



Infection of Eurasian badgers (*Meles meles*) with *Mycobacterium bovis* and *Mycobacterium avium* complex in Spain [☆]

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ABSTRACT

The prevalence, distribution and pathology related to infection with *Mycobacterium bovis* and other mycobacteria were determined in trapped ($n = 36$) and road-killed ($n = 121$) badgers in Spain from 2006 to 2010. The prevalence of *M. bovis* based on bacteriological culture from road-killed badgers was 8/121 (6.6%) and from trapped badgers was 0/36 (0%). Tuberculosis/*M. bovis* infection was evident in 15/121 (12.4%) road-killed badgers when bacteriology and histopathology were combined. *Mycobacterium avium* complex was isolated by culture from the tracheal aspirate of 1/36 (2.8%) trapped badgers and from tissue pools from 8/121 (6.6%) road-killed badgers.

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Introduction

The *Mycobacterium tuberculosis* complex (MTC; *M. tuberculosis*, *M. bovis*, *M. caprae*, *M. pinnipedii*, *M. africanum* and *M. microti*) causes disease in humans and domestic and wild animals (Grange et al., 1990; Aranaz et al., 2003). Control of bovine tuberculosis (TB) in cattle can be compromised in areas where a reservoir of infection exists in wildlife. In the United Kingdom (UK) and Republic of Ireland (RoI), Eurasian badgers (*Meles meles*) are involved in the transmission of *M. bovis* to cattle (Bourne et al., 2007; Murphy et al., 2010).

The first case of bovine TB (*M. bovis*) in a Spanish badger was identified in 2003 in Cabañeros National Park in Central Spain (Sobrinho et al., 2008). *M. bovis* was also isolated from lymph nodes of a badger from León in Northern Spain in 1997 (J.F. García Marín, personal communication). In Doñana National Park in Southern Spain, 23% of badgers were seropositive (Martín-Atance et al.,

2006). Elsewhere in continental Europe, *M. bovis* infection in badgers has been confirmed only in France, with a prevalence of 7.2% (Hars et al., 2010).

Mycobacterium avium complex (MAC) spp. were detected by culture of tissues from 7.4% of badgers in Spain and 0.5% of badgers in the UK (Balseiro et al., 2011). *M. avium paratuberculosis* (*Map*) has been isolated from the intestine and mesenteric lymph nodes of a badger in Scotland (Beard et al., 2001). *M. intracellulare* was isolated from the faeces of two badgers in Ireland (Hughes et al., 1993) and from tissues of a badger in Spain (Sevilla et al., 2005). In this study we present data on the prevalence, distribution and pathology of *M. bovis* and other mycobacteria from trapped and road-killed badgers in Spain.

Materials and methods

Collection of samples

Road-killed badgers

From 2006 to 2010, postmortem examinations were performed on 121 badgers (10 cubs and 111 adults; 57 males and 64 females) killed on roads in Spain, mostly from Northern Spain, with smaller numbers from Southern Spain (Fig. 1). Samples of the lungs, intestine and retropharyngeal, submandibular, tracheobronchial,

