were calculated on the basis of the number of colonies isolated from a 1:1000 dilution of the soil specimens.

1. Emmons, C. W., *Public Health Rep.*, 1949, v64, 892.

2. Larsh, H. W., Hinton, A., Furcolow, M. L., *J. Lab. Clin. Med.*, 1953, v41, 478.

3. Ajello, L., Zeidberg, L. D., *Science*, 1951, v113, 662.

4. Stewart, R. A., Meyer, K. F., PROC. SOC. EXP. BIOL. AND MED., 1932, v29, 937.

5. Furcolow, M. L., Larsh, H. W., *ibid.*, 1952, v80, 246.

6. Smith, C. D., *Mycopathologia*, 1963, in press.

Received October 29, 1963. P.S.E.B.M., 1964, v115.

Variations of Acute Iodide Toxicity Among Inbred Strains of Mice. (28965)

FRED G. BOCK AND JOHN J. WRIGHT

Biological Station, Roswell Park Memorial Institute, New York State Department of Health, Springville, N. Y.

Although there is scant information concerning the lethal effects of acute iodide intoxication, most evidence indicates individual variability. In man, individual idiosyncrasies predispose certain persons to iodism, a relatively mild reaction to low levels of administered iodides(1). Occasionally, death occurs in newborn infants whose mothers receive repeated doses of iodides during gestation. The lethal effects in these cases may result from the development of huge goiters which interfere with respiration(2). Such occurrences are infrequent, suggesting that individual sensitivity is involved in these deaths. In rats and dogs, massive doses of iodide cause pulmonary edema, often followed by death; Trotter suggests that production of pulmonary edema by iodide is related to its antithyroid activity(3).

That some aspects of iodine metabolism are under genetic control is indicated by variations of thyroid activity among several lines of inbred mice and their F1 hybrids(4). Evidence presented here shows that acute iodide intoxication is also controlled by genetic factors.

Materials and methods. Mice of various strains (Table I), 30-90 days of age, were injected intraperitoneally with 10% aqueous NaI and observed daily for 14 days. In early experiments, it was noted that all mice killed by the treatment died within 5 days. Thereafter, mice still alive after 14 days were listed as survivors. The LD_{50} and its 99% confidence interval were calculated using the Spearman-Karber method(5). Approximately equal number of males and females were used in each group. With the exception of the ICR Swiss and CFW stocks, the mouse strains had been inbred for at least 20 generations. The CFW stock was obtained from Carworth Farms, inbred for 10 generations and maintained subsequently by random breeding. The ICR Swiss mice have been random bred since the stock was established.

To provide material for genetic analysis, F1 hybrid mice were obtained by reciprocal crosses between the F and CFW stocks. One hundred and seventy-eight of the F1 mice were injected with different levels of NaI to determine the LD_{50}. Other of the F1 hybrids were bred among themselves to provide 170 F2 mice. Finally, the remaining F1 hybrids were mated to animals of the F stock to provide 388 backcross mice. The F2 and backcross mice were injected once with 1.0 g/K of NaI. This dose was found sufficient to kill 100% of the F stock, but none of the CFW mice. The treatment thus served to divide the test animals into 2 groups, those like the F parental stock (sensitive) and those like the CFW stock (resistant).

Three additional groups of CFW mice, 1 month, 4 months, and 8 months of age were treated with either 1.6 or 2.4 g/K of NaI to provide information relating variations in

TABLE I. LD_{50} for Various Strains of Mice.

Strain*	LD_{50} (g/K)	99% confidence range for the LD_{50} (g/K)
F	.43	.40- .47
N	.70	.65- .75
C57/St	.78	.71- .85
L	.78	.72- .84
CBA	.83	.74- .93
PBR	.84	.76- .93
C3H	.88	.82- .94
Poly 2	.91	.88- .94
A	.92	.87- .97
AK	1.18	1.10-1.26
ICR Swiss	1.50	1.19-1.88
DBA/2	1.53	1.42-1.66
CFW	2.01	1.87-2.16
DBA/1	2.03	1.93-2.13

* The ICR Swiss, DBA/1, and DBA/2 mice were from the breeding colonies of Roswell Park Memorial Inst., West Seneca, N. Y. All other mice were from the breeding colony of Dr. L. C. Strong, Biological Station, Springville, N. Y.

animal age to the experimental results. To determine the reversibility of NaI intoxication, a group of 120 ICR Swiss mice was injected with 0.75 g/K of NaI (½ LD_{50}). Fourteen days later, the LD_{50} of a second injection of NaI was measured.

Results. Large differences in sensitivity to NaI were noted among various strains (Table I). The results for the DBA, C3H, and C57 strains were in substantial agreement with earlier unpublished observations in which closely related mice were used. The range of sensitivity was more than 4-fold; the LD_{50} for the F strain being 0.43 g/K and that for the DBA/1 mice being 2.03 g/K. The toxic effects of NaI were not cumulative over a 14-day time span. The ICR Swiss mice, injected 14 days earlier with 0.75 g/K of NaI, showed an LD_{50} of 1.68 g/K as compared to 1.50 g/K found in untreated ICR Swiss mice. This difference was not statistically significant ($P>.2$).

Six CFW males were injected with 10% aqueous NaCl at a dose of 2 g/K. All survived. In terms of sodium ion content, these mice received the equivalent of 4.2 g/K of NaI, an amount well above the 100% lethal dose. Survival of these mice demonstrated that deaths after NaI administration were due to the effects of the iodide ion.

