XVII. THE EFFECT OF DIET ON MAMMARY SECRETION.

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I. INTRODUCTION.

THE literature of research work performed to elucidate the effect of diet on the secretion of milk does not give the impression that there is a definite relation between the two things. On the other hand, the great experience of the farmer indicates how closely these are related, and any precise information would be not only of value to him, but might also assist in overcoming the disability so frequently met with in modern women, many of whom are unable to breast-feed their children owing to a deficient secretion.

Various methods are available for studying this problem.

1. The animals could be fed on diets of well-known foods, used in different proportions, such as bread, meat, butter, etc.

2. Synthetic diets could be employed, consisting of isolated carbohydrates, proteins, fats, vitamines, salts and water.

3. A combination of both the above methods is possible.

It seemed probable that some factors could be most easily found in one way and some in another.

Since man lives on natural foods and not on a diet of synthetic foodstuffs, the first method is more likely to yield results, directly applicable to human mothers and babies, than either of the others. Hence in the experiments described in this paper, the diets were chosen from ordinary foods and the rate of growth of the young during the lactation period was gauged by the daily increase in weight. The mother's weights were also recorded, since an ideal diet would keep up the mother's weight, and at the same time provide for the rapid growth of her young, while an inadequate diet might fail in one or both respects.

An omnivore had to be used and rats were chosen, because they readily eat all foods normally taken by man, and psychic effects resulting from forcing an animal to eat unpalatable food were prevented. Also rats were most convenient, since they are small, have short gestation and lactation periods and will breed throughout the year, provided they are kept warm. One rat will give birth to and rear as many as six healthy litters in a year.

The drawback to work on rats is that it is difficult, if not impossible, to differentiate whether the results obtained are due to alteration in quantity or quality of the milk.

Although very little appears to be known about the actual factors concerned in the production of milk, it is generally accepted that a nursing mother should have a large proportion of protein in her diet. The experiments described later tend to show that this statement needs modifying to some extent. Up to a certain limit protein is certainly beneficial, it not only promotes good growth in the young, but also helps to maintain the mother's weight and prevent excessive loss, but above that limit, decidedly harmful effects are obtained and the baby rats usually die, showing very typical symptoms.

The experiments of McCollum, Simmonds and Pitz [1916] were made on similar lines to those described in this paper, but the mothers were fed on synthetic diets. These observers showed that a maximum rate of growth in the baby rats can only be obtained when the mother is receiving a full diet, and that with a poor diet, although the litter does gain, yet the gain is not normal, and is at the expense of the mother's own tissues, as shown by the fact that her weight goes down during the lactation period. They further pointed out that there are limits beyond which the mother cannot meet the demands on her tissues and certain factors (especially fat-soluble A and water-soluble B) must be present in the diet and cannot otherwise be supplied by the mother (or only to a very slight extent for a limited time). On the whole the experiments described below agree with those of McCollum, Simmonds and Pitz, but it is shown here that the loss in weight of the mother, which is so characteristically noticed with a poor diet, can be prevented, except for an initial loss, by adding extractives to the mother's diet, in spite of the fact that it may still be inadequate in other respects. In speaking of the work of these observers one comment suggests itself with regard to the duration of their experiments. Their curves extend over a period of 24 days, but a baby rat usually begins to eat at about the 18th day and is quite capable of living without its mother from the 21st day, so that the last part of the curve may not be entirely due to the nourishment obtained from the mother.

On the other hand the babies might have been prevented from eating for themselves, but this would hardly be working under normal conditions and would put an extra strain on the mother rat.

Hart, Nelson and Pitz [1918] proved that the mammary gland has not the power to synthesise lysine, and that as far as proteins are concerned, the milk secretion is ultimately dependent upon the quality and quantity of amino-acids ingested in the food. The actual dependence of the milk secretion on the food ingested is also noticeable in the experiments described later, in which the diet is changed during the lactation period, and an immediate effect on the rate of growth of the young can be seen.

These observers further state that the average time taken by a young rat to double its birth weight is 14 days, but they do not support their statement by experimental evidence.

On the other hand McCollum, Simmonds and Pitz show by actual weight curves that the time taken is much shorter, viz. 7 days with a full diet and 8-9 days on a deficient diet. This latter fact is in accordance with results of experiments described below, which are also based on actual figures. Therefore it seems permissible to say that the time taken to double the weight of a new-born rat is 7-9 days and not 14 as stated by Hart, Nelson and Pitz.

A great deal of work has been done on the composition of milk as varied by different diets, but in all cases larger animals were used and consequently a certain quantity of milk could be obtained and analysed. In experiments such as are described in this paper, it is of course impossible to analyse the milk, and its quality can only be surmised from the rate of growth of the young.

