

# Seasonal effects on the prevalence of bluetongue in small ruminants in west Azarbaijan, Iran

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## Key words:

Bluetongue virus, ELISA test, antibodies, relevant factors, seroprevalence.

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## Abstract:

**BACK GROUND:** Bluetongue virus (BTV) is recognized to infect ruminants all around the world, and its prevalence mainly depends on the BTV situation, vectors distribution, weather patterns and susceptibility of the host. **OBJECTIVES:** The purpose of this study was to draw a correlation between the prevalence of BTV antibodies and climate changes in Iranian Azerbaijan Province. **METHODS:** Sera samples were collected from sheep, in a period of a year between 2008- 2009. The seroprevalance of Bluetongue antibodies were evaluated using an Enzyme-linked Immunosorbent Assay, considering monthly weather changes in the Province, recorded by the Iran Meteorological Organization. **RESULTS:** The infection rate was found to be 55.9% and there was a peak BTV prevalence of at 81% ( $p < 0.001$ ). The Pearson correlation coefficient ranges were found to be between 0.36 and 0.005. **CONCLUSIONS:** The vector distribution is based on climatic changes and environmental conditions. It was concluded that the phenotypic expression of the BTV gene may be influenced by the weather.

## Introduction

Bluetongue Virus (BTV) is an infectious, non-contagious, genus (or subfamily) within the reoviridae family and an insect born virus disease found in ruminants with varying clinical severity. Clinical disease is primarily found in sheep, although cattle and goats may be affected (Baldet et al., 2003). Twenty-five serotypes of BTV have been recognized worldwide (Bando et al., 1975; Gard et al., 1989).

Serodiagnosis of this disease is based on both antigen (P2 & P7) and antibody detection and can be determined based upon the presence of the P7 protein (Martyn et al., 1990).

BTV was first reported more than 125 years ago with the introduction of European breeds of sheep to Southern Africa. BTV is traditionally understood as occurring around the world in a broad band stretching from about 35° S to 50° N, although in certain areas it may extend up to around 50° N. This large distribution

is a reflection of the distribution of its Culicoides vectors and the temperature required for BTV replication in, and transmission by, the vectors (Dulac et al., 1989; Zhang et al., 1999, 2004; Lundervold et al., 2003; Carporade et al., 2003). BTV is currently recognized to infect domestic ruminants on the continents of Africa, Asia, North America, South America and Australia; and, several islands in the tropics and sub-tropics (Zhang et al., 2004; Mac Lachlan et al., 2003). Prevalence of BTV in Iran depends on the BTV situation, the vectors distribution, the weather patterns and the susceptibility of the host (Homepage of Handistatus, 1966-2009). BTV incidence is determined largely by the distribution of insect vectors (midge Culicoides species) which is, in turn, primarily driven by weather patterns, including temperature, humidity, wind stream and rainfall. For a year, monthly weather reports were recorded officially according to reports of the Iran Meteorological Organization. The aim of

this study was to investigate the seroprevalence of BTV in sheep in the Iranian western provinces, without considering the ruminant's age and sex, and to apply the scientific assumption that alkalization makes an inhospitable environment for the larval reproductive sites in the western provinces. A study concerning the reproductive behavior of BTV vectors has been reported (Baldet et al., 2003; Dulac et al., 1989). The first data about Bluetongue (BT) history in Italy, especially in Sardinia, has been gathered. This report has provided an epidemiologic retrospective view of the events in the whole island. The present study was issued for disease prevalence; and how the environmental and geo-pedagogical factors can change the prevalence. It was also conducted on pilot farms in selected areas of the western provinces where the first hypothesis has been tested and all the information about larval development has been collected. During the whole experiment, data was collected which included all data about captures in the light traps; and, captures in the traps where midges were emerging from the larval case, alkalized or not.

## Materials and Methods

Serum samples from nine hundred eighty one sheep were collected from the Iranian province of Azerbaijan from April, 2008, to March, 2009. Following the manufacturer's instructions, the ELISA kit (BT, France) was used for the detection of BTV antibodies. Suspected serum samples were diluted and incubated in the wells pre-coated with VP7 protein, to find an antibody specific to VP7. An anti-VP7 antibody, coupled to peroxidase, was added in the wells. If the suspected serum contained specific VP7 antibodies, the conjugate can bind on the VP7. Plates were washed twice, the enzyme substrate, tetramethyl benzidine was added to the conjugate forming a blue compound which become yellow after blocking. The intensity of the color was in inverse measure of the proportion of anti-VP7 antibodies present in the suspected serum samples.

## Results

Results showed, 548 (55.9%) out of the total 981 sera samples were found to be positive and have specific antibodies against BTV (Table 1).

Table 1. The frequency of the serological (ELISA) test in different months.

