INHERITANCE OF AGE AT SEXUAL MATURITY IN THE DOMESTIC FOWL¹

D. C. WARREN

Kansas Agricultural Experiment Station, Manhattan, Kansas

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THE PROBLEM

Dr. RAYMOND PEARL (1912) was the first worker to suggest that sex-linked factors were involved in the inheritance of fecundity. PEARL used as a measure of fecundity the eggs produced during the winter period. He proposed that two major dominant factors were responsible for high egg production, one of which resided in the sex chromosome. GOODALE (1919) and HURST (1921) each failed to confirm the earlier statement of PEARL that a sex-linked factor was involved in the inheritance of fecundity.

GOODALE (1919) called attention to the inadequacy of PEARL's method of attacking the problem of the inheritance of fecundity. He pointed out that there were a number of more or less independent genetic factors responsible for high egg production and that PEARL's method of considering as a unit the total number of eggs produced could not be expected to yield critical results. GOODALE (1922) held that the annual egg record was dependent upon the combined action of the following five characteristics: maturity, intensity, broodiness, persistency, and winter pause.

HAYS (1924) proposed that maturity (age at sexual maturity) was dependent upon two pairs of factors, one of which was sex-linked. He held that the early maturity factor was dominant in each case. The data presented in support of the theory are rather meager. HAYS has suggested that since PEARL's study of inheritance of fecundity utilized only egg production during the winter period, he was probably considering primarily age at sexual maturity. It is readily seen that differences in age at which birds started to lay would materially influence the number of eggs produced in that period.

PUNNETT (1930) outlined what he considered a crucial test for the theory of PEARL that sex-linked factors were involved in the transmission of high fecundity. It had been shown previously that the two genes, silver-gold and barred-non-barred were located on the sex chromosome about 45 units apart (AGAR 1924, SEREBROVSKY and WASSINA 1927, WARREN 1928). PUNNETT stated that if these two genes and the one for fecundity were all on the sex chromosome, through segregation some evidence for linkage among them should be found. In following the segregation of these

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three factors through a male heterozygous for all three, he found no evidence for linkage. His conclusions were based upon the mean egg production of birds segregating for the silver-gold and barred-non-barred pairs of factors. Since he found no pronounced difference in the mean egg production of the various color segregates, PUNNETT concluded that there was no evidence for sex-linked factors influencing egg production. In view of the rather general acceptance of the opinion that the inheritance of the egg production is too complicated to consider the annual record as a unit; PUNNETT's method of attack appears somewhat obsolete. This is especially true when we consider the fact that the only characteristic which has been proposed to show sex-linked inheritance is age at sexual maturity. Even though age at sexual maturity were sex-linked the influence of this factor on the annual record could easily have been obscured by the other factors influencing egg production, especially in view of the small numbers with which he dealt.

EXPERIMENTAL RESULTS

Selection

During the six-year period, 1926-1931, selection was carried out on Single Comb Rhode Island Reds at the KANSAS AGRICULTURAL EXPERIMENT STATION for the establishment of strains differing in age at which they reach sexual maturity. Very little new stock was obtained during the period of the experiment so that the flock remained relatively homogeneous. Age at sexual maturity was measured by the age attained at the laying of the first egg. This limited to females the classification with regard to age at sexual maturity since no satisfactory measure was found which would apply to both sexes. In carrying out selection a somewhat inaccurate estimate was made of the age at sexual maturity in the males, being based upon the plumage and comb development at six months of age. This classification of the males was not used in measuring the progress of selection but probably did increase the effectiveness of selection.

Throughout the period of the experiment care was exercised to house the pullets early enough to obtain an accurate record of the age at first egg. Most of the pullets were hatched between February 15 and April 15 of each year thus avoiding the influence of wide seasonal differences on maturity. The young stock during this period was relatively free from diseases and parasites which might influence the age of attainment of sexual maturity. It is realized, however, that even under the best conditions the health of the bird will influence the onset of production and is probably a source of considerable error which could not be entirely avoided. The pullets were closely observed during the early part of the laying year and any bird showing an inclination to lay eggs outside the trapnests was

eliminated from consideration because of likelihood of error in the record as to date of starting.

During the 1926 season six matings were made for the study of maturity, three being selections for later maturity and three for earlier maturity. In the selection for early maturity, early maturing females were mated to males from early maturing mothers. In each case care was exercised to use for the most part individuals from early averaging families. The same rule held for the late maturity matings and as selection progressed the use of individuals of proved breeding qualities was stressed.

In the selection of birds to be used for continuation of the early and late maturity strain, no individual maturing at an age of more than 225 days was used in the early strain and none having maturity age of less than 235 days was considered for the late strain. The difference in maturity age for the females used in the two strains was usually much greater than the limits mentioned.

Although the data recorded in table 1 show the daughters of the late maturity matings in 1926 to have a mean age of 7.2 days greater than

TABLE 1
Selection for early and late maturity in Rhode Island Reds.

