tomarily measured by mortality. Data collected by this method are often inadequate because only the existence or non-existence of anaphylaxis is recognized. A method of observation which measures the extent of the reaction, rather than its mere presence or absence, is more satisfactory because variation within groups of mice can be more precisely estimated. An efficient method for determining the extent of anaphylaxis should have these characteristics: 1) There should be regularity in the dose-response relationship; 2) experimental error should be small enough so that moderate differences in the response can be measured in rather small groups of animals; and 3) individual observations should be distributed continuously so as to rule out qualitative observations. The data presented herein indicate that these criteria are approached when anaphylaxis in mice is determined by change in hematocrit. Furthermore, this method was shown to be reliable for two widely divergent anaphylactogenic procedures. This method has other advantages in that the collection of blood samples does not seem to harm the animal; each mouse serves as its own prechallenge control, and the apparatus required is simple, inexpensive, and seems to have little intrinsic variation(1). Therefore, the use of hematocrit determinations for drawing quantitative comparisons in mouse anaphylaxis studies is recommended.

Summary. Results indicate that hematocrit rise in anaphylactic mice is a sigmoidal function of the log dose bovine albumin in the challenge antigen preparation. Over a given range, the relationship tends to be linear. Indications are that hematocrit change is a continuous function, and not a ratio of "all or none" responses. This method is recommended for drawing quantitative comparisons in studies of mouse anaphylaxis.

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Effect of Artificial Changes in Egg Composition on Hatchability and Chick Growth.* (23635)

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Egg size. as well as the weight and physical characteristics of yolk and albumen, is known to affect the hatchability of hen's eggs. The earlier work on this subject has been reviewed by Landauer(1). Lerner(2) has shown that optimal hatchability is found in eggs slightly below the mean egg size for any stock in which egg size has been favored by artificial selection.

The weight and quality of albumen in the hen's egg can be reduced artificially by local X-irradiation of the oviduct(3). The influence of these changes in the egg on hatchability and chick growth is considered in the present report.

Materials and methods. Mature White Leghorn hens were used in these experiments. Oviducts were exposed by laparotomies and X-irradiated; other parts of the hen's body were protected from irradiation by lead shielding. The details of the irradiation procedure have been described elsewhere(3). Surgical controls were prepared for each irradiated group. In the initial experiments the entire oviducts of experimental birds were irradiated. However, this procedure resulted in such numerous and severe shell defects in eggs laid following treatment that only a few were suitable for hatchability studies. This report deals only with later experimental groups in

* This research was performed under U. S. Atomic Energy Commission contract with the University of California.

which only the magnum (albumen-producing region of the oviduct) was irradiated with 3500 r X-rays. A small amount of whole-body irradiation (<2% of the treatment dose) resulted from stray radiation from the machine and from passage of X-rays through the lead shielding. While the deleterious effect of whole-body X-irradiation is well known (4), this small quantity was considered negligible. Whole-body X-irradiation of chickens up to 300 r has been shown to have no effect on fertility or hatchability of eggs laid after treatment(5). The X-irradiation procedure used in this experiment may be considered merely as a means of producing eggs with a diminished albumen content; its results are similar to those of the more difficult method of magnum resection(6). The operative procedures used interrupted the normal sequences of ovulation and oviposition for approximately 3 weeks. When operated birds resumed lay, they were inseminated artificially at weekly intervals. The last 20 eggs of each 30 laid by each hen after treatment were incubated; the first 10 eggs were broken-out for other studies(3,7). All eggs were incubated in a forced-draft commercial type of incubator under the usual conditions of temperature and humidity. Eggs were candled twice during the incubation period; infertile eggs and those containing dead embryos were removed. Dead embryos were classified as to cause of death and age at death. Since embryos may die at such an early stage of development as to be considered infertile(8), all blastodiscs were examined carefully for any evidence of cell proliferation to distinguish between infertile eggs and early embryonic death. All chicks hatched were weighed on removal from the incubator and at weekly intervals thereafter throughout the 6-week brooding period.

