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ESTABLISHMENT OF AN INTERFERON-GAMMA-RELEASE-ASSAY FOR GLANDERS DIAGNOSIS IN HORSES – FIRST RESULTS

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Purpose

The mallein test, as the only available assay to analyze the cellular immune response in *Burkholderia mallei* -infected animals, was removed recently from the OIE list of mandatory methods. This *in vivo* test can induce a seroconversion in animals leading to false-positive results in the required complement fixation test (CFT). Our intention was to establish a new *in vitro* assay to detect pathogen-specific T-cell immune responses based on the release of interferon-gamma (IFN- γ).

Methods

Initially, a culture of whole blood is incubated with a commercially available ppd mallein and a whole-cell-lysate antigen for 24 h and 48 h. In the second step a sandwich ELISA was adapted to quantify the amount of equine IFN- γ in supernatants, which is an indicator for the activation of specific memory T-cells by antigen-presenting macrophages.

Results

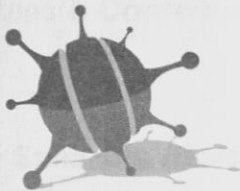
Blood samples of 20 non-infected horses, previously tested negative for glanders by CFT and immunoblot, and one sample of a horse immunized with a whole-cell-lysate-antigen were analyzed. The blood samples of the immunized horse showed a significant increase of IFN- γ concentrations after incubation with whole-cell-lysate-antigen or mallein. With the exception of a few animals, most of the non-infected horses showed no increased IFN- γ levels in response to mallein.

Conclusions

The IFN- γ -release-assay is as easy to handle as the mallein test. However, based on some false-positive results in several non-infected horses we conclude that both antigens used are not suitable. In order to establish a better diagnostic tool, a broad analysis is necessary to study more specific antigens.

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