Voit's experiments on a bitch showed that the composition of the milk secretion only varied slightly when the mother received different diets, such as meat alone, meat and starch, meat and fat, starch alone, fat alone and also when she was starved. However the composition of the milk did vary to a slight extent and this variation might possibly have been greater had the animal been fed on the same diet for more than 2-3 days.

Lusk [1917] fed a goat on a diet of hay, cornneal and bran, then starved it for two days, after which it was fed again on the original diet. He found that when the animal was starving the fat content of its milk was greater than normal and nearer in quantity to that found in the milk of the dog. When the goat received its normal diet again, the fat content of the milk was reduced.

It is somewhat difficult to correlate experiments such as are described in this paper with known facts about dairy feeding, because the object in view is different. When feeding animals on the commercial scale, it is necessary to consider what foods produce the greatest quantity of milk and butter fat for the least cost. Although the amount of butter fat and the amount of protein in the milk are generally closely related, at any rate in some animals, yet this may not always be the case.

Again it is difficult to draw conclusions with regard to human milk secretion from results obtained by feeding cows, since the metabolism of a herbivore must be different from that of an omnivore.

It seems quite possible that a milk which is highly suitable for cheese or butter-making might not be so suitable for rearing the young.

In order to obtain an abundant milk secretion, farmers have found it necessary to consider the palatability of the food and this factor has probably the same significance with all animals. The experiments described later show that in many cases the young grow better when their mother eats more of any given food and therefore the question of palatability must certainly have a bearing on this question.

Eckles [1911] states that in cows it is almost impossible to restore the milk flow to its original amount after once allowing it to run down from lack of food. With rats, however, it appears that a good diet after a poor one succeeds in pulling up the milk secretion practically to a maximum.

Eckles also points out that with cows, if the ration is changed suddenly from grain to grass, a peculiar taste is noticed in the milk, but this is scarcely detected if the ration is changed by degrees; in other words, the milk supply seems to be dependent, to some extent, on the food taken at the time. This fact is also shown by altering the diet of the rat, as a result of which a difference is at once evident in the growth curve of the young.

Eckles further states that for a heavy-milking cow, the food must be concentrated, since sufficient grass to provide material for the secretion cannot be eaten and, therefore, grain must be supplied in addition.

On the whole farmers think that diets rich in protein are the best for getting a good supply of milk and this is certainly supported by the experiments done on rats, provided the ration does not contain a large excess of protein.

A well-balanced diet is recommended for cows and it is evident from the experiments described later that this is an important factor. It is found with rats that butter may have practically no effect when fed with bread only, but may be quite beneficial if fed with a better, more mixed type of diet.

A heavy milking cow usually loses weight for 2-3 weeks (and sometimes more) after the birth of the calf, probably because she is not taking enough food for all her requirements. This feature is also exhibited in the curves shown later. Many of the rats lost a considerable amount of weight for the first few days after the birth of the litter, and for about 48 hours after the birth they rarely ate more than the average rat.

This loss of weight of the mothers may be due to one of the following factors:

1. The energy of the diet might be insufficient to supply food for the young and to replace the mother's heat loss.

2. The diet might be adequate from the energy standpoint, but lacking in certain necessary constituents, which are then supplied from the mother's own tissues, thus causing a loss of weight.

This latter supposition is supported by an experiment of Hart and Humphrey [1915]. They fed a cow on a food with a "nutritive ration" of 1:8, i.e. one part of protein to eight parts of carbohydrate and fat. Provided the protein given was milk protein, this diet gave a positive nitrogen balance, but if wheat protein was substituted a negative nitrogen balance resulted. The amount of protein in the milk was unchanged and the loss of maternal nitrogen must have resulted from tissue breakdown, consequent on the lower biological status of wheat protein as compared with milk protein.

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II. METHODS EMPLOYED.

The actual experiment was started as soon as the litter was born. In the majority of cases the babies were born in the night, but a litter born during the day was not weighed until the next morning, because it was found desirable not to disturb the mother too soon.

During the gestation period the females were fed on an ordinary mixed diet, consisting chiefly of bread, with small amounts of milk and green and other vegetables. They were removed from the males a few days before the birth of the litter. The number in each litter was always reduced to six. The mothers and litters were weighed every day, as nearly as possible at the same time and after weighing they were given their 24 hours' rations. It was impossible to give definite and equal amounts to the various rats, because their appetites exhibited great differences, and what was ample for one rat proved to be starvation diet for another. Hence the method was adopted of keeping the proportions of the constituents of the food constant and allowing the rats to eat what they liked, the amount eaten, of course, being noted. If a rat ate all its food, then the diet was increased the next day. In this way, with a little experience it was easy to gauge the animals' appetites, and feed them so that only small quantities were left each day. It seemed essential that they should have sufficient to satisfy them, otherwise abnormal curves were obtained for the litters, even though the mother was given a very good diet, such as bread and milk (which will later be shown to be one of the best diets for a nursing rat).

