VARIATIONS WITHIN A STRAIN OF RHODE ISLAND REDS IN THEIR REQUIREMENT FOR VITAMIN B_{12}

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by

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CONTENTS

INTRODUCTION

The basis of all knowledge is experience. In science it is experimentation. The raw materials of experimentation are variations. Causes of variations in biological studies are differences in heredity, with which the individuals start life, and differences in environment, known and unknown, to which they were exposed during their development. The interaction of these two main causes may have a joint effect which can not be ascribed to either one alone.

The lack of ability to reproduce experimental results can probably be attributed to this effect of interaction, especially in environmental experiments which have no precautions controlling the individuals' heredity either by uniform selection of individuals or by breeding for uniformity. Variation that is ascribed to environment alone may be the combined results of both heredity and environment.

Lack of uniformity of any biological factor in individuals of a population is certainly expected. Selection of uniform individuals to be used in an experiment arouses the curiosity. Can that uniformity be obtained by breeding? Basic genetic principles are involved; specific genotypes allowing the development of a degree of expression of a certain characteristic. The cause of the lack of uniformity is the heterozygosis of the individuals. By phenotypic selection, relatively homozygous individuals can be obtained in some future generation.

A great deal of work is being carried on today concerning hereditary resistance and susceptibility to certain diseases. Hereditary differences in anatomy causing variations in some physiological processes are also being studied.

Whether it is a difference in anatomy or a difference directly caused by a specific genotype, the difference in individual efficiency of utilization of a specific nutrient has received very little attention experimentally.

The experiment presented in this thesis concerns itself with a study of this type, namely a study of the variations in two-week weights of chicks fed a vitamin B_{12} deficient diet.

PURPOSE OF STUDY

The purpose of this study was to determine if variations in two-week weights of chicks on a vitamin B_{12} deficient diet could be partially attributed to heredity; and if so, to test the possibility of developing strains relatively resistant and relatively susceptible to a deficiency of vitamin B_{12} in the diet.

PREVIOUS WORK

Inheritance as a Factor Influencing Efficiency of Food Utilization

An intensive genetic study on the efficiency of utilization of a specific nutrient was made by Lamoreux and Hutt (1948). Based on relative resistance or susceptibility of chicks to a riboflavin deficiency, selective breeding was practiced for six generations. The analysis of the progeny mortality and weight data showed lower mortality and higher weights in the resistant strain as compared with the mortality and weights of the susceptible strain. Chicks from both strains made comparable growth gains when fed an adequate diet. These researchers believed that between the two strains there were genetic differences which were specifically concerned with the utilization of riboflavin. The data of an incomplete study by Lerner and Bird (1948) supported the results of Lamoreux and Hutt with respect to genetic differences in the chicks' response to a riboflavin deficiency.

Many nutritional researchers have demonstrated or suggested breed and strain differences in the dietary requirement of certain nutrients. These differences have been especially prominent in biological assays in which the response of the animals is used to determine the potency of a specific nutrient within the supplement fed the animals. Biological assays which utilize chicks as experimental animals have another variable to contend with, i.e., the amount of the specific nutrient in the egg at hatching time as it may vary between breeds, strains, families, or even individuals within a family.

Selecting on the merit of efficiency of food utilization, Morris, et al., (1933) inbred nine generations of rats. A line for high efficiency and a line for low efficiency were developed. Each generation was compared statistically with the second generation and significant differences were found. The average level of efficiency of food utilization appeared fairly uniform in both lines after the sixth generation. The evidence of this study supported the belief that complex heritable factors influence the efficiency of food utilization.

Strains of rats fed a low vitamin D, high calcium diet were found by Gowen (1936) to have an average life of five to six months, half of the average life span of rats on a normal diet. Males lived longer on this deficient diet than the females, while on a non-deficient diet the females lived longer. The evidence of this experiment points to distinct genetic differences in the dietary requirements of the different strains. However, resistant and susceptible strains to vitamin D dietary deficiency were not developed.

Ringrose and Norris (1936) found strain differences in chicks used for the biological assay of vitamin A.

Breed differences in resistance to a deficiency of thiamine were observed by Lamoreux and Hutt (1939). In each of four experiments the ability of White Leghorn chicks to survive on a diet deficient in thiamine was greater than that of Rhode Island Reds. The progeny of a White Leghorn male and Rhode Island Red females were intermediate between the parental lines with respect to a deficiency of thiamine.

White Leghorns were also found to be superior in resistance to manganese deficiency when compared with New Hampshires in a study by Gallup and Norris (1939). Within the two breeds there were also strain differences in manganese requirements.

Golding, et al., (1940) found White Leghorns superior to Barred Rocks in the low requirement of manganese.

Studies in the nutrition of the mouse by Fenton and Cowgill (1947) showed that one highly inbred strain of mice had a low requirement for riboflavin, while another highly inbred strain had a high requirement.

Indications resulting from a study by Bethke, et al., (1936) show that the variability of vitamin G in eggs from different hens is due in part to strain differences. The majority of the variability came from the various levels of vitamin G in the diet of the hen. Hens' diets also affect the vitamin E content of the egg (Barnum, 1935) and the vitamin A content of the egg (Ellis, 1933).

Using a modified macro-fermentation assay procedure, Scrimshaw, _et al._, (1945) found that the mean thiamine content of eggs from White Leghorns was significantly greater than the thiamine content of eggs from Rhode Island Reds and Barred Rocks, the latter two having comparable stores of thiamine in their eggs. All these hens were kept under comparable conditions and had the same thiamine content in their diets.

Jackson, _et al._, (1946) found variations in the concentration of riboflavin in the eggs from various breeds and strains on the same ration. There were also variations between hens of the same strain.

Bird, _et al._, (1946) have clearly shown that the manifestations of animal protein factor deficiency in the hens' diet are poor hatchability and low viability of their chicks, indicating that the stores of this factor in the egg are decreased when the diet of the hen is deficient in this factor.

Studies by Bird, _et al._, (1947) show that chicks vary widely with respect to their stores, at hatching time, of the animal protein factor and their ability to withstand a deficiency. Hens fed a deficient diet for eleven months were found to vary in the hatchability of their eggs. Hens characterized by high and by low hatchability were not found to vary with respect to the growth promoting properties of their excreta or their efficiency of food utilization. The progeny of hens characterized by high, intermediate, and low hatchability, respectively, showed high, intermediate, and low viability and growth rate to six weeks of age. The effect of dietary supplements upon the growth of chicks varied with the maternal diets and with the ability of the dams to withstand the dietary deficiency.

EXPERIMENTAL PROCEDURE

I. REARING OF PARENTAL STOCK AND SELECTION OF BREEDERS

The initial preparations for this study began in January, 1951, when the replacement stock of Rhode Island Reds for that year was hatched. The parental stock (P_1) was housed in nine different pens, with fifteen females and one male per pen. Seven hundred F_1 chicks were hatched in January and seven hundred F_1 chicks were hatched in February.

The chicks were wingbanded and brooded in batteries on a standard chick starting mash. Mortality was recorded daily and individual weights, to the nearest gram, were obtained at the end of two weeks. At four weeks of age the chicks were removed to a brooder house. Six weeks later the flock was culled heavily. The birds were ranged on good pasture from May to August, at which time the breeders were selected and housed.

