Introduction

In recent decades, we have witnessed the emerging and re-emerging of mosquitoes and mosquito-related diseases. Providing the tools for better management of mosquito-related issues requires much better in-depth knowledge of the ecological requirements of the main disease vectoring species.

Environmental DNA (eDNA) is a potentially powerful new method that can be used to attain that, but requires rigorous testing across a broad range of ecological conditions. Here we used paired trapping of adults and aquatic larvae in Kruger and fringing rural areas to improve understanding of mosquito ecology.

Experimental setup

To investigate changes in mosquito communities across Kruger National Park (KNP) and fringing rural communities we used different traps to catch adult specimens (see below), and in parallel, we used an aquatic eDNA-survey method. At each site we collected: (i) adult mosquitoes, (ii) mosquito predators, (iii) aquatic eDNA samples, (iv) environmental variables (temp., pH, etc.).

*DNA collection and DNA extraction*

At each site 30*25 mL eDNA aquatic subsamples were collected, and concentrated over a 0.2 micron PES-filter and stored in Longmire buffer until DNA extraction.

*Primer design and PCR*

Family specific eDNA primers for mosquitoes and dragonflies were designed with PrimerMiner, Genious R10 and Primer3. Using a degenerate oligonucleotide-primed PCR approach. Additionally we used the primers BF1+BR1 as general macroinvertebrate primers.

*NGS sequencing and data analysis*

Amplicons were generated using a hot-start PCR. These amplicons will be sequenced with the Ion Torrent NGS. After sequencing a OTU/species list per site will be created using the Galaxy-pipeline (Naturalis), and compared with the site specific species list of the adult mosquitoes.

### Hypotheses

**H1** Mosquito communities differ between natural areas and fringing rural communities (fig.1)

**H2** These trends are similar for adult communities (using traps) and larval (using eDNA)?

### Preliminary results

Our preliminary results shows a major shift in mosquito community composition and abundance between inside and outside KNP (fig.2). We expect that our eDNA samples will reflect these dynamics for the aquatic stage.

![Figure 1. Community composition and species abundance, under natural conditions (Inside parks) and with anthropogenic stressors (OUTside parks). The community composition and species abundance can change due presence or absence of anthropogenic stressors.](image)

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