GENETICAL STUDIES ON THE SKELETON OF THE MOUSE
V. 'INTERFRONTAL' AND 'PARTED FRONTALS'

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(With Four Text-figures)

Introduction

The name 'interfrontal' was given to a small bone between the frontals which occurred commonly in two mouse strains bred at the Bussey Institution (Keeler, 1933). It was also found in a wild female caught in a Boston house which, mated to a normal male, produced seven normal offspring. As, in F_2, there were six normal and two abnormal young, Keeler concluded that 'the character cannot be dominant because it disappeared completely in the first generation. The second hybrid generation shows a perfect three-to-one ratio which is the recombination proportion expected if the character is determined by a single pair of recessive Mendelizing genes.'

Keeler (1930) had previously described a condition in the mouse skull which he called 'parted frontals'. It occurred commonly among the mice of his rodless retina stock and, on the basis of a pedigree of 100 animals, Keeler suggested that 'it is apparently inherited as a dominant unit character because (1) offspring of two affected parents are not all affected, (2) offspring of an affected parent and a normal of unrelated strain not bearing "parted frontals" may be all affected, some affected, or none affected. Only the last-named condition would be compatible with behavior as a recessive character.'

In view of the complex genetic basis encountered by Grüneberg (1952) in the case of several skeletal variants, it seemed of interest to reinvestigate the behaviour of these two minor skull variants in genetically homogeneous material. This became possible when both characters were rediscovered in the pure lines, C57 Black, CBA and A.

Interfrontal

The interfrontal bone occupies a position in the suture between the frontals anterior to the transverse sinus. It may articulate anteriorly with the nasals, as in Fig. 1B, but is sometimes completely surrounded by the frontals. The interfrontal bone differs from ordinary suture or Wormian bones by the regularity of its outline and position and by the fact that the sutures surrounding it are invariably smooth. Most CBA mice have a fairly large interfrontal bone which may be about 3 mm. long and 0·2–0·4 mm. wide. In some animals inspection from the dorsal side shows no interfrontal; some of these have a small interfrontal bone situated ventrally between the frontals; such animals would probably have been classified as normal by Keeler. Two groups of sublines from the C57BL stock which had been separated since 1941 (at least twenty generations) and which were known to differ in other skeletal features (Searle, unpublished), were examined. These two sublines differ from each other and from CBA; both of them having a lower incidence of interfrontal and the bone also tends to be smaller. In both sublines there is a tendency for the interfrontal to fuse secondarily with its neighbours (Fig. 1D); this has not been seen to happen in the CBA strain. Finally, in the A strain, dorsal inspection generally does not reveal the presence of an interfrontal at all, but inspection from the inside shows that some of the animals have hidden (ventral) interfrontal bones which are always rather small in size. A group of thirteen wild mice caught in an animal house of the Zoological Gardens London, lacked the interfrontal.

