The maternal effects on growth and conformation in Shire horse-Shetland pony crosses

By ARTHUR WALTON AND JOHN HAMMOND, F.R.S.

School of Agriculture, University of Cambridge

[Plates 14, 15]

INTRODUCTION

The relative importance of heredity and environment in determining the size, development, and ultimate proportions of animals has attracted some scientific study and discussion, but little planned experimentation has been attempted with mammals. In the mammal the early and formative stages of development take place in the uterus, where the environment is remarkably constant, and it is not easy to subject the foetus to varying conditions and at the same time not overstep the normal physiological limits. Previous work on the rabbit (Hammond 1934 a; Wishart and Hammond 1933), however, has shown that the size of young at birth is approximately inversely proportional to the number in the litter, and it appears probable that some internal secretion or metabolic product of the mother forms a limiting factor in the growth of the embryo. In a monotocous species this effect does not appear, although exceptional twinning or polytocous births reveal its existence, the young at birth being regularly smaller. That the absolute amount of this hypothetical growth-promoting substance may be related in some way with the size of the mother is suggested by the figures of Gregory (1930), which show that the average number of ova of rabbits increases with the average size of the strain. In a monotocous species therefore the size of the mother might determine the size of the offspring at birth. This is also suggested by the results of reciprocal crosses between donkey and horse. The mule (donkey ♂ × horse ♀) is larger than the hinny (horse ♂ × donkey ♀) (Plumb 1916). Since, however, in a species cross the objection may be raised that abnormal genetic segregation might affect the result, we selected for our experiment two breeds of the same species, but of markedly different size, namely, the

[311]

Shire horse and the Shetland pony, and reciprocal crosses were made to see how far the size of the mother would affect the size of the offspring. No genetical analysis of size inheritance in horses has yet been attempted, and therefore it may be said that the genetic potentialities of our material were unknown. In the absence of evidence to the contrary, however, we considered it justifiable to assume, on the basis of much genetical experimentation on size inheritance, that a large number of factors would be involved, that segregation and recombination would be at random, and that the offspring of the two breeds would be genetically intermediate, irrespective of which way the cross was made. Any difference between the offspring of the reciprocal crosses would therefore be due not to chromosomal differences but to differences in the environment brought about by difference in the size of the mother. In other words we would have a controlled experiment in which "Mother-size" was the only or predominating variable.

The horse is particularly well adapted for studying the effects of maternal influence on the offspring, since the duration of pregnancy is relatively long (11 months) and the foal is born in an advanced stage of development, particularly with regard to the skeleton (see Meek 1901). Consequently, the effects of maternal influence acting over a long and formative period should be well marked.

The specific problems which we set out to investigate were:

(1) Would the offspring at birth be intermediate in size between the normal offspring of the parent breeds? If this were to be so the foetus in the small mother would certainly be too large and would either cause death of the mother or very difficult parturition or possibly premature abortion, while in the large mother the foal would be very much smaller at birth than normal for the breed.

(2) Would, on the other hand, the size of the mother by virtue of the growth-promoting substance available exert a controlling influence on the size of the foetus, so that in the case of the small mother the foetus would be limited in growth to such a size as she would normally give birth to, while in the case of the large mother the foetus would develop up to the size normal to the large mother?

(3) If a difference in size at birth were established, would this affect the subsequent growth and permanent proportions of the body?

(4) If permanent differences in adult life were established, would these differences be transmitted to subsequent generations?

Naturally a complete answer to the last question cannot be given for several years but we now have evidence to present on the first three.

The experiments relate to several problems of scientific interest, such as

the relative importance of heredity and environment, the effect of maternal nutrition on the growth and development of the offspring and the inheritance of "acquired characters". From an agricultural standpoint the experiments bear on the practical problems of making crosses between breeds of different size, on grading up stock for size, and on the influence of early nutrition on subsequent growth and proportions.

EXPERIMENTAL PROCEDURE

The small stud of Shetland ponies kept at the Animal Research Station, Cambridge, was used for this experiment. We obtained the use for breeding of a Shire mare from the University Farm close at hand. Pedigree Shire stallions were only available at some distance from Cambridge, but this difficulty was overcome by the collection of semen at the stud and its transportation to Cambridge. Owing to the difference in size of the parents natural coitus was of course impossible and artificial insemination had to be used. The methods were those developed in Russia and by ourselves, and previously described (see Imp. Bureau Animal Genetics 1933; Walton 1938). The mares at the Animal Research Station were tried daily with a sterile (vasectomized) stallion to determine oestrus. When one or more of the Shetland mares were on heat arrangements were made by telephone to collect Shire semen at the distant stud. Usually an oestrous mare (Shire) was available at the stud, and collection was made by means of the artificial vagina. The semen was brought back to Cambridge as soon as possible and the Shetland mares inseminated. The usual time between collection and insemination was about two hours.

When the Shire mare on the University Farm was in oestrus she was brought to the Research Station. Semen was collected from the Shetland stallion by means of the artificial vagina, using one of the Shetland mares as a mount, and the Shire mare was inseminated with the fresh semen. The percentage of successful inseminations was low (about 20 %). This, however, may have been due to inexperience with the method, or merely chance, and does not necessarily imply any incompatibility between the parents.

Details of the successful inseminations are shown in Table I.

PREGNANCY AND PARTURITION

Pregnancy was first recognized in the inseminated mares by the absence of heat and was confirmed by means of the mucus test (see Hammond

1934 *b*). During pregnancy the Shire mare was used on farm work until shortly before foaling and was kept in good working condition. The Shetland mares were run out in grass paddocks with plenty of wild white clover. They were only fed hay during the winter. They did no work and were in very good condition. An open shed gave shelter when wanted. The Shetland pony is very hardy and accustomed to the rigors of the open hillside or moor. Foaling takes place naturally in the open. For this reason no special precautions were taken on approach of parturition except to provide separate paddocks for the foaling mares.

In 1934 the Shire mare ("Daisy") in foal to the Shetland stallion, and the Shetland mare (S 1) in foal to the Shire stallion, foaled during the night without assistance, and the foals were found running with the mothers in the morning. In 1936 the two Shetland mares (S 1 and S 5) which were in foal to Shire stallions were given the same treatment, but the foals were found dead shortly after parturition. As will be seen from Table I, the weights of these foals were perfectly normal for the weights of the dams. There was no sign of dystokia. The placentas and foetuses were perfectly normal in appearance. The cause of death was accidental suffocation by the foetal membranes, as birth occurred during the night when they were not under observation. Had there been any difficulty at birth the mares would have required help.

In 1936 the Shire mare "Daisy" produced a second cross-bred foal. In addition to the cross-bred animals we had pure Shetland foals with which to make comparison of weights and growth rates. From birth onwards weighings, measurements and photographs of the foals were made periodically. The data are recorded in Tables I, II and III, and photographs of the parents and offspring, all brought to the same scale, are shown in Plates 14 and 15.

The principal features shown in Table I will be briefly noted here but a full discussion will be given later.

Duration of pregnancy. The average duration of pregnancy in the horse is about 340 days, but varies considerably with the time of year the foal is born and other circumstances (see Hammond 1934 *b*). As the date of service of the pure Shires was not recorded, the average duration for the species is given in brackets. The table shows that the duration of pregnancy varied considerably, but that there is no obvious relationship to the size of dam nor size of foetus. There is no significant difference between groups.

Weight of dam. The size of Shetland ponies varies considerably. Much higher prices are paid for the smallest animals. The ones we were able to

TABLE I. DATA RELATING TO PARTURITION

	Pure Shetland				Shetland-Shire crosses					Pure Shire	
					From Shetland dam			From Shire dam			
No. and sex of foals	S 5♀	S 6♀	S 7♂	S 50♀	D 2♀	D 4♀	D 5♀	D 1♀	D 3♀	Sh 1	Sh 2
Dam	S 4	S 3	S 5	S 1	S 1	S 1	S 5	Daisy	Daisy	Rosie	Pedigree mare
Sire	S W	S W	S W	S W	Bower Romancer	Bower Duke	Bower Duke	S W	S W	Ouse Bridge Albert	Pedigree Stallion
Date of insemination	*	17. viii. 33	22. v. 36	12. vii. 36	24. vii. 33	9. viii. 35	27. v. 35	30. vi. 33	16. v. 35	*	*
Date of birth	15. iii. 33	15. vii. 34	30. iv. 37	31. v. 37	6. vi. 34	28. vi. 36	10. v. 36	30. v. 34	23. iv. 36	9. v. 36	18. v. 36
Duration of pregnancy (days)	207	334	343	323	319	337	349	336	343	(340)	
Wt. of dam (kg.)	—†	142	185	229	229	229	185	797	797	(800)	(800)
Birth wt. of foal (kg.)	19·5	18·2	22·2	18·6	17·2	13·6	22·7	53·5	45·3	59·4	82·6
Wt. of placenta: Chorion	—†	1·056	1·553	1·030	0·910	1·210	1·672	3·860	2·790	3·810	5·098
Amnion	—†	0·421	0·464	0·297	0·412	0·213	0·580	1·180	0·685	1·027	1·805
Chord	—†	0·090	0·077	0·080	0·088	0·259	0·107	0·257	0·203	0·184	0·405
Total	1·513	1·567	2·094	1·407	1·410	1·682	2·259	5·297	3·678	5·021	6·308
Wt. of foal and placenta	21·0	19·8	24·3	20·0	18·6	15·3	25·0	58·8	49·0	64·4	88·9
$\dfrac{\text{Wt. of placenta}}{\text{Wt. of foal and placenta}} \times 100$	7·2	7·9	8·6	7·0	7·6	11·0	8·8	9·0	7·5	7·8	7·1
As percentage of weight of dam											
Birth wt. of foal	9·4	12·8	12·0	8·1	7·5	6·0	12·2	6·7	5·7	7·4	10·2
Foetal membranes	0·7	1·1	1·1	0·6	0·6	0·7	1·2	0·7	0·5	0·6	0·8
Foal and membranes	10·2	13·9	13·1	8·7	8·1	6·7	13·4	7·4	6·2	8·0	11·0
Group means											
Wt. of dam	190·7				214·1			797·0		(800)	
Wt. of foal	19·6				17·8			49·4		71·0	
$\dfrac{\text{Wt. of foal}}{\text{Wt. of dam}} \times 100$	10·3				8·3			6·2		8·9	

