

TRANSFERRIN POLYMORPHISM IN THE AUSTRALIAN MARSUPIAL  
MOUSE *SMINTHOPSIS CRASSICAUDATA* (GOULD)\*

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A number of polymorphisms involving characteristics of the blood have been described in Australian marsupials, but the genetical control of these has been established only for variation in the iron-binding serum protein transferrin in the red kangaroo *Macropus rufus* (Desmarest) (= *Megaleia rufa*) (Cooper and Sharman 1964), and the eastern and western grey kangaroos *Macropus giganteus* (Shaw) and *Macropus fuliginosus* (Desmarest) (Kirsch and Poole 1967). Transferrin variation in the brush-tail possum *Trichosurus vulpecula* (Kerr), first detected by Kirsch (personal communication), is being studied in this laboratory. The inheritance of the protein patterns developed on a starch gel after electrophoresis may be ascribed to the actions of two or more autosomal allelic genes without dominance.

In addition, polymorphisms for transferrin have been described in the euro *Macropus robustus* (Gould) (Kirsch, unpublished data), the tammar wallaby *Macropus eugenii* (Desmarest)§ (Kirsch 1967), the ring-tail possum *Pseudocheirus peregrinus* (Boddaert), and the marsupial mouse *Sminthopsis larapinta* (Spencer) (both Hope, unpublished data). Lai (1966) has described variation of red cell acid phosphatase in the red and grey kangaroos, Owen and Thomson (1964) have described an electrophoretic variant of haemoglobin in the mountain possum *Trichosurus caninus* (Ogilby), and Barker (1961) has investigated a polymorphism involving the level of erythrocytic sodium and potassium in the brush-tail possum, similar to that described by Evans (1954) in sheep. The genetical control of these polymorphisms has yet to be established.

The present communication reports the discovery of heritable variation in the electrophoretic mobility of serum transferrin in the fat-tailed marsupial mouse *Sminthopsis crassicaudata* (Gould) and indicates the present knowledge of the inheritance of this variation.

The animal is a small, nocturnal, carnivorous marsupial, widely distributed over the Australian continent, but apparently most plentiful in the drier inland areas. There are several distinct geographical races, differing in size, pelage, and body proportions (Finlayson 1933, 1961; Troughton 1964).

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§ See Calaby (1966) for note on generic names of wallabies.

The animals used in the present study were either captured in the field or were from crosses set up in the laboratory for other purposes. The main capture localities are given in the following tabulation:

Population	Locality	Approx. Radius of Capture (miles)
1	Glengyle Homestead, south-western Queensland (24' 48" S., 139' 37" E.)	3
2	Patacoona Homestead, Lower Flinders Ranges, S.A. (32' 01" S., 138' 10" E.)	3
3	Waramboo and other localities, Eyre Peninsula, S.A. (33' 14" S., 135' 36" E.)	50
4	Beachport and other localities, south-eastern South Australia (37' 30" S., 140' 01" E.)	50

Blood samples were obtained from etherized animals, and in some instances from animals recently killed for other purposes, by puncturing the orbital sinus with a fine needle, and drawing the resulting drops of blood (average volume 0.25 ml)

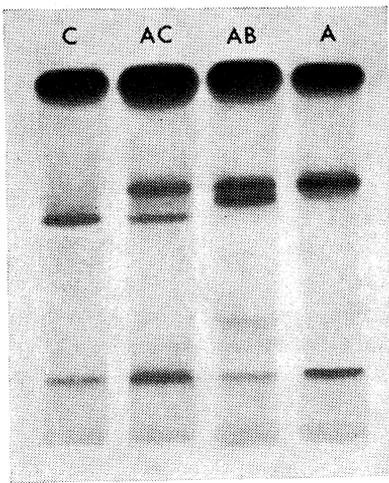


Fig. 1.—Starch gel electrophoretogram of plasma samples of *Sminthopsis crassicaudata*, showing transferrin phenotypes.

into an heparinized syringe. Plasma was subjected to vertical starch-gel electrophoresis using water-cooled gel racks and the buffer system of Gahne (1966), the gel being prepared as described by Kristjansson (1963). Autoradiographs, to enable localization of transferrin, were made using  $^{59}\text{FeCl}_3$  basically as described by Cooper and Sharman (1964). One-half of each sliced gel was stained with amido black (Gurr) to detect protein, and the other half was stained for plasma esterase activity (Gahne 1966). Addition of a small amount of haemolysate to the plasma enabled the localization of the haptoglobin-haemoglobin complex on the gel, by staining with dianisidine reagent for peroxidase activity as described by Owen and Smith (1961). Haemolysates were examined for haemoglobin variation by starch-gel electrophoresis, using the buffer system of Smithies described by Huehns and Shooter (1965).