Month	Results			
	-	100%	+	0%
April	12	100%	0	0%
May	41	58.6%	29	41.4%
June	45	34.1%	87	65.9%
July	55	53.9%	47	46.1%
August	29	30.2%	67	69.8%
Sep	10	16.4%	51	83.6%
Oct	24	92.3%	2	7.7%
Nov	21	25.9%	60	74.1%
Dec	19	21.8%	68	78.2%
Jan	78	59.5%	53	40.5%
Feb	83	67.5%	40	32.5%
March	16	26.7%	44	73.3%
Total	433	44.1%	548	55.9%

The proportion of the positive cases in different months was significant ( $p < 0.001$ ).

The frequencies of positive cases were from spring to autumn, especially in the summer.

## Discussion

In Iran, the usual range for Bluetongue disease is between latitude 35°S and 40°N. The ranges, especially in the western provinces of Iran, are between 42° to 44° N. It is common to find seasonal migrations of animals in the study areas in these provinces. This may suggest that BTV has been endemic for some time in the western provinces of Iran (Homepage of Handistatus II 1066-2009). During this study, the majority of the samples were collected from different areas along seasonal migrations (for both men and animals), however, there is no relationship between seroprevalence and movement patterns, either spatially or by out distribution. The apparent absence of the BTV antibody in negative sera (44.1%) may reflect a difference between areas of the provinces, their inhabitants, and their interaction with vectors and climate changes (Jenning et al., 1980; Sellers et al., 1978). Analysis of weather variables in the provinces are worthy of mention in that there was no correlation between temperature and wind speed. The Pearson Correlation Coefficient between temperature and the mean of sunny hours is  $r=0.965$  ( $p < 0.001$ ). The correlation between temperature and rainfall was  $r=0.93$  ( $p < 0.001$ ); the correlation between temperature and humidity  $r=0.975$  ( $p < 0.001$ ); the correlation between the mean of sunny hours and rainfall was  $r=0.885$  ( $p < 0.001$ ) (Maclachlan et al.,

Table 2. Mean and standard deviation of climate factors in positive and negative sera. The significant criteria were considered as a correlation between the incidence of disease (or life cycle of Culicoides) and the weather (to account for linearity in the seroprevalence-weather relationship). Data were analyzed statistically by use of Chi square ( $p < 0.001$ ). \*SD = Standard Deviation.

Climate changes	Number		Positive results		Negative results		P-Value
	Positive	Negative	Mean	SD*	Mean	SD*	
Temperature (°C)	548	433	12.38	9.9	10.41	10.2	0.002
Speed of wind (Km/hr)	548	433	12.81	1.09	13.22	1.29	0.000
Sunny hours	548	433	245.88	113.42	226.03	105.750	0.005
Rainfall	548	433	52.30	47.83	64.70	49.61	0.000
Humidity (%)	548	433	61.57	6.40	62.77	6.35	0.003

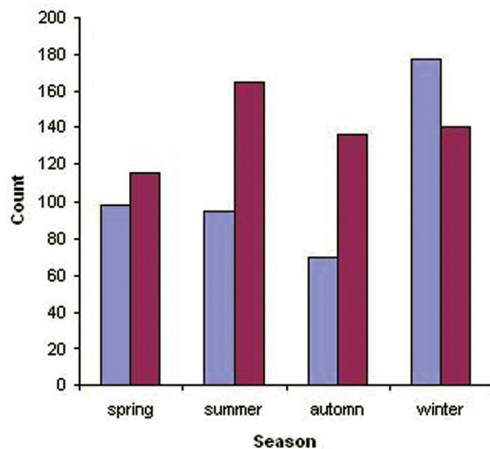


Figure 1. Incidence of Bluetongue disease in different seasons.

■ Negative ■ Positive

2003). In all these conditions there is a higher probability of the incidence of BTV. There was not a significant correlation between the speed of wind and the mean of sunny hours ( $r=0.36$ ); and between the speed of wind flow and humidity ( $r=0.005$ ). In comparison, the correlation between the climate changes and serological tests showed there was a significant relationship between positive and negative sera vector distribution based on climatic and environmental conditions. Also, because one gene controls BTV competency in the vector, phenotypic expression of the gene may be influenced by temperature, rainfall, soil PH, and other factors. (Martyn et al., 1990). The role played by these vectors in the entering of BTV in *Culicoides* spp, appears to be based on temperature. In the cases when the environmental temperature is not sufficient for complete viral protein assembly, the incomplete virus will remain in the intestinal cells of the vector until the critical temperature is reached to allow for virus assembly. So, there is a relationship between climate changes in different months and seroprevalence of

the disease. It was found, that positive sera samples represent an increase in the rate of infection at the end of March to the middle of November. There is a peak, in numbers of incidence, during the summer (the end of June) at the rainy season and before the beginning of the hot season. Therefore, the provinces are located in incursion zones where BT appears every decade (Viral Animals Disease Research and Diagnosis Department, Razi Institute Report, 2000). So, the association with climate changes seems to be one of the most probable factors for the appearance of BT. Research has shown the correlation between ecological specification of the vector (*Culicoides*) and different weather throughout seasons (Sellers et al., 1978).