* From normal matings.   † Not recorded.

obtain for experiment were about average size and not the smallest pro-
curable, yet even so there is a very great difference in size between a pony
weighing 200 kg. and a horse weighing 800 kg. Exact weights of the
Shire mares were not available, but the mare used in the experiment
weighed 797 kg., and 800 kg. or 16 cwt. is recognized as about average
weight.

Weight of foals. It will be seen that the weights of the cross-bred foals
out of the Shetland dams are not significantly different from the weights of
pure Shetland foals. The weights of the reciprocal cross are smaller than
the weights of the pure Shire, but obviously there are not sufficient data to
show whether this is significant, although it may be so. Of the pure Shires,
one was small for the breed and the other large. Mr A. G. Holland,
Secretary of the Shire Horse Society, informs us that the weights of good
pure-bred Shire foals at birth are between 68 and 77 kg. (150 and 170 lb.).
It is obvious, however, that in each reciprocal cross there has been very
complete regulation of the size of the foal, the foals in the small mothers
being limited in size to that of Shetland offspring and those in the large
mother growing unchecked, although perhaps not quite reaching the full
size of a Shire foal. When the weights of the foals relative to the weights
of the dams are compared it will be seen that the percentage weights are
not significantly different, except possibly that the cross-breds from the
Shire dam are below those of the other groups. The weights of all foals are
about 8 % of the weights of the dams.

Weights of placentas. The placenta of the horse is non-decidual so that
the afterbirth represents only foetal material. Up to birth the placenta is
an integral part of the whole animal of which it constitutes about 8 %.
Since the placenta is an organ of foetal nutrition and may also have an
endocrine function, its size may be an important correlate of foetal size and
development.

POSTNATAL GROWTH IN WEIGHT

All the data for postnatal growth are given in Table II. It will be seen
that within the two groups for which we have more than one representative
there is fairly close agreement. For the purpose of our analysis, however,
we shall confine attention to the three animals—S 6, D 1 and D 2—repre-
senting respectively pure Shetland, Shetland-Shire cross from Shire dam,
and Shetland-Shire cross from Shetland dam. All three foals are of the
same sex (female) and were born in the same year, within a period of about

TABLE II. WEIGHTS AND GROWTH RATES OF FOALS (ALL FEMALES)

S 50 Pure Shetland

Age in months (4 weeks)	Weight in kg.	Growth rate per month	Relative growth rate % per month
0	18·2		
		24·4	85·1
1	42·6		
		11·0	23·0
2	53·6		
		15·8	25·8
3	69·4		
		10·9	15·8
4	81·3		
		7·6	8·9
5	88·9		
		4·6	5·1
6	93·5		

S 6 Pure Shetland

Age in months (4 weeks)	Weight in kg.	Growth rate per month	Relative growth rate % per month
0	18·1		
		18·2	69·6
1	36·3		
		12·3	29·2
2	48·6		
		10·9	20·2
3	59·5		
		17·7	26·0
4	77·2		
		9·5	11·6
5	86·7		
		2·3	2·6
6	89·0		
		3·6	4·0
7	92·6		

Calendar months	Weight in kg.	Growth rate per month	Relative growth rate % per month
0	18·1		
		11·3	29·5
5·4	89·0		
		6·6	6·1
11·0	125·8		
		6·1	4·5
16·6	160·7		
		− 2·0	− 1·3
20·7	152·5		
		11·9	6·4
27·0	227·2		
		− 3·3	− 1·5
31·8	211·3		
		8·2	3·6
37·1	255·0		

D 2 Cross-bred from Shetland dam

Age in months	Weight in kg.	Growth rate per month	Relative growth rate % per month
0	17·2		
		22·7	84·2
1	39·9		
		14·5	31·0
2	54·5		
		17·7	28·2
3	72·1		
		9·2	12·0
4	81·3		
		9·5	11·1
5	90·8		
		10·4	10·9
6	101·2		
		11·4	10·7
7	112·6		

Calendar months	Weight in kg.	Growth rate per month	Relative growth rate % per month
0	17·2		
		15·3	32·2
5·5	101·2		
		7·3	6·1
11·1	142·1		
		13·2	7·6
16·4	212·0		
		− 3·7	− 1·7
22·0	191·1		
		16·9	7·0
28·3	297·8		
		− 4·8	− 1·7
33·1	274·9		
		10·3	3·4
38·4	329·4		

D 1 Cross-bred from Shire dam

Age in months	Weight in kg.	Growth rate per month	Relative growth rate % per month
0	53·5		
		43·6	59·6
1	97·1		
		27·7	15·1
2	124·8		
		23·1	27·0
3	147·9		
		26·4	16·4
4	174·3		
		− 0·9	− 0·5
5	173·4		
		16·8	9·3
6	190·2		
		23·2	11·5
7	213·4		

Calendar months	Weight in kg.	Growth rate per month	Relative growth rate % per month
0	53·5		
		24·9	23·1
5·5	190·2		
		9·2	4·5
11·0	240·6		
		10·6	3·9
16·6	300·1		
		− 5·4	− 1·9
22·2	269·7		
		24·0	7·1
28·5	421·0		
		− 8·6	− 2·2
33·3	379·5		
		14·8	3·6
38·6	458·0		

D 3 Cross-bred from Shire dam

Age in months	Weight in kg.	Growth rate per month	Relative growth rate % per month
0	45·7		
		33·2	54·6
1	78·9		
		35·3	37·0
2	114·2		
		21·8	17·5
3	136·0		
		12·7	8·9
4	148·7		
		− 0·5	− 0·3
5	148·2		
		62·6	35·2
6	210·8		
		− 4·6	− 2·2
7	206·2		

Calendar months	Weight in kg.	Growth rate per month	Relative growth rate % per month
0	45·7		
		28·0	26·4
5·8	210·8		
		9·7	4·8
10·8	268·0		
		11·3	3·8
15·8	324·4		

6 weeks. They were on pasture together with their mothers until weaned at 4 months. During the rest of their lives they have been grazed together in the open, getting only grass and in winter some hay. Although losing weight during winter they have never fallen out of condition and have been perfectly healthy. Each foal was weighed at 4-week intervals from birth up to 28 weeks, i.e. over the period of most rapid growth. The foals were then weighed when about 11 calendar months old and at approximately 6 months interval after that. The biennial weighings were so arranged that one fell about August and the other about March in order that full effect might be given to the differences in nutrition during spring and early summer when there is a plentiful supply of grass, as contrasted with autumn and winter when the nutritive value of the pasture is low and grazing has to be supplemented with hay.

