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RESEARCH NOTE

Human alveolar echinococcosis in Slovenia

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ABSTRACT

Between January 2001 and December 2005, 1263 patients suspected of having echinococcosis were screened serologically by indirect haemagglutination assay (IHA). IHA-positive patient sera were then retested by western blot for confirmation and differentiation between *Echinococcus granulosus* and *Echinococcus multilocularis* infection. Of 43 sera confirmed as *Echinococcus*-positive, nine appeared to be specific for alveolar echinococcosis (AE) caused by *E. multilocularis*. AE-positive serological results corresponded to the clinical and/or imaging findings concerning the patients' liver cysts. The detected incidence of AE was $0.45/10^5$ inhabitants, which suggests that clinicians and health authorities in Slovenia should give greater attention to AE in the future.

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Keywords Alveolar echinococcosis, diagnosis, *Echinococcus*, indirect haemaglutination assay, Slovenia, western blot

Original Submission: 4 September 2006; **Revised Submission:** 10 November 2006; **Accepted:** 18 December 2006

Clin Microbiol Infect 2007; **13**: 544–546
10.1111/j.1469-0691.2007.01701.x

Human echinococcosis is caused mainly by the larvae of the tapeworm *Echinococcus granulosus*, which cause cystic echinococcosis (CE), and by the larvae of *Echinococcus multilocularis*, which can cause alveolar echinococcosis (AE). *E. granulosus* occurs worldwide, but *E. multilocularis* is found only in the temperate northern hemisphere. The main endemic areas of this tapeworm are Alaska, Canada, central North America, some parts of central western Europe, western Turkey, Russia, China, central Asia and northern Japan. At the adult stage, *E. multilocularis* is 1.2–3.7 mm in length and is harboured in the intestine of definitive hosts, typically foxes, and also domestic dogs and cats. The tapeworm eggs are excreted with the faeces. Following accidental ingestion of these eggs, the larval stage of AE may develop in many species of small rodents, and sometimes in humans as intermediate hosts, usually in the liver. AE is potentially fatal and is chronically progressive as a tumour-like hepatic disease [1–6]. The aim of the present study was to examine serologically whether patients in Slovenia suspected of having echinococcosis had been infected by the larvae of *E. multilocularis*.

Between 1 January 2001 and 31 December 2005, 1263 patients suspected of having echinococcosis

(because of lesions in the liver and/or lung, and/or findings from ultrasound scanning or computerised tomography, e.g., different forms, size and number of lesions) were examined serologically. Sera were obtained from patients of both genders and different ages, and from varying areas of Slovenia. The sera were screened by an indirect haemagglutination assay (IHA) (Cellognost-Echinococcosis; Dade Behring, Marburg, Germany).

Low positive IHA titres of 1:32–1:128 should only be accepted as positive for echinococcosis when they are confirmed in conjunction with a second serological method (e.g., immunofluorescence test, ELISA or western blot (WB)). For confirmation and differentiation between CE and AE, all IHA-positive sera were retested by WB IgG (LDBIO Diagnostics, Lyon, France). According to the manufacturer's interpretation data and the evaluation of Liance *et al*. [7], immunoblot patterns P1, with only a 7-kDa band, and P2, with a band at 7 kDa and at least a diffuse band at 16–18 kDa, are specific for CE. Pattern P3, with at least one band at 26–28 kDa plus two sharp bands at 16 and 18 kDa, is the most specific pattern for AE. Pattern P4, with one 26–28 kDa band only, is also *c.* 88% specific for AE. Pattern P5, with a band at 7 kDa and a band at 26–28 kDa only, cannot be used to discriminate between the two species [7]. Because of possible cross-reactivity with neurocysticercosis, *E. multilocularis*-positive sera were retested using an immunoblot assay (Cysticercosis WB IgG; LDBIO Diagnostics).