## References

1. Bando, B.M. (1975) Isolation of Bluetongue and Epizootic Hemorrhagic Disease Viruses in Cell Culture, 18<sup>th</sup> Annual Proceeding. American Association of Veterinary Laboratory Diagnosticians.
2. Baldet, T., Baylis, M., Belli, G. (2003) Vector of Bluetongue Virus, 3<sup>th</sup> International Symposium on Bluetongue, Taormina, Sicily (Italy), Oct) 26-29.
3. Carporade, V., Osburn, B., Daniels, P. (2003) Control and Trade BT, 3<sup>rd</sup> International Symposium on Bluetongue. Taormina, Sicily (Italy).
4. Dulac, G.C., Dubuc, C., Myers, D.J., Afshar, A., Taylor, E.A. (1989) Incursion Bluetongue Virus Type 11 and Epizootic Hemorrhagic Disease of Deer Type 2 for Two Consecutive Years in the Okanagan Valley. *Can. Vet.J.* 30: 351-353.
5. Gard, G.P., Melville. (1989) Investigation of bluetongue

- and other arboviruses in the blood and serum of naturally infected bulls. *Vet. Microbiol.* 20: 315-322.
6. Homepage of Handistatus II (1966-2009) Annual Animal Disease Status. Reports on outbreaks of BT. 1966-2002.
  7. Jennings, M., Boorman, J. (1980) Use of the indirect fluorescent antibody technique for the detection of BTV antigen in tissue smears from *Culicoides varipennis*. *Vet. Microbiol.* 5: 13-18
  8. Knipe, D.M., Howley, P.M. (2001) *Fundamental Virology*. (4<sup>th</sup> ed.). Lippincott Williams and Wilkins, Philadelphia, USA.
  9. Linder Vold, M., Milner-Gulland, E.J., Ocallagan, C.J., Hamblin, C. (2003) First evidence of bluetongue virus in Kazakhstan. *Vet. Microbiol.* 92:281-187.
  10. Martyn, C.J., Gould, A.R., Eaton, B.T. (1990) High Level Expression of the Core Protein VP7 and the Non-structural Protein NS3 of Bluetongue Virus in Yeast: Use of Expressed VP7 as a Diagnostic Group-reactive Antigen in a Blocking ELISA. *Virus Record.* 18: 165-178
  11. Maclachlan, N.J., Caporale, V., Pearson, J.E. (2003) Critical Conclusion and Finding from the Symposium on BT. 3<sup>rd</sup> International Symposium on BT. Taormina, Sicily, Italy.
  12. Sellers, R.F., Pedgley, D.E., Tucker, M.R. (1978) Possible Windborne Spread of BT to Portugal. *J. Hyg. Camb.* 81:189-196.
  13. *Viral Animals Disease Research and Diagnosis Department* (2000) Razi Institute, Annual Report, Karaj, Iran.
  14. Zhang, N., Li, Z., Zhang, F., Zhu, J. (2004) Studies on Bluetongue Disease in the People's Republic of China. *Vet. Ital.* 50:31-36.
  15. Wittmann, E.J., Baylis, M. (2000) Effects on *Culicoides* transmitted viruses and implication for the U.K. *Vet. J.* 16:107-117.

## اثرات فصل در شیوع بیماری زبان آبی نشخوارکنندگان کوچک در استان آذربایجان غربی- ایران

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### چکیده

**زمینه مطالعه:** ویروس بیماری زبان آبی عامل آلودگی نشخوارکنندگان کوچک در سراسر جهان می شود و اساساً میزان شیوع بیماری بستگی به وضعیت ویروس، میزان انتشار ناقلین ویروس، شرایط آب و هوایی و حساسیت میزبان دارد. **هدف:** هدف اصلی از این مطالعه یافتن رابطه ای بین میزان شیوع بیماری و تغییرات آب و هوایی در استان آذربایجان بود. **روش کار:** نمونه های سرمی گوسفندان در فاصله زمانی سال های ۲۰۰۸-۲۰۰۹ جمع آوری گردید و میزان آنتی بادی سرمی با استفاده از آزمون الیزاشناسائی گردید. تغییرات آب و هوایی هم بصورت ماهیانه در این استان بوسیله سازمان هواشناسی ثبت گردید. **نتایج:** میزان آلودگی ۵۵/۹٪ و نقطه اوج شیوع بیماری ۸٪ (۰/۰۰۱ < p) بود. تغییرات ضریب همبستگی پیرسون بین ۰/۰۰۵ - ۰/۳۶ محاسبه گردید. انتشار ناقلین ویروس زبان آبی بستگی به تغییرات آب و هوایی و شرایط محیطی دارد. **نتیجه گیری نهایی:** نتیجه گرفته می شود که نمایش فنوتایپی ژن ویروس زبان آبی ممکن است تحت تاثیر آب و هوا قرار داشته باشد.

**واژه های کلیدی:** ویروس زبان آبی، آزمون الیزا، آنتی بادی، فاکتورهای وابسته، شیوع سرمی.

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