Selection was based on pedigree estimates of the breeders. These estimates were obtained from the records of the two-week body weights of the breeder and body weights of its sibs from the two replacement hatches. Two-week body weight records of the chicks from two previous hatches fed a vitamin B_{12} deficient diet also contributed to the estimates.

All P_1 individuals were classified as Resistant (R) or Susceptible (S) based upon the reaction of their progeny obtained from four hatches. The F_1 individuals were classified as R if their sibs were in the top 20% of the weight range and suffered less than 25% mortality, or S if their sibs were in the lowest 20% of the weight range and had more than 25% mortality.

II. PLAN OF MATINGS

The plan of each experiment was a modification of the conditions of Fisher's (1925-1944) "Latin Square" design. The modification is that these were polyallel matings involved in each experiment and analyses were made within the polyallel mating arrangement instead of rows, columns, and treatments.

Contrary to procedure used in Drosophila, i.e., the female parent is listed first, the symbols used herein follow the procedure of the animal breeders where the male parent is listed first.

The subscript number refers to a specific male and the subscript letter refers to a specific pen.

The designs of the experiments are shown in Figure 1 on the following page.

Figure 1 - Designs of Experiment I and Experiment II

Experiment I

Hatch	Pen A (S) Male		Female	Pen B (S) Male		Female	Pen C (R) Male		Female	Pen D (R) Male		Female
1 and 2	S_1	x	S_a	S_2	x	S_b	R_1	x	R_c	R_2	x	R_d
3 and 4	S_2	x	S_a	S_1	x	S_b	R_2	x	R_c	R_1	x	R_d
5 and 6	R_2	x	S_a	R_1	x	S_b	S_2	x	R_c	S_1	x	R_d
7 and 8	R_1	x	S_a	R_2	x	S_b	S_1	x	R_c	S_2	x	R_d

Experiment II

Hatch	Pen E (S) Male		Female	Pen F (S) Male		Female	Pen G (R) Male		Female	Pen H (R) Male		Female
1 and 2	S_3	x	S_e	R_4	x	S_f	R_3	x	R_g	S_4	x	R_h
3 and 4	R_3	x	S_e	S_4	x	S_f	S_3	x	R_g	R_4	x	R_h
5 and 6	R_4	x	S_e	S_3	x	S_f	S_4	x	R_g	R_3	x	R_h
7 and 8	S_4	x	S_e	R_3	x	S_f	R_4	x	R_g	S_3	x	R_h

Two pens, each housing 20 R pullets, and two pens, each housing 20 S pullets, were set up in Experiments I and II. Two R cockerels and two S cockerels were rotated in each of the four pens of each experiment. Two hatches were obtained from each mating. Males were removed from the pens and placed in individual cages on approximately the 28th day after their initial entry into the pen. Two days later, semen was collected from the next male to be introduced in the particular pen and pullets of that pen were artificially inseminated with 0.2 cc. of semen. If the volume of semen was not sufficient on the first collection, the male was returned to the battery and the remaining inseminations took place the following afternoon. The male was then placed in that pen after all the pullets had been inseminated. Eggs laid after a lapse of 36 hours were saved for hatching. It is generally accepted that the newly introduced sperm fertilizes the ova (Warren and Gish, 1943; Warren and Kilpatrick, 1929).

Eggs were set at intervals of approximately twelve days. All chicks were pedigreed and hatchability, fertility and abnormal embryos were recorded. The chicks were placed in batteries equipped with raised bottoms and fed a basal diet deficient in vitamin B_{12}, (Table 1). The wing band numbers of all dead chicks were recorded each day and the survivors were weighed at two weeks of age.

Table 1 - Composition of Basal Chick Diet

	Pounds
Soybean oil meal............................	50.0
Ground yellow corn..........................	39.4
Dehydrated alfalfa meal.....................	5.0
Steamed bonemeal............................	3.0
Oyster shell flour..........................	1.5
B-Y feed (500 micrograms riboflavin per gram)......................................	0.3
Fish oil (400 D 2000 A).....................	0.2
Salt (iodized)..............................	0.5
Choline chloride............................	0.1
Nicotinic acid..............................	0.005
Manganese sulfate...........................	0.022
	100.027

Calculated analysis:

Protein.....................	26.6 %
Calcium.....................	1.5 %
Phosphorus..................	0.8 %

III. STATISTICAL ANALYSIS

As stated previously, the design of the matings was to simulate a "Latin Square" where each cell was analyzed respectively and later combined in all pertinent comparisons. The Analysis of Variance according to the procedure of Fisher (1924), as presented by Snedecor (1946), was used to test significance.

However, in the analyses of variance only one criterion of measurement can be used. The R line had greater weights and very little mortality, while the S line had low weights and a high mortality. It was recognized that mortality is a necessary criterion of selection and can not be eliminated in any analysis. Therefore, the experimenter chose the one criterion of weight, assuming the weight of every dead chick as 30 grams. It is logical that the chick showing vitamin B_{12} deficiency symptoms will decrease in initial weight; the initial weight of a chick is approximately 40 grams or 65% of the weight of the egg (Jull and Quinn, 1925). It is postulated that because no chick weights were recorded under 31 grams, the assumed weight of 30 grams is reasonable for all chicks that died during the two week period. It is logical that the use of the figure of 30 grams as a constant for all dead chicks is superior in an analysis to giving these chicks no weight at all and yet including them in the average.

If a female of a pen was represented in a hatch by only one chick that chick's performance was eliminated from the data.

One hundred and sixty-two analyses of variance were made of all pertinent comparisons of the breeders' performance involving four thousand chick weights of Experiments I and II.

In Experiments I and II the data were analyzed initially to obtain differences of progeny performance between hens of a pen. Further, in Experiments I and II two consecutive hatches of chicks were produced by each of the sixteen different pen matings to obtain sufficient numbers of chicks for proper statistical analysis. Thirty-two analyses of variance were made of the two-week weights of the chicks from two consecutive hatches from each of the sixteen different pen matings. Each pen mating consisted of one male and twenty females. On an average, only eleven of the twenty females of a pen produced chicks in each hatch. The design of the sixteen different pen matings and their duplicate hatches may be seen in Figure 1. These initial thirty-two analyses of variance gave estimates of variance of progeny weights between hens of the same pen.

The data from the analysis of variance of each individual mating of one hatch were combined with the data of the analysis of variance from the same mating of the next consecutive hatch. A new analysis of variance was made to calculate the estimate of the average variance between the hens of a pen from the data of two hatches;

thus having gained an estimate of variance between consecutive hatches involving the same mating.

In order to obtain a final analysis of variance giving comparisons between the sires of both classifications (R and S), the statistics of the combined consecutive hatches had to be compared in a special manner. It is designated as Approach No. 1 and is illustrated in Figure 2.

Approach No. 2 (Figure 3) is necessary to obtain comparisons between the pens of both classifications (R and S).

Approach No. 3 (Figure 4) is necessary for comparisons between the unlike matings R x R vs. S x S and R x S vs. S x R.

S.V.* Bet. Like Pens i.e. $S_2 \times S_a$ vs. $S_2 \times S_b$

(8 Comparisons in the Experiment)

$S_1 \times S_a$	
$S_2 \times S_a$	

vs.

$S_2 \times S_b$	
$S_1 \times S_b$	

S.V. Bet. Like Sires i.e. $S_1 \times S_{ab}$ vs. $S_2 \times S_{ab}$

(4 Comparisons in the Experiment)

S_a	S_b
$S_1 \times S_a$	---
---	$S_1 \times S_b$

vs.