Bread alone was taken as the poorest type of diet. In every case, except where 100 cc. milk were given, the animals were allowed water to drink.

It was found that 15.0 g. bread was enough to start with and the amount was increased as necessary. When other foods were to be given, they were mixed with the bread, so that an even mixture was obtained. The bread etc., was always slightly moistened with warm water, because the animals would not eat much dry food, and it was also advantageous because they did not scatter the paste thus made about their cages, as they did dry food. The bread ration was increased by 5.0 g. per diem when necessary and the other foods in corresponding proportions.

The chief diets used were:

Various combinations of the above were also made, e.g. bread, butter and meat extract.

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The day on which the babies were first seen eating, generally the 18th, is indicated by * on the curves. The mothers were removed on the 21st day, unless the babies did not eat at the usual time, in which case the mother was allowed to remain three days after the babies began to eat.

III. EFFECT OF VARIATION IN THE MOTHER'S DIET ON THE GROWTH OF THE LITTER.

(a) General conditions.

Considerable differences were shown in the weights of the various litters. In some cases the average gain per day was about 4-5 g., while in others it was as much as 11-12 g. It might be expected that the daily gain would be greater towards the end of the lactation period than at the beginning, but this was not always the case. On a good full diet, e.g. bread and milk, there was a slight actual increase in weight gained per day towards the end of the lactation period, tending to keep the percentage increase constant, but with a poor diet, e.g. bread only, the actual increase in weight per day was usually greater at the beginning than at the end of the period. This latter fact seems what would be expected because such a diet is lacking in certain constituents which are normally present in milk and, it may be inferred, were present in the milk of these rats, as the litters were normally healthy, but small. The constituents are probably supplied from the mother's tissues. Therefore towards the end of the lactation period it is probable that the available reserves of these special substances are coming to an end and consequently the babies do not gain as much as previously. In keeping with this explanation is the fact shown in the curves that the mothers themselves lose weight on poor diets. On other poor diets and especially those rich in extractives there seems to be a fairly uniform daily gain in the weights of the litters and the mothers' weights remain constant.

There are a few remarks which apply generally to the curves. Later they will be treated separately.

1. The relation of the milk secretion to the amount of food taken by the mother.

In the majority of cases, given any definite diet, a litter belonging to a mother which ate considerably more than another, did better than the litter of a mother which ate less.

2. Relation of milk secretion and age and weight of mother.

The age and weight of the mother seem to be unimportant factors. In some instances the heavier rats brought up their litters better, *i.e.* curves of the young were better but this was by no means a general rule. In some cases a first litter did not grow as rapidly as the later ones, but this again is not always true.

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3. Condition of litters.

(a) At birth. Litters of very low weight *i.e.* about 20.0 g. for six (normal weight of six is about 30.0 g.) were generally weakly, but with a good mother they usually pulled up after a few days In a few cases the six weighed a small amount and yet did well. This was when they belonged to exceptionally large litters and this fact alone accounted for their low weight. Also such babies seemed physically fit, while weakly litters were easily detected by their general appearance as well as low weight.

(b) During lactation. In all cases (except where excess protein in the form of caseinogen was given) the litters were healthy and the babies quite strong and able to look after themselves when separated from their mothers, but the large differences in weight were parallelled by differences in size, e.g. a 10-11 days' "bread and milk" baby was often about the size of a 20-21 days' "bread" baby. With the poorer diets, especially bread only, bread and butter, bread and dextrin, the babies' coats were rather thin, more like an animal's summer coat. An exception to this statement was found in the case of bread and meat extract where the babies had a normal coat, rather thick and silky. All the babies (except when the mother was given excess protein, as before mentioned) were able to run about at the usual age. Those belonging to the less well-fed mothers were, if anything, more lively than the fatter litters of the well-fed mothers. The babies belonging to the rats fed on (1) meat extract and bread, (2) meat extract, bread and butter were specially active. The general good health and activity of the young rats, even when the mother's diet is deficient and their own weight small, indicate that it is the quantity rather than the quality of the milk which suffers as the result of defective feeding in the mother.

(b) Individual diets.

