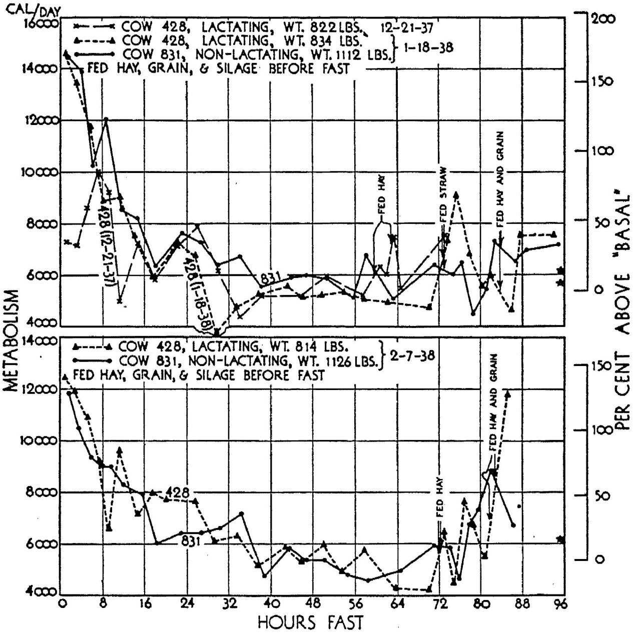

Fig. 5a.—Time curves of heat production with increasing fast and refeeding in the 800-pound lactating cow 428, and 1100-pound dry cow 831. Note that the heat production is the same in the two cows despite the 300-pound difference in body weight in favor of the non-lactating animal. These curves indicate a definitely higher heat production (with respect to body weight) in the lactating cow than the non-lactating even under basal-metabolism conditions of fast when milk production was greatly depressed. Note that the heat production shortly after feeding is about 150% above the "basal metabolism" level. But these enormously high initial metabolism values are independent of lactation. The stars on the extreme right, which represent "basal metabolism" values of dry dairy cows of approximately the same weight as our milking cow interpolated from Ritzman and Benedict,[7] were inserted by way of comparison and check with our data.

Fig. 5 represents metabolism in terms of heat production per day not corrected for body size of the animal. We next compare the heat production of the lactating (L) and non-lactating (NL) cows per unit "physiologic weight," which is tentatively taken to be the 0.73 power of body weight[12], with results shown in Fig. 6.

[12]Cf. Brody, S., Relativity of physiologic time and physiologic weight. GROWTH, 1, 60, 1937.

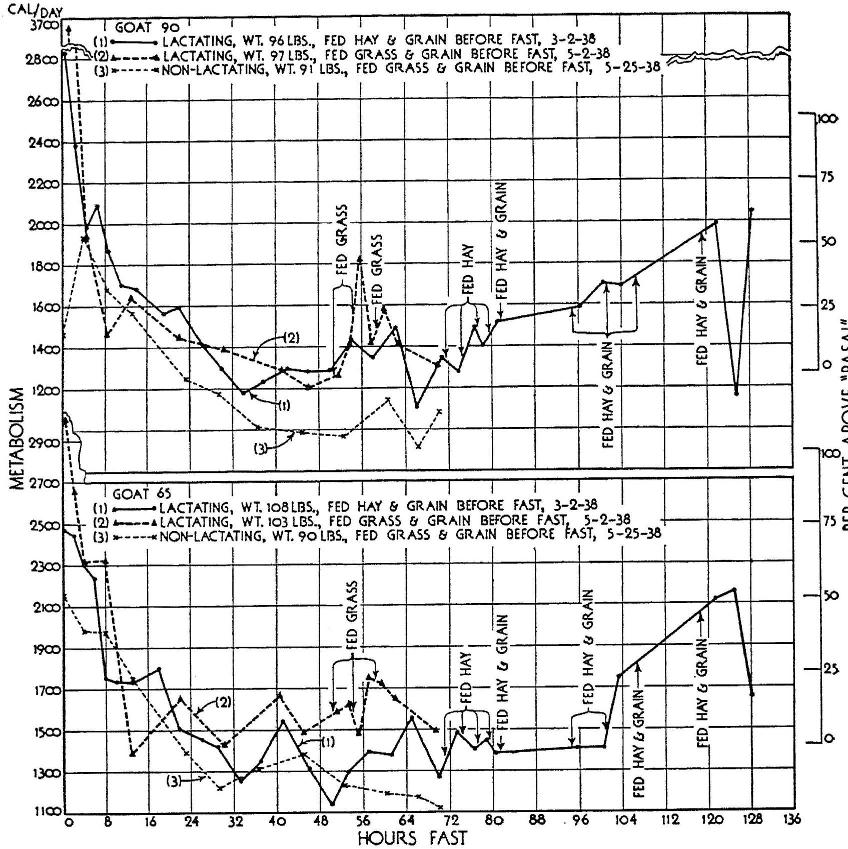

Fig. 5b.—Same as 5a but for goats.