S_a	S_b
---	$S_2 \times S_b$
$S_2 \times S_a$	---

S.V. Bet. Unlike Matings i.e. $S_{12} \times S_{ab}$ vs. $S_{12} \times R_{cd}$

(2 Comparisons in the Experiment)

S_a	S_b
$S_1 \times S_a$	$S_2 \times S_b$
$S_2 \times S_a$	$S_1 \times S_b$

vs.

R_c	R_d
$S_2 \times R_c$	$S_1 \times R_d$
$S_1 \times R_c$	$S_2 \times R_d$

S.V. Bet. Unlike Sires - Includes all cells of the Latin Square design in the Comparison

S_a	S_b	R_c	R_d
$S_1 \times S_a$	$S_2 \times S_b$	---	---
$S_2 \times S_a$	$S_1 \times S_b$	---	---
---	---	$S_2 \times R_c$	$S_1 \times R_d$
---	---	$S_1 \times R_c$	$S_2 \times R_d$

vs.

S_a	S_b	R_c	R_d
---	---	$R_1 \times R_c$	$R_2 \times R_d$
---	---	$R_2 \times R_c$	$R_1 \times R_d$
$R_2 \times S_a$	$R_1 \times S_b$	---	---
$R_1 \times S_a$	$R_2 \times S_b$	---	---

*S.V. - Source of variation

S.V.* Bet. Like Sires i.e. S_1 x S_a vs. S_2 x S_a

(8 Comparisons in the Experiment)

S_1 x S_a		S_2 x S_b
vs.		vs.
S_2 x S_a		S_1 x S_b

S.V. Bet. Like Pens i.e. S_{12} x S_a vs. S_{12} x S_b

(4 Comparisons in the Experiment)

S_a		S_b
S_1 x S_a	vs.	S_2 x S_b
S_2 x S_a		S_1 x S_b

S.V. Bet. Unlike Matings i.e. S_{12} x S_{ab} vs. R_{12} x S_{ab}

(2 Comparisons in the Experiment)

S_a	S_b
S_1 x S_a	S_2 x S_b
S_2 x S_a	S_1 x S_b

vs.

S_a	S_b
R_2 x S_a	R_1 x S_b
R_1 x S_a	R_2 x S_b

S.V. Bet. Unlike Pens - Involves all the cells of the Latin Square Experiment

S_a	S_b		R_c	R_d
S_1 x S_a	S_2 x S_b		R_1 x R_c	R_2 x R_d
S_2 x S_a	S_1 x S_b	vs.	R_2 x R_c	R_1 x R_d
R_2 x S_a	R_1 x S_b		S_2 x R_c	S_1 x R_d
R_1 x S_a	R_2 x S_b		S_1 x R_c	S_2 x R_d

* S.V. - Source of variation

S.V.* Bet. Pens - (Same as Figure 2)

S.V. Bet. Like Sires - (Same as Figure 2)

S.V. Bet. Unlike Matings i.e. S_{12} x S_{ab} vs. R_{12} x R_{cd}

(2 Comparisons in the Experiment)

S_a	S_b		R_c	R_d
S_1 x S_a	S_2 x S_b	vs.	R_1 x R_c	R_2 x R_d
S_2 x S_a	S_1 x S_b		R_2 x R_c	R_1 x R_d

S.V. Bet. matings of extreme classification and reciprocal matings

S_a	S_b	R_c	R_d
S_1 x S_a	S_2 x S_b	R_1 x R_c	R_2 x R_d
S_2 x S_a	S_1 x S_b	R_2 x R_c	R_1 x R_d

vs.

R_2 x S_a	R_1 x S_b	S_2 x R_c	S_1 x R_d
R_1 x S_a	R_2 x S_b	S_1 x R_c	S_2 x R_d

*S.V. - Source of variation

RESULTS AND DISCUSSION

In the P_1 population both the R individuals and the S individuals were found to be heterogeneous with respect to transmitting the hereditary factors for a uniform body weight to all their offspring. The degree of phenotypic expression within families ran from resistant (body weight in upper 20 per cent of weight range, less than 25 per cent mortality) through intermediate and even to susceptible (body weight in lower 20 per cent of weight range, more than 25 per cent mortality). However, classification of the P_1 individuals was based on the average of their F_1 progeny performances. The criteria of selection of the F_1 breeders were pedigree estimates of the genotypes of the individuals. The decision to keep or reject a bird for breeding was based on the average of its sibs' two-week weight and mortality records. The estimated intensity of resistance for any one individual was in all probability not equal to the intensity of resistance of its sibs. Thus, individual merit of those selected was lowered every time a breeder, which would be rejected by progeny test results, was accepted because it had unusually excellent sibs.

Therefore, it is expected that the F_1 breeders would also demonstrate heterogeneity with respect to producing chicks that would have relatively equal weights at two weeks of age when fed a vitamin B_{12} deficient diet. From the analyses of the progeny tests of Experiment II, the pedigree estimates of male R_2 and male S_2 were found to be erroneous. Fortunately, however, they were interchangeable, R_2 became S_2 and S_2 became R_2.

The data from a study by Bird, et al., (1947) showed that the amounts of vitamin B_{12} stored in eggs at hatching time varied. These researchers suggested that the variation within a family in the ability of chicks to withstand a vitamin B_{12} deficiency was due to heredity.

All other environmental factors were relatively equal for all chicks or controlled by statistical treatment. It is suggested that the variability of body weights within families is due either to a lack of uniform deposition by the hen of the vitamin in the egg, an inherited characteristic in the chick affecting the efficiency of vitamin B_{12} utilization, or a combination of both. Since the deposition of the vitamin in the egg is affected only by the female, the difference in the progeny performance of the R males and the S males can be ascribed solely to heredity if all other effects, including that of the females, can be statistically eliminated.

The analyses of variance of chick weights from Experiment I are presented in Tables 2, 3 and 4. Each of the three analyses of variance has a common estimate of error, estimate of variance between hens, estimate of variance between hatches, and total variance. The approach of the analysis of variance in Table 2 is to facilitate the comparison of unlike sires. The approach of the analysis of variance in Table 3 is to facilitate the comparison of unlike pens. In Table 4, the approach of the analysis of variance is to facilitate the remaining comparison of unlike matings.

The above description of Tables 2, 3, and 4 of Experiment I also describes respectively Tables 5, 6, and 7 of Experiment II.

Some elaboration on the terminology used in the analyses of variance is warranted. Variation between the performances by progeny from hens of a pen is referred to as variation between hens. Variation in the performance by progeny from a specific pen mating of one hatch and the performance by progeny of that same specific pen mating of the next consecutive hatch is referred to as variation between hatches. Variations in the performances by progeny of females of one pen and the performance by progeny of the females of another pen is referred to as variation between pens. The variation in performance by progeny of males is referred to as variation between sires. The variation of performance by progeny of particular types of matings is referred to as variation between matings.

I. VARIANCES BETWEEN HENS WITH RESPECT TO THEIR PROGENY PERFORMANCES

Eight of the thirty-two analyses of variance of Experiment I showed no significant difference in the variance between hens of a pen, and seven of the thirty-two analyses of variance of Experiment II data showed no significant difference in the variance between hens of a pen. These non-significant differences in the variance between performances of hens of a pen, even though these hens had been selected by pedigree estimates, show the relatively homozygosity among hens with respect to producing offspring of relatively equal average weights at two weeks of age on a vitamin B_{12} deficient diet.