The majority of the figures consist of two sets of curves:

1. The mothers' weights.

2. The weights of the litters.

The corresponding mother and babies are indicated by the same symbols. At least three experiments were made with each diet, and an average growth curve given in Fig. 6.

1. Bread only. Basal diet. Water to drink.

The diet consisted of white bread, no crust being used. It was slightly moistened with water to prevent it from getting too dry. An initial ration of 15.0 g. per diem was given and this was increased by 5.0 g. at a time when necessary.

In each experiment the mothers lost a considerable amount of weight and their curves show a varied loss throughout the lactation period. A large initial drop in weight was shown by two of the three animals and this is typical of many other experiments. The litters show a steady but not large daily increase, which gradually gets less towards the end of the lactation period. Roughly, the curve might be divided into three parts, the first showing a daily increase of about 4.5 g., the second of 3.6 g., and the third of 2.2 g., after which the babies began to eat for themselves (see Fig. 6 and table on p. 158).

This diet consisted chiefly of carbohydrate in the form of starch and protein. It was, therefore, partially deficient in fat, fat-soluble A, water-soluble B, and the anti-scorbutic factor. Since the young rats were healthy throughout and showed normal activity, it is probable that the mother can supply these deficiencies from her tissues. And, also, as the daily gain gradually becomes less, it might be inferred that the mother's tissue supplies cannot entirely meet the demands made on them.

It is seen from the figures given below that I at more than either of the other rats and her litter showed the best growth; further, L at more than Y and the L babies did better than the Y babies.

I ate 520 g. bread in 17 days L ,, 500 ,, ,, 19 ,, Y ,, 370 ,, ,, 18 ,,

Result. The rat can bring up a healthy litter, normal in all respects except size, on a diet deficient in fat and vitamines. The mothers lose weight throughout the lactation period (see tables on p. 158).

2. Effect of adding meat extract to basal diet. Water to drink.

The initial diet consisted of 15.0 g. bread and 2.0 g. meat extract (a commercial preparation "Jardox" was used). The extract was dissolved in hot water, poured over the bread and an even mixture made. When necessary the diet was increased by 5.0 g. bread and a proportionate amount of meat extract added. This diet is slightly better than the basal diet, but is again deficient in fat and probably in vitamines as well. Nevertheless there are remarkable differences in the results.

The most noticeable feature when meat extract is given, is that the mothers' weights show very little or no loss, and in one case a decided gain was exhibited throughout the lactation period. Also the initial loss in the mothers' weights (pointed out previously when discussing the bread diet) is absent in two of the three animals. In addition the mothers show a steadier weight curve than that usually obtained, when the animals frequently lose or gain about 10.0 g. per day.

The greater increase in growth of the litter as a result of the mother's eating more is again illustrated.

 3 ate 490 g. bread and 66 g. meat extract in 18 days

 6 ,, 490 ,, ,, 66 ,, ,, 18

 W ,, 460 ,, ,, 74 ,, ,, 17

The litters show a more even increase in rate of growth throughout the lactation period and do not exhibit the slower growth towards the end of the

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period, as shown by litters whose mothers received only bread (see Fig. 6 and table on p. 158).

Result. When extractives are added to the basal diet, the mothers lose very little, if any, weight and their curves are much more regular. As compared with bread diet the milk secretion is slightly improved, more especially at the end of lactation and the litters gain practically equal amounts throughout the lactation period.

3. The effect of additional carbohydrate in the diet. Water to drink.

The initial ration consisted of the usual 15.0 g. bread to which 5.0 g. dextrin were added. The dextrin was dissolved in a small quantity of hot water, the bread added and an even mixture made.

The curves representing the rate of growth of the litters are practically identical with those of litters whose mothers fed on bread only. From this it appears that an excess of carbohydrate in the diet makes no difference to the milk supply of the mother, which is very different from the results obtained when excess protein was given to the mother (see curves in Figs. 1, 2, 3 to be described later).

The mothers' curves also are very similar to those of the rats fed on bread only, a typical loss in weight being again a characteristic (see table on p. 158).

Result. Additional carbohydrate in the form of dextrin has no effect on the milk secretion.

4. Effect of adding fat and fat-soluble A to basal diet. Water to drink.

Two types of diet were used in these experiments:

(a) 15.0 g. bread, and 2.0 g. butter, for initial diet.

(b) ,, ,, 5.0 g. ,,

The bread was moistened with hot water, the butter melted and an even mixture made.

The curves of the growth of the litters are again similar to those obtained when the mothers were given a bread diet.

From the experiments made under these conditions, it seems that the extra fat has a slightly depressing effect on the curves of the litters, but this may be an indirect effect, because it was found that with a fatty diet of any kind, the mothers tended to eat less than the normal amount. In such a diet the calorific value was not appreciably depleted, but the animals were getting less protein.