YEAR	EARLY MATURITY STRAIN		LATE MATURITY STRAIN		DIFFERENCE IN MATURITY AGE IN DAYS	DIFFERENCE
	NUMBER OF DAUGHTERS	MEAN MATURITY AGE IN DAYS	NUMBER OF DAUGHTERS	MEAN MATURITY AGE IN DAYS		p.e.
1926	89	269.0±2.00	85	276.2±1.70	7.2±2.6	1:2.8
1927	96	230.8±2.70	84	249.5±3.29	18.7±4.3	1:4.3
1928	73	221.9±3.55	97	247.6±2.85	25.7±4.6	1:5.6
1929	92	211.5±3.13	78	252.3±3.84	40.8±5.0	1:8.2
1930	20	216.7±4.65	29	249.3±4.38	32.6±6.4	1:4.9
1931	45	222.2±5.13	71	269.0±3.37	46.8±6.1	1:7.7

those from the early maturity mating, the difference was not sufficiently large to have statistical significance. During the second year (1927) there was a much better opportunity to apply the progeny test within the late and early maturity strains. This resulted in a greater difference in the mean age at which the two strains matured. The 96 female offspring of the four early maturity matings had a mean age at the time of laying of the first egg of 230.8 days while 84 pullets from the three late maturity matings began laying at a mean age of 249.5 days. The difference in age is 18.7 days or 4.3 times its probable error.

After the third year of selection (1928) there was very little change in the mean maturity age of either strain. In fact there was very little change secured in the late maturity strain, practically all of the difference between the two strains resulting from the reduction of the mean age at laying in the early maturity strain,

During 1930 and 1931 when it became evident that further change in the maturity of the two strains was not probable, an increasing portion of the facilities was given over to the crossing of the two strains. This accounts for the reduction in number of females tested in the two strains during the last two years. By this time it seemed to be well demonstrated that a heritable difference in age at sexual maturity had been brought about by selection. The next step undertaken was to analyze the mode of inheritance of the factor or factors involved.

Reciprocal matings of the early and late maturity strains

During the 1930 and 1931 breeding seasons reciprocal matings were made between the early and late maturing strains. A comparison of the age at sexual maturity of the female offspring of reciprocal matings should indicate whether sex-linked factors influenced the age at which pullets started to lay. An equal number of pens were headed by males from the late and early maturity strains and each pen carried females not only from the strain to which the male belonged but also from the other strain. By this method each male was checked upon by matings within his own strain at the same time that he was being out-crossed to females of a different maturity age.

It chanced during the first year (1930) in which crosses were made between the early and late maturity strains, that the difference between the pure early and late strain matings was less than those for the preceding or succeeding years. There were two matings of early maturity males by late maturity females and three of late maturity males by early maturity females. The 31 female offspring (table 2) from the former mating had a

TABLE 2
F₁ generation results from crossing early and late maturity strains of Rhode Island Reds.

YEAR	EARLY MATURITY MALE BY LATE MATURITY FEMALE				LATE MATURITY MALE BY EARLY MATURITY FEMALE			
	MALE BAND	NUMBER OF	NUMBER OF	DAUGHTERS' MEAN	MALE BAND	NUMBER OF	NUMBER OF	DAUGHTERS' MEAN
	NUMBER	MATES	DAUGH- TERS	MATURITY AGE	NUMBER	MATES	DAUGH- TERS	MATURITY AGE
1930	1082M	4	28	221.8 ± 2.69	1081M	1	4	245.8 ± 9.57
	1088M	1	3	231.3 ± 10.70	1083M	3	20	247.0 ± 4.57
					1084M	3	18	214.3 ± 5.56
Total and mean			31	222.7 ± 2.66			42	232.9 ± 3.75
1931	1086M	1	11	200.8 ± 8.57	1083M	2	10	223.6 ± 9.39
	1128M	3	13	230.8 ± 8.87	1130M	2	7	245.7 ± 12.19
	1131M	2	12	219.7 ± 9.67	1132M	4	34	231.5 ± 4.80
					1133M	3	29	268.7 ± 6.76
Total and mean			36	217.9 ± 5.42			80	244.8 ± 3.83

mean age of 222.7 days at the laying of the first egg while the latter mating produced 42 daughters that had a maturity age of 232.9 days. If sex-linked factors influenced the age at sexual maturity it would be expected that the female offspring of the latter type of mating would show later maturity than those of the former. The difference (10.2 ± 4.59) is in the expected direction but its probable error is too large for it to have much significance.

