

a different chromosomal number. That this may have been the case is suggested by the fact that we have found a single plant which was markedly abnormal throughout and distinct from the $(2n-1)$ branches previously investigated, but which was found also to lack one of the same Rolled chromosomes.

(b) *Chimeras with chromosome excess.* A plant otherwise normal, has been found with one branch bearing leaves and capsules which resembled the $(2n+1)$ Globe mutant. Chromosome counts have not yet been secured; but offspring from the normal branches were normal, while offspring from the abnormal branch showed the proportion of Globe seedlings expected from Globe parents. The evidence is clear, therefore, that the subepidermal tissue of the abnormal branch of this chimera was $(2n+1)$ with the extra chromosome in the Globe set. That the epidermal tissue was possibly of a different chromosomal constitution is suggested by the fact that neither the leaves nor capsules on the abnormal branch were fully typical for Globe characters.

(c) *Chimeras with doubled chromosome number.* Several cases have been found chiefly after treatment with cold, in which a single branch on an otherwise normal $2n$ plant has shown resemblances to a tetraploid. Growth and bud formation in these cases has been poor, but these abnormal branches have been shown to be $4n$ in generative tissue by the sizes of their pollen grains as well as by the tetraploid offspring which they have produced as contrasted with the $2n$ offspring produced by the normal branches.

Other and possibly more complicated chimeras which may have a basis in differences in chromosome number are under investigation. The evidence already obtained, however, is sufficient to indicate that chromosomal aberrations may be an important cause in the production of bud sports.

Figures and a more detailed description of the chromosomal chimeras mentioned in the present paper will appear shortly in the *Journal of Heredity*.

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UNIQUE DIETARY NEEDS FOR LACTATION¹

INVESTIGATION of the dependence of specific bodily functions upon specific nutritive elements is possible because the maintenance of life and, indeed, approximately normal growth are independent of some of those functions. Animals may be reared and will

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continue to live indefinitely despite disorder, for instance, of the osseous system. It is a matter of practical knowledge centuries old that reproduction may be normal in animals and the function of lactation subnormal or held in abeyance. It has been possible for us to show that active, normal sized and normal appearing rats may be reared by dietary régimes which sterilize them. It has, furthermore, been possible to show that this "dietary" sterility is due to the absence of minute quantities of a specific so-called vitamine substance X, the stability, solubilities and other characteristics of which have now been studied.² The commonest dietary régime employed by us in such studies consisted of a well-known mixture of "pure" or isolated foodstuffs (casein 18, cornstarch 54, lard 15, salts 4)³ together with an abundance of the growth vitamins A and B in the form of butter and yeast. The butter constitutes 9 per cent. of the ration, but the yeast must be fed daily separately in a dose of from 400 to 600 milligrams.

Work with this basal ration of pure food and an abundance of the vitamins at present known should enable us to detect whether or not the function of lactation has other and special dietary dependencies. It is clear that hitherto one could not have amassed the requisite data for such study, since animals do not usually reproduce upon such synthetic mixtures. The detection of the vitamine substance X makes it possible to convey minute but adequate amounts of this substance to animals upon the classic pure food régime and to secure at will reproduction on the régime and to study lactation upon it. Other studies, moreover, have shown that at least one of the foods involved in the pure ration itself, namely, milk fat, possesses seasonally a sufficient contamination with vitamine X to enable animals reared upon this régime to have their first litters. This fact has unfortunately led certain workers to deny the existence of a vitamine which determines reproduction. We have designated this phenomenon "first litter fertility," for upon the same régime the same animals are subsequently sterile. It is due, we believe, to a low amount of dietary vitamine X augmented by

² Evans and Bishop, *SCIENCE*, Vol. 56, p. 650, Dec. 8, 1922; *Jour. of Metabolic Research*, Vol. 3, No. 2, Feb., 1923; and Evans and Bishop and Evans and Burr, *Proc. Amer. Assoc. Anat., Anatomical Record*, Vol. 27, No. 4, April, 1924.

³ Salts. The salt mixture employed was identical with that used by E. V. McCollum and consisted of

NaCl	0.173
MgSO ₄ (anhyd)	0.266
NaH ₂ PO ₄ + H ₂ O	0.347
CaH ₄ (PO ₄) ₂ + H ₂ O	0.540
Fe citrate	0.118
Ca lactate	1.300
K ₂ HPO ₄	0.954

low but constant amounts of X already in the tissues of young animals, since we have proved that the substance is transmitted to them in intrauterine life.

