A COMPARATIVE BIOMETRIC STUDY OF ALBINO AND COLOURED GUINEA-PIGS FROM THE POINT OF VIEW OF THEIR SUITABILITY FOR EXPERIMENTAL USE.

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(With 3 Diagrams.)

Introduction.

Prior to the year 1923 most of the guinea-pigs required for experimental work at the National Institute for Medical Research were obtained by purchase from selected dealers. In the previous year certain work on biological standardisation had been begun, involving the use of large numbers of guinea-pigs of a definite age and weight, and the method of animal supply then in vogue was found to be unsatisfactory for the following reasons:

(1) It was difficult to obtain a regular supply of sufficient animals of the required age and weight.

(2) It was difficult to avoid the introduction of infected animals, in spite of elaborate quarantine precautions.

(3) It was demonstrated experimentally that some, at least, of the variability in the results then being obtained was due to the use of animals of varying and unknown origin and pedigree.

Accordingly, towards the end of 1922 it was decided to build up a breeding stock of guinea-pigs of our own. As the change from an external source of supply to a self-supporting one could not be effected immediately, the following plan was adopted. A number of albino animals and a number of cream, and cream and white, coloured animals were selected and mated. The offspring were left with the mothers until they reached about 150 grm. in weight. The young were then separated with regard to sex and kept, two in a cage, until fully grown and sexually mature. During this time the young animals were weighed daily, and any animal which failed to grow at a normal rate, or showed any signs of illness, was rejected along with its cage companion. In this way some twenty females and twelve males were selected as the source of the breeding stock, from which the animals used at the Institute for the past six years have been drawn. The chosen animals, when mature, were mated and records kept of the progeny. As the population increased the method of accommodating the breeding stock was altered somewhat. Pens

about five feet square were constructed and ten to twelve females and two or three males were run together. As the females became advanced in pregnancy they were transferred to separate boxes, 2 ft. 6 in. long by 1 ft. 3 in. deep by 1 ft. 3 in. high, each box having a darkened half and a netting-fronted half. When the young were born, the date of birth, the number in the litter and colour of the individual offspring were recorded, and, for the purpose of the enquiry the results of which are given in the following pages, the weight at birth, the daily weight for the first twelve to fourteen days of life, and deaths were also recorded.

It will be noticed that guinea-pigs of two colours only were retained for breeding purposes, viz. (1) albino, and (2) cream, or cream and white, coloured. The reason for this is that the skin of an animal, rather than the whole animal, is being used in immunological work to an ever-increasing extent, and white or cream coloured animals are equally suitable for skin testing or for whole-animal experiments. In other words, it is uneconomical to breed animals with dark coloured skins on account of their relatively limited use. As was to be expected, the progeny of albino parents were invariably white in colour. It was expected that the offspring of cream and cream and white guinea-pigs would be variously coloured, but from the start the progeny of these animals were, in nearly all cases, similar in colour to the parents. Castle (1912) states that there are races of yellow and white guinea-pigs which breed true, and it is possible that the original animals in our experiment were of this type. The intensity or shade of the colouring varied from a very pale cream to the familiar light brown. The only exception which has been observed during a period of some six years is that, occasionally, one or more members of a family bred from cream or cream and white parents exhibited a patch of rat-coloured hair usually on the head or rump; by removing the animals which produced these young from our breeding stock we appear to have succeeded in eliminating the tendency for this type of animal to be produced.

With regard to the feeding of the guinea-pigs during the past six years, this has varied but little throughout the period, and the scheme adopted has been based upon the work of Glenny and Allen (1921), which one of us had been privileged to see carried out. Glenny and Allen showed conclusively that an epizootic among guinea-pigs could be controlled by an efficient diet. This work was continued and extended by Boock and Trevan (1922). It is of interest to note that Murray, Webb and Swann (1925–6) investigated a disease which occurred among their stock rabbits, and remark that "the impression gained was that really adequate food would terminate the epidemic." Glenny and Allen showed that, of the readily available fresh green food, cabbage ranks very high in the merit scale as foodstuff for guinea-pigs, and accordingly every effort has been made to supply this vegetable in adequate amounts to the guinea-pigs. At certain times of the year the supply has been short and other green food (*e.g.* kale) has been given. We are able to record that, during the past six years, we have had no epidemic among our guinea-

pigs, and on the rare occasions on which a few cases of deaths have been attributable to bacterial infection (and due, apparently, to an organism of *B. enteritidis*, Gaertner type) the spread of the disease has been prevented by attention to the diet. Throughout the whole period the death-rate has been very low; whether this is due to the particular care which has been given to the diet, as would be suggested by the work of Glenny and Allen, or to the fact that no new guinea-pigs have been imported since 1923 and thus the introduction of a fresh infection avoided, it is impossible to say.

As a result of the precautions taken, a healthy stock of animals was, perhaps, to be expected. Certain other results, however, were unexpected, and became obvious at an early date by an inspection of the record cards. The chief of these unexpected results was the large number of large litters obtained. Litters of four were common, litters of five by no means infrequent, and occasionally litters of six were born. During one period of eight months in 1924 the average size of the litters was about 3·5, and during the period under review the average size of the litters was three. This appears to us to be higher than is usually believed to be the case with guinea-pigs bred in this country. Whether the large family size is due to the accident that, in the original choice of the first parents of our present breeding stock, some naturally prolific animals were unconsciously selected, or whether the high fertility rate is due to the regular and liberal supply of a diet containing, possibly, some accessory food factor concerned with fertility and reproduction, we cannot definitely say. From certain observations we have made, however, we are inclined to ascribe the large family size to the latter cause.