The second year's (1931) results were much more definite. The 36 daughters of the mating of the early maturity male by late maturity female had a mean age of 217.9 days and the reciprocal cross produced 80 female offspring which had a maturity age of 244.8 days. The difference between the two types of matings was 26.9 days which was 4.1 times its probable error. The fact that the mating of males from the early maturity strain to females from the late maturity strain produced daughters starting to lay at an earlier age than did those from the pure early maturity mating for that year is of interest. The age at sexual maturity for these cross-strain females was very similar to the pure early-strain birds during the last few years of selection. This would indicate that the major factorial difference was sex-linked or that any autosomal factors for late maturity must be completely recessive. The results for the mating of late maturity males to early maturity females were not so similar to those from the pure late maturity matings. This mating produced daughters which had a mean age of 244.8 days at the laying of the first egg while the pure late maturity matings for the same season gave daughters with a mean maturity age of 269 days. These results would tend to support the view that autosomal factors were also involved and that the influence of the autosomal and sex-linked factors were cumulative. The intermediate maturity here may be interpreted as a result of the conflicting effects of the sex-linked and autosomal factors. HAYS has suggested that both autosomal and sex-linked factors are involved in the inheritance of age at sexual maturity and in referring to the early maturity factor stated that "either combined or alone, simplex or duplex, these factors produce a bird that normally begins to lay at 215 days or less." A comparison of the results given in table 2 would indicate that a female carrying the sex-linked factor for late maturity (and having only one sex chromosome she can only carry it in a single dose) and being heterozygous for the autosomal factor or factors, matures somewhat earlier than one having the same sex chromosomal constitution and being homozygous for late maturity. Thus it would appear that the expression of the early maturity gene is in certain combinations dependent upon the individual's constitution with regard to the other genes influencing maturity age.

Backcross generation

The backcross results further substantiate the conclusions drawn from the F_1 generation results. Two types of matings were made and in each case the male was from the early maturity strain and was mated to females from the reciprocal F_1 crosses. If we assume that a sex-linked and an autosomal factor are involved, then the one type of F_1 generation female used would have the constitution $E E'e'$ and the other the constitution $e E'e'$. In the foregoing use of symbols (E) is the sex-linked factor and (E') the autosomal factor. The 132 female offspring from the backcross of F_1 generation females from the cross of early maturity males by late maturity females, by males from the early maturity strain, had a mean maturity age of 214.4 ± 2.17 days. F_1 generation females (249 birds) from the reciprocal cross when backcrossed to males from the early maturity strain had a mean maturity age of 217.5 ± 1.54 days. There was found to be no significant difference between the two types of backcrosses. The maturity age obtained from these matings is very similar to that characteristic of the pure early maturity strain. The duplication here of the mean of the pure early maturity strain substantiates the conclusion drawn from the F_1 generation that when a female carries the sex-linked factor for early maturity it makes no difference whether she is heterozygous or homozygous for the autosomal factor for early maturity. In each type of backcross half the female offspring would be homozygous for the autosomal factor for early maturity and half heterozygous for this factor. If the effect of the autosomal factor had been different in a homozygous or heterozygous condition when in combination with the sex-linked early maturity factor, the mean age here should have been greater than for the pure early strain and there should have been a bimodal shape to the distribution curve for the female offspring in the backcross generation. The maturity ages of the female offspring from the larger (backcross of female from cross of late male by early female) of the two backcross series are presented in the form of a distribution in the following table:

1. Maturity age—170 days or less.....	5
2. Maturity age—171–190 days.....	47
3. Maturity age—191–210 days.....	67
4. Maturity age—211–230 days.....	71
5. Maturity age—231–250 days.....	24
6. Maturity age—251–270 days.....	12
7. Maturity age—271–290 days.....	8
8. Maturity age—291–310 days.....	8
9. Maturity age—311 days or more.....	7
Total.....	249

There is here no evidence of any bimodality. The curve would not descend rapidly in the later maturity classes but this increase in number of the slowest maturing birds is probably due to factors other than genetic.

The results of the backcross and F_1 generation support the hypothesis of Hays in indicating that both sex-linked and autosomal factors are involved in determining the age at which birds reach sexual maturity. The results obtained in the F_1 generation indicate rather strongly that Hays was in error in believing that either the sex-linked or the autosomal factor could when acting alone produce early maturity. No birds were obtained in which the sex-linked early maturity factor was acting alone but the cross of a late maturity male to early maturity females produced female offspring of the constitution $e E'e'$ which had a mean maturity age of 244.8 days while pure early maturity females ($E E'E'$) matured in 222.2 days

TABLE 3

Comparison of maturity age of daughters of individual males when mated to females of their own strain and to females of differing maturity age.

YEAR	MALE BAND NUMBER	LATE MATES		EARLY MATES	
		NUMBER OF DAUGHTERS	DAUGHTERS' MEAN MATURITY AGE	NUMBER OF DAUGHTERS	DAUGHTERS' MEAN MATURITY AGE
1930	Late males				
	1081M	11	244.4 ± 6.5	4	245.8 ± 9.6
	1083M	6	258.8 ± 8.2	20	247.0 ± 4.6
	1084M	12	249.0 ± 7.5	18	214.3 ± 5.6
	Total and mean	29	249.3 ± 4.4	42	232.9 ± 3.8
#	Early males				
	1082M	28	221.8 ± 2.7	9	219.7 ± 5.5
	1088M	3	231.3 ± 10.7	11	214.2 ± 7.1
	Total and mean	31	222.7 ± 2.7	20	216.7 ± 4.7
1931	Late males				
	1083M	21	268.5 ± 6.0	10	223.6 ± 9.4
	1130M	8	278.9 ± 10.9	7	245.7 ± 12.2
	1132M	36	261.8 ± 4.5	34	231.5 ± 4.8
	1133M	6	296.0 ± 12.2	29	268.7 ± 6.8
	Total and mean	71	269.0 ± 5.1	80	244.8 ± 3.8
	Early males				
	1086M	11	200.8 ± 8.6	19	228.9 ± 8.4
	1128M	13	230.8 ± 8.9	20	201.8 ± 3.9
	1131M	12	219.7 ± 9.7	6	269.0 ± 18.1
	Total and mean	36	217.9 ± 5.4	45	222.2 ± 5.1

