

Distribution of bluetongue in the United States of America, 1991-2002

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Summary

Bluetongue virus (BTV) distribution in the United States of America (USA) is limited by the range of the vector *Culicoides* spp. Regional differences exist with the north-eastern states being free of BTV, while the central and north-western states are seasonally free of virus. Activity of the virus can be observed throughout the year in the southern USA. Serological evidence defining the distribution of BTV in selected regions of the USA is gathered regularly through serological surveys conducted on samples from slaughter cattle. From 1991 to 2002, ten serological surveys were completed. Results from Alaska, Hawaii, Michigan, Minnesota, New York, Wisconsin and New England consistently demonstrated a seropositive rate of less than 2%, confirming BTV-free status. Antibody against BTV was sporadically detected in cattle originating from states contiguous to the BTV-free regions. Additional information on BTV distribution in the USA is obtained through identification of BTV or BTV RNA in diagnostic, surveillance and export specimens submitted to the National Veterinary Services Laboratories. Results confirm that BTV serotypes 2, 10, 11, 13 and 17 are present in the USA.

Keywords

Bluetongue virus – *Culicoides* – Epizootic haemorrhagic disease virus – Isolation – Serotype – Survey – United States of America.

Serological surveys

To assess the distribution of bluetongue (BT) virus (BTV) in the United States of America (USA), a nationwide survey was conducted in the winter of 1977/1978 (4). In this comprehensive survey, market cattle samples from eighteen northern and north-eastern states including Alaska and Hawaii had $\leq 1.0\%$ seropositive samples in the BTV complement fixation test. Subsequently, 19 regional surveys incorporating subsets of the 50 states have been completed. Results of nine regional surveys conducted prior to 1991 were reported previously (6). Since 1991, ten BTV serological surveys utilising market cattle samples have been completed.

The agar gel immunodiffusion (AGID) test supplanted the complement fixation test as the primary BTV antibody detection method for regional surveys completed between 1979 and 1992 and the BTV competitive-enzyme-linked immunosorbent assay (c-ELISA) was used to test serum collected in surveys from 1993 to 2002. Both c-ELISA and AGID are current OIE-prescribed BT serological methods for international trade (1). For each survey,

samples were collected in late autumn and early winter in order to optimise detection of current year exposure to BTV. In this report, the surveys are described by calendar year of sample collection. For each survey, the majority of serum samples were collected in the specified calendar year; in some surveys a few samples were collected as late as mid-January of the subsequent year. At least 600 samples per region were tested for each survey. If all 600 samples tested negative, a 95% confidence level indicated that the true incidence rate for the region fell between 0.0% and 0.5%. Historically, a seropositive rate of less than 2% per region has been required to mitigate United States cattle export requirements for movement into Canada.

‘Traditional states’ survey results (1991-2002)

Surveys of cattle from 18 traditionally BTV-free states comprising 11 regions were conducted annually from 1991 to 1996. Biennial surveys were conducted in 1998, 2000 and 2002. States included in all regional surveys were as follows:

- Connecticut
- Delaware

- Indiana
- Maine
- Maryland
- Massachusetts
- Michigan
- Minnesota
- New Hampshire
- New Jersey
- New York
- North Dakota
- Ohio
- Pennsylvania
- Rhode Island
- Vermont
- West Virginia
- Wisconsin.

Regions consisted of a single state with the exceptions of New England (Connecticut, Maine, Massachusetts, New Hampshire, Rhode Island and Vermont) and the combinations of Maryland/Delaware and Pennsylvania/New Jersey. Ohio and West Virginia were sampled as separate regions for surveys from 1991 to 1996 but were combined into a single region for surveys from 1998 to 2002. An abbreviated survey was conducted in 1997 and included only six traditionally surveyed states comprising four regions namely: Indiana, Ohio, Maryland/Delaware and Pennsylvania/New Jersey.

For five regions, i.e. New England, Michigan, Minnesota, New Jersey and Wisconsin, BTV seropositive samples did not exceed 2.0% in any survey. The numbers of BTV antibody-positive samples for all surveys from 1991 to 2002 were as follows:

- New England 28/8 808 (0.3%)
- Michigan 16/5 999 (0.3%)
- Minnesota 40/6 298 (0.6%)
- New Jersey 15/6 912 (0.2%)
- Wisconsin 24/6 028 (0.4%).

