Available online at www.sciencedirect.com
SCIENCE DIRECT®
Veterinary Parasitology 140 (2006) 177–180
veterinary parasitology
www.elsevier.com/locate/vetpar

Short communication

Human outbreak of trichinellosis in the Mediterranean island of Sardinia, Italy

Edoardo Pozio [a,*], Pietro Mesina [b], Franco Sechi [c], Michele Pira [c], Manuele Liciardi [d], Pasquale Cossu [d], Gianluca Marucci [a], Giovanni Garippa [e], Antonio Firinu [d]

[a] *Department of Infectious, Parasitic and Immunomediated Diseases, Istituto Superiore di Sanità, viale Regina Elena 299, 00161 Rome, Italy*
[b] *Presidio Ospedaliero San Francesco, Department of Infectious Diseases, 08100 Nuoro, Italy*
[c] *Azienda Sanitaria Locale, Veterinary Service, via Manzoni 40, 08100 Nuoro, Italy*
[d] *Istituto Zooprofilattico Sperimentale della Sardegna, via Duca degli Abruzzi 8, 07100 Sassari, Italy*
[e] *Department of Animal Biology, Section of Parasitology, University of Sassari, via Vienna 2, 07100 Sassari, Italy*

Received 23 November 2005; received in revised form 8 March 2006; accepted 13 March 2006

Abstract

Trichinella sp. infection has been documented in both humans and animals in most Mediterranean countries, yet in the past 60 years no infections have been reported on the Mediterranean islands. We describe the first outbreak of *Trichinella* sp. infection to have been reported on the island of Sardinia. The outbreak occurred in two villages in 2005 and involved 11 persons who had eaten raw sausages made from the same free-ranging sow. All 11 persons developed signs and symptoms of trichinellosis and seroconverted within 48 days of consuming the infected meat. The etiological agent was *Trichinella britovi*. Sardinia, like all Mediterranean islands, had been considered to be *Trichinella*-free. The present report, together with a recent report of *T. britovi* infection in animals on the nearby island of Corsica (France), raises questions as to the validity of the concept of *Trichinella*-free areas or regions.

Keywords: Trichinellosis; *Trichinella britovi*; Sardinia; *Trichinella*-free; Outbreak; Pig; Mediterranean sea; Humans; Zoonosis

1. Introduction

Trichinella sp. infection has been documented in animals and/or humans in most Mediterranean countries. The most prevalent species is *Trichinella britovi*, although *Trichinella spiralis* has also been reported (Pozio and Zarlenga, 2005). On the Mediterranean islands, *Trichinella* sp. infection has never been detected, except on Sicily, where between 1933 and 1946 four human outbreaks caused by the consumption of pork from domestic pigs infected with *T. spiralis* were reported (Pozio and La Rosa, 1998).

* Corresponding author. Tel.: +39 06 4990 2304; fax: +39 06 4938 7065.
E-mail address: pozio@iss.it (E. Pozio).

0304-4017/$ – see front matter © 2006 Elsevier B.V. All rights reserved.
doi:10.1016/j.vetpar.2006.03.012

On the Mediterranean islands, the lack of human outbreaks and infections in both domestic and wild animals has strongly reduced the attention of the local veterinary services to controls for *Trichinella* at slaughterhouses. In the present work, we describe the first outbreak of *Trichinella* sp. infection to have been reported on the island of Sardinia.

2. Materials and methods

In early April 2005, five persons living in the same household in the village of Orgosolo (Province of Nuoro) were hospitalised for suspected food intoxication in the town of Nuoro. The clinical patterns (fever, palpebral, facial and/or limb oedema, itching spotted-papular rash, myalgia) and laboratory features (eosinophilia, leukocytosis, increase of muscle enzyme values) were suggestive of trichinellosis. The serological test confirmed the diagnosis. Several days later, five persons living in the village of Lanusei (located several hours from Orgosolo) were hospitalised with a similar clinical pattern at the hospital in the city of Cagliari.

The veterinary services were alerted, and the epidemiological investigation ascertained that all members of the family in Orgosolo had consumed raw homemade sausages from a domestic pig; all of the persons first ate the sausage on the same day (a sixth household member had also eaten the sausage yet was not hospitalized). Some sausages had also been sent as a gift to friends and relatives in Lanusei. The infected sausages had been made with the meat of a 3-year-old free-ranging sow weighing about 100 kg, which was illegally slaughtered at the home of the family in Orgosolo in early February 2005. The free-ranging sow had been bred in Bargagia, a remote mountain area covered by forest, which is close to Orgosolo. The breeder reported that he had provided the sow with food only sometimes and that otherwise the sow was left to find food on its own.

The local veterinary service of Orgosolo confiscated the remaining sausages in the house of the infected persons. Sausages were sent to the International *Trichinella* Reference Centre in Rome to detect *Trichinella* larvae and for further studies. Since the sausages were still frozen and dried, they were rehydrated by cutting them into small pieces and soaking them in water for 24 h. The fat was removed, and *Trichinella* larvae were collected from the sausage by artificial digestion, following standard protocol (Gamble et al., 2000). To perform histology, a piece of muscle was collected from a sausage, fixed in formalin, and embedded in paraffin. Single larvae were identified at the species level by a multiplex PCR analysis (Pozio and La Rosa, 2003), using as reference larvae those of the strains of *T. spiralis* (ISS03), *T. britovi* (ISS02) and *Trichinella pseudospiralis* (ISS013), the only three species that are known to circulate in central-southern Europe (Pozio and Zarlenga, 2005) and are potentially present on Sardinia. Sequencing was performed to investigate the presence of a deletion or a substitution in the highly polymorphic sequence of the expansion segment 5 (ES5) of the large subunit of rDNA. Serum samples were tested by an in-house ELISA using excretory/secretory antigens (Gamble et al., 2004) at the International *Trichinella* Reference Centre in Rome. Briefly, *T. spiralis* ES antigen was diluted in 0.1 M carbonate-bicarbonate buffer (pH 9.6) and used at a concentration of 5.0 μg/ml. Peroxidase-labelled anti-human IgG conjugate (Kirkegaard and Perry Laboratories, Inc., Maryland) was used at 1:10,000 dilution and incubated at 37 °C for 1 h. Control samples consisted of serum samples from five uninfected individuals and from five known to be infected with *Trichinella*, stored at the International *Trichinella* Reference Center. Each serum was diluted 1:200.

3. Results

A mean of eight larvae per gram of sausage meat (excluding the fat) were detected after HCl-pepsin digestion. All of the larvae were dead at this time. The larvae belonged to *T. britovi* (isolate code ISS1571) (Fig. 1). The sequence of the ES5 did not show any difference from that of the *T. britovi* reference strain. The thickness of the collagen capsule and the presence of cellular infiltrates around the larva detected in the histological section suggested that the sow had acquired the infection at least 4 weeks earlier.

Serum samples from the six persons in Orgosolo, taken 24 days after having first eaten the sausage, were positive for anti-*Trichinella* IgG. In Lanusei, serum samples were taken from two of the infected persons 20 days after having first eaten the sausage; one