

THE GENETICS OF FRIZZLING IN FOWLS.

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by

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## INTRODUCTION.

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Frizzled fowls are neither very common nor of any special economic importance, but as one of the most unique of the many variations of the species they are prized by fanciers in various parts of the world. The variety assumed a special interest for the geneticist when it was discovered that in the experience of some breeders frizzled fowls did not breed true but always "threw" a certain proportion of normally-feathered birds. Arising from this observation, the theory has become current that the homozygous frizzle is non-viable and presumably is killed off at an early stage of embryonic development. The present investigation was undertaken to substantiate or disprove this theory and to determine the mode of inheritance of the character.

## HISTORICAL

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Frizzled fowls were described as early as 1645 by ALDROVANDUS, to whom a specimen had been sent from Parma. WILLOUGHBY (1676) refers to its occurrence in England and states/

states that it was at that time referred to as the "Frisland" breed. Temminck states that the "coq à plumes frisées" is kept under domestication in Java, Sumatra, Southern Asia and the Philippines, the prevailing colour being white.

TEGETMEIER (1867) quotes a correspondent in Ceylon as writing that the frizzled fowl is called by the Cingalese, Caprikukullo. It is found here but rarely, and the natives say they came from Batavia. This agrees with Temminck. DARWIN (1868) states that it is not uncommon in India.

Data regarding the viability of frizzled fowls are controversial. TEGETMEIER (loc. cit.) quotes a breeder's statement that they are susceptible to injuries from exposure and "are of a somewhat sickly appearance". On the other hand, WRIGHT (1917) says "contrary to the common belief every breeder who has had real experience with the Frizzled Fowl that we have yet met with, pronounces it distinctly hardy".

#### DESCRIPTION

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The peculiarity of the frizzled fowl is that the contour feathers, instead of conforming to the shape of the body have the shaft recurved so that the exterior of the feather becomes concave. The resulting general appearance is/  
is/

is as if these feathers had been rubbed the wrong way (Fig. 1). In good specimens a ruff is formed in the neck region. The shafts of the rectrices and remiges are recurved only in the more slender region at the tip of the feather, but usually in certain regions of these feathers small groups of the barbs are twisted so that they appear to curl around the shaft, instead of forming the normal web. These barbs are usually eventually worn off so that only the shafts of these feathers remain. As a result the birds are unable to fly and must be given low roosts. It is doubtful if the mutation could persist long in a state of nature.

The character is distinctly variable in its expression. In some cases the contour feathers have not only the shaft recurved but also the barbs so curled that not a feather on the body has the normal flat web. The degree of curvature of the shaft varies so that in some birds, the feathers are only slightly curved and still point in a posterior direction, while in others the feathers are curled right around so that the tips point toward the head.

TEGETMEIER found in examining Darwin's collection of domesticated birds from all over the world that some specimens exhibited frizzling in all parts of the body while in others the character was confined to the neck region.

Frizzled fowls are found in various colours - black, white, red, buff and blue - the main preference of the/

the breeder being for birds all one colour and for uniformity in show pairs, trios or pens. Those used in the present experiment were all red (i.e., of the Rhode Island Red) excepting four bantams of which the cock was blue and the hens blue, black and buff.

DARWIN (loc.cit.) states that the periosteum of the bones of the frizzled fowls of India is black but this character has not been observed in any of the writer's material and there is no reason to suppose that it is necessarily linked with frizzling.

The only published data concerning the mode of inheritance of frizzling known to the writer is that of DAVENPORT (1906) who raised only one generation from a cross of Frizzled x Silky fowls but found that the former character was dominant and that his four female Frizzles were all heterozygous.

The writer was unable to detect any satisfactory indication of this character in the newly-hatched chick, but frizzled birds could usually be positively identified as soon as the wing feathers had grown to half an inch or even less. In one case, the classification of 47 chicks at 6 days of age, agreed exactly with a second classification at 14 days. Another lot of 66 chicks sorted out at 9 days was found when re-checked at two weeks to have been accurately classified on the first occasion. In some cases of extremely slow feathering individuals, the type of plumage/

plumage could not be accurately decided till the chicks were over two weeks of age. In the data given below only those chicks are included in which determination of the frizzled or normal type of feathering was definite and accurate. With the exception of those dying early, all descriptions were re-checked at two weeks or over.

#### MATERIAL

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In the present study 29 birds were used of which 7 were normal and 22 frizzled. All of the latter came directly or indirectly from the flock of Major G. S. Williams, Tredrea, Perranwell, Cornwall, some having been purchased in 1927, a few raised here from this stock and ten loaned for the breeding season of 1928. The writer wishes to express his sincere thanks to Major Williams who not only supplied valuable data concerning his own flock but also, at his own suggestion, very generously loaned six large frizzled hens and four bantams for the entire breeding season. Thanks to this assistance the writer was enabled to obtain much more extensive data than would otherwise have been possible.

#### METHODS/

## METHODS

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In order to get sufficient numbers of chicks, matings were begun early in February 1928 and chicks were hatched right up to July 26th of the same year.

Matings were made as follows:

### A. Frizzle x Normal.

1. Eight frizzled females were mated with ♂ 283, a normally feathered cockerel extracted in 1927 from a frizzle x frizzle mating. One of these eight, ♀ 129, and another hen had also been mated earlier in the season to a Brown Leghorn male, No. 142.
2. Two normal females, Nos. 272 and 300, hatched in 1927 from a frizzle x frizzle mating were bred in separate pens to the frizzled cockerels. P 101 and M 102 respectively. Both of these males had been hatched in 1927 from a frizzle x frizzle mating.
3. Three normal Bantam females were mated to a blue frizzled Bantam cock B 18 (Fig. 3).

### B. Frizzle x Frizzle.

1. Sixteen frizzled females were mated with the two frizzled cockerels P 101 and M 102, each male having eight females in a separate pen. These matings were made concurrently and in the same pens as those referred to in A 2. Ten of these females had/

had been previously used in the series A 1 but care was taken to ensure that the influence of the normal male was lost before their eggs were included in the B 1 series.

2. Three frizzled bantam females were mated to the blue Frizzled Bantam cock, B 18, these birds being in the same pen as the normal bantams mentioned above, A 3.

C. Normal x Normal matings.

1. The two normally feathered females 272 and 300, were mated concurrently with the A 1 series, to the normal male 283. All three of these birds had been extracted from matings of frizzle by frizzle.

INCUBATION.

All eggs were incubated as nearly as possible in the same manner. The majority were started in a gas-heated Phipps incubator and moved to Hearson electric incubators a week before hatching. All hens were trap-nested, all eggs were marked and the chicks were hatched in pedigree bags so that the ancestry of each chick was definitely known. The eggs were candled at least twice during each hatch. Every dead embryo was examined and the/

the estimated period of its death recorded as well as any evidence of abnormality. Since there was a possibility that a lethal factor if present might be operative at an early stage of development of the zygote, all eggs which appeared infertile on candling, were broken for more accurate observation and where necessary an examination was made under the dissecting microscope.

### RESULTS.

In view of the fact that the investigation as thus far conducted does not yet definitely answer the question of whether or not the homozygous frizzled condition is lethal, the incubation records and progeny ratios for each hen and each mating are presented in detail. (Tables 1, 2 & 3).

TABLES/



TABLE 2.

MATINGS OF EXTRACTED NORMALS.

Series	♂	♀	Eggs Set	Infer- :tile	1-2	3-4	5-6	7-8	9-10	11-12	13-14	15-16	17-18	19-21	No. (fert. eggs)	%	Class- :filed	Frlz- :zle	Mor- :mal
C 1	283	272	19								1			5	13	68.4	13	0	13
	"	300	19											2	17	89.5	14	0	14
	Totals		38											7	30		27	0	27



INTERPRETATION OF RESULTS.

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On reference to Table 1, it is seen that in Series A every frizzled bird tested, whether male or female, produced both frizzled and normal offspring when mated to normal fowls. (No. 301 had only one chick in this series but was subsequently shown in Series B to be heterozygous). The numbers of the two classes were 64 Frizzles to 76 Normals. The deviation from the expected ratio (Table 4) is only 1.47 times the probable error of the ratio and may therefore be considered as occasioned only by the fluctuations of sampling. (The probable error has been determined by the formula  $P.E. = + .6745 \sqrt{pq \times n}$  where  $n$  is the number of individuals in the population, and  $p$  and  $q$  the proportions expected in each class expressed as a fraction of the whole).

The results in Series A may therefore be construed as indicating (1) that frizzling is a dominant character dependent for its expression upon a single factor and (2) that the 9 females and 3 males tested were all heterozygous with respect to that character. Since the crossing of frizzled males with normal females gave results similar to those in the reciprocal cross, it is evident that no sex-linked factors are involved.

TABLE 4./

TABLE 4.

RATIOS OBTAINED IN SERIES A, B, AND C.

Series		Frizzle	Normal	Deviation	Probable Error	Dev. P.E.
A	Observed	64	76	6		1.47
"	Expected	70	70		$\pm 4.08$	
B	Observed	230	96	14.5		2.75
"	Expected	244.5	81.5		$\pm 5.27$	
C	Observed	0	27			
"	Expected	0	27			

In the Series B matings (Table 3) normally feathered chicks were obtained from every one of the 19 hens, 10 of which had not previously been tested. Since all of these Frizzles, male and female, had thus been shown to be heterozygous, the expectation in the progeny, considering the findings in Series A, was a simple 3:1 ratio. While the deviation of 14.5 from expectation (Table 4) seems fairly large for a population of 326 individuals it is only 2.75 times the probable error of the ratio and is therefore still within the limits of fluctuations due to sampling.

In conformation with the results of these two series of matings, it was found in Series C that breeding together/

together normally feathered birds extracted from Frizzle x Frizzle matings produced only normals (Table 3) and that these were therefore true simple recessives.

The results of all three series can be accommodated by the assumption that frizzling is a monofactorial dominant character and that if any lethal factors are operative their influence is not manifest by the time the chicks are old enough to be classified.

At the same time it should be pointed out that in both Series A and B there is a deficiency of frizzled chicks. In fact, the ratio in the latter series fits a 2:1 expectation equally as well as one of 3:1. On the basis of 2:1 the calculated ratio is 217.2 frizzles:108.6 normals and the deviation of 12.8 is only 2.1 times the probable error of the ratio -  $\pm 6.08$ .

However, in Series B the deficiency of frizzles results from an extreme deviation from a 3:1 ratio in the progeny of certain females mated to males P 101 and B 18. The progenies from the other male, M 102 make up an almost perfect 3:1 ratio, there being 105 frizzles to 39 normals where the expectation is 108:36. Of the females mated to ♂ P 101, Nos. 341, 342 and 344 gave deviations from the expected 3:1 ratio so marked that they might be considered as indicating a differential production of gametes by this male were it not for the facts that from the same male ♀ 345 produced an excess of frizzled chicks, ♀ 346 an exact/

exact 3:1 ratio, and moreover that in series A 2 (Table 1) it had been shown that ♂ P101 was producing the two classes of gametes in equal proportions.

It may, therefore, be considered that the deficiency of frizzled chicks in Series B arose from fluctuations of sampling.

It is more difficult to explain why every one of the 22 frizzled birds tested in this investigation and four frizzled females mated by DAVENPORT (1906) should have been heterozygous. Since one would expect that in an unselected population of this size considerably more than one-third would be homozygous, there is every reason to suggest that a lethal factor is involved.

Such a lethal might be either gametic or zygotic. The possibility of a gametic lethal is eliminated by the results in Series A of this experiment which showed that 10 females and 3 males tested were producing viable gametes carrying the factor for frizzling in the same proportion as gametes carrying the recessive factor.

If a zygotic lethal were involved it might be operative either during embryonic development or any time after hatching. Consideration of the time of effect of lethals in other animals and the few known in fowls leads one to suspect such a factor to be effective during the period of incubation. The percentage mortality for each 4-day period of all eggs set in Series A and B is shown in Table 5.

TABLE/

TABLE 5.

COMPARISON OF EMBRYONIC MORTALITY IN SERIES A and B.

Series	Mating	Fertile Eggs	% mortality in 4-day periods					Total	% Hatch.
			1-4	5-8	9-12	13-16	17-21		
A	Frizzle x Normal.	210	6.19	2.38	1.43	1.90	12.38	24.28	75.71
B	Frizzle x Frizzle	482	4.98	2.90	2.07	2.28	12.86	25.10	74.89

If homozygosity for frizzling were lethal to the embryo a peak of mortality would be expected at some stage of incubation for the eggs set from matings of frizzle x frizzle. In addition one would expect approximately 25% greater mortality in this series than in eggs from matings of frizzle x normal. Neither of these conditions was present. The death rate in one series ran closely parallel to that of the other. The peak of the mortality during the last five days is quite usual and represents to a considerable extent fully formed chicks dying from being in positions which make it impossible or extremely difficult for them to hatch.

Embryos representing various types of teratological monsters, chondrodystrophic chicks, and abnormalities in position were found but in no greater numbers in one series than the other, and in no greater proportion altogether than were observed by HUTT and GREENWOOD (1928) in a survey of a large number of unhatched eggs from different sources.

In view of these findings it seems reasonable to conclude that homozygosity for frizzling is not lethal to the embryo. There remains the possibility that the homozygous individual may be killed off at some time after hatching. Unfortunately, in the present investigation brooding conditions were unsatisfactory and mortality was usually high among chicks of both series in this investigation/

investigation so that no statement is possible regarding differential mortality or a possible peak of mortality in Series B chicks after hatching.

Another alternative, which seems the more probable key to the problem of the lost homozygous Frizzle is that this individual may be of an undesirable type and therefore discarded by the fancier prior to the breeding season. In the population raised in the present investigation distinct types of frizzling are distinguishable. One of these gives the bird a quite woolly appearance while the other presents the more desirable show type in which the feathers stand out stiffly from the body.

Major Williams writes that it is his policy "to discard in all cases the narrow feathered and unevenly feathered birds". He states further that with his Bantams, mating of plain cock with frizzled hens, or the reciprocal cross, produce frizzled birds having the quality of feather and curl desired, whereas mating frizzled birds together produces "perhaps 80% frizzles, and of these about three-quarters are narrow feathered and useless".

These facts fit perfectly with the hypothesis that the homozygous frizzled fowl is viable but is discarded by the breeder, with the exception that one would expect about one-third rather than three-quarters of the last/

last class mentioned to be undesirable. The difference probably indicates a selection for the desired type so rigid that it effects the heterozygous as well as the homozygous individuals.

In consideration of all these facts it is reasonable to assume that the reason no homozygous frizzled fowls were present among the 22 tested in this investigation is that these birds constituted not a random sample but a population of individuals selected according to a breeder's standard. Presumably this standard barred out the homozygous individual. If this view be correct the case of the Frizzle is parallel to that of the Blue Andalusian in which the individual preferred is heterozygous and cannot breed true. In the latter case the phenotypes segregate distinctly in a 1:2:1 ratio; with the frizzled birds the incompleteness of dominance results in a gradation of phenotypes from that of the desired heterozygote to the undesirable homozygote.

In order to submit to a critical test the hypothesis presented above it is planned to test out in 1929 a truly representative sample of the population raised in the present investigation in which the undesirable as well as the preferred type will be included.

SUMMARY./

SUMMARY.

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1. An investigation has been conducted to determine the mode of inheritance of frizzling in the domestic fowl and to substantiate or disprove the current theory that homozygosity for frizzling is lethal.
2. One hundred and forty chicks classified from matings of frizzled x normal fowls exhibited a 1:1 ratio of each class.
3. Three hundred and twenty-six chicks classified from matings of Frizzle x Frizzle approach a 3:1 ratio of Frizzle to normal closely enough to be within the limits of fluctuations due to sampling.
4. Offspring of normally feathered fowls extracted from matings of Frizzle x normal were all normal.
5. Incubation records of 758 eggs fail to show any evidence of a zygotic lethal factor believed to be associated with frizzling being operative during embryonic development.
6. The possibility of a gametic lethal is discussed and discarded.
7. It is concluded that frizzling is a monofactorial dominant character.
8. The hypothesis is suggested that the reputed inability of frizzled fowls to breed true arises from a preference by the fancier for the phenotype exhibited by/

by the heterozygous individual resulting in the exclusion from the breeding pen of all the homozygous individuals.

9. A critical experiment to prove or disprove this hypothesis is under way.

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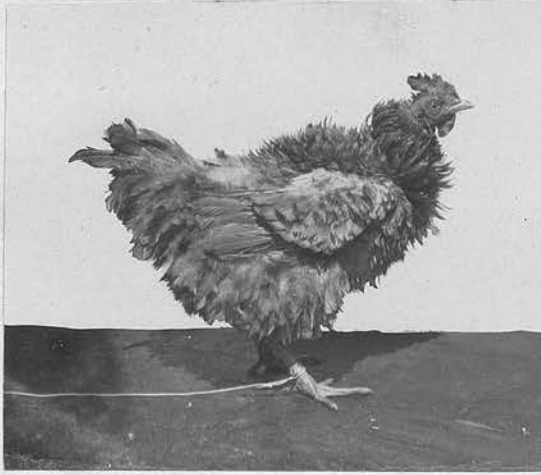
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## CAPTIONS FOR ILLUSTRATIONS.

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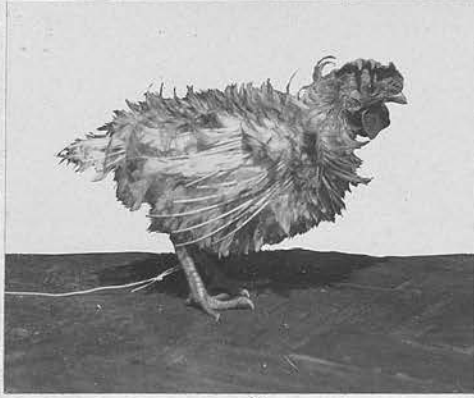
1. A typical frizzled female in good plumage.
2. A frizzled female at the end of the breeding season. The bare back and broken tail feathers are found in nearly all females at this time.
3. The blue frizzled Bantam cock used in Series C matings. The naked wing quills seen here and in Fig. 2 are characteristic of most individuals at all seasons.
4. A frizzled chick at one week of age. The wing feathers of the chick plumage curl away from the back instead of conforming to the shape of the body.



1



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3



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On the Relation of Fertility in Fowls to the Amount  
of Testicular Material and Density of Sperm  
Suspension.

By F. B. Hutt, B.S.A.



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#### INTRODUCTION.

STUDIES on fertility in the domestic fowl so far reported have been concerned more with the influence of the female than with that of the male. At the same time it is common knowledge among poultry breeders that there is often a great difference in the fertility of different cocks of the same breed under the same environmental conditions. Such differences are conceivably dependent upon three different factors, these being (1) the frequency of copulations, (2) the number of spermatozoa ejaculated, and (3) the physiological efficiency of the spermatozoa. The present study was undertaken to determine the influence on fertility of the second of these factors.

In attacking this problem it was assumed that the number of spermatozoa elaborated by different males would be dependent to some extent

upon the amount of testicular tissue present. Presumably the same assumption is the basis of the very general objection on the part of breeders of horses, cattle, and other domestic animals to males with only one testis or having one or both testes undersized. While there is considerable variation in the size of testes among normal cocks of the same breed, such differences could be accurately determined only by post-mortem weighing. If normal birds were used, a great number would be necessary to ensure that among them appreciable differences in the amount of testicular material present would be found upon examination at the conclusion of the experiment. Accordingly it was considered that the most suitable material would be cocks which had been castrated in various degrees.

MATERIAL.

The males used in the experiment were eleven Single Comb Brown Leghorns hatched in the spring of 1927. The degree of castration in each is shown in Table I.

TABLE I.—MALES TESTED.

Bird Number.	Age at Operation. Days.	Tissue removed.
142	7	Left testis.
152	7	" "
149	7	" "
157	7	Almost all of left testis.
176	14	Right testis. Removed tissue implanted subcutaneously.
5	7	Left testis and about half of right testis.
21	38	Left testis and about half of right testis.
172	12	Left testis and anterior and posterior ends of right testis. Removed tissue implanted subcutaneously under right wing.
382	control	None.
384	control	"
387	control	"

In cases where part of a testis was removed care was taken at the time of operation not to destroy the epididymal connection. These operations were performed by Dr A. W. Greenwood, and to him are due the writer's sincere thanks for the excellent material provided.

The development and growth of these birds were unexceptional, and at six months of age they were indistinguishable in any way from normal males.

The females to which these cocks were mated were mostly Brown Leghorns, but included also some Rhode Island Reds and cross-bred birds, which

were distributed as equally as possible among the different males. The majority were pullets.

