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# Role of wild ruminants in the epidemiology of bluetongue virus serotypes 1, 4 and 8 in Spain

Ignacio García-Bocanegra<sup>1\*</sup>, Antonio Arenas-Montes<sup>1</sup>, Cristina Lorca-Oró<sup>2</sup>, Joan Pujols<sup>2</sup>, Miguel Ángel González<sup>3</sup>, Sebastián Napp<sup>2</sup>, Félix Gómez-Guillamón<sup>4</sup>, Irene Zorrilla<sup>5</sup>, Elena San Miguel<sup>6</sup> and Antonio Arenas<sup>1</sup>

## Abstract

Although the importance of wild ruminants as potential reservoirs of bluetongue virus (BTV) has been suggested, the role played by these species in the epidemiology of BT in Europe is still unclear. We carried out a serologic and virologic survey to assess the role of wild ruminants in the transmission and maintenance of BTV in Andalusia (southern Spain) between 2006 and 2010.

A total of 473 out of 1339 (35.3%) wild ruminants analyzed showed antibodies against BTV by both ELISA and serum neutralization test (SNT). The presence of neutralizing antibodies to BTV-1 and BTV-4 were detected in the four species analyzed (red deer, roe deer, fallow deer and mouflon), while seropositivity against BTV-8 was found in red deer, fallow deer and mouflon but not in roe deer. Statistically significant differences were found among species, ages and sampling regions. BTV RNA was detected in twenty-one out of 1013 wild ruminants (2.1%) tested. BTV-1 and BTV-4 RNA were confirmed in red deer and mouflon by specific rRT-PCR.

BTV-1 and BTV-4 seropositive and RNA positive wild ruminants, including juveniles and sub-adults, were detected years after the last outbreak was reported in livestock. In addition, between the 2008/2009 and the 2010/2011 hunting seasons, the seroprevalence against BTV-1, BTV-4 and BTV-8 increased in the majority of provinces, and these serotypes were detected in many areas where BTV outbreaks were not reported in domestic ruminants. The results indicate that wild ruminants seem to be implicated in the dissemination and persistence of BTV in Spain.

## Introduction

Bluetongue (BT) is a vector-borne disease caused by a group of viruses belonging to the genus *Orbivirus*, which is mainly transmitted between vertebrate hosts by biting midges of the genus *Culicoides*. To date, 24 distinct BT virus (BTV) serotypes have been identified; three of which were found in Andalusia (southern Spain) during the last decade. In October 2004, BTV-4 was detected in Cádiz Province (south-west Andalusia), and a total of 316 outbreaks were reported in livestock. The last BTV-4 outbreak was detected in October 2005, and the country was declared free of BTV-4 in March 2009 [1]. However, in October 2010 a new BTV-4 outbreak was detected in Cádiz, and until December 2010 eight further outbreaks were reported.

In July 2007, a new BT epidemic, caused by BTV-1, affected southern Spain. BTV-1 causes clinical disease to

both sheep and goats and 4446 outbreaks were detected from July 2007 through December 2008 in Andalusia. While no BTV-1 outbreaks were detected during 2009, two new cases were confirmed at the end of 2010 in the Jaén Province (northern Andalusia) [2]. Furthermore, BTV-8 was detected in northern Spain in January 2008, and arrived in Andalusia in October 2008. Until March 2009, 23 BTV-8 outbreaks were reported, followed by a period without apparent cases. In November 2010, a new BTV-8 outbreak was confirmed.

A mandatory vaccination program in domestic ruminants against serotype 4 started in 2004 and was maintained until 2008, two years after the last outbreak was reported. In 2010 vaccination against BTV-4 was resumed in the southern provinces of Andalusia to prevent introduction from northern Africa [3]. Since November 2006, animals have also been vaccinated against serotype 1, and since the end of 2008 also against BTV-8. Currently, BTV-1, BTV-4 and BTV-8 are endemic in Andalusia, and therefore the southern regions of Spain are considered restriction zones for

\* Correspondence: nacho.garcia@uco.es

<sup>1</sup>Departamento de Sanidad Animal. Facultad de Veterinaria, UCO, Campus Universitarios de Rabanales 14071, Córdoba, Spain

Full list of author information is available at the end of the article