

# TRANSFERRIN POLYMORPHISM AND POPULATION STRUCTURE OF THE WEDDELL SEAL *LEPTONYCHOTES WEDDELLI* (LESSON)\*

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Variation in the electrophoretic mobility of the iron-binding serum protein transferrin has been reported by Naevdal (1966a) in the harp seal *Pagophilus groenlandicus* (Erxleben), and in a serum protein thought to be transferrin in the ringed seal *Pusa hispida* (Schreber) (Naevdal 1966b). Transferrin variation has also been found in southern fur seals *Arctocephalus* spp. of the Australasian region (Shaughnessy, unpublished data).

This communication reports the discovery of transferrin variation in the Weddell seal *Leptonychotes weddelli* (Lesson), and discusses the results in relation to the population structure of the species. The Weddell seal (Phocidae) breeds on the fast ice around the Antarctic continent and adjacent offshore islands, and is also seen in the pack ice, where it is less abundant.

## Methods

Blood samples were collected at five localities on the Antarctic coast: near the Australian National Antarctic Research Expedition bases at Mawson, Davis, and Wilkes; on the eastern side of McMurdo Sound near Scott Base; and at nearby White I. in the Ross Ice Shelf. These localities are shown in Figure 1.

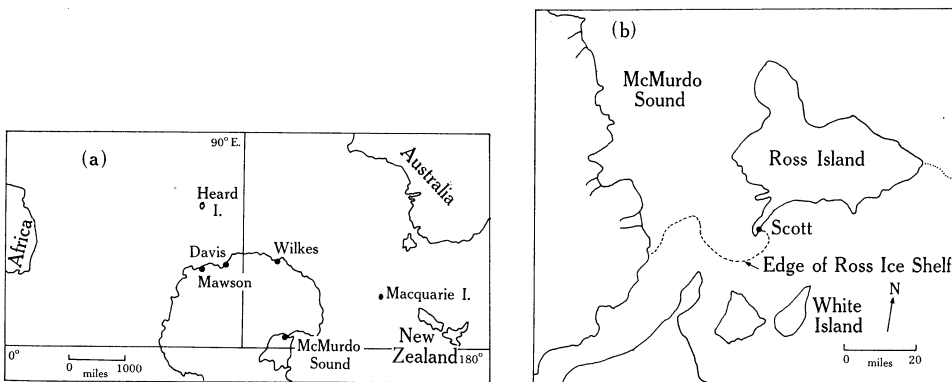


Fig. 1.—(a) Antarctica, showing sampling locations of *Leptonychotes weddelli*. (b) McMurdo Sound, showing Scott Base and White I.

Blood was collected either from seals shot for dog food or, in the case of some live animals at McMurdo Sound, from the hind flipper. Collections at the Australian bases were made by medical officers, those at McMurdo Sound by Dr. I. Stirling. Serum samples were stored at  $-20^{\circ}\text{C}$ , and transported to Australia by ship. Analyses were performed in Adelaide.

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Serum was subjected to vertical starch-gel electrophoresis using water-cooled gel trays with the Tris-cacodylate buffer system of Kristjansson and Hickman (1965), and the electrolyte of Gahne (1966). After electrophoresis, gels were sliced longitudinally and stained for protein with amido black. Autoradiography to locate transferrin zones was carried out with radioactive iron as described by Cooper and Sharman (1964).

To locate haem-binding proteins, a small amount of haemolysate was added to the serum samples before electrophoresis in another gel which was stained with *o*-dianisidine for the peroxidase activity exhibited by haem groups (Owen and Smith 1961). Haemolysates were examined for haemoglobin variation by starch-gel electrophoresis using the buffer system of Smithies (Huehns and Shooter 1965).

### Results

Each animal was found to possess two haemoglobin zones of equal intensity, and a single haem-binding zone. No variation was observed in the mobility of the haemoglobins of 17 Mawson animals, nor in the haem-binding zone for 70 animals from Mawson, Wilkes, and McMurdo Sound.

Three transferrin types were observed and have been named *TfS*, *TfSF*, and *TfF* (Fig. 2). Transferrins *S* and *F* each possessed three zones, of which the slowest was strongest, and the fastest was faintest. The mobility of the intermediate zone of *S* was the same as that of the slowest zone of *F*. In *SF* only three of the expected six zones were visible; one of the other three overlapped with the slowest *F* zone (as described above), while the other two zones were probably too faint to be visible.