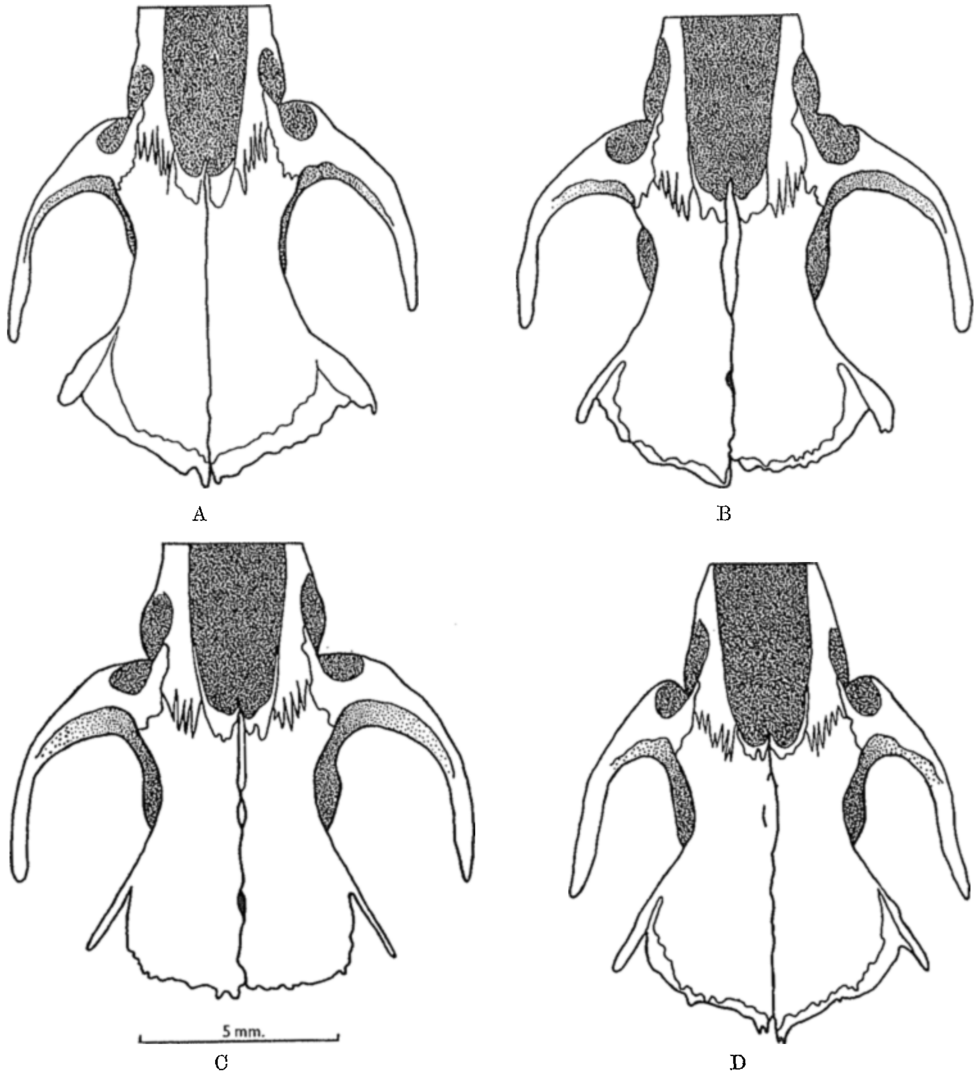

Fig. 1. Normal (A, wild mouse) and interfrontal (B–D). B, large interfrontal (CBA ♂). C, divided interfrontal, a rare variant (CBA ♂). D, interfrontal with two secondary fusions to the left frontal (C57BL ♂). In B there is a small, and in C a medium-sized fontanelle between the frontals ('parted frontals'). Papain preparations (nasals and parietals missing). Camera lucida drawings.

In sections through the heads of newborn animals, one can see that the interfrontal bone has no relation to the nasal septum. There is no doubt that the interfrontal is an independent entity (Figs. 2, 3), and that it is a membrane bone like the frontals which flank it.

The distributions according to sex and stock are given in Tables 1 and 2. While the sexes do not differ significantly as regards the interfrontal there are striking and highly significant

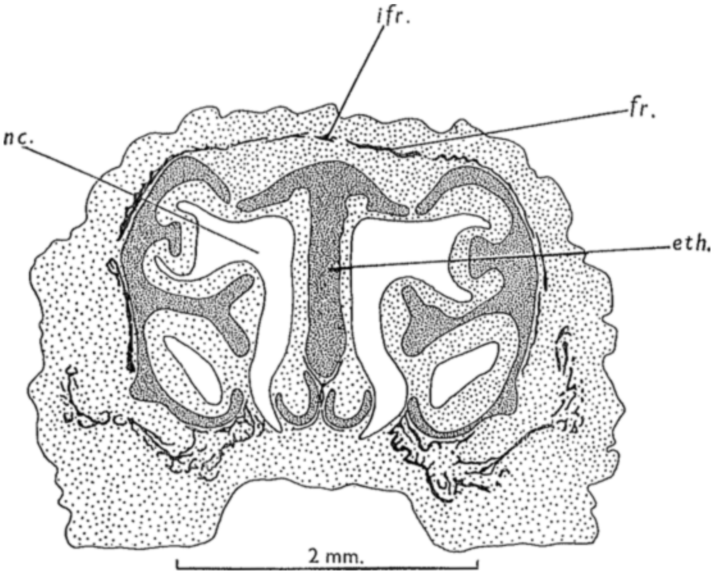

Fig. 2. Transverse section through the region of the interfrontal bone in a newborn CBA mouse. Note that the bone is completely independent of the cartilage of the nasal septum and clearly a membrane bone. Camera lucida drawing; bone spicules, black; cartilage, heavily stippled. *eth.* ethmoid cartilage in nasal septum; *fr.* frontal; *ifr.* interfrontal; *nc.* nasal cavity.