We have not yet had the opportunity to subject such views as we hold, regarding the cause of the large family size and the freedom from epidemic disease, to such strict experimental enquiry as could furnish absolute proof. Our prime concern has been to produce a healthy stock of animals in numbers adequate for our needs, and to avoid at all costs the introduction of infection and the development of an epidemic. In this we have been successful; in addition we have been fortunate in obtaining large litters; and, in experiments made with the animals we have bred, we have obtained clear evidence that their reaction to certain biological stimuli is remarkably uniform (Hartley, *1925, 1926).*

In 1926 we decided, for various reasons, to make a comparative study of the two classes of guinea-pigs maintained at the farm—albino and cream and cream and white coloured—in respect of fertility, mortality, weight at birth and rate of growth. In the preceding three years the two kinds of animals had been used indifferently for a variety of biological experiments, and no evidence had been obtained to show that one class behaved or reacted in any way differently from the other. From our records up to that time we were not able to decide whether albino guinea-pigs were more or less prolific, more or less difficult to rear, or differed in weight at birth, or in rate of growth, or in any other way from the cream and cream and white coloured animals.

If either group was really superior to, or in any way more satisfactory than the other, our breeding stock was then sufficiently large to enable us to increase the number of the more satisfactory group and eliminate the less suitable animals. Other reasons which led to our making the survey, the results of which are recorded and analysed below, were the following:

(i) For purposes of administration, supply of animal food (particularly green food), housing of animals, allocation of subordinate staff, etc., we desired to know whether any particular season of the year favoured the size and number of litters, the robustness of the progeny and the death-rate and, if so, to what extent.

(ii) Experience had shown that occasionally, for the purpose of some particular experimental work, a demand was made for a hundred or more guinea-pigs. We desired to know whether such demands could be met at any time during the year, or whether at certain seasons larger stocks were available than at others.

(iii) Reference has been made to the large litters obtained. We considered it of importance to discover the relationship of initial weight at birth to the family size, to determine the rate of growth of the individuals occurring in families of different size, and to determine whether the death-rate among the young was related in any way to family size.

From the data thus obtained we hoped to be able to ascertain, for example, whether individual members of a large family were markedly inferior in weight at birth, or showed an abnormally slow rate of growth or an abnormally high death-rate in early life; and to decide whether it might not prove more economical to reduce the number of such families immediately after birth by sacrificing one or more of the less robust members.

In order to collect the necessary data to enable us to come to a decision regarding these various questions it was decided to make more extended observations regarding the guinea-pigs born at Rhodes Farm during at least one year. Actually records were kept from August 1926 to September 1927—a period of fourteen months. In regard to each family born the following particulars were recorded:

(1) Date of birth.

(2) Number in the litter.

(3) Weight at birth of each member of a family.

(4) The daily weight of each member of every family during the first twelve to fourteen days of life, or until the animal reached 150 grm. in weight.

(5) Number of deaths, and age at death.

(6) The age at which the young were withdrawn from observation.

When the experiment was completed, a preliminary examination of the numerical data was made by one of us; it was evident, however, that the data should be subjected to a critical statistical analysis.

The nature and results of this analysis will now be described. All the collaborators in this work are jointly responsible for its general conclusions but

two of us (G. W. Dunkin, P. Hartley) are directly responsible for the experimental and observational work, and two (E. Lewis-Faning and W. T. Russell) for the statistical analysis. We are deeply indebted to Prof. Greenwood for the assistance he has given. We wish also to record our appreciation of the conscientious and unremitting attention which Mr L. Gammage, who has been in charge of the small animals at the Farm during the whole period of the enquiry, has devoted to the collection of data.

The data utilised in the analysis consist of the pigs born by crossing albino bucks and does, cream and cream and white bucks and does at the Medical Research Council's Farm, at Mill Hill, between August 1926 and September 1927. Particulars of the available data have been given above. Environmental conditions were identical for the two types of guinea-pig. The pens and cages were situated indoors throughout the year, and in the winter months the animal house was efficiently heated. The animals were reared in cages of the same dimensions and given the same form of diet, so that any differentiation that may exist between the two classes is one of physiological importance and not due to the force of external circumstances.

DISTRIBUTION OF SIZES OF LITTER.

The experience covers 404 litters of albino and 278 of cream, cream and white guinea-pigs (hereafter called, for convenience, coloured guinea-pigs) and the distribution according to colour and number in the litter is shown in Table I. The smallest and largest litters were the most infrequent. Litters

Table I. *Showing the frequency of the various size litters.*

	Albino		Cream, cream and white		Albino + cream, cream and white	
No. born in litter	No. of litters	Percentage of total	No. of litters	Percentage of total	No. of litters	Percentage of total
1	18	4·5	11	3·9	29	4·3
2	93	23·1	58	20·8	151	22·1
3	184	45·5	122	43·9	306	44·9
4	91	22·4	68	24·5	159	23·3
5	16	4·0	18	6·5	34	5·0
6	2	0·5	1	0·4	3	0·4
	404	100·0	278	100·0	682	100·0

containing only one animal formed 4·5 per cent. of the total albino litters; the proportion increased to 23·1 per cent. in the next group and attained a maximum of 45·5 per cent. in the case of families of three guinea-pigs. The number of families with four births declined to 22·4 per cent., and there were only two instances, or less than 1 per cent. of the total, in which litters of six pigs were born. The corresponding proportions for the mixed colour guinea-pigs were almost identical. Litters containing two guinea-pigs constituted 20·8 per cent. of the total for this class, and those in which there were three guinea-pigs born formed 43·9 per cent. of the total mixed colour litters. Generally it may be said that the distribution according to size, that is, the

number born in the litter, is markedly symmetrical in albino litters and in a slightly less degree in those of mixed colour. Combining the two groups so as to form one frequency distribution, we obtain the actual values and their relative proportions as set out in the fifth and sixth columns of the table. The apparent symmetry, which has already been mentioned, will be seen in Diagram 1, where the ordinates, plotted to form a frequency polygon, suggest that a normal curve would describe the distribution.

When this was fitted it was found that the equation was

$$y = 293{\cdot}07\, e^{(-x^2/1{\cdot}7241)},$$

where x is the distance from the origin. The theoretical frequencies deduced from this equation are given in apposition to the actual values in Table II, and shown on Diagram 1. Application of the usual "Goodness of Fit" test shows that the graduation is satisfactory.

