

Gli esami delle urine, relativi a questo lavoro scientifico sono stati eseguiti presso il laboratorio di analisi veterinarie



QUALITATIVE DETERMINATION OF PROTEINURIA BY SDS-AGE IN THE HEALTHY DOG

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The determination of proteinuria by Sodium Dodecyl Sulphate Polyacrilamide Gradient Gel electrophoresis (SDS-AGE) makes it possible to evidence in the urine protein with a molecular weight (MW), from 9000 (β_2 microglobulin) to 900,000 (α_2 macroglobulin). The method has a sensitivity for each protein fraction of 20 mg/l.

The proteins identifiable with SDS-AGE ARE: β_2 microglobulin, lysozyme, retinol binding protein, light chains (κ and λ), α_1 microprotein, dimers of light chains, albumin, transferrin, IgG, IgA, haptoglobin and α_2 macroglobulin. The method is based on the migration of each protein fraction according to its mass (not charge).

In this way it is possible to eliminate interference by high-molecular-weight proteins on the migration of the low-molecular-weight proteins. Distinction of the individual protein bands makes it possible to identify proteins of tubular origin (MW <70,000) and proteins of glomerular origin (MW >70,000).

The scope of the study was the qualitative determination of physiological protein in the dog. Fourteen dogs aged 24 to 48 months (6 whole males, 1 castrated male, and 7 females), with a body surface of 0.60 to 0.92 m², were evaluated preventively to exclude extrarenal causes of proteinuria.

After a period of dietetic pretreatment and restricted physical activity of 4 months, each dog was sedated and subjected to echo-assisted renal biopsy with an 18-gauge Tru-cut.

The biopsy samples were preserved in 10% buffered formalin for conventional optical microscope examination and in liquid nitrogen, after immersion in the appropriate medium, for immunofluorescence examination.

Conventional optical microscope examination was done on samples stained with hematoxylin-eosin, periodic acid-Schiff, trichrome according to Goldner, methenamine, and Congo red. Immunofluorescence examination was performed by testing the samples with IgG, IgA, C₃ complement fraction and fibrinogen.

At the time of the biopsy, a pre-established volume (10 ml) of urine was collected by centesis and kept (after the addition of 1% sodium azide equal to 1 μ l/ml of urine) at 4-8°C. The following analyses were performed on the urine samples: physico-chemical, sediment, proteinuria/creatinuria ratio, determination of proteinuria by a quantitative (staining with pyrogallol) and a qualitative method (SDS-AGE).

All the dogs with inert sediment and with normal conventional optical microscopic, immunohistochemical and immunofluorescence examinations at the qualitative study of proteinuria showed exclusively a minimum band of albumin or the complete absence of protein bands.

SDS-AGE can be considered a useful method, specific and sensitive, for the qualitative determination of physiological proteinuria in the dog.