Results. Response of the birds to magnum X-irradiation was variable; in some cases, a reduction of albumen quality† was the principal effect, while in others marked reductions in both albumen quality and quantity were observed. In the surgical controls neither of these factors was affected adversely. The mean effect of this level of X-irradiation (3500 r) on egg composition has been reported elsewhere(3).

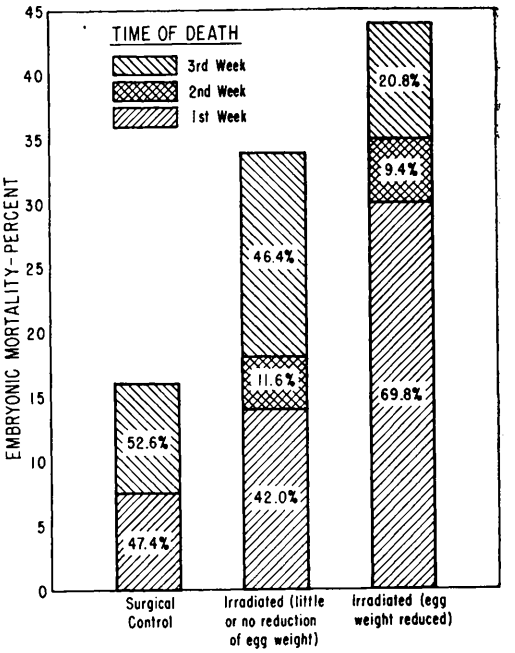

FIG. 1. Embryonic mortality in eggs laid after oviduct X-irradiation. Ordinate indicates embryo mortality observed in each of the treatment groups. Chronological distribution of embryo death (as % of total deaths) is indicated. Surviving populations of these groups are the same as in Groups 1, 2 and 3 in Table I.

Accordingly, for convenience in analyzing hatchability and growth, the birds were divided into 3 groups: 1) surgical controls; 2) X-irradiated with no marked decrease in egg weight; 3) X-irradiated with egg weight decreased. Not enough experimental birds were prepared to support a more detailed analysis of X-irradiation-induced changes in egg and their effect on hatchability.

Fig. 1 indicates that total mortality, as well as its chronological distribution, was influenced by the X-irradiation procedure. There are two critical periods in chick embryo development, and under normal conditions most embryonic mortality is associated with them. The first occurs between the second and fourth days of incubation when most of the organ

† Albumen quality is a commercial term denoting, semiquantitatively, its viscosity. Standard methods for evaluating albumen quality are available(9). Decreases in albumen viscosity increase the likelihood of contact between the shell and yolk (embryo) which may interfere with normal development.

TABLE I. Egg Size, Hatching Weight and Growth of Chicks.

Treatment group	Control 1	Irradiated; no effect on egg wt 2	No post-treatment reduction of egg size 1 & 2	Irradiated with reduction of egg size 3
No. hens	7	7	14	11
No. chicks hatched	86	84	170	66
Egg wt (g)–Pre-treatment	59.1 ± 2.0	58.3 ± 1.8	58.7 ± 1.3	58.0 ± .8
Post-treatment	61.2 ± 1.6	59.8 ± 1.7	60.5 ± 1.1	53.5 ± 1.0
Chick wt—g	41.9 ± .4	40.6 ± .5	41.2 ± .3	35.6 ± .4
% egg	68.0 ± .4	66.0 ± .4	67.0 ± .3	65.5 ± .5
Body wt of chick as multiple of hatching wt at:				
1 wk	1.67 ± .03	1.64 ± .02	1.66 ± .02	1.74 ± .02
2	2.96 ± .06	2.67 ± .10	2.80 ± .06	3.19 ± .06
3	4.75 ± .11	4.71 ± .09	4.73 ± .07	5.32 ± .10
4	6.97 ± .15	7.10 ± .20	7.04 ± .12	7.96 ± .22
5	9.90 ± .21	9.86 ± .24	9.88 ± .16	10.86 ± .25
6	13.05 ± .43	13.50 ± .36	13.39 ± .29	14.90 ± .41

systems are formed. Any developmental deficiency results in the death of the embryo, and ordinarily this accounts for slightly more than one-third of the total embryo mortality. The second critical period occurs between the 17th and 21st days of incubation (*i.e.*, the last four days of that process) when various prehatching changes take place. Ordinarily, about two-thirds of total embryo mortality occurs during this period.