Growth curves of absolute weight (kg.), monthly gains (kg./month) and percentage monthly increments [$100 \log_e 2 - \log_e 1$] are shown in figs. 1, 2 and 3 respectively for the early period of growth. From these graphs it will be seen that D 1, the cross-bred from the large mother, grew rapidly, putting on large monthly gains. This rapid growth may be due in part to the plentiful supply of milk from the large mother. We have no actual measurement of the milk flow, but we assume that it would be sufficient to maintain a normal Shire foal. That the growth of the foal was to some extent dependent upon the milk supply of the dam is shown by the rapid drop in the growth curve which follows weaning at 4 months. Unfortunately, we have no actual measurements of Shire foals with which to compare the growth of D 1, but even in this early period it was apparent that the foal was growing less rapidly than a normal Shire. Fig. 4 gives some data on Shire growth, and from this curve it is obvious that D 1 diverges from the normal Shire curve at an early stage. In contrast to D 1 the cross-bred from the small mother grew less rapidly, and this was possibly due to some extent to the limitation in the milk supply from the small mother. The foal did, however, grow more rapidly than S 6 (pure Shetland), although it was receiving approximately the same nutrition. During lactation therefore the growth of the foals is not entirely determined by nutrition. Genetic differences make their appearance—D 1 grows less rapidly than a pure Shire foal although receiving the same nutrition. D 2 grows more rapidly than S 6 (pure Shetland), although both are suckled by dams of the same size. This genetic effect is of course not independent of nutrition. Growth in bulk can only be obtained from the food supply, but apparently genetic differences determine the amounts of the food which will be assimilated and retained for the growth of additional tissues.
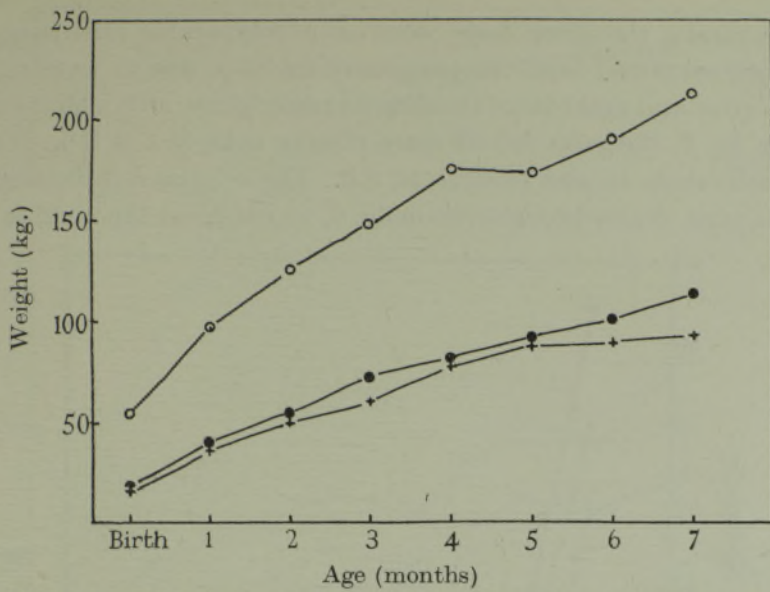
FIG. 1. Early growth of foals. ○ Shetland-Shire cross from Shire dam. ● Shetland-Shire cross from Shetland dam. + Pure Shetland.
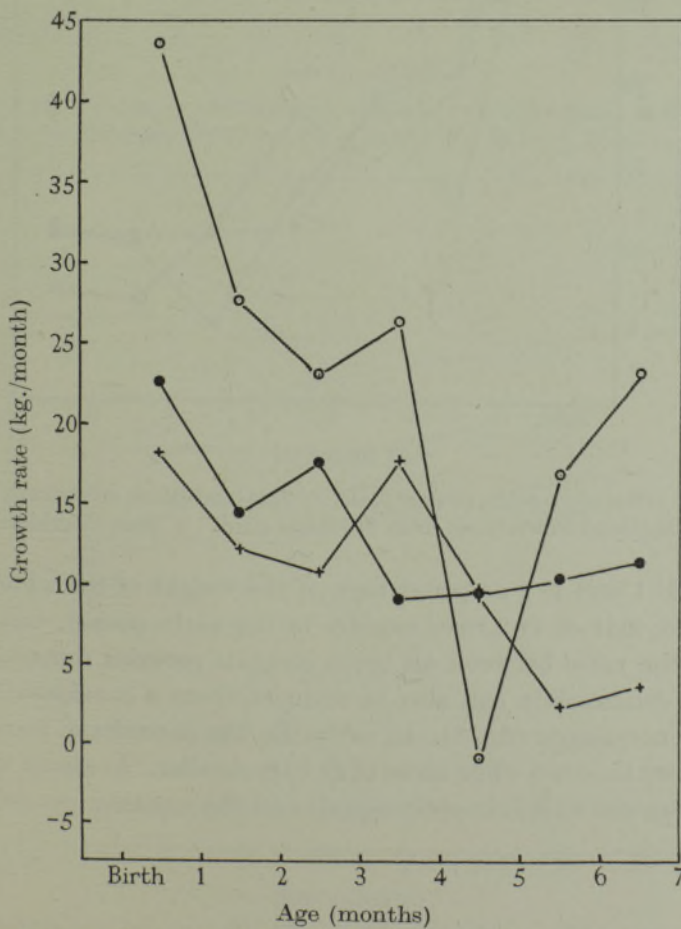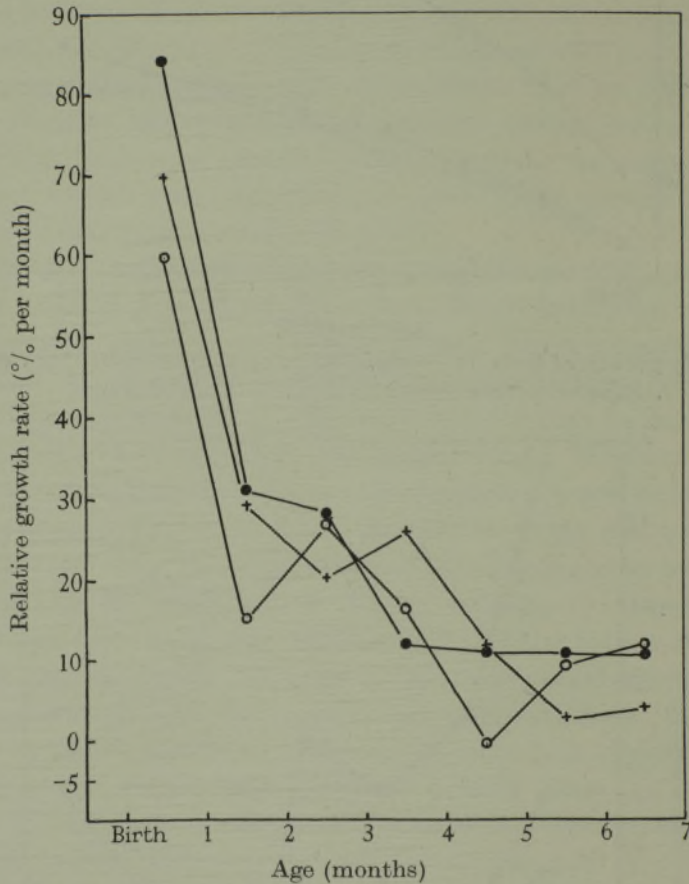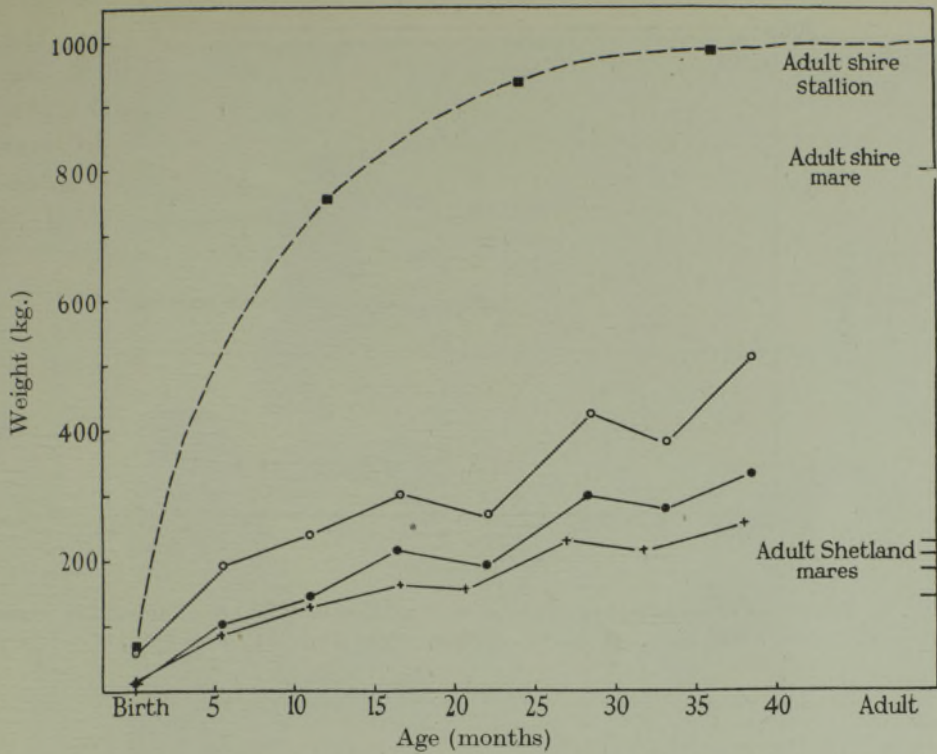

FIG. 2. Early growth rates of foals. ○ Shetland-Shire cross from Shire dam. ● Shetland-Shire cross from Shetland dam. + Pure Shetland.

