An automated biochemical assay for haptoglobin: prevention of interference from albumin

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Abstract: Measurement of the acute phase serum protein, haptoglobin (Hp), is performed by biochemical methods based on haemoglobin binding, in many veterinary diagnostic laboratories. During attempts to develop a robust biochemical assay for serum Hp it was discovered that serum albumin interfered with the assay system increasing results by as much as 0.28 mg/ml, which could affect interpretation of results especially in species with low normal Hp concentrations. A reagent cocktail (SB-7) was devised which inhibited the interfering effect of albumin. An automated assay for Hp utilising SB-7 was developed for production as a biochemical assay kit and was evaluated for use in veterinary diagnosis. The intra-assay coefficients of variation were of 0.9%, 0.9% and 1.3% for Hp concentrations of 2.0, 1.0 and 0.23 mg/ml, respectively and interassay coefficients of variation of 1.7% and 4.5% for Hp of 2.08 mg/ml and 0.24 mg/ml, respectively. The lower limit of detection of was 0.02 mg/ml, linearity extended to 8 mg/ml and recovery was 101±7% (mean ± SD). The assay had correlation coefficients (R^2) of 0.96 and 0.90 when compared with immunodiffusion assays of canine Hp and bovine Hp, respectively. Lipaemia and bilirubinaemia caused no interference. Haemolysis did not affect measurement of low levels of Hp, but at serum Hp concentrations of 0.4 and 1.8 mg/ml the apparent Hp concentration was decreased. Elevated concentrations of Hp were measured in cattle with mastitis, dogs with polyarthritis and rats experimentally infected with *Bordetella pertussis*. The automated assay is precise and has negligible interference from albumin.

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