From Fig. 6, there is no question that per unit "physiologic weight" metabolism of the lactating animals is greater than of the non-lactating. However, the authors expected that three days fasting would bring the curves of the lactating and non-lactating animals together. This expectation was nearly approached in the cows, but not in the goats (see Fig. 5b). The non-lactating goats were dried off only two weeks before the experiment so that the lactation-endocrine stimulus may not have been eliminated by the time that the animals were considered to be dry.

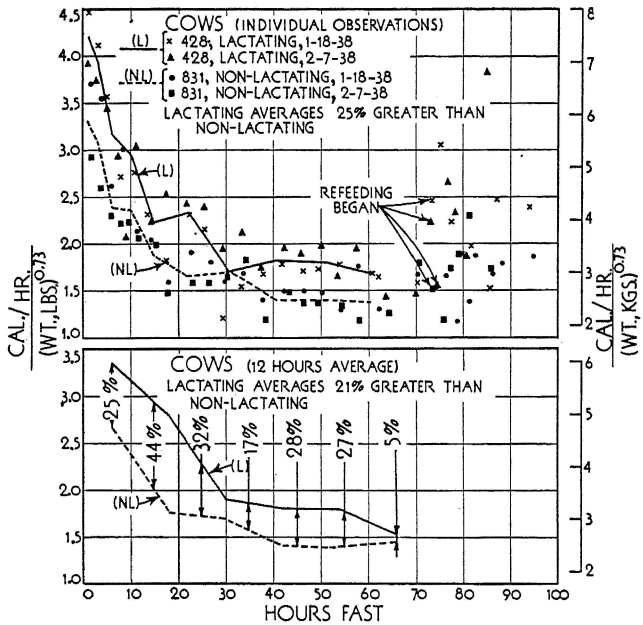

Fig. 6.—Heat production per unit of "physiologic weight" (Wt.$^{0.73}$) in lactating (L) and non-lactating (NL) cows. In Fig. 6 the time curves of heat production of the dry 1100-pound cow coincided with the lactating 800-pound cow in spite of the 300-pound difference in live weight—indicating a higher heat production with respect to body size in the lactating than dry cow. But when the heat production is presented with reference to the 0.73 power of body weight ("physiologic weight"), the heat production of the milking cow is considerably above the dry cow as shown in this chart.

5. Comparison of the Relative Time Changes in Milk Production, Heat Production and Respiratory Quotient.

It is evident that the time curves of milk production with advancing fast in Fig. 2 strongly resemble the time curves of: 1) methane production in Fig. 3, 2) respiratory quotient in Fig. 4, and 3) heat production in Fig. 5. In other words, *normal* milk production appears to be inseparably tied up with feed consumption and heat production. They fall and rise together.

The parallelism between the time changes with fast of milk production, heat production, and R. Q., may be demonstrated in a more quantitative fashion by plotting both on the same axes in terms of percentage of the prefasting (control) periods. Such comparisons shown in Figs. 7a (goats) and 7b (cows) speak for themselves. There is no doubt of the strong quantitative interrelation between milk