After weaning the three foals were on a comparable nutrition. D 1, after it had recovered from the temporary set-back due to weaning, continued to grow and make large absolute monthly gains, although, as will be seen from fig. 2, the gains fell off more rapidly than D 2 or S 6. D 2 continued to increase in size relative to S 6. The relationship between the animals is most clearly brought out in fig. 6, which shows the weights of the

FIG. 3. Early relative growth rates of foals. ○ Shetland-Shire cross from Shire dam. ● Shetland-Shire cross from Shetland dam. + Pure Shetland.

cross-breds D 1 and D 2 as percentage of the weight of S 6. The curve of D 1 falls and that of D 2 rises rapidly in the early period, but at about 18 months the ratio between all three animals remains remarkably constant. This relationship can also be deduced from a consideration of the percentage increments (fig. 8). In early life the percentage increments of D 2 are greater than S 6 while those of D 1 are smaller. At about 18 months, however, they are approximately equal, and the equality persists in spite

FIG. 4. Growth of foals. ○ Shetland-Shire cross from Shire dam. ● Shetland-Shire cross from Shetland dam. + Pure Shetland. ■ Pure Shire (stallion).
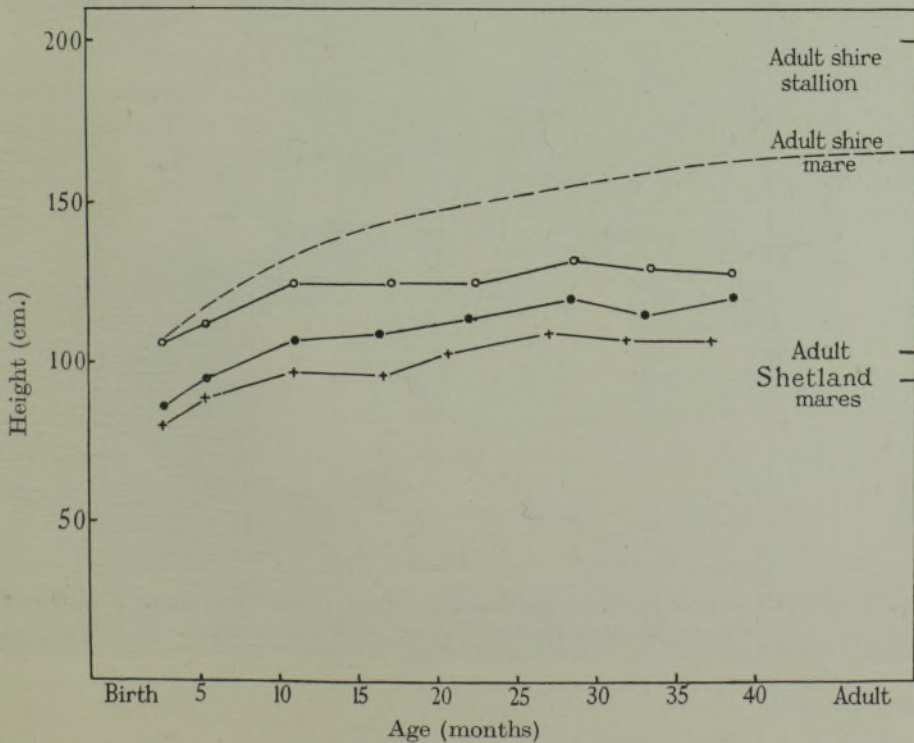

FIG. 5. Growth in height of foals. ○ Shetland-Shire cross from Shire dam. ● Shetland-Shire cross from Shetland dam. + Pure Shetland.
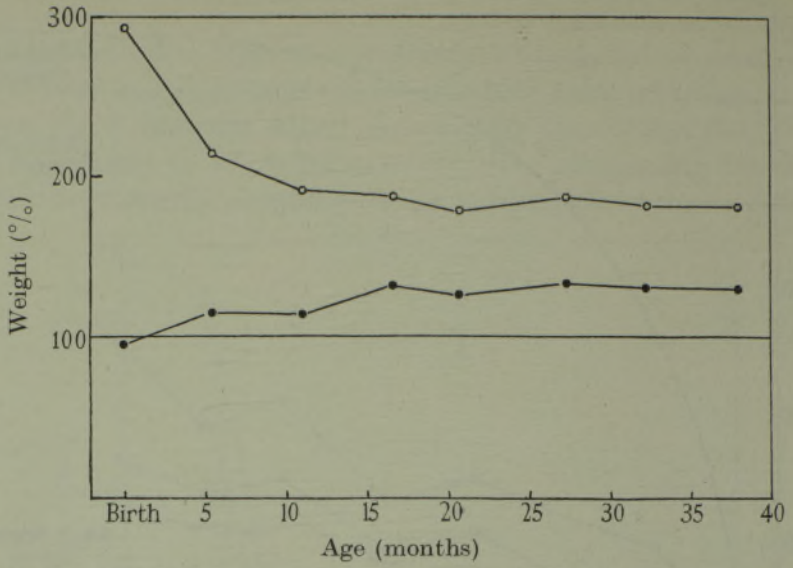
FIG. 6. Weight of cross-breds as per cent of Pure Shetland. ○ Shetland-Shire cross from Shire dam. ● Shetland-Shire cross from Shetland dam.
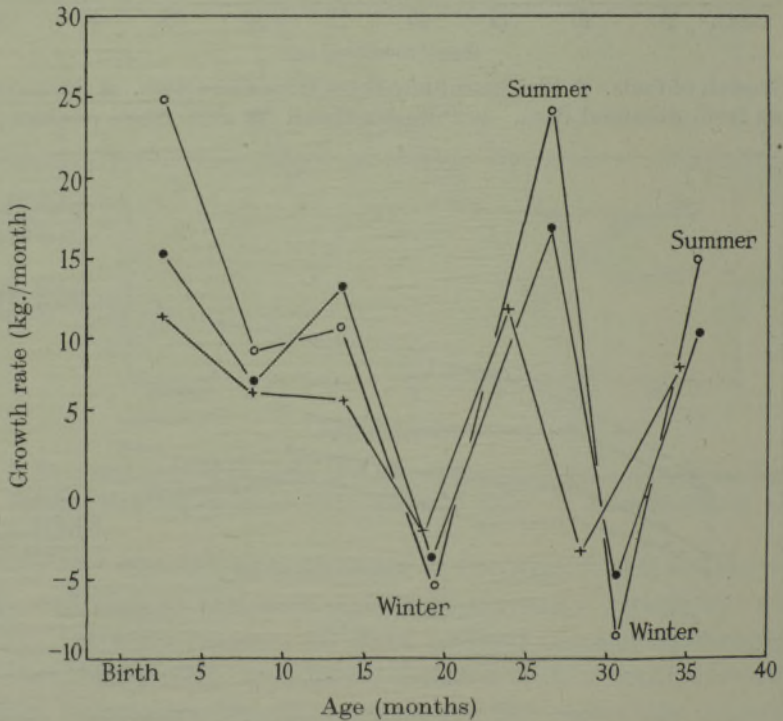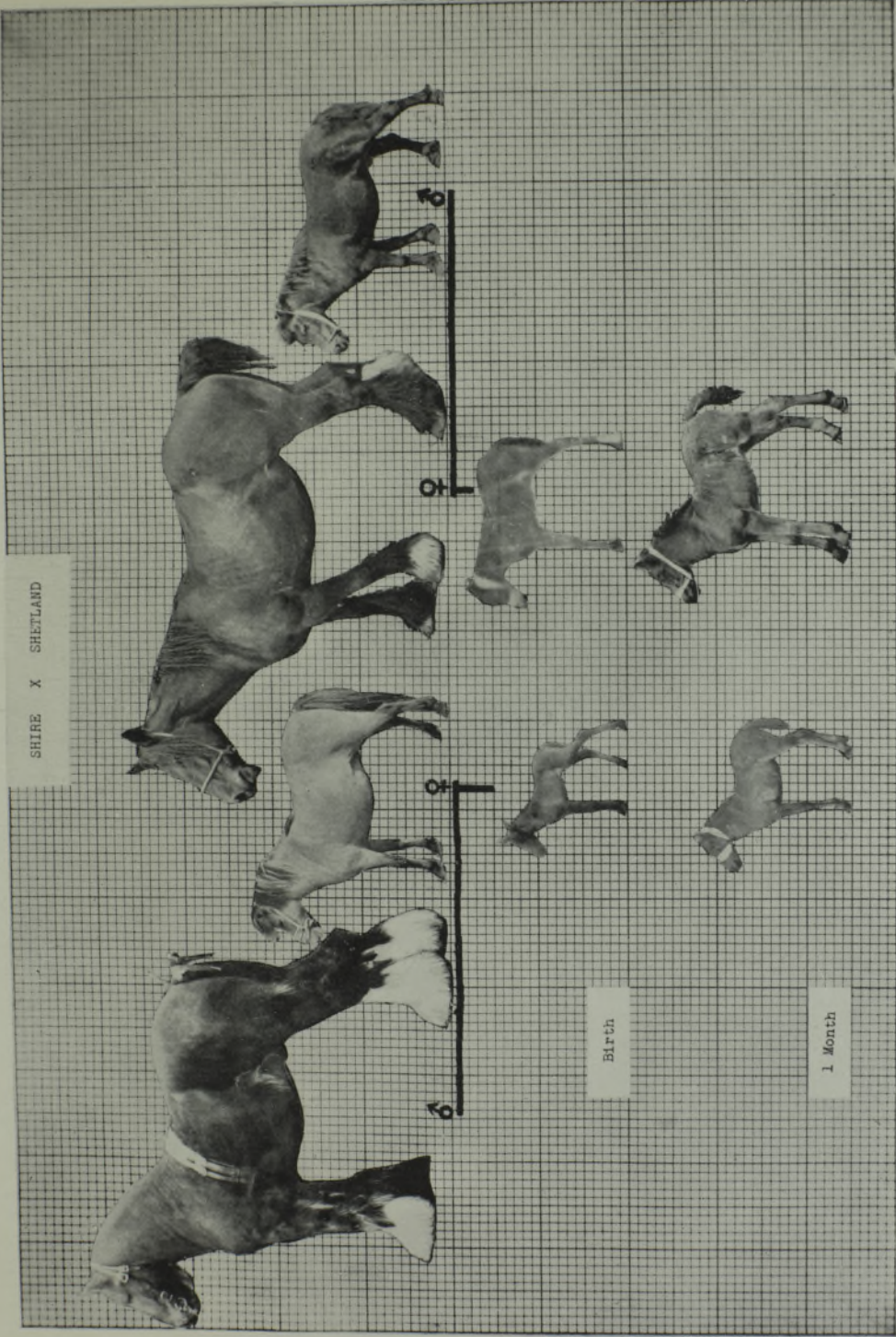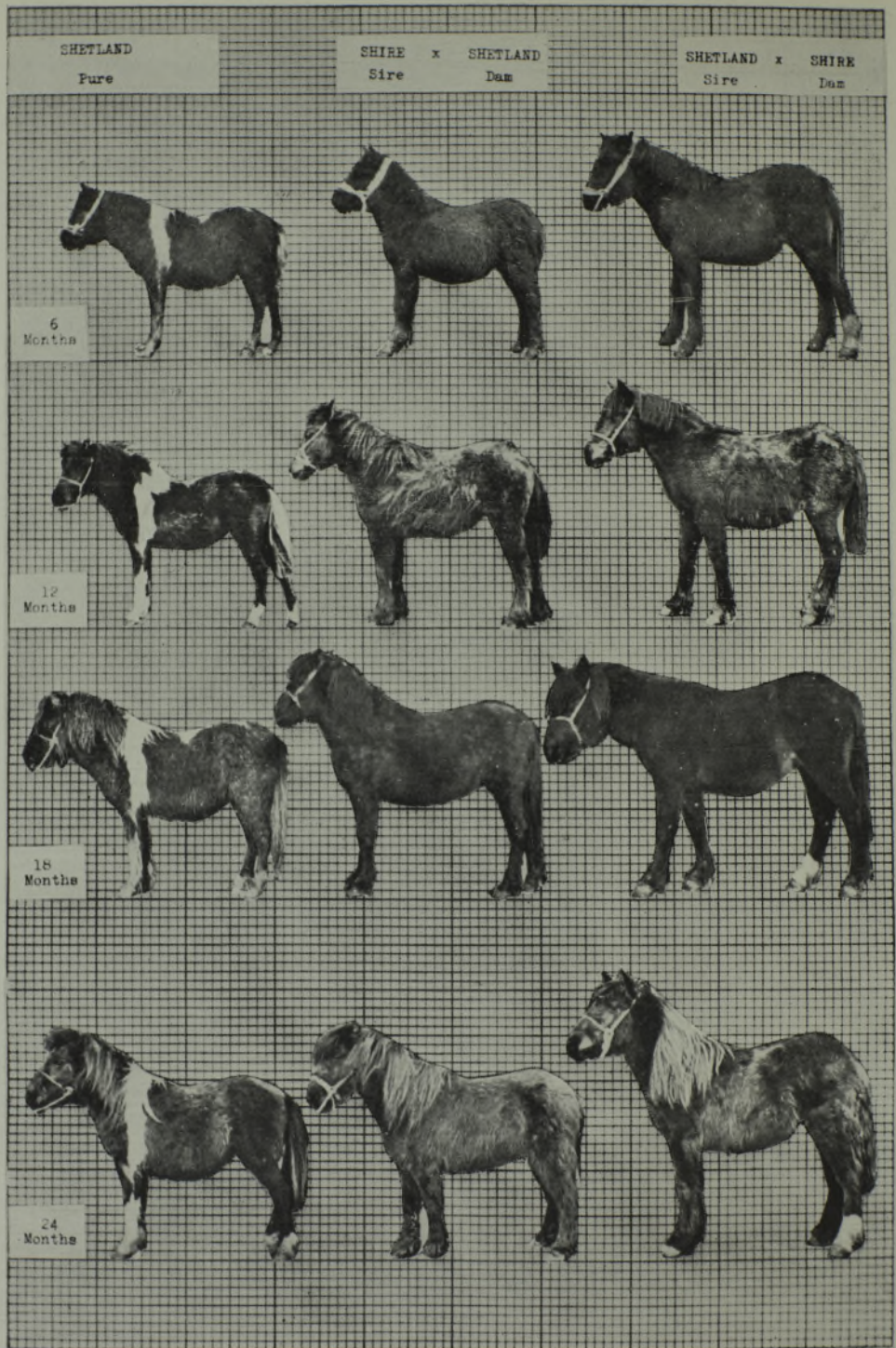