#### METHODS.

(a) *Stud Matings*.—The accommodation available did not permit giving each male a separate pen with a flock of females, and therefore the “stud-mating” system was used for the earlier part of the experiment. The cocks were housed in two separate pens, and kept confined till late in the afternoon. Fifty-eight females were used in the first part of the experiment, these being housed in three separate pens, with continual access to an outdoor yard. All were leg-banded and trap-nested. To each male were assigned the leg-band numbers of from five to seven females, and whenever a hen was taken from a trap-nest she was put in a small breeding-coop along with the male indicated by her number. After a period of fifteen or twenty minutes both hen and cock were returned to their original pens.

In assigning the females to the different males care was taken to see that females of different ages and breeds were distributed as evenly as possible among the ten males used for stud mating. The system is open to the criticism that the hen laying six times a week is mated more often than one laying only twice, but since the hens were assigned regardless of their laying ability, the probability of any one male getting a high proportion of good or poor layers among his five to seven hens was not great. The actual records indicated that the chance distribution of frequent layers and poor layers was fair enough to permit comparable tests of the fertility of the various cocks.

All birds were given a grain ration of wheat, oats, and maize; mash of Sussex ground oats, wheat middlings, bran, maize meal, soy-bean meal, and fish-meal, with cabbage occasionally, and oyster-shell *ad libitum*. Milk was supplied irregularly. Cod-liver oil was fed at a level of two per cent. in the mash.

Before records were begun, the females were isolated from males for a period of two weeks, and those laying were not used till their eggs were found, after incubation, to be infertile. Matings were begun on 1st January 1928, and continued till 8th March. Matings with Cocks 172, 176, 157, 382, and 387 were not begun till 27th January owing to there being insufficient laying hens before that date.

All eggs laid after the beginning of the experiment were incubated and examined for fertility by candling in the usual way at five or six days of incubation. All doubtful eggs were broken and the germinal disc examined under a dissecting microscope.

(b) *Flock Matings*.—In a stud-mating system such as has just been described it is obvious that the sexual activity of all males would be appreciably less than if they were at liberty with a flock of the usual fifteen or twenty hens. It was conceivable, therefore, that while the fertility of partially castrated males might compare favourably with that of normal cocks when each male was given only two to five hens per day and sometimes less, it might be different under conditions where sexual activity was unrestricted. Accordingly, three pens of flock matings were established after the termination of the first series in the early part of March. Cocks 149, 5, and 382 were each given free run of a large yard with nineteen, eighteen, and seventeen females respectively. Male 5 had not been previously tested, but both ♂ 149 and the control ♂ 382 had been used in the stud-mating series.

In these flock matings a period of ten days was allowed for fertility to become established, and then all eggs from each pen over a period of from seven to eleven days were incubated and examined as before for fertility. Some of the hens in this series had also been used for stud matings. However, in view of Crew's (1926) findings that on removal of one male and introduction of a second the influence of the first is lost after seven to ten days, it is fairly certain that the ten-day interval allowed in these experiments was ample to ensure that eggs gathered from the eleventh to the twenty-second day indicated only the fertilising power of the second male.

During the course of the experiment samples of semen were obtained and counts made of the spermatozoa therein. At the conclusion of the experiment all males were killed and weights of testes were determined.

#### OBSERVATIONS ON FERTILITY.

In the stud-mating series it was necessary to establish some arbitrary standard for the number of potentially fertile eggs. The work of Crew (1926) and Dunn (1927) indicates that in single matings a fertile egg may rarely be obtained on the first day following mating, but that fertility is well established by the second day. In Dunn's cases fertility was complete and maximum forty-eight hours after mating, but Crew found that with some males the time required for the onset of fertility was three and even five days. In these experiments all eggs laid on or after the third day from the first mating were considered as potentially fertile. The duration of fertility after removal of the male has been shown by the writers quoted and others to decrease after the first week. In these records it was considered that all eggs laid up to five days after the last

mating ought to be fertile, but any fertile eggs laid after that were also included. Infertile eggs intervening between the first and the last fertile egg counted for any hen were also considered potentially fertile.

The results observed on this basis, in both stud and flock matings, are shown in Table II.

TABLE II.—FERTILITY OF MALES TESTED.

Cock.	Number of Hens.	Potentially Fertile Eggs.	Fertile Eggs.	Fertility per cent.	Days to last Fertile Egg after removing Male.
<i>Stud Matings.</i>					
Experimental :					
21	6	127	91	71.65	18
142	5	119	110	92.44	9
152	6	119	86	72.27	10
149	7	118	98	83.05	7
172	6	67	54	80.59	12
176	5	53	35	66.04	7
157	5	58	41	70.69	
			Average*	76.67	
Controls :					
382	6	64	47	73.44	11
387	5	62	48	77.42	13
384	7	100	80	80.00	13
			Average*	76.95	
<i>Flock Matings.</i>					
Experimental :					
5	18	84	72	85.71	
149	19	126	117	92.85	
Control :					
382	17	47	33	70.21	

\* *I.e.* the average fertility of the cocks, *not* of the potentially fertile eggs.

It is evident that in the stud matings the fertility of partially castrated cocks was equally as good as that of normal cocks, the averages for the two groups being, by a coincidence, practically equal. In the flock matings ♂ 149 gave better fertility than when stud-mated, and both his record and that of the previously untested ♂ 5 were excellent. The one control cock gave slightly lower fertility than in stud matings, but this was probably due to a chance difference in the smaller number of eggs tested from his pen. The duration of fertility after removal of the male was on the average practically as long in the experimental group as in the controls.

Table III presents a biometrical analysis of the individual performances of the females in the stud-mating series.

TABLE III.—STATISTICAL CONSTANTS FOR FEMALES IN THE STUD-MATING SERIES.

Class.	Number of Individuals.	Mean Fertility and Probable Error per cent.	Standard Deviation and Probable Error per cent.
Experimental . . . .	40	76.45 ± 2.38	22.33 ± 1.68
Control . . . .	18	76.22 ± 2.76	17.41 ± 1.95

It is evident that there is no significant difference between the mean fertility in each group. The difference in the standard deviations is  $4.93 \pm 2.37$ . Since this difference is only 2.08 times its probable error, it is not statistically significant. It may therefore be concluded that the results are not affected by any undue variability of the females in either group.

#### SPERMATOZOA COUNTS.

The hypothesis suggested by these results, namely, that fertility of the male is entirely independent of the amount of testicular tissue present, could not be proved until that tissue had been weighed after death. Both Benoit (1925) and Domm and Juhn (1927) found a compensatory hypertrophy of the remaining testis following unilateral castration of young chicks. It was therefore to be expected that compensatory hypertrophy would take place to an unknown extent in some or all of the material used in this experiment. An attempt had been made to prevent such hypertrophy in ♂ 172 by grafting subcutaneously the portions of testis removed at the time of operation. If the compensatory hypertrophy were occasioned by the demand for production of a certain degree of physiological activity of the testis this demand might be met (and hypertrophy prevented) by a functioning testis in any part of the body, while obviously only the testicular material in communication with the vas deferens would be available for reproduction. Nevertheless, the amount of testis in any of the males was entirely unknown. To determine whether or not the density of sperm suspension in the semen bore any relation to the size of testis, or could be accepted as a measure of potential fertility in mating, an examination was made of thirty-six samples of semen from ten different males.

(a) *Collection of Semen.*—Determinations of the density of sperm suspension in semen of the fowl have been previously made by Payne (1914) and Craft, M'Elroy, and Penquite (1926). These workers obtained their samples from the cloaca of the hen after coitus, and it is therefore

difficult to see how a certain amount of dilution of semen by fluids in the cloaca could have been avoided. Since this method was hardly accurate, an attempt was made in the present study to secure normal semen from the ejaculate.

It was found that after the cocks had become accustomed to being handled and had been used in stud matings for over a week, they would readily copulate when a willing female was introduced, regardless of the presence of an observer. With a little practice it was possible to intercept the ejaculate and collect it in a watch crystal. By using this method, samples were obtained from the ten cocks stud-mated. Male 5, which was used only for flock matings, could not be induced to copulate when confined in a small coop with an observer present, and therefore no semen was obtained from him.

(b) *Technique of Counting.*—The densities of the sperm suspensions in the semen were determined with a Thoma-Hawksley hæmacytometer having a depth of 0.10 mm. Ringer's solution, to which had been added 3.5 per cent. of formalin, was used as a diluting fluid and gave excellent results. Without any formalin, movement of the spermatozoa was not arrested, so that accurate counting was difficult. More than 3.5 per cent. formalin caused the sperms to curl up and become more difficult to see and to distinguish from minute masses of debris. For each sample counts were made of 160 out of the 400 squares ruled off on the hæmacytometer.

"Student" (1907) has demonstrated that when the technique of dilution and counting is accurate, the distribution of yeast cells on the squares of a hæmacytometer conforms to a Poisson Series. Moreover, Fisher (1925) points out that the standard error of a random sample from such a distribution is  $\pm \sqrt{m}$ , where  $m$  (the number of cells counted) is a large value. The same error should apply to any large hæmacytometer count if the technique be accurate. Two or three counts of the same dilution of semen were compared on several occasions. It was found that the deviations from the mean were within the standard error in most cases, and in no case significantly greater than the limits of that error. It may, therefore, be considered that the technique used was satisfactory, and that the counts reported below are accurate measurements of the density of sperm suspension in the various samples of semen examined.

It was found that the numbers of spermatozoa per cubic millimetre varied somewhat between different individuals, but even more in samples from the same individual secured following various degrees of sexual

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activity. Thus, on 30th March the first sample for several days from ♂ 142 contained 5,500,000 sperms per cu. mm. After forty minutes' freedom with a hen, a sample was again obtained which contained 1,690,000 per cu. mm. The cock was left with another hen for forty-five minutes, and a sample secured thereafter had only 277,600 per cu. mm. Accordingly an attempt was made to secure representative samples from each male, *i.e.* after similar degrees of sexual activity. This presented some difficulty because of the unwillingness to copulate on the part of some of the males after one or two previous matings. In most cases this could be overcome by leaving the cock with one hen for an hour or so, then removing her and introducing another hen of a different colour, but with others a similar artifice was unsuccessful (*e.g.* ♂ 149). The data in Table IV give the average number of spermatozoa per cu. mm. in representative samples from each male, including in each case (except ♂ 149) the first ejaculation of a day, and two or more other semen samples taken after various degrees of sexual activity.

TABLE IV.—SPERMATOZOA COUNTS, TESTIS WEIGHTS, AND FERTILITY.

Bird Number.	Body Weight (grams).	Testis Weight (grams).			Ratio Body Wt. Testis Wt.	Spermatozoa.		Fertility.	
		R.	L.	Total.		Samples.	Av. No. p. cu. mm.	Flock.	Stud.
Experimental :									
21	1927	5.77	...	5.77	333.9	4	1,782,650	...	71.6
142	1843	15.95	0.55	16.50	111.7	7	1,836,650	...	92.4
152	1757	24.00	...	24.00	73.2	3	825,960	...	72.2
149	1786	13.59	...	13.59	131.4	1	5,300,000	92.8	83.0
172	1814	8.98	...	8.98	202.0	2	5,032,500	...	80.0
176	1474	...	10.52	10.52	140.1	5	3,994,380	...	66.0
157	1956	15.55	0.94	16.49	118.6	4	7,328,500	...	70.6
5	1360	1.06	3.51	4.57	297.6	...	...	85.7	...
Average :	1740	...	...	12.55	201.1	...	3,728,663	89.2	76.6
Controls :									
382	1843	9.82	8.54	18.36	100.4	4	3,615,000	70.2	73.4
387	1814	6.07	8.00	14.07	128.9	3	4,848,667	...	77.4
384	1672	6.22	4.83	11.05	151.3	3	5,425,333	...	80.0
Average :	1776	...	...	14.49	126.8	...	4,629,667	...	76.9

## WEIGHTS OF TESTICULAR MATERIAL.

In the same table are given the weights of testes found in each fowl at post-mortem examination. Only that tissue is included which was in communication with the vasa deferentia, as evidenced by active sperm being found in these ducts. In one or two cases small nodules of testes had regenerated after the operation but were not in communication with the vas and so are not included.

## INTERPRETATION OF RESULTS.

(a) *Fertility in Relation to Size of Testis.*—Inspection of Table IV shows that while the testis size and spermatozoa counts of the controls were on the average slightly higher than in the experimental males, the average fertility was practically identical in each group in the stud matings. In the flock matings the two partially castrated cocks excelled the control. The latter bird did no better in flock matings than in stud matings, while ♂ 149 showed considerable improvement.

It is particularly significant that ♂ 5, tested only in flock matings, yielded a fertility of 85.7 per cent., a figure excelled by only two of the whole eleven males, in spite of the fact that he had only 4.57 grams of testis material. Male 172, with 8.98 grams of testis, gave 80 per cent. fertility. In contrast to these, ♂ 152, with 24 grams of testis, gave only 72.2 per cent. fertility. Cock 157, with 16.49 grams of testis, secured only 70.6 per cent. fertility; while ♂ 142, with exactly the same amount of testicular material, induced 92.4 per cent. fertility.

From these data it is reasonably certain that within the ranges covered by the eleven males tested (*i.e.* fertility from 66 per cent. to 92 per cent. and testis from 4.5 to 24 grams) fertility is entirely independent both of the absolute amount of testicular tissue present and of the amount in proportion to body weight.

(b) *Fertility in Relation to Density of Sperm Suspension.*—It is also evident that within the same limits of fertility a density of sperm suspension ranging from 825,960 per cu. mm. to nine times that figure has no relation to the resultant fertility. Thus Cocks 142 and 157 had equal amounts of testicular tissue, but while the former had an average sperm count of 1,836,650 and 92.4 per cent. fertility, the latter had an average count of 7,328,500 and a fertility of only 70.6 per cent. Similarly the fertility of ♂ 152 and of ♂ 382 was practically the same, although the number of sperm calls per cu. mm. was four times as great in the semen of the latter as in the former. The cock giving the lowest fertility in

the stud-mating series had over twice as many sperms per cu. mm. of semen as the cock with the highest fertility in the same series—♂ 142.

Walton (1927) found that with rabbits, a sperm suspension of less than 1,000,000 in 3 c.c. resulted in reduced fertility, and that below 10,000 in 3 c.c. sterility occurred. Possibly the same dilutions might produce similar results in the fowl if only one insemination were made. However, since for none of the ten cocks examined was the average density of sperm suspension less than *eight hundred times* Walton's critical figure, and in all but one it was *over one thousand times that figure*, it is extremely doubtful if in normal cocks density of sperm suspension is a factor contributing to the differential fertility often observed. Moreover, insemination in the fowl occurs not just once as in rabbits, but is repeated in an irregularly continuous manner often several times daily.

It is quite conceivable, however, that in certain pathological conditions the density of sperm suspension may become so low as to affect fertility. It is quite probable that the partial sterility observed in the first breeding year of Gowen's (1926) case of a fowl with cystic testes, reflected a degree of that occlusion of the vasa deferentia which post-mortem examination revealed had later become complete. In such cases one would expect a sperm suspension low enough to affect fertility even if the sperm were physiologically normal.

The extremely high average sperm count and the low fertility of ♂ 157 may have both resulted from a mild cloacitis with which he was affected throughout the entire breeding season. It is perhaps possible that the local irritation may have induced hyper-active spermatogenesis. No great difference in motility or appearance of his spermatozoa was evident when compared with those of others.

In view of these findings, it seems reasonable to infer that the differential fertility of cocks is dependent (except in cases of obviously unwilling breeders) upon differences in the physiological efficiency of the spermatozoa. No difference in motility of sperm cells from the ten cocks was noticed, but it was observed that the motility depends to a marked extent upon the temperature of the semen when examined.

Williams and Savage (1925) have shown that even a small proportion of certain types of abnormal sperms in the semen of bulls indicates low fertility. In a later paper Savage, Williams, and Fowler (1927) have also shown that the breeding efficiency of an unsound bull can be detected by measuring the head lengths of a representative sample of spermatozoa, determining the statistical constants for the distribution and the degree

of skewness of the curve of frequency distribution. A coefficient of variation greater than certain physiological limits or a statistically significant skewness indicated a poor breeder. Such results were usually confirmed by cytological and clinical findings, but in some cases of low fertility no evidence of unsoundness was found except by the statistical analysis of head lengths of spermatozoa. This means that a great variation in size of sperm, or a certain proportion of large or small cells, indicates some unknown condition which results in poor breeding efficiency. It is probable that similar conditions apply in the case of the fowl.

(c) *Amount of Testis and Density of Sperm Suspension*.—Contrary to expectation, the density of sperm suspension does not appear to bear any definite relation to the size of testis. In the first ejaculations of the day, or after a rest period of several days, practically no difference was observed between the counts of control and experimental males. It was only after being allowed to copulate several times that variations became marked. Such differences were not necessarily in accord with the amount of testis tissue present, although the decrease was less marked in the control males. For example, the lowest count obtained was one of 18,700 per cu. mm. from ♂ 152 after he had been 1½ hours with a hen. After a longer period at liberty with two different hens ♂ 142 gave a count of 277,600. The latter had 16.5 grams of testes, the former 24. Such a difference may, of course, reflect only different degrees of activity, but in this case both were keen breeders. In general, the slightly lower average counts of experimental cocks from which three or more samples were obtained reflect reduced counts following sexual activity, this reduction being equally apparent in cocks with a large but unilateral testis (e.g. ♂ 152) as in those with less testicular tissue.

The data of Craft *et al.* (1926), when re-arranged, also indicate that the density of sperm suspension is not dependent upon the size of testis (Table V). The counts made by these workers are much below those observed by the writer. Nevertheless it is probable that the degree of error in them (*vide infra*) was approximately the same in each case, especially since the number of samples from each pen (of three White Leghorn yearling cocks) ranged from eight to twenty-two, and that therefore the average counts are comparable one with another.

It seems probable that the density of sperm suspension in the semen is a reflection more of the degree of spermatogenic activity than of testis mass, and that at any one season differences in this activity depend upon the individual peculiarities of different birds as well as upon environmental conditions.

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TABLE V.—RELATION OF AMOUNT OF TESTICULAR TISSUE TO THE DENSITY OF SPERM SUSPENSION IN THE SEMEN.

(Rearranged from data of Craft, M'Elroy, and Penquite.)

Pen.	No. of Males.	Samples counted.	Average No. Spermatozoa per cu. mm.	Average Total Testis Weight (grams).
1	3	7	105,400	22.30
2	3	20	177,500	10.80
3	3	12	225,300	13.46
4	3	11	224,900	17.01
5	2	8	96,500	12.17
6	3	10	676,000	22.66

Polowzow (1927) has recently shown that the rhythm of sperm production in the horse differs in different individuals besides being influenced by the frequency of breeding and length of rest periods.

(d) *Average Sperm Counts.*—The individual counts observed by Craft, M'Elroy, and Penquite ranged from 2000 to 4,000,000 per cu. mm. The semen was collected from the cloaca of the female *post coitu*, usually from the first matings in the morning. Since, in the present work, samples taken at this time were much higher than those taken after several matings, the writer is of the opinion that the low average counts of Craft *et al.* indicate that their samples were somewhat diluted by fluids of the cloaca. The determinations made in the present experiment ranged from 18,700 to 8,864,000 per cu. mm., with an average of 4,015,088 for 36 counts. The average counts for individual males ranged from 825,000 to 7,328,500, with an average of 3,998,642 for each of the ten cocks tested. Payne (1914) found that the density of sperm suspension in the semen of five different cocks ranged from 1,920,000 to 5,470,000 per cu. mm., with an average of 2,928,000. These figures are in accord with those observed in this experiment.

(e) *Amount of Ejaculate.*—It was found exceedingly difficult so to improve upon the method of collecting semen used in the present study that one could always secure the entire ejaculate. It was also difficult to measure the exact volume of the very small samples invariably obtained; the weight, however, could be accurately determined. The weights of eight samples of semen definitely known to contain the entire ejaculate are given in Table VI.

These data merely indicate that there is considerable variation in the amount of the ejaculate of different individuals and of the same individual

at different times. It is also apparent that the density of sperm suspension is independent of the amount of ejaculate. The available data do not permit of any statement concerning the relation between the total number of sperms ejaculated and fertility.

TABLE VI—WEIGHT OF EJACULATE WITH CORRESPONDING SPERM COUNTS.