These experiments suggest that fat is an unimportant factor in the question of milk production.

The curves of Fig. 2 suggest that fat may play a more important part if more protein is given, because litters K and M in the upward parts of their curves did better than litter O, the mother in this case receiving no fat in her diet. The significance of this is by no means clear and the point awaits further investigation.

Result. When fat and fat-soluble A are added to the basal diet, the mothers lose in weight as on the basal diet and the rate of growth of the litters is practically the same as when the mothers are fed on bread alone (table, p. 158).

5. The effect of adding milk to the basal diet.

The milk ration was kept constant at 100 cc. per diem. An initial amount of $15 \cdot 0$ g. bread was given and increased by $5 \cdot 0$ g. as in previous experiments. This is decidedly the best diet used as it contains all the necessary factors. The daily gain of the litter exceeded that obtained with any other diet (see Fig. 6, curve D). When milk was given to the mothers it was noticeable that the actual daily increase in weight of the litters became greater towards the end of the lactation period, the exact opposite of what was found with poor diets, *e.g.* bread, or bread and dextrin, or bread and butter. The mothers' curves were somewhat irregular but speaking generally, a loss in weight was followed by a gain, so that there was no very great permanent loss.

Result. Milk added to the basal diet forms a most efficient food and the litters show maximum gains in weight (table, p. 158).

6. (a) Effect of adding meat to basal diet. Water to drink.

Equal weights of bread and meat were given. The bread was moistened with water as before, the meat minced finely (after removal of fat) and an even mixture made.

The mothers' curves are characterised by gain rather than by loss of weight. The litters gained practically equal amounts daily throughout the lactation period. This result is similar to that described with respect to those litters whose mothers were fed on bread and meat extract. The addition of meat to the bread had an obviously beneficial effect on the secretion of milk (see Fig. 6, curve 2).

Result. Meat added to the basal diet of bread promoted rapid growth of the young, but not so marked as when the mothers were fed on bread and milk (table, p. 158).

(b) Effect of feeding with lean meat only. Water to drink.

This diet was not so good as that of bread and meat, possibly because the carbohydrate was insufficient for the effective metabolism of the protein taken. The mothers lost weight and the daily gain of the litters was less than half that of the litters whose mothers received bread and meat (see Fig. 6, curve 10 and tables on p. 158).

When fed on bread and meat or meat only the rats ate large quantities, e.g. 30.0 g. meat plus 30.0 g. bread, and 60.0 g. meat. These are exceptionally large amounts considering that the rats were by no means fully grown and only weighed 190.0 and 150.0 g. respectively. Even then, when eating meat only, the rats would not be getting an energy value equivalent to that of the basal diet. The average amount eaten per day on a meat diet was $45 \cdot 0$ g. and on a bread diet the average consumption was about $28 \cdot 0$ g. per day. The energy value of $45 \cdot 0$ g. meat is about $460 \cdot 0$, while that of $28 \cdot 0$ g. bread is $690 \cdot 0$. Hence on a meat diet the mother may not be able to eat sufficient food to get the necessary amount of energy for a good milk secretion.

Result. Meat only is not a diet suitable for producing a good mammary secretion, possibly because the energy of the food is too little to allow for the supply of a good milk secretion.

(c) The effect of adding bread to a diet consisting entirely of meat. Water to drink.

A rat was fed entirely on meat for nine days of the lactation period and then her diet was changed to an equal mixture of bread and meat. When the mother was eating meat alone, the litter gained an average of 4.0 g. per day, but when her diet was altered they gained on an average 6.8 g. per day. She lost less weight on the mixed diet and after a few days even showed a tendency to put on weight. The question of change of diet during the lactation period is discussed more fully later.

7. The effect of adding butter and meat extract to basal diet. Water to drink.

The proportions used were 15.0 g. bread, 2.0 g. butter and 2.0 g. meat extract (commercial preparation "Jardox"). The meat extract was dissolved in sufficient hot water to moisten the bread. The butter was melted and the whole made into an even mixture.

An interesting point arises here in connection with the mothers' weights. In other experiments, where meat extractives were fed, the mothers did not lose weight to any appreciable degree, but either maintained or slightly increased their original weight. In these earlier described experiments, no fat was given, while in the ones now being discussed, two or more g. of fat were taken per day. The action of the fat appears to inhibit that of the extractives since the mothers lost weight to about the same extent as when they were fed on the basal diet of bread only. A further point is brought out by these results. One mother lost more weight than either of the others which received the same diet. At the same time her litter gave a better growth curve, the improvement being greater than could be accounted for by the extra food eaten by their mother. Similar results were obtained in other experiments, which might be taken to indicate a connection between loss of weight in the mother and gain of weight in the young.