FIG. 7. Growth rates of foals. ○ Shetland-Shire cross from Shire dam. ● Shetland-Shire cross from Shetland dam. + Pure Shetland.

SHIRE X SHETLAND

Birth

1 Month

Parents and offspring of reciprocal Shetland-Shire crosses.

Growth and changes in body conformation of pure Shetland and reciprocal
Shetland-Shire crosses.

of the considerable growth that is still being made, and the extreme
fluctuation in absolute value of the increments occasioned by seasonal
variation in nutrition as shown both by the curves of absolute weight
(fig. 4) and monthly gains (fig. 7). What this implies, in other words, is
that from about 18 months onwards growth was not influenced by genetical
differences and was proportional to the size of the animal already established
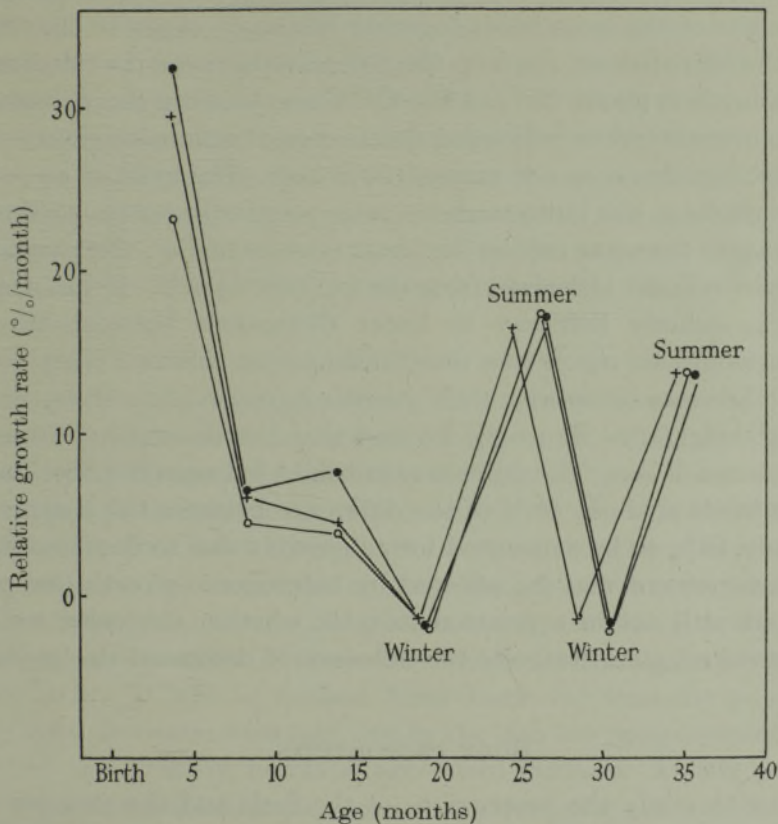although subject to considerable fluctuation in nutrition.

FIG. 8. Relative growth rates of foals. ○ Shetland-Shire cross from Shire dam.
● Shetland-Shire cross from Shetland dam. + Pure Shetland.

The ultimate size which the three foals will attain will not be known for
another year or so, but it is obvious from the growth curves that there will
be relatively little change in that time and that the ultimate weight is
already nearly attained. The pure Shetland, for example, at 3 years old
weighs 255 kg., which is above the weight of any of our adult Shetland
mares.