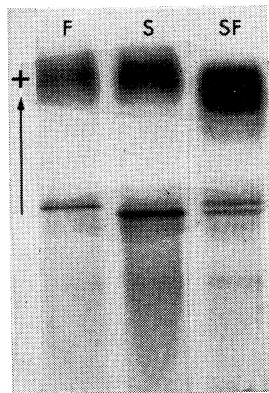


Fig. 2

Extensive family data from other animals, e.g. cattle, sheep, pigs, pigeons (Lush 1966), are in agreement with the hypothesis that transferrin variation is controlled by a number of allelic genes. If transferrin types in the Weddell seal are controlled by two allelic genes, *TfS* and *TfF*, without dominance, then types *S* and *F* would be homozygotes (*TfS*/*TfS* and *TfF*/*TfF*), and *SF* a heterozygote (*TfS*/*TfF*). A small amount of family data, consisting of two mother-pup pairs from White I., are in agreement with this hypothesis. In both cases

the transferrin types of the mother and pup are *S*.

Population data for transferrin phenotypes, and gene frequencies, calculated on the basis of the above hypothesis, are given in Table 1. The observed phenotypic frequencies for the Mawson, Davis, and Wilkes samples are very close to expectations predicted by Hardy-Weinberg equilibrium. However, the Scott sample differs significantly from expectations ( $P = 0.01$ ) because of a deficiency of heterozygotes. If the eastern McMurdo Sound population is considered as a single breeding unit, then small population size alone is an unlikely explanation for this deficiency, for Stirling (1968) estimated that this population contains about 900 breeding females and at least 100 breeding males, giving an effective breeding population size (Wright 1931) of about 400. However, there is evidence that the population is subdivided into breeding isolates (Stirling 1968), and this may well be the cause of the observed deficiency of heterozygotes.

The population of Weddell seals at White I. is separated from the eastern McMurdo Sound population by 16 miles of the Ross Ice Shelf. Inspection of transferrin frequencies in the White I. and Scott samples shows that both alleles are present in each population. From a study of tagged animals at White I. and eastern McMurdo Sound, Stirling (1968) concluded that these populations are isolated from each other. However, the transferrin data neither support nor contradict this conclusion.

The three largest samples, from Mawson, Wilkes, and Scott, are heterogeneous with respect to transferrin gene frequency ( $\chi^2_2 = 6.52$ ,  $0.02 < P < 0.05$ ). Considering the three samples pairwise, the difference in gene frequency between the Mawson and Scott samples is not significant ( $\chi^2_1 = 0.13$ ,  $0.70 < P < 0.80$ ), but both gene frequencies are significantly different from that in the sample from Wilkes which is situated between them ( $\chi^2_1 = 5.14$ , with Yates' correction,  $0.02 < P < 0.05$ ; and  $P = 0.044$ , respectively).

TABLE 1  
TRANSFERRIN TYPE FREQUENCIES (O) AND RANDOM-MATING EXPECTATIONS (E) IN WEDDELL SEAL POPULATIONS

Location	Transferrin Phenotypes						No. of Seals	<i>Tf</i> <sup>F</sup> Gene Frequency	95% Confidence Limits*
	<i>S</i>		<i>SF</i>		<i>F</i>				
	O	E	O	E	O	E			
Mawson	19	19.4	11	10.3	1	1.3	31	0.210	0.117–0.332
Davis	6	6.1	2	1.8	0	0.1	8	0.125	0.016–0.384
Wilkes	22	22.0	2	1.9	0	0.1	24	0.042	0.005–0.143
Scott	17	14.7	2	6.6	3	0.7	22	0.182	0.082–0.327
White I.	7†		1		0		8		

\* Calculated using Stevens' table (Fisher and Yates 1963).

† Includes two mother-pup pairs.

Differences in gene frequency between animals in widely spaced localities are not unexpected if Weddell seals breed in the vicinity of their birthplace. There is some evidence that they do. Perkins (1945) found three seals in the Bay of Whales, Ross Sea, which had been branded there as pups 6 years earlier by Lindsey (1937). Also, a female tagged as a pup in 1948 by Laws at Signy I. in the South Orkney Is. was observed to breed there in 1960 and in 1962 (see Carrick 1964). Further, Stirling (1968) from observations of branded and tagged animals in McMurdo Sound, over several years, suggested that Weddell seals generally breed in the vicinity of their birthplace.

The present data do not permit determination of the relative importance of natural selection and random sampling fluctuations in maintaining the observed differences in transferrin gene frequency. Nor do they enable the effect of migration on these differences to be assessed. The Weddell seal is distributed around the Antarctic continent, so that collections at several localities between those sampled in this study would enable the role of these factors to be investigated.

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