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ISOLATION, CHARACTERIZATION, AND QUANTITATIVE ANALYSIS OF SERUM α_2 -MACROGLOBULIN FROM HORSES

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Alpha₂-macroglobulin (α_2 M) was isolated from equine serum by polyethylene glycol precipitation, gel filtration, and anionexchange chromatography. Purified α_2 M was identified by antichymotrypsin activity and its immunological cross-reaction with human α_2 M.

Equine α_2 M migrated at the α_2 -globulin region under cellulose acetate electrophoresis, and its isoelectric point was 5.2. In SDS polyacrylamide gel electrophoresis, the α_2 M subunit dissociated into a molecular weight of 177,000 after reduction with 2-mercaptoethanol. Accordingly, horse α_2 M molecular weight was estimated to be approximately 708,000 because of its molecular form of a tetramer of identical glycosylated subunits. The polypeptide portion of equine α_2 M contained 4,800 amino acid residues, whose composition was similar to that of human α_2 M. Spur formation in a double immunodiffusion test demonstrated that equine α_2 M shared a common antigen region with human α_2 M.

The concentration of equine serum α_2 M was measured by the single radial immunodiffusion method. In clinically normal horses, the concentration of serum α_2 M was lowest in newborn foals (1.25 ± 0.19 mg/ml), maintained low values from 0 to 1 month old (1.73 ± 0.80 mg/ml), then showed a tendency to increase with aging from 3 to 12 months old (5.85 ± 1.08 mg/ml), and finally stabilized in adult horses (4.48 ± 1.17 mg/ml). The mean value of α_2 M was 3.99 ± 1.88 mg/ml in horses of all ages. The serum α_2 M concentration in mares during the perinatal period started to decrease from 2 months pre-partus, and returned to its normal level within 2 months after delivery. In horses with experimentally induced inflammation, there was no significant change in serum α_2 M concentration with the exception of slight increases on days 10 and 14 after treatment. In most of the horses with clinically inflammatory signs, serum levels of α_2 M were statistically lower than the normal values.

It was concluded that α_2 M is not an acute-phase reactive protein in the case of horses. However, the serum level of equine α_2 M might change in some pathological states of various diseases.