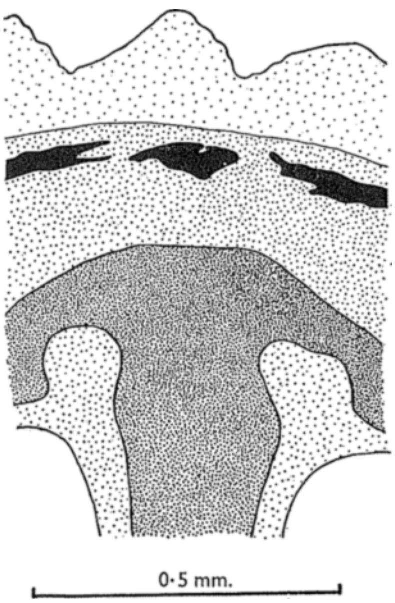

Fig. 3. The region of the interfrontal in Fig. 2 at a higher magnification. Camera lucida drawing.

differences between the pure lines (Table 2). The data for CBA and the two sublines of the C57 BL stock are broken down according to the mating type of the parents (interfrontal × interfrontal, interfrontal × normal, and normal × normal) in Table 3. In none of the stocks is there any evidence for a parent-offspring correlation. Hence it must be concluded that each of the three stocks (and presumably the A stock as well) breeds true to the production of its own characteristic incidence of interfrontal. Within each stock, presence or absence of the bone is thus determined by the environment; as the interfrontal is present at birth, the decision must have occurred during embryonic life.

Table 1. *Incidence of the interfrontal bone in the pure lines* CBA, C57 BL (*two different sublines*) *and* A. *Arbitrary size classes; the symbol* (+) *means that the interfrontal is entirely ventral*

Stock	Sex	+ + +	+ +	+	(+)	–	Total
CBA	♂♂	56	79	38	14	26	213
	♀♀	49	77	27	14	31	198
Total		105	157*	65	28	57	412*
C57 BL	♂♂	12	18	14	16	19	79
Sublines 3 and 4	♀♀	12	21	16	27	33	109
Total		24	39	30	43	52	188
C57 BL	♂♂	3	18	16	34	46	117
Sublines 1 and 2	♀♀	4	13	19	21	60	117
Total		7	31	35	55	106	234
A	♂♂	—	—	—	8†	59	67
	♀♀	—	—	—	5	39	44
Total		—	—	—	13	98	111

* Sex of one animal unknown.
† In one of these animals a little of the interfrontal could just be seen through the suture by dorsal inspection.

Table 2. *Interfrontal. Incidence in three inbred strains*

Stock	Normal	Interfrontal	Total	% interfrontal
CBA	57	355	412	86·2
C57 BL, sublines 3 + 4	52	136	188	72·3
C57 BL, sublines 1 + 2	106	128	234	54·7
A	98	13	111	11·7

Table 3. *Incidence of interfrontal according to the mating type. Mice with unknown parental phenotype are omitted*

Stock	Mating type	Normal (N)	Inter-frontal (I)	Total	χ^2	n	P
CBA	I × I	38	232	270			
	I × N	7	52	59			
	N × N	11	43	54			
	Total	56	327	383	1·838	2	~0·4
C57 BL	I × I	24	73	97			
Sublines 3 and 4	I × N	23	49	72			
	N × N	4	8	12			
	Total	51	130	181	1·241	2	~0·5
C57 BL	I × I	14	20	34			
Sublines 1 and 2	I × N	41	50	91			
	N × N	47	44	91			
	Total	102	114	216	1·386	2	~0·5

No special study has been made of the environmental factors which determine manifestation within the various pure lines. However, maternal age seemed to have a marked effect in the CBA stock (Table 4). The percentage of interfrontal is lowest in litters produced by females of 160–189 days of age, while in both older and younger females the incidence is higher. A homogeneity test shows beyond doubt that the deviations of the individual age groups from the common mean cannot be ascribed to chance alone ($\chi^2=34{\cdot}74$; $n=7$; $P<0{\cdot}000,1$). However, a maternal age effect which first rises and then falls again in a similar fashion, though not impossible, is unusual. Hence the possibility must be kept in mind that the heterogeneity between the age groups in Table 4 might have been introduced by a variable other than maternal age. That this may be so is suggested by the behaviour of the two sublines of the C57BL stock (Table 5). Sublines 1+2 again show strong evidence of heterogeneity between age groups ($\chi^2=24{\cdot}575$; $n=6$ (pooling the first and last two age

Table 4. *Interfrontal. Manifestation in relation to maternal age.* CBA *stock*

Maternal age (days)	Normal	Interfrontal	Total	% interfrontal
<100	1	44	45	97·7
100–129	6	56	62	90·3
130–159	12	64	76	84·2
160–189	21	39	60	65·0
190–219	12	65	77	84·4
220–249	1	34	35	97·1
250–279	2	27	29	93·1
280 and over	2	26	28	92·9
Total	57	355	412	86·2