Table II. *Showing the comparison between the actual number of litters and the expected values deduced from the equation of the normal curve.*

No. born in litter	No. of litters		$(\text{Act.}-\text{Exp.})^2$	$\frac{(\text{Act.}-\text{Exp.})^2}{\text{Exp.}}$
	Actual	Expected		
1	29	26	9	0·35
2	151	156	25	0·16
3	306	293	169	0·58
4	159	172	169	0·98
5	34	32	4	0·13
6	3	2	1	0·50
				$\chi^2 = 2{\cdot}70$
				$P = 0{\cdot}75$

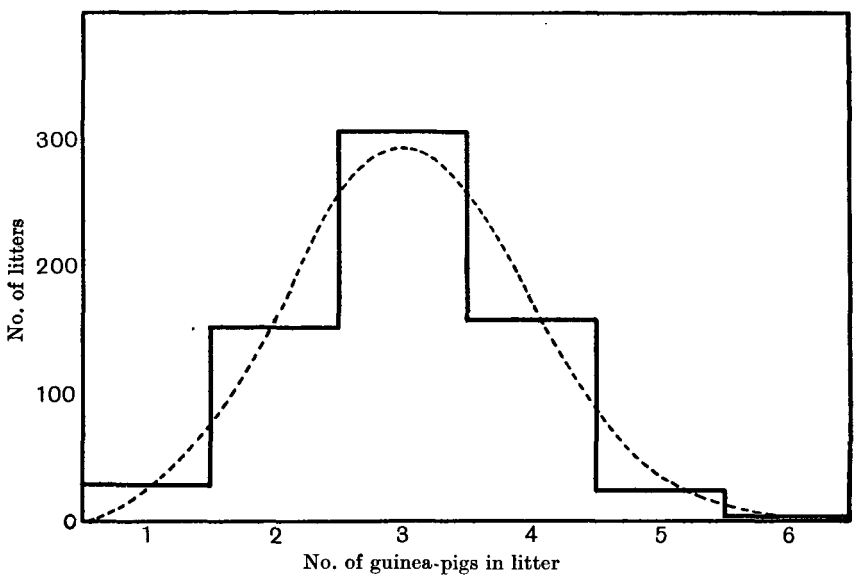

Diagram 1. Showing the frequency of size of litter.

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Seasonal incidence of litters.

As will be observed from Table III there is little evidence to indicate a definite seasonal distribution in the frequency of the litters. The highest incidence in the case of the albino guinea-pigs occurred in September 1926, when the proportion of litters born was 10·3 per cent. of the total but, in the same month in the succeeding year, the ratio was as low as 4·7 per cent. The

Table III. *Showing the seasonal incidence of litters.*

Albino

		No. born in litter								
		1	2	3	4	5	6	Total	%	
1926.	August	1	6	13	9	2	—	31	7·7	18·0
	September	4	11	21	6	—	—	42	10·3	
	October	2	4	18	5	—	—	29	7·2	23·3
	November	1	8	12	9	1	—	31	7·7	
	December	—	5	19	9	1	—	34	8·4	
1927.	January	—	9	17	3	—	—	29	7·2	15·7
	February	1	7	5	3	—	—	16	4·0	
	March	—	11	6	1	—	—	18	4·5	
	April	1	5	11	10	—	—	27	6·7	23·7
	May	2	10	19	3	1	—	35	8·6	
	June	1	7	12	11	2	1	34	8·4	
	July	2	3	11	6	5	—	27	6·7	19·3
	August	3	6	11	9	3	—	32	7·9	
	September	—	1	9	7	1	1	19	4·7	
	Total	18	93	184	91	16	2	404	100·0	

Cream, cream and white

		No. born in litter								
		1	2	3	4	5	6	Total	%	
1926.	August	1	3	11	8	2	—	25	9·0	15·1
	September	—	5	6	3	3	—	17	6·1	
	October	3	8	9	6	—	1	27	9·7	27·3
	November	1	3	10	11	4	—	29	10·4	
	December	—	2	14	4	—	—	20	7·2	
1927.	January	2	2	9	3	—	—	16	5·8	16·2
	February	—	4	7	3	—	—	14	5·0	
	March	1	9	5	—	—	—	15	5·4	
	April	—	9	8	2	1	—	20	7·2	17·3
	May	—	3	8	4	—	—	15	5·4	
	June	1	1	6	4	1	—	13	4·7	
	July	1	5	14	8	2	—	30	10·8	24·1
	August	1	3	5	6	2	—	17	6·1	
	September	—	1	10	6	3	—	20	7·2	
	Total	11	58	122	68	18	1	278	100·0	

maxima in the mixed group occurred at two different periods of the year, 10·4 per cent. in November and 10·8 per cent. in July, but it is extremely doubtful if any importance can be attached to these values for the maximal incidence, as the months in which they occur are not coincident for the two classes of guinea-pig. They probably arise merely by chance. To eliminate the rapid fluctuations in the monthly values the figures were summed for periods

of three months: this resulted in four periods of three months and one period of two months' duration. For the purpose of maintaining correspondence with the ordinary quarter of the year, August and September of 1926 were taken to form the two-monthly period. In the quarter, October–December, the proportion of albino litters was 23·3 per cent. of the total. During the January to March period the ratio was at a minimum value of 15·7 per cent. and, although the proportion increased to 23·7 per cent. in the succeeding interval, it declined to 19·3 per cent. in the last quarter of the period under review. In the case of the mixed colour guinea-pigs, the percentage frequency was at a maximum at the end of 1926, when the value was 27·3 per cent. It decreased to 16·2 per cent. during the first quarter of 1927 and, although it showed only a slight increase during the April to June period, the value for the summer months reached 24·1 per cent. As regards the comparability of the seasonal incidence for the two classes of guinea-pigs, it will be seen that, in both instances, the ratios for the three-monthly values start at maximal points, 23·3 per cent. and 27·3 per cent. respectively, and reach their lowest values of 15·7 and 16·2 per cent. during the same quarter, January–March 1927. Afterwards they pursue irregular courses.

Relation of size of litter to coat colour.