Eggs in which albumen quantity and quality were reduced were characterized by a marked increase in mortality during the first week of incubation. An examination of embryos dying in this period indicated that much of this mortality resulted from a failure in very early differentiation, rather than the more commonly observed later defects in organogenesis. The occurrence of appreciable second-week mortality was also unusual. Mortality associated with the terminal period of development, however, was of the type normally expected (*viz.*, failure of the embryo to orient properly, late yolk sac absorption, difficulties in change from allantoic to pulmonary respiration, etc.).

Differences in hatch weight (absolute and relative to egg weight) were found between the chicks from control and irradiated hens (Table I). These differences are large only between the controls and the group in which egg weight was decreased. Although the observed differences between the groups in relative chick weights (calculated as percent of egg weights) are statistically significant, they are small. Other investigators, whose results have been reviewed by Hutt(10), have consistently found that within a single breed day-old chick weight is determined largely by egg weight.

Whether or not differences in the growth rates of the 3 groups of chicks exist was found to depend upon the basis of the comparison (*i.e.*, absolute or relative to hatch weight). There is no systematic difference by either comparison between the control chicks (Group 1) and those from X-irradiated hens in which egg size was not markedly decreased (Group 2). The growth rate (relative to hatch weight) was significantly greater in the chicks from the X-irradiated hens in which egg size was reduced (Group 3). However, on an absolute basis, there is no statistically significant difference in the growth rate after the first week of brooding.

Discussion. Decreases in albumen quality and quantity brought about by oviduct irradiation are associated with a decreased hatchability. This artificial alteration, however, does not necessarily relate to the effects of natural variations in albumen size and quality on hatchability. The effect of local oviduct X-irradiation on the egg, and especially the adverse effect on albumen quality, is simi-

lar to that of certain respiratory diseases(3). Hatchability of eggs from flocks after an outbreak of respiratory disease, such as Newcastle disease, is decreased, and characteristically very early embryonic mortality, similar to that reported here, is observed.

Other conditions leading to higher embryo mortality do not follow the pattern observed after qualitative or quantitative decreases in the albumen (*i.e.*, oviduct X-irradiation or respiratory disease). The embryo mortality encountered after excessive inbreeding, for example, is much more variable, but deaths occur mainly at the usual critical periods of development.‡ No reproducible pattern is associated with this cause of embryo death.

Increased embryo mortality in the second week of incubation is also encountered in eggs from birds deficient in certain specific nutrients (*viz.*, riboflavin(11)) and in some stocks which carry specific genetic lethals. These intermediate deaths may result from metabolic abnormalities.

It appears that the "growth potential" of the chicks is not affected by the X-irradiation treatment of the dams. Although chicks from the lighter eggs are smaller at hatching, this difference soon disappears. Similar results have been obtained with rats which were stunted early in life by a protein-deficient ration(12). Upon return to a normal feeding regime, stunted rats made up their early deficiency and eventually out-grew their controls. Several studies with normal birds(10) have also indicated that the size of the chick at hatching has little effect on its subsequent growth.

Summary and conclusions. Artificial decrease in albumen quality and quantity caused by local X-irradiation of the oviduct reduces the hatchability of eggs. The distribution of embryo mortality is different from that encountered in normal eggs: very early mortality is markedly increased. Chicks from such eggs have a lower hatch weight; however, no effect on subsequent chick growth was observed.

‡ Unpublished.

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