

ELISA and Western Blot: two useful tools in the diagnosis of *Besnoitia besnoiti* infection

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Abstract

Besnoitia besnoiti is a bovine parasite endemic in many tropical and subtropical areas. In some Mediterranean countries such as Portugal, the prevalence seems to be increasing. Most infections are mild or subclinical, characterized by the formation of numerous cutaneous and sub-cutaneous cysts, which may lower the quality of skins for leather industry. The definitive host of this parasite is still unknown and no treatment is available for the disease so far. Therefore, the present control of *B. besnoiti* infection only relies upon vector control and identification and culling of infected animals. We now describe two assays for the detection of anti-*Besnoitia besnoiti* serum antibodies in cattle, including a somatic tachyzoite antigen-based enzyme-linked immunosorbent assay (ELISA) and a Western Blot, using the same antigen of the Bb1Evora03 parasite strain. To validate and stan-

darize these methods, the diagnostic sensitivity of the methods was investigated with sera from animals with chronic stage of besnoitiosis and asymptomatic but infected animals. Specificity was tested with parasitologically confirmed *Toxoplasma gondii* and *Neospora caninum* sera, including also negative sera from areas of different endemicity. The ELISA offers the possibility for large-scale screening of animals such as in epidemiology survey studies. Sera from animals with besnoitioses correlated well with the Western Blot. The Western Blot offers the advantage to detect potential cross-reactions that can occasionally be found in ELISA. This study provides evidence that ELISA and Western blot, using somatic *B. besnoiti* antigens, are useful tools in the serological detection of animals that have been exposed to *Besnoitia* and which subsequently may either result in an asymptomatic or a symptomatic course of infection.