When we come to consider the fertility in the two classes of guinea-pig, certain considerations may possibly affect the figures, but it has been impossible to make any allowance for them. Firstly, there were no data available relating to the does as regards their previous natality. In other words, it has been impossible to decide whether large litters alternated with small ones, or whether the does always had an average sized litter. Secondly, there was no record kept of the age of the parents. If the age distribution of the parent guinea-pigs, especially the female, is different in the two classes, this introduces a difficulty in the comparison of their relative fertility, because fertility declines with age. In the absence of any information on this point, the age distribution was regarded as identical for both classes. This assumption is possibly not very wide of the mark. The average number of guinea-pigs born in litters of different colour and at different periods of the year is stated in Table IV, where it will be seen that there is little or no relationship between the colour of the guinea-pigs and the number of their progeny. The average number of guinea-pigs born in albino litters is 3·000 and the corresponding value for the mixed colour is 3·097. There is thus a difference of 0·097 between the means, but the probable error of this difference is $\pm$ 0·049. If the variation indicated is to be regarded as statistically significant, it should exceed three times its probable error and, as will be seen, it fails to fulfil this criterion. Hence, we are justified in concluding that the fertility of guinea-pigs is not differentiated according to colour.

Table IV. *Showing the seasonal incidence in fertility.*

ALBINO.

		Total litters	Total young	Average per litter		Coefficient of variability
1926.	August ...	31	98	3·16	2·89 ± ·071	31·2
	September ...	42	113	2·69		
	October ...	29	84	2·90		
	November ...	31	94	3·03	3·04 ± ·055	26·2
	December ...	34	108	3·18		
1927.	January ...	29	81	2·79		
	February ...	16	42	2·63	2·65 ± ·059	26·2
	March ...	18	44	2·44		
	April ...	27	84	3·11		
	May	35	96	2·74	3·03 ± ·064	30·7
	June	34	111	3·26		
	July	27	90	3·33		
	August ...	32	99	3·09	3·29 ± ·082	32·7
	September ...	19	68	3·58		
	Total ...	404	1212	3·000 ± ·031		30·5

CREAM, CREAM AND WHITE.

		Total litters	Total young	Average per litter		Coefficient of variability
1926.	August ...	25	82	3·28	3·26 ± ·102	30·0
	September ...	17	55	3·24		
	October ...	27	76	2·81		
	November ...	29	101	3·48	3·14 ± ·076	31·3
	December ...	20	62	3·10		
1927.	January ...	16	45	2·81		
	February ...	14	41	2·93	2·67 ± ·079	29·6
	March ...	15	34	2·27		
	April ...	20	55	2·75		
	May	15	46	3·07	2·98 ± ·083	28·7
	June	13	42	3·23		
	July	30	95	3·17		
	August ...	17	56	3·29	3·31 ± ·077	28·2
	September ...	20	71	3·55		
	Total ...	278	861	3·097 ± ·038		30·5

All periods combined: difference between mean of all albino and mean of all cream, cream and white pigs = 0·097 ± 0·049.

SEASONAL VARIATIONS IN FERTILITY.

As regards the seasonal distribution, the results support the idea that fertility varies according to the period of the year.[1] During the two-monthly period, August–September, the average size of albino litters is 2·89, in the succeeding quarter the size increases to 3·04 per litter, but during January–March the value declines to a minimum of 2·65 ± 0·059. Afterwards there is a steady increase, the fertility in the period, April–June, being 3·03 ± 0·064, and it attains its maximum value of 3·29 ± 0·082 in the last quarter of the period under review. The seasonal trend of the fertility of the guinea-pigs of mixed colour is almost identical with that for the albino guinea-pigs. The average number of young per litter is 3·26 ± 0·102 in the first two months, the rate declines in the two succeeding quarters, having its minimum value of

[1] The gestation period of the guinea-pig is about 70 days duration.

2·67 ± 0·079 during January to March, and becoming almost equal in size to that for the albino guinea-pigs. In the remaining periods there is a decided increase in the rate, and in the last quarter of our experience the maximum fertility prevails when the average number of guinea-pigs is 3·31 ± 0·077. Generally it may be said that there is a relationship between fertility and the period of the year, but there is no significant difference in the average number of animals born to albino litters and the average number born to coloured litters. It will be noted that the period at which the fertility is at its lowest point is January–March, and this is the same period at which we found the fewest litters were produced. It would thus appear that the winter months are unfavourable to the production of guinea-pigs, since not only are fewer litters produced but the size of such litters is small.

Weight at birth and size of litter.

The mean weight of the young animals at birth is stated according to the size of litter in Table V. The average weight at birth of an albino guinea-pig is 81·2 grm. and the corresponding weight of one drawn from the coloured group is 82·6 grm. There is thus a difference of 1·4 grm. between the average weight of the two types of guinea-pig but, as this difference is less than three times its probable error, which is ± 0·56 grm., it cannot be regarded as statis-

Table V. *Showing the mean weight in grm. at birth according to the size of the litter.*

No. born in litter	Albino		Cream, cream and white		Average weight of albino minus average weight of cream, cream and white
	No. of young	Average weight in grm.	No. of young	Average weight in grm.	
1	18	103·3 ± 3·89	11	75·5 ± 6·17	+ 27·8 ± 7·29
2	186	92·3 ± 1·06	116	97·7 ± 1·17	− 5·4 ± 1·58
3	551*	83·7 ± 0·48	366	85·6 ± 0·63	− 1·9 ± 0·79
4	362†	73·5 ± 0·55	272	76·5 ± 0·61	− 3·0 ± 0·82
5	80	69·8 ± 1·07	90	70·9 ± 1·06	− 1·1 ± 1·50
6	12	72·5 ± 2·64	6	68·3 ± 2·72	+ 4·2 ± 3·79
Total	1209	81·2 ± 0·36	861	82·6 ± 0·43	− 1·4 ± 0·56

* One animal died shortly after birth and was not weighed.
† Two animals died shortly after birth and were not weighed.

tically significant. In families which occur rather frequently, namely, those in which two, three and four births occur, the mean weight of the mixed colour guinea-pig is in all instances greater than that for the albino one, yet, in only two instances, namely, in litters of two and four guinea-pigs, can the difference in the average weights be regarded as statistically sensible and, even then, it is only slightly beyond the range of significance. In albino litters containing two animals, the mean weight of the young at birth is 92·3 grm.; the corresponding value for the coloured is 97·7 or a difference of 5·4 grm. with a probable error of ± 1·6 grm. Similarly in the case of litters containing four guinea-pigs the mean weight at birth of the albino animals is 73·5 grm., and that for the mixed class is 76·5 grm. or a difference of 3·0 grm. to which is

attached a probable error of ± 0·8. With regard to litters containing three and five guinea-pigs respectively, it was found that no real importance could be attached to the variation in the mean weight between the two colours.