No difference in iodide sensitivity was noted between the reciprocal F × CFW hybrids; the data have therefore been grouped together in Fig. 1. The LD_{50} of the F1 hybrid mice was nearly equal to that of the CFW parental stock, 1.67 g/K as compared to 2.01 g/K. Thus it appeared that, between these two stocks, the major factors responsible for resistance to injected I^- were dominant. The results with backcross mice injected with 1.0 g/K of NaI suggested a single gene effect for these 2 strains (Table II). Two hundred and five of the 388 mice

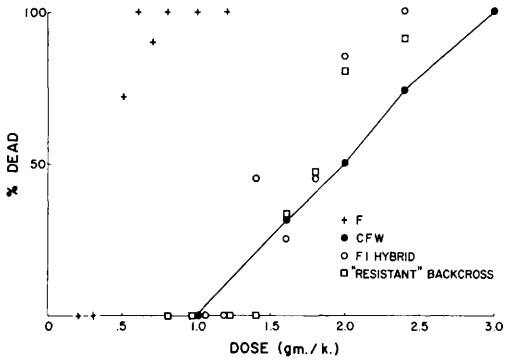

FIG. 1. Dose-response data for mice of the F and CFW stocks, F × CFW F1 hybrids, and "resistant" (see text) backcross mice.

died, whereas 194 deaths were predicted on the basis of a single dominant Mendelian gene conferring resistance to NaI. After 14 days, the surviving backcross mice were injected with varying doses of NaI. The response ($LD_{50} = 1.81$) was similar to that obtained for the F1 parents (Fig. 1), *i.e.* the backcross mice which survived the injection of NaI were, indeed, like the CFW mice.

The results with the F2 mice were not as clear cut. Thirty of the 170 mice died whereas 42.5 deaths were predicted on the basis of a single Mendelian gene. Chi square for this difference is 4.9 ($P\sim.03$). Unfortu-

TABLE II. Sensitivity of F2 and Backcross Mice to 1.0 g/K NaI.

Stock	No. mice	Dead		% predicted for a single gene
		No.	%	
Backcross (F × F1 hybrids)	388	205	53	50
(F × CFW) F2	170	30	18	25

TABLE III. Effect of Age and Sex on Sensitivity to NaI.

	%* of mice killed by			
	1.6 g/K		2.4 g/K	
Age (mo)	♂	♀	♂	♀
1	36	12	88	83
4	56	5	100	85
8	81	62	95	100

* Of 18-28 mice per group.

nately, further test of the survivors was not undertaken.

The study of aging mice showed that, on a dose/unit weight basis, older mice were more sensitive than young mice and males were more sensitive than females (Table III).

Discussion. Although the CFW stock has not been inbred for the customary 20 generations, there is evidence that the mice were homozygous for the genes affecting sensitivity to I^-. The dose response curve of the CFW mice appeared to be of a simple sigmoid shape. The variance of the mean log dose was as small as that found for other inbred strains. Likewise, the dose response curve of the F1 hybrids failed to disclose a mixed population. If the CFW stock were heterozygous for the genes controlling iodide sensitivity, one might expect evidence of two populations in the CFW stock, and particularly in the F × CFW hybrids. Evidence for two such populations was clearly apparent in the backcross mice; half of the mice were killed by 1.0 g/K NaI although the survivors showed no deaths after subsequent administration of 1.4 g/K. Exactly this result would be predicted on the basis of a single gene for which the CFW mice were homozygous.

The differences in iodide sensitivity between males and females and among mice of different ages may have been due to the differences in body weight. Because the dose per unit of weight was the same, larger animals received a greater total dose of drug. It seemed desirable to check whether the LD_{50} would be less variable if determined on a dose per mouse basis. A limited examination of this possibility, however, showed that older (heavy) mice were more resistant than young (light) mice when treated with equal amounts of NaI.

Regardless of the mechanism, however, the differences among mice of different ages and sexes impose a limitation on the analysis of the data presented here. Small or moderate differences in the observed LD_{50} or the dose response curves may be due to differences in weight of the animals used for the study. Large differences, however, are undoubtedly due to factors more directly related to the metabolism of iodide. This is particularly true of differences between mice such as the F, which are of light weight, and the CFW, which are heavier. In this case, a difference in sensitivity was observed in spite of the weight effect rather than because of it.

CFW mice were resistant to injected NaI; F mice were sensitive. The results with the F1, F2, and backcross offspring suggested that the difference between the parental stocks was due mainly to the action of a single gene, with resistance to NaI being dominant. There were slight differences between the F1 hybrids and the CFW parents and between the "resistant" backcross mice and the CFW stock. These differences may have been due to experimental variables such as weight or they may have been due to modifying genetic factors. There is insufficient evidence to choose between these possibilities. It is clear, however, that genetic factors are capable of causing much of the variability in response to iodine administration which is exhibited both in experimental animals and in clinical patients.

1. Grollman, A., *Pharmacology and Therapeutics,* 4th ed., Lea & Febiger, Philadelphia, 1960, p923.

2. Galina, M. P., Avnet, N. L., Einhorn, A., *New Eng. J. Med.,* 1962, v267, 1124.

3. Trotter, W. R., *Brit. J. Exp. Path.,* 1952, v33, 202.

4. Amin, A., Chai, C. K., Reineke, E. P., *Am. J. Physiol.,* 1957, v191, 34.

5. Finney, D. J., *Statistical Methods in Biological Assay,* Hafner Publishing Co., New York, 1952, p524.

Received October 30, 1963. P.S.E.B.M., 1964, v115.