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Banding Pattern Indicative of Echinococcosis in a Commercial Cysticercosis Western Blot

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Abstract
Objective: A commercial cysticercosis Western blot was evaluated for serological cross-reactivity of sera from patients with alveolar (AE) and cystic echinococcosis (CE).
Methods: A total of 161 sera were examined, including 31 sera from AE-patients, 11 sera from CE- patients, 9 sera from patients with other parasitic diseases and 109 sera from patients with unrelated medical conditions. All AE- and CE-sera were also examined by the echinococcosis Western blot.
Results: More sera from patients with AE than with CE showed cross-reactivity in the form of ladder-like patterns ("Mikado aspect") and untypical bands at 6-8 kDa (71% and 77.4% versus 27.3% and 45.5%, respectively). In contrast, triplets of bands in the area above 50 kDa and between 24 and 39-42 kDa were more frequent in CE than in AE sera. The fuzzy band at 50-55 kDa typical for cysticercosis was absent in all AE and CE sera.
Conclusions: Atypical banding patterns in the cysticercosis Western blot should raise the suspicion of a metacestode infection different from Taenia solium, i.e. Echinococcus multilocularis or E. granulosus, especially when the Mikado aspect and an altered 6-8 kDa band is visible in the absence of a fuzzy 50-55 kDa band.

Key words: Alveolar echinococcosis, Cystic echinococcosis, Cysticercosis, Western blot, Banding pattern, Serology, Cross-reactivity

Introduction

Like cysticercosis, alveolar echinococcosis (AE) and cystic echinococcosis (CE) may affect the central nervous system (CNS) and subcutaneous tissue. In contrast to cysticercosis however, CNS and skin involvement in echinococcosis, especially AE [1] is rare. Still, the multivesicular appearance of the racemose cysticercus in the CNS may be confused with either form of echinococcosis. A peculiar banding pattern seen on a cysticercosis Western blot in a patient with cerebral AE [2] prompted us to further investigate cross-reactivity of AE and CE sera on a commercially available cysticercosis Western blot.

Materials and Methods

Sera: A total of 161 sera from 161 patients were examined in the cysticercosis Western blot. Among these, 31 sera were from patients with parasitologically proven AE, 11 sera were from patients with CE and 1 serum was obtained from a patient with cysticercosis. 3 sera from patients with each schistosomiasis, toxocariasis and trichinellosis, respectively, were also tested. 109 serum samples were derived from patients with unrelated medical conditions. All sera from patients with AE and CE were also examined with the echinococcosis Western blot.

Cysticercosis and Echinococcosis Western blots: The Cysticercosis Western Blot IgG and the Echinococcus Western Blot IgG (both LDBIO Diagnostics, Lyon, France) were used according to the manufacturer's instructions.

Results and Discussion

To our knowledge, no systematic investigation on cross-reactivity of AE sera on cysticercosis blots has been reported. Recently, cross-reactivity of CE sera on an enzyme-linked immunoelectrotransfer blot (EITB) for cysticercosis has been published [3]. In our study, none of the AE and CE sera exhibited a pattern typical for cysticercosis. Instead, cross-reactivity in the form of ladder-like patterns ("Mikado aspect" [4]), untypical bands at 6-8 kDa and the absence of a fuzzy band at 50-55 kDa were observed.

The Mikado aspect was shown in 22 out of 31 AE sera (71%) and in only 3 out of 11 CE sera (27.3%) (Fig. 1). This pattern was also seen in 2 out of 3 sera from patients with schistosomiasis, in 1 out of 3 patients with toxocariasis and in 4 out of 109 patients with unrelated diseases (1 patient with a brain abscess, borreliosis, idiopathic eosinophilia and psychosis each, respectively). According to the manufacturer [4], this non-specific binding pattern may concern only one part of the blot strip and was shown as an example in sera of 1 patient with CE and 2 patients with AE. No frequencies in echinococcosis patients are reported, however.