♂.	Ejaculate (grams).	Sperm per cu. mm.
152	0.079	787,200
152	0.184	...
157	0.124	8,864,000
176	0.036	5,935,200
142	0.055	564,800
383	0.067	2,720,000
"	0.141	5,962,000
"	0.217	5,864,000
Average :		0.113

(f) *Compensatory Hypertrophy*.—With regard to compensatory hypertrophy, it is evident (Table IV) that in males 142, 152, 149, and 157 the retained testis had hypertrophied to an extent approximately equivalent to the combined weight of both testes in the three controls. Indeed, in ♂ 152 the remaining testis was larger by 5.64 grams than the total testis material in the largest control pair. In another cock (♂ 147) unilaterally castrated at sixteen days, but untested for fertility owing to shortage of females, the retained right testis was found on autopsy thirteen months after the operation to be 17.1 grams. The results in this case are uniform with those of the other four.

In general these findings agree with those of Benoit (1925) and Domm (1927), but they differ from the latter's results in one important particular. Domm found that unilateral castration of birds 16, 24, or 40 weeks of age resulted in compensatory hypertrophy of the retained gonad whether right or left was removed, but that if one testis were removed from a week-old chick, compensatory hypertrophy resulted only when the left testis was retained and *not when the right one remained*. In all of the five birds mentioned above compensatory hypertrophy of the *right* testis was manifest to an extent approximating the normal weight of two testes.

On the other hand, the right retained testis of ♂ 21, operated on at five weeks of age, did not hypertrophy at all, and represents exactly what was left at the operation, *i.e.* half of a testis. Similarly ♂ 5, from whom

the left testis and half of the right were removed at seven days of age, had only 1.06 grams of testis on the right side at post-mortem but had regenerated a 3.51 nodule of testis tissue on the left. Neither the data now presented nor those of the other two workers afford a satisfactory explanation of why there should not have been hypertrophy in these two cases to the same extent as in the others. The theory suggests itself that mutilation of the surviving testis may prevent its hypertrophy, but the numbers are too small to substantiate the theory.

Cocks 172 and 176 present another aspect of the case. Both these birds were operated upon at twelve to fourteen days (Table I), but the tissue removed was grafted subcutaneously under the wing. These grafts persisted and were present at autopsy over a year after the operations. That of ♂ 172 was 2.41 grams, while ♂ 176's graft was only 0.125 gram. In neither of these cases was there any distinct evidence of hypertrophy (see Table IV). Another cock, untested for fertility (♂ 162), fell in the same class with a retained right testis of 7.58 grams, and an unweighed testis graft 1.9 cm. × 1.5 cm.

While the present data support the previous workers' establishment of the occurrence of compensatory hypertrophy, they are not sufficient to account for that phenomenon. The obvious assumption is that a certain amount of testis tissue is necessary to maintain the balance between different organs of the body. Nevertheless, some of the cases reported above have matured and reproduced in every respect like normal males, yet had only about half of a normal testis. The fact that hypertrophy was not evident in the three birds carrying grafts suggests that the balancing action of the testis calls for production of an optimum amount of testicular hormone, and that if this be in part supplied from a graft the testis retained need not hypertrophy. However, the grafts of these birds, plus retained testis, did not amount to much more than a single normal testis.

#### SUMMARY.

1. Fertility tests of eight partially castrated cocks and three controls show that within quite wide limits of fertility and of testis size the fertility of the male fowl is not in any way dependent upon size of the testis.
2. The average density of sperm suspension was found to be approximately four million spermatozoa per cubic millimetre for the thirty-six samples examined from ten males.
3. The variations in the average density of sperm suspension from

different birds ranged from 825,000 to over 7,000,000 cells per cu. mm., but within this range the density of sperm suspension bore no relation to fertility.

4. The number of sperm per cubic millimetre of semen appears to be entirely independent of the size of the testes within the ranges covered by this experiment.

5. Compensatory hypertrophy to a degree approximating to the normal weight of both testes was observed in the retained right testes of cocks castrated unilaterally on the left side at one week of age.

6. Exceptions to the rule of compensatory hypertrophy included birds with subcutaneous testis grafts and two in which one whole testis and part of the other had been removed.

7. It is suggested that fertility in the male fowl is dependent upon the physiological efficiency of the spermatozoa rather than upon their quantitative production.

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X.-XII.—Studies in Embryonic Mortality in the Fowl.

I. The Frequencies of Various Malpositions of the Chick Embryo and their Significance.

By F. B. Hutt, B.S.A.

II. Chondrodystrophy in the Chick.

III. Chick Monsters in Relation to Embryonic Mortality.

By F. B. Hutt, B.S.A., and A. W. Greenwood, Ph.D.



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X.—Studies in Embryonic Mortality in the Fowl. I. The Frequencies of Various Malpositions of the Chick Embryo and their Significance. By F. B. Hutt, B.S.A., Animal Breeding Research Department, University of Edinburgh. *Communicated by* Dr F. A. E. CREW. (With One Plate.)

(MS. received October 5, 1928. Read January 7, 1929.)

INTRODUCTION.

THE loss to the poultryman from mortality among embryos during incubation is probably second only to that from disease. In the attempt to reduce this loss many investigations have been made of the influence on incubation of such environmental factors as temperature, humidity, oxygen supply, methods of turning and cooling, etc., as well as of the influence of the ration fed to the breeding stock. However, even when conditions of nutrition and incubation are ideal so far as present knowledge goes, there is still a mortality in the incubator that may range as high as 50 per cent. of the fertile eggs and is seldom less than 25 per cent.

The investigations reported in this and the two following papers of the series were made to determine the extent to which various types of abnormalities occasionally observed in the chick embryo were responsible for the high mortality rate.

MATERIAL.

This investigation is based upon examination of over 12,000 eggs which had failed to hatch, including dead germs at first test (eight or nine days) and eggs unhatched at twenty-one days, from the following four sources, all in Scotland:—

Animal Breeding Research Department.

Romanno Bridge Poultry Farm, West Linton, Peeblesshire.

W. Johnston & Co., Cowden, Comrie, Perthshire.

Sussex Hatcheries, Glasgow.

To the three firms mentioned we express our thanks for the eggs and data so willingly supplied and without which this investigation could not have been made. These sources will hereafter be designated A, B,

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C, and D, but these letters will not represent the firms in the same order as they are listed above.

The number of eggs received from each source, together with the breeds kept and incubators used, are given in Table I.

TABLE I.—MATERIAL USED IN THE INVESTIGATION.

Source.	Dead Germs. 1-10 days.	D.I.S. 10-21 days.	Breeds.*	Incubators.	Times turned daily.
A	1775	8059	W.L., Bl.L., W.W.	Blue Hen Miller	3 1
B	271	744	W.L., R.I.R., W.W., B.R., L.Suss. × R.I.R.	Glevum (mammoth)	5
C	...	654	W.L., Anc., R.I.R., R.I.R. × L.S.		
D	104	190	Friz., Br.Leg.	Hearson	2
	2150 †	9647 †			

\* Abbreviations as follows :—

W.L.—White Leghorn  
Bl.L.—Black Leghorn  
Br.L.—Brown Leghorn  
L.S.—Light Sussex  
Friz.—Frizzle

W.W.—White Wyandotte  
R.I.R.—Rhode Island Red  
B.R.—Barred Plymouth Rock  
Anc.—Ancona.

† Exclusive of eggs from "chondrodystrophy matings" at this Department.

Practically all of the eggs examined came from settings between 12th February and 1st May, with the exception of some later and earlier settings at this Department. The total of 11,797 unhatched eggs examined represents settings of 39,760 eggs from nine different breeds, and in several types of incubators under different management, and may therefore be considered as a fairly representative sample of incubated eggs in general. In some cases eggs removed at first test were not sent to us, and from some lots (*e.g.* all from C) only those dead at hatching were received. Actually, therefore, the total number of dead embryos from the 39,760 eggs set was somewhat greater than the 11,797 examined by us. However, since data was obtained from each hatch, even where

the eggs were not examined, we are able to determine for each source the average infertility, dead at first test, dead at hatching, and chicks hatched during the season's operations. These data are given in Table II.

TABLE II.—SEASON'S AVERAGE INCUBATION RESULTS FOR EACH SOURCE OF MATERIAL.

Source.	Of Eggs set.		Of Eggs fertile.		
	Infertile. Per cent.	Hatched. Per cent.	Dead Germs. Per cent.	Dead-in-shell. Per cent.	Hatched. Per cent.
A	6.75	57.07	9.75	29.04	61.21
B	8.19	72.95	6.80	17.24	75.96
C	8.48	65.11	8.81	20.06	71.13
D	5.04	64.08	11.50	21.02	67.48

"Dead germs" in this paper refers to those eggs in which the embryo was distinctly dead at eight or nine days when the first test was made. It includes embryos ranging from one to eight days old.

"Dead-in-shell" refers to those embryos found in eggs unhatched after twenty-two days of incubation. Since doubtful cases are often left in at first test, this class will include some embryos of less than eight days, but the majority will range from eight to twenty-one days. The proportion of embryos eighteen days and over is given in Table III.

#### METHOD.

Eggs were received from the contributors a few days after the termination of each hatch. Each egg was broken, the embryo examined and recorded as "too young to detect abnormality," "18-day normal," "Position II," "teratological abnormality," etc., in its proper category by checking it off in a counting-board consisting of six rows of differently coloured wooden beads. All teratological abnormalities were separated and resorted into their various classes.

The abnormalities observed in this survey were of three distinct classes, and will therefore be discussed separately under the following heads:—

1. The Frequency of Various Malpositions of the Chick Embryo and their Significance.
2. Chondrodystrophy in the Chick.
3. Chick Monsters in Relation to Embryonic Mortality.

## I. THE FREQUENCY OF VARIOUS MALPOSITIONS OF THE CHICK EMBRYO AND THEIR SIGNIFICANCE.

During the examination of the first two lots of eggs from one of these farms, it was found that many of the fully formed dead embryos were apparently abnormal only in their positions. Accordingly data were kept on the frequency of the most common malpositions found among the eggs subsequently examined. Such abnormal positions were recorded only among the embryos approximately eighteen days old or over, since prior to this stage of development, persistent malpositions could not be determined with any degree of accuracy. Among 8295 unhatched embryos (excluding all that had been discarded at first test on the eighth day of incubation), 1935 had died before the tenth day and 929 from the tenth to seventeenth days. The remaining 5050 which were approximately fully formed provided the data given below.

The normal position for the chick just before emerging is as follows: The head is towards the larger end of the egg (or toward the air-cell in oval eggs). The neck is so bent as to bring the head to the right side of the body and backwards with the beak under the right wing and just external to the femorotibial joint (fig. 1). As a consequence of this position the tip of the beak rests near the shell at a point just where the inner shell membrane separates from the outer to form the air-cell. The feet are folded on the ventral side of the body almost exactly the same as in a trussed fowl, except that the toes reach to the head.

The deviations from this position which we have most frequently observed, the first three of which have also been noted by Sanctuary (1925), are as follows:—

*Position I.*—The head is not bent to the right but extends down the median ventral line and is buried between the thighs (figs. 2 and 3). In some cases of this class the head is turned slightly to the left, but the beak is still caught between the thighs. In the less extreme cases of this type of abnormality in position the beak is just held enough by the tibia to prevent it from reaching the shell. A number of embryos which had died at twelve to fourteen days were found in Position I.

*Position II.*—The chick is turned completely upside down so that the head is in the small end of the egg (figs. 4 and 5).

*Position III.*—The head of the chick is in the large end of the egg but is turned to the left instead of to the right. In most cases this brings

the beak near the shell at a point directly opposite the air-cell (figs. 6 and 7).

*Position IV.*—The chick lies with the head toward the large end and the beak under the right wing as in the normal position, but is rotated within the shell, so that the beak is buried opposite to the air-cell (fig. 8). This abnormality was not noticed as being of frequent occurrence till toward the end of the incubation season, hence data concerning it are available for only 1085 dead embryos of eighteen days or over.

Several chicks were found in combinations of Positions I and II or of Positions II and III. Other abnormalities found in much smaller numbers included chicks normal in every respect except that the head was not under the wing, and some in which the beak pointed toward the large end of the egg instead of toward the side.

The frequency of embryos in Positions I, II, and III is shown in Table III.

TABLE III.—FREQUENCY OF POSITIONS I, II, AND III.

Source.	Dead-in-shell at Hatching.	18-day Embryos.	Normal (incl. Pos. IV.)	I.	II.	III.	I and II.	II and III.	Total Abnormal.
A	6707	4114	2693	363	758	272	16	12	1449
B	744	458	270	41	85	56	2	4	194
C	654	380	229	48	62	37	2	2	155
D	190	98	53	15	20	9	...	1	46
Totals.	8295	5050	3245	467	925	374	20	19	1844

It should be particularly noted that in the above table the embryos classified as normal include those in Position IV, since records of this abnormality were not kept till towards the end of the season. The data on Position IV are given in Table IV.

It is reasonable to assume that these smaller lots may be taken as a representative sample of the dead-in-shell from each source, and that the percentage of Position IV embryos found in these samples would be approximately correct for the entire lot of dead-in-shell from each source. On this basis we have deducted from the embryos classed in Table III as normal the calculated number in Position IV, and have thus been able to determine the frequency in percentage of all four abnormal positions in proportion to the true normal positions among all the 18-day

TABLE IV.—FREQUENCY OF POSITION IV.

Source.	Dead-in-shell at Hatching.	18-day Embryos.	Position IV.	
			No.	Per cent. of 18-day Embryos.
Poultry Farm A	1222	850	176	20·70
" B	129	117	21	17·95
" C	230	188	35	18·08
" D	79	41	6	14·63

(or over) embryos from each source. These data are presented in Table V.

TABLE V.—RELATIVE FREQUENCY OF POSITIONS I, II, III, AND IV.

Source.	18-day Embryos.	Normal, per cent.	Pos. I, per cent.	Pos. II, per cent.	Pos. III, per cent.	Pos. IV, per cent.	I and II, per cent.	II and III, per cent.	Total Abnormal Positions, per cent.
A	4114	44·77	8·82	18·42	6·61	20·70	·38	·29	55·22
B	458	41·00	8·95	18·56	12·23	17·95	·44	·87	59·00
C	380	42·20	12·63	16·29	9·74	18·08	·53	·53	57·80
D	98	39·65	15·31	20·20	9·18	14·63	...	1·02	60·34
Average (weighted)	5050	44·14	9·25	18·31	7·41	20·12	·39	·37	55·85

## SIGNIFICANCE OF ABNORMAL POSITIONS.

It cannot yet be definitely stated to what extent all these abnormal positions prevent chicks otherwise healthy and normal from emerging.

It is reasonably certain that none of the chicks in Position I (head buried between the thighs) would be able to break the shell. Even the normal chick is unable to raise the neck till some time after it has left the shell, and in most cases chicks in Position I had the head so tightly wedged between the thighs that to raise it would be much more difficult than in the normal chick. In a very few of these chicks the beak was close enough to the shell to break it if the chick had been able to do so. Since none of the 467 embryos found by us in Position I nor the 196 reported by Sanctuary, had pipped the shell it is reasonable to conclude that chicks in Position I are unable to hatch. From our data this would account for 9·25 per cent. of the fully formed dead-in-shell at hatching

time. Sanctuary found this abnormality in 196 cases out of 1490 "full-time" embryos, or 13.15 per cent., it being more common in his material than either Positions II or III.

In order to realise the extent to which Positions II, III, and IV may prevent or hinder hatching, some understanding of the normal process is essential. The most detailed available account of the actual process of hatching is still that of Réaumur (1751), first published in 1749. He describes the position of the chick in the shell, elaborates on the difficulty of its task to pierce the tough membrane and the hard shell wall, and goes on to describe how the first crack is extended till the shell is broken almost all the way around the egg, thus permitting the chick to burst the two portions asunder and tumble out. Since the head of the chick remains under the right wing up to the time when the two parts of the shell are separated, this extension of the first opening can be made only if the chick rotates in the shell. Réaumur was perplexed to know whether this rotation was done by means of the beak, or the only other probable agency, the feet. He finally satisfied himself by a simple experiment that the latter were responsible for the rotation. He first determined that the fracture was lengthened from left to right, then by removing shell and membrane in advance of the chick's beak, removed the only object against which the beak could exert any force. Since the chick was able to rotate and hatch as in normal cases, Réaumur concluded that the feet were responsible for the rotation.

Our own observations are essentially in accord with those just quoted, except that we are inclined to think that one of the most important steps in hatching occurs before any of the process described by Réaumur—*i.e.* the penetration of the beak through the inner shell membrane into the air-cell, so that the allantoic respiration is supplemented by normal breathing through the lungs. That this does happen is attested by the peeping of the chicks, which is usually heard before the eggs are pipped. Possibly Réaumur took this part of the process for granted. The number of chicks which we have found dead-in-shell with the head far removed from the air-cell suggests that the initiation of pulmonary respiration is of prime importance if the chick is to emerge successfully.

The physiological changes in the embryo prior to hatching as described by von Baer also emphasise this point. (We quote from Lillie's (1919) summary.)

"Important changes preparatory to hatching take place on the seventeenth to the nineteenth days. The fluid decreases in the amnion. The neck acquires a double bend so that the head is turned forwards, and in

consequence the beak is towards that part of the membranes next to the air-chamber. The intestine is retracted completely into the body-cavity, and on the nineteenth day the yolk-sac begins to enter the body-cavity. On the twentieth day the yolk-sac is completely included, and practically all the amniotic fluid has disappeared. The chick now occupies practically all the space within the egg, outside of the air-chamber. The umbilicus is closing over. The ductus arteriosi begin to contract, so that more blood flows through the lungs. The external wall of the allantois fused with the chorion still remains very vascular.

"Now, if the chick raises\* its head, the beak readily pierces the membranes and enters the air-chamber. It then begins to breathe slowly the contained air; the chick may be heard, in some cases, to peep within the shell two days before hatching, a sure sign that breathing has begun. But the circulation in the allantois is still maintained, and it still preserves its respiratory function. When the chick makes the first small opening in the shell, which usually takes place on the twentieth day, it begins to breathe normally, and then the allantois begins to dry up and the circulation in it rapidly ceases. It then becomes separated from the umbilicus, and the remainder of the act of hatching is completed, usually on the twenty-first day."

Since the allantoic circulation is maintained till the twentieth day it is quite possible that strong chicks may be able to pierce the shell without ever having breathed through the air-cell. This is evidenced by the chicks occasionally found hatching (or pipping the shell) at the small end of the egg. Although in certain of these cases the air-cell may be at the small end, it is more likely that such chicks were strong enough to break the shell and begin normal breathing before allantoic circulation stopped, and without the intermediate use of the air-cell. It is probable, however, that the extra energy consumed by the physical exertion attendant upon the attempts to pierce the shell puts a burden on the respiratory apparatus greater than can be accommodated by the allantois alone. The normal chick has both pulmonary and allantoic respiration to meet this burden. The chick in Position II, III, or IV is in most cases unable to reach the air-cell, and is therefore dependent on the allantois alone. It is reasonable to assume that only the strongest chicks are able to overcome this handicap and hatch successfully.

\* The chick does not "raise" its head in the usual sense of the expression, *i.e.* extension (in contradistinction to flexion) of the head upon the neck. Actually it thrusts the beak forward really toward its back, and so pierces the inner shell membrane to enter the air-cell (see fig. 1).

Some idea of the importance of pulmonary respiration at this stage is conveyed by the observations of Dr F. R. Horner (quoted by Tegetmeier, 1867), that before breaking the shell the chick breathes at the rate of about eighty times a minute, but that this rate decreases after a hole has been made in the shell large enough to admit air freely.

Histological examination of lung tissue from chicks found fully formed, but dead in Positions II and III, showed plainly that the lungs, though well supplied with blood, had never been used.

In addition to the handicap imposed by the inability to breathe through the lungs, many of the chicks in Positions II and III are hampered mechanically in their efforts to break the shell. It is manifest that a chick with the head in the small end of the egg is very cramped. Many have been observed in positions where the head had apparently no room whatsoever to move. Others were placed so that even if they did strike the shell the blows would be in such a slanting direction that they would have very little effect.

It should be noted that the breaking of the shell membranes, and shell is facilitated—if not accomplished entirely—by the egg tooth. The position of this structure on top of the upper beak suggests that the taps on the shell are made by upward movements of the beak rather than by direct forward hits. The observations of Breed (1912) confirm this suggestion. Some of the chicks found by us in Positions II and III were so situated that it was difficult to see how the egg tooth could be brought into efficient action against the shell, even had the cramped quarters of the chick permitted any upward thrusts of the beak.

In some cases ruptured blood-vessels in the chorio-allantoic membrane surrounding chicks dead in abnormal positions, in unpipped eggs, suggested that their efforts to break the shell had not only failed but had probably wrought their own destruction by injuring the only available means of respiration.

It is difficult to say whether or not the failure of the head to be under the wing affects the ability to hatch of the chicks occasionally found in this position. Réaumur was of the opinion that the wing served to guide the thrusts of the beak.