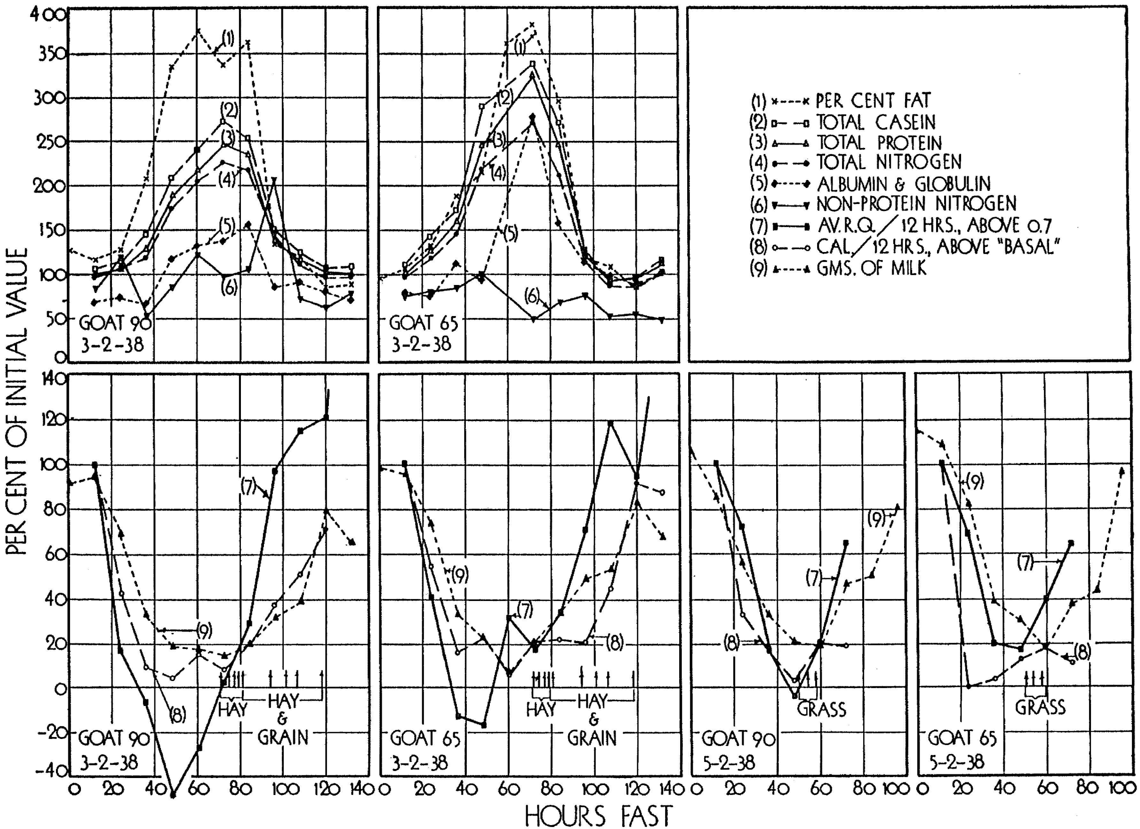

Fig. 7a.—Goats. The lower half represents comparison of the time curves of milk production (curve 9), heat production above base (curve 8), and R. Q. above 0.7 (curve 9), all in terms of percentage of normal initial values. Note the striking parallelism of these curves for goats and compare to the parallelism of the curves in Fig. 1c for rats. The upper curve represents the time changes in composition of the milk with advancing fast and refeeding. Campare to Fig. 2.

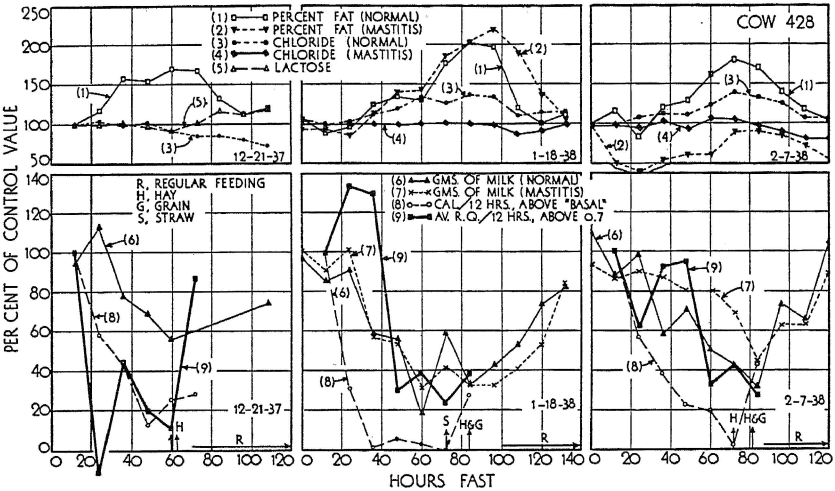

Fig. 7b.—Same as Fig. 7a but for cow 428, and showing in addition a comparison of the milk from the three normal and one mastitic quarter discussed in the text.

production, feed consumption, heat production (above base level) and R. Q. (above 0.7). This relation is also exhibited for the goats in Fig. 9 in which the heat production above the base level is plotted against milk production (both for the same 12-hour periods).

A linear equation was fitted to the data in Fig. 8, although it is almost certain that the distribution of the data is logarithmic rather than linear. It appears that the absolute heat production rises more rapidly than the milk production, as might be expected from the law of diminishing returns. No attempt, however, was made to fit a logarithmic equation to the data because of their limited amount. The coefficient of correlation, 0.897 is however highly significant indicating an unquestionable interrelation between heat production above the basal level and milk production.

