DNA-barcoding: an efficient tool for rapid identification of native and exotic mosquito species intercepted in Belgium

Due to international trade, tourism, and climate/ecological changes, mosquito species are transported, dispersed, introduced and may eventually become established in new territories. The introduction of potential disease vector species constitutes a threat to human and animal health. Since July 2017, a nationwide three year monitoring project funded by the Belgian federal authorities and the federated entities, is ongoing in Belgium (MEMO: Monitoring of Exotic Mosquitoes), and is coordinated by the Institute of Tropical Medicine (ITM). DNA-based technologies are used to validate the morphological identifications of intercepted exotic mosquito species (EMS), as well as of a 5% subset of the yearly sampling (quality control). Additionally, a DNA sequence reference database is being compiled.

Aim MEMO project: Detecting and evaluating the occurrence of exotic mosquito species in Belgium + risk analyses.

**INTRODUCTION**

**MATERIAL, METHODS AND RESULTS**

**Sampling: 23 PoEs (Point-of-Entry)**
- Larval sampling
- Oviposition traps
- Adult traps

**Voucher collection**
- Adults
- Larvae

**Species identification Morphology-based**

**Sp identification validation DNA-based**
- PCR and sequencing of COI, ND4, ITS2
- Size fragment analyses of ACE2, CQ11

**Sp validation of intercepted exotic mosquitoes**
Four EMS collected once or multiple times at one or multiple PoEs

Map of Belgium indicating the PoEs where EMS were detected in 2017 and 2018 (preliminary data; ITM)

**CONCLUSION**

**Annual validation**: The DNA-based identifications up to species complex level were in agreement with the morphology, validating the morphology-based species identifications. Yet, DNA-based methods allowed to discriminate between species of the same complex.

**EMS validation**: EMS collected at the nine distinct PoEs were validated using DNA-based techniques. EMS eggs were most often morphologically mis-identified with *Aedes geniculatus* eggs (native). EMS seem to enter effectively via different introduction pathways: through lucky bamboo, tyre transport and ground traffic, but possibly also by natural dispersal. Early interceptions and rapid DNA-based verifications should help the authorities in their efforts to contain the spread and eradicate EMS populations.