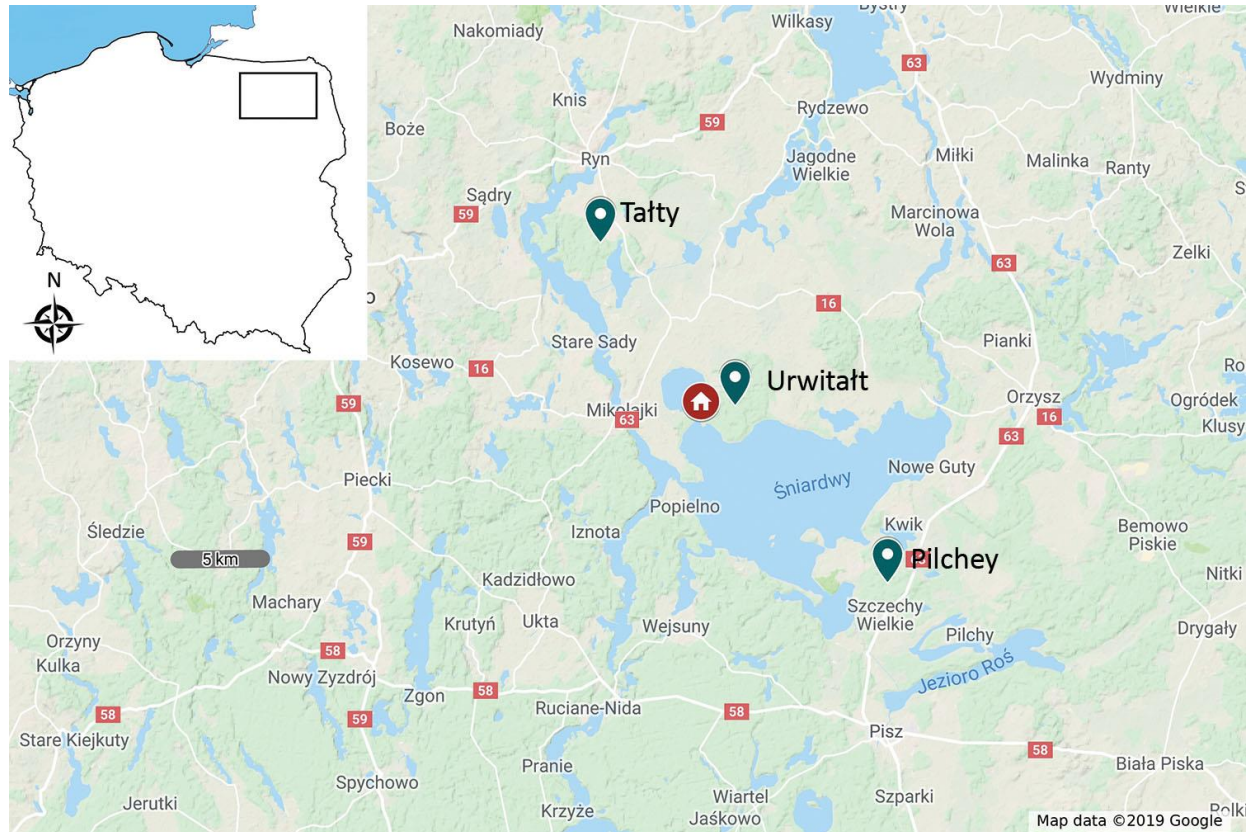
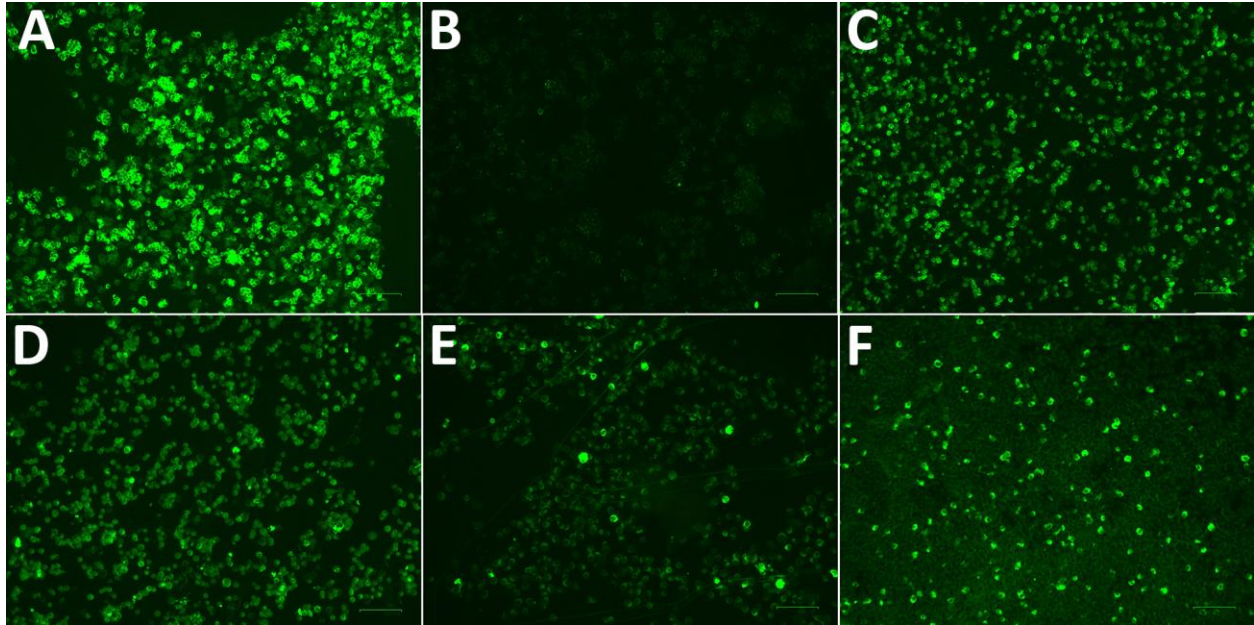


# Zoonotic Virus Seroprevalence among Bank Voles, Poland, 2002–2010

## Appendix



**Appendix Figure 1.** Locations of study sites in the Mazury Lake District in northeastern Poland. Red pointers, study sites. Blue pointer; Warsaw University Field Station. Source: Google Maps.



**Appendix Figure 2.** Immunofluorescence assay (IFA) for: PUUV (A) positive control, (B) positive sample; LCMV (C) positive control, (D) positive sample; CPXV (E) positive control, (F) positive sample. The serum samples were diluted 1:10 in PBS, and the reactivity of the samples to hantaviruses were tested with PUUV-IFA, to cowpox viruses with CPXV-IFA and arenaviruses with LCMV-IFA. Briefly, PUUV (Sotkamo strain), CPXV, and LCMV (Armstrong strain)-infected Vero E6 cells were detached with trypsin, mixed with uninfected Vero E6 cells (in a ratio of 1: 3), washed with PBS, spotted on IFA slides, air-dried, and fixed with acetone as described earlier (1). The slides were stored at  $-70^{\circ}\text{C}$  until use. IFAs were carried out with seropositive human serum as a positive control for the PUUV- and CPXV-IFA; and LCMV mouse monoclonal antibody (Progen, Heidelberg) for the LCMV-IFA. The slides were read under a fluorescence microscope, and pictures were taken with a ZOE fluorescent cell imager (BioRad).

## Reference

1. Hedman K, Vaheri A, Brummer-Korvenkontio M. Rapid diagnosis of hantavirus disease with an IgG-avidity assay. *Lancet*. 1991;338:1353–6.