In fig. 4 the growth curves of pure Shire are shown for the purpose of

comparison. The figures for stallions were given us by Mr A. G. Holland, Secretary of the Shire Horse Society, as being exceptionally good weights. The figure for the mare is that of the mother of D 1. We have not had an opportunity of making more extensive measurements, and the data and the curves should be regarded as only rough approximations. They serve their purpose for making a rough comparison. Now if we compare the weights of the cross-breds with those of the parents we see that while the weight of the cross-bred from the large mother is nearly intermediate, the mean weight of the cross-breds together lies much closer to the Shetland than to the Shire parent. In fact, the difference between the Shetlands and the cross-breds is about 25 % of the difference between the parents. This might be interpreted as indicating dominance of small-size genetic factors over large, but this does not necessarily follow. The bulk of a solid body which increases in size but retains the same relative linear proportions does not increase at the same rate as the linear measurements. For example, the volume of a cylinder is derived from the formula $v = \pi r^2 h$. Hence a cylinder which was exactly half-way in linear dimensions between two other cylinders would be much less than half-way in volume. Now when we compare the cross-breds with their parents in respect to a linear measurement, e.g. height (see figure 8), we find that the discrepancy from intermediacy is much less. The difference in height between the Shetlands and the cross-breds is about 40 % of the difference between the parents. This leaves only 10 % to be accounted for as possibly due to dominance, but a linear measurement may be affected by heterogonic growth (see p. 325), and we are still not in a position to state whether the value we obtain represents at all quantitatively the influence of dominant size genes.

BODY PROPORTIONS

In order to study the proportions of the foals and the changes during postnatal growth a number of measurements were taken at intervals. Linear measurements taken on the surface of the body of a living animal are neither so accurate nor so discriminating of growth differences as measurements by weight. As has been pointed out earlier, the weight of a body roughly cylindrical and maintaining approximately the same linear proportions increases geometrically where a linear measurement would increase arithmetically, but in addition to this there are several sources of error involved in getting the animal to stand still in exactly the same position, in defining exact points of reference on the surface of the body from which measurements are taken and in variation due to thickness of hair,

dirt, and so forth. The data are recorded in Table III. Height at withers is the vertical distance from the crest of the withers to the ground with the animal standing four-square. It was taken with a measuring rod with horizontal side arm. Heart girth is the circumference of the trunk, just behind the forelimb. Length of body was measured from muzzle to poll, poll to withers and withers to root of tail. Each measurement was recorded separately, but the sum only is shown in the table. Similarly, length of forelimb was measured from withers to point of shoulder, point of shoulder to elbow, elbow to knee, knee to fetlock and fetlock to ground. Similarly, the hind-limb was measured from middle of rump to stifle, stifle to hock, hock to fetlock, fetlock to ground. Right and left sides of the body were measured separately. The figures in the tables represent the mean of the two sides. In order to determine the fineness of bone, measurements of the circumference of the forecannon, knee, hindcannon and hock were taken on each side of the body and the means recorded in the table. As a measure of comparative fineness all four measurements are added and related to the length of hindcannon.

As a measure of the "stockiness" of the animal the heart girth in relation to height at withers may be taken. The ratio increases in all animals from about 110 % at 3 months to about 150 % in the adult. In the larger animals it is slightly greater. In thickness of bone there is also a steady increase with age, and again the larger animal has the thicker bone. The cross-breds therefore tend towards the respective proportions of their mothers, but since the size of the animal affects also the proportions, it is not clear whether size or specific genetic influence is responsible. More exact definition is possible in the case of the ratio of length of limbs and length of body. It will be noticed from Table III that the percentage length of limb decreases with age, but in the last few measurements there is a distinct rise. This is, however, not a real increase. As the body fills out and the muscles develop the tape laid on the surface of the limbs from point-to-point becomes more convex and a systematic error is introduced increasing apparently the measurement. For this reason in the analysis which follows only measurements up to 2 years can be relied upon to give an exact picture of growth.

The relationship between the relative growth of the limbs and body is most clearly demonstrated by logarithmic plotting. Fig. 9 shows the length of forelimb plotted against body length and fig. 10 length of hind-limb also against body length. It will be seen that relative growth follows approximately the simple heterogonic formula of Huxley (1932) ($y = bx^K$), indicating that there is a constant ratio between the rate of growth of the

TABLE III. MEASUREMENTS (IN CM.) AND PROPORTIONS OF THE BODY

S 6

Age (calendar months) ...	2·7	5·4	11·0	16·6	20·7	27·0	31·8	37·1
(a) Height at withers	80	89	97	96	103	109	107	107
(b) Heart girth	85	100	111	124	120	137	135	155
Ratio $b/a \times 100$	106	118	114	129	117	126	126	145
(c) Length of body	142	162	177	199	192	197	206	197
(d) Length of forelimb	110	121	129	138	139	147	151	153
Ratio $d/c \times 100$	77	75	73	69	71	75	73	78
(e) Length of hindlimb	109	117	122	134	132	140	144	143
Ratio $e/c \times 100$	77	72	69	67	69	71	70	73
(f) Min. circumference of forecannon	11	13	13	14	14	15	16	15
(g) Circumference of knee	18	20	21	22	22	22	24	24
(h) Min. circumference of hindcannon	12	13	14	15	15	16	17	17
(i) Circumference of hock	22	25	25	27	25	28	29	27
(j) Length of hindcannon	28	29	29	30	31	31	31	31
Ratio $(f+g+h+i)/j \times 100$	225	245	252	260	245	262	277	274

D 2

Age (calendar months) ...	2·8	5·5	11·1	16·4	22·0	28·3	33·1	38·4
(a) Height at withers	86	95	107	109	114	120	115	120
(b) Heart girth	92	107	123	144	134	162	152	175
Ratio $b/a \times 100$	107	113	115	132	118	124	132	146
(c) Length of body	152	156	183	197	200	214	205	211
(d) Length of forelimb	118	130	142	151	157	161	167	170
Ratio $d/c \times 100$	78	83	78	77	79	75	81	81
(e) Length of hindlimb	111	125	133	146	147	155	157	161
Ratio $e/c \times 100$	73	80	73	74	74	72	77	76
(f) Min. circumference of forecannon	12	15	14	16	16	16	18	17
(g) Circumference of knee	20	22	23	24	24	24	25	26
(h) Min. circumference of hindcannon	12	15	15	17	16	18	20	18
(i) Circumference of hock	22	25	26	28	25	30	30	30
(j) Length of hindcannon	30	32	32	33	31	33	32	33
Ratio $(f+g+h+i)/j$	220	240	244	258	261	267	291	276

D 1

Age (calendar months) ...	2·7	5·5	11·0	16·6	22·2	28·5	33·3	38·6
(a) Height at withers	106	112	125	125	125	132	129	128
(b) Heart girth	117	134	142	161	141	174	182	188
Ratio $b/a \times 100$	110	120	114	129	113	132	141	147
(c) Length of body	178	188	210	226	222	236	239	237
(d) Length of forelimb	144	158	169	169	170	180	185	191
Ratio $d/c \times 100$	81	84	81	75	77	76	77	81
(e) Length of hindlimb	143	149	162	164	164	172	177	180
Ratio $e/c \times 100$	80	79	77	73	74	73	74	76

TABLE III (*continued*)

(*f*) Min. circumference of forecannon	16	18	19	19	19	20	22	20
(*g*) Circumference of knee	26	27	28	28	28	29	29	30
(*h*) Min. circumference of hindcannon	17	18	19	21	20	22	24	22
(*i*) Circumference of hock	29	33	35	34	31	36	37	36
(*j*) Length of hindcannon	38	38	41	38	41	39	40	39
Ratio $(f+g+h+i)/j \times 100$	232	253	246	269	239	274	280	277

D 3

Age (calendar months) ...	Birth	2·8	5·8	10·8	15·8
(*a*) Height at withers	90	96	115	121	124
(*b*) Heart girth	73	110	129	150	161
Ratio $b/a \times 100$	81	115	112	124	130
(*c*) Length of body	125	166	187	206	216
(*d*) Length of forelimb	120	144	156	170	173
Ratio $d/c \times 100$	96	87	83	83	80
(*e*) Length of hindlimb	116	139	151	161	167
Ratio $e/c \times 100$	93	84	81	78	77
(*f*) Min. circumference of forecannon	12	15	17	18	18
(*g*) Circumference of knee	21	24	26	28	28
(*h*) Min. circumference of hindcannon	12	16	18	20	20
(*i*) Circumference of hock	23	28	32	32	33
(*j*) Length of hindcannon	33	36	37	38	37
Ratio $(f+g+h+i)/j \times 100$	206	230	251	258	268