When the weight is studied in conjunction with the size of or the number born in a litter, it was found that the mean weight declines as the families become larger. In litters containing one animal, although the frequency of this class is rather small, the mean weight of albino guinea-pigs at birth is 103·3 grm. It declines to 81 per cent. of this value in the case of litters of three guinea-pigs and, in families in which five animals are born, the average weight at birth is only 68 per cent. of that in the smallest litter. Owing to the paucity of the data no reliance can be placed on the mean weight of the mixed colour litters containing one guinea-pig, but, if the weights in the succeeding litters are expressed as percentages of that with two births, it will be observed that the rate of declension is in close concordance with that for

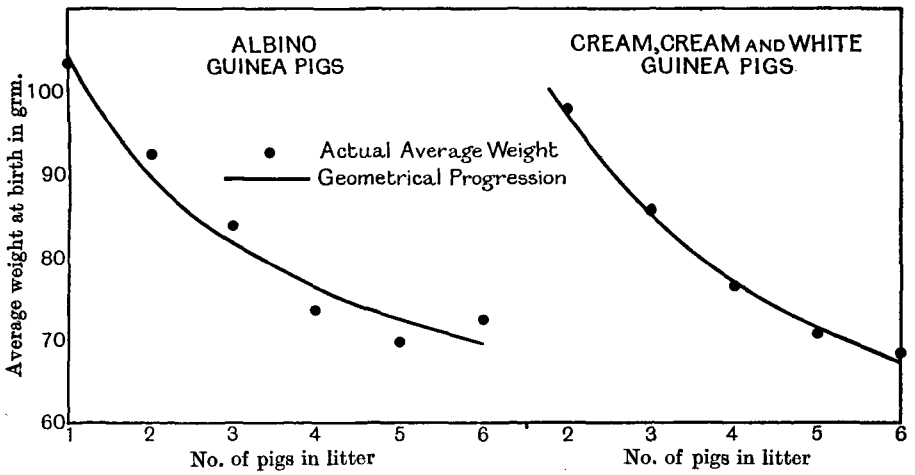

Diagram 2. Showing the actual and graduated weights at birth in litters of different size.

albino guinea-pigs in the corresponding groups. The average weight at birth of mixed coloured litters in which there are two guinea-pigs is 97·7 grm., and the corresponding value in the case of families with five guinea-pigs is 70·9 grm., a decrease of nearly 27 per cent. The trend of the mean weights according to the size of the litter is shown in Diagram II, and here it will be seen that the relationship between the size of the family and the average weight at birth cannot be described satisfactorily by a linear function. In view of the fact that the ratio of each ordinate to the previous ordinate is roughly constant, we are led to suppose that the relationship might be described by a geometric progression, which is based on the fact that a quantity grows or declines in such a way that its increase or decrease in value during any equal interval is proportional, not to its original value, but to the value at the beginning of that interval. It is well known that a curve of this description can be suitably applied to many problems in the field of biology. The observations were fitted

by least squares to an equation of this nature and the theoretical values deduced. As will be seen from the diagram, there is close agreement between the actual values and the theoretical curve. The mean weight in families with one guinea-pig in the litter in the case of the mixed colour class was excluded from the calculations, as it is extremely doubtful if this value of 75·5 grm.

Table VI. *Showing the comparison between the actual mean weight at birth and the corresponding graduated values.*

No. born in litter	Albino. Mean weight at birth		Cream, cream and white. Mean weight at birth	
	Actual	Graduated	Actual	Graduated
1	103·3	104·8	75·5	—
2	92·3	89·5	97·7	97·5
3	83·7	81·6	85·6	85·1
4	73·5	76·4	76·5	77·3
5	69·8	72·7	70·9	71·7
6	72·5	69·7	68·3	67·4
	$\chi^2=0{\cdot}495$	$P=0{\cdot}981$	$\chi^2=0{\cdot}033$	$P=0{\cdot}997$

truly represents the actual weight of animals in this group. Table VI shows the actual and graduated values of the mean weight at birth. Testing the correspondence between them, by the usual method P is found to be 0·981 for the albino group and 0·997 for the coloured.

Weight and season.

The next point considered was to ascertain if any variation occurred in the mean weight of litters born at different periods of the year. The information on this point is contained in Table VII, where the mean weights at different seasonal periods for litters of varying size are expressed as percentages of the

Table VII. *Showing* (A) *the total number born in litters of given size at different periods, and* (B) *their mean weight at birth expressed as a percentage of the average value for the whole period.*

Size of litter	Aug.–Sept. 1926		Oct.–Dec. 1926		Jan.–Mar. 1927		April–June 1927		July–Sept. 1927	
	A	B	A	B	A	B	A	B	A	B
Albino.										
1	5	109	3	100	1	92	4	96	5	96
2	34	103	34	105	54	89	44	109	20	95
3	101*	108	147	97	84	87	126	108	93	97
4	58†	104	92	99	28	93	96	101	88	100
5	10	104	10	114	—	—	15	88	45	100
6	—	—	—	—	—	—	6	100	6	100
Cream, cream and white.										
1	1	79	4	83	3	108	1	146	2	109
2	16	95	26	104	30	93	26	101	18	109
3	51	97	99	102	63	90	66	102	87	106
4	44	99	84	97	24	91	40	109	80	102
5	25	103	20	102	—	—	10	94	35	99
6	—	—	6	100	—	—	—	—	—	—

* One animal died shortly after birth and was not weighed.
† Two animals died shortly after birth and were not weighed.