However, the experimental evidence is insufficient to allow of a definite statement either with regard to the metabolism of simultaneously ingested butter and extractives, or the mutual relation between the weights of the mother and her offspring.

Result. When fat is added to a diet of bread and meat extract it does not have any marked stimulating effect on the production of milk and the litters

do not show any great increase in weight (table, p. 158). On the other hand the addition of fat now brings about loss in the mother's weight, which is not evident in its absence (table, p. 158).

8. The effect of excess protein in the mother's diet (Figs. 1, 2, 3).

The protein employed was pure caseinogen used in various diets described below.

The results were striking, the litters did well for a time, but eventually died, showing typical symptoms. In most cases they gained weight normally at first, then ceased to gain, finally lost weight and died. The first indication that something was wrong with the babies was that they became very excitable, and when taken out to be weighed, kicked and struggled, whereas normal babies would have kept fairly still. The age at which the symptoms developed varied in the different litters according to the diet chosen, but all exhibited them sooner or later. About two days after the excitability was first noticed, the babies usually showed signs of inability to crawl normally and had a marked tendency to drag one hind leg and roll over towards that side.

Later extensor and contractor spasms were evident and in very bad stages the young rats dashed madly round their cages, bit each other and their mother and knocked themselves wildly against the sides of the cage. The fits generally lasted 3-5 minutes, after which the babies were exhausted and slept.

If the symptoms were not really bad until the babies were about 18 days old, they frequently recovered, if they ate for themselves. When the babies died at about 15–16 days, death was always preceded by extreme weakness and they had the appearance of being starved, although starvation alone could not cause these symptoms. On examination their alimentary canals were found to be practically empty, which naturally suggested interference with the milk supply of the mother. Accordingly the mother was killed and the mammary glands were found to contain no milk, but only a small quantity of a lymph-like substance, while the glands of a well-fed mother contained a large amount of milk, which oozed out directly the tissue was cut. Thus it appears that excess protein in the diet first causes the production of abnormal milk and then stops the milk supply.

The mothers appeared to be normally healthy but in some cases developed diarrhoea to a slighter or greater extent and the faeces were light in colour.

This question of the relationship between protein metabolism and milk secretion arose purely incidentally in the experiments. Its importance was at once recognised and therefore experiments to elucidate the problem were begun and are still in progress.

The mothers gained noticeably in weight and remained apparently in good health even when the young all died. Whether an excess of protein in the diet has any relation to the development of fits in suckling children or even to the abnormal excitement and restlessness that is often seen in such children, remains to be examined by further investigation.

(a) Effect of adding excess caseinogen to basal diet. Water to drink (Fig. 1).

The proportions used were 15.0 g. bread and 5.0 g. caseinogen. The bread was well moistened and thoroughly mixed with the caseinogen.

The litter P did not develop the symptoms early, possibly because their mother ate less caseinogen than N or T. However three died; probably the others survived because they ate for themselves.



Litter N all died, in spite of the fact that the mother's diet was changed to bread and milk on the 13th day when the symptoms became evident. It may be that the milk supply started again as found in experiment O (p. 154), but the babies were too weak to suck.

Litter T showed symptoms similar to those of litter N, one was found dead on the 17th day, two more died that day and the remainder were killed together with their mother. The babies' stomachs were empty and their intestines almost so. The mother appeared normal, except that on cutting the mammary glands, no milk oozed out, but only a thin lymph-like secretion.

Result. Addition of excess caseinogen to the basal diet causes the mother to put on weight, but the milk supply is affected in such a way that the litters do well up to the 10th day, after which they cease to gain weight and finally die. It appears that the quality of the mother's milk, after being very good, is detrimentally altered before being completely suppressed.

(b) Effect of adding caseinogen to other diets. Water to drink (Fig. 2). The diets given were:





The yeast extract used was "Marmite" and the foods were mixed as before described.

The mothers' weights again show a decided increase. The weighings of M were discontinued because she was found to be pregnant.

With regard to the litters, all the K babies died, in spite of the fact that the mother's diet was changed. It seems probable, as suggested previously, that the milk supply was not re-established until the babies were too weak to suck.

In the case of M the diet was changed to bread and milk before the symptoms became very severe, and as shown by the curve, the babies recovered. Later they were given to a foster mother as their own mother was pregnant and there was the possibility of the birth of another litter. (This second litter proved to be quite normal in every respect.) It may be that the rat's becoming pregnant again, while no others did so, is a factor which must be taken into account, but the work is insufficient to warrant any suggestions upon this point.