and pure late maturity females ($e e'e'$) matured in 269.0 days. This tendency is noted from the data in table 3 where a comparison is made of the results from individual males when mated to both early and late maturing females. The results of the two generation crosses seem to give the following mean maturity ages for the various genotypes in the females.

$E E' E'$ —222 days maturity

$e e' e'$ —269 days maturity

$E E' e'$ —217 days maturity

$e E' e'$ —245 days maturity

From these results it may then be concluded that the sex-linked early maturity factor in combination with the autosomal factor either in a homozygous or heterozygous condition produces early maturity individuals. The autosomal factor for early maturity when in a heterozygous condition and in combination with the sex-linked factor for late maturity gives intermediate maturity. Since maturity age cannot accurately be measured in males it cannot be stated what influence the sex-linked early maturity factor would have in a heterozygous condition. It is realized that the backcross data do not provide a crucial test for the existence of sex-linked factors for age at sexual maturity. Since we are limited to the female sex in classifying birds for maturity age no backcross or F_2 generation cross will provide such a test. We must, therefore, be satisfied with evidence from the F_1 generation and the fact that the later generation results do not contradict this interpretation.

TABLE 4

Distribution of maturity age among males resulting from various types of matings between early and late maturity strains.

	NUMBER OF MALES	PERCENTAGE OF EARLY MATURITY	PERCENTAGE OF MEDIUM MATURITY	PERCENTAGE OF LATE MATURITY	RATIO OF EARLY TO LATE	CONSTITUTION OF MALE OFFSPRING
Early ♂ × early ♀	52	19.2	69.2	11.5	1:0.60	EEE'E'
Late ♂ × late ♀	46	8.7	60.9	30.4	1:3.49	eee'e'
Late ♂ × early ♀	96	33.3	46.9	19.8	1:0.59	EeE'e'
Early ♂ × late ♀	41	17.1	73.2	9.8	1:0.57	EeE'e'
Backcross of F_1 ♀ (early ♂ × late ♀) to early ♂	61	13.1	70.5	16.4	1:1.25	EEE'E' EEE'e'
Backcross of F_1 ♀ (late ♂ × early ♀) to early ♂	91	20.9	60.4	18.7	1:0.89	EeE'E' EeE'e'

Maturity in males

In table 4 are recorded the data on inheritance of age at sexual maturity in males. As was earlier stated the estimation of the age at sexual maturity in males is subject to considerable error. The results are given here only because of the value they have in supplementing the data on females and due to already mentioned analytical difficulties arising from being limited to the classification of one sex.

In obtaining these data males reared from the maturity study matings were segregated by hatches when they were approximately six months

old and graded as early, medium, and late. Judgment was based upon plumage and comb development. Although the method of classification is somewhat crude and subject to considerable error, the data supply some evidence as to the mode of inheritance of age at sexual maturity that cannot be obtained in any other manner. In table 4 the data are recorded in percentage of those falling in the three groups, early, medium, and late. The ratio of those of early classification to those in the late group is also given for each type of mating.

The ratio of 1:0.60 obtained in the early by early mating is very similar to those (1:0.59 and 1:0.57) obtained for F_1 generation males from reciprocal crosses. Since the males from these two latter matings were heterozygous for the sex-linked and autosomal factors ($EeE'e'$) it would seem that when the two factors for early maturity are present it makes little difference whether they are in a heterozygous or homozygous condition. Since the standard for classification was somewhat indefinite it may not be well to attempt to compare the first four matings listed in table 4 with the two backcrosses since the former were made in 1931 while the latter two were carried out in 1932. The evidence from the two backcrosses is not entirely consistent with the other data. The male offspring from the two matings differ only in the fact that the first listed ones are homozygous for the sex-linked factor while the latter ones are heterozygous for this factor. The first mating gave a lower percentage of early maturing sons than did the latter. This difference is not to be expected from the genetic constitution of the two groups of males. From these results it can at least be said that we have here confirmatory evidence for the earlier stated conclusions that the sex-linked factor has the same effect in a heterozygous condition that it has in a homozygous state.