For surveys (1991-1996) in which West Virginia was evaluated as a region, the seropositive rate was less than 2.0% each year with an overall average of 0.7%. The New Jersey/Pennsylvania region samples averaged 1.2% seropositive in surveys conducted from 1991 to 2002 and fell below 2.0% positive samples in all surveys, except in 1996. In 1996, this region revealed 2.2% seropositive samples. For seven of nine surveys that included North Dakota (1991, 1993-1996, 1998 and 2000), the seropositive

rate fell below 2.0%. The North Dakota region samples exceeded 2.0% positive only in 1992 (2.7%) and 2002 (5.7%). The region of Delaware/Maryland had less than 2% positive samples in 1991-1995 and 1997-1998, but exceeded 2.0% positive samples in 1996 (2.5%). In 2000 and 2002, insufficient numbers of samples were collected from Delaware/Maryland for inclusion in the survey data. For surveys (1991-1996) where Ohio was considered a region, Ohio samples exceeded 2.0% seropositive twice with 2.2% in 1992 and 2.6% in 1996. When Ohio/West Virginia was examined as a region, the seropositive rate fell below 2.0% in 1998 and 2002 but was 3.6% in 2000. The BTV seropositive rate for samples from Indiana was less than 2% for seven of ten surveys but exceeded 2.0% in 1992, 1993 and 1996, with 4.2%, 2.3% and 2.5% seropositive rates, respectively. Table I illustrates the seropositive rates per survey for the traditional regions.

Samples from 'non-traditional' states (1991-2002)

In addition to the traditional regions surveyed, samples from additional states were included in some of the serological surveys of market cattle. Samples from Alaska and Hawaii were examined in annual surveys from 1991 to 1996 and BTV antibody testing revealed negligible seropositive rates (0.0-0.9%) for these regions. Virginia was included in surveys from 1991 to 1993, Illinois in 1992 and 2002, eastern and western Washington in 1996, South Dakota in 2000 and 2002, Oregon in 2000 and Idaho, Iowa, Montana and Wyoming in 2002. Samples from western Washington had a 1.3% BTV seropositive rate in the 1996 survey. However, samples from Iowa, Idaho, Illinois, Montana, Oregon, South Dakota, Virginia, eastern Washington and Wyoming exceeded 2.0% seropositive in all surveys in which they were included. Figure 1 illustrates the composite results of BTV antibody detection in regional surveys conducted from 1991 to 2002.

Bluetongue virus isolation and bluetongue virus RNA detection in animals in the United States of America (1991-2002)

Disease due to BTV infection is observed only sporadically in domestic animals in the USA. The OIE-prescribed BTV isolation method of intravenous inoculation of embryonating chicken eggs followed by cell culture passage (1) is lengthy, requiring 4 to 5 weeks, and is limited to facilities that have appropriate resources and personnel with expertise in the procedures. Consequently, few diagnostic laboratories in the USA attempt BTV isolation. The nested polymerase chain reaction

isolation and PCR methods were employed. BTV isolates were subjected to further testing by PCR (2) or virus neutralisation to determine the serotype. When PCR was the only method identifying BTV nucleic acid in a sample, serotype identification was not performed.

In all years from 1991 to 2002, BTV was isolated from blood or tissues submitted to the NVSL. Species from which BTV isolates were obtained include bighorn sheep, bison, cow, elk, goat, domestic sheep, Persian gazelle and white-tailed deer. Additional species in which BTV was detected only by PCR were gerenuk, mule deer and pampas deer. BTV serotypes 17 and 11 were isolated most frequently during this period followed by BTV serotypes 13 and 10. Although there had not been evidence of BTV serotype 2 activity since 1986 (6), it was isolated from sheep residing in Florida in 1999. Figure 2 illustrates the states from which BTV was detected by PCR and/or virus isolation and shows the distribution of BTV serotypes observed in submissions to the NVSL for the years 1991-2002.