In connection with Fig. 7b incidental note is made concerning the relative influence of fasting on milk yield and composition in the three apparently normal quarters, and on one mastitic quarter in cow 428. Under normal feeding conditions (prior to the first fast) the foremilk from the left rear quarter contained considerable curd-like chunks and strings of mucous-like material typical of mastitis. After 36 hours fast, the abnormal milk disappeared, and did not reappear until after the second fast when the animal was turned on pasture. During the 3rd fast, it disappeared again after 36 hours of fast, and did not reappear during the remainder of the observation period of

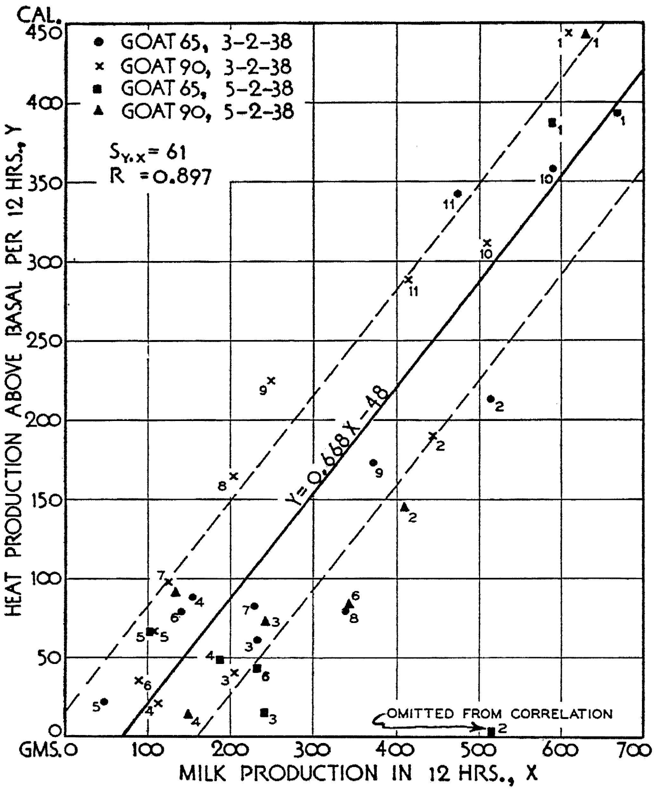

Fig. 8.—Heat production per 12-hour period above the basal level plotted against milk production during the corresponding 12-hour period, during fast and refeeding of the goats. The data points representing successive 12-hour periods are numbered serially. A linear equation was fitted to the data although it seems that the distribution of the data is not linear. The space between the broken curves includes ⅔ of the data points. Sy.x, the standard error of Y about the equation line (Y as a function of x), has a value of 61, which means that the heat production above basal can be estimated from the equation with the odds 68 out of 100 that the error will not exceed 61 Calories.

3 weeks. The mastitis and normal milks were kept separately during the second and third fasts, with the results in milk yield and composition exhibited in Fig. 7b. Washburn associated the disappearance of the mastitis condition with increase in the globulin and albumin fraction in milk (difference between total protein and casein protein) and consequently possible increase of immune bodies which are thought to be associated with these proteins, with the advance of fast. There is a large literature indicating increased permeability of epithelial cells during fast. These results suggest possible beneficial therapeutic effects of fasting on mastitis.

III. SUMMARY AND CONCLUSIONS

It is established that the heat production in lactating animals is very much higher than—sometimes double—that of non-lactating animals. (For evidence see Figs. 1, 5, 6 in this bulletin; for substantiating evidence see Missouri Res. Bul. 176, 1932; pp. 15 and 23 and references 6 and 7.)

It is established that on fasting rats, cows, and goats, there is a parallel decline in percentage milk production, heat production and respiratory quotient. Hence it is concluded that it is not possible to measure fasting or "basal metabolism" of animals producing milk at their *normal* level, because fasting profoundly depresses milk production. In other words, fasting or basal metabolism is not compatible with milk production at the *normal* (i.e., customary) level. There is an inseparable interrelation between milk production, feed consumption, and heat production.

The theoretical significance of these results is that the "basal metabolism" of a lactating animal can not be evaluated simply because it is not possible to attain the post-absorptive condition requisite for "basal metabolism" measurements and yet maintain lactation at its normal level. The practical significance of the dependence of a good milk flow on an *immediate* abundant food supply is too well known to need elaboration. Milk flow is very sensitively dependent on a *continuous* food supply. Missing even one meal affects milk flow, but missing one or even several meals does not impair the milk producing *capacity,* because it is here demonstrated that milk flow is promptly brought back to normal on refeeding the fasting animal.

IV. APPENDIX: NUMERICAL DATA.

(See Insert)