Franke(4), respectively. However, Touster, using the bromine-labile sulfur method(5) as well as the Hunter method of ergothioneine assay, has failed to confirm the latter results. Hunter recently, unlike Eagles and Vars, did not find that maize meal contains an ergothioneine precursor(6). The definite dietary influence on blood ergothioneine reported here, while possibly a species characteristic, is quite likely at least in part also attributable to the fact that the animals were started on the experimental diet at weaning.

Absence of an ergothioneine protective action against oxidation of the hemoglobin could account for the occurrence of an unidentified hemoglobin derivative previously observed in erythrocytes of the purified diet rabbits(7). Likewise, ergothioneine differences may be implicated in the individual and age differences previously observed in MHb reversion(8).

Summary. 1. Erythrocytes of rabbits fed a purified diet are nearly or completely devoid of ergothioneine. 2. The amount of ergothioneine in the red cells of purified diet rabbits increases with a supplement of liver or ergothioneine. 3. MHb formation and reversion rates after $NaNO_2$ are related inversely to the ergothioneine blood levels. 4. The diets used do not influence the blood glutathione

The authors are indebted to Mr. Howard F. Brubach for valuable assistance in aspects of this work.

4. Potter, V. R., and Franke, K. W., *J. Nutrition,* 1935, v9, 1.

5. Touster, O., *J. Biol. Chem.,* 1951, v188, 371.

6. Hunter, G., *Biochem, J.,* 1951, v48, 265.

7. Spicer, S. S., Wooley, J. G., and Clark, A. M., *J. Biol. Chem.,* 1950, v182, 445.

8. Spicer, S. S., and Reynolds, H., *Am. J. Physiol.,* 1949, v159, 47.

Received May 26, 1951. P.S.E.B.M., 1951, v77.

Effect of Diet on Obesity of Yellow Mice in Inbred Lines.* (18800)

PAUL F. FENTON AND HERMAN B. CHASE.

From the Department of Biology, Brown University, Providence, R. I.

It has been well known for many years that obesity is associated with yellow coat color in mice. That continued inbreeding reduces the obesity is less widely known but has been the experience of several mouse geneticists who have developed inbred lines of this stock. In mice the gene (A^y) for yellow coat color is dominant over the other alleles (A^w, A, a^t, a) at this agouti locus, and the homozygote (A^y A^y) is lethal. Consequently yellow mice are always heterozygous for yellow and produce both yellow and non-yellow offspring. By continued inbreeding of a yellow stock (successive brother-sister matings with one or both parents yellow), complete homozygosity at all loci is approached except at the A locus. In this manner yellow and non-yellow littermates are obtained having the same genetic background and differing only in that the yellows have one A^y gene and the non-yellows have none. Danforth(1), by crossing yellows to certain unrelated lines, showed that it was possible to increase the expression of obesity in the F_1 hybrid yellows. This practice of obtaining F_1 obese yellows has been employed in other studies on obesity(2,3). In the experiments to be reported here, however, such hybrid yellows were not used.

Two inbred yellow sublines (one pink-eyed and the other not) have been developed by one of us (H.B.C.). In view of the failure of the yellows of these sublines (and certain ones

*This investigation was supported by grants from the American Cancer Society upon recommendation of the Committee on Growth of the National Research Council; The National Cancer Institute, U. S. Public Health Service; and the Anna Fuller Fund.

1. Danforth, C. H., *J. Hered.,* 1927, v18, 153.

2. Dickerson, E. G., and Gowen, J. W., *Records Gen. Soc. Am.,* 1946, v14, 44.

3. Dickie, M. M., and Woolley, G. W., *J. Hered.,* 19..., v37, 365.

in other laboratories) to become obese under the usual conditions of colony maintenance, the question arose whether dietary manipulations could still bring out a capacity to become obese not shared by the non-yellow littermates.

Methods. Exp. 1. Two inbred yellow sublines were developed from the yellow stock of Jackson Laboratory. One subline is of the genotype CCA^y/abbDDppss and is pink-eyed yellow and café-au-lait (non-yellow). The other subline is of the genotype CCA^y/aBBDDPPss and is normal-eyed yellow and black (non-yellow). Both sublines are white-spotted. Since there is no difference in weight between these sublines, they will be considered together as the yellow strain. Yellow and non-yellow littermates were weaned between 21 and 28 days of age. They were maintained in plastic cages, one litter per cage, and fed a commercial stock ration. Food and water were supplied *ad libitum*.

Exp. 2. Yellow and non-yellow littermates of the sublines used in Exp. 1 were weaned when 21 days of age. At this time they were placed in individual screen-bottom metal cages and fed diet No. 226 or a commercial stock ration. Diet No. 226 was composed of casein 30%, sucrose 17.5%, corn oil 5%, hydrogenated cottonseed oil 42.5%, and salt mixture A(4) 5%. Fat- and water-soluble vitamins were added to this mixture in the amounts previously reported(4). Food and water were supplied *ad libitum*. The animals were weighed every 3 days until 51 days of age. Thereafter they were weighed once each week. They were sacrificed at 6 months of age and carcass fat determined by the method described by Feyder(5). A total of 72 mice were used in these 2 experiments.

Results. Exp. 1. At 6 months of age yellow mice were only slightly heavier on the average than their non-yellow littermates. At one year of age only one yellow male reached a weight of 50 g, all other animals weighing less than 41 g.