Epizootic haemorrhagic disease

Owing to similarities in transmission and clinical presentation, epizootic haemorrhagic disease virus (EHDV) infection is frequently considered among differential diagnoses with BTV. Virus isolation in cell culture and PCR were used to identify EHDV in specimens submitted to the NVSL (5, 7). Figure 3 illustrates EHDV isolate distribution data for NVSL submissions from 1991 to 2002. EHDV serotypes 1 and 2 exist in the USA. From 1991 to 2002, isolation of EHDV serotype 2 was more common than isolation of EHDV serotype 1. EHDV serotype 2 was isolated from bighorn sheep, cattle, mule deer and white-tailed deer during this period. EHDV serotype 1 was isolated from deer (species not specified) in California and white-tailed deer in New Jersey. EHDV was also detected by PCR in bovine samples from Illinois and Florida but the serotype was not determined. Although EHDV infections are often subclinical in domestic species, EHDV contributed to morbidity in cattle from the mid-western portion of the USA during the late 1990s (3).

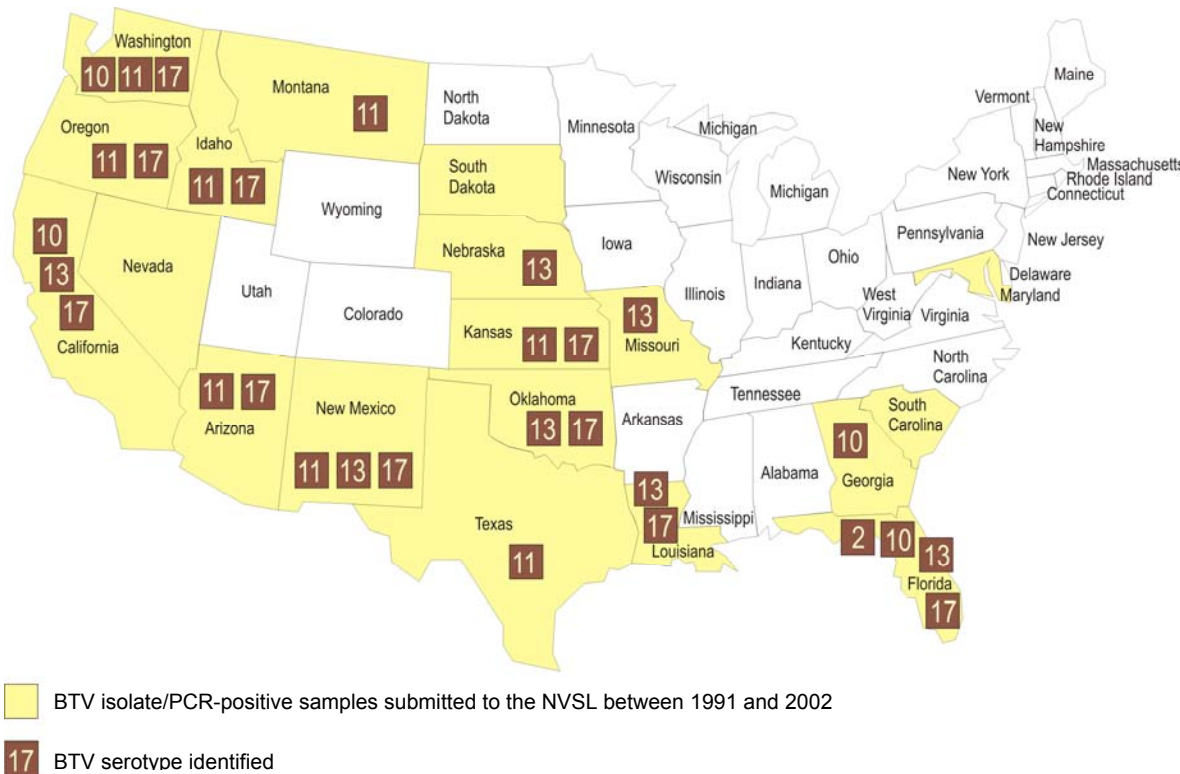


Figure 2
Bluetongue virus (BTV) isolation and bluetongue virus RNA identification between 1991 and 2002
States from which BTV isolates and PCR positive samples were identified from samples submitted to the NVSL between 1991 and 2002
Source: National Veterinary Services Laboratory, Ames, Iowa