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corresponding values for the total duration of fourteen months. The chief feature exhibited in this table is that once again attention is centred on the January–March quarter as the most unfavourable period, the tendency being for guinea-pigs born in these months to be below the average in weight. Taking the most frequent sized litters, namely, those in which three births occur, we find that in August–September 1926, albino guinea-pigs are 8 per cent. above the general average in weight. In the succeeding quarter there is a decline of 3 per cent., and the rate of decrease becomes more accelerated during the January–March period as the value is 13 per cent. below the normal. In the ensuing quarter the mean weight increases. During April to June the value is 8 per cent. above the average, and in the last interval of the period under review normality is almost reached. As regards the mixed colour families we find for litters of three that, during August–September 1926, the average weight of coloured guinea-pigs is 3 per cent. below the standard, but during October to December there is an excess of 2 per cent. In the January to March quarter we have a decrease amounting to 10 per cent. After this period the values rise above the general average.

GROWTH AND AGE.

The weight of an immature animal of course increases, as it grows older, at a rate which slackens as maturity approaches. It follows that the law of growth with age cannot be linear although for many purposes—for instance, a rough graduation of changes in weight with age of children from three or four years to puberty—a straight line may describe the change with sufficient accuracy. It has been found that, when the present data are treated as wholes, that is to say, all the available observations tabulated by size and litter and colour (so forming six separate groups) and graduated by straight lines, these latter represent the course of events fairly well through the earlier days of life, but diverge very sensibly from the observations at the later ages, especially in the case of litters containing two animals. We surmised that the explanation of this might be that animals removed from the litters and brought into laboratory use within the early days of life were heavier than average guinea-pigs, so that the subsequent averages of the remnant might represent an unfavourable selection. An examination of Table VIII fully confirms this expectation. Thus of 186 animals born to albino parents in litters of two, 119 or 64 per cent. were removed within the first thirteen days of life and 74 or 40 per cent. of the births were taken away before the eleventh day. The corresponding proportions during the same period for coloured animals in litters of the same size were 66 per cent. and 53 per cent. respectively. There was less stringent selection in the case of litters with three in family, as the numbers removed before the eleventh day formed only 12 per cent. of the total births in albino families and 21 per cent. in coloured families. In the largest sized litters of numerical importance, namely, those containing four births, the influence of selection on the rate of growth was almost negligible,

Table VIII. *Showing* (I) *the total number of guinea-pigs weighed each day and their average weight,* (II) *the number of removals each day and their average weight on day prior to removal.*

(I) A = Total number weighed each day.
B = Their average weight each day.

(II) C = The number removed each day.
D = Average weight on day prior to removal.

Day ...	0	1	2	3	4	5	6	7	8	9	10	11	12	No. born	Total removed (0–12 days inclusive)	Percentage of births	Total removed (0–10 days inclusive)	Percentage of births
ALBINO.																		
2 in litter:																		
A	186	164	158	146	148	126	117	144	105	100	80	54	32					
B	92	95	100	107	114	122	130	131	132	135	137	136	132					
C	—	—	—	—	2	5	6	11	22	12	16	25	20					
D	—	—	—	—	168	156	154	164	160	159	166	159	157	186	119	63·9	74	39·7
3 in litter:																		
A	551	479	456	480	449	407	375	494	438	403	405	353	297					
B	84	86	87	93	98	104	109	114	118	121	126	127	130					
C	—	3	—	—	—	—	—	1	20	20	24	32	30					
D	—	97	—	—	—	—	—	150	152	157	154	154	140	551	130	23·6	68	12·3
4 in litter:																		
A	362	333	313	272	314	275	245	330	310	271	255	287	239					
B	74	75	77	81	87	91	95	100	105	109	112	115	117					
C	—	—	—	—	—	—	—	4	7	—	—	8	14					
D	—	—	—	—	—	—	—	96	160	—	—	158	156	362	33	9·1	11	3·0
CREAM, CREAM AND WHITE.																		
2 in litter:																		
A	116	108	102	97	88	83	61	86	74	58	41	29	26					
B	98	101	106	113	120	127	130	138	139	141	136	134	142					
C	—	—	—	2	—	2	10	8	10	16	14	15	—					
D	—	—	—	148	—	153	159	151	158	159	159	155	—	116	77	66·4	62	53·4
3 in litter:																		
A	366	326	309	303	274	286	248	323	273	251	213	181	151					
B	86	88	92	97	104	110	113	118	121	126	126	129	131					
C	—	—	—	—	—	—	9	6	15	18	30	21	29					
D	—	—	—	—	—	—	154	151	157	144	155	146	146	366	128	31·9	78	21·3
4 in litter:																		
A	272	236	230	228	215	215	183	249	238	222	213	192	173					
B	77	79	80	87	90	96	101	105	110	111	117	118	122					
C	—	—	—	—	—	—	—	—	—	4	—	12	20					
D	—	—	—	—	—	—	—	—	—	105	—	153	145	272	36	13·3	4	1·5

because only 3 per cent. of the albino births and 1·5 per cent. of the coloured births were taken away before the eleventh day. In practically every instance the animals removed on particular days were heavier than those kept under observation for longer periods. If we take as an example the recorded weights for albino guinea-pigs with two in family, we find that six animals were taken away on the sixth day, and their mean weight on the previous day (the animals were not weighed on day of removal) was 154 grm. as compared with an average of 122 grm. for all animals weighed on that particular day. It will be observed that there is less divergence between the values for animals which were removed, and between those for what may be conveniently termed the residual animals in litters with three and four in family. This is, of course, what we should expect, since fewer animals were taken away from these groups for experimentation before the eleventh day.

Table IX. *Showing the mean weights of guinea-pigs under observation for different periods.*

A = Average daily weight of those guinea-pigs under observation from day of birth until the expiration of the twelfth day.

B = Average daily weight of all guinea-pigs excluding those removed at any time prior to the eleventh day.

C = Average daily weight of all guinea-pigs including those weighed up to and on day prior to removal.

ALBINO.

2 in litter

Day...	0	1	2	3	4	5	6	7	8	9	10	11	12
A	77	79	82	89	92	99	106	110	116	121	125	127	132
B	84	84	89	95	101	108	116	120	126	133	137	136	132
C	92	95	100	107	114	122	130	131	132	135	137	136	132

3 in litter

A	81	82	83	89	94	100	103	108	112	116	123	126	130
B	82	84	85	91	96	102	105	110	115	119	126	127	130
C	84	86	87	93	98	104	109	114	118	121	126	127	130

4 in litter

A	73	73	75	79	85	88	93	98	103	107	109	110	117
B	73	74	76	81	86	90	94	99	105	109	112	115	117
C	74	75	77	81	87	91	95	100	105	109	112	115	117

CREAM, CREAM AND WHITE.

