Tick-Borne Encephalitis Virus, *Coxiella burnetii* & *Brucella* spp. in Milk, Kazakhstan

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**Introduction**

*Brucella* spp., *Coxiella burnetii*, and tick-borne encephalitis virus (TBEV) are believed to be enzootic in the Republic of Kazakhstan, and pose a particular public health risk due to their transmissibility in unpasteurized milk and dairy products. We established a milk surveillance methodology employing immuno and molecular assays to identify these agents, and applied this methodology to milk samples collected in western Kazakhstan in winter 2014-2015.

**Methods**

Raw milk was collected from cows in the area around Uralsk in western Kazakhstan over the winter of 2014-2015. Samples were defatted and frozen at -20°C, then tested as follows for the presence of infectious agents. For TBEV, 65 samples were tested using the VectorBest TBEV antigen capture kit. For *C. burnetii*, 50 samples were assayed using a species-specific qPCR assay. For *Brucella* spp., PCR, ELISA and FPA testing was initiated and is ongoing.

**Results**

For TBEV, of 65 samples tested, nine percent were found to be positive, indicating that these milk samples contained the virus at the time of collection and thus that the source animals were infected. Source animals of TBEV negative samples may or may not have been infected, suggesting a need for blood sample collection for antibody assessment in conjunction with milk sample collection in future studies. This additional activity may also provide valuable information regarding how long infected animals shed the virus. For *C. burnetii*, all 50 samples tested were found to be negative, though positive controls were consistently positive. Because *C. burnetii* exhibits seasonal increases in bacterial load during parturition, these results may be related to the time of sample collection during winter months and may not be representative of year-round presence of *C. burnetii* in milk, such that additional samples from other seasons will be tested in future studies. For *Brucella* spp., ongoing testing has yielded some positive results by PCR, ELISA and FPA.

**Conclusions**

Our data suggest that consumption of raw cow’s milk in western Kazakhstan is a risk factor for tick-borne encephalitis and brucellosis. The risk for Q fever seems to be small during winter, but may be present at other times of the year. Milk samples will be collected year-round in future work, and may be accompanied by collection of blood samples for comparative analysis.

**Keywords**

Tick-borne encephalitis; Q fever; Brucellosis; surveillance; milk