DISCUSSION OF RESULTS

The results for the study of maturity in both female and male Rhode Island Reds would tend to substantiate the findings of HAYS that there are both sex-linked and autosomal factors involved in the inheritance of age at sexual maturity. The data also support the view that the sex-linked factor for early maturity is completely dominant and has the same effect regardless of the constitution with respect to the autosomal factor. There is also evidence that the autosomal factor for early maturity is not completely dominant when acting in the presence of the sex-linked late maturity. For simplicity of treatment the results have been analyzed as if there were only one pair of sex-linked and one pair of autosomal factors involved. It is realized that the results do not exclude the possibility of several pairs of factors having an influence upon maturity age but the evidence supports the view that at least some sex-linked and autosomal factors are concerned.

LOCATION OF THE GENE FOR AGE AT SEXUAL
MATURITY ON THE SEX CHROMOSOME*Evidence for sex linkage of maturity differences
in crossbreeding*

The foregoing studies on Single Comb Rhode Island Reds have indicated that differences in age at sexual maturity in this breed were inherited in a sex-linked manner. The Mediterranean breeds are known to mature at an age considerably less than that found in the heavier dual purpose breeds. It is then of interest to determine whether the difference in age at sexual maturity among these breeds is due to sex-linked factors.

WARREN (1930) made reciprocal crosses between Single Comb White Leghorns and Single Comb Rhode Islands Reds. The hybrids were half-sisters to the purebreds with which they were being compared. The 51 White Leghorns had a mean maturity age of 172.4 days and the 49 Rhode Island Reds 247.6 days. The cross of a White Leghorn male by a Rhode Island Red female produced 58 hybrid females maturing at the average of 175.9 ± 2.27 days while the reciprocal of this mating produced 52 daughters with a mean maturity age of 206.4 ± 3.78 days. Since the White Leghorns had a much earlier maturity age than the Rhode Islands Reds the difference between the maturity ages of reciprocal crosses is in the expected direction provided sex-linked factors are responsible for the difference.

A second comparison was made of the maturity age of hybrids from reciprocal matings of White Leghorns and Rhode Island Reds. In this case the Rhode Island Red stock came from the late maturity strain which had a mean age at maturity of 249 days for that year (1930). The particular females used in the mating with the White Leghorn males had a mean age at maturity of 269 days. The White Leghorn stock used had a mean maturity age of 175 days for that year. The 77 female offspring from the cross of the White Leghorn male by Rhode Island Red females matured at the mean age of 197.7 ± 2.94 days while the 171 daughters of the reciprocal cross, Rhode Island Red male by White Leghorn females, had a mean maturity age of 209.6 ± 2.18 days. Here again the earlier maturing hybrids came from the cross in which the father was from the early maturity stock. The actual difference in maturity age was 11.9 days which was 3.26 times the error of the difference. In both the crosses cited here it is seen that statistically significant differences were found between the reciprocal crosses and in each case the difference was in the expected direction provided sex-linked factors were involved. It would, therefore, appear that differences in age at sexual maturity between the Mediterranean and the larger breeds are at least partially due to sex-linked factors.

Order of genes

Some attention has already been given to the mapping of the sex-linked factors in the fowl (SEREBROVSKY 1927, WARREN 1928, HERTWIG and RITTERSHAUS 1929, and MACARTHUR 1933). The best known sex-linked factors are gold-silver, rate of feathering, barring, and shank color. Workers are in fair agreement by finding 10 to 19 percent of crossing over between the genes for gold-silver color and rate of feathering. The genes for barring and shank color have shown themselves to be only loosely linked with either gold-silver or rate of feathering (crossing over percentage 40-48). No very satisfactory data have been published on the linkage relations of shank color and barring but their reaction with other factors in the sex chromosome would indicate that they are closely associated.

The writer (1928) tentatively proposed the order of the genes on the sex chromosome to be barring-silver-rate of feathering. Since both silver and rate of feathering show very loose linkage with barring it remained a question as to the exact order of these two factors. In the 1928 publication the crossover value for the rate of feathering and barring was 48 and for silver and barring 46. In table 5 are presented two additional years' results on the linkage relations of this pair of sex-linked factors. Some inconsistencies are seen in the two members of the non-crossover and crossover classes but these can probably be attributed to the difficulty of recognition of the phenotypes when both barring and gold are involved. Gold is not well expressed in a barred bird carrying extended black and the barring factor has little opportunity of expression when the black is restricted to the degree that it is in the Columbian pattern. In the latter group evidence of the existence of the barring factor was usually found in the feathers of the hackle but in some cases the black area was so restricted that decision was difficult. This fact is probably responsible for the shortage of birds in the barred class. In the barred birds carrying extended black (Barred Plymouth Rock pattern) the presence of gold was not easily recognized. If such individuals showed any buff or red in the neck or breast region they were classified as gold, but in the absence of these colors, were called silver. The difficulty of recognizing gold and silver when in combination with barring and other pattern factors resulted in some variations in the ratio of these colors in the different classes. The two years' results given in table 5 and the writer's previously published results are all in agreement in indicating a higher percentage of crossing over between barring and rate of feathering than between barring and gold-silver. These results would tend to confirm the view that although both rate of feathering and gold-silver are widely separated from barring, the latter (gold-silver) is located between rate of feathering and barring.

