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## SOME BIOCHEMICAL AND BIOPHYSICAL CHARACTERISTICS OF CANINE AND BOVINE ERYTHROCYTES.

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This study was done to provide a better understanding of the structures and functions of erythrocytes of domestic animals under both normal and pathological conditions. In the present study, the cause of poikilocytosis in calves and an abnormality of amino acid transport in canine erythrocytes were investigated.

Nine calves exhibiting poikilocytosis (poikilocytic calves), 1 to 2 months old, were examined. The proportion of poikilocytes, such as spicule-, teardrop- or pear-shaped red cells in their blood was more than 70% in two calves (severe poikilocytosis), while it average 30% in the remaining seven calves (moderate poikilocytosis). All of the poikilocytic calves exhibited mild anemia. The concentration of plasma iron in 8 of them was 20–30% of that in normal calves. Hemoglobin preparations from these 9 calves and from 13 normal calves, 2 months old, were analysed on an anion-exchange column, using a high-performance liquid chromatography system. The column chromatography of the hemoglobin preparation from all the calves examined revealed 5 distinct peaks. Two peaks were HbF (fetal hemoglobin) and HbA (adult hemoglobin), respectively. The other peaks were tentatively designated Hb-2, Hb-3 and Hb-5, respectively. The amount of Hb-2 in 2 calves with severe poikilocytosis was two times that in the other calves, including those with moderate poikilocytosis, and it decreased as the calves aged. The decrease of Hb-2 in a severely poikilocytic calf was related to a reduction of the proportion of poikilocytes in its peripheral blood. In addition, the amount of HbF in poikilocytic calves was higher than that in normal calves. Furthermore, SDS-PAGE analysis of the red cell membrane revealed that band 4.2 consisted of two polypeptides in five poikilocytic calves, while it consisted of a single polypeptide in the other calves examined.

These results indicated that the abnormalities of hemoglobin composition and band 4.2 protein in the erythrocyte membrane observed in calves with poikilocytosis might be responsible for the morphological changes in the erythrocytes.

The Na<sup>+</sup>-dependent L-glutamate (Na/Glu) transport in canine erythrocytes was investigated. In this experiment, dogs having erythrocytes characterized by hereditary high concentrations of K<sup>+</sup> and reduced glutathione (GSH) [HK dogs], dogs with a high concentration of K<sup>+</sup> and low GSH in their erythrocytes [HK/LG dogs], dogs with low K<sup>+</sup> and low GSH erythrocytes [LK/LG dogs] and normal dogs [LK dogs] were used. The initial uptake rate of L-glutamate (L-glu) into erythrocytes was 1.70

$\mu$ moles/l cells/hr in HK cells and 0.53 in LK cells. This increased uptake of L-glu by HK cells was due to the presence of  $\text{Na}^+$ ,  $\text{K}^+$ -ATPase in these cells, while LK dog erythrocytes completely lack the enzyme. In contrast, there was no significant uptake of L-glu into either HK/LG or LK/LG cells. Dog reticulocytes, immature erythrocytes, are known to contain  $\text{Na}^+$ ,  $\text{K}^+$ -ATPase. The L-glu uptake by reticulocytes from LK dogs greatly increased compared to that by LK erythrocytes. In contrast, no uptake of L- $^3\text{H}$ glu by reticulocytes from HK/LG dogs was observed.

These results strongly indicate that both HK/LG and LK/LG dog erythrocytes completely lack the Na/Glu transport which is specific to canine erythrocytes.