Short communication

First evidence of bluetongue virus in Kazakhstan

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Abstract

We report the results of the first serological survey for bluetongue virus in Kazakhstan. We analysed blood samples collected from 958 livestock and 513 wild saiga antelopes over a large area of the country, and found 23.2% seroprevalence in livestock and 0% in saigas. Seroprevalence in livestock did not vary by species, but increased significantly with age. There was no evidence for variation in seroprevalence at the regional level, but there was significant clustering at the farm level. Bluetongue has never before been reported in Kazakhstan, yet our results suggest that it may be endemic. We found seropositive animals at the furthest known northern limits of the bluetongue virus in this region of the world. Recorded vectors are not known to be present in Kazakhstan, so a novel vector is likely to be operating. The lack of evidence for bluetongue virus in saigas is unexpected and suggests a need for further investigation.
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1. Introduction

Bluetongue, an Office International des Epizooties list A viral disease transmitted by midges of the genus *Culicoides*, has never been reported in Kazakhstan. None of the proven vectors of the bluetongue virus (BTV) are known to exist in the country, but other potential vectors of the sub-genus *Avaritia* are present (*Culicoides dewulfi*, *C. montanus*, *C.*

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obsoletus, *C. scoticus*, *C. ukumensis*—G. Auezova, personal communication). The usual range for BTV is between latitudes 35°S and 40°N, however the virus can extend further north into large land-locked areas, e.g. to 48°N in Xinjiang, China (Qin et al., 1996) and to almost 50°N in western North America (Dulac et al., 1989).

In Soviet times, bluetongue virus was not thought to occur in Kazakhstan, and was not tested for. Since independence in 1991, the disease has been reported in Russia (Vishnyakov et al., 1994), but it has not been surveyed for in Kazakhstan. In this study, we carried out a serological survey for antibodies to BTV in domestic livestock and in saiga antelopes (*Saiga tatarica*). Kazakhstan's agricultural economy is based on pastoralism on its extensive semi-arid rangelands, hence livestock and saiga antelopes come into contact when grazing. Nomadic livestock herding with seasonal migrations continued throughout the Soviet period, but at the time of our study withdrawal of State support for agriculture had led to the cessation of the majority of seasonal livestock movements, and the depletion of saiga herds due to poaching (Robinson et al., in press).

2. Materials and methods

Blood samples from 279 cattle, 542 sheep, 137 goats and 513 saiga antelopes were collected across a broad swathe of Kazakhstan between latitudes 41.2°N and 49.3°N, in the period 1996–1998 (Fig. 1). Our study was focused on Central Kazakhstan, where 23 ex-collective farms, now villages, are located, of which we sampled 17. This area is within the range of a population of saiga antelopes. The ex-collective farms were large, many around 80,000 Ha, and at the time of our study contained several kinds of new farm, all of which we sampled; these were smallholders based in the main village centre, larger privately owned herds based some kilometres from the main village, and remnants of the collective flocks. For comparative purposes we also sampled herds from one village in west Kazakhstan, within the range of a second saiga population, and three villages in the south-east of Kazakhstan, outside the saiga range. In total, we sampled livestock belonging to 54 owners, based at 35 farms in 21 villages (Table 1). These were in 11 *raions* (districts) of 6 *oblasts* (provinces). Saigas were sampled at sites throughout Central Kazakhstan after they were shot by commercial hunters, and in west Kazakhstan under scientific licence. Sera were assayed using a monoclonal antibody based competitive ELISA (Anderson, 1984), that has been validated both for domestic and wild ungulates (Afshar et al., 1995).