In 93 of 1263 sera from patients with suspected echinococcosis, antibodies to *Echinococcus* spp. with low-to-high titres were detected by IHA. Of 93 IHA-positive sera, 43 were also positive for echinococcosis by WB. Of these 43 sera, 20 sera with patterns P1 or P2, and 14 sera with pattern P5 were not studied further. Nine of 43 sera appeared to be positive for AE caused by *E. multilocularis*, as the antibodies in these sera bound specifically to antigens of 26–28 kDa. Seven of these nine sera (four with IHA titres of 1:32–1:128, one with a titre of 1:256, one with a titre of 1:512, and one with a titre of 1:1024) were from women, aged 60, 31, 57, 46, 68, 79 and 62 years, respectively, while two sera (IHA titres 1:32 and 1:64) were from men, aged 66 and 65 years, respectively. The serological results corresponded with the clinical data (jaundice, epigastric pain, hepatomegaly, anaemia) and/or the imaging findings (single irregular and/or small multiple, and some calcified lesions in the patients' livers). All AE-positive sera were negative for cysticercosis according to the immunoblot assay.

AE develops almost exclusively in the liver (99% of cases), with a fatality rate of >90% in untreated patients. The typical transmission cycle in Europe involves red foxes as final hosts, and small rodents (different species of voles and mice) as intermediate hosts. In many cities, towns and villages in southern central Europe, foxes have adapted to urban environments and bring the parasite to human settlements. This urban cycle of the parasite in red foxes and their intermediate hosts is likely to lead to an increased risk of infection from domestic dogs and possibly cats [8–10].

In the 1980s, human AE occurred in Europe only in Austria, France, Germany and Switzerland, but since the 1990s, sporadic cases have also been reported in Belgium, Poland, the Czech Republic, Slovakia and Greece [2,3,8,11,12]. AE usually manifests in older patients because of its characteristic asymptomatic incubation period of 5–15 years. The mean age of AE patients in the present study was 59.3 years, which is in agreement with findings published previously [8,12]. The incidence of AE in Slovenia in 2001–2005 has been estimated as $0.45/10^5$ inhabitants, with a mean annual incidence of 0.09 cases/10^5 inhabitants. The patients in the present study were from southern and north-eastern parts of Slovenia. This region is agricultural and is populated by small rodents that are the primary intermediate hosts for transmission of the parasite. Based on a report of AE in Slovenian cattle in 1966 [13] and a report of AE in the *Apodemus flavicollis* mouse in 1984 [14], it seems that human AE may also have been present previously in Slovenia, but remained undetected or overlooked because of insufficient investigations and serological tests unsuitable for accurate confirmation and differentiation between CE and AE. It is also possible that AE might have been transmitted from the southern region of Austria, bordering Slovenia, where *E. multilocularis* was reported in foxes during the 1980s [8].

It is difficult to understand the precise ecology of the parasite and the route of infection of humans. None of the AE-positive patients in the

present study was a farmer or hunter, but most owned, or formerly kept, dogs or cats, and these could be a possible risk-factor for this infection [15,16]. Overall, the results of this study suggest that clinicians and health authorities in Slovenia should give greater attention to AE in the future.

ACKNOWLEDGEMENTS

This work was supported by Grant no. P3-0083 from the Ministry of Higher Education, Science and Technology of the Republic of Slovenia.

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RESEARCH NOTE

Paediatric varicella hospitalisations in France: a nationwide survey

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ABSTRACT

Paediatric patients hospitalised with varicella (n = 1575) were reported to a French national network between March 2003 and July 2005. Superinfection was identified in 50.3% of cases, principally of skin and soft-tissue (36.5%). The risk of superinfection increased with fever relapse, use of non-steroidal anti-inflammatory drugs, prolonged fever, an age of 1–5 years, and contamination at the childminder's home. Neurological complications were observed in 7.8% of cases, while pulmonary complications were less frequent (3.1%). Forty-nine patients had sequelae and eight patients died. Surveillance should continue in France with a view to the future implementation of a universal vaccination programme.

Keywords Complications, paediatric patients, risk-factors, superinfection, surveillance, varicella

Original Submission: 25 July 2006; **Revised Submission:** 11 December 2006; **Accepted:** 19 December 2006

Clin Microbiol Infect 2007; **13**: 546–549
10.1111/j.1469-0691.2007.01706.x

Varicella is a mild infectious disease preventable by vaccination [1,2]. Only high-risk individuals are

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