In the final analysis of variance of the data of Experiment I and of the data of Experiment II (Tables 2, 3, 4, 5, 6 and 7) the individual sum of squares of each analysis of variance were totaled and divided by the total degrees of freedom to give an estimate of average variance between hens. In Experiment I (Tables 2, 3 and 4) the estimate of average variance between hens was 1271.0, which was shown to be highly significant ($F = < 0.01$). The combined degrees of freedom were 351. This figure was derived from the averaged eleven hens from each duplicated mating of the sixteen different matings. In Experiment II the estimate of the average variance between hens was 1247.0 with 359 degrees of freedom. This was shown to be highly significant ($F = < 0.01$, Tables 5, 6 and 7).

Two factors are suggested as causing these highly significant variances between hens. Firstly, the lack of homogeneity between individuals selected by pedigree estimates may partially account for this highly significant difference. Secondly, individual females varied with respect to the amounts of vitamin B_{12} in their eggs at hatching time and this may, in part, contribute to this variance.

II. VARIATIONS BETWEEN HATCHES

The pooling of the progeny weight data from each of the sixteen specific matings of one hatch with the data from each of the same sixteen specific matings of the next consecutive hatch gave estimates of variance between hatches. Thirteen of the sixteen new estimates of Experiment I showed that no significant differences between hatches existed. Six of the sixteen new estimates of Experiment II also showed no significant differences between hatches. In both Experiments I and II, the data of the new estimate were combined and divided by the combined sixteen degrees of freedom to obtain an estimate of average variance between hatches. In Experiment I, the estimated average variance between hatches was 1000.0, which was shown to be highly significant ($F = < 0.01$, Tables 2, 3 and 4), and in Experiment II, the estimate of average variance between hatches of 2403.0 was found to be highly significant ($F = < 0.01$, Tables 5, 6 and 7).

A suggested explanation for the highly significant variance between hatches is that in each of the two consecutive hatches the representation of chicks was not from the same eleven females of the pen. Since the differences between hens has been demonstrated to be highly significant, it can therefore be expected that there will be significant variations between hatches.

III. APPROACH NO. 1, VARIATIONS BETWEEN PENS

We next proceed to the source of variation between like pens of the diallel matings of Experiments I and II, (Table 2 and 5, respectively). The individual comparisons (Comparisons 8 to 15, inclusive) of like pens are illustrated in Figure 2.

Highly significant differences in the variance between pens in Experiment I are presented in Table 2 (Comparisons 8, 9, 10, 12, 13, 14, and 15). In Comparison 11, male R_2 x females S_a produced 88 chicks which had an average weight of 61.1 grams and 32 per cent mortality. Male R_2 x females S_b produced 113 chicks which had an average weight of 61.4 grams and 23 per cent mortality. No significant differences in the variance between these pens were found to exist.

In the final analysis of variance (Table 2), the sum of squares of these eight comparisons were pooled and divided by the eight degrees of freedom to obtain the mean square of 4219.0. This estimate of the average variance between pens was found to be highly significant ($F = < 0.01$).

We now turn our attention to a consideration of the final analysis of variance of Experiment II (Table 5).

Comparisons 13, 14 and 15 showed that the differences between like pens were highly significant ($F=<0.01$). Comparisons 8, 9, 10, 11 and 12 showed that the differences between like pens were non-significant. The results in the comparison of like pens wherein highly significant differences were shown to exist in another comparison are conflicting, i.e., results of Comparison 12 as compared with results of Comparison 13 (Table 5). In Comparison 12, the individuals of the S_3 x S_e mating produced 154 chicks averaging 66.4 grams and had a 10 per cent mortality. The individuals of the S_3 x S_f mating produced 114 chicks which averaged 64.9 grams and had a 15 per cent mortality. The progeny weights of Pen S_e did not differ significantly from the progeny weights of Pen S_f. In Comparison 13, male S4 mated to females S_e produced 130 chicks which averaged 52.2 grams and suffered 32 per cent mortality. Male S_4 mated to females S_f produced 130 chicks which averaged 43.1 grams and had a 54 per cent mortality. This showed that Pen S_e differed to a highly significant degree ($F=<0.01$) from Pen S_f.

In the final analysis of variance for Experiment II, (Table 5), the sum of squares of these eight comparisons were pooled and divided by the eight degrees of freedom obtaining the mean square of 1831.0. This estimate of the average variance between pens was found to be highly significant ($F=<0.01$).

It is hypothesized that the lack of genetic homozygosity of the hereditary factors for resistance to this deficiency within the male may well be the cause of this contradiction. S_3 appears more homozygous than does male S_4. The lack of homogeneity between breeders and differences in vitamin B_{12} storage in the egg may also affect the resulting variance between like pens. Although differences between consecutive hatches have been eliminated from the estimate of error, there still remains the difference between diallel matings of non-consecutive hatches, which may also have an effect on the estimate of variance between pens.

IIIa. APPROACH NO. 2, VARIATIONS BETWEEN LIKE SIRES

The data from the analyses of variance which gave estimates of variance between hatches were pooled in the manner illustrated in Figure 3. This facilitated comparisons of like sires of the various diallel matings.

The new estimates of variance between like sires for the sires of Experiment I and sires of Experiment II are given in Comparisons 23 to 30 inclusive, (Tables 3 and 6, respectively). As shown in Table 3, only three of the eight comparisons showed non-significant differences in the variations between like sires. Similarly, in Table 6, only three of the eight comparisons showed non-significant differences in the variations between like sires.

These results are conflicting, i.e., in Table 3, Comparison 28 showed a non-significant difference in the variation between sires S_1 and S_2. Individuals in the $S_1 \times R_d$

mating produced 42 chicks which had an average weight of 61.1 grams and 36 per cent mortality. The individuals of the S_2 x R_d mating produced 121 chicks which had an average weight of 57.5 grams and 31 per cent mortality. In Comparison 29, S_1 x S_a individuals produced 126 chicks which averaged 53.1 grams and exhibited 40 per cent mortality. The 145 chicks produced by the individuals of the S_2 x S_a mating had an average weight of 64.7 grams and had 30 per cent mortality. In the comparison of the variances of S_1 and S_2 highly significant differences were found ($F = < 0.01$).

These same conflicting results are shown in Table 6, i.e., in Comparison 25, variances between S_3 and S_4 were found to be highly significant in the matings S_3 x R_g vs. S_4 x R_g. In Comparison 26, variance between S_3 and S_4 in the matings S_3 x R_h vs. S_4 x R_h were non-significant.

The pooling of the statistics from the eight comparisons gave an estimate of the average variance between like sires. Table 3 shows an estimate of average variance between like sires to be 5397.0 with 8 degrees of freedom. This variance is highly significant ($F = < 0.01$). The estimate of average variance between like sires of Experiment II is 9363.0 with 8 degrees of freedom, as shown in Table 6. This is also a highly significant difference ($F = < 0.01$).

The suggested reasons for the significant differences in the variation between like sires is the same as those given in Section III for variations between like pens.

IV. APPROACH NO. 1, VARIATIONS BETWEEN LIKE SIRES

The data from the analyses of variance which gave estimates of variance between pens were pooled in the manner illustrated in Figure 2. These combinations facilitated the comparisons of variance between like sires in diallel matings.