Because the data were spatially hierarchical, we used a multiple-variable hierarchical model to test for clustering at different spatial levels, while assessing hypothesised fixed-effect differences in seroprevalence. We considered the levels of the province, district and farm as random effects, because the data were not sufficient to distinguish between the village, farm and owner levels. The fixed effects that were considered were age, $(age)^2$ (to account for non-linearities in the age-seroprevalence relationship), species origin (bought in or born on the farm) and gender. The data were aggregated for all three livestock species (cattle, sheep and goats) because neither the age profile nor seroprevalence were significantly different between species (age: $\chi^2 = 3.98$, d.f. = 6, NS; species: $\chi^2 = 0.03$, d.f. = 2, NS). The model was developed in MLwiN (Multilevel Models Project, Institute of Education, London; Lundervold, 2001).
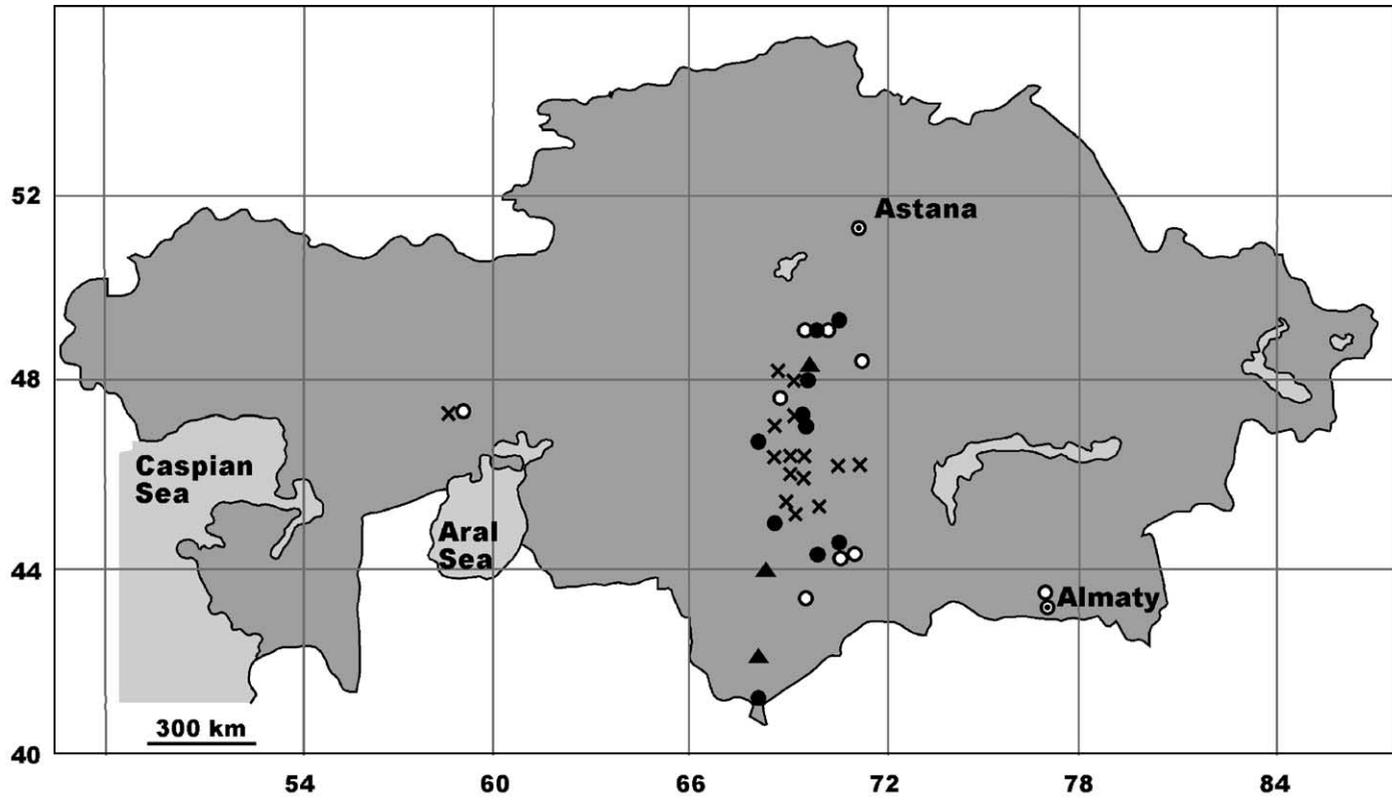
Fig. 1. A map showing the locations where samples were taken. The filled triangles indicate locations where more than 50% of the animals sampled were sero-positive, the filled circles indicate locations where 20–49% of the animals sampled were sero-positive, and the open circles locations where <20% of the animals sampled were sero-positive. Crosses indicate the locations where saigas were sampled.

Table 1
Summary of number of animals sampled in each village, divided into cattle and sheep and goats

Village	Cattle		Sheep and goats	
	Seropositive (%)[a]	Sample size	Seropositive (%)	Sample size
Luchivostock	8.3	12	0	8
Akkum	22.2	9	100	9
Shardarinski	0	10	–	0
Chu	16.7	12	42.9	35
Suzak	20	5	73.3	15
Zhuantobe	–	0	35	20
Ulan Bel'	–	0	0	10
Chiganak	37.5	8	16.7	12
Kalinina	–	0	8.3	12
Kamkaly	50	10	30	10
Sarysu	0	3	16.7	12
Zhailma	33.3	15	23.5	34
Moybulak	16.7	6	35.7	70
Sarysuyskiy	17	53	14.3	133
Druzhba	9.8	61	11	91
Zhenis	41.9	31	28.8	80
Amantau	100	10	10	20
Arshelinski	0	5	21.7	23
Taldesay	40	10	5	20
Tkenekta	50	10	6.7	15
Zhanakonys	11.1	9	14	50
Overall	25.4	279	22.2	679

[a] The % seroprevalence to BTV is also given.

3. Results

Specific BTV antibodies were detected in 25.4% of cattle, 21.4% of sheep, 25.5% of goats and 0% of saigas (Table 1). Positive results were obtained throughout the study area (Fig. 1), but prevalence varied from 0 to 100% between livestock owners. The most parsimonious model showed a significant non-linear increase in seroprevalence with age (Table 2, Fig. 2). None of the other fixed effects were significant. We found significant clustering at the farm level, which may be related to local variations in the distribution of the vector. Once clustering by farm was accounted for, no significant clustering at the district or province level was detected, which might have been expected if vector abundance varied regionally. The spatial pattern reported here was similar to that found by Ward and Carpenter (2000) for blowfly strike in Australia.