These cases of first litter fertility in animals upon the basal "pure" diet have proved of great value to us in our studies upon lactation. Furthermore, in the cure of sterile animals by the administration of alcoholic or ether extracts of a food substance high in X—wheat germ—it has been possible to study the lactation of animals with induced fertility upon the "pure" régime, modified by the insignificant addition involved in a minute amount (one to six drops) of wheat germ oil daily. About 100 young have been weaned from mothers showing first litter fertility on the basal régime and over 300 have been suckled by mothers likewise on the basal ration but whose fertility was provoked by minute doses of the lipoids found in the embryo of wheat. A singular correspondence obtains in the results secured with both of these large groups. *Lactation is always seriously impaired upon the "pure" diets.* The average weaning weight of the animals resulting from such lactations is almost exactly half that which is normal, i.e., 20 grams instead of 40 grams on the twenty-first day of life. Distribution graphs of the actual weaning weights in the two groups show no overlap (Figures 1 and 2). In practically none of the pure food young is a normal weaning weight secured. Furthermore, the mothers lose approximately five per cent. of their body weight in the performance. *It hence seems to us established that the function of lactation demands for its normal expres-*

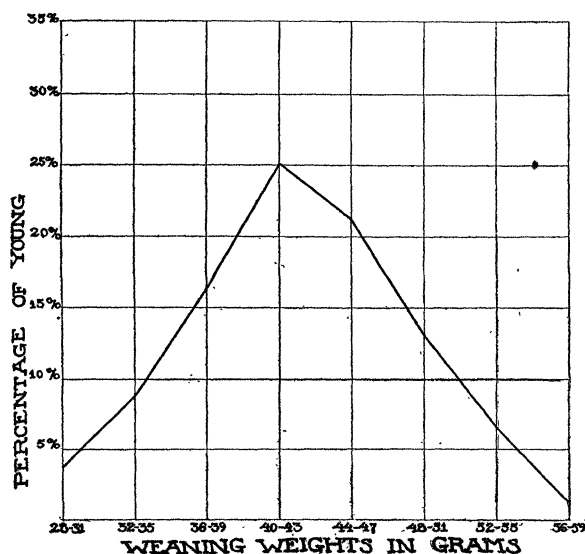


FIG. 1. Distribution of weaning weights (twenty-first day of life) of 503 rats, the mothers of which were on Standard Diet 1 (whole wheat 67, whole milk powder 10, casein 15, NaCl 1, CaCo₃ 1.5, milk fat 5).

sion either one or more dietary elements different from those adequate for normal growth and for normal reproduction, or else larger quantities of certain dietary elements.

Striking as are such results, it is conceivable that they are due to the impairment of the young. It may be stated that the new-born of rats on natural foodstuffs are slightly heavier (one gram) and apparently more vigorous than are the pure food nurslings. Some slight abnormality of the young born from "pure food" gestations might be the cause of their inability to grow well during the lactation period. But they grow normally afterwards and a conclusive answer to this contention is furnished, we believe, by a group of exchange experiments which we have instituted. As many as a hundred young from cases of sporadic fertility have now been interchanged with a hundred young from normal mothers upon a generous table scrap diet. The experiment was done in such a way that the normal mother was

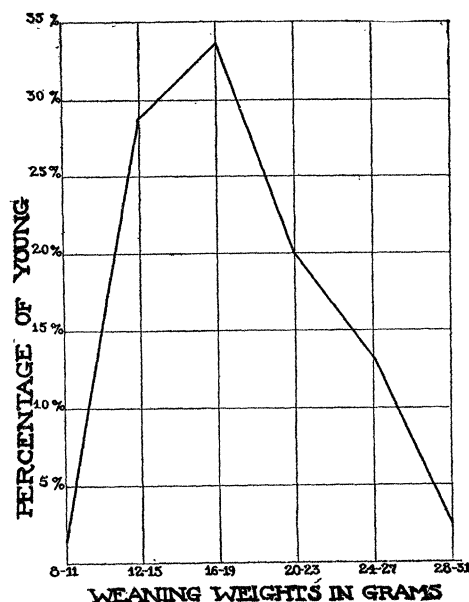


FIG. 2. Distribution of weaning weights of 134 young of mothers on Basal "Pure Food" Diet (casein 18, cornstarch 54, lard 15, milk fat 9, salts 4, yeast daily .5 gram).

forced to accommodate equal numbers of her own and of the synthetic food young, and, conversely, the synthetic food mothers were required to suckle normal young as well as their own. In all cases the litter size was reduced to six. The experiment has shown us that *the young from pure food mothers when suckled by normal mothers possess the same capacity to grow during lactation as do normal animals suckled by their own mothers*, for during the three weeks' lactation period they increase their body weight about seven times. With their

own mothers this increase would have been, roughly, four times. Conversely, normal young suckled by pure food mothers increase their body weight but four times in spite of the somewhat superior vigor with which these animals start life.