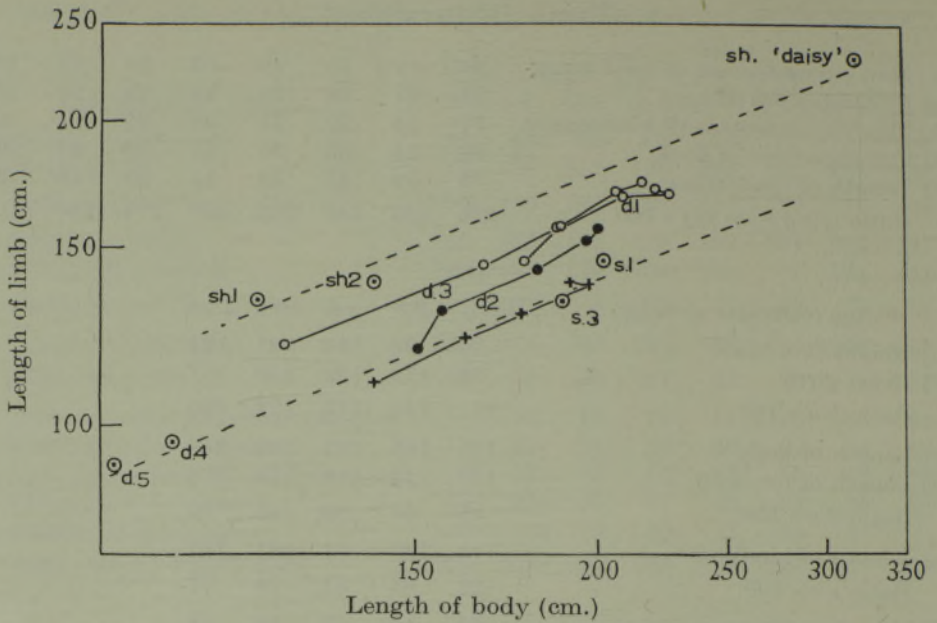|	S 1	S 3	D 4	D 5	"Daisy"	Sh 1	Sh 2
Age ...	Adult	Adult	Birth	Birth	Adult	Birth	Birth
(*a*) Height of withers	104	95	77	64	166	95	110
(*b*) Heart girth	151	113	55	43	210	83	94
Ratio $b/a \times 100$	145	119	71	67	126	87	85
(*c*) Length of body	202	187	109	102	318	122	143
(*d*) Length of forelimb	145	134	94	82	234	134	138
Ratio $d/c \times 100$	72	72	86	80	74	91	97
(*e*) Length of hindlimb	131	120	96	80	215	133	140
Ratio $e/c \times 100$	65	64	88	78	68	109	98
(*f*) Min. circumference of forecannon	14	12	10	7	29	14	17
(*g*) Circumference of knee	21	19	16	13	41	26	29
(*h*) Min. circumference of hindcannon	14	13	10	8	34	15	17
(*i*) Circumference of hock	25	23	19	16	49	31	36
(*j*) Length of hindcannon	29	29	28	24	48	37	41
Ratio $(f+g+h+i)/j \times 100$	255	231	196	183	319	232	241

FIG. 9. Relative growth of forelimb to body length, (logarithmic plotting). ○ Shetland-Shire crosses from Shire dams. ● Shetland-Shire crosses from Shetland dams. + Pure Shetland. ⊙ Single point observations.
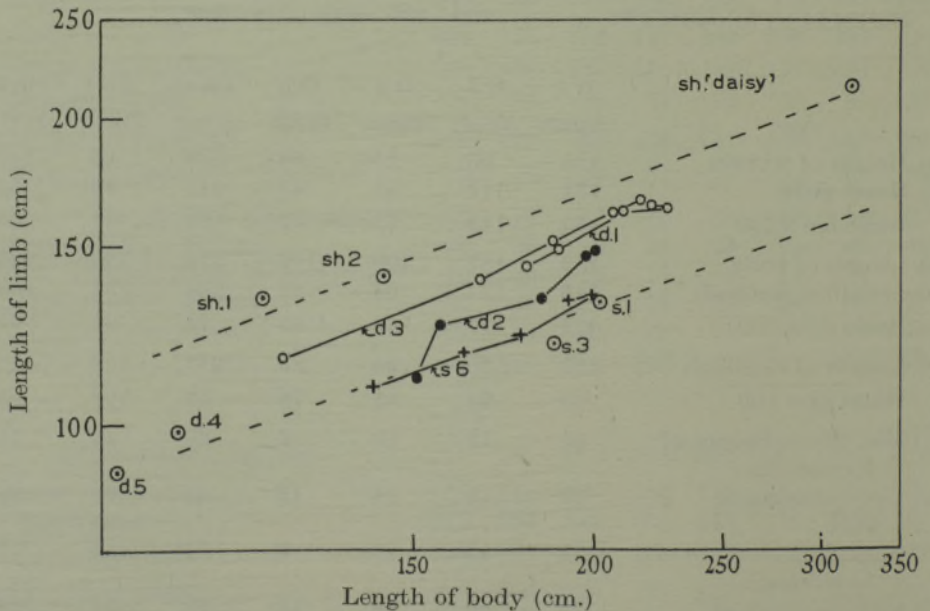

FIG. 10. Relative growth of hind limb to body length (logarithmic plotting). ○ Shetland-Shire crosses from Shire dams. ● Shetland-Shire crosses from Shetland dams. + Pure Shetland. ⊙ Single point observations.

limbs and the rate of growth of the body. K, the ratio constant which determines the slope of the curves, is approximately the same in all animals, the possible exception being D 2, the cross-bred from the small mother; this will be discussed later. There is, however, a considerable difference in the fractional constant b as denoted by the distance apart of the curves. In words this implies that the pure Shetland is relatively shorter in the limb than the pure Shires and the crosses are intermediate, and that this is true at any body length. For example, at body length 160 the values read from the graph are approximately: Pure Shetland, 121; D 1, 132; D 3, 140; Shire, 155 for the forelimb; and Shetland, 116; D 2, 126; D 3, 135; and Pure Shire, 155 for the hindlimb. With regard to the relative influence of genetic and nutritional factors it is to be noted that the cross-breds occupy an intermediate position between the Shetland and the Shire but are themselves different. D 2 at 3 months and D 4 and D 5 at birth, have a body limb ratio which falls on the pure Shetland curve, but at 6 months D 2 definitely reaches a higher level and follows a line distinctly above that of the pure Shetlands. Up to birth therefore the influence of maternal nutrition predominates, and not until after birth do genetic differences appear to affect the body limb proportions. On the other hand, in D 3 at birth, and in D 1 at 3 months, the proportions are not similar to those of the Shire but distinctly nearer the Shetland type. This shows that the proportions of the body have been affected by genetic differences acting before birth. Subsequently they follow the curve of heterogonic growth established at this early period.

DISCUSSION

In mammals parturition occurs at a time when the foetus reaches a size characteristic of the species and is still able to pass through the genital passage. In most species this size approaches the maximum and parturition is laboured and frequently critical. There is undoubtedly synchronization between the development of the foetus and the termination of pregnancy. The advantage of this synchronization to the animal is obvious and is no doubt an evolutionary adaptation. In the large and small mothers in these experiments the foetus at birth is approximately proportional to the size of the mother, and this proportion is not significantly altered in the reciprocal crossings. Although the offspring differ genetically from their respective maternal parents, as is shown by subsequent growth, at birth the size of the foetus is approximately that to which the mother would normally give birth. The degree of maternal regulation of size

in these experiments is very marked. It is, however, not absolutely complete; the cross-breds from the large mother are perhaps not quite so large at birth as normal shires, and the limbs are relatively shorter as seen from figs. 9 and 10. In the small mothers regulation appears to be more complete.

In agricultural practice there is a division of opinion whether the mating of large sires to small dams is liable or not to cause dystokia, although up to the present no critical experiments have been done to test the point. Possibly those cases in which difficulty at birth is experienced are to be attributed not so much to the general size of the offspring as to a disproportionate lack of regulation of some particular part which may give rise to difficulty at parturition, for example the heavy head and shoulders of the Bison cattle-crosses, or the width of skull in certain breeds of sheep and in bull dogs. Difficulty in parturition, although attributed to the size of the foetus, may be due to an abnormal pelvis or to lack of development of the maternal reproductive tract. For example, in the Dexter breed of cattle the "bull dog" monster calf is actually smaller than the normal calf of the same age, but, as it is usually aborted before term, parturition is difficult owing to lack of development of the mother's cervix and vagina.

The mechanism of maternal regulation must remain at present a matter for speculation. There is ample proof in the rabbit (Hammond 1935) that the size of the young is not limited by uterine space, for one horn of the uterus is capable of accommodating double the normal number of young, and at birth the young are not smaller than an equal number of young distributed throughout two horns. In species which are normally monotocous, multiple births are not uncommon, and in these cases the total content of the uterus exceeds that of a single birth, showing that considerable distention is possible. It does not appear probable that uterine surface and placentation are determining factors. In the rabbit experiments quoted above, the placentas were not smaller in the crowded uterus than normal. The size of the placenta is very closely correlated with the size of the foetus, but it is not affected by the space available in the uterus. The foetal placenta is an organ of the foetus and varies with the foetal weight, and is affected by the same changes as the foetus itself.

To look for the causes of maternal regulation, therefore, we must go beyond the spacial and anatomical features of the uterus and placenta. Regulation may take place through limitation in the supply of nutrient material to the foetus. On this hypothesis the growth of the foetus may be determined by the level of nutrient substances available, and this level in

turn may be a function of the size of the mother. This explains the well-known fact that in polytocous species the larger the litter the smaller is the individual young, as due to competition for a common food supply. The hypothesis is not upset by the fact that in large litters the total uterine content is greater than in small litters. It is a feature of nutrition that the law of diminishing returns operates so that the larger the litter and the smaller the young the more efficient will be the use made of available nutrients.