TABLE 5

Linkage relations of the sex-linked factors barring with gold-silver and with rate of feathering.

FACTORS	NON-CROSSOVERS			CROSSOVERS			GRAND TOTAL	PERCENTAGE CROSSING OVER
	NON-BARRED GOLD	BARRED SILVER	TOTAL	NON-BARRED SILVER	BARRED GOLD	TOTAL		
BARRING AND GOLD-SILVER								
Brown Leghorn by Barred Rock	51	81	132	46	60	106	238	44.5
White Leghorn by Rhode Island Red	203	59	262	69	123	192	454	42.3
BARRING AND RATE OF FEATHERING	NON-BARRED EARLY	BARRED LATE	TOTAL	NON-BARRED LATE	BARRED EARLY	TOTAL	GRAND TOTAL	PERCENTAGE CROSSING OVER
	NON-BARRED LATE	BARRED EARLY	TOTAL	NON-BARRED EARLY	BARRED LATE	TOTAL		
Brown Leghorn by Barred Rock	51	76	127	52	65	117	244	48.0
White Leghorn by Rhode Island Red	59	36	95	50	39	89	184	48.4

Additional evidence was obtained from the 1931 data which involved rate of feathering, silver and barring in the same cross. They were as follows: 25 early feathering, silver, barred; 48 late feathering, gold, non-barred; 9 early feathering, gold, barred; 3 late feathering, silver, barred; 24 early feathering, silver, non-barred; 24 early feathering, gold, non-barred; 3 late feathering, silver, non-barred; and 33 late feathering, gold, barred. If we assume that silver is located between rate of feathering and barring, then breaks between barring and rate of feathering should frequently affect the relationships of silver and rate of feathering. It would also be true that breaks between silver and rate of feathering would not affect the relationships between silver and barring except in cases of double crossing over where breaks occurred simultaneously between silver and barring and silver and rate of feathering. In 84 breaks between barring and rate of feathering, 27 were accompanied by changes in the relationships of silver and rate of feathering. There were only 12 out of 69 cases where the relationships of silver and rate of feathering were affected by breaks between barring and silver. Likewise, if silver occupied the end position having rate of feathering between it and barring, breaks between silver and rate of feathering should not affect the relationships of rate of feathering and barring. A summarization of the breaks between silver and rate of feathering revealed the fact that in a total of 39 such breaks, 27 also changed the relative positions of rate of feathering and barring. The only way to account for these 27 cases is by double crossing over and it seems improbable

that the percentage would be so high. Therefore, all the results seem to indicate strongly that silver is located between barring and rate of feathering. It should be noted that this arrangement of the genes agrees with the findings of SEREBROVSKY and WASSINA (1927) but not with the conclusions of MACARTHUR (1933).

Position of sex-linked gene for maturity

Having accumulated evidence that the differences in age at sexual maturity both in Rhode Island Reds and between Rhode Island Reds and White Leghorns are inherited in a sex-linked manner, it is of interest to determine the linkage relations between the responsible gene or genes and the other known genes in the sex chromosome. The method followed is similar to that made use of by PUNNETT (1930) except that consideration is here given to age at first egg instead of annual record. The sex-linked factor for age at sexual maturity should show linkage with the factor with which it is most closely associated on the sex chromosome. If early maturity and silver plumage were characteristic of a breed, and the two factors were closely associated on the sex chromosome, then there should be a tendency for these two characters to be associated in segregation. In other words, if the early maturity silver stock was mated to a late maturity gold stock, the silver segregates should be of earlier maturity than the golds. If the golds and silvers should have similar maturity then it could be assumed that the genes for gold-silver and for maturity age were not closely associated on the sex chromosome. Then if several known sex-linked genes were involved at the same time the age at maturity of the segregates should indicate the approximate location of the gene for age at sexual maturity.

The Single Comb Brown Leghorn and the Barred Plymouth Rock breeds were chosen for the first test for the linkage relations of the factor for age at sexual maturity. These two breeds were crossed because they produced a hybrid male heterozygous for three sex-linked factors, barred, rate of feathering, and gold-silver. The Brown Leghorn carried the recessive gold, recessive early feathering, and recessive non-barring. The Barred Plymouth Rock carried the three dominant alleles. It was also known that there was the characteristic difference in age at sexual maturity between these two representatives of the Mediterranean and American classes, the Brown Leghorn having the earlier maturity. It would have been desirable to have known the average difference in age at sexual maturity between these two breeds but since the available stock of each was limited this was not feasible.

A Brown Leghorn male was mated to three Barred Plymouth Rock females for the production of males heterozygous for the four sex-linked

factors, rate of feathering, barring, gold-silver, and age at sexual maturity. Since age at sexual maturity could be measured only in the females and since the female bird has only one sex chromosome, it made little difference what type of females were mated to the multiple heterozygotes. The female offspring having only one sex chromosome and receiving it from their sire only, must show the sex-linked constitution of the gamete which they received from him.

