

| Case | Tot. dosage of benzol in minims. | Period of administration. | Leucocytes |        | Erythrocytes |       | Result                   |
|------|----------------------------------|---------------------------|------------|--------|--------------|-------|--------------------------|
|      |                                  |                           | Before     | After  | Before       | After |                          |
| 1    | 3495                             | 47 days                   | 340,000    | 11,200 | 5.6m         | 6.1m  | Symptomatically improved |
| 2    | 555                              | 25 days                   | 292,000    | 300    | 2.1          | 1.0   | Purpura and death        |
| 3    | 7104                             | 3 1/3 yrs.                | 430,000    | 11,600 | 3.3          | .8    | No untoward symptoms     |

4. The effect of benzol on the bone marrow persists after withdrawal of the drug.

Benzol was routinely discontinued when the leucocyte count reached 30,000 but in many cases these cells continued to fall to a level well below normal.

The action of benzol in these cases, though parallel to the action on normal subjects, is not in itself conclusive because the patients were diseased and many of them received radium or Roentgen ray or both simultaneously with the benzol.

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### Fertility of the white rat on purified rations.

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It has been observed that animals (rats) fed exclusively on milk<sup>1</sup> or on purified rations made to simulate milk<sup>2</sup> seldom reproduce. In those instances where reproduction has occurred, few young were born and only a small percentage have survived the suckling period. The failure in the reproduction function of

<sup>1</sup> Daniels, A. L., and Hutton, M. K., *J. Biol. Chem.*, 1925, lxiii, 143.

<sup>2</sup> The purified ration consisted of casein 18 grams; lard 23 grams; butter 5 grams; cod liver oil 2 grams; cornstarch 47 grams; NaCl 0.514 grams; K<sub>2</sub>HPO<sub>4</sub> 2.587 grams; NaH<sub>2</sub>PO<sub>4</sub>·4H<sub>2</sub>O 1.172 grams; magnesium citrate 0.76 grams; CaCl<sub>2</sub>·H<sub>2</sub>O 2 grams; iron citrate 0.75 grams; potassium iodide 2 per cent solution, 0.4 cc.; alcoholic extract of 22 grams of wheat embryo.

these animals has been attributed to a vitamin deficiency,<sup>3</sup> the vitamin in question being an ether-soluble complex associated with certain fats, which is present in green leaves, egg yolk, and many natural foods. Another group of investigators working with milk<sup>4</sup> and so called purified mixtures in which milk has formed the basis of the ration have postulated that milk is deficient in certain essential inorganic substances. Rats fed on milk to which given amounts of iron, iodine, sodium silicate, sodium fluoride, manganese sulphate and aluminum potassium sulphate were added produced through six successive generations an average number of normal young. With combinations of two or three, or, when single salts were fed, the results were confusing, so that no conclusions could be drawn as to whether all or part of these salts were essential. When, however, these salts were added to a purified ration, in which the fat component consisted of 23 per cent lard, 5 per cent butter fat and 2 per cent cod liver oil, all animals failed to reproduce.

In order to determine whether the reproductive failure on our purified ration is due to a vitamin or inorganic deficiency we have repeated our experiments with purified rations, using (1) larger amounts of those salts which were effective in correcting the deficiencies of milk, and (2) substituting, in some cases, cotton seed oil for the lard, and in others different amounts of butter in place of an equivalent amount of lard. We have also extended our experiments with milk, testing the effect of the ether extract of lettuce as well as the ash of various foods which have been demonstrated to be potent in correcting the sterility in our milk fed rats.

The addition of the ash of soy beans, lettuce, and yeast respectively, to milk in amounts equal to those furnished by the non-incinerated foods which had been effective in overcoming the sterility on the milk diets, resulted in the birth of normal young. Two generations of rats fed these milk modifications have been successfully raised. These results confirm our earlier

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<sup>3</sup> Mattill, H. A., and Conklin, R. E., *J. Biol. Chem.*, 1920, xliv, 137; Mattill, H. A., and Stone, N. C., *J. Biol. Chem.*, 1923, lv, 443; Mattill, H. A., Carman, J. S., and Clayton, M. M., *J. Biol. Chem.*, 1924, lxi, 729; Sure, B., *J. Biol. Chem.*, 1923-24, lviii, 681, 693; Evans, H. M., and Bishop, K. S., *J. Am. Med. Assn.*, 1923, lxxxii, 889; *Anat. Rec.*, 1924, xxvii, 203.

<sup>4</sup> Anderegg, L. J., and Nelson, V. E., *J. Ind. and Eng. Chem.*, 1925, xvii, 453; Daniels, A. L., and Hutton, M. K., loc. cit.

findings, and indicate that milk is not lacking in the reproductive vitamin but is low in certain essential inorganic constituents.

On the other hand when lettuce ash, and larger amounts of those salts which were effective in correcting the deficiency of milk were added to our purified mixture, the animals failed to reproduce; whereas the addition of the ether extract of lettuce (0.3 gram per rat per day) to these rations, or the substitution of 23 per cent butter fat or cotton seed oil for the lard in our purified ration, resulted in the production of young at a comparatively early age. Sixty-four per cent of these have been reared through the suckling period. It would appear, therefore, that failure to reproduce on our former purified ration was due to a lack in the reproductive vitamin.

When yeast was used as a source of Vitamin B in the purified mixture containing an adequate amount of butter, lettuce extract, or cotton seed oil the second generation at two months of age was of average size (124 grams and 159 grams respectively in two groups) while the young of mothers receiving a similar purified ration in which a clear alcoholic extract of wheat embryo was used as a source of the antineuritic vitamin weighed only 44 grams (average of 8) at two months of age. The marked difference in the size of these two groups cannot be explained by the amount of Vitamin B furnished in the two rations, since in the latter the animals were given daily 100 cc. of the wheat germ extract in addition to that incorporated in the food. It seems probable that yeast carries essential inorganic substances and in studies aimed to determine the potency of certain salts some other source of Vitamin B must be used.