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#### SEROLOGICAL STUDIES ON MARINE MAMMALS

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#### WHALES

Erythrocyte antigens of different whale species have been extensively studied by Fujino and collaborators. The results up to 1963 have been reviewed by Cushing (1964) and only the main points will be mentioned here.

By immunizing rabbits by erythrocytes of the striped dolphin, Prodelphinus caeruleo albus, Yamaguchi and Fujino (1952) discovered a blood group system containing two factors named  $Dc_1$  and  $Dc_2$ . Weak, and occasionally strong, naturally occurring antibodies were found partly to correspond to the  $Dc_1$  and  $Dc_2$  factors, but they were usually too weak to allow the blood types to be classified this way.

Fujino (1953) described two antigens (agglutinogens and hemolysinogens) in each of four baleen whales, the Sei-whale, Balaenoptera borealis, the Fin-whale, B. physalus, the Blue whale, B. musculus, and the Humpback whale, Megaptera novaeangliae. All specimens of each of the species could be classified into four groups on the basis of these antigen variations. In the Fin whale a remarkable change in frequencies of the blood group factors, in this species named  $Bp_1$  and  $Bp_2$ , was observed during the whaling season in the Northern Pacific Ocean in 1952, indicating mixing of different population units.

In two later papers (Fujino 1956, 1958) two new antigens, named Ju 1 and Ju 2, were described in the Fin whale. Ju-antibodies occurred in normal rabbit and fowl sera, but were normally obtained from immune rabbit sera.

In later papers (Fujino 1960, 1962, 1963 a, 1963 b, 1964) a subdivision of the Ju 2 factor into five subfactors (Ju 2<sub>1</sub> ..... Ju 2<sub>5</sub>) was described. Four breeding populations of Fin whales differing in the frequencies of the Ju-antigens, could be distinguished in the Antarctic (Fujino 1962, 1963b, 1964), and a total of five populations of Fin whales could be classified on the basis of frequencies of the Ju- and Bp-factors in the Pacific (Fujino 1960, 1963c). The Ju-factors could be used with a higher precision in blood typing than the Bp factors.

In the Sperm whale, Physeter catodon, two independent blood group systems were described by Fujino (1954), and a third system resembling the Ju-group of fin whale, was described by Cushing, Fujino and Calaprice (1963). Fujino (1963d) found great differences in frequencies of Ju 2-positive sperm whales between samples taken in waters around Japan and samples taken around the Aleutians. Seasonal changes occurred, and the populations seemed to mingle in waters near Japan.

Blood group systems were also described in Baird's beaked whale, Berardius bairdi, (Fujino 1954), and frequency variation were observed also in this species.

Cushing, Fujino and Takahashi (1959) found that whale blood cells were well preserved when frozen with glycerol, and this method was frequently used in the studies referred to.

Fujino and Cushing (1959) described an alternative method to the conventional blood type technique which may be potentially useful for large scale blood typing under unfavourable conditions. They found that blood specimens could be typed with I<sup>131</sup>-labelled antibodies after the blood had been dried on filter paper.

Fujino (1962, 1963a, 1963b) found that pregnancies were less frequent in Ju 2 negative Pigmy Blue and Fin whales than in Ju 2 positive females, because Ju 1 positive females possess lytic isoantibodies for Ju 2 cells causing maternal-foetal incompatibility and thus intrauterine selection. Possibly the blood type gene frequencies are maintained in the populations by differences between blood groups in relative viability in accordance with increment of age (Fujino 1964).

Post mortem changes of blood types due to specific inhibition of agglutination by soluble substances in serum were observed by Cushing et al. (1963). This effect may be avoided by using blood from the heart of recently killed animals for blood typing.

As a conclusion blood typing of whales have contributed considerably to our knowledge of the existence and dispersal of population units of Fin and Sperm whales. The results by and large are in accordance with marking experiments (Fujino 1960, 1964).

A search for blood protein polymorphism to be applied on Atlantic populations of whales, especially Fin-whale, has been started with Arctic Biological Station, Fisheries Research Board of Canada, Hafransoknastfrunin, Reykjavik and Institute of Marine Research, Bergen, cooperating in the project.