2 in litter

A	82	83	90	89	98	106	110	116	120	124	124	133	142
B	89	91	97	102	110	115	121	131	133	141	136	134	142
C	98	101	106	113	120	127	130	138	139	141	136	134	142

3 in litter

A	80	81	85	89	94	99	104	109	114	118	121	126	131
B	82	83	87	92	98	102	107	113	118	122	126	129	131
C	86	88	92	97	104	110	113	118	121	126	126	129	131

4 in litter

A	76	78	79	85	87	93	99	102	107	108	112	115	122
*B	78	80	81	88	90	96	101	105	110	111	117	118	122
C	77	79	80	87	90	96	101	105	110	111	117	118	122

* Some of the guinea-pigs removed were smaller than those kept in.

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To exhibit more clearly the influence of the selective factor, the data were re-classified and the mean daily weights were then calculated for the following categories:

(A) Those animals under observation throughout the entire period.

(B) All animals weighed except those removed before the eleventh day.

(C) All animals weighed on particular days.

The results are presented in Table IX. It will be observed that there are distinct differences between the mean weights in Groups A and C for the smallest sized family. A graduation of the values in C, the total weighings, cannot be expressive of the rate of growth as the observations from age to age are not homogeneous. On the other hand, although the values in Group A are strictly comparable, yet they relate to guinea-pigs which were sub-normal in weight at birth, and a graduation of the mean weights will not represent the actual relationship between growth and age in the present experience. Under the circumstances, the values in Group B were accepted as offering the best criterion, because only animals removed on or before the eleventh day were excluded from our calculations. Accordingly straight lines were fitted to the first ten weighings, *i.e.* from birth to the ninth day inclusive, and the extrapolates of these lines compared with the actual weights. The results are shown in Table X and in Diagram 3. It will be seen that in the three-in-litter series and in the four-in-litter series the concordance is excellent but, in the smallest sized family, the extrapolates exceed the observed values. Taking then the results for the three- and four-in-litter series as more fairly comparable—being less stringently selected—a comparison of the albino with the coloured guinea-pigs does not disclose any important difference between the respective rates of growth. To measure the possible influence of a seasonal factor the data were distributed in quarterly periods, but an analysis of the results revealed no material difference between the rates of growth of guinea-pigs born in the various quarters of the year.

Mortality.

We now come to examine the final phase of these statistics, namely, the mortality amongst young guinea-pigs. In obtaining the values for the population exposed to risk, a certain hypothesis was made. In the present experience the total births, apart from those which died, were affected by another consideration. Some animals were taken away at different ages for experimental purposes, and these were accepted as withdrawals in an actuarial sense. They were regarded as half units, or, in other words, they were accepted as being exposed to risk for half a day on the day of their transfer. For instance, if six guinea-pigs were born in a family and three were taken away on the next day then, according to the method adopted in the analyses, six animals were regarded as being exposed to risk at age 0–1 day and 4·5 on the succeeding day. This practice was pursued in all cases.

As regards the number of deaths amongst our stock during the initial

Table X. *Showing* (A) *the actual and* (B) *the graduated weights of guinea-pigs in litters from which all animals removed on or before the eleventh day were excluded.*

	ALBINO						CREAM, CREAM AND WHITE					
	2 in litter		3 in litter		4 in litter		2 in litter		3 in litter		4 in litter	
Day	A	B	A	B	A	B	A	B	A	B	A	B
0	84	79	82	79	73	70	89	86	82	79	78	76
1	84	85	84	84	74	74	91	92	83	84	80	80
2	89	91	85	88	76	78	97	98	87	89	81	84
3	95	97	91	92	81	82	102	104	92	93	88	88
4	101	103	96	97	86	87	110	110	98	98	90	92
5	108	109	102	101	90	91	115	116	102	103	96	96
6	116	114	105	105	94	95	121	122	107	108	101	100
7	120	120	110	110	99	99	131	128	113	112	105	104
8	126	126	115	114	105	104	133	134	118	117	110	108
9	133	132	119	119	109	108	141	140	122	122	111	112
10	137	138	126	123	112	112	136	146	126	127	117	116
11	136	144	127	127	115	116	134	152	129	131	118	121
12	132	149	130	132	117	121	142	158	131	136	122	125

N.B. The graduated values for the tenth, eleventh and twelfth days are extrapolated.