#### CAUSES.

From the fact that the proportions of the various abnormalities do not vary a great deal among the eggs from different sources, it is doubtful if they are due to any one method of incubation, turning, or handling. It would be of interest in this connection to determine the

frequency of these abnormal positions in eggs incubated under hens, which are turned much more than those in incubators. It seems possible that Position IV in which the embryo is normal except in being turned from the air-cell, may be caused by its being turned to that position just before it becomes too large to move freely within the shell. If so, the critical point after which it would be safer not to turn the eggs, comes before the period at which turning is usually stopped, *i.e.* the end of the 18th or 19th day. In this connection the recent experiments at the South Dakota Experiment Station (1927) are of special interest. It was found that eggs turned only to the 15th day gave better hatches than those turned to the 19th day. If turning beyond the critical point be the cause of Position IV, the fact that only a part of the embryos examined by us were affected can be explained by their being not all at exactly the same stage of development.

It is probable that some of the abnormalities (particularly Positions II or III of the full grown embryos) result directly from an incorrect orientation established by the first few cleavage divisions. Normally the embryo first lies at right angles to the long axis of the egg, and in such a way that when the small end of the egg is pointed away from the observer the head of the embryo is directed to the left. This orientation is preserved until about fourteen days of incubation, at which time the embryo becomes adjusted to the shape of the egg.

Duval (quoted from Lillie) found that out of 166 fertile eggs observed at an early stage, 124 embryos were oriented exactly as described, in 39 the axis was slightly oblique, 2 had the head toward the large end and 1 was completely reversed from the normal position. Similarly, rearrangement of Féré's (1899) data shows that of 296 embryos examined by him, 42 deviated from the correct axis—3 by 15°, 27 by 45°, 7 by 90°, and 5 by 180°. Féré classed these with other abnormalities induced by exposure to fumes of alcohol, but since they occurred just as frequently in his controls as in treated eggs, it is evident that the incorrect orientation occurred naturally and was not due to experimental procedure.

These observations afford quite a likely explanation of at least some of the abnormalities in position, but it remains to be shown what factors cause the embryonic axis to vary from its normal position at right angles to the long axis of the egg as delineated by the chalazæ. The extent to which different methods of storing, handling, turning, and incubating eggs can cause these abnormal positions must also be determined experimentally.

The incidence of embryos in two malpositions combined suggests that no single cause is responsible for more than one abnormal position. The average frequency of Position I is 9.25/100 (Table V). The average frequency of Position II is 18.31/100. If each of these malpositions was caused by entirely independent agencies one would expect them to occur together purely by chance in  $(9.25/100)(18.31/100) = 1.69/100$  cases. If both of these positions could be induced by the same cause, the frequency of embryos combining Positions I and II would be greater than 1.69 per cent. expected by purely chance association. Actually these two malpositions occurred together in only 0.39 per cent. of the total number, *i.e.* considerably less than would be expected on the basis of two independent causal agencies. It is difficult to believe that any embryos in both Positions I and II could possibly have hatched. The data may therefore be taken to indicate not only that two separate factors are responsible for Positions I and II, but also (since their coincidence is less than would be expected by chance association) that the two causes are probably somewhat antagonistic.

Similarly the frequencies of Positions II and III are 18.31 and 7.41 per cent. respectively of all the embryos examined. On the basis of a purely chance association, one would expect them to occur together in 1.35 per cent. of the population. Only 0.37 per cent. were recorded in II and III together. Here, again, the evidence shows that two distinct agencies are at work, one causing Position II, the other Position III, and that they tend to be antagonistic rather than associated. Such an antagonism may be nothing more than the greater mechanical restriction imposed upon an embryo with the head in the small end, than on one with the head in the large end, so that in the former case it is more difficult for the head to become turned to the left away from the normal position.

The analysis just given indicates that in the attempt to eliminate malpositions a distinct cause for each one must be sought.

Since in the case of the majority of the eggs examined it was impossible to trace those from individual hens, we are unable to say if the tendency to produce embryos in these abnormal positions is hereditary or not. There is some evidence from our own flock that this may be so.

No significant difference was observed between the frequencies or proportions of these abnormal positions in eggs examined early in the season (February) and those from April and May settings.

SUMMARY.

1. An examination has been made of 11,797 eggs which had failed to hatch, among which were 5050 embryos which had died after the eighteenth day.

2. Four major malpositions of the chick embryo are described and the frequency of each given.

3. It is concluded that one of these, in which the head is buried between the legs, definitely prevents hatching. In the material examined, this abnormality was responsible for 9.25 per cent. of the mortality among embryos of eighteen days or over.

4. It is suggested that the other three malpositions usually result fatally, by reason of their preventing pulmonary respiration in the embryo as well as by mechanical hindrance.

5. Of the embryos over eighteen days, nearly 56 per cent. were in one or another of the four major malpositions.

6. Possible causes are discussed, and the suggestion made that some of the abnormal positions result from an incorrect orientation of the embryo established early in cleavage.

ACKNOWLEDGMENTS.

The writer is indebted to the Romanno Bridge Poultry Farm, West Linton, W. Johnston & Co., Comrie, and the Sussex Hatcheries, Glasgow, for material supplied for this investigation. He is especially indebted to Dr A. W. Greenwood for valuable assistance in examining the eggs. Thanks are also due to Professor F. A. E. Crew for constructive criticism and advice, and to Mr J. M. A. Chisholm, of this Department, who did all the photography.

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EXPLANATION OF PLATE.

Fig. 1. Normal position prior to hatching.

Figs. 2 and 3. Position I. Head buried between the thighs.

Fig. 4. Position II. Head in the small end of the egg.

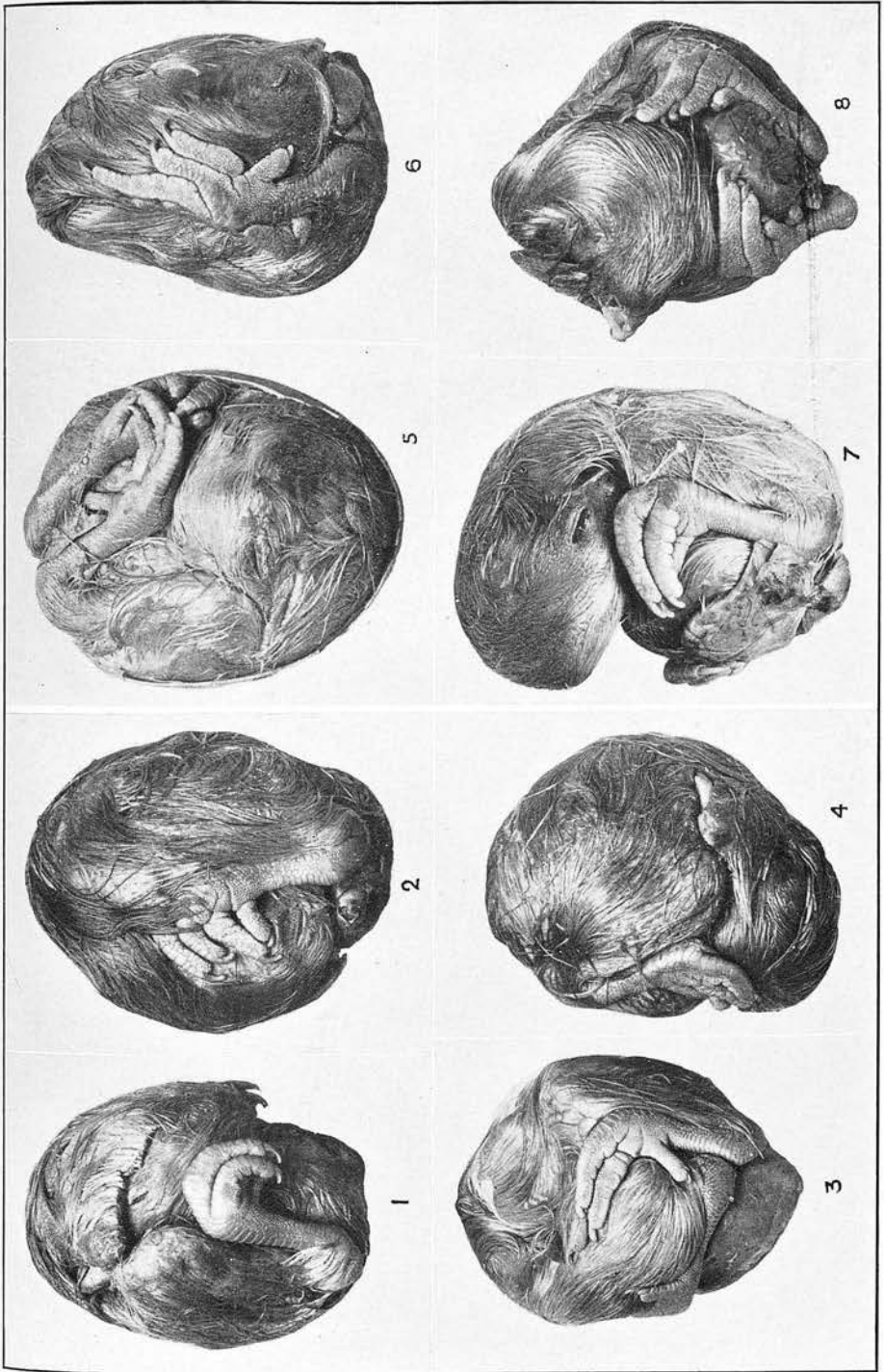
Fig. 5. Positions II and III combined. Head in the small end of the egg, and turned to the left instead of to the right.

Fig. 6. Position III. Head turned to the left and in this case extending to the small end of the egg.

Fig. 7. Position III. Head to the left.

Fig. 8. Position IV. The embryo is turned so that the beak is buried away from the air-cell.

(Issued separately April 1, 1929.)



XI.—Studies in Embryonic Mortality in the Fowl. II. Chondrodystrophy in the Chick. By F. B. Hutt, B.S.A., and A. W. Greenwood, Ph.D., Animal Breeding Research Department, University of Edinburgh. (With One Plate and One Text-figure.)

(MS. received October 5, 1928. Read January 7, 1929.)

CHONDRODYSTROPHY.

AMONG the eggs incubated in January 1928 at this Department a number of embryos were found with the parrot beak and short legs peculiar to that abnormality first observed in chick embryos by Dunn (1923), and tentatively termed by him the "parrot" embryo. More recently Landauer's (1927) extensive study of the morphology and histology of these embryos has shown that the condition corresponds closely to the *chondrodystrophia fetalis* or *achondroplasia* of mammals, of which the "bull-dog" calf of the Dexter and the achondroplastic dwarf in the human are well-known examples.

All embryos dying during incubation at this Department were thereafter systematically examined till by March it was found that 21 hens out of 76 from which eggs had been incubated had produced one or more chondrodystrophic individuals. Of these the 18 which gave most were put in one pen on 7th March, and all their eggs, till 1st April, were examined at 11 to 13 days of incubation. Thereafter all their eggs were allowed to incubate 21 days. When it was found that very few chondrodystrophics were obtained from the April eggs, breeding was discontinued except for random samples from these 18 hens set during May and June.

*Description of Chondrodystrophy.*

The more extreme cases of chondrodystrophy are characteristically and markedly abnormal. The lower beak is much shortened, and the upper beak bends down over it, giving the characteristic "parrot beak" appearance (Pl. I, figs. 1, 2, and 4). The frontal and parietal bones of the skull are vaulted anteriorly to make a much rounder head than is found in the normal embryos. The legs are exceptionally short. This is caused partly by a general shortening and thickening of the bones and partly by a bending which may be in the femur, tarso-metatarsus, or tibia (Pl. I, fig. 6). In our material, bending of the tibia was most

evident, although in some cases the tarso-metatarsus or femur was also badly bent. The effect of this bending is to turn the plantar surface of the foot toward the body. The epiphyses of the long bones are often markedly enlarged.

In less extreme cases there may be no trace of prognathia in the upper beak, but the bend in the tibia is still present in varying degrees, some of which can be best detected by microscopic examination of sectioned bones.

In our examination of eleven- and thirteen-day living embryos it was noticed that feather growth was so retarded in extreme chondrodystrophics that in some cases less down was present at 13 days than is found on the normal embryo at 10 days (Pl. I, figs. 1, 2, and 3). In chondrodystrophic embryos found dead in other material at 16 to 18 days it was noted that the down was somewhat thinner and seemed somewhat curled, so that in some cases one could identify a chondrodystrophic individual before the beak or tibia had been examined (Pl. I, fig. 5).

In the young living embryos (11-13 days) it was also noticed that the embryos had a swollen appearance, caused by a gelatinous substance just beneath the skin. Traces of this were apparent in older embryos.

Extreme cases which have survived to the 18th day present the characteristic appearance shown in Pl. I, fig. 5. The yolk-sac has been retracted, and is so much larger in proportion to the dwarfed "chondro" embryo than to the normal chick, that the general appearance of the former is a small head protruding from a spherical mass, mostly yolk. Less extreme cases, 18 days or over (Pl. I, fig. 6), may approach the normal so closely as to require sectioning of the tibia and microscopic examination.

For a complete account of the pathology and histology of the chondrodystrophic skeleton the reader is referred to Landauer's (1927) detailed study.

Dead chondrodystrophic embryos were found by us at all stages from 8 to 21 days of incubation. The majority of the eggs from our own chondrodystrophy matings were examined at 13 days, at which time the abnormality is easily recognised. In some cases the embryos had died before the tenth day. In the eggs from outside sources the majority of chondrodystrophic embryos had died during the second week of incubation. We have not known one to hatch, but found one chondrodystrophic embryo alive in the shell at the twenty-second day.

Chondrodystrophy has been found combined with microphthalmia and exencephaly, but only in a very small and insignificant number of cases.

*Frequency of Chondrodystrophic Embryos.*

It was very difficult to determine the exact number of chondrodystrophics in the material from the three outside sources—Commercial Poultry Farms A, B, and C. (Full details of this material are given in the first paper of this series.) Decomposition had become well advanced in many cases, so that in most of the embryos under 10 days definite diagnosis was impossible. Only 6 chondrodystrophics were positively identified in the 2046 “first test” eggs (*i.e.* dead before 9 days) examined, and these were all from one lot of 280 such eggs removed at Poultry Farm A from eggs set 26th February. Of the eggs classed as dead-in-shell, 24 per cent. were too young or too decomposed to be of value. The incidence of chondrodystrophy in the remaining “valid” embryos is shown in Table I. Only those in which identification was positive have been included.

TABLE I.—INCIDENCE OF CHONDROS FROM CONTRIBUTING FLOCKS.

Source.	D.I.S. examined.	“Valid” Embryos.	Chondrody- strophics.	Per cent. of valid Embryos.
Poultry Farm A	8059	5990	48	.80
” ” B	744	653	52	7.96
” ” C	654	492	12	2.44
Our own stock D	...	1900 (eggs set)	124	6.53

At this Department 124 chondrodystrophic embryos were found. They first appeared in the eggs from some of 65 hens being used to test the fertility of partially castrated males. These eggs were incubated only till the sex of the embryo could be easily determined, *i.e.* to about the eleventh day. As soon as chondrodystrophic embryos were observed careful examination was made for them in all eggs set in this and other experiments. By 8th March, 21 hens had been found to produce one or more specimens of this abnormality, and 18 of these were then penned separately. One other hen subsequently produced 2 chondrodystrophic embryos. All eggs from these 18 were opened at the eleventh or thirteenth day, until early in April, when it became apparent that the chondros were decreasing. Thereafter the chicks were allowed to hatch, and only the unhatched eggs were examined for abnormalities.

Out of a total of approximately 1900 eggs set at this Department from 126 hens, 124 chondrodystrophic embryos were obtained, all in the progeny of 22 hens.

*Seasonal Incidence of Chondrodystrophy.*

The data just given and those in Table I give little indication of the extent to which chondrodystrophy may be responsible for embryo mortality. This is because the condition was found much more frequently early in the season than in the later hatches. Thus, of the 48 chondrodystrophics out of 5990 "valid" embryos from A, 37 appeared among 2453 embryos from February incubations. The remaining 3537 dead embryos from March and April settings contained only 11 specimens of this abnormality. Similarly, in B's eggs the frequency of chondrodystrophics was 8.5 per cent. of the valid embryos in February, 6.9 per cent. in March, and 3.3 per cent. in April.

In our own "chondrodystrophy matings" (*i.e.* the 21 chondrodystrophy-producing hens) the frequency of this abnormality was as high as 34.78 per cent. of the eggs set in January, but declined steadily to a complete absence in June (Table II).

TABLE II.—SEASONAL DISTRIBUTION OF CHONDRODYSTROPHIC EMBRYOS FROM 21 CHONDRODYSTROPHY-PRODUCING MOTHERS AT THE A.B.R.D.

Month of Laying.	Hens Laying.	Fertile Eggs Set.	Chondrodystrophic Embryos.	
			No.	Per cent.
January . . . . .	6	23	9	34.78
February . . . . .	21	150	48	32.00
March . . . . .	20	204	59	28.92
April . . . . .	13	47	5	10.64
May . . . . .	8	18	1	5.55
June . . . . .	9	38	0	0
Total . . . . .		480	122	

Dunn (1927) found a distinct tendency toward a decrease in chondrodystrophy in his "C" series as the season advanced, but was dealing with smaller numbers over a shorter season, and therefore made no definite conclusion. The data in Table II, supplemented by that from the A, B, and C eggs, establish fairly conclusively that the occurrence of chondrodystrophy is influenced by season, and drops to a comparatively low figure in the summer months. Two chondrodystrophics have been obtained here from eggs laid in July, and therefore it cannot be said that it does disappear entirely. It was noted, particularly in the eggs from

Poultry Farm A, that chondrodystrophic embryos found later in the season were "low grades," *i.e.* much less abnormal than those observed from February settings.

One hen, Brown Leghorn 115, had a chondrodystrophic embryo in her egg of 4th February, but seventeen eggs recorded between that date and 7th May contained only normal embryos. The egg laid on the latter day yielded an extreme chondrodystrophic embryo, with parrot beak and an angle of almost  $90^\circ$  in the tibiae. Another hen produced two chondrodystrophics only—1 in an egg laid in February and 1 from the egg of 9th May. Our data suggest no reason for such a discontinuous distribution.

♀ No. 92 produced 28 chondros out of 40 fertile eggs laid between 19th January and 22nd March. Three of the remaining 12 eggs contained normal embryos and 9 died at an early stage of development. This laying period was one continuous cycle, with the good record of 40 eggs in 73 days. The hen went broody on 23rd March, and did not resume laying till 6th April. The eggs laid on 19th, 20th, and 22nd March all gave rise to chondrodystrophic embryos, but in not a single embryo out of 23 eggs incubated after the broody period was there any trace of this abnormal condition. This suggests that the two weeks' abstinence from laying permitted the righting of whatever condition had been abnormal during the long winter cycle.

#### *Evidence of a Genetic Basis for Chondrodystrophy.*

From the facts that the incidence of chondrodystrophy from different hens ranged from 1 up to 28, and that there was no consistent semblance of genetic ratios, evidence for the abnormality being due to the genetic constitution of the embryo is negative. However, the greater frequency of the abnormality in the progeny of certain birds of our flock than from others suggests strongly that the tendency to produce it is inherited. In addition to No. 92's 28 chondrodystrophics, another hen had 14, and still two others 10 each. In contrast to these, many hens under identical conditions in the same pen had no chondrodystrophics whatever in progenies of over 25 embryos examined. The records of the 21 hens in the chondrodystrophy-matings are given in Table III.

Since not all the eggs from each hen were incubated during April and May, we have not attempted to compare the frequency of chondrodystrophics from different mothers on a percentage basis. If the chondrodystrophic progenies of the same size were grouped, the distribution of these groups would not even approximate a Poissons series

TABLE III.—PRODUCE OF FEMALES IN CHONDRO MATINGS.

Hen.	Fertile Eggs Incubated.	Chondrodystrophic Embryos. No.
92	61	28
142	37	14
35	24	10
68	25	10
105	19	8
156	26	8
8	17	6
143	38	4
114	30	4
20	10	5
61	31	3
370	13	3
139	41	3
95	9	3
106	14	3
117	20	2
30	20	2
115	20	2
38	15	2
58	12	1
24	9	1

such as would be expected if the higher incidence of the abnormality in the progeny of certain females were due to chance. The chance of getting progenies of 28 and 14 in such a series would be infinitely small, and the progenies of only one chondrodystrophic would be much more frequent than were actually found.

Since the high incidence of chondrodystrophic embryos in the progenies of certain hens cannot be due to chance, and since only certain hens showed the tendency while others under identical environmental conditions produced only normal embryos, we are led to believe that the causal agency is an hereditary physiological abnormality in the dam which, under certain environmental conditions, results in the production of chondrodystrophy in the embryo.

