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The Effects of Synthetic Diets on Fertility and Lactation.

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The existence of a dietary complex essential for reproduction (vitamin E) first described by Evans and Bishop,[1] now corroborated by Sure,[2] Mason[3] and Matill and his associates,[4] has not been generally accepted. Evans and Bishop found that on a synthetic diet composed of casein 18, corn starch 54, lard 15, milk-fat 9, salt mixture 4, plus a daily ration of 0.4 to 0.6 gm. of yeast, rats were for the most part sterile in the first generation, and wholly so in the second. This deficiency is corrected by the addition of lettuce, wheat germ, egg-yolk, meat, and to a less extent milk-fat, or a small quantity of a fat-free concentrate prepared by extraction from wheat germ oil (Evans,[5] and Sure[6]). On the other hand, Nelson, Heller and Fulmer,[7] using a synthetic diet consisting of casein 18, salt mixture 3.7, filtered butter fat 5, yeast 1-8 and dextrine up to 100, encountered no difficulty in securing litters, although only a small percentage of the young could be reared. Likewise, Hogan, Harshaw[8] employing synthetic rations containing casein 20, milk-fat 10, cod liver oil 5, yeast 6, salts 4, corn-starch 50-55 and agar 2-5, reported no sterility in their rats, but rather failure in lactation.

From data accumulated for the past two years in this and in Professor Mendel's laboratory in New Haven, (which are summarized in Tables I and II) and from a review of the more recent

TABLE I.

Synthetic Ration Used.

Ration	Casein	Cornstarch	Salts*	Yeast	Butter-fat	Lard	Cod liver oil	Mixed extract	Yeast extract	Soy bean ash	Fresh lettuce
	gm.	gm.	gm.	gm.	gm.	gm.	gm.	gm.	gm.	gm.	gm.
1	20	41	5	10	9	15	—	—	—	—	—
2	20†	59†	5	10†	5	1	—	—	—	—	—
3	20†	58†	5	10†	5	—	—	2	—	—	—
4	20†	59†	5	10†	5	—	—	—	1	—	—
5	20	59	5	10	5	—	1	—	—	—	—
6	20	59	4.5	10	5	—	1	—	—	0.5	—
7	20	59	5	10	5	—	1	—	—	—	10‡
8	Stock breeding diet: steamed bread, milk, green vegetables, wheat bran and, occasionally, meat.										

*McCollum's salt mixture 185.
†Extracted with absolute alcohol.
‡10 grams to each adult rat per day.

TABLE II.

Fertility and Lactation Record on Diets Exhibited in Table I.

Ration	Number females	Advanced pregnancies terminated by death	Number fetuses found	Number of litters	Total young born*	Young survived beyond 5th day	Young weaned at age of 4 weeks.	Number males on same diet
1	11	0	—	1	5	0	0	6
2	19	1	9	25	187	140	36	12
3	8	1	10	12	91	73	48	4
4	6	0	—	10	77	52	8	4
5	12	6	47	5	32	14	0	5
6	10†	3	24	4	26	15	2	5
7	5	0	—	4	30	27	27	2
8	4	0	—	4	38	36	36	4

*As counted on second day after birth.
†Not including two mother rats that died soon after birth of young.

literature, it seems clear that what appears to be contradictory evidence regarding the existence of vitamin E is only apparently so. On Ration 1, young rats grew more or less normally but were practically sterile; on Ration 2 they reproduced repeatedly. Thus 5 per cent butter-fat in Ration 2 is apparently an adequate source of vitamin E, provided the caloric value of the diet is not raised too high, as in Ration 1. That 9 per cent butter-fat in a high lard diet is not conducive to the production of young is also in harmony with the more recent work of Nelson and his associates[9] who are opposed to the vitamin E theory. A high fat diet, *per se,* is not deficient for reproduction as shown by Evans and Bishop,[1] who secured

litters when all of the 24 per cent fat in the diet is supplied in the form of butter-fat. More recently Kennedy and Palmer[10] found that crisco (hydrogenated cotton seed oil), replacing lard in a high fat diet, allows the production of young. Finally, a more definite test for the existence of vitamin E in butter-fat has been shown by Matill and associates,[11] and Sure,[2] who found that rats fed on synthetic diets containing cod liver oil in place of butter-fat were sterile.