The results of the performances of the males used in Experiment I and the males used in Experiment II are given in Tables 2 and 5, respectively, Comparisons 4 to 7, inclusive. Each of these individual comparisons showed highly significant differences in the variation between like sires ($F = < 0.01$). In Table 2, the estimate of average variance between like sires of 9989.0 with 4 degrees of freedom is highly significant ($F = < 0.01$). These results conform with theoretical expectations. In Table 5, the estimate of average variance between like sires is 16,591.0 with 4 degrees of freedom and is highly significant ($F = < 0.01$).

IVa. APPROACH NO. 2, VARIATIONS BETWEEN LIKE PENS

The data from the analyses of variance which supplied estimates of variance between like sires were pooled in the manner illustrated in Figure 3. These pooled statistics facilitated the comparison of variance between two like pens, with two males mated to the females in each pen. In each like pen, discussed in Section III, only one male was involved.

The results of the comparisons of the pens in Experiment

I and the comparisons of the pens in Experiment II are shown in Table 3 and Table 6, respectively.

Comparison 19 (Table 3) summarizes the results of the R_{12} x R_e mating which produced 347 chicks which had an average weight of 71.3 grams and 13 per cent mortality, and the R_{12} x R_d mating which produced 320 chicks which averaged 69.7 grams and had a 33 per cent mortality. The differences in the variations between R_e and R_d were non-significant. Again, these are conflicting results. In Comparison 20 (S_{12} x R_e vs. S_{12} x R_d), highly significant variations were found ($F=<0.01$).

These four estimated variances were pooled and 7634.0 was obtained for the estimate of average variance between like pens of Experiment I. This figure was shown to be highly significant ($F=<0.01$).

Comparisons 19, 20 and 21 (Table 6, Experiment II) showed that the differences between like pens was also non-significant. However, when these variances were pooled the estimate of average variance between like pens of 1526.0 with 4 degrees of freedom and was found to be highly significant ($F=<0.01$).

V. APPROACH NO. 1, VARIATIONS BETWEEN UNLIKE MATINGS

The data from the analyses of variance which gave estimates of variance between like sires (Section IV) were further pooled to facilitate estimating the variance between unlike matings. The system of pooling the data is illustrated in Figure 2.

In Experiment I (Table 2) the matings represented by R_{12} x R_{cd} produced 667 chicks which had an average weight of 70.5 grams and 12 per cent mortality. The R_{12} x S_{ab} matings produced 455 offspring which had an average weight of 65.4 grams and 17 per cent mortality. The difference of 5.1 grams between the average weights of the progeny from the individuals in the two unlike matings resulted in an estimated variance of 7091.0. This variance was demonstrated to be highly significant ($F = < 0.01$, Comparison 2). Theoretically, it is expected that the progeny from the R x R matings would have a significantly higher average weight than the average weight of the progeny produced by the R x S matings.

In Comparison 3 (Table 2), the S_{12} x S_{ab} individuals in the matings produced 535 progeny which had an average weight of 53.6 grams and 42 per cent mortality. The individuals in the S_{12} x R_{cd} matings produced 308 chicks which had an average weight of 64.1 grams and 23 per cent mortality. The calculated estimate of variance between the two unlike matings was 21,635.0. The difference of 10.5 grams in the average weights of their chicks was shown to be highly significant ($F = < 0.01$). It was concluded that theoretical expectations had been realized. Further, when these two estimates of variance were combined and divided by the 2 degrees of freedom, the estimate of average variance between unlike matings was 14,363.0. This variance was found to be highly significant ($F=<0.01$).

In Experiment II (Table 5, Comparison 2) the individuals in the R_{34} x R_{gh} mating produced 536 progeny which had an average weight of 71.8 grams and 11 per cent mortality, as compared with the individuals in the R_{34} x S_{ef} matings that produced 511 chicks with an average weight of 71.3 grams and 10 per cent mortality. The difference in average weight was 0.5 grams. The calculated estimate of variance between these unlike matings showed that this difference was non-significant. These findings are not in agreement with the expected results. The effect of the varied amounts of vitamin B_{12} in the eggs from different hens at hatching time and the effect of heterozygosity among breeders may well have caused this deviation from the expected.

In Comparison 3 (Table 5), the individuals in the S_{34} x S_{ef} matings produced 528 chicks. Twenty-seven per cent of these chicks died. When these data were compared with the data obtained from the S_{34} x R_{gh} matings (460 chicks, average weight of 60.8 grams and 25 per cent mortality) the difference of 3.9 grams in the average body weight was found to be highly significant ($F=<0.01$). This difference was theoretically expected. When the mean squares were combined and averaged, the estimate of variance between these unlike matings of 1907.0 with 2 degrees of freedom was also shown to be highly significant ($F=<0.01$).

Va. APPROACH NO. 2, VARIATIONS BETWEEN UNLIKE MATINGS

The data from the analyses of variance used to obtain estimates of variance between like pens (Section IVa) were pooled in the manner illustrated in Figure 3. The purpose of this approach was to facilitate the estimation of variance between unlike matings.

In Comparison 17 (Table 3, Experiment I), the individuals in the R_{12} x R_{cd} matings produced 667 offspring. These chicks had an average body weight of 70.5 grams and a mortality rate of 12 per cent. When compared with the S_{12} x R_{cd} matings, the individuals of which produced 308 chicks which averaged 64.1 grams in body weight and had a mortality of 23 per cent, the difference of 6.4 grams in average body weight in favor of the chicks produced by the individuals of the R_{12} x R_{cd} matings was highly significant ($F=<0.01$) is in accord with theoretical expectations.

In Comparison 18 (Table 3, Experiment I), the individuals in the S_{12} x S_{ab} matings produced 535 chicks which had an average body weight of 53.6 grams and a mortality rate of 42 per cent. The individuals in the R_{12} x S_{ab} matings produced 455 chicks. These chicks averaged 65.4 grams in body weight and 17 per cent of the chicks died. The difference of 11.8 grams in average body weight in favor of the chicks produced by the latter matings was highly significant ($F=<0.01$).

When the two mean squares of the individual comparisons were combined and averaged, highly significant differences ($F = < 0.01$) were found in the variance between unlike matings.

The unlike matings of Experiment II (Table 6) were found to differ to a highly significant degree ($F = < 0.01$). The 536 offspring produced by the individuals in the R_{34} x R_{gh} matings (Comparison 17) had an average body weight of 71.8 grams and a mortality rate of 11 per cent. The 460 progeny from the individuals in the S_{34} x R_{gh} matings weighed 11 grams less (60.8 grams) and 25 per cent of the chicks died. This difference in the average body weight of the chicks from the individuals of the two different matings was highly significant ($F = < 0.01$). In Comparison 18, the individuals of the S_{34} x S_{ef} matings produced 528 chicks. The average body weight of the chicks was 71.3 grams and 10 per cent of these chicks succumbed. The comparison of these unlike matings showed that the difference of 14.4 grams in average body weight was highly significant ($F = < 0.01$). The estimate of average variance between these unlike matings of 42,283.0 with 2 degrees of freedom was also highly significant ($F = < 0.01$).

Vb. APPROACH NO. 3, VARIATIONS BETWEEN UNLIKE MATINGS

The data of the analyses of variance used to obtain estimates of variance between like sires (Section IV) were pooled in the manner illustrated in Figure 4. From these pooled data estimates of variance between unlike matings were obtained.