4. Discussion

The distribution of seropositive animals, the relative homogeneity of seroprevalence observed between the three domesticated species and the age profile suggest that BTV has

Table 2
Parameter estimates for the most parsimonious mixed logistic-normal hierarchical model for seroprevalence of bluetongue

Parameter[a]	Estimate	S.E.
Random effects		
Farm-level variance[b]	0.779	0.255
Animal-level extra-binomial variance[c]	0.953	0.044
Fixed effects		
Intercept	−2.078	0.256
Age	0.334	0.080
$(Age)^2$	−0.015	0.006

[a] Parameters are at two levels; random effects at the farm level and fixed effects for age.
[b] Individual farms may contain several herds.
[c] Animal-level extra-binomial variance measures the degree to which assumptions of binomial variance are violated. A value of 1 implies variance is binomial; the estimate obtained suggests no significant departure from this assumption.

been endemic for some time. However, because of the paucity of information it is not possible to estimate when BTV entered the country. Most animals sampled were born in their villages, so were likely to have been infected in situ. Before independence in 1991, seasonal movement of livestock from farms in the south of the study area to grazing areas further north was common. However during the study the majority of the farms visited no longer carried out seasonal migrations, and there is no evidence of any relationship
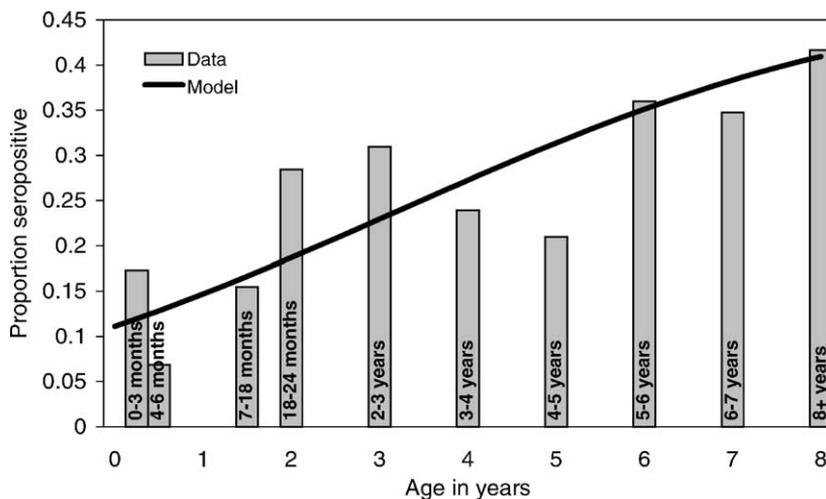

Fig. 2. Age-related seroprevalence to BTV among domestic livestock in Kazakhstan, showing a classical decline in percentage of sero-positives as maternal antibodies wane, followed by an increase with age following BTV exposure and infection. The raw data, expressed as a histogram, and the hierarchical mixed logistic-normal model fit (━) are superimposed. Because there were no significant differences in the BTV seroprevalence recorded for cattle, sheep and goats at each age group, the results obtained for the three species were combined.

between seroprevalence and movement patterns either spatially or in terms of age distribution.

The negative reactions recorded for the 513 saigas were interesting, given the relatively high prevalence of antibody recorded for domestic livestock from the same areas. BTV antibodies have been reported previously in many wild ungulates, including antelopes (Verwoerd and Erasmus, 1994). The sample size was sufficiently large to give a 95% chance of detecting at least one positive saiga with a true prevalence as low as 3.6%. The apparent absence of BTV antibody in saigas may reflect differences between saigas and livestock in their habits or habitats and their interactions with vectors. Either the vector is not biting saigas or they only rarely come into contact with infected vectors. Vector distribution, abundance, infection rates, efficiency and host preferences may all be important. Although saigas and livestock use the same water sources and grazing areas while vectors are active, they may not interact where vector populations are high. Even when BTV is endemic, infection rates are very low (Mohammed and Mellor, 1990; Al-Busaidy and Mellor, 1991), and in Africa, cattle are used to protect sheep from BTV infection because vectors preferentially feed on cattle (Nevill, 1979). Further research is needed into the ecology of saigas and vector midges in order to clarify the reason for our results.

This study shows the presence of BTV in areas as far north as $49.30°N$. The absence of clinical disease is not unexpected because bluetongue is rare in cattle and goats, and in sheep clinical signs can vary considerably both between individuals and particularly between breeds. Nevertheless, the presence of antibody against BTV in Kazakhstan is epidemiologically important because it demonstrates the presence of BTV at the furthest known northern limits of the virus. The absence of a proven BTV vector in Kazakhstan strongly suggests that a novel vector is operating in this region (Mellor and Whittmann, 2002). Further research into the prevalence and transmission of BTV in Kazakhstan is important for developing a comprehensive animal health policy for this region.

Acknowledgements

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