The effects of synthetic diets on lactation are more clearly defined. All investigators are agreed concerning the striking failure of rats to rear their young when they are fed certain synthetic diets. This is chiefly due to deficiency in lactation, for the majority of the young lived from a few days to several weeks, and finally died in an emaciated condition before they could be weaned. This is strikingly true in the case of rats fed on Rations 2, 3 and 4.

It was first thought that the casein, starch, and yeast used in synthetic diets might be carriers of a dietary complex necessary for lactation as well as reproduction. The effect of feeding Ration 3 as contrasted with Ration 2, corroborates this view. A much higher percentage of young were reared on a synthetic diet with the addition of the mixed extract than without it. The extract was prepared by treating separately, casein, starch, and yeast, with absolute alcohol at boiling temperature under a reflux. The alcohol was filtered and the filtrate concentrated over a steam bath to a thick brownish liquid. The addition of 2 per cent of the mixed extract, as in the case of Ration 3, introduced several times more of the substance than was originally present in an unextracted ration. The addition of 1 per cent of yeast extract alone, as in the case of Ration 4, proved, however, ineffective. Subsequent experiments with commercial casein, corn starch and Northwestern yeast powder (Ration 5) also failed to demonstrate any appreciable difference between these and extracted products as far as lactation is concerned. Ration 6 was suggested by the work of Daniels and Hutton,[12] who found that the sterility of rats fed on milk diets could be relieved by supplementing the milk diet with soybean ash. The addition of soybean ash to a synthetic ration (Ration 6) exerted no favorable influence on lactation. In marked contrast, lactation was very successful when the rats were fed, in addition to the synthetic diet, 10 grams of fresh lettuce daily, as in the case of Ration 7. It seems evident that lettuce contains something which promotes not only fertility, as shown by Evans and Bishop and others, but also promotes lactation.

All the experiments were started on albino rats 3½ to 4 weeks old, weighing 40 to 50 gm. and were carried out in three series, each at a different time. Thus, Rations 1, 2, 3, and 8 constituted the first series, Rations 2, 4, 5, and 6 the second, and Rations 5, 6, 7, and 8 the last series. In each series except the second, two sets of controls were employed, one with litter mates fed on either Rations 2 or 5, and the other with litter mates fed on Ration 8, which is a stock breeding ration. It seems strange that such a striking number of advanced pregnancies were terminated by death of the animals as in the last series of experiments. There are also other observations which are not brought out in the tables. Although a few rats on Rations 2 and 3 reproduced repeatedly (4 or 5 litters to each rat) several second generation animals raised on the same diet were sterile. Again, the feeding of a number of rats, particularly those on Ration 4, was discontinued before they were given the opportunity to have a second or third litter. It was further observed that the rats fed on the high lard diet did not grow as normally as those on 5 per cent butter-fat rations, and the latter not as vigorously as litter mates raised on a stock breeding diet.

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[1] Evans, H. M., and Bishop, K. S., *Science*, 1922, lvi, 651.

[2] Sure, B., *J. Biol. Chem.*, 1923, lviii, 681.

[3] Mason, K. E., *Proc. Nat. Acad. Sci.*, 1925, xi, 377.

[4] Matill, H. A., Carman, J., and Clayton, M., *J. Biol. Chem.*, 1924, lxi, 729.

[5] Evans, H. M., and Bishop, K. S., *J. Am. Med. Assn.*, 1923, lxxxi, 889.

[6] Sure, B., *J. Biol. Chem.*, 1924, lxii, 371.

[7] Nelson, V. E., Heller, V. G., and Fulmer, E. I., *J. Biol. Chem.*, 1923, lvii, 415.

[8] Hogan, A. G., and Harshaw, H. M., *J. Met. Res.*, 1924, v, 111.

[9] Nelson, V. E., Jones, R. L., Heller, V. G., Parks, T. B., and Fulmer, E. I., *Am. J. Physiol.*, 1926, lxxvi, 325.

[10] Kennedy, C., and Palmer, L. S., *Am. J. Physiol.*, 1926, lxxvi, 316.

[11] Matill, H. A., and Clayton, M., *J. Biol. Chem.*, 1926, lxviii, 665.

[12] Daniels, A. L., and Hutton, M. K., *J. Biol. Chem.*, 1925, lxiii, 143.