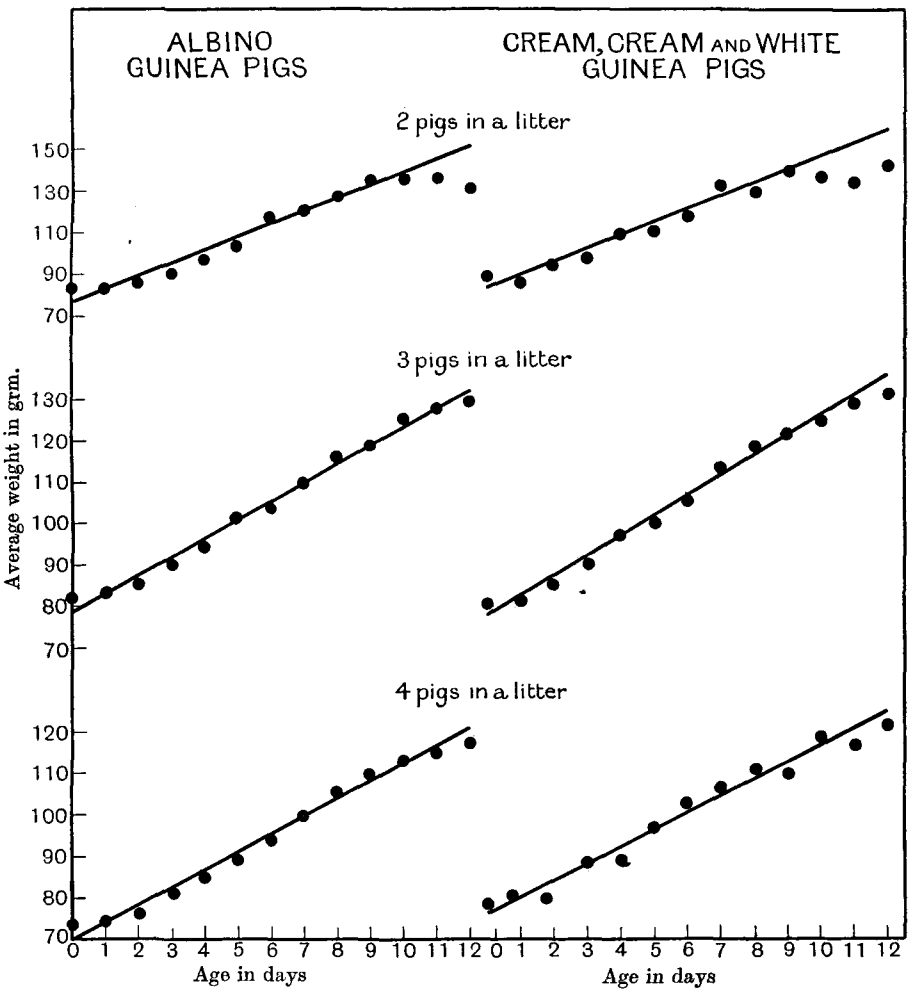

Diagram 3. Showing the actual and graduated daily weights of all guinea-pigs except those removed before the eleventh day.

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thirteen days of life, there were 80 deaths amongst the albino and 48 amongst the coloured guinea-pigs. When allowance was made for any variation that might be present in the age constitution of the respective populations, it was found that the standardised death-rate amongst the albino animals was 5·58 per 1000 per day as compared with a value of 4·73 for the coloured. The number of still-births was exceptionally low. There was only 1 still-birth amongst the albino and none amongst the coloured. In view of the records published by other observers, it would appear that the returns for the still-births in our data were very deficient but, as far as can be ascertained, they represent the actual facts. During the period which the stock was under observation this peculiarity was noted by the superintendent of the animal department and, as the pregnant does were examined at least twice a day, the possibility of any still-birth being overlooked was considerably reduced. To measure the mortality in the different series, we applied the death-rates at specific ages for the whole experience to the numbers exposed to risk at the various ages in the different sized litters, and so obtained the numbers of expected deaths which we compared with the actual values. Assuming, for all practical purposes, that the standard error of the expected number of deaths is measured by the square root of that value, we arrived at the results in Table XI. We found that in the total series of albino guinea-pigs there should

Table XI. *Showing the actual and expected deaths in litters of different size.*

Size of litter	No. of young	Deaths		(Actual-Expected) deaths / Standard Error of Expected
		Actual	Expected	
Albino.				
1	18	2	0·84	+1·28
2	186	16	10·43	+1·72
3	552	29	34·74	−0·97
4	364	25	23·19	+0·38
5	80	5	5·03	−0·01
6	12	3	0·69	+2·80
Total ...	1212	80	74·90	+0·59
Cream, cream and white.				
1	11	2	0·59	+1·85
2	116	4	6·58	−1·01
3	366	27	22·34	+0·99
4	272	9	17·40	−2·01
5	90	4	5·85	−0·77
6	6	2	0·36	+2·71
Total ...	861	48	53·12	−0·70

have been, on the average, 74·90 deaths. There were actually 80 deaths, but the difference does not exceed the standard error. Similarly, we predicted 53·12 deaths amongst the coloured animals as against an actual occurrence of 48 deaths. In the different sized litters the excess or defect between the actual and expected results never exceeds three times the standard error in any instance, and there is practically no uniformity in the size or sign of the differences. Furthermore, the excess which we noted in the standardised

death-rate for the total albino animals, as compared with the coloured, is of no significance, because it will be observed that the results for the important litters, namely, those containing three and four births, are in the reverse direction, *i.e.* the actual number of deaths is in defect of the expected for albino litters having three births and in excess for coloured animals, but in litters in which extra birth occurs, the mortality amongst the albino is slightly above the standard whilst that for the coloured is decidedly in defect. Hence we conclude that the mortality in the present data is independent of size and colour of litters, and is indeed contrary to what might be expected, as generally one would expect to find a fair degree of relationship between mortality and size of family.

An attempt was made to study the death-rate in the first year of life in accordance with the number of births in the litter, but the figures were rather insufficient for such a purpose.

SUMMARY.

1. The average number of guinea-pigs born in albino litters is 3·000 as against 3·097 in the case of the mixed group, but the difference between the mean values is not statistically significant. Hence, it will be seen that there is little or no relationship between the colour of guinea-pigs and the number of their progeny.

2. It may be said that there is a relationship between fertility and the period of the year. There is a tendency for fewer litters to be born during the quarter, January to March, and likewise for the fertility to be lowest during this period, as the mean number of births per litter is 2·65 for albino guinea-pigs and 2·67 for the mixed class, both values being significantly below the mean for the whole period.

3. The mean weight of the albino guinea-pigs at birth is 81·2 $\pm$ 0·36 grm., the corresponding value for the cream, cream and white class is 82·6 $\pm$ 0·43, but the difference is of no statistical importance. Hence we conclude that the weight of a guinea-pig at birth is not affected by its colour. Once again attention is centred on the January–March quarter as the most unfavourable period, since there is a tendency for guinea-pigs of either colour born in these months to be below the normal weight.

4. When allowance was made for the effects of selection on our data, there was no material difference between the rates of growth for the two types of guinea-pigs and, furthermore, the period of the year at which littering occurred exercised no apparent influence.

5. The rate of mortality during the first thirteen days of life amongst albino guinea-pigs is 5·58 per 1000 per day, and amongst cream, cream and white guinea-pigs 4·73 per 1000 per day, but the difference probably represents nothing fundamental because, when the mortality is studied according to the size of the litter, the rates are sometimes in the reverse direction.

6. Finally, there is, in the present data, nothing to suggest that albino guinea-pigs are as regards fertility, growth and mortality, significantly different from cream, cream and white guinea-pigs.

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