ISOLATION AND CHARACTERIZATION OF A BOVINE ACUTE PHASE PROTEIN BY AFFINITY CHROMATOGRAPHIES

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Different affinity chromatographies were performed in order to isolate an acute phase protein, which was proposed to be bovine C-reactive protein (CRP), from sera of bovine with differet inflammations. Both pneumococcal C-polysaccharide (CPS)-conjugated Sepharose4B (agarose beads) and Sepharose4B bound the protein in the presence of Ca^{2+} and eluted with 10mM EDTA solution. Sepharose4B chromatography was superior to CPS-Sepharose4B chromatography because of less contamination. The eluate from Sepharose4B column was subsequently applied to DEAE-cellulose or phosphorylcholine-coupled TSK-GEL (TOYOPEARL HW-65C). The acute phase protein was purified by the latter affinity column.

The protein bound to either CPS, agarose or phosphorylcholine in the presence of Ca^{2+}. The molecular weight of the subunit dissociated from the protein by SDS was 23,000 daltons. Moreover, the protein was detectable in three sera from cows having different inflammations. The protein similarly charged as IgG as demonstrated in native electrophoresis or ion-exchange chromatography. These results strongly indicated that the purified protein is bovine CRP.