#### *Breeds.*

The majority of the birds used for breeding at this Department are Brown Leghorns. Eighteen of the 22 hens which produced chondrodystrophic progenies were of this breed, the other four being White Wyandotte, Rhode Island Red, Frizzle, and Cross-bred. Our own data and that from the three outside sources give little indication that any one breed is more likely to produce chondrodystrophic embryos than another,

except that they were more frequent in the progeny from Light Sussex ♀ × R.I.R. ♂ at Poultry Farm B than in other breeds at the same place. Their occurrence, however, was not as frequent there as in the Brown Leghorns at this Department; and, since records of individual hens at B were not available to us, we are unable to say if their greater frequency in this cross was due to a general tendency of many hens toward chondrodystrophy or to one or two individuals with a marked tendency such as was exhibited by our ♀ No. 92. The latter is a White Wyandotte. Chondrodystrophic embryos were found in eggs from all the nine breeds and varieties represented by the material examined from the four sources, a list of which is given in the first paper of this series.

#### *Age of Dam.*

Of the 22 chondrodystrophy-producing mothers found at this Department, 1 was 3 years old, 3 were 2 years old, and the rest were pullets almost a year old at breeding. Since this represents fairly well the proportions of hens of these ages bred this season, it is doubtful if age of dam has much to do with the incidence of chondrodystrophy.

#### *Sex of Chondrodystrophic Embryos.*

Males predominated among the first chondrodystrophic embryos to appear at this Department. This was not in accord with Dunn's finding that of 51 chondrodystrophics sexed by him 26 were males and 25 were females, but it seemed explicable on the basis of the proportion of males being normally higher early in the season, as the work of Jull (1924) would indicate. We have determined the sex of 83 chondrodystrophics obtained from chondrodystrophy-producing matings at this Department before 1st April, the ratio being 50 males to 33 females. Of 91 normal embryos from the same hens and of the same period, 41 were males and 50 females. The sex-ratio for the whole 174 embryos was 109:64.\* The numbers expected on this basis in the four classes are given in Table IV.

At first glance it would seem that the number of males is significantly high in the chondrodystrophics and proportionately low in the normals. However, on application of the  $X^2$  test for goodness of fit to this distribution, it is found that the value of  $P$  is 0.107. This has been calculated by the method and table of Fisher (1928), taking  $n$  as 2. This means that one would expect as poor fit (or worse) of observed

\* Males per 100 females.

TABLE IV.—SEX OF CHONDRODYSTROPHIC AND NORMAL EMBRYOS AND EXPECTED PROPORTIONS IN EACH CLASS ON THE BASIS OF A SEX-RATIO OF 109:64 FOR THE ENTIRE POPULATION.

Class.	Observed.	Expected.
Chondrodystrophic ♂ . . . . .	50	43
Chondrodystrophic ♀ . . . . .	33	40
Normal ♂ . . . . .	41	48
Normal ♀ . . . . .	50	43
Total . . . . .	174	174

to expected ratios in about 11 per cent. of cases, and that the higher proportion of males observed in chondrodystrophic embryos is not significant.

*Relation of Chondrodystrophy to Mortality.*

From Dunn's observations and those made at this Department it may safely be assumed that chondrodystrophic embryos do not hatch but usually die before 19 days. Table I shows that this abnormality caused 0.80, 7.96, and 2.44 per cent. of the mortality among such dead-in-shell as could be classified at Poultry Farms A, B, and C respectively, covering the whole season. Allowing for doubtful cases discarded and for the many embryos of less than 8 days in which classification was not attempted, it is probable that these percentages would not be far wrong if applied to all the mortality at each place. However, based on positively identified chondrodystrophics only, the condition was responsible for 0.55 per cent. of the total mortality at all ages at A and 5.12 per cent. of the same at B. These figures cover the whole season. As stated previously, chondrodystrophy has a much greater effect on the earlier hatches.

The records of our own flock show that the abnormality may be more serious than at A, B, or C. Out of approximately 1900 eggs set from January to June, 122, or 6.4 per cent., produced chondros. Table II shows that the frequency of this abnormality from the group of chondro mothers ranged from 34.78 per cent. to 0.0 per cent. of the fertile eggs set according to season. The record of individual hens may be much worse, e.g. that of ♀ No. 92, who had only 3 normal embryos out of 40 fertile eggs set up to 22nd March.

There is some evidence from the birds mated here that chondrodystrophy may be associated with an unusually high number of deaths

before 7 or 8 days, *i.e.* apart from the recognised chondrodystrophics. In addition to No. 92's record, hen No. 156, from whom 31 fertile eggs were set up to 2nd April, produced 5 normal embryos, 8 chondrodystrophics, and the remaining 18 embryos died at various stages, practically all before 7 days. Other females producing this abnormality showed a similar tendency. Unfortunately, there are not adequate controls to establish this point.

It should be noted that the percentage frequency of chondrodystrophy varied considerably among eggs from the four sources from which they were obtained. To some extent this would be expected, even if the actual frequency of chondrodystrophics were the same in all stocks. Since the percentage hatch was lower in some cases than others (*vide* Table II in the preceding paper of this series), one would expect a greater number of normal embryos among the dead-in-shell from such hatches, and consequently a lower percentage of abnormal. Possibly this accounts to a slight extent for the low percentage of abnormal from Poultry Farm A. However, at Farms B, C, and at this Department, where the percentage hatches were much the same, the frequency of chondrodystrophics varied much more than did that of teratological abnormalities and malpositions. It must not be inferred, therefore, that chondrodystrophy is responsible for mortality to the same extent in all flocks.

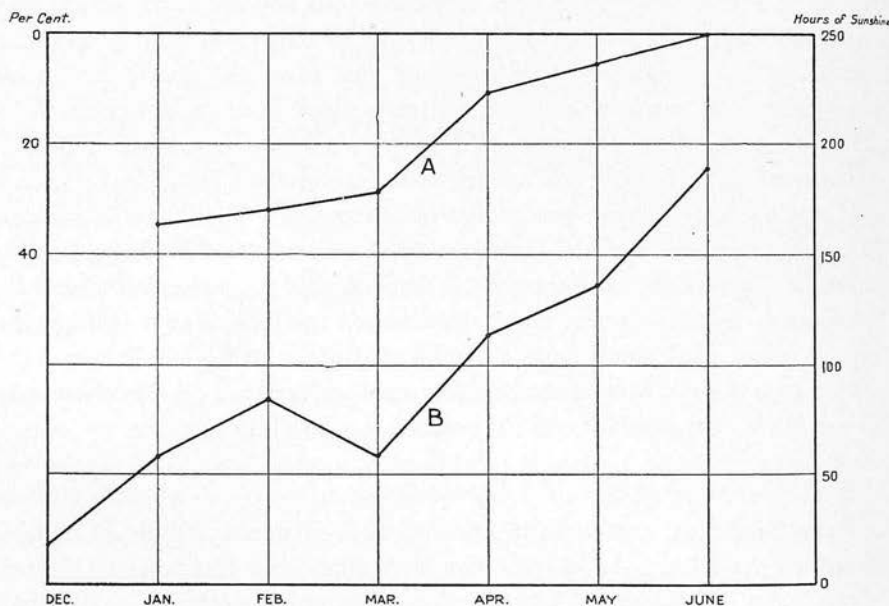
#### DISCUSSION.

The fact that, so far as we are aware, chondrodystrophy has been found in America only at the Storrs Agricultural Experiment Station, Connecticut, suggests that it may be less common there than here, where it was obtained in all four sources from which eggs were received. Additional evidence for this is found in Dunn's report that the abnormality constituted about 1.5 per cent. of 4644 embryos dead after the 7th day examined at Storrs, whereas, in three out of four flocks here, the frequency was much higher.

In attempting to determine factors concerned in the etiology of chondrodystrophy this difference must be considered. The marked decline in its frequency from January to June in our own chondro matings, together with a similar decline in eggs from Poultry Farms A, B, and C over a shorter period, is still more important. Both together suggest that some difference in weather is a factor. To find that factor, one must look for something which is aggravated in this locality in January, February, and March, but becomes ameliorated from March on, and which is more acute in this climate than in the United States. Lack of

sunshine is at once suggested. Edinburgh is at  $55^{\circ} 36'$  North Latitude, and is therefore subject to very short days in winter. This condition is often aggravated in winter by mist and cloudy skies, so that the lack of sunshine is quite evident to any visitor.

Examination of the meteorological data at the Royal Observatory, Edinburgh, led to the conclusion that sunlight was of more importance than temperature, humidity, barometric pressure, or any other determiner of climate. Text-figure 1 shows in graphic form the total hours of sun-



TEXT-FIG. 1.—Graphs showing (A) the monthly frequency, in percentage, of chondrodystrophic embryos from the 21 hens in the chondrodystrophy-producing matings at the Animal Breeding Research Department from January to June 1928. (B) The total hours of sunshine per month at Edinburgh, from December 1927 to June 1928.

shine as recorded at the Observatory, half a mile from the poultry-yards of this Department, from December 1927 to June 1928. The decrease in the frequency of chondrodystrophic embryos at this Department, also recorded, parallels the increase in sunshine closely.

In support of this hypothesis, the average hours of sunshine per month from December to June, over a period of 28 years at Edinburgh, is compared in Table V with the same data for a period of 45 years at New York. The latter data are taken from the records of the New York Meteorological Observatory, kindly furnished by the Royal Observatory, Edinburgh.

TABLE V.—AVERAGE HOURS OF SUNSHINE PER MONTH AT EDINBURGH AND NEW YORK.

Month.	Edinburgh, 28 years.	New York, 45 years.
December . . .	41.0	141.0
January . . .	44.8	140.7
February . . .	70.7	159.2
March . . .	106.1	205.7
April . . .	142.2	229.3
May . . .	162.1	270.5
June . . .	192.4	292.2

Admittedly the hours of sunshine at New York do not hold for all other parts of the United States, but they should apply reasonably closely for nearby Connecticut. The difference between Edinburgh and New York sunshine is so marked as to suggest at once why chondrodystrophy has been of such rare occurrence at the Storrs Connecticut Station and in the United States generally, compared with the A.B.R.D. flock here and other flocks nearby.

Chondrodystrophy must not be confused with leg weakness or avian rickets, which is caused by a lack of Vitamin D or of adequate minerals or both, and cured by direct sunshine, ultra-violet light, or cod-liver oil plus proper minerals. At this Department the flock received a grain ration, consisting of maize, wheat, and oats, and a mash mixture of wheat middlings, bran, ground oats, soy-bean meal, alfalfa meal, fish meal, bone meal, and salt. Oyster-shell was supplied in hoppers, and crude cod-liver oil was fed in the mash till well on in April. Cabbage was given occasionally. All birds were out of doors daily. There was no evidence of leg weakness in the laying stock, and no unusual number of soft- or thin-shelled eggs. No. 92's eggs were all of particularly firm, sound shell. The rations of Poultry Farms A and B had no deficiency of any of the vitamins or minerals recognised as essential. Both were feeding cod-liver oil. Farm C's rations are not known.

Hart, Steenbock, *et al.* (1925) have shown that the hatchability of eggs from hens deprived of sunshine for several weeks declines to a low figure, but can be quickly restored by irradiation with ultra-violet light and much more slowly restored by feeding cod-liver oil. The deficiency of ultra-violet light in this locality during the fall and winter months is even more acute than is indicated by the small number of hours of sunshine. The relative humidity here averaged 85 per cent. for December, January, February, and March. Under such conditions the amount of

ultra-violet light actually reaching the fowls would be considerably less than in a drier atmosphere. It seems possible that under such acute shortage of ultra-violet light, cod-liver oil may supply enough Vitamin D to permit normal calcium assimilation, but cannot make up for *other beneficial effects* of the radiant energy. The same reason may explain its slower curative action in the experiments just cited. That the value of sunshine is not confined to its catalytic rôle in calcium and phosphorus metabolism is evidenced by its bactericidal powers, its value in the treatment of tuberculosis, and its ability to increase the resistance of the body to toxins and to invading micro-organisms.

Evidence of a genetic basis for chondrodystrophy has already been given, and the belief expressed that the tendency to produce the abnormality is inherited. Such a statement is incompatible with Dunn's (1927) finding that there was no greater incidence of this abnormality among his inbred chondrodystrophy series than among controls. However, it is evident that, whether or not the hereditary tendency be present in the same degree in all affected individuals, it is not manifested in the same degree in their offspring, and in any case is manifested only under unfavourable conditions. Many of our fowls produced only 1 or 2 chondrodystrophic embryos; none of Dunn's produced more than 6. If the abnormality is most frequently expressed under sunshine starvation, it is quite conceivable that most of the individuals in the flock at Storrs were safely past the border-line of such starvation by the middle of February, when incubation was begun, and that the tendency to produce chondrodystrophic embryos was not manifested to the same extent as it might have been under less favourable conditions. Table V indicates that the average sunshine is almost as great at New York in February as it is here in May, at which latter season chondrodystrophy had decreased in our chondrodystrophy-matings to one-sixth of its earlier intensity.

What rôle sunlight may play in the etiology of chondrodystrophy the writers cannot state. Since two embryos exhibiting the abnormality were obtained in eggs from Poultry Farm B, set in July, it is doubtful if lack of sunshine is the sole cause. The provisional hypothesis is suggested that extreme lack of sunshine superimposed upon an inherited susceptibility results in the production of eggs deficient or incorrectly balanced with respect to some essential constituent, and that such eggs give rise to chondrodystrophic embryos.

The writers are indebted to the Romanno Bridge Poultry Farm, West Linton; W. Johnston & Co., Comrie; and the Sussex Hatcheries,

Glasgow, for eggs supplied for this investigation. Thanks are also due to Professor Crew for his interest and constructive criticism; to the Royal Observatory, Edinburgh, for meteorological data; and to Mr J. M. A. Chisholm for the photographs.

#### SUMMARY.

1. Among 7135 chick embryos of nine days or over, obtained from unhatched eggs from three poultry farms, 112 cases of chondrodystrophy were observed. From 1900 eggs set at this Department, 124 chondrodystrophic embryos were obtained.

2. The mortality caused by this condition varied in different flocks from 0.80 to 7.96 per cent. of the embryos dying after the eighth day; at this Department it was responsible for loss of 6.5 per cent. of the eggs set.

3. The incidence of chondrodystrophic embryos was found to be greatest in January and February, and to decline steadily thereafter to an almost complete absence in June.

4. The occurrence of chondrodystrophy appears to be independent of breed of fowl, sex of embryo, and age of dam.

5. The number of these abnormal embryos produced by individual hens ranged from 1 to 28.

6. Evidence is advanced in support of the theory that the causal agency is an hereditary physiological abnormality in the dam which, under certain unfavourable conditions, results in the production of chondrodystrophy in the embryo.

7. It is shown that the incidence of chondrodystrophy appears to be inversely proportional to the amount of sunshine, and the suggestion is made that lack of direct sunlight is a factor in the etiology of the abnormality.

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EXPLANATION OF PLATE.

(All photographs are natural size.)

Fig. 1. Three eleven-day embryos from eggs set on the same day and in the same incubator. The one on the right is normal; the other two are from ♀ No. 92, and are typically chondrodystrophic. In addition to the vaulted skull, parrot beak, and shortened bent legs of the two abnormal embryos, a difference in the rate of growth of the down is already apparent.

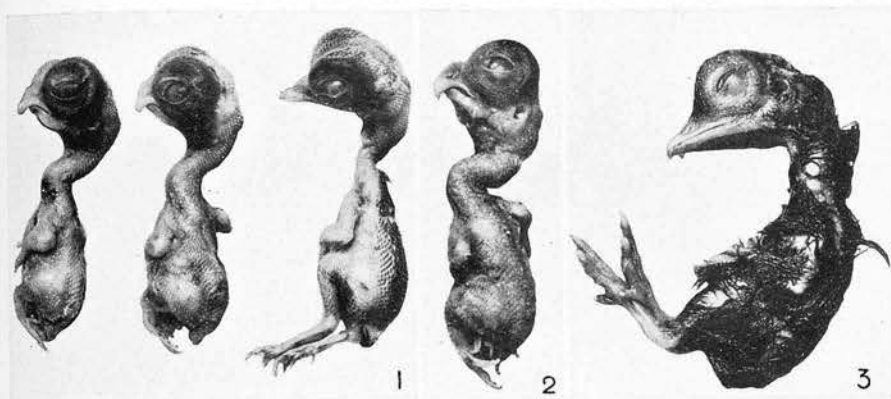
Fig. 2. A thirteen-day chondrodystrophic embryo from ♀ No. 92. The retarded growth of down is characteristic of acute chondrodystrophy.

Fig. 3. A thirteen-day normal embryo from the same setting as fig. 2. The difference in length of the legs and in the direction of the plantar surfaces of the feet in fig. 2 and fig. 3 is well marked.

Fig. 4. A specimen, estimated about 18 days, exhibiting extreme chondrodystrophy complicated with ectopia. The sharp angle of the bend in the tibia is evident; a similar bend is also present in the much reduced tarso-metatarsus.

Fig. 5. Extreme chondrodystrophy in an embryo, estimated at 20 days. The yolk-sac has been drawn into the body cavity, and gives the embryo the appearance of a round ball, from which the feet project. The curly appearance of the down is characteristic of extreme chondrodystrophy.

Fig. 6. A less extreme case of chondrodystrophy in a twenty-one-day chick. The slight protrusion of the upper beak might have passed unnoticed, but the backward turning of the feet suggested the typical bending of the tibia revealed when the muscles covering that bone were removed.



XII.—Studies in Embryonic Mortality in the Fowl. III. Chick Monsters in Relation to Embryonic Mortality. By F. B. Hutt, B.S.A., and A. W. Greenwood, Ph.D., Animal Breeding Research Department, University of Edinburgh. (With Three Half-tone Plates.)

(MS. received October 5, 1928. Read January 7, 1929.)

DURING the course of an examination of some 12,000 eggs which had failed to hatch, abnormal embryos of three distinct classes were encountered. Abnormalities of position and chondrodystrophy have been discussed in previous communications of this series (Hutt 1928, Hutt and Greenwood 1928).

The present paper deals with various types of monsters already well described in the literature of teratology. Their description in this paper will therefore be no greater than is necessary to establish their identity.

Full details of the material examined, which came from three large commercial poultry farms and from this Department, have been given in the first paper of the series.

#### TYPES OF MONSTERS ENCOUNTERED.

1. *Hyperencephaly*.—The characteristic features of this monstrosity include a complete absence of the roof of the cranium, marked reduction or absence of the upper beak, and almost invariably absence of both eyes. There is usually extreme shortening of the neck, and ectopia is conspicuous in over 75 per cent. of the specimens (figs. 1, 2, and 3).

2. *Exencephaly*.—Included in this class are various types of meningocele in which a portion of the brain extrudes through the cranium but is still confined by the meninges. It is usually due either to an incomplete ossification of the brain case or to portions of the brain being pushed up so that the bones cannot meet in the normal suture lines. Such monsters are easily recognised by the protrusion from the head of a bare mass of tissue free from down (figs. 4, 6, and 7). In some cases this is in the occipital region (notencephaly), where it consists of an extrusion of the optic lobes; but in our material it occurred more commonly as an extrusion of part of the cerebral hemispheres in the anterior part of the skull (proencephaly). The terminology used is that of Dareste (1891), who classified hyperencephaly as a subdivision of the exencephaly group.

3. *Microphthalmia*.—The characteristic feature of this group is the failure of one or both eyes to develop normally (fig. 5). Dissection of several of these specimens showed that a small pigmented optic cup is present in many cases which appear to have no eye whatever. It is evident that anophthalmia without other complication is merely an advanced degree of microphthalmia, and therefore these two conditions are classed together in this report. The defect may be bilateral, but more commonly only one side is affected. Unilateral microphthalmia is accompanied by shortening of the face on the affected side, with the result that the upper beak is twisted to the short side (fig. 5).

Embryos were found with both microphthalmia and exencephaly (figs. 13, 14, and 15). In some of these there was also a reduction of the fronto-nasal process, suggesting that the condition was allied to a low grade of hyperencephaly. Ninety-three per cent. of all monsters observed in this survey were of one or another of the three types described above. Other miscellaneous abnormalities occurring in small numbers included the following:—

*Ectopia*—Eversion of the viscera.

*Prognathia*—Protrusion of the upper or lower beak beyond its fellow.

*Cyclopia*—A condition in which there is a single median eye. The cerebral hemispheres are absent, the mandible and tongue reduced, and the upper beak represented by a small proboscis-like structure above the eye (figs. 8 and 9).

*Duplicity*—Duplication of parts ranging in degree in this material from two upper beaks to two separate heads or duplication of the whole body except the head (figs. 10 to 12 and 16 to 20).