#### SEALS

Fujino and Cushing (1960) described four blood types in the Fur seal, Callorhinus ursinus, differentiated by absorption of rabbit anti-fur seal serum. Isoagglutinins also occurred at high frequencies. A special non-random distribution of types during the migratory season indicated the existence of special breeding stock.

Borisov (1966) studying sera of the Harp seal, Pagophilus groenlandicus, by immunoelectrophoresis and agar diffusion technique, could not find differences between representatives of the breeding herds in the White Sea, in the Jan Mayen area and the area northeast of Newfoundland.

As part of a program to determine whether the breeding herds of the Harp seal and the Hooded seal, Cystophora cristata, are selfsustaining population units, serological studies on these species were started at the Institute of Marine Research, Bergen, in 1962, with a search for intraspecific variations in erythrocyte antigens in Hooded seals. Strong rabbit antisera against hooded seal antigens were obtained, but absorbtions removed all the antibodies, and no variation in the erythrocyte antigens could be found. The study was highly complicated by lack of fresh material for absorbtions, and a freezing technique was found not to be successful for preserving blood cells of this species. Therefore attention was shifted to electrophoretic studies on blood proteins.

Blumberg, Allison and Garry (1960) described intraspecific variation in postalbumins and haptoglobins in the Fur seal. The variations seemed to be genetically controlled as far as could be determined from a limited material.

Electrophoretic analyses of hemoglobins were performed on about 600 specimens of Harp seals and 400 specimens of Hooded seals. For comparison 17

specimens of ringed seal, Pusa hispida, and 46 specimens of bearded seal, Erignathus barbatus, were also analyzed (Nævdal 1966a, 1966b). One strong anodic and one weaker cathodic component was the normal pattern, but three specimens of harp seals each showed different additional anodic components, indicating that these three animals were heterozygotes for rare hemoglobin-controlling genes. These genes, however, were too rare to be of any value for comparison of samples.

Analyses of serum proteins of harp seals (Nævdal 1966a) revealed polymorphism in the serum transferrins. Six phenotypes were found, and the observed distributions were in accordance with a hypothesis of control by three alleles at one autosomal locus. The material comprised samples from the Jan Mayen area (132 specimens), the White Sea (105 specimens), the Barents Sea (119 specimens) and the area northeast of Newfoundland (208 specimens). A t-test of gene frequencies showed significance at the 5 percent level, but not at the 2 percent level, between the sample from Newfoundland and the total of the northeast Atlantic samples, indicating genetic isolation. However, samples collected in 1967 northeast of Newfoundland (the Front) and in 1968 in the Gulf of St. Lawrence showed that no significant differences existed between any of the breeding herds of harp seals (Nævdal 1969).

A total of about 500 specimens of Hooded seals from the Jan Mayen area (130), the Denmark strait (83) and northeast of Newfoundland were analyzed for serum proteins (Nævdal 1966b and unpublished). Considerable intraspecific variation was observed, but no variation in the transferrins could be detected by the present method. Variations in haptoglobins were conspicuous, but the variations were complicated and no proof of genetical control could be given. Haptoglobin differences were also observed between pups and adult hooded seals.

In another group of unidentified serum proteins, intraspecific variations, possibly controlled by a pair of autosomal alleles, were demonstrated. The two components, tentatively called II and III, occurred at different frequencies in the analyzed samples, as the component of higher anodic mobility (III) was considerably more frequent in the samples from Newfoundland than in the samples from the Jan Mayen area and the Denmark Strait. However, because the components were weak and the phenotypes were not always easily recognized, any conclusion based on the observed differences between the samples is doubtful.

A limited number of sera from Ringed seals (15) and Bearded seals (40) were analyzed for comparison. In the Ringed seal variation in components supposed to be transferrins (not tested autoradiographically) was seen, and although few sera were analyzed, it is probable that this variation corresponds to the transferrin variation of Harp seals. In the Bearded seal no clear intraspecific variation was seen, but haptoglobin variations were indicated.

Analyses of pancreas amylase has been carried out on a limited material of Harp seals (73), Hooded seals (30), and Bearded seals (2), using the method described by Sick and Nielsen (1964). A clear intraspecific variation was found only in the Hooded seal, and the results indicated a control by at least three alleles.

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