No variation was observed in the haemoglobin or the haemoglobin-haptoglobin complex. These differed in mobility but in all samples were recognized in each case as a single band on the gel. Numerous plasma esterase bands were apparent, but no clear variation between individuals was found.

Insufficient blood was available from these animals for analysis of erythrocytic sodium and potassium levels.

Four transferrin phenotypes could be distinguished and are named *Tf A*, *Tf AB*, *Tf AC*, and *Tf C*, referred to in the remainder of this paper as *A*, *AB*, *AC*, and *C* respectively (see Fig. 1). *A* possessed a single iron-binding protein band. *AB* showed two bands of equal intensity, the faster of which was of similar mobility to the single band of the *A* type. *AC* also showed two bands of equal intensity. The faster was

TABLE 1  
FAMILY DATA SHOWING THE INHERITANCE OF TRANSFERRIN  
PHENOTYPES IN *SMINTHOPSIS CRASSICAUDATA*

Transferrin Phenotype of Parents*	No. of Matings	Frequency of Transferrin Phenotypes in Offspring		
		<i>A</i>	<i>AB</i>	<i>AC</i>
<i>A</i> × <i>A</i>	12	31	—	—
<i>A</i> × <i>AB</i> †	12	19	12	—
<i>A</i> × <i>C</i>	3	—	—	10
? × <i>A</i>	12	16	3	—
? × <i>AB</i>	2	1	4	—

\* Parents and offspring are not grouped according to sex.

†  $\chi^2_1$  (based on an expectation of 1 *A*: 1 *AB* amongst the progeny) = 1.58,  $0.2 < P < 0.3$ .

identical to the *A* type band, the slower was equivalent to the single band of the *C* type—which was slower than the slow band of *AB* type. It is postulated that the phenotypes are controlled by three autosomal allelic genes without dominance (*Tf<sup>a</sup>*, *Tf<sup>b</sup>*, and *Tf<sup>c</sup>*) each giving rise to a characteristic single transferrin band. Thus the *A* and *C* types are homozygotes (*Tf<sup>a</sup>/Tf<sup>a</sup>* and *Tf<sup>c</sup>/Tf<sup>c</sup>*) and the *AB* and *AC* types are heterozygotes (*Tf<sup>a</sup>/Tf<sup>b</sup>* and *Tf<sup>a</sup>/Tf<sup>c</sup>*). Family data on the inheritance of these phenotypes, given in Table 1, are not inconsistent with this hypothesis.

Two other transferrin types would be expected to occur on the basis of the above hypothesis, namely *Tf B* and *Tf BC*. The absence of these types is attributed to a low frequency of the *Tf<sup>b</sup>* allele in the populations sampled. Attempts are being made to produce such animals by controlled matings.\*

Table 2 gives the population data for the transferrin gene frequencies. As some of the females caught in the wild had pouch young, the inclusion of these relatives necessitated appropriate correction of these frequencies (see Finney 1948). The gene frequencies do not differ significantly between the first three populations.

\* Note added in proof.—Three *Tf B* animals have now been obtained from these matings.

However, the samples are small, and the detection of any slight differences must await more data.

The animals from Glengyle, in south-western Queensland, are recognized as a separate subspecies, *S. crassicaudata centralis* (Thomas, 1902), distinguished from the nominate race by their diagnostic pattern of granulations on the sole-pad (Troughton 1964 and personal communication). The taxonomic status of the other populations of *S. crassicaudata* has not yet been resolved. The animals captured near Beachport and in other localities in the south-east of South Australia could be clearly distinguished by their darker pelage and shorter tails, and it seems probable that this dark form found to the east of the Murray R. will eventually be recognized as a distinct subspecies. These seven animals showed a transferrin band not found in the other populations.

TABLE 2

POPULATION DATA FOR THE TRANSFERRIN POLYMORPHISM IN *SMINTHOPSIS CRASSICAUDATA*

Population	Transferrin Phenotypes					Pouch Young Included in Sample	<i>Tf<sup>b</sup></i> Gene Frequency*	95% Confidence Limits†
	<i>A</i>	<i>AB</i>	<i>AC</i>	<i>C</i>	Total			
1	17	3	—	—	20	2	0.08	0.02–0.22
2	19	4	—	—	23	12	0.05	0.01–0.16
3	16	11	—	—	27	16	0.19	0.06–0.31
4	—	—	1	6	7	—	—	—

\* These frequencies have been corrected for the inclusion of pouch young in the data (Finney 1948).

† Based on Stevens' tables (Fisher and Yates 1957).

The potential of *S. crassicaudata* as a laboratory animal has recently been discussed by Martin (1965). The transferrin locus may prove a most useful genetic marker for further laboratory and taxonomic studies of *S. crassicaudata*.

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