In the comparison of the $R_{12} \times R_{cd}$ matings (Table 4, Comparison 2), the individuals of which produced 667 chicks (average weight of 70.5 grams, 12 per cent mortality), with the $S_{12} \times S_{ab}$ matings, the individuals of which produced 535 chicks (average weight of 53.6 grams, 42 per cent mortality), highly significant differences were found ($F=<0.01$). Of all the comparisons of the performance of progenies from unlike matings, these matings were theoretically expected to produce offspring which showed the greatest differences in average body weights and mortality.

In Comparison 33 (Experiment I, Table 4) and Comparison 33 (Experiment II, Table 7), which involved reciprocal matings of the two parental lines, i.e., $R_{12} \times S_{ab}$ vs. $S_{12} \times R_{cd}$ and $S_{34} \times R_{gh}$ vs. $R_{34} \times S_{ef}$, conflicting results were obtained.

In Experiment I, the differences between the average body weights of the chicks produced by the individuals of the two contrasting matings were non-significant. In Experiment II, the average body weight differences of the chicks produced by the individuals in the reciprocal matings were highly significant ($F=<0.01$).

It is suggested that further experimentation is necessary before reliable conclusions may be drawn regarding the performance of the progenies produced by these reciprocal matings. Stud matings instead of pen matings would have to be practiced.

The remaining unlike matings, S_{34} x S_{ef} vs. R_{34} x R_{gh} of Experiment II are shown in Table 7, Comparison 32. The individuals of the S_{34} x S_{ef} matings produced 528 chicks which had an average body weight of 56.9 grams. Twenty-seven per cent of the chicks died. The individuals of the R_{34} x R_{gh} matings produced 536 offspring which had an average body weight of 71.8 grams. The mortality rate of these latter chicks was 11 per cent. The difference of 14.9 grams in average body weight was highly significant ($F = < 0.01$).

VI. APPROACH NO. 1, VARIATIONS BETWEEN UNLIKE SIRES

Data from the analyses of variance which estimated variances between unlike matings were pooled in a manner illustrated in Figure 2. Estimates of variance between unlike sires were calculated.

In Comparison 1 (Table 2, Experiment I) the individuals in matings R_{12} x R_{cd} and R_{12} x S_{ab} produced 1122 progeny which had an average weight of 68.5 grams and 14 per cent mortality. The individuals in matings S_{12} x R_{cd} and S_{12} x S_{ab} produced 843 chicks which had an average weight of 57.4 grams and 35 per cent mortality. The two-week weights of chicks sired by R_{12} when compared with the two-week weights of chicks sired by S_{12} were found to be highly significant ($F = < 0.01$).

In Comparison 1 (Table 5, Experiment II), the individuals in matings R_{34} x R_{gh} and R_{34} x S_{ef} produced 1047 offspring which had an average weight of 71.6 grams and 10 per cent mortality. The individuals in matings S_{34} x S_{ef} and S_{34} x R_{gh} produced 988 offspring which had an average weight of 58.7 grams and 26 per cent mortality. The difference of 12.9 grams favored the progeny sired by R_{34}. The comparison of the progeny weights of chicks sired by S_{34} and those sired by R_{34} demonstrated highly significant differences ($F=\langle 0.01$).

It is suggested that these variations in the performance of progeny from unlike sires is due solely to hereditary effects.

VIa. APPROACH NO. 2, VARIATIONS BETWEEN UNLIKE PENS

Data from the analyses of variance which estimated variances between unlike matings were pooled in a manner illustrated in Figure 3. From this pooled data estimates of variances between unlike pens were calculated.

In Comparison 16 (Table 3, Experiment I), the individuals in matings R_{12} x R_{cd} and S_{12} x R_{cd} produced 975 chicks which had an average weight of 68.5 grams and 15 per cent mortality. The individuals in matings R_{12} x S_{ab} and S_{12} x S_{ab} produced 990 chicks which had an average weight of 59.0 grams and 30 per cent mortality. A mean square of 44,168.0 was calculated from the weights of chicks from females of the R_{cd} pens and from the weights of chicks from females of the S_{ab} pens. This figure was found to be highly significant ($F=\langle 0.01$).

In Comparison 16 (Table 6, Experiment II), the individuals of pen matings R_{34} x R_{gh} and S_{34} x R_{gh} produced 996 chicks which had an average weight of 66.7 grams and 17 per cent mortality. The breeders of pen matings R_{34} x S_{ef} and S_{34} x S_{ef} produced 1039 chicks which had an average weight of 64.0 grams and 19 per cent mortality.

The difference in body weights of chicks from females of R_{gh} pens and in the body weights of chicks from females of S_{ef} pens is shown to be highly significant ($F = \langle 0.01$).

These results are in accord with theoretical expectations.

VIb. APPROACH NO. 3, VARIATIONS BETWEEN RECIPROCAL AND EXTREME MATINGS

Data from the analyses of variance which estimated variances between unlike matings were pooled in a manner illustrated in Figure 4.

In Comparison 31 (Table 4, Experiment I), the 1202 progeny of the individuals involved in matings R_{12} x R_{cd} and S_{12} x S_{ab} had an average weight of 63.0 grams and a 25 per cent mortality rate. The 763 progeny of the individuals involved in matings R_{12} x S_{ab} and S_{12} x R_{cd} had an average weight of 64.9 grams and 20 per cent mortality. The differences in variance between the average weights of chicks from breeders of the extreme matings (R_{12} x R_{cd} and S_{12} x S_{ab}) and the average weight of chicks from breeders of the reciprocal matings (R_{12} x S_{ab} and S_{12} x R_{cd})

were found to be highly significant (F=$<$0.01).

In Comparison 31 (Table 7, Experiment II), the offspring from matings S_{34} x S_{ef} and R_{34} x R_{gh} numbered 1064. The average weight of these chicks was 64.4 grams and their mortality rate was 19 per cent. The individuals that were in matings S_{34} x R_{gh} and R_{34} x S_{ef} produced 971 chicks which had an average weight of 66.3 grams and 17 per cent mortality. The differences in variance between the average weights of chicks from breeders of the extreme matings (S_{34} x S_{ef} and R_{34} x R_{gh}) and the average weights of chicks from breeders of the reciprocal matings (S_{34} x R_{gh} and R_{34} x S_{ef}) were found to be highly significant (F=$<$0.01).

Theoretically no significant difference would be expected in this comparison if homozygosity between hens and between sires, with respect to the chicks' withstanding of the deficiency to an equal degree, could be demonstrated. This has not been demonstrated; therefore, highly significant differences can not be considered unexpected.

The average weight of progeny of the R males in Experiment I was 19 per cent higher than the average weight of progeny of the S males. The average weight of progeny of the R males in Experiment II was 20 per cent higher than that of the S males. It is again suggested that these variations in the performance of progeny from unlike sires is due solely to genetic effects.

According to Lush (1949) the breeding value of two males involved in diallel matings is twice the difference of the average merit of their offspring. The R males in these experiments were then 40 per cent more resistant to the deficiency of vitamin B_{12} in their diet than the S males. It appears very probable that a relatively resistant strain and a relatively susceptible strain, with respect to a deficiency of vitamin B_{12}, could be developed.

TABLE 2

The results of an analysis of variance of two week weights (gms) of chicks on a vitamin B_{12} deficient diet.