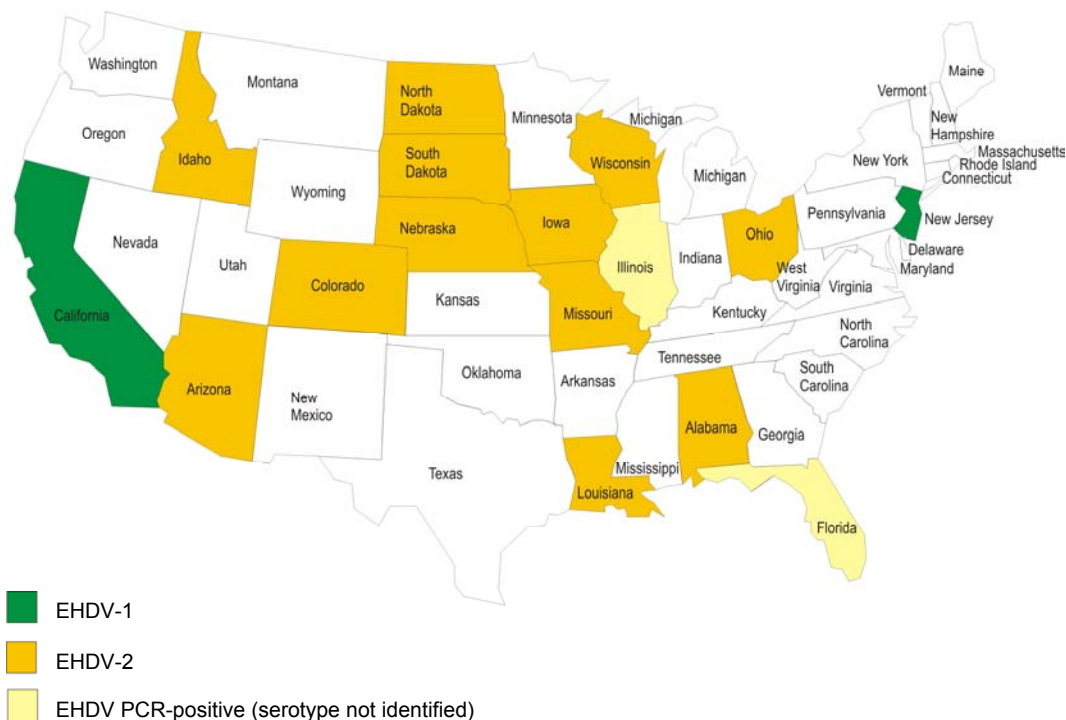


Figure 3
Epizootic haemorrhagic disease virus (EHDV) isolation and RNA identification between 1991 and 2002
States from which EHDV isolates and PCR-positive samples were identified from samples submitted to the NVSL between 1991 and 2002

Source: National Veterinary Services Laboratory, Ames, Iowa

Conclusions

Taken together, the results of regional serosurveys and virus isolation/identification help define the distribution of BTV in the USA. Based on serosurveys conducted between 1977 and 1997, the states of Alaska and Hawaii are BTV-free. In all 19 market cattle surveys conducted since 1977, cattle from the geographic region of New England and the states of Michigan, Minnesota, New York and Wisconsin have had low (less than 2%) seroprevalence for BTV antibodies. BTV isolation has not been reported from animals in these regions. These northern and north-eastern states comprise a BTV-free zone of the continental USA. Animals from regions bordering the BTV-free zone are customarily seronegative for BTV antibodies. The regions of Delaware/Maryland, Ohio/West Virginia, Pennsylvania/New Jersey, Indiana and North Dakota have had less than 2% BTV antibody-positive samples in the majority of market cattle surveys. No BTV isolation from animals in these states was obtained at the NVSL in the past decade although BTV RNA was detected in one sheep from Maryland in 2000. Agent identification and serological survey results for remaining states indicate presence of BTV, at least seasonally. Since

1991, all five known USA serotypes of BTV (types 2, 10, 11, 13 and 17) and both EHDV serotypes 1 and 2 have been isolated, documenting that these viruses are present in the USA. The distribution of EHDV is similar to that of BTV. No additional serotypes of either virus have been identified in animals in the USA.

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