*Otocephaly*—Characterised by various degrees of approach of the ears on the ventral surface of the head, and different grades of reduction of the brain, extending down to entire absence of the head.

*Malformed Limbs*—Including thickened and flattened tarso-metatarsus, unilateral absence of leg muscles, and absence of one or more toes. Minor cases of twisted feet or toes were not included.

*Absence of Premaxillæ*—One embryo was abnormal only in the absence of these bones; another lacked also the nasals.

#### FREQUENCY.

The number of these various monsters observed among 11,797 unhatched eggs examined are shown in Table I.

## Chick Monsters in Relation to Embryonic Mortality. 147

TABLE I.—FREQUENCY OF TERATOLOGICAL ABNORMALITIES.

Source.	Eggs.	Hyperen- cephaly.	Exen- cephaly.	Microph- thalmia.	Both Micro. and Exenceph.	Miscel- laneous.	Total.
A	9834	161 1·74%	80 ·81%	72 ·73%	22 ·22%	22 ·22%	357 3·63%
B	1015	16 1·57%	13 1·29%	11 1·08%	4 ·39%	3 ·29%	47 4·63%
C	654	1 ·15%	5 ·77%	7 1·07%	3 ·46%	2 ·30%	18 2·75%
D	294	1 ·34%	2 ·68%	6 2·04%	0 0%	2 ·68%	11 3·74%
Total	11797	179	100	96	29	29	433
Per cent.		1·52	·84	·81	·25	·25	3·67

These defects can be recognised in chick embryos of 6 days or even less without microscopic examination, and were quite common in the dead germs removed at 8 or 9 days. For this reason the percentages given in Table I are calculated for the total numbers of eggs examined, including both dead-in-shell at hatching and those dying before the first test (usually made at 8 days). Many of the latter were too small or too decomposed to permit recognition of teratological abnormalities; but, since in our material these were found dead at all stages from 20 days right down to the limit of recognition, it is probable that the same types occurred beyond that limit. Dareste, who studied chiefly the earlier stages of chick embryos, found other types of abnormalities (*e.g.* omphalocephaly and arrested development of blood islands) which invariably caused death in the first week of incubation. It is practically certain, therefore, that the total loss from teratological monsters is much greater than is indicated by the percentage given above. Indeed, Stockard (1921), in his exhaustive studies of abnormalities induced experimentally in *Fundulus* (minnow), found that "there may be such minor defects as would escape observation until the hatched embryos were found to be unable to right themselves and swim." Possibly a similar condition is responsible in part for some of the chick embryos found fully formed dead in the shell, apparently normal but unable to hatch, and generally classed as "too weak to hatch."

Out of 96 specimens of microphthalmia encountered in this study, the defect was bilateral in 13 and unilateral in 83. Data for 55 specimens

of unilateral microphthalmia show that the left eye was affected in 31 cases and the right one in 24.

Ectopia, or extrusion of the viscera, was not recognised as being a distinct type of abnormality till well on in the investigation. Its frequency, therefore, is probably greater than the six specimens recorded would indicate.

Dareste states that otocephaly is a very rare occurrence in the chick. Only one specimen in our material, a chick with practically no head, fitted into his definition of this class, although, if Wright's (1923) grades of otocephaly were followed, the two cases of cyclopia observed would also be included.

The remainder of the 29 miscellaneous monsters included 8 cases of prognathia, 8 specimens exhibiting duplicity, 3 with malformed limbs, and 2 with prenexillæ or nasal bones absent.

#### SEASONAL FLUCTUATIONS.

The monthly distribution of teratological monsters in the eggs from Poultry Farms A, B, and C is given in Table II. The total number of monsters from each source is given in brackets, and their monthly frequency is expressed as a percentage of the number of unhatched eggs examined. The month refers to the dates of setting, not of hatching.

TABLE II.—SEASONAL DISTRIBUTION OF MONSTERS.

Month.	Farm A (357).	Farm B (47).	Farm C (18).
	Monsters.	Monsters.	Monsters.
February . . . . .	4.77	5.16	...
March . . . . .	3.51	5.21	2.80
April . . . . .	1.65	2.40	2.61

A tendency toward a decrease in teratological abnormalities as the season advanced is evident in all stocks. The material from Farms B and C is rather small, and permits of greater variation and error; but the numbers from Farm A are large enough to indicate a steady decline from February to April, which may be considered reasonably significant.

## SEX.

Wright found that otocephaly was much more common in females than males in his inbred guinea-pigs, the ratio being 55 ♀:26 ♂, or 32.1 per cent. males. We have records of sex of 147 monstrosities, representing the types most common in our material, but are unable to find any significant deviation from the normal sex-ratio in any class. The data are given in Table III.

TABLE III.—SEX OF MONSTERS.

Class.	♂	♀	Sex-Ratio.*
Hyperencephaly . . . .	30	34	88.23
Exencephaly . . . . .	22	17	129.41
Microphthalmia . . . . .	21	17	123.53
Micro. + Exenceph. . . . .	3	3	100.00
All classes . . . . .	76	71	107.04

\* Expressed as the number of males per 100 females.

## MORTALITY.

Chicks exhibiting a low grade of microphthalmia recognisable by their twisted beaks occasionally hatch, but perish sooner or later. The comparatively large numbers found dead in this investigation lead to the belief that the majority affected with this abnormality die in the shell. Similarly, exencephalic chicks may very rarely hatch, but their death soon follows. We know of no case in which a hyperencephalic chick has hatched. Table IV shows the proportions of the different monstrosities found dead in the first, second, and third weeks of incubation.

TABLE IV.—PERIOD OF MORTALITY OF TERATOLOGICAL ABNORMALITIES.

	First Week, per cent.	Second Week, per cent.	Third Week, per cent.
Hyperencephaly . . . . .	28.57	32.97	38.46
Microphthalmia . . . . .	7.59	13.92	78.48
Exencephaly . . . . .	...	12.50	87.50
Micro. + Exenceph. . . . .	...	9.52	90.48

The majority of the last three classes in this table were embryos of 18 to 20 days. In many cases it was possible to estimate the age of

hyperencephalic embryos only by the size of the posterior limbs. Very few of those that survived to the third week were fully formed. Apart from the occasional exceptions listed above, there is every reason to concur in Dareste's statement that "les monstres simples autosites produit chez les oiseaux périssent fatalement dans la coquille, avant l'éclosion."

#### DISCUSSION.

The older teratologists expressed the belief that monsters were caused by an arrest in development which in some cases was believed to be brought about by pressure of the amnion or injury to that organ. In recent years it has been shown that the same abnormalities can be produced experimentally in fish embryos (Stockard 1921 and others) in which no amnion is present, as in the Amniota, and hence that the amnion is not responsible to the extent hitherto believed. The same abnormalities can be produced by different means, chief of which are chilling, lack of oxygen, exposure to fumes of alcohol, ether or other gases, and the use of such chemical substances as magnesium and butyric acid. An excellent review of such experimental teratology in the chick has recently been given by Hyman (1927). The common feature of all these methods is that they cause an arrest in the development of the embryo. Fairly conclusive proof of this is given by Stockard's demonstration that by chilling the embryo of *Fundulus* at a critical period he could produce every known type of monster, such types being dependent upon the period of interference with growth.

Whether or not all monsters occurring in the absence of experimentation are due to this same cause is another question; but the discontinuous mode of development of the chick embryo suggests that an arrest of growth may well be the cause of abnormalities in that species. Stockard found that, prior to the formation of the gastrula, the embryo was particularly susceptible to an arrest of development. The fowl's egg is ready to be laid at 21 to 27 hours after ovulation. The variation in the time required to traverse the oviduct results in corresponding differences in development at the time of reaching the vagina. This variation is made still greater by the fact that some eggs may be ready for laying at the end of the day but be retained by the hen till the following morning. Others may reach the end of the oviduct in the morning and be laid at 21 or 22 hours after ovulation. It has been shown by Patterson (1909) and others that gastrulation takes place very close to the time of laying, and it is therefore quite conceivable that an egg laid only 21 or 22 hours after ovulation may be just at the commence-

ment of gastrulation. Such an egg would be very susceptible to the chilling to which it is subjected after laying. The majority have completed gastrulation at laying, and are therefore better able to withstand the cessation or retardation of growth.

Another possibility must be considered. Alsop (1919) has shown that high or low incubation temperatures during the first three days will produce various types of defects in the eye and brain. However, since the abnormalities produced experimentally were dissimilar to those found in her controls it is questionable whether faulty temperature is responsible for the monsters found in our material. If there were a great number in any one hatch this would be suspected; but, since the actual frequency is very low, sometimes only one in the eggs remaining from an 85 per cent. hatch, it seems more probable that incubator conditions are not responsible in the majority of cases. There may be a differential susceptibility to slightly high or low temperature, but this again would depend largely on the degree of development when the egg went into the incubator.

The actual *modus operandi* of the arrest is best explained on the assumption that there is a critical period for the development of each organ of the body. At this time the anlage of that organ develops at a greater rate than other parts of the embryo. An arrest in development at such a stage slows or stops growth, so that all parts of the embryo are equal. When cell division is resumed, the particular region previously accelerated is not again able to assume its differential rate, and is therefore suppressed or poorly developed. The rest of the body may develop normally, but the affected anlage can never recover from its reverse and fulfil its original destiny.

In most experimental teratology with the chick, embryos are not incubated and observed beyond the first week, and it is therefore doubtful to what extent the abnormalities induced artificially correspond to those commonly found in our material which was incubated under supposedly optimum conditions. Our data afford one interesting case of agreement under natural conditions with results obtained experimentally. Stockard (1921) found that the period at which an arrest of development could affect the eyes of *Fundulus* was close to the critical period for the primary brain ventricles, and that as a result combinations of defects in these regions were common in groups of experimental embryos subjected to the same treatment. In our material the frequency of eye defects was 0.81 per cent. and of exencephaly 0.84 per cent. If these were induced by different causes one would expect them to be associated

by chance in only 0.0068 per cent. of cases, or less than once in the 11,797 eggs examined. Actually 29 embryos were found with both microphthalmia and exencephaly, a frequency of 0.25 per cent., which is over 33 times the expectation. This indicates that in the chick embryo, defects of eye and brain arising under optimum conditions tend to be associated, and that presumably the critical periods of development for each are close together. The chief difference between Stockard's findings in the minnow and these in the fowl is that in the former case a continuous development was arrested by experimental procedure, while in the latter a naturally discontinuous development was subjected under natural conditions to an arrest equally effective because operative at a critical period.

Injuries have been induced simultaneously in brain and eye regions of the chick by Hyman (1927) and Hinrichs (1927), but, since these were done with chemicals and ultra-violet light at various stages of incubation, they are not comparable with those abnormalities occurring naturally in the chick or induced by a chilling in the minnow. The work of these two investigators, however, shows that there is an antero-posterior gradient of susceptibility to injury in the chick, and that the greater frequency of head abnormalities than all other types is probably due to a higher level of metabolism in the anterior end of the embryo. Such an interpretation is in accord with Child's axial gradient hypothesis.

Monsters produced in the departmental stock were few in number, and no evidence was found of an hereditary tendency underlying their production. Wright (1923) found that otocephaly (similar in some respects to monsters described in this paper) increased to a marked extent in certain inbred families of guinea-pigs. In one branch the frequency of this abnormality was 20 per cent. in the nineteenth generation, compared with 1.5 per cent. for the family and 0.2 per cent. for the entire population. While no genetic ratios were demonstrable, the increase of abnormalities thus brought about by inbreeding could only be interpreted as the intensification of genetic factors rendering the individual carrying them susceptible to any unfavourable environmental condition predisposing toward the abnormality.

It is probable that a non-lethal grade of exencephaly has become incorporated as a breed characteristic in such breeds as the Polish, Crèveœur, and Houdan, in which cerebral hernia is typical. Dunn (1927) believes that this character is entirely distinct from exencephaly. Dareste (1891), on the other hand, after making a study of young chicks of these breeds, concluded that the condition was quite comparable to

proencephaly, differing only in that the cerebral hemispheres were upthrust entirely instead of only in part. This resulted in spreading apart the frontal bones (which were normally developed), and the overlying membrane gradually became ossified to form the large brain case typical of such fowls. Tegetmeier's (1867) study of Polish skulls supports Dareste's findings. Dareste concluded that these races were produced by the spontaneous appearance and hereditary transmission of an exencephaly of a grade or kind that was not fatal to its exhibitor. If this be so, then the tendency to produce similar abnormalities, lethal in effect, may also be hereditary.

#### SUMMARY.

In an examination of 11,797 eggs which had failed to hatch, 433 specimens exhibiting teratological abnormalities were obtained.

The various types of monsters observed are briefly described and the frequency of each given.

The most common types were characterised by various degrees of abnormality either in the brain, cranium, or eyes, or in two of these structures, or in all three. Hyperencephaly, exencephaly, and microphthalmia constituted 93 per cent. of all the monsters observed.

Other teratological abnormalities encountered included prognathia, duplicity, ectopia, cycloopia, otocephaly, malformed limbs, and absence of premaxillæ.

Monsters accounted for at least 3.6 per cent. of all mortality in the material examined. Since this material included many embryos too decomposed for detection of teratological abnormality, the actual loss is probably considerably higher.

No significant deviations from the normal sex-ratio were evident in any class of monsters.

A decline in the frequency of such abnormalities from February to April was evident.

Evidence is adduced from Stockard's experimental teratology in fish eggs, to show that a very probable cause of the production of monsters in the chick is an arrest of development of the embryo at a critical stage. Presumably such a condition results from the chilling of those eggs laid in the early stages of gastrulation.

Some evidence to indicate the possibility of an hereditary tendency to produce monsters is briefly discussed.

## ACKNOWLEDGMENTS.

The writers are greatly indebted to the Romanno Bridge Poultry Farm, West Linton; W. Johnston & Co., Comrie; and the Sussex Hatcheries, Glasgow, for supplying the majority of the eggs examined. Thanks are also due to Professor F. A. E. Crew for suggestions and advice, and to Mr J. M. A. Chisholm, of this Department, for all the photographs.

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## EXPLANATION OF PLATES.

## PLATE I.

Fig. 1. Hyperencephaly in an embryo of about 8 days.  $\times 3$ .

Fig. 2. A hyperencephalic embryo of about 16 days enclosed in the amnion. Natural size.

Fig. 3. A hyperencephalic embryo of about 18 days, with the membranes and yolk-sac removed, showing the characteristic absence of upper beak, eyes, and brain covering. Ectopia is conspicuous. Natural size.



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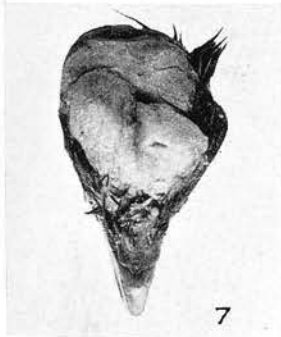
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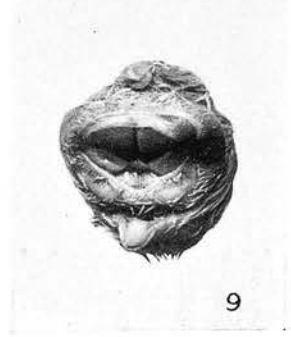
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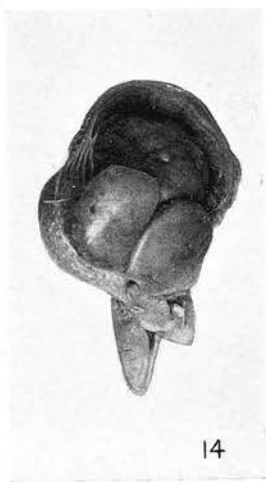
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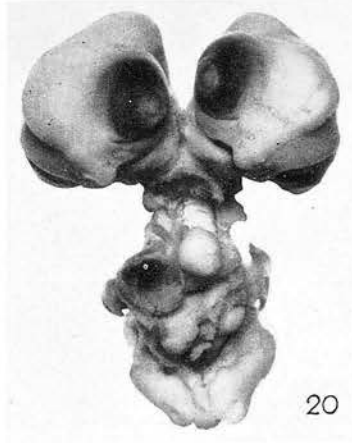
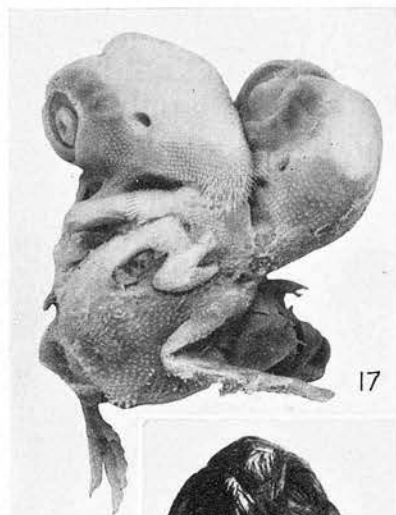
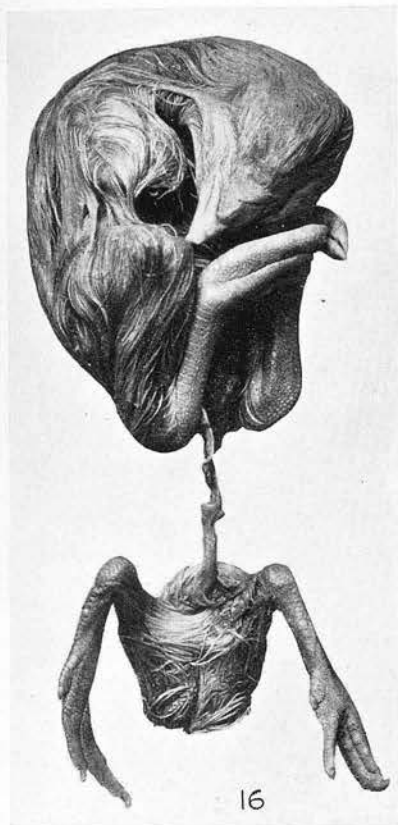


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Figs. 4 and 6. Exencephaly. Nineteen-day embryos exhibiting anterior encephalomeningoceles (proencephaly) of different sizes.  $\times 1.4$ .

Fig. 5. Unilateral microphthalmia in a twenty-day chick. The down has been plucked from around the ear and eye. The black dot is made by the reduced eyelids. The twisted upper beak is characteristic.  $\times 1.4$ .

Fig. 7. Exencephaly extended to include both the occipital region and that of the cerebral hemispheres.  $\times 1.4$ .

Fig. 8. Anterior aspect of the head of a cyclopean embryo.  $\times 1.4$ .

Fig. 9. Anterior aspect of the head of an embryo in which cyclopa is not complete. The two lenses have come together in the median line, but are not completely fused. The reduced lower beak, the tongue, and the characteristic proboscis above the eye are conspicuous. The absence of cerebral hemispheres is less so.  $\times 1.4$ .

PLATE II.

Fig. 10. Partial duplicity and exencephaly in the head of a sixteen-day chick. The two upper beaks cross the lower ones, which are seen in part from the side. Two combs are evident, but duplication of the rest of the head, though present, is less obvious.  $\times 1.4$ .

Fig. 11. One lower beak, but two upper ones and two combs. The dark spot between the two combs is a small opening from the median eye, resulting from fusion of the inner eyes of the parts which almost became two heads.  $\times 1.4$ .

Fig. 12. Duplicity more advanced than in figs. 10 and 11. The opening of the median eye is distinct.  $\times 1.4$ .

Figs. 13, 14, and 15. Various degrees of exencephaly combined with unilateral microphthalmia.

PLATE III.

Fig. 16. A fully formed embryo to which were attached by a strand of tissue the hind limbs only of a second embryo. Natural size.

Fig. 17. Cranial dichotomy in a ten-day embryo.  $\times 2$ .

Fig. 18. A chick of about 16 days exhibiting duplicity extending anteriorly as far as the neck. Down has been plucked to show the four wings. Natural size.

Fig. 19. Caudal dichotomy in a microphthalmic chick of about 9 days.  $\times 2$ .

Fig. 20. Cranial dichotomy in a chick of about 7 days.  $\times 3$ .

## Potentially Fatal Fatigue of Cervical Muscles of the Fowl Resulting from an Excessively Large Comb.

By F. B. HUTT,

*Animal Breeding Research Department, University of Edinburgh.*

THERE have recently occurred at this department two cases in which a cock was reduced to the verge of death as a result of an excessively large comb, and was restored to perfect health by the single expedient of cutting off that structure. These cases are unique in the experience of the writer, and are considered worthy of record as illustrating a condition that would seem to be a potential cause of death in the fowl.