Table 5. *Interfrontal. Manifestation in relation to maternal age. Two sublines of the* C57BL *stock*

Maternal age (days)	Sublines 1+2			Sublines 3+4		
	Normal	Interfrontal	Total	Normal	Interfrontal	Total
<100	3	1	4	5	10	15
100–129	19	24	43	5	8	13
130–159	16	27	43	15	27	42
160–189	29	16	45	3	17	20
190–219	13	21	34	7	11	18
220–249	4	6	10	9	30	39
250–279	15	5	20	5	19	24
280–309	4	12	16	2	6	8
310 and over	3	16	19	1	8	9
Total	106	128	234	52	136	188

groups), and $P\sim0{\cdot}000,5$). However, there is no clear trend with maternal age; the main contributions to χ^2 come from the age groups above 250 days when the incidence of interfrontal first drops below and then shoots up above the common mean. By contrast, sublines 3+4 show no evidence of heterogeneity at all ($\chi^2=7{\cdot}284$; $n=6$; $P\sim0{\cdot}3$). It is difficult to escape the conclusion that the heterogeneity brought out in the data of Tables 4 and 5, though apparently one of maternal age, is really of a different origin. Data not given here in detail do not indicate that heterogeneity between matings or seasonal factors might be involved. So, for the present, the source of heterogeneity must remain in doubt.

PARTED FRONTALS

Even in papain preparations which are greatly superior in cleanness to those obtained by other methods of preparation, the classification of this character is somewhat arbitrary. Usually there is a spindle-shaped widening of the suture between the frontals behind the transverse sinus (Fig. 1B, C). It is thus well behind the position of the interfrontal bone which is always in front of the transverse sinus, and the two features are in fact often found in the same skull. However, while there cannot be much doubt about a spindle-shaped widening in the typical position which has at least a well-defined anterior end, sometimes a widening of the suture is so diffuse and slight that two observers may disagree about its very existence. In normal mouse skulls the two frontals are joined ventrally by a plate of bone across the midline in the neighbourhood of the transverse sinus and for a variable distance behind it; in this region the sagittal sinus is thus entirely enclosed in a bony canal. Where parted frontals are present, the opening leads from the outer surface of the skull into this canal; the opening thus does not penetrate directly into the skull cavity (although sometimes the floor of the canal is pierced by a vascular foramen just under the spindle-shaped opening).

While the 'true' percentage of animals with this variant may be a matter for argument, the existence of great inter-strain differences is beyond doubt. Contrary to Keeler's interpretation, I have not been able to find any blood vessel or other structure entering or leaving this opening; in dissections of freshly killed animals, the opening always seemed completely closed by tough connective tissue. Moreover, in some macerated specimens there is evidence that the opening is occasionally secondarily closed over by bone; this would hardly happen if the opening were traversed by a blood vessel. It also seems that

Table 6. *Incidence of parted frontals in inbred stocks*

Stock	Parted frontals	Doubtful	Normal	Total	% interfrontal
CBA	83	4	13	100	86·2
C57BL, sublines 3+4	2	0	98	100	72·3
C57BL, sublines 1+2	1	1	98	100	54·7
A	29	8	63	100	11·7

vascular foramina in the mouse skeleton are usually rounder and more sharply defined. Keeler's 'parted frontals' is probably to be regarded as a fontanelle, i.e. as a place where the frontals have failed to make contact with each other in the middle and which thus remain closed by a membrane only. The difference between it and other fontanelles is that the frontals of either side are actually fused underneath the opening. The mechanical reasons which are responsible for the persistence of this hole are obscure.

Samples of 100 mice each of the strains CBA and A and of the two C57BL sublines have been examined. The results (Table 6) which are based on a conservative classification show great differences between the three strains while the two sublines of C57BL do not differ in this character. It is also clear that interfrontal and parted frontals behave independently of each other; in CBA both features are frequent; C57BL has very few parted frontals, but numerous interfrontals, while A which has few interfrontals has a fair number of parted frontals. Table 6 thus shows that there are three different stable frequencies for this character; within each stock, the decision concerning presence and absence of parted frontals is purely environmental.

As usual with characters of this kind, manifestation does not appear to be random throughout the stock. In the CBA strain, the 100 animals examined came from five matings (Table 7); of these, two matings with a total of only twenty-six young contributed ten out of a total of thirteen normal animals (without parted frontals); moreover, there seems to be a tendency for the normals to occur in clusters in certain litters. The 100 animals came from twenty-six litters; ignoring differences in litter size, the mean expectation of normals is thus one-half per litter; hence most normals should occur as single animals in a litter if the distribution were random. Actually, there were two litters each with one, two and three normals respectively; in addition, the normals in the first mating occurred in two successive litters (1 and 3 respectively), while five out of six of the CBA 57/58 mating occurred similarly (3 and 2 respectively). Thus it appears that non-manifestation of parted frontals is favoured in certain litters, and that the maternal conditions responsible tend to carry over from one litter to the next.

