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# Unraveling the Molecular Evolution of Usutu Virus in Germany using Nanopore Sequencing

### Inhalt

In Germany, the mosquito-borne flavivirus Usutu virus (USUV) is the causative agent of annually reoccurring epizootics in the avifauna, primarily of blackbirds and owls. USUV was first documented in 2010 in the southwest of Germany in *Culex* mosquitoes and in 2011 in blackbirds. Ongoing surveillance efforts identified a spread of the virus to the north and east of the country, upon where it reached all Federal States in 2018. The same year in which the largest recorded epizootic occurred in the country (1,208 USUV-positive birds). Despite its relevance for not only veterinary but also public health, little research in the past has focused on gathering USUV whole genome sequences and analyzing the virus's molecular evolution. Therefore, in the frame of this study, a Nanopore sequencing tool was implemented for USUV and compared to that of Illumina sequencing as well as to a protocol with and without prior target-enrichment. The subsequent cost- and time-effective amplicon-based Nanopore platform was then used to generate 118 USUV whole genome sequences from wild and captive birds from Germany. Phylogenetic and phylodynamic analyses revealed the prevalence of lineages Europe 3 and Africa 3 in Germany and that their most-recent common ancestors confirm a 3-year time-lag between their possible introduction and their actual identification. Furthermore, the study provides indices for the genetic evolution of USUV within the country as well as frequent new introductions thereof.

## Keywords

Germany, Nanopore, Surveillance, Usutu virus, Whole genome sequencing

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### **Junior Scientist Status**

Yes, I am a Junior Scientist.

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