In this study, 24 out of 31 AE sera showed a narrow single or double band at 6-8 kDa in the cysticercosis

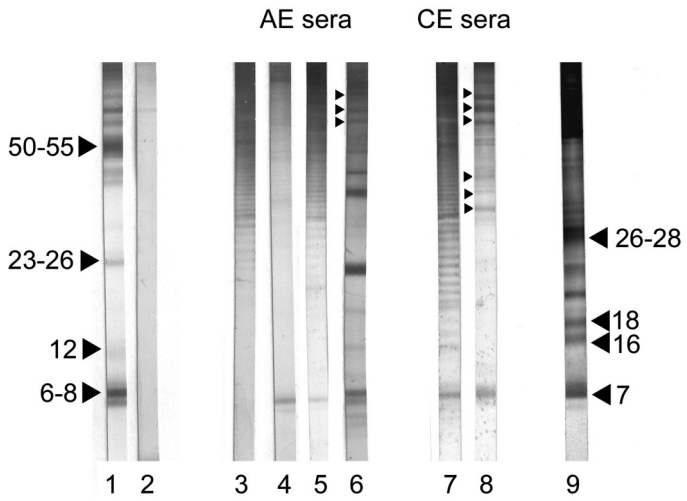

Fig. 1. Examples of cross-reactivity profiles in sera of patients with alveolar and cystic echinococcosis tested in the cysticercosis Western blot.
Strip1 1, cysticercosis positive control. Molecular size markers in kDa are shown. The band at 12 kDa is absent in this serum. **Strip 2**, cysticercosis negative control. **Strip 3**, AE serum with Mikado-aspect only. **Strip 4**, AE serum with band at approx. 7 kDa. **Strip 5**, AE serum with both Mikado-aspect and 7 kDa-band. **Strip 6**, AE serum which demonstrated the T1 triplet (small triangles) and various other bands. **Strip 7**, CE serum with both Mikado-aspect and 7 kDa-band. **Strip 8**, CE serum with 7 kDa-band and triplets T1 (small triangles above 50-55 kDa) and T2 (small triangles below 40 kDa). **Strip 9**, Echinococcus Western blot positive control with AE serum. Molecular size markers in kDa are shown. Abbreviations: AE, alveolar echinococcosis; CE, cystic echinococcosis

Western blot (77.4%). A corresponding large single band at 7 kDa was present in all these sera when tested by the echinococcosis Western blot (Fig. 1). In the remaining 7 AE sera, both the 6-8 kDa and the 7 kDa band in the cysticercosis and echinococcosis Western blot were absent, respectively. In patients with CE, a single or double 6-8 kDa band was present in only 5 out of 11 sera when tested in the cysticercosis blot (45.5%), but all 11 sera showed the 7 kDa band in the echinococcosis blot. Bands at 6-8 kDa were absent in all sera from patients with schistosomiasis, toxocariasis, trichinellosis and unrelated medical conditions in the cysticercosis blot. Much lower frequencies of 50% and 10% for the presence of the 6-8 kDa band in AE and CE sera, respectively, were shown by the manufacturer [4]. In all AE and CE sera that cross-reacted with the 6-8 kDa band in the cysticercosis blot, the band corresponded to the 7 kDa band in the echinococcosis blot. This low molecular-weight band could thus be tapeworm-specific. A combination of both the Mikado aspect and the presence of an atypical band at 6-8 kDa was detected in 58.1% of AE sera and in 27.3% of CE sera.

The so-called T1-triplet of bands above a 50 kDa band seen in 78% of CE sera tested on an EITB for cysticercosis as decribed by van Doorn et al. [3] were not readily identified in our study. However, 2-3 similar bands directly above the 50-55 kDa area were seen in 17 out of 31 AE sera (54.8%) and in 10 out of 11 CE sera (90.9%) in our survey. These 3 bands were also visible in the presence of the fuzzy 50-55 kDa band on the control strip incubated with a cysticercosis positive serum. In all AE and CE sera, however, the 50-55 kDa band itself was absent (Fig. 1). In van Doorn's study [3], a 50 kDa band was present in cysticercosis sera, but also absent in many, if not all CE sera tested (no concise data published). The so-called T2-triplet of bands between 24 and 39-42 kDa, which was infrequently detected in CE sera by EITB [3] was not found in any of our AE sera, but in 7 CE sera (Fig.1). Although the EITB and the Western blot use different methodology, a concordance of 98% was shown for these techniques [5].

In conclusion, atypical banding patterns in the cysticercosis Western blot should be interpreted with caution and should raise the suspicion of a metacestode infection different from *Taenia solium* in conjunction with the clinical status of the patient. The Mikado aspect may possibly indicate an infection with a metazoan parasite other than *T. solium*. The presence of an altered 6-8 kDa band in the absence of a fuzzy 50-55 kDa band could signal a larval infection with a different cestode than *T. solium*, i.e. *E. multilocularis* or *E. granulosus*. A combination of these blot patterns further favours the diagnosis of a possible echinococcal infection. Different serological assays with a higher specificity should then be performed.

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