Against the hypothesis may be cited the fact that the growth of young *in utero* is not markedly affected by increasing or decreasing the supply of food given to the mother. This relative immunity of the foetus is, however, by no means absolute (see Hogan 1928). Furthermore, if the mother is fed on a low plane of nutrition the foetus will take priority of the available nutrients and deplete the food reserves in the mother's body. On the other hand, if the mother is well fed food reserves will be laid down. The availability of nutrients to the foetus may remain relatively constant whether the mother is gaining or losing weight.

So far we have discussed nutrition in very general terms, but we know that the organism requires a variety of special substances, some of which although small in amount have marked effects upon growth. The vitamins and growth hormones fall into this category. It is possible that regulation of foetal size is brought about through one, or more than one, of these substances. For example, it is possible that growth-promoting hormone of the maternal pituitary may be the regulating factor. This hypothesis is attractive as it at once suggests a mechanism by which the mother's own size and that of the foetus she bears may be co-ordinated.

Another possible explanation of maternal regulation may be cytoplasmic inheritance. It is possible that the ovum receives a quantity of some growth-regulating substance proportional to the size of the mother, and that this determines the size that the foetus will reach at birth. Marshak (1936) found evidence of cytoplasmic inheritance in reciprocal crosses in mice. Cytoplasmic inheritance, however, will not alone explain why after birth the growth rates of the crosses change so that the foals follow a different growth curve to that of the pure-bred foals of the respective maternal parents. This change can, however, be explained on the assumption that maternal regulation acts continuously on the foetus during uterine life and ceases on the termination of pregnancy. It is the uterine environment rather than factors inherent in the foetus which determines regulation.

After birth maternal regulation ceases, perhaps not entirely, since difference in milk supply from the large and small mother may still exert

some influence on the growth rate, but after weaning the foals are on the same plane of nutrition. The growth curves tend to converge. The relative increments of the foal from the large mother are less than the relative increments of the foal from the small mother. At about 18 months (see figs. 6 and 8) equilibrium is established and the relative increments remain constant, in spite of considerable fluctuation in absolute growth rates (see fig. 7) due to season variation. It is clear, however, that the differences induced by maternal regulation during pregnancy are not entirely lost, and that the foals are permanently affected, at least as far as the experiment to date carries us. This permanent influence of maternal regulation is not general in all animals (see Green 1931), possibly the long period of gestation, the relatively late stage of development at which the embryo is born and the relatively large difference in nutritional level between the large and small mothers may in this experiment have determined the permanency of the effect.

The marked influence of nutrition on growth which we see illustrated in these experiments is not without theoretical importance. Robertson (1923) and more recently Brody (1928) have made attempts to analyse the integral growth curve in terms of some fundamental intrinsic reaction. This assumes that the organism can be regarded as an isolate in a neutral environment, and that nutrition does not limit growth nor set the pace. This is clearly not the case in this experiment, and further evidence on this point is obtainable from the numerous nutritional experiments the object of which is to improve upon the ration and obtain maximum growth. Until this hypothetical perfect ration is obtained and applied throughout the growth period including pregnancy it cannot be shown that the growth curve is determined by intrinsic reactions and independent of the food supply.

With regard to the genetical implications of the experiment we see that at birth as far as weight is concerned the cross-bred young show little influence of their mixed heritage. Maternal regulation has masked the genetical differences, which, however, appear shortly after the maternal regulation is withdrawn. At about 18 months onwards also, growth is apparently proportional to the size of the animal, and genetical differences do not appear to affect the course of growth. The question of dominance of size factors has already been discussed (see p. 324). In regard to the proportions of the body it is clear that before any comparison of genotypes can be made the influence of size as such must be taken into consideration. Figs. 9 and 10 show that as far as fore and hindlimb lengths are concerned there is a simple heterogenic relationship with body length. The difference

of genotype appears however in the difference in the fractional coefficients. To the same body length the Shetland pony is shorter in the limb than the Shire, and the reciprocal crosses are intermediate. The influence of nutrition is not so apparent as in the case of the weight curves. D 2, D 4 and D 5 (foals from the small mothers) perhaps show maternal regulation at birth, and D 2 shows a "genetic" tendency away from the maternal parental type at 6 months. D 3 and D 1 (foals from the large mother) show no definite signs of maternal regulation. The difference between D 2 on the one hand and D 1 and D 3 on the other, is, however, a nutritional effect if our original postulate of genetic similarity (see p. 312) is allowed. The fact that body proportions (bone growth) behaves differently from weight (muscle growth) may possibly be related to the difference in time of maximal development of these two tissues. This experiment well illustrates the interplay of genetic and nutritional factors which are involved in the development of the animal. When offspring are obtained from the cross-bred foals it is hoped that the analysis may be carried further.

SUMMARY

1. Reciprocal crosses between the large Shire horse and the small Shetland pony have been made by means of artificial insemination.

2. At birth the foals were approximately proportional in weight to the weights of their mothers and about equal to foals of the pure breeds to which the mothers belonged. The cross-foals from the Shire mare were three times the size of the cross-foals from the Shetland mares. Maternal regulation of foetal growth was very marked and obscured any genetic differences.

3. After weaning, when the foals were under the same nutritive conditions, genetic differences appeared. The foals from the Shire mares grew much less rapidly than pure Shire foals, and the foals from the Shetland mares grew much more rapidly than pure Shetlands. At about 18 months

an equilibrium point was reached at which the relative growth rates of the cross-foals and the pure Shetland remained constant. At 3 years the difference between the reciprocal crosses is still marked and is apparently permanent.

4. Differences in the proportions of the animals, when size differences are eliminated, were not so marked as differences in weight and the influence of nutrition not so obvious.

5. The mechanisms, by which maternal regulation may be brought about, are discussed and three possibilities suggested: (*a*) Maternal regulation of foetal nutrition; (*b*) Maternal hormonal control; (*c*) Cytoplasmic inheritance.

6. The bearing of these results on the theoretical concept of growth is discussed.

7. The experiments illustrate the interplay of nutritional and genetical factors which are involved in development.

REFERENCES

Brody, S. 1928 See Robbins *et al.*
Green, C. V. 1931 *J. Exp. Zool.* 59, 213.
Gregory, P. W. 1930 *Carnegie Inst. Washington Publication*, no. 407, p. 141.
Hammond, J. 1934 *a J. Exp. Biol.* 11, 140.
— 1934 *b Act 16ème Congr. int. Agric., Budapest.*
— 1935 *Trud. Dinam. Razvit*, 10, 95.
Hogan, A. G. 1928 See Robbins *et al.*
Huxley, J. S. 1932 "Problems of Relative Growth." London.
Imperial Bureau of Animal Genetics 1933 "The Technique of Artificial Insemination." Edinburgh.
Marshak, A. 1936 *J. Exp. Zool.* 72, 497.
Meek, A. 1901 *Veterinarian*, 74, 8.
Plumb, C. S. 1916 "Types and Breeds of Farm Animals." Boston.
Robbins, W. J. *et al.* 1928 "Growth." Yale and London.
Robertson, T. B. 1923 "The Chemical Basis of Growth and Senescence." Philadelphia.
Walton, A. 1936 "Notes on Artificial Insemination of Sheep, Cattle and Horses." London (2nd ed. 1938).
Wishart, J. and Hammond, J. 1933 *J. Agric. Sci.* 23, 463.

DESCRIPTION OF PLATES

Plate 14. Parents and offspring of reciprocal Shetland-Shire crosses (all to same scale).

Top line	Parent	Shire ♂ × Shetland ♀	Shire ♀ × Shetland ♂
Middle line	Offspring at Birth	Crossbred foal D 2	Crossbred foal D 1
Bottom line	Offspring at 1 month	Ditto	Ditto

The animals were photographed against a squared background, and prints made with the squares enlarged to the same size. Each animal was then cut out from the prints and all mounted on squared paper and rephotographed. Relative sizes and body proportions can be compared.

Plate 15. Growth and changes in body conformation of pure Shetland, and reciprocal Shetland-Shire crosses (all to same scale).

	Pure Shetland (S 6)	Cross-bred from Shetland dam	Cross-bred from Shire dam
Top line 6 months	,,	,,	,,
2nd line 12 months	,,	,,	,,
3rd line 18 months	,,	,,	,,
4th line 24 months	,,	,,	,,

Method of photography and reproduction as in Plate 14.

The secretion of protein material in the parasympathetic submaxillary saliva

By G. O. Langstroth, Ph.D., D. R. McRae, Ph.D.

Research Associates, Department of Physics,

and G. W. Stavraky, M.D.

Research Associate, Department of Physiology,

McGill University, Montreal

This paper deals with a study of the secretion of protein material by the submaxillary gland of the cat in response to electrical stimulation of the chorda tympani nerve. In the attempt to make a more generous use of certain advantages offered by physical methods in research on the digestive glands, the submaxillary gland was chosen for the initial investigation because of the relative ease of physiological experimentation. The study is characterized by (1) the use of quantitative spectroscopic methods of analysis which permit an extension in the range of the investigation over that possible with a purely chemical procedure, and (2) the application of mathematical analysis in the interpretation of the results.