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ISOLATION, CHARACTERIZATION AND QUANTITATIVE ANALYSIS OF EQUINE C-REACTIVE PROTEIN

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C-reactive protein (CRP) was isolated from equine serum by calcium-dependent affinity chromatography using Sepharose-4B conjugated pneumococcal C-polysaccharide, DE-52 anion-exchange chromatography and Sephacryl S-300 gel filtration. It was identified as genuine CRP by its immunochemical cross-reactivity with anti-human CRP.

Purified equine CRP had a molecular weight which was estimated to be approximately 118,000. It was composed of five identical, non-glycosylated and non-covalently associated subunits (molecular weight; 23,000). Intact equine CRP showed the typical cyclic pentameric disk-like structure of the pentraxin family under electron microscopy. Equine CRP had 201 amino acid residues and its amino acid composition was similar to that of human CRP. In immunoelectrophoresis, equine CRP had $\beta \sim \gamma$ mobility while human CRP had γ mobility, but in cellulose-acetate membrane electrophoresis, equine CRP had fast γ mobility in both the presence and absence of calcium. The isoelectric point of the protein was estimated to be about 7.0 by isoelectric focusing.

The serum CRP concentrations in horses were measured by a single radial immunodiffusion method. In normal horses, the concentration (mean \pm SD) was 7.4 ± 2.0 (5.0~9.6, n=10) μ g/ml. In horses suffering from pneumonia, it was 19.0 ± 9.0 (10.4~38.5, n=10) μ g/ml, whereas it was 16.0 ± 6.4 (7.4~26.9, n=10) μ g/ml in enteritis cases and 11.5 ± 3.3 (8.3~19.5, n=10) μ g/ml in horses with arthritis. The difference ($p < 0.05$) between the value of normal horses and that of the patients was statistically significant. The serum CRP concentration of the patients was two or three times higher than that of normal horses. After injection of turpentine oil or castration, serum CRP levels increased to about six times their pre-levels.