Six Brown Leghorn-Barred Plymouth Rock hybrid males were mated to a group of 70 Rhode Island Red females. Females of the Rhode Island Red breed were chosen primarily because of their availability. The heterozygous males received early feathering, gold, non-barred, and early maturity in one gamete and late feathering, silver, barred, and late maturity in the other. Therefore, the allelic pair of characters which shows the greatest difference in maturity between its segregating members should indicate the approximate location of the gene for age at sexual maturity.

In table 6 are presented comparisons of the mean age at sexual maturity for the members of the three sex-linked pairs of factors in the Brown Leghorn-Barred Rock cross. There was very little difference in maturity age for the early and late feathering females, the 146 late feathering pullets having a mean age of 194.4 ± 2.17 days and for the 149 early feathering ones, 193.8 ± 1.92 days. When classified for gold and silver plumage the difference was also of no significance, the silver ones maturing in 192.4 ± 2.14 days and the golds in 195.5 ± 1.95 days. The only case where the difference is at all great is for the barred and non-barred groups. The barred birds had a maturity age of 200.7 ± 2.31 days while the non-barred ones had a mean maturity age of 189.5 ± 1.80 days. The difference of 11.2 days in age at sexual maturity of the barred and non-barred groups approaches statistical significance and emphasized the desirability of a repetition of the experiment.

In order to make the results more critical the experiment was repeated with breeds of fowl concerning which there was a more dependable knowledge of the age at sexual maturity. The two breeds used were the Single Comb White Leghorn and Single Comb Rhode Island Red. White Leghorn males from a strain with a mean maturity age of 175 days were mated to Rhode Island Red females which came from a strain that had been selected for a period of years for late maturity (mean maturity age 249 days). The F_1 generation results from reciprocal matings between these two strains have been given under the discussion on evidence for sex linkage of maturity age in crossbreeding. The data indicated that sex-linked factors were involved. The chief reason that this mating was not originally used was the fact that the presence of dominant white in the White Leghorn interfered with the expression of barring. This fact neces-

sitated the discarding of one-half of the offspring in the backcross generation.

The F_1 generation males from the above mentioned cross were heterozygous for barring, silver, rate of feathering, maturity age, and dominant white. The combination of barring, silver, early feathering, early maturity was brought in from the White Leghorn and non-barring, gold, late feathering, and late maturity from the Rhode Island Red breed. The White Leghorn carries barring and silver in a hypostatic condition. Previous observations had demonstrated that the two strains used were homozygous for the two types of feathering.

The F_1 males were mated to Rhode Island Red females and the female offspring were classified for barring, gold-silver, rate of feathering, and age at sexual maturity. As has been mentioned already it was necessary to discard all white chicks produced in this mating because of the dominant white masking the expression of barring and gold-silver. Since dominant white was known to be an autosomal factor, the elimination of the white chicks should in no way effect the segregation of the sex-linked factors. Since half of the chicks were discarded because of white and since only females could be classified for age at sexual maturity, only one-fourth of the chicks hatched were of any value. Allowing for the mortality normally occurring before maturity, it was necessary to hatch about 900 chicks to obtain a test for maturity on 186 females.

TABLE 6

Comparison of mean age at sexual maturity of allelic pairs in the case of three sex-linked factors. Differences in maturity age should indicate the approximate location of the maturity gene on the sex chromosome.

ALLELES	BROWN LEGHORN—BARRED ROCK CROSS		WHITE LEGHORN—RHODE ISLAND RED CROSS	
	NUMBER OF FEMALES	MEAN AGE AT SEXUAL MATURITY	NUMBER OF FEMALES	MEAN AGE AT SEXUAL MATURITY
Late feathered	146	194.4 $\pm$ 2.17	89	215.5 $\pm$ 2.71
Early feathered	149	193.8 $\pm$ 1.92	82	198.8 $\pm$ 2.68
Silver	147	192.4 $\pm$ 2.14	62	189.4 $\pm$ 2.70
Gold	148	195.5 $\pm$ 1.95	120	217.5 $\pm$ 2.25
Barred	121	200.7 $\pm$ 2.31	75	208.3 $\pm$ 3.10
Non-barred	174	189.5 $\pm$ 1.80	109	207.3 $\pm$ 2.28

The data in table 6 under the heading White Leghorn-Rhode Island Red cross are from this experiment. In the pair of factors responsible for the barred or non-barred condition there was no significant difference in age at sexual maturity of the two groups of segregates. The 75 barred females had a mean maturity age of 208.3 days and the non-barred ones,