Now it is conceivable that the excellent lactatory powers of the normal animals are not actually due to the food consumed by them during the lactation period, but to "reserve" substances in their tissues which can be called upon in this unique need. An answer to this is given by the imperfect lactation resulting when an animal is shifted from a satisfactory diet to the "pure" one. The mammary stimulant must hence be an element of the food.

The natural foods therefore contain a substance, or substances, essential for the normal function of the mammary gland. We have been able to shed some light upon which particular natural foods contain and which do not contain this material, which may be variously termed the lactation auximone, or auximones, or nutritive galactagogue according to one's preference. Early in our studies various amounts of different natural foods were added to the basal pure diet in order to induce fertility, and we took occasion to study not only the function of reproduction but, when young were born, that also of lactation. Many substances (*e.g.*, vegetable oils) which induce reproduction do not improve the lactation of animals upon the basal régime; on the other hand, some of them greatly improve mammary function. Among the substances studied are leaves, grains and animal tissues. A considerable body of data has been secured with the use of lettuce, wheat, egg yolk and beef muscle. Fresh leaves in high amounts improve lactation; when dried, they do not do so. Wheat embryo does so when fed as a considerable portion of the ration. Egg yolk and meat help lactation markedly. Furthermore, the fat has been extracted from both egg yolk and wheat embryo, and in their fat-free form these foods were as effective in galactagogic action as they were formerly. It would appear, therefore, that the food material necessary for normal mammary function is not soluble in fats. Implication of a protein factor is suggested by the potency of meat and grains, even though experiments with milk itself make it seem less likely that either protein or inorganic matter constitutes the dietary substance conditioning normal mammary performance. The total milk solids (we have used as much as a third by weight of the entire ration in the form of whole milk powder) do not repair lactation delinquency so effectually as do the other substances mentioned. Yet the difference between bovine and murine milk in these very constituents robs such an argument of crucial value. Attention is being paid to the distribution

and possible isolation of the food constituent in question. It is by no means ascertained that this is not a well-known substance. A suggestion, however, that this is not the case and that we may be dealing here with one of those specific stimulants to the biochemical mechanism constituted by the so-called auximones like *bios*, is furnished by the superior effect of fresh leaves when contrasted with the same desiccated leaf substance. Be that as it may, a single conclusion—yet an important one—seems clearly validated by the "pure food" lactation studies. Food requirements for normal lactation in the rat are not fulfilled by the classical "synthetic" dietaries of fat, carbohydrate and protein, together with salts and an abundance of the known vitamins A, B, C, D and X—dietaries entirely adequate for growth and reproduction.

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(Continued)

3-amino-4-hydroxyphenylarsine: G. W. RAIZISS and B. C. FISHER. This dihydrochloride of 3, 3'-diamino-4, 4'-dihydroxyarsenobenzene, known under the names of arsphenamine and salvarsan, is a remedy of great value in medicine. Its oxidation product, the 3-amino-4-hydroxyphenylarsineoxide, possesses a still greater destructive effect upon parasites, but it is more toxic and therefore has not been used in treatment of diseases. The authors were interested to study the biological properties of the reduction product of the first-named compound and therefore it became necessary to secure the product as pure as possible. German and English patents in which the preparation of the 3-amino-4-hydroxyphenylarsine is described, when followed, gave extremely small yields and a very impure product. A systematic and prolonged study led to a satisfactory method which gave comparatively good yields and what is very important, a chemically pure product. This arsine was made from arsphenamine by the reduction with zinc dust and hydrochloric acid at 40° C. It was precipitated out by means of a saturated solution of sodium acetate. The crude product was extracted with ether, the work having been performed strictly under pure nitrogen gas. The crude product has been purified four times, dissolving it in dilute alkali and reprecipitating by acetic acid. This again was done under nitrogen. The product so obtained represents a pure white substance easily oxidizable to the yellow arsenobenzene. It is an interesting substance from the biological standpoint, being highly trypanocidal, more so than arsphenamine and the corresponding arsineoxide.

The use of silicon tetrachloride for the synthesis of acid chlorides. (By title): R. E. MONTONNA and H.