Table 7. *Parted frontals. Distribution in five matings of the* CBA *stock*

Mating	Parted frontals	Doubtful	Normal	Total
CBA 51/52	4	1	4	9
53/54	34	1	—	35
55/56	29	1	1	31
57/58	11	—	6	17
59/60	5	1	2	8
Total	83	4	13	100

DISCUSSION

The data presented prove beyond doubt that the simple factorial interpretation given by Keeler (1930, 1933) for interfrontal and parted frontals is untenable. On the other hand, both entities show certain features regarded as typical for quasi-continuous characters (Grüneberg, 1952). Interfrontal occurs in four and parted frontals in three different stable frequencies in pure lines. The former shows a parallelism between 'penetrance' and 'expressivity'; in the CBA stock, where the character is common, most interfrontals are large in size; by contrast, in the A strain in which interfrontal is rare, the bone is small and hidden ventrally under the frontals. Interfrontal shows striking heterogeneity in manifestation which suggests hitherto unidentified environmental influences. For parted frontals, some bunching within certain litters suggests that there exist transitory prenatal conditions which favour (or inhibit) the manifestation of this entity. There is thus good evidence that both characters are quasi-continuous variants, and now that they are again available for study, further details may be filled in by other workers interested in this group of phenomena.

From the morphological point of view, the interfrontal cannot be regarded as a suture or Wormian bone, at least if these terms are used in their usual sense, i.e. for irregular ossicles of more or less accidental origin. The regularity of shape and position and particularly the considerable degree of genetical control raise the bone from the accidental to the level of structural significance. The character is in no sense pathological; indeed, in the CBA pure line it has become the normal, and though it has not yet been found in the wild except sporadically a more thorough investigation of wild mice may well reveal mouse populations in which the interfrontal is as common or commoner than in the CBA strain. The interfrontal is a structural element which, it seems, must be taken seriously as a potential addition to the mammalian skull in the evolutionary future. However, the interfrontal does not seem to have much of an evolutionary past. Cornevin (1883) described an 'os wormien fronto-nasal' as common in cattle (about one in twenty animals); its comparative frequency indicates some genetic basis. Fig. 4 shows that it occupies a position similar to that of the interfrontal in the mouse, and the two bones appear to be 'homologous'. Le Double (1903) also mentions a similar bone as a rare variant in Man. One cannot assume that there has been a common ancestor with this feature, but a probable interpretation is that the genetic capacity to form an interfrontal is derived from a remote common ancestor and is presumably present in many mammals; its phenotypical realization being the exception rather than the rule. As discussed by de Beer (1951) the apparent polyphyletic origin of a phenotype thus does not exclude the possibility that it rests on a common genotype of monophyletic origin ('latent homology').

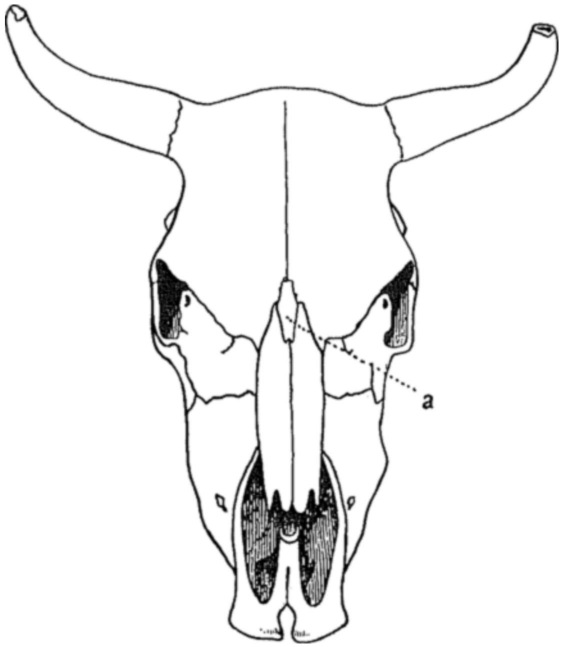

Fig. 4. An interfrontal bone in the cow. From Cornevin (1883).

Summary

The skull variants interfrontal and parted frontals originally described by Keeler are shown to have a complex genetic basis.

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