Litter O all developed the typical symptoms; one was dead on the 15th day and the remaining five died the next day. The mother O was killed on the 16th day (24 hours after the diet had been changed to bread and milk) and her mammary glands contained a small quantity of milk which led to the conclusion that the milk supply was just being restarted. The babies, like those of litter N, had nothing in their stomachs and very little in their intestines.

Result. The milk supply of the mother is stopped by excess caseinogen in any diet, but the secretion can be re-established by changing the diet to bread and milk.

> (c) Comparison of bread and milk diet with one containing excess caseinogen (Fig. 3).

The diets used were:

1. 100 cc. milk, bread 15.0 g. (increased when necessary) ... Exp. D

2. Bread 15.0 g., caseinogen 5.0 g., meat extract 2.0 g., butter 2.0 g. ... Exp. K

An interesting point is brought out by this experiment. The two curves run parallel until the 10th day. This is a curious fact for it is to be remembered that bread and milk diet gives the best growth curve. After the 10th day, the litter K ceased to gain weight and finally died. Possibly a similar diet to that used in experiment K, *i.e.* bread, caseinogen, meat extract and butter, would give a maximum growth curve, if less caseinogen were used. It is hoped to work out this point later.

9. (a) Comparison of animal and yeast extractives (Figs. 4, 5).

The diets used were:

A.	1.	Meat	extract,	bread,	caseinogen,	butter	Exp. K) Fig. 4
	2.	Yeast	, , ,	,,	99 ,	,,	Exp. M/ 11g. 4.
В.	1.	Meat	extract,	bread	and butter	•••	Exp. I)
	2.	Yeast			55		Exp. A



The diets were mixed as before described. The meat and yeast extracts used were the commercial preparations "Jardox" and "Marmite" respectively.

The work on this question is scanty, but tends to show that the growth curves of the young are better when the mother receives animal extractives, than when she is given yeast extractives in her diet.



(b) Comparison of various diets in relation to growth of litters (Fig. 6).

In the following comparisons typical curves taken from the previously described experiments are used (see p. 158).

- 1. Comparison of (a) bread (basal diet),
 - (b) bread and fat,
 - (c) bread and additional carbohydrate.

The growth curves obtained when the mothers were fed on the above diets were almost identical. Hence fat, fat-soluble A and extra carbohydrate added to a poor diet appear to have no effect on the mammary secretion. The mothers show a distinct loss in weight during the lactation period.

2. Comparison of (a) bread (basal diet),

- (b) bread and meat extract,
- (c) bread and meat extract and fat.



The typical "bread curve" is compared with curves obtained by feeding the mothers on bread and meat extract, and bread, meat extract, and butter respectively. This shows that better results are obtained with the improvement in diet and also that (as pointed out previously) butter has not very much effect. The difference in growth is so slight that it could even be obtained using the same diet, and it may be concluded that a diet of bread and meat extract is practically equivalent to a similar one with butter added so far as the secretion of milk is concerned.

- 3. Comparison of (a) bread (basal diet),
 - (b) bread and meat,
 - (c) bread and meat extract,
 - (d) bread and milk,
 - (e) meat only.

Fig. 6 represents a comparison of the typical growth curves obtained with widely different diets. To avoid complication in the figure, bread is taken as representing the curves of bread, bread and fat, and bread and additional carbohydrate, previously described as almost identical. Similarly bread, meat extract and fat is omitted owing to its resemblance to bread and meat extract with regard to effect on mammary secretion.

Comparing the diets in this way accentuates the fact that differences in the curves are greater towards the end of the lactation period, than at the beginning, e.g. the curves of (D) (bread and milk) and (2) (bread and meat) run together till the 9th day and then gradually diverge. Again the curve of L (bread only) runs parallel with that of (3) (bread and meat extract) until about the 7th day after which the extractives produce a better effect.

Table	showing	relative	gain in	weight	of	litter	when
	mothe	ers recei	ve differ	ent die	ts.		

		Average gain of litters			
Mother's diet	Days 1-7		Days 7-14	Days 14—eating	
		g.	g.	g.	
Bread only (basal diet)	•••	4.5	3.6	2.2	
Bread and butter		4 ·3	3.06	2.5	
Bread and dextrin		4.5	3.25	2.0	
Bread and meat extract	•••	5.1	5.25	5.0	
Bread, meat extract and butter	•••	5.3	6.1	4.7	
Bread and meat	•••	8.3	8.66	9.0	
Meat only	•••	3.5	3.2	4.5	
Bread and milk	•••	9.0	10.5	11.0	
Bread, caseinogen, butter, meat ext	ract	8.0	5.3	-8.0	

Table showing initial and final weights of litters and mothers, when latter received different diets.