The first bird observed was a Single Comb Brown Leghorn cockerel, No. 21 on the records of this department, hatched in May, 1927, and operated upon at six weeks of age, when the left testis and half of the right were removed. The cockerel developed normally, and at eight months was indistinguishable in appearance or behaviour from normal cocks of the same age, except that it had a rather large comb.

On January 1st, 1928, he was put in a small house with six other cockerels of the same breed and about the same age. These seven birds were used for individual matings with selected hens, the object being to determine how No. 21 and other partially castrated males compared in fertility with normal cockerels. No. 21 was the largest of the seven and was recognised by the others as ruler of the roost. By this time his comb had lopped over on one side.

On the morning of February 11th this bird was noticed standing motionless in the pen with his head bent down almost to the floor. The only signs of impaired health previously observed had been a slightly reduced activity on the preceding two days. There was no evidence of fighting or of anything else that might have injured the bird. When placed by himself in a good-sized cage his head, at first erect, slowly but inevitably sagged till his beak rested on the floor. When roused he raised the head slightly, but almost at once it began to sink gradually back to the floor. It seemed as though his head were far too heavy for him to stand erect for any length of time.

Since the bird took no interest whatsoever in food, he was forcibly



fed twice daily with pellets of moist mash. When unassisted, he made no attempt to drink. When his beak was put in water, the patient would gulp several times, but could swallow only when someone lifted his head.

On February 12th, the day after he was first noticed, all power of the flexor muscles seemed entirely gone, and when the head was lifted it dropped straight down again to the floor (Fig. 1). The extensor muscles were able to exert pressure, and did so when the head was lifted too high or pushed back toward the body. The symptoms were not in the least suggestive of botulism or of any injury to the auditory labyrinths, either of which conditions would be accompanied

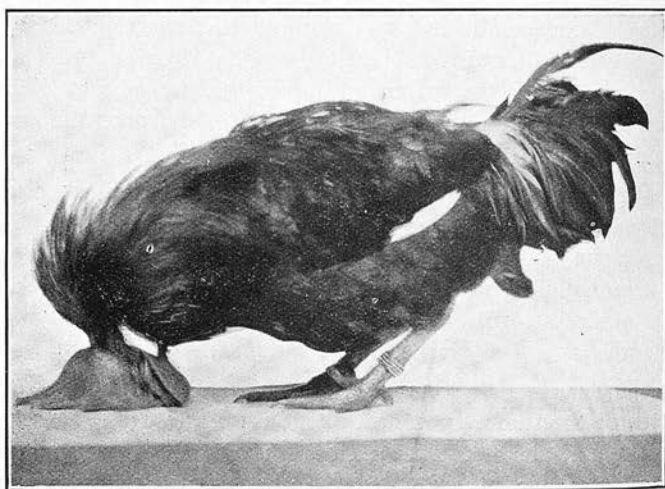


Fig. 1.

by a twisting of the head to one side. Both eyes were bright and showed the usual reflex winking.

On February 14th, No. 21 was distinctly weaker. Though it was recognised that a pathological disturbance in the central nervous system might be one cause of the condition exhibited, it was decided to adopt the view that the condition was one in which the comb had increased in size by virtue of its own developmental impulses to an extent that overtaxed the power of the muscles of the neck to support it. This view could be put to the test of experiment. If the idea were correct, a reduction in the weight of the comb should be followed by an amelioration of the condition. Accordingly, after measurements had been taken, the comb was sliced off with a razor about one half-

inch from the head. In spite of its vascular appearance, the comb of the cock is composed almost entirely of connective tissue. In the present case a few drops of adrenaline stopped bleeding so effectively that not a drop of blood fell from the comb.

Measurements taken prior to its removal showed that No. 21's comb was 14.6 cm. in its longest dimension, 8 cm. high and 2.6 cm. through at its thickest point. The portion cut off weighed 58.7 gm. The body weight was 2,041 gm. on February 7th and only 1,814 gm. on February 11th.

When the bird was placed on the floor he staggered drunkenly

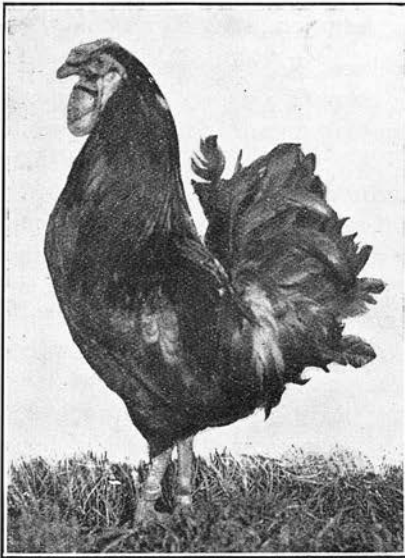


Fig. 2.

right across the room, holding his head about 8 or 9 in. above the floor. When he ceased walking the head again sagged down, but when roused the bird was able to hold his head off the floor for over a minute. On being offered grain and water, the cockerel drank thirstily and ate voluntarily for the first time in three days.

The following morning No. 21 was found in his old position with the head on the floor, but a marked improvement was noticeable in his general demeanour when aroused. Thereafter a gradual daily improvement was noticeable, and on February 19th, five days after removal of the comb, the bird was returned to an outdoor yard, where he celebrated his recovery by lusty crowing (Fig. 2). Matings

were resumed, and the fertility of his hens was soon as good as before he became incapacitated, and equal to that of normal cocks.

The history of the other case, also a Single Comb Brown Leghorn cockerel, was so like that given above that detailed description is unnecessary. In this case 19.9 gm. of wattles and 37.85 gm. of comb were removed on March 13th. Only the posterior half of the comb was removed, and the remainder seemed to weigh downward on the anterior part of the head, causing the beak to be tucked in close to the neck. Recovery was not complete till most of the remaining comb (18.2 gm.) was cut off on April 3rd. Altogether 75.95 gm. of comb and wattles were removed from this cockerel, whose body weight was 1,927 gm. prior to the operation.

This second cockerel, No. 25 of our records, had been completely castrated at nine days of age, and the removed testes implanted subcutaneously one under each wing. It is possible that the interference with the testes may have been in part responsible for an excessive growth of the comb. Greenwood and Crew<sup>(1)</sup> and Blyth<sup>(2)</sup> have put forth evidence that the development of the comb is dependent to a considerable extent upon the degree of spermatogenic activity of the testes. No. 21 was found on *post-mortem* examination five months later to have only 5.77 gm. of testis on the left side and a regenerated nodule of .05 gm. on the right side, but not connected with the vas deferens. It is possible that in the small fragments of testis material retained by this bird the process of spermatogenesis was proceeding during the breeding season at a much faster rate than in normal cocks and that this condition was responsible for excessive comb growth as the hypothesis of the writers mentioned would suggest. No evidence of any gross abnormality of the central nervous system was encountered. No. 25 is still living, and therefore can contribute little to this discussion.

However, several other cockerels similarly operated upon had no trouble with their combs and therefore, in the opinion of the writer, the large combs of these two cases described represent less an abnormal condition resulting from the operations to which the birds were submitted than the natural result of breeding for a large comb. In Great Britain the exhibition standard for Leghorns calls for a much larger comb than is desired in Canada and the United States. The writer has seen combs on normal Leghorn cockerels in poultry shows in this country, which were quite as big as those removed from Nos. 21 and 25. The suggestion is made that this process of breeding for large combs has brought that structure to a size where its wearer may be taxed to the limit to support it. In such exceptional cases as here

described it may even grow beyond the limits of endurance of the supporting muscles and unless the cause is removed the muscle fatigue thus induced may cause death from starvation and exhaustion.

After No. 25 had completely recovered, a circlet of leaden washers weighing 50 gm. was fastened around the stub of comb remaining. This weight he was able to carry quite easily, and did so for a week. Two months later a 76 gm. leaden crown (equal to the weight of comb and wattles removed) was fastened to the top of the cockerel's head. It was not possible to attach this so that its weight acted in the same direction as the removed comb, but after four days' observation, it was evident that the extra weight was reducing the bird to a condition similar to that induced by the comb, and it was therefore removed.

Champy<sup>(3)</sup> and others have adduced considerable evidence to show that in all probability the disharmonious increase in size of a part in relation to the body as a whole has led to the extinction of species. It is easy to recognise the inevitable end of the Irish Elk, for example, if, as the individuals of that species increased in general body size at a given rate for generation after generation, the antlers, already large, increased disproportionately in size and weight. Ultimately the head would become too heavy for the neck muscles to bear. Such heterogony (i.e. the growth of a part of the body out of proportion to the growth of the organism as a whole) must, indeed, limit the ultimate body size attainable by many species.

It would seem to be the case that there is a limit to the weight of comb which a body of limited size can sustain. If, by selection, the breeders seek to produce a Leghorn with an excessively large comb, they will probably also have to adopt the practice of "dubbing" (i.e. cutting off the comb), which, in turn, would defeat the object of breeding, or else the breed might be led to extinction. The writer does not suggest that the Leghorn fowl as a breed is doomed to such a fate, but the cases described above show that there is a limit beyond which a large comb can so interfere with the normal function of the bird as to render it non-viable.

Another reason for the preference of combs of medium rather than large size is the fact that in areas having cold winter climate loss from frozen combs is a serious problem. In Canada and the United States loss by this cause has been instrumental in establishing a preference for medium-sized combs, and an exhibition standard that would eliminate from show classes some of the large-combed Leghorns which carry off red and blue ribbons in this country.

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SEX DIMORPHISM AND VARIABILITY IN THE APPEN-  
DICULAR SKELETON OF THE LEGHORN FOWL

F. B. HUTT

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## SEX DIMORPHISM AND VARIABILITY IN THE APPENDICULAR SKELETON OF THE LEGHORN FOWL

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During the past five years there has been sent to this department a number of male fowls suspected by their owners of having been originally females and of having turned into functional males in a manner similar to that of the cases described in the fowl by Crew<sup>5</sup> and in the pigeon by Riddle.<sup>17</sup> In nearly every instance it has been impossible to verify the accuracy of the case history by breeding tests or by post mortem examination.

Sex dimorphism, in size as well as in plumage and colour, is more conspicuous in the Gallinae than in the majority of other birds. It seemed possible that this dimorphism might be so great that skeletal measurements would give a clue to the original sex of such individuals as those mentioned above. It was recognized that the range in length of any single bone in one sex would probably overlap the range of the same bone in the other sex, as Schneider and Dunn<sup>18</sup> have already demonstrated for several bones of the fowl. However, casual observations suggested that the difference in size between male and female fowls is more marked in the tarso-metatarsus than in other bones of the appendicular skeleton. This bone was not measured by the workers quoted. There was also some evidence from a pseudo-gynandromorph studied at this department that the sex dimorphism is greater in the proximal bones of the extremities than in the distal ones.\* Accordingly, at the suggestion of Professor Crew, the writer undertook to determine the extent of sex dimorphism in the appendicular skeleton of the fowl and to see if a significant dimorphism prevailed either in absolute length, or in proportional length of one or more bones, which might be of value in

\* In this bird, a report of which is being given elsewhere by Prof. Crew, the measurements of the appendicular skeleton were those of a male on one side and approximately those of a female on the other. Differences in shank colour showed that the disturbance had resulted from the loss of an autosome, not of a sex chromosome as would be at first suspected.

determining the original sex of fowls which had undergone partial or complete sex reversal.

#### MATERIAL

The material studied included practically all the mature fowls which died or were killed at this department from September 1927 to September 1928, as well as a number of carcasses kindly supplied by the Royal (Dick) Veterinary College, Edinburgh, to which they had been sent for autopsy. One hundred and fifty-three skeletons were measured but the data used in the present report are confined to those of Leghorn fowls only. Of these there were 53 females, 36 males and 16 capons. Theoretically the measurements for all varieties of this breed should be comparable but it is probable that some, notably the whites, are slightly larger on the average than others. The majority of the birds used in the present investigation were Brown Leghorns which were in some cases slightly below standard weights for the breed.

The fact that a number of the birds were diseased should in no way prejudice the reliability of the bone measurements since most diseases are probably unable to affect the size of any bone once growth is completed. Malformed and broken bones were not measured and one skeleton affected with erythromyelosis was discarded.

#### METHODS

After autopsy both legs and both wings of each bird were skinned and boiled until the flesh could be easily removed. When two or more skeletons were boiled together, cotton bags were used to keep them separate. The individual bones were scraped clean and allowed to dry for at least 18 hours at room temperature after which any remaining pieces of cartilage were removed from the epiphyses and measurements were taken. This was done by marking on a sheet of paper the extremities of each bone (using a sharp pencil) and measuring the distance between the two points. The procedure was simplified by using a vertical line as the starting point for all measurements. Care was taken to ensure that distances were always measured between the same points on each bone. These were as follows:

*Humerus*—from the proximal point of the head to the distal extremity of the ulnar tubercle.

*Radius*—from the margin of the concavity in the proximal end to the extremity of the tubercle on the distal convex surface.

*Ulna*—from the proximal point of the olecranon to the distal extremity of the inferior flange of the trochlear surface.

*Carpo-metacarpus*—from the extremity of the flange dorsal to the articular surface on the proximal end to the outermost point of the tuberculum articulare on the distal end.

*Two phalanges of third digit\**—the maximum distance between the proximal and distal extremities.

*Femur*—from the proximal extremity of the trochanter to the end of the external condyle.

*Tibio-tarsus*—from the projecting point of the cnemial process to the farthest point on the external condyle.

*Tarso-metatarsus*—from the end of the ridge between the two surfaces for articulation of the tibio-tarsal condyles to the extremity of the central trochlea.

*Four phalanges of third digit*—from the tubercle dividing the articular grooves on the proximal surface to the extremity of the condyles on the distal end. The claw was removed from the ungual phalanx before that bone was measured.

Measurements of all bones were made to the nearest half millimetre. In every case (except where odd ones were lost) all the bones listed above were measured in duplicate, *i. e.* in both right and left limbs, the average of each pair being taken as the correct length for that bone. It is evident therefore that the average measurement might be expressed to 0.25 mm. In the majority of cases the measurements were coincident and only in rare cases was there a greater difference than 0.5 mm.

In statistical analysis of the absolute lengths, the humerus, radius, ulna, femur, tibio-tarsus and tarso-metatarsus were grouped in 1.0 mm. classes and the remaining bones into 0.5 mm. classes.

#### I ABSOLUTE BONE LENGTHS

The biometric constants for absolute lengths of the bones of

\* This terminology is based on the embryological evidence as presented by Lillie<sup>12</sup> which indicates that the first and fifth digits are lost in development while the second, third, and fourth persist. Kaupp<sup>8</sup> and Bradley<sup>2</sup> refer to these two phalanges as constituting the second digit.

TABLE 1. BIOMETRIC CONSTANTS OF ABSOLUTE BONE LENGTHS

Bone	No.	Mean. (mm)	Standard Deviation (mm)	Coefficient of Variation (%)
(a) Females				
Humerus	53	72.377 ± .350	3.778 ± .247	5.219 ± .342
Radius	52	64.154 ± .287	3.065 ± .203	4.777 ± .316
Ulna	52	70.943 ± .334	3.575 ± .236	5.039 ± .333
Carpo-metacarpus	51	39.206 ± .094	1.000 ± .067	2.551 ± .170
Phal. 1 of Dig. III	52	15.019 ± .038	.404 ± .027	2.623 ± .173
Phal. 2 of Dig. III	52	14.221 ± .047	.506 ± .033	3.558 ± .235
Femur	53	82.075 ± .401	4.330 ± .283	5.275 ± .346
Tibio-tarsus	53	116.566 ± .627	6.699 ± .439	5.748 ± .376
Tarso-metatarsus	52	78.615 ± .476	5.088 ± .366	6.472 ± .428
Phal. 1 of Dig. III	51	19.235 ± .063	.667 ± .044	3.415 ± .228
Phal. 2 of Dig. III	51	15.412 ± .048	.507 ± .034	3.289 ± .219
Phal. 3 of Dig. III	52	14.375 ± .046	.492 ± .032	3.422 ± .226
Phal. 4 of Dig. III	50	12.970 ± .046	.488 ± .032	3.762 ± .253
(b) Males				
Humerus	36	80.639 ± .419	3.720 ± .296	4.613 ± .367
Radius	36	72.505 ± .385	3.427 ± .272	4.723 ± .375
Ulna	36	80.472 ± .421	3.745 ± .286	4.654 ± .369
Carpo-metacarpus	36	44.319 ± .103	.921 ± .073	2.078 ± .165
Phal. 1 of Dig. III	35	16.686 ± .056	.429 ± .030	2.571 ± .207
Phal. 2 of Dig. III	35	16.386 ± .056	.489 ± .035	2.984 ± .240
Femur	36	92.288 ± .496	4.413 ± .350	4.782 ± .380
Tibio-tarsus	36	133.528 ± .709	6.316 ± .502	4.731 ± .376
Tarso-metatarsus	35	91.629 ± .609	5.340 ± .430	5.828 ± .469
Phal. 1 of Dig. III	35	20.900 ± .069	.589 ± .047	2.818 ± .227
Phal. 2 of Dig. III	35	16.843 ± .049	.438 ± .035	2.600 ± .209
Phal. 3 of Dig. III	35	15.829 ± .054	.478 ± .038	3.019 ± .243
Phal. 4 of Dig. III	35	13.857 ± .058	.509 ± .041	3.673 ± .296
(c) Capons				
Humerus	16	82.700 ± .633	3.753 ± .447	4.538 ± .541
Radius	16	74.012 ± .589	3.495 ± .417	4.587 ± .547
Ulna	16	82.419 ± .638	3.757 ± .448	4.557 ± .543
Carpo-metacarpus	16	45.312 ± .368	2.186 ± .260	4.824 ± .575
Phal. 1 of Dig. III	15	16.913 ± .131	.754 ± .093	4.458 ± .549
Phal. 2 of Dig. III	15	16.127 ± .116	.942 ± .116	5.841 ± .719
Femur	16	95.094 ± .698	4.138 ± .493	4.351 ± .519
Tibio-tarsus	16	138.109 ± 1.143	6.779 ± .808	4.908 ± .585
Tarso-metatarsus	16	95.218 ± 1.106	6.029 ± .719	6.332 ± .755
Phal. 1 of Dig. III	15	21.846 ± .236	1.355 ± .166	6.202 ± .764
Phal. 2 of Dig. III	16	17.425 ± .172	1.021 ± .121	5.859 ± .699
Phal. 3 of Dig. III	16	16.328 ± .196	1.165 ± .139	7.135 ± .851
Phal. 4 of Dig. III	16	14.969 ± .185	1.099 ± .131	7.342 ± .875

the males, females and capons studied are given in Table 1. Variation in the numbers measured in any class merely indicates that some bones were lost, broken or malformed.

(a) *Variability*

In such material it is to be expected that the absolute variation, as measured by the standard deviation, would be in general proportional to the magnitude of the object measured. For comparison of organs of different size the coefficient of variation must be employed and when this is done the present data afford some interesting exceptions. In males, females and capons the tarso-metatarsus proved to be relatively more variable, as indicated by the coefficient of variation, than any other bone in the appendicular skeleton, though it was exceeded in mean length by the tibia in all three classes and by the femur in males and females.

There is in all three classes distinct evidence that the variability of the phalanges (in both wing and leg) increases progressively toward the distal extremity while the mean lengths of these bones decrease progressively in the same direction. If this increased variability were apparent only in the standard deviation, it might be construed as having arisen from the progressive decrease in the means, but since it is also evident in the coefficients of variation (which are corrected for the magnitudes of the means), a definitely greater variability in the lengths of the distal phalanges is indicated. The same phenomenon is apparent in the measurements and variation constants recently given for the phalanges of all four digits of the fowl by Alpatov and Boschko-Stepanenko.<sup>1</sup> Assuming that the length of any bone is a function both of the rate of growth and of the duration of the growth period, the problem arises of why either or both of these processes should be more variable in the distal than in the proximal phalanges. No solution can be attempted in this paper.

The coefficients of variation for the humerus, ulna, femur and tibia in this material are slightly higher than those given by Schneider and Dunn (*loc. cit.*) for the same bones from 348 female White Leghorns and 46 males of the same variety. Since the difference is equally as apparent in the males, where the numbers were approximately equal, as in the females, where their

number of individuals was over six times that of the writer, it may be concluded that the greater variability of the writer's material results from the greater heterogeneity of the stock rather than from the comparatively small numbers available.

(b) *Sex Dimorphism*

In the mean lengths of each bone measured there is a statistically significant sex dimorphism as shown in Table 2. The males are consistently larger than the females and the difference is in no case less than twelve times its probable error.

