PURIFICATION AND IDENTIFICATION OF PORCINE ERYTHROCYTE ADENTLATE KINASE (ATP: AMP PHOSPHOTRANSFERASE, EC 2. 7. 4. 3)

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Porcine erythrocyte adenylate kinase showed three components, Ak-a, Ak-b and Ak-c, from origin to anode starch gel electrophoresis. The genetic isozyme pattern, however, could not be observed in this experimental scale of 50 specimens, which differed from human erythrocyte adenylate kinase having electrophoretic variants. By isoelectrofocusing, 96% of the total erythrocyte adenylate kinase focused at pH 9.25, and this predominant enzyme form corresponded to Ak-a on starch gel electrophoresis. Porcine skeletal adenylate kinase showed the same profiles as those of the erythrocyte enzyme on starch gel electrophoresis and on isoelectrofocusing.

The predominant enzyme form, Ak-a, was purified approximately 31,000 fold from porcine erythrocyte in an overall yield of 46% to a final specific activity of 2,100 units per mg of enzyme. The purified enzyme had a molecular weight of 21,500 and migrated in gel electrophoresis as a single band. A mixture prepared together with purified skeletal muscle enzyme also gave a single narrow band in sodium dodecylsulfate gel electrophoresis. The amino acid composition of porcine erythocyte Ak-a was found to be Asx_{13}, Thr_{14}, Ser_{11}, Glx_{25}, Pro_{6}, Gly_{19}, Ala_{8}, Val_{17}, Met_{6}, Ile_{9}, Leu_{18}, Tyr_{7}, Phe_{5}, His_{2}, Lys_{21}, Arg_{11}, Cys_{2}, (as cysteine), and no Trp. Peptide mapping showed the correspondence of all spots of the tryptic peptides from erythrocyte and skeletal muscle enzyme preparations.

These results strongly suggest that the predominant adenylate kinase in porcine erythrocyte is identical to porcine skeletal muscle adenylate kinase.