

SHORT TERM SCIENTIFIC MISSION (STSM) SCIENTIFIC REPORT

This report is submitted for approval by the STSM applicant to the STSM coordinator

Action number: CA17108 - Aedes Invasive Mosquitoes STSM title: Training on molecular methods for the identification of mosquito species, origin of blood meals and genotyping of insecticide resistance alleles STSM start and end date: 15/05/2022 to 28/05/2022 Grantee name: Marina Bisia

PURPOSE OF THE STSM:

In the current Short Term Scientific Mission (STSM) the first objective was to gain experience with the molecular methods used for the molecular identification of the mosquito species *Aedes albopictus*as an extra tool for the identification of mosquitoes species, at the University "La Sapienza", Rome, Department of Public Health and Infectious Diseases.

The second objective was to be trained on the molecular protocols for a. the identification of the origin of the blood meal and b. the genotyping of insecticide resistance alleles in DNA samples extracted from specimens collected in Greece.

Finally, this STSM provided me the opportunity to acquire a set of practical skills acquired for all laboratory procedures. Not only this STSM allowed me to improve my practical work but also offered me an ideal opportunity to advance my knowledge on molecular methods, which is important for our capacity building. I consider gaining more experience in this field will help my future research regarding my PhD.

In conclusion, I firmly believe that this Short Term Scientific Mission at the University "La Sapienza", Rome, Department of Public Health and Infectious Diseases, under the guidance of Professor Alessandra della Torre, helped me to the future goal of the Benaki Phytopathological Institute, where I currently work on, to improve molecular protocols for processing invasive mosquito species.

DESCRIPTION OF WORK CARRIED OUT DURING THE STSMS

During the first day (16/5/2022) the main focus was to be informed on the molecular methods and protocols used in the laboratory of the Department of Public Health and Infectious Diseases, on which I was trained on the following days. In addition, I was introduced to the facilities and the equipment of the laboratory that I used for the molecular procedures.

Furthermore, during the next three days (17/5 - 19/5/2022) I performed several PCRs for the identification of the species *Ae. albopictus*, on the DNA samples from the speciments that were collected in Greece during the year 2021. The procedures were performed following the specific protocols used by the staff of the laboratory and included:

- 1. Cleaning of the bench and the equipment that were used during the experimental procedures.
- 2. Centrifuge of the DNA samples (spin down)
- 3. DNA amplification by PCR following the protocol adapted in the laboratory for the preparation of the Master Mix and the cycling conditions adapted in the laboratory
- 4. Preparation of the agarose gel (concentration 3%)
- 5. Gel electrophoresis (loading the PCR products and appropriate ladder to the agarose gel)

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- 6. Gel documentation under UV illuminator
- 7. Analysis of the results







Figure 2. Preparing the PCR product Figure 3. Loading the agarose gel

Figure 1. Adding the master mix to the plates

During the three days (20/5/2022 and 23/5-24/05/2022) I focused on the blood meal analysis of the same samples collected from Greece, in order to be able to combine the results (species identification and origin of the blood meal). The two days that followed (25/5-26/5/2022), I was trained on the PCR procedure regarding the genotyping of insecticide resistance alleles. The last day (the 27th of May) we discussed the results we obtained.

in order to load it to the agarose gel

DESCRIPTION OF THE MAIN RESULTS OBTAINED

During my stay at the La Sapienza University of Rome, Department of Public Health and Infectious Diseases, I strongly believe that I have accomplished the main goal of my Short Term Scientific Mission, which was to get familiar with PCR protocols regarding the identification, blood meal analysis and genotyping incecticide resistance alleles for the *Aedes spp* mosquitoes and particularly for *Aedes albopictus*. The samples collected in Greece where identified as *Ae. albopictus* and further analysis of the results obtained is needed.

FUTURE COLLABORATIONS (if applicable)

We continue to collaborate by exchanging ideas about the combined results from the different PCRs we conducted. Hopefully, we will establish collaboration for further research on these topics between our Institute and the research team involved in this STSM from the Department of Public Health and Infectious Diseases of La Sapienza University, Rome.