Exp. 2. The weight gains of yellow and non-yellow males and females fed diet No. 226 are shown in Fig. 1. Only one yellow male gained less weight during the experimental period than did the heaviest of the non-yellow males. Table I summarizes the fat content of the several experimental groups fed the highly purified diet. At the time of autopsy all yellow mice were found to have fatty livers, and two of them had liver tumors of moderate size. No comparable difference in weight gains was noted between yellow mice and non-yellow littermates when raised on the commercial stock ration.

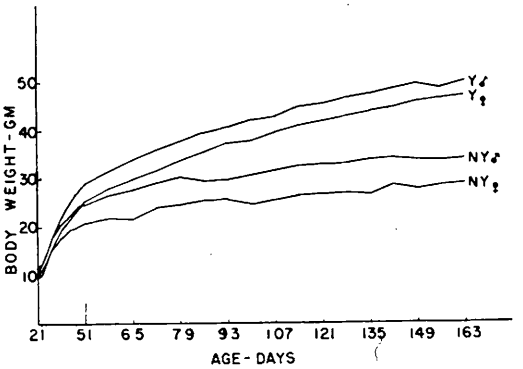

FIG. 1. Growth Curves of Yellow and Non-yellow Mice Fed Diet #226.

Discussion. The experiments reported here confirm the observation that yellow mice maintained under the usual laboratory conditions (housed either singly or in groups) did not become obese although they attain a body weight slightly greater than that of their non-yellow littermates. It is clear, however, that mice with the yellow gene, A^ya, still retain their capacity to become obese when fed suitable diets. The littermates homozygous for the non-yellow allele, aa, do not possess this capacity as far as we have been able to determine. The induction of obesity in yellow

TABLE I. Fat Content of Yellow and Non-Yellow Mice Fed Diet #226 until 6 Months of Age.

	Total body wt, g	Total fat, g	Fat-free wt, g	Fat content, %
Yellow males	50.7	17.2	33.5	33
" females	49	17.7	31.3	35.9
Non-yellow males	34	5.6	28.4	16.3
" females	27.7	4.2	23.5	14.6

4. Fenton, P. F., and Carr, C. J., *J. Nutrition,* 1951, v43, 441.

5. Feyder, S., *J. Nutrition,* 1935, v9, 457.

mice by the feeding of a diet of the "synthetic" type might be attributed to the greater nutritional adequacy of the synthetic diet as compared with the commercial pellet, or to the greater fat content of diet No. 226. Further experiments are needed to answer this question. Fatty livers such as shown by the yellow mice fed diet No. 226 have been reported in mice of the A and C_3H strains by Fenton and Mershon(6).

The occurrence of two liver tumors seems worthy of mention although no data are available on the incidence of liver tumors in these yellow sublines. The existence of such tumors in animals only 6 months of age merits further attention.

Summary and conclusions. 1. The observation has been confirmed that yellow mice on continued inbreeding no longer become obese if maintained under the usual laboratory conditions. 2. Feeding a diet of the "synthetic" type made it possible to induce obesity in the yellow mice but not in their non-yellow littermates. 3. The obese yellow mice were observed to have fatty livers. Two of these animals were found to have liver tumors.

6. Fenton, P. F., and Mershon, J. S., *Fed. Proc.*, 1951, v10, 382.

Received May 28, 1951. P.S.E.B.M., 1951, v77.

Effect of Nitro-Compounds and Aldehyde Semicarbazones on Virus of Primary Atypical Pneumonia.* (18801)

MONROE D. EATON, MAUDE E. PERRY, AND ILA M. GOCKE.

From the Department of Bacteriology and Immunology, Harvard Medical School, Boston, Mass.

Since the atypical pneumonia virus is intermediate in size between the agents of the psittacosis-LGV group and the so-called "true" viruses like influenza it may occupy in relation to chemical inhibition a strategic position one step down the scale from the "bacteria-like" viruses and rickettsiae for which many inhibitory substances have already been found. For this reason further exploration of the relation of chemical structure of certain substances to antiviral activity has been undertaken with this agent.

In a previous publication(1) it was reported that chloramphenicol had little effect on the pulmonary lesions produced in cotton rats by intranasal inoculation of the atypical pneumonia virus, but more recent studies to be reported here indicate activity in chick embryos. Because of the similarity of action of other p-nitro-benzene and 5-nitrofuraldehyde derivatives particularly those containing a semicarbazone group to that of chloramphenicol against respiratory infection in mice with viruses of the psittacosis-LGV group(2) it was considered of interest to study the effect of these substances against the atypical pneumonia virus. Experiments were also done with para-amino-benzaldehyde thiosemicarbazone and its acetyl derivative which have recently been reported to show slight inhibitory activity against vaccinia virus in mice and chick embryos(3).

Materials and methods. The methods of performing the experiments in chick embryos and cotton rats were essentially the same as those described(1). Chick embryos inoculated by the amniotic route with virus received the test substances in the yolk sac. Drugs used for the chick enbryos were sterilized by heat or filtration but for intraperitoneal injection of cotton rats the semicarbazones and thio-semicarbazones were

* This work was supported in part by grants from the Division of Research Grants and Fellowships of the National Institutes of Health, U. S. Public Health Service, and the Eugene Higgins Trust.

1. Eaton, M. D., PROC. SOC. EXP. BIOL. AND MED., 1950, v73, 24.

2. Eaton, M. D., Huang, C., and Levenson, C. G., PROC. SOC. EXP. BIOL. AND MED., 1949, v71, 501.

3. Hamre, D., Bernstein, J., and Donovick, R., PROC. SOC. EXP. BIOL. AND MED., 1950, v73, 275.