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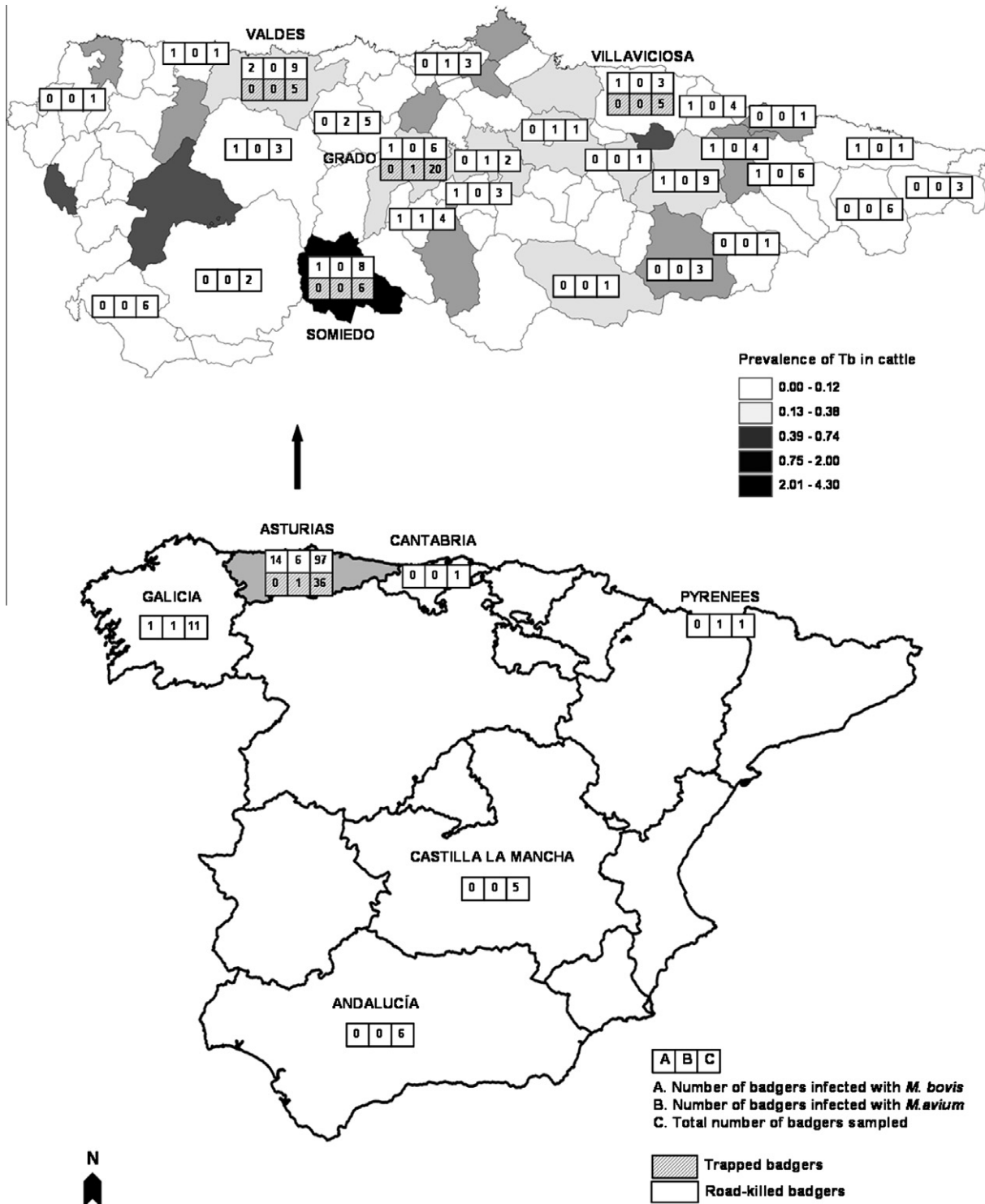


Fig. 1. Geographical distribution of badgers (trapped and killed by road traffic) sampled and infected with *M. bovis* and *M. avium* spp. in Spain. The exploded view of Asturias shows the number and geographical distribution of badgers in Asturias, as well as the badgers infected with *M. bovis* and *M. avium* spp. Note the geographical prevalence of TB in cattle in Asturias (MARM, 2009).

mediastinal, hepatic and mesenteric lymph nodes were collected for bacteriology, molecular studies and histopathology. Serum samples were collected for serology. Samples for culture and real time PCR were frozen at -20°C before processing.

Trapped badgers

Thirty-six badgers (1 cub and 35 adults; 16 males and 20 females) were captured during trapping operations in Asturias, Northern Spain, from April to July in 2009 and 2010 (Animal Research Ethics Committee of SERIDA register number 041/06-01-2008). Traps were located at active sets in four areas with a high incidence of TB in cattle (Fig. 1). Trapped badgers were anaesthetised (de Leeuw et al.,

2004) and samples of faeces (anal swabs), urine (external palpation of the bladder) and sputum (tracheal aspiration) were collected, along with clotted and heparinised blood samples.

Bacteriology

Mycobacterium tuberculosis complex and *Mycobacterium avium* complex

The Mycobacteria Growth Indicator Tube (MGIT) liquid medium system, Löwenstein–Jensen solid media with sodium pyruvate and Coletos solid media (Coletos, 1960; Idigoras et al., 2000) were used to isolate members of the MTC

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