

THE PLASMA PROTEOME OF EQUUS CABALLUS: PATTERNS UNDER BASELINE CONDITIONS AND DURING ANTHELMINTIC TREATMENT

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Introduction

The present study focuses on the plasma proteome of *Equus caballus*, and investigates its changes during infestation and anthelmintic treatment, paying special attention to acute phase proteins and immunologic reactions. Intestinal parasitic infection causes a variety of clinical symptoms which may occur with different severity, and regular anthelmintic treatment is known to contribute to the animals' wellbeing.

Materials and Methods

Sera from several healthy horses of Norican breed were used to establish the serum protein map. Classical two-dimensional electrophoresis (2-DE) was applied; proteins were identified either by mass spectrometry methods or by immunoblotting and (cross-) reactive antibodies.

For the infestation study, a group of six horses was subjected to oral treatment with ivermectin, blood samples were collected in regular intervals from day 0 to 14. Effectivity of the treatment was checked by parasitological investigations (nematode egg counts from feces). Besides determining hematological values as well as cytokine and IgE expression levels, serum protein patterns were studied as a function of time. A combination of different methods was applied, ranging from Elisa (for serum amyloid A) to SDS-PAGE and 2-DE (including differential image gel electrophoresis).

Results

Equus caballus is one of the species with yet rather incompletely characterized genomes, and thus protein databases lack informations on horse proteins. Part of these problems can be solved by cross-species comparison and check for similarity with homologues from other species. Thus, we were able to identify almost all moderate to high abundance proteins stained in the plasma 2-DE pattern. Compared to maps of other species (human, bovine, pig), the horse map shows several differences concerning positions/patterns and levels of single proteins. Known polymorphisms, e.g. for-antitrypsin (pi), could be detected in specimens from individuals.

Horses in the anthelmintic study showed temporarily increased IgE levels and eosinophile counts as a reaction to treatment. Longer-term investigations of haptoglobin revealed a significant increase a few days after treatment, in all investigated animals. Similarly, a temporary increase of fibrinogen was noticed. In contrast to that, time-courses of serum amyloid A and overall IgG(T) were individual, with considerable to minor fluctuations. Selected samples, from the most interesting period between day 0 and 7, were investigated in more detail by 2-DE. This method allowed resolution and quantification of proteins which cannot be sufficiently separated in one-dimensional methods, and thus gave more insight into variations of single proteins.

Conclusion

The present study shows that - although used as a single dose administration - the drug causes an inflammatory response which is biphasic. This might be taken as a sign that the treatment was effective against different developmental stages of equine intestinal parasites.

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