TABLE 2. COMPARISON OF MALES WITH FEMALES

Bone	Difference in Mean Length in mm. $\bar{\sigma} - \bar{\eta}$	Range in mm.		$\frac{(\bar{\sigma} - \bar{\eta})}{\bar{\eta}} \times 100$
		$\bar{\sigma}$	$\bar{\eta}$	
Humerus	8.262 ± .553	14.75	15.50	11.41
Radius	8.351 ± .480	13.25	13.75	13.02
Ulna	9.529 ± .537	15.50	14.00	13.43
Carpo-metacarpus	5.113 ± .139	7.50	8.50	13.04
Phal. 1 of Dig. III	1.667 ± .067	3.75	3.50	11.09
Phal. 2 of Dig. III	2.165 ± .072	3.50	4.50	15.22
Femur	10.213 ± .637	18.00	17.25	12.44
Tibio-tarsus	17.062 ± .946	23.25	27.25	14.55
Tarso-metatarsus	13.014 ± .772	21.75	21.00	16.55
Phal. 1 of Dig. III	1.665 ± .093	4.00	6.25	8.66
Phal. 2 of Dig. III	1.431 ± .068	4.00	4.50	9.29
Phal. 3 of Dig. III	1.454 ± .070	4.50	4.50	10.11
Phal. 4 of Dig. III	.887 ± .073	4.50	4.75	6.84

In Table 2 the extent of the range between minimum and maximum bone length in each sex is also given. Comparison of these with the means (Table 1) shows that for every bone the range of absolute bone length in one sex overlaps the limit of that bone in the other sex. The absolute length of any one bone is therefore of no value in diagnosing the sex of doubtful individuals despite the significant differences in mean length. Since the range of any of these bones in one sex may overlap the limit of variation of the same bone in the other sex, it follows that the range of the total length of all these bones in males may overlap the limits of the same figure in females and therefore that total absolute length of any number of bones in the appendicular skeleton will not give a figure confined solely to either sex.

The range for any one bone is approximately the same in each sex. This confirms the suggestion of Schneider and Dunn (*loc. cit.*) that the distinctly smaller ranges found in the bones of their males were due to the smaller numbers of that sex studied.

Considering only the humerus, ulna, femur and tibio-tarsus, the male bones are 13.4 per cent greater on the average than those of the female. For these same bones Schneider and Dunn give a difference of 16 per cent. In the data of Latimer<sup>10</sup> for these four bones from approximately 20 mature individuals of each sex, a difference (computed by the present writer from Latimer's averages) of 15.6 per cent is found. The greater sex difference in the material of these other workers may possibly have resulted from the somewhat greater average size of their birds.

The difference between males and females is most extreme in the tarso-metatarsus. This is of special significance when considered along with the fact that in both sexes this bone displays a greater relative variability than any other of those measured. The observations of Latimer (*loc. cit.*) on the ages at which ossification of various bones is completed explain both of these phenomena. Whereas in the humerus, radius, ulna, femur and tibio-tarsus ossification was found to be completed from 10 to 47 days later in males than in females, ossification in the tarso-metatarsus was not completed in males till 56 days later than in females. Latimer's data suggest that in comparison with other bones of similar size, ossification of the tarso-metatarsus is completed relatively early in females and relatively late in males. Why this phenomenon should occur is an unsolved problem, but it is attested both by Latimer's observations and by the greater variability and sex dimorphism observed in this bone by the present writer.

The greater percentage growth of the male skeleton is less in the toe bones than in the longer bones of the leg and wing.

## II. PROPORTIONAL BONE LENGTHS

In order to determine if a significant sex dimorphism existed in the proportional lengths of bones in the appendicular skeleton, for every bird measured there was calculated the ratio of each bone to a more proximal bone in the same limb. The hum-

erus and femur were chosen purely arbitrarily as bases for the indices. In every skeleton the length of each of these bones was taken as 100 and the proportional length of every other bone in the same limb expressed on this basis. The biometric constants for these data are shown in Table 3.

(a) *Variability*

As is to be expected, the variability in the proportional length (Table 3) of every bone is much less than that in the absolute lengths (Table 1). In general those bones which showed the greatest variability in absolute length also exhibit the greatest variability in proportional length. As the extremity of each limb is approached the ratio of each phalanx to the humerus or femur becomes more variable. In the ratio of the tarso-metatarsus to the femur, a greater variability is evident than in any other index.

The ratios found and the degrees of variability are not in entire accord with the report of Latimer (*loc. cit.*) who states that the ratio of radius to humerus remains constant at 92.5 per cent after 1300 grams of body weight. In the present writer's material this index ranged from 85 to 91 per cent in females and from 87 to 93 per cent in males with a mean of 88.5 for 53 females and of 89.9 for 36 males. A few of the females were under 1300 grams but for these Latimer gives an even higher index. Similarly his statement that "the adult tibio-femoral ratio in the chicken is 147" can hardly apply to all individuals since in the material described in this paper the tibio-femoral index ranged from 137 to 148 with a mean figure of 141.7 for females and of 144 for males. However, Latimer was dealing not only with smaller numbers but also with larger birds of a less heterogeneous stock and on that account would probably find less variability. His measurements were taken to include the maximum length of each bone and should therefore be comparable with those made by the present writer.\*

Among the skeletons of other breeds not included in this

\* Latimer's statement (page 37) that in females the ulna is on the average 0.05 cm. greater than the humerus is obviously a misprint since his averages are 7.52 cm. for the humerus and 7.47 cm. for the ulna.

TABLE 3. BIOMETRIC CONSTANTS OF PROPORTIONAL BONE LENGTHS

Bone	No.	Mean. (mm)	Standard Deviation (mm)	Coefficient of Variation (%)
(a) Females				
Humerus	53	100.	0	0
Radius	52	88.529 ± .073	.779 ± .051	.879 ± .058
Ulna	52	98.144 ± .056	.599 ± .039	.613 ± .039
Carpo-metacarpus	51	54.171 ± .024	.257 ± .017	.474 ± .031
Phal. 1 of Dig. III	52	20.793 ± .018	.190 ± .012	.914 ± .060
Phal. 2 of Dig. III	51	19.705 ± .024	.259 ± .017	1.314 ± .087
Femur	53	100.	0	0
Tibio-tarsus	53	141.735 ± .109	1.172 ± .076	.827 ± .054
Tarso-metatarsus	52	95.548 ± .128	1.367 ± .090	1.431 ± .094
Phal. 1 of Dig. III	51	23.333 ± .023	.244 ± .016	1.045 ± .069
Phal. 2 of Dig. III	51	18.731 ± .016	.174 ± .011	.929 ± .062
Phal. 3 of Dig. III	52	17.562 ± .017	.187 ± .012	1.065 ± .070
Phal. 4 of Dig. III	50	15.775 ± .019	.201 ± .013	1.274 ± .085
(b) Males				
Humerus	36	100.	0	0
Radius	36	89.972 ± .082	.731 ± .058	.813 ± .065
Ulna	36	100.055 ± .045	.565 ± .045	.564 ± .045
Carpo-metacarpus	36	54.833 ± .026	.232 ± .018	.423 ± .034
Phal. 1 of Dig. III	35	20.757 ± .017	.152 ± .012	.732 ± .059
Phal. 2 of Dig. III	35	20.200 ± .026	.232 ± .019	1.152 ± .093
Femur	36	100.	0	0
Tibio-tarsus	36	144.279 ± .116	1.033 ± .082	.716 ± .057
Tarso-metatarsus	35	99.201 ± .161	1.431 ± .114	1.443 ± .115
Phal. 1 of Dig. III	35	22.707 ± .023	.198 ± .016	.872 ± .073
Phal. 2 of Dig. III	35	18.257 ± .014	.127 ± .010	.696 ± .056
Phal. 3 of Dig. III	35	17.135 ± .024	.214 ± .017	1.248 ± .101
Phal. 4 of Dig. III	35	15.328 ± .025	.218 ± .017	1.422 ± .114
(c) Capons				
Humerus	16	100.	0	0
Radius	16	89.623 ± .158	.936 ± .112	1.044 ± .124
Ulna	16	99.690 ± .102	.606 ± .072	.608 ± .072
Carpo-metacarpus	16	54.314 ± .158	.939 ± .112	1.729 ± .206
Phal. 1 of Dig. III	15	20.524 ± .079	.451 ± .056	2.197 ± .270
Phal. 2 of Dig. III	15	19.566 ± .234	1.342 ± .165	6.859 ± .845
Femur	16	100.	0	0
Tibio-tarsus	16	145.233 ± .338	2.004 ± .241	1.310 ± .156
Tarso-metatarsus	16	100.085 ± .479	2.843 ± .339	2.845 ± .339
Phal. 1 of Dig. III	15	23.002 ± .120	.690 ± .085	2.999 ± .369
Phal. 2 of Dig. III	16	18.366 ± .112	.667 ± .079	3.632 ± .433
Phal. 3 of Dig. III	16	16.908 ± .115	.686 ± .082	4.057 ± .484
Phal. 4 of Dig. III	16	15.726 ± .133	.788 ± .094	5.011 ± .597

study were those of three dwarf fowls in which, owing to an unfavourable environment, growth had been stunted. In these, the tarso-metatarsus exhibited a greater deviation from its normal ratio to the femur than did any other bone (Table 4).

TABLE 4. RATIO TO FEMUR OF LEG BONES OF THREE DWARF FOWLS

	♀ 35	♂ 68	♂ 69
Tibio-tarsus -----	139.13	143.57	136.54
Tarso-metatarsus -----	84.78	88.86	84.62
Phalanx 1 of third digit-----	22.82	22.43	21.77

In fowls of other breeds the proportional lengths of the bones were found to be very similar to those in Leghorns. Comparison of the data for these three dwarfs with the ratios in Table 3 shows that while the phalanx and the tibio-tarsus exhibit little deviation from the normal ratio, the tarso-metatarsus is much shorter in every case. The inference is that this bone has not only greater potentialities for growth than the others in the appendicular skeleton, but is also distinctly more susceptible to conditions unfavourable for growth.

(b) *Sex Dimorphism*

A statistically significant sex dimorphism exists in the mean ratio of each wing bone (except Phalanx 1) to the humerus and of each leg bone to the femur. The differences, though not given here, have been calculated and (except in Phalanx 1 of the wing) are in each case more than 13 times their probable errors. In the phalanges of the toe, the proportional length is greater in females than in males. For the other leg bones and those of the wing the ratio is greater in males than in females. The first phalanx in the third digit in the wings presents an exception for which no explanation can be offered.

As in absolute length, the range of proportional length for any bone in the male was found to overlap the limit of that index in the same bone in the female. Such an index can not therefore be used with accuracy in determining the sex of an individual fowl. The same finding applies to several other ratios tried.

Latimer (loc. cit.) observed a sex difference in the weights, plotted against gross body weight, of the humerus, tibio-tarsus and tarso-metatarsus but not in other long bones studied. He also

noted sex-dimorphism in the lengths of the three proximal bones in the wing and leg when plotted against body weight and age. The present writer attempted to establish indices of bone length to body weight but found that these overlapped in the two sexes even more than did the other ratios reported above. Schneider and Dunn<sup>18</sup> and Kopec<sup>9</sup> present data to show that body weight is three to six times as variable as are bone measurements. Latimer also found a sex difference in the weight of bone per unit of length in the humerus and tarso-metatarsus (but not in others) when plotted against body weight.

### III. CAPON SKELETONS

The range from minimum to maximum in every bone is less in the capons than in the normal males as is to be expected from the smaller numbers measured. The greater variability of the capons in absolute bone lengths (Table 1) suggests that a disturbance in growth results from castration but larger numbers are desirable before this point can be considered proven.

With the exception of the distal phalanx of the wing the mean absolute lengths for all bones in the capons are greater than the means for the same bones in normal males (Table 5). The 6 larger bones are on the average 2.9% greater in capons than in the males.

Similar results have been observed following castration in rabbits by Poncet<sup>16</sup>, in guinea-pigs by Pirsche<sup>15</sup>, in rats by Hatai<sup>7</sup> and in steers and ovariectomised cows by Tandler and Keller<sup>22</sup>.

In these capons the increased size of the bones compared with those of males is statistically significant in the humerus, tibio-tarsus and three phalanges of the toe. If greater numbers had been available the probable errors of the means would have been considerably less and the differences would probably be significant in most cases, since the majority of them are even now either greater than three times their probable error or almost that magnitude. It is also significant that the increase in size is consistent for all the bones except one phalanx of the wing.

One important principle is evident, viz. that the increased growth of capons is greater in the posterior than in the anterior limb. The same phenomenon has been observed in eunuchs and

TABLE 5. COMPARISON OF CAPONS WITH MALES

Bone	Mean Absolute Length		
	Capon minus ♂ mm.	Difference P. E.	(Capon—♂)x100 ♂
Humerus	2.061 ± .759	2.7	2.56
Radius	1.507 ± .703	2.1	2.08
Ulna	1.947 ± .764	2.7	2.42
Carpo-metacarpus	.993 ± .382	2.6	2.24
Phal. 1 of Dig. III	.227 ± .142	1.6	1.36
Phal. 2 of Dig. III	— .259 ± .128	2.0	— 1.58
Femur	2.806 ± .856	3.3	3.04
Tibio-tarsus	4.581 ± 1.345	3.4	3.44
Tarso-metatarsus	3.589 ± 1.262	2.8	3.92
Phal. 1 of Dig. III	.946 ± .245	3.9	4.52
Phal. 2 of Dig. III	.582 ± .178	3.3	3.46
Phal. 3 of Dig. III	.499 ± .202	2.4	3.15
Phal. 4 of Dig. III	1.112 ± .193	5.7	8.03

eunuchoid individuals by Tandler and Grosz<sup>21</sup>, Launois and Roy<sup>11</sup> and in rabbits by Poncet (*loc. cit.*). Launois and Roy cite the investigations of several other workers also reporting the same phenomenon in eunuchs and cases of gigantism associated with infantilism. The writer has been unable to find any other measurements of capon skeletons but the principle is amply demonstrated by the indices in Table 6, taken from the material described in this paper.

It is evident that in capons not only is the femur greater in proportion to the humerus but the tarso-metatarsus is greater in proportion to the femur and the whole posterior limb is greater in proportion to the anterior limb than in males. Comparison of the data on capons and males in Table 3 reveals that, whereas in the anterior limb the proportions of all bones to the humerus are less in capons than in males, in the posterior limb the ratios of all bones to the femur are greater than in males except in one phalanx. It is evident, however, that in all three of the indices used there is a marked difference between females and normal males. As stated previously this difference results largely from growth being protracted in males after it has ceased in females. In the opinion of the writer the still greater differences in these indices for the capons result from a protraction of growth in that class after it has ceased in the males. The increase in these

TABLE 6. INDICES SHOWING THE ELONGATION OF THE POSTERIOR LIMB IN CAPONS

	No.	Femur x 100 Humerus	7 leg bones x 100 6 wing bones	Tarso- metatarsus x 100 Femur
Female	52	113.4	122.9	95.5
Males	35	114.4	123.7	99.2
Capons	16	114.9	125.7	100.0

indices and the elongation of the posterior limb result, therefore, from a continuation of a differentiating process which is already operative, rather than from any special heterogony to be included among the sequelae of gonadectomy.

The slight bilateral asymmetry observed in eunuchs by Tandler and Grosz (loc. cit.) was not apparent in the capons.

Mitchell, Card and Hamilton<sup>14</sup> found that before 18 weeks and after 32 weeks there was little difference between the weights of cockerels and capons in White Plymouth Rocks but their records show the capons to have exceeded the cockerels in weight from the 18th to the 32nd week. Waite<sup>24</sup>, also using White Rocks, found that up to 24 weeks of age there was little difference but that from 24 to 42 weeks the capons exceeded the cockerels. Titus and Jull<sup>23</sup> computed from the equation of a curve of diminishing increment that in Rhode Island Reds, cockerels and pullets mature at 21 to 23 months and capons at 17.4 months.

While there is a great difference between the rates of growth of Leghorns and the heavier breeds, it seems logical to assume that if in the latter the capons do not grow faster than normal cockerels up to 5 or 6 months of age, neither will they do so in Leghorns up to a corresponding period of 4½ to 6 months. Nevertheless the capon skeletons are larger than those of the males and one is therefore led to the conclusion that skeletal growth is protracted in capons after it has ceased in normal males. This is in accord with the findings of Mitchell, Card and Hamilton and of Waite, although it is impossible to say how much of the greater gains made by their capons after six months represent growth of skeleton and how much is accumulation of fat. Such a conclusion is not necessarily a contradiction to the computation of Titus and Jull that capons mature sooner than normal males

since the question of what constitutes maturity is open to different interpretations and theirs can obviously not be based on skeletal growth. It is particularly difficult to decide what constitutes maturity in a capon, or if a capon may be said to mature at all in the same sense as a normal male, since its condition corresponds to one of indefinitely protracted sexual infantilism. The data of Schneider and Dunn<sup>18</sup> show that in Leghorns maximum growth of the skeleton is attained before eight months.

## DISCUSSION

The data in Table 2 shed some light on the case of the gynandromorph fowl described by Macklin<sup>13</sup> and her bird in turn gives a clue to the principles underlying sex dimorphism in general. Macklin's fowl (judging from photographs of the head) was a Barred Plymouth Rock. Nevertheless it is evident (Table 7) that the difference in skeletal measurements between left and

TABLE 7. COMPARISON OF NORMAL SEX DIMORPHISM WITH THE BILATERAL ASYMMETRY OF MACKLIN'S GYNANDROMORPH

	Hutt	Macklin
	Ratio $\frac{\text{♀}}{\text{♂}}$	Ratio $\frac{\text{Left}}{\text{Right}}$
Humerus -----	.89	.86
Radius -----	.88	.87
Ulna -----	.88	.85
Femur -----	.88	.86
Tibio-tarsus -----	.87	.82

right sides of this bird was almost identical with that between females and males in the present writer's material.

In Macklin's gynandromorph both testis and ovotestis were found at autopsy. Whether or not these had both been present during the growth period is not known but as Macklin says "the secretions of any endocrine glands concerned with growth were equally available to the two sides, so that the explanation must rest upon the different genetic constitutions of the two halves." Presumably sex dimorphism in normal fowls is also due to the genetic constitution of the soma rather than to the endocrine secretion of the gonad. This does not mean that endocrine secretions play no role whatever in growth. The data on gonadecto-

mized animals show a distinctly abnormal growth in that class resulting apparently from a disturbed inter-endocrine balance affecting the pituitary. It is evident, however, that the ultimate growth of any individual depends upon the ability of the soma to respond to all the factors stimulating growth, and that presumably this ability is determined by its genetic constitution.

Contrary to the opinion of Castle<sup>3</sup> <sup>4</sup>, but in accord with those of Davenport<sup>6</sup>, Wright<sup>25</sup>, Sumner<sup>19</sup> <sup>20</sup> and Kopec<sup>9</sup> the writer is led to the conclusion that while there are genetic size factors affecting all parts of the body, there must also be special factors affecting only certain regions and certain individual bones. This is attested by the extreme variability and sex dimorphism in the tarso-metatarsus, by the fact that the greater size of bones in males is not uniform for all bones and by the elongation of the posterior limb to a greater extent than the wing when growth is protracted in males and capons.

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#### SUMMARY

The appendicular skeletons of 53 females, 36 males and 16 capons were measured and subjected to biometrical study.

A statistically significant sex-dimorphism was found in the mean absolute length of all bones measured. With the exception of one phalanx in the wing, a statistically significant sex dimorphism was also found in the mean ratio of every wing bone to the humerus and of every leg bone to the femur. In the phalanges of the toe the length proportional to the femur is greater in females than in males. In all other bones the ratio to the humerus or femur is greater in males than in females.

The longer bones in males fowls were 13.4 per cent greater than in females. The increase in the males was not constant for all bones but varied from 6.8 to 16.5 per cent.

The tarso-metatarsus exhibited a greater variability both in absolute length and in proportional length than any other bone. It also represented the extreme sex dimorphism being 16.5 per

cent greater in males than in females. In dwarfed fowls the growth of the tarso-metatarsus was found to be retarded more than that of other bones. It is concluded that this bone differs from others in its potentialities for growth and its susceptibility to environmental conditions.

Variability in the absolute and proportional lengths of the phalanges was found to increase progressively toward the distal extremity of either limb.

Capons were found to be slightly larger in all bones (except one) than males. In the six longer bones the average difference was 2.9 per cent. It was shown that in capons the increase in size is greatest in the posterior limb but reasons are given for the opinion that the elongation of that limb results from the continuation of a process already operative in normal males and not from a heterogonic growth induced by gonadectomy.

It is concluded that sex dimorphism results from a differential response of the soma, caused by its genetic constitution, to the different factors promoting growth.

No index was found which would permit diagnosis of sex in doubtful individuals from a study of the appendicular skeleton.

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