Bovine coronaviruses from the respiratory tract: antigenic and genetic diversity

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Introduction

Bovine coronaviruses (BoCV) have been associated with bovine respiratory diseases (BRD) in cattle pulled for treatment in feedlots and from healthy cattle in the US with BoCV identified by virus isolation from nasal swab and bronchoalveolar lavage (BAL) samples, and also by seroconversions for the detection of active infections. Bovine coronaviruses have been identified in pneumonic lungs from field cases, often in combination with other viruses and bacteria including *Mycoplasma spp*. Experimentally, BoCV infections have caused respiratory tract lesions. It has been suggested there is a dual tropism of BoCV for the respiratory and enteric tracts. Historically, BoCV was associated with neonatal enteric disease and adult winter dysentery. Respiratory isolates were identified later and focused on post-weaning cattle entering feedlots. Control measures are limited. There are USDA-licensed killed and modified live virus (MLV) vaccines for BoCV; however, these are for control of the enteric disease in cattle. There are no USDA-licensed vaccines for control of BoCV associated BRD in cattle. The purpose of the study was to compare BoCV isolates from the respiratory tract of cattle in Oklahoma to reference respiratory tract and enteric strains via antigenic and genetic procedures.

Materials and Methods

The evaluated BoCV were from cattle at an experimental feedlot, and included samples from three studies in 2009 and one in 2011. Cattle were auction market origin and were commingled at auctions and transported to OSU. Nasal swab samples, or in some cases BAL fluids, were collected at entry and post arrival. Acute and convalescent serum samples were collected and tested for antibodies by a virus neutralization test (VNT). Filtered nasal swab samples and BAL fluids were inoculated on HRT cells for virus isolation.

Reference strains for the VNT used included the reference BoCV NVSL enteric strain and representative respiratory BoCV from the above studies to determine their ability to neutralize the respective challenge virus. A reference monoclonal antibody prepared against enteric BoCV was also included.

Genetic analysis was made with primers which amplified the polymorphic region of the S glycoprotein region (envelope).Those sequences were compared to 17 sequences listed in GenBank for BoCV, including sequences from field strains from Japan and Korea, reference, and vaccine BoCV strains. The phylogenetic analysis was done by use of the Mega program. There were 72 Oklahoma isolates submitted and 56 were able to generate sequence. There were multiple isolates from the same animal representing nasal swab and BAL samples plus different collections with identical sequences, resulting in 22 unique samples.

Results

From the 56 BoCV samples, there were 22 unique sequences that were submitted to Genbank. On the basis of phylogenetic analysis, these BoCV group into a clade with viruses previously isolated from cattle with BRD. The clade (Clade 2) was distinct from a clade comprised of viruses isolated from calves with enteric infection/ disease and vaccine strains (Clade 1). Viruses from the two clades could be distinguished antigenically using convalescent antisera from US cattle recovering from respiratory disease.

Significance

This is the first large scale study of recent BoCV of US origin to evaluate the genetic and antigenic diversity of the virus. Bovine coronaviruses have been isolated from multiple BRD outbreaks, indicating that BoCV plays a role in BRD. Also, current BoCV vaccines, licensed with a label to control BoCV enteric disease, may have limited efficacy in control of respiratory BoCV.

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