EXPERIMENT I - Comparison of Unlike Sires

S.V.	d.f.	m.s.	Comparison No.	Mating	No. Chicks	Ave. Wt. (gms)	Mortality (%)
Total	1964	1146705			1965	63.7	23
Bet. Unlike Sires	1	**58582	1	R_{12} x R_{cd} R_{12} x S_{ab}	1122	68.5	14
				S_{12} x R_{cd} S_{12} x S_{ab}	843	57.4	35
Bet. Unlike Matings	2	**14363					
	1	** 7091	2	R_{12} x R_{cd} R_{12} x S_{ab}	667 455	70.5 65.4	12 17
	1	**21635	3	S_{12} x S_{ab} S_{12} x R_{cd}	535 308	53.6 64.1	42 23
Bet. Like Sires	4	** 9989					
	1	**11688	4	R_1 x R_{cd} R_2 x R_{cd}	329 338	74.8 66.4	5 18
	1	** 6205	5	R_1 x S_{ab} R_2 x S_{ab}	254 201	68.7 61.3	9 27
	1	**19300	6	S_1 x S_{ab} S_2 x S_{ab}	261 274	47.4 59.4	52 32
	1	** 2761	7	S_1 x R_{cd} S_2 x R_{cd}	136 172	67.5 61.4	21 25
Bet. Like Pens	8	** 4219					
	1	** 6300	8	R_1 x R_c R_1 x R_d	152 77	79.5 70.7	3 7
	1	1076	9	R_2 x R_c R_2 x R_d	195 143	64.9 68.6	19 17
	1	* 1343	10	R_1 x S_a R_1 x S_b	135 119	70.9 66.3	9 10
	1	7	11	R_2 x S_a R_2 x S_b	88 113	61.1 61.4	32 23
	1	** 7909	12	S_1 x S_a S_1 x S_b	126 135	53.1 42.1	40 62
	1	** 8404	13	S_2 x S_a S_2 x S_b	145 129	64.7 53.6	30 34
	1	** 2470	14	S_1 x R_a S_1 x R_d	94 42	70.3 61.1	15 36
	1	** 6248	15	S_2 x R_c S_2 x R_d	51 121	70.7 57.5	10 31
Bet.Hatches	16	** 1000					
Bet.Hens	351	** 1271					
Error	1582	331					

ignificant ghly Significant

TABLE 3

The results of an analysis of variance of two week weights (gms) of chicks on a vitamin B_{12} deficient diet.

EXPERIMENT I - Comparison of Unlike Pens

S.V.	d.f.	m.s.	Comparison No.	Mating	No. Chicks	Ave. Wt. (gms)	Mortality (%)
Total	1964	1146705			1965	63.7	23
Bet. Unlike Pens	1	**44168	16	R_{12} x R_{cd} S_{12} x R_{cd}	975	68.5	15
				R_{12} x S_{ab} S_{12} x S_{ab}	990	59.0	30
Bet. Unlike Matings	2	**21570	17				
	1	** 8715		R_{12} x R_{cd}	667	70.5	12
				S_{12} x R_{cd}	308	64.1	23
	1	**34425	18	S_{12} x S_{ab}	535	53.6	42
				R_{12} x S_{ab}	455	65.4	17
Bet. Like Pens	4	** 7634					
	1	404	19	R_{12} x R_c	347	71.3	13
				R_{12} x R_d	320	69.7	33
	1	**11077	20	S_{12} x R_c	145	70.5	12
				S_{12} x R_d	163	58.4	11
	1	1091	21	R_{12} x S_a	223	67.0	18
				R_{12} x S_b	232	63.9	16
	1	**17965	22	S_{12} x S_a	271	59.3	35
				S_{12} x S_b	264	47.7	48
Bet. Like Sires	8	** 5397					
	1	**18265	23	R_1 x R_c	152	80.0	3
				R_2 x R_c	195	64.9	19
	1	395	24	R_1 x R_d	177	70.7	7
				R_2 x R_d	143	68.9	17
	1	** 5107	25	R_1 x S_a	135	70.9	9
				R_2 x S_a	88	61.1	32
	1	* 1357	26	R_1 x S_b	119	66.3	10
				R_2 x S_b	113	61.4	23
	1	6	27	S_1 x R_c	94	70.3	15
				S_2 x R_c	51	70.7	10
	1	396	28	S_1 x R_d	42	61.1	36
				S_2 x R_d	121	57.5	31
	1	** 8981	29	S_1 x S_a	126	53.1	40
				S_2 x S_a	145	64.7	30
	1	** 8667	30	S_1 x S_b	135	42.1	62
				S_2 x S_b	129	53.6	34
Bet. Hatches	16	** 1000					
Bet. Hens	351	** 1271					
Error	1582	331					

-ignificant ghly Significant

TABLE 4

The results of an analysis of variance of two week weights (gms) of chicks on a vitamin B_{12} deficient diet.

EXPERIMENT I - Comparison of remaining unlike matings

S.V.	d.f.		m.s.	Comparison No.	Mating	No. Chicks	Ave. Wt. (gms)	Mortality (%)
Total	1964		1146705			1965	63.7	23
Bet. Extreme Matings and Reciprocal Matings	1		** 1678	31	R_{12} x R_{cd} S_{12} x S_{ab}	1202	63.0	25
					R_{12} x S_{ab} S_{12} x R_{cd}	763	64.9	20
Bet. Unlike Matings	2		**42815					
		1	**85314	32	R_{12} x R_{cd} S_{12} x S_{ab}	667 535	70.5 53.6	12 42
		1	316	33	R_{12} x S_{ab} S_{12} x R_{cd}	455 308	65.4 64.1	17 23
Bet. Like Sires	4		** 9989					
Bet. Like Pens	8		** 4219					
Bet.Hatches	16		** 1000					
Bet. Hens	351		** 1271					
Error	1582		331					

* Significant ** Highly Significant

TABLE 5

The results of an analysis of variance of two week weights (gms) of chicks on a vitamin B_{12} deficient diet.

EXPERIMENT II - Comparison of Unlike Sires

S.V.	d.f.	m.s.	Comparison No.	Mating	No. Chicks	Ave. Wt. (gms)	Mortality (%)
Total	2034	1115675			2035	65.3	18
Bet. Unlike Sires	1	**84655	1	$R_{34} \times R_{gh}$ $R_{34} \times S_{ef}$	1047	71.6	10
				$S_{34} \times R_{gh}$ $S_{34} \times S_{ef}$	988	58.7	26
Bet. Unlike Matings	2	** 1907					
	1	82	2	$R_{34} \times R_{gh}$	536	71.8	11
				$R_{34} \times S_{ef}$	511	71.3	10
	1	** 3731	3	$S_{34} \times S_{ef}$	528	56.9	27
				$S_{34} \times R_{gh}$	460	60.8	25
Bet. Like Sires	4	**16591					
	1	** 2983	4	$R_3 \times R_{gh}$	246	74.4	9
				$R_4 \times R_{gh}$	290	69.7	13
	1	** 5374	5	$R_3 \times S_{ef}$	303	68.6	13
				$R_4 \times S_{ef}$	208	75.2	5
	1	**43485	6	$S_3 \times S_{ef}$	268	65.8	12
				$S_4 \times S_{ef}$	260	47.6	43
	1	**14522	7	$S_3 \times R_{gh}$	243	66.1	14
				$S_4 \times R_{gh}$	217	54.8	38
Bet. Like Pens	8	** 1831					
	1	46	8	$R_3 \times R_g$	175	74.7	8
				$R_3 \times R_h$	71	73.7	10
	1	67	9	$R_4 \times R_g$	127	69.1	15
				$R_4 \times R_h$	163	70.1	11
	1	339	10	$R_3 \times S_e$	179	67.7	15
				$R_3 \times S_f$	124	69.9	11
	1	360	11	$R_4 \times S_e$	134	76.2	6
				$R_4 \times S_f$	74	73.4	4
	1	155	12	$S_3 \times S_e$	154	66.4	10
				$S_3 \times S_f$	114	64.9	15
	1	** 5420	13	$S_4 \times S_e$	130	52.2	32
				$S_4 \times S_f$	130	43.1	54
	1	** 2675	14	$S_3 \times R_g$	135	69.0	10
				$S_3 \times R_h$	108	62.4	19
	1	** 5585	15	$S_4 \times R_g$	79	48.1	56
				$S_4 \times R_h$	138	58.6	28
Bet.Hatches	16	** 2403					
Bet.Hens	359	** 1247					
Error	1644	280					

