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**Mosquito ID: sequencing out of the mobile suitcase lab as an early**

**warning system for emerging infectious diseases**

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**Abstract:**

**Background/Objective:** Mosquito-borne diseases are responsible for spillover to around 700 million people each year, including many neglected tropical diseases such as Dengue fever and Chikungunya virus disease. Case reports are rising over the last decades, as mosquito species flare-up as a result of climate change and globalization. Surveillance tools and early warning systems are essential to prevent disease spread to humans and animals. Next generation sequencing technologies offer great advantages for disease outbreak investigation. The aim of this study was to develop a field deployable sequencing platform to identify potential mosquito pathogens, species and host from blood meals.

**Methods:** A rapid extraction reverse purification method was developed using mosquito specimens collected in the field and/or from laboratory colonies in Greece (Athens) and Spain (Barcelona), including *Culiseta longiareolata*, *Culex pipiens*, *Aedes albopictus*, *-cretinus* and *-aegypti.* Nucleic acids from the specimens were isolated using a rapid “all-in-one” extraction protocol based on lysis buffer, glass beads, magnetic beads, heating and vortexing. Oxford Nanopore Technologies rapid barcoding sequencing was operated using a MinION MK1C device. A reverse transcription step was performed for RNA targets. All steps were carried out in the fully equipped suitcase lab. A specific offline BLAST database was created to semiautomatically identify mosquito species, host in blood meal and pathogens.

**Results:** The species was correctly identified in all samples. Both animal and human DNA could be detected in the mosquito blood meal. Mosquito-associated viruses were detected.

**Conclusion:** The protocol performed in the suitcase lab allows fast mosquito “footprint” analysis directly in the field, allowing an early warning for mosquito-borne diseases and on-site outbreak investigation. The mobile system is deployed in field in Bangladesh to support the local surveillance program.

**Keywords:** early warning system, mosquito-borne diseases, rapid diagnostics