207.3 days. The original combination was early maturity with barring and late maturity with non-barring. The very slight difference is in the direction opposite to be expected and, therefore, would indicate that the factors for barring and for age at sexual maturity were not closely associated on the sex chromosome. Since the barring factor is widely separated from the factors for rate of feathering and for gold-silver and not closely associated with the factor for age at sexual maturity, it might be expected that there would be linkage between the latter factor and those for rate of feathering and gold-silver. Since the parental combinations were early maturity and silver, and early maturity and early feathering, it would be expected that the alleles of silver and early feathering would show later maturity. The results given in table 6 show the silver females to have matured 28 days earlier than did the gold ones and the early feathering females to have started to lay 17 days earlier than did the late feathering ones. The difference in the first case is 8 times its probable error and in the second 4.4 times its probable error. These differences are statistically significant and would, therefore, indicate that the factor for age at sexual maturity is located on the sex chromosome somewhere between those for rate of feathering and barring. The results here are more significant when we consider the fact that linkages of the maturity factor with three known points on the sex chromosome are in agreement. The difference in results between this experiment and the one using the cross of the Brown Leghorn and Barred Rock is difficult to account for unless the former cross did not involve sex-linked factors influencing maturity. In the former case differences in age at sexual maturity were assumed and not actually proved.

The findings here are also in agreement with the results in table 5 in showing closer linkage with silver-gold than with rate of feathering. The earlier work had indicated that these three genes were located on the sex chromosome in the following order: rate of feathering, gold-silver, and barring. If the proposed order of genes is correct and the factor for age at sexual maturity is located between barring and gold-silver, it would necessarily show closer linkage with gold-silver than with rate of feathering. If the factor for age at sexual maturity were located between gold-silver and rate of feathering, it might also show closer linkage with the former than the latter but since these two factors are themselves fairly closely associated (about 15 units apart), the maturity factor would necessarily need to be very closely linked with each. This does not appear to be true and the order of the genes on the sex chromosome is probably rate of feathering, gold-silver, age at sexual maturity, and barring.

In order to determine approximately the position of the gene for age at sexual maturity on the sex chromosome, calculations were made of the percentage of crossing over between this factor and the other sex-linked fac-

tors. It is realized that in working with a physiological factor of this type considerable error will enter into the calculation. Many environmental factors are known to interfere with the expression of the factor for age at sexual maturity. An additional problem is to establish a dependable dividing line between the late and early maturity groups. For purposes of making the calculation, the dividing line was arbitrarily placed at 205 days, thus segregating the birds into two approximately equal groups. Theoretically, the segregation of the sex-linked factors for maturity should produce equal numbers of early and later maturing individuals. Then in determining the linkage relations for the gene for age at sexual maturity and the gene for gold-silver the non-crossover classes would be silver-early maturity and gold-late maturity and the crossover classes would be silver-late maturity and gold-early maturity. In the case of the relations of age at sexual maturity and rate of feathering the classes are obtained by substituting early feathering for silver and late feathering for gold.

The data for the linkage relations calculated in the manner described are given in table 7. In the measurement of linkage between age at sexual maturity and gold-silver it was found that the percentage of crossing over

TABLE 7

Percentage crossing over between the gene for age at sexual maturity and the sex-linked genes, gold-silver, rate of feathering, and barring.

FACTORS	NON-CROSSEOVERS			CROSSEOVERS			GRAND TOTAL	PERCENTAGE CROSSING OVER
	SILVER EARLY	GOLD LATE	TOTAL	SILVER LATE	GOLD EARLY	TOTAL		
Age at sexual maturity and gold-silver	65	44	109	38	22	60	169	35.5
	EARLY-FEATH. EARLY	LATE-FEATH. LATE	TOTAL	EARLY-FEATH. LATE	LATE-FEATH. EARLY	TOTAL	GRAND TOTAL	PERCENTAGE CROSSING OVER
Age at sexual maturity and rate of feathering	50	50	100	32	37	69	169	40.8
	BARRED EARLY	NON-BARRED LATE	TOTAL	BARRED LATE	NON-BARRED EARLY	TOTAL	GRAND TOTAL	PERCENTAGE CROSSING OVER
Age at sexual maturity and barring	34	46	80	36	53	89	169	52.7

was 35.5. For rate of feathering and age at sexual maturity the percentage of crossing over was found to be 40.8. The barring factor and age at sexual maturity were found to give 52.7 crossing over, indicating no linkage. No attempt was made to treat statistically the foregoing data since errors of classification were probably much greater than those of random sampling. Although these calculations are considered of little value in determining

the exact location of the gene for age at sexual maturity they do supply some information as to the approximate position of this factor.

These results would support the view that the order of the genes in the sex chromosomes is as follows: rate of feathering, gold-silver, age at sexual maturity, and barring. The available evidence indicates that the gene for age at sexual maturity is located somewhat nearer gold-silver than barring.

SUMMARY

1. By selection it was possible to establish from a flock of fowls, two strains of Single Comb Rhode Island Reds differing considerably (about 40 days) in the mean age at sexual maturity.

2. A genetic analysis indicated that both sex-linked and autosomal factors determined age at sexual maturity in this breed of fowl.

3. Evidence was obtained to show that the difference in age at sexual maturity between the White Leghorn and Rhode Island Red breeds was at least partially due to sex-linked factors.

4. Linkage results from the White Leghorn-Rhode Island Red cross indicate that the sex-linked factor for age at sexual maturity is located between the factor for barring and that for gold-silver.

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