	Weig	hts of litters	Weights of mothers		
Mother's diet	At birth	When eating for themselves	After birth of litter	When litter eating	
	g.	g.	g.	g.	
Bread only (basal diet)	30	90	120	105	
Bread and butter	30	. 85	148	125 _	
Bread and dextrin	30	88	173	144	
Bread and meat extract	33	114	132	132	
Bread, meat extract and butter	28	122	176	168	
Bread and meat	26	143	140	140	
Meat only	24	81	· 168	148	
Bread and milk	35	185	163	161	

(c) The effect of change of diet during the lactation period (Fig. 7).

The effect of changing the diet during the lactation period is most striking. It is obtained quickly, the difference in daily gain being noticeable within 24 hours after changing the food. The curve G represents the typical curve of a bread and milk diet and L that of a bread diet.



In experiment U the mother was fed on bread and milk for the first eight days and then the diet was changed to bread only. A decrease in the daily gain of the litter was noticed 24 hours after the change of diet (see Fig. 7).

In experiment S the converse was tried, *i.e.* bread for the first period, changing to bread and milk later and in this case the improvement in diet produced a corresponding increase in the daily gain of the litter.

It is interesting to note how the parts of the curves due to a bread diet run parallel with the typical bread curve, and those due to bread and milk diet with the typical bread and milk curve. This experiment indicates that it makes no difference whether the good diet be given at the beginning or the end of the lactation, since the milk supply is largely dependent on the daily intake of food.

This point was also noticed on changing from a diet composed entirely of meat to one of bread and meat.

IV. THE SUPPLY OF ESSENTIAL SUBSTANCES FROM THE MOTHER'S TISSUES (Fig. 8).

Since the mothers receiving such inadequate diets as bread only, bread and dextrin etc., were able to bring up healthy litters, normal in all respects except that they were smaller than those of litters whose mothers received good diets, it seems evident that the mothers can supply various constituents for their milk from their own tissues.

The adequacy of the different diets was tested by keeping the young (after removal of the mother) on the same diets used during the lactation period. The results of these experiments are shown in Fig. 8. The day on which the babies began to eat for themselves is indicated by * and the removal of the mother is shown by an arrow.

From this diagram it is quite evident that with a full diet, *e.g.* bread and milk, bread and meat, the young will continue to grow at a fair rate after their mother has been taken away. On the other hand, with poor diets, the young rats gained weight very slowly after their mother had been removed.

From these "poor diet" results, it appears probable that the mother is able to supply certain food constituents absent from the diet.

The supply is cut short when the mother is removed and the young rats at once show a decline in the rate of growth.

Bread diet. Experiment L:

Baby rats gained 60 g. in 18 days when fed by mother.

,, ,, 24 g., 11 ,, ,, eating bread.

Bread and fat diet. Experiment Q:

Baby rats gained 68 g. in 17 days when fed by mother.

" " 18 g. " 14 " " eating bread and butter.

These experiments indicate that the mother can furnish from her own tissues, fat, fat-soluble A, water-soluble B, and the anti-scorbutic factor.



V. GENERAL CONCLUSIONS.

1. With certain typical diets, definite and different curves can be obtained for the rate of growth of the litters which obviously are an indication of differences either in quantity or quality or both of the mother's secretion of milk.

When the mother is given a good diet, *e.g.* bread and whole milk, the baby rats at the end of lactation weigh twice as much as when she receives a poor diet, *e.g.* bread only.

2. The loss or gain in the weight of the mother is not definite, but liable to great variation. Speaking generally the mother loses less weight as the diet contains more physiologically good elements. In the majority of cases the mother shows a typical initial loss of weight at the beginning of the lactation period.

3. Extractives tend to keep up the mother's weight. Protein has a similar effect.

4. Excess protein in the mother's diet probably alters the composition of the milk and after a time causes the milk supply to cease. On altering the diet to bread and milk, the secretion is again established.

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5. Excess carbohydrate appears to have no effect at all.

6. Excess fat in the mother's diet has a slightly depressing effect on the growth of the litter, but the absence of fat seems to make practically no difference.

7. On any given diet, the litter belonging to a rat which eats a large amount, does better than one the mother of which eats a less amount.

8. The mother can supply certain essential substances from her own tissues, viz. fat, fat-soluble A, water-soluble B and the anti-scorbutic factor during the normal period of lactation.

9. The milk supply to a large extent is dependent on the food taken and can be increased or diminished within 24 hours by changing the diet.

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