Significant ghly Significant

TABLE 6

The results of an analysis of variance of two week weights (gms) of chicks on a vitamin B_{12} deficient diet.

EXPERIMENT II - Comparison of Unlike Pens

S.V.	d.f.	m.s.	Comparison No.	Mating	No. Chicks	Ave. Wt. (gms)	Mortality (%)
Total	2034	1115675			2035	65.3	18
Bet. Unlike Pens	1	** 3902	16	R_{34} x R_{gh} S_{34} x R_{gh}	996	66.7	17
				R_{34} x S_{ef} S_{34} x S_{ef}	1039	64.0	19
Bet. Unlike Matings	2	**42283					
	1	**30482	17	R_{34} x R_{gh} S_{34} x R_{gh}	536 460	71.8 60.8	11 25
	1	**54084	18	S_{34} x S_{ef} R_{34} x S_{ef}	528 511	56.9 71.3	27 10
Bet. Like Pens	4	** 1526					
	1	174	19	R_{34} x R_g R_{34} x R_h	320 234	72.4 71.2	11 11
	1	122	20	S_{34} x R_g S_{34} x R_h	214 246	61.3 60.3	26 24
	1	3	21	R_{34} x S_e R_{34} x S_f	313 198	71.3 71.2	11 9
	1	** 5805	22	S_{34} x S_e S_{34} x S_f	284 244	59.9 53.3	20 36
Bet. Like Sires	8	** 9363					
	1	** 2268	23	R_3 x R_g R_4 x R_g	175 127	74.7 69.1	8 15
	1	654	24	R_3 x R_h R_4 x R_h	71 163	73.7 70.1	10 11
	1	**21828	25	S_3 x R_g S_4 x R_g	135 79	69.0 48.1	10 56
	1	832	26	S_3 x R_h S_4 x R_h	108 138	62.4 58.6	19 28
	1	** 5482	27	R_3 x S_e R_4 x S_e	179 134	67.7 76.2	15 6
	1	588	28	R_3 x S_f R_4 x S_f	124 74	69.9 73.4	11 4
	1	**14296	29	S_3 x S_e S_4 x S_e	154 130	66.4 52.2	10 32
	1	**28959	30	S_3 x S_f S_4 x S_f	114 130	64.9 43.1	15 54
Bet. Hatches	16	**2403					
Bet. Hens	359	**1247					
Error	1644	280					

Significant ghly Significant

TABLE 7

The results of an analysis of variance of two week weights (gms) of chicks on a vitamin B_{12} deficient diet.

EXPERIMENT II - Comparison of remaining unlike matings

S.V.	d.f.	m.s.	Comparison No.	Mating	No. Chicks	Ave. Wt. (gms)	Mortality (%)
Total	2034	** 115675			2035	65.3	18
Bet. Extreme Matings and Reciprocal Matings	1	** 1809	31	S_{34} x S_{ef} R_{34} x R_{gh}	1064	64.4	19
				S_{34} x R_{gh} R_{34} x S_{ef}	971	66.3	17
Bet. Unlike Matings	2	**43330					
	1	**54084	32	S_{34} x S_{ef} R_{34} x R_{gh}	528 536	56.9 71.8	27 11
	1	**30482	33	S_{34} x R_{gh} R_{34} x S_{ef}	460 511	60.8 71.3	25 10
Bet. Like Sires	4	**16591					
Bet. Like Pens	8	** 1831					
Bet.Hatches	16	** 2403					
Bet.Hens	359	** 1247					
Error	1644	280					

* Significant ** Highly Significant

SUMMARY AND CONCLUSIONS

It was hypothesized that the two-week body weight of the individual chick on a vitamin B_{12} deficient diet is a phenotypic expression of its genetic constitution and that this genetic makeup may be modified by an uncontrollable environmental factor, namely vitamin B_{12} storage in the egg at hatching time. No attempt was made to determine or even suggest any genotypic identification. This was a study of progeny tests of pedigree-selected breeders, the results of which evaluated the breeders' worth.

The data of 4000 chick weights were collected from two experiments, each involving four pens of 20 females and a male. The plan of matings simulated the design of a 4 x 4 Latin Square, and further, within the Latin Square, diallel mating designs were used.

It was demonstrated that there were variations within breeders with respect to producing offspring that had relatively equal weights at two weeks of age on a vitamin B_{12} deficient diet. Highly significant differences in variations were found between hens with respect to their ability to produce families giving relatively equal performances in withstanding this dietary deficiency.

Highly significant differences were found between the average body weights of chicks from consecutive hatches. Highly significant differences were found in the variances between progenies of individuals of like pens and highly significant differences were found in the variances between offspring of like sires. It may be concluded from these significant differences that there is a lack of homogeneity between breeders of similar classifications.

The four types of matings involved in each experiment were R x R, R x S, S x R and S x S. Of the six possible comparisons which were made of these data in Experiment I, all conformed to theoretical expectations. The R x R progeny had a 16.9 gram higher weight and 30 per cent lower mortality than the S x S progeny.

In Experiment II, similar comparisons were made of the six possible comparisons of unlike matings of that experiment. R x S vs. S x R show highly significant differences. S x R vs. R x R showed non-significance. These two deviations from the expected are due, in general, to the lack of homogeneity among the breeders of one classification with respect to resistance to the vitamin B_{12} dietary deficiency.

The R hens of Experiment I had progeny with an average weight 16 per cent above the average progeny weight of the S hens. The R hens of Experiment II had progeny with an average weight only 4 per cent above the average weight of progeny of the S hens. It is suggested that these differences are due to a combination of genetic effects transmitted by the hen and those effects due to varied amounts of vitamin B_{12} in eggs of hens at hatching time.

Since the deposition of the vitamin in the egg is affected only by the female, the difference in the progeny performance of the R males and the S males can be ascribed solely to heredity if all other effects, including that of the females, can be statistically eliminated. In these experiments, they were eliminated. The R males of both experiments were 40 per cent more resistant to the deficiency than the S males. It may be concluded that these differences are hereditary and it is further concluded that it is highly probable that a strain relatively resistant and a strain relatively susceptible to a vitamin B_{12} dietary deficiency could be developed.

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