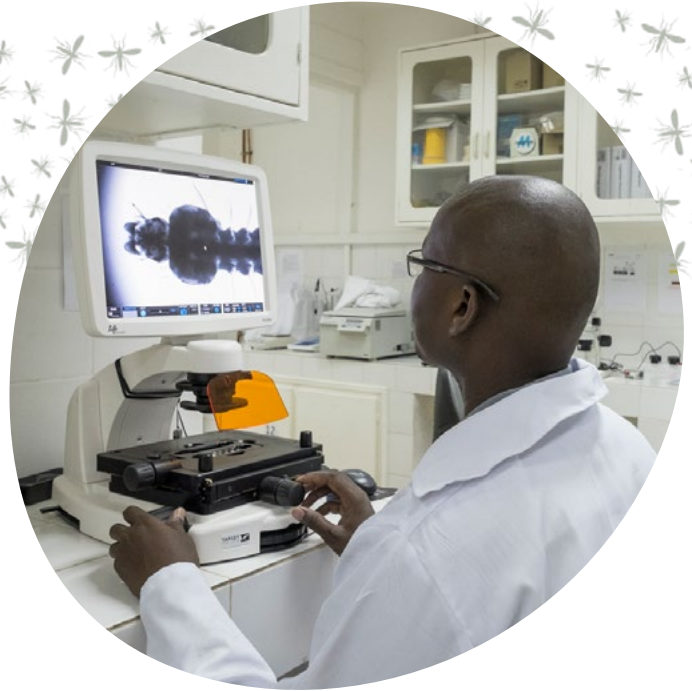


# Results of laboratory studies on non gene drive genetically modified sterile male mosquitoes in Mali



## Summary

In 2021, Target Malaria Mali concluded its studies on the non gene drive genetically modified sterile male mosquitoes that were housed in the Arthropod Containment Level 2 (ACL-2)<sup>1</sup> insectary based at the Malaria Research and Training Center (MRTC) at the University of Sciences, Techniques and Technologies of Bamako (USTTB). In this factsheet, we provide a summary of the results of the studies conducted by the team over the last two years (2019 to 2021).

In 2022, the MRTC ended its participation in the Target Malaria consortium to launch a new project to train African scientists on molecular engineering.

## 1. Importation

In June 2019, Target Malaria Mali received approval from the Malian Ministry of Environment, Sanitation and Sustainable Development (MEADD) to import a strain of non gene drive genetically modified sterile male mosquitoes to be housed and studied in the ACL-2 insectary based at MRTC-USTTB in Bamako.

In September 2019, the package containing the eggs of this strain, sent from our collaborating partner at Polo d’Innovazione di Genomica, Genetica e Biologia (PoloGGB) in Italy, was received at Président Modibo Keita airport by

the Malian regulators. The package containing the eggs was then transferred to the Target Malaria team, who took it to the ACL-2 insectary where the studies were conducted.

## 2. Overview of sterile males

This phase of our research programme involved conducting experiments in an ACL-2 insectary on a strain of non gene drive sterile male mosquitoes. The sterile male mosquitoes have been modified so that only the males are sterile. To create these sterile males, we used a nuclease gene that, when activated during sperm production, fragments the X chromosome of the sperm and of the egg upon fertilisation. This modification means that the eggs do not hatch, and therefore no offspring are produced. The modified females of this strain are instead fertile allowing breeding to happen in the lab under specific management conditions.

The sterile strain was generated in the laboratory of Imperial College London in the United Kingdom in 2008. After molecular characterisation, the mosquitoes were further evaluated in large indoor cages under a defined set of environmental parameters in an ACL-2 insectary at PoloGGB in Italy.

1 ACL guidelines are a widely recognised reference for research laboratories to assess risk and establish protocols for the safe handling of arthropods, such as *Anopheles* mosquitoes. For more information, please see our factsheet on “Safety measures for work on genetically modified mosquitoes in containment”

### 3. Study objectives

Our team studied the characteristics and behaviours (survival rates, development, fecundity, feeding, etc.) of the non gene drive sterile male *Anopheles* mosquitoes, in an ACL-2 insectarium.

### 4. Key findings of the studies on sterile male mosquitoes in a contained environment

The Target Malaria Mali insectary team developed considerable expertise, thanks to the successful importation and breeding of genetically modified mosquitoes in a contained environment. It should be noted that this import was a first for Mali. The work performed was significant in terms of transfer of knowledge and development of skills to support implementation of the next phases of the project.

- The gene for the modification became part of the genetic makeup of the locally derived colony housed in the ACL-2 insectary after 6 generations of breeding between modified females and local males, as expected according to the Mendelian model, with a 50/50 chance of inheritance.
- Our scientists then maintained the sterile male mosquito colony by continually selecting and breeding modified females and local males in containment.
- The stability of the modification was confirmed by the lack of viable eggs from cross-breeding between modified males and local females.
- The reliability of sorting in the larval and pupal stage was highly successful with greater than 99% sorting accuracy.
- Detailed studies of life history parameters were conducted and compared between genetically modified and local mosquitoes. These included longevity, mating competitiveness, female blood feeding avidity and egg production. Results were as expected based on previous studies and confirmed the introgression of the transgene in the local genetic background did not have any unexpected effects, thus confirming the expected results from risk assessments and the system safety.

- Out of eight insecticides tested, no resistance has been found, neither in the genetically modified mosquitoes nor in the local strain.

All planned experiments were completed successfully, enabling the team to gain a more in-depth understanding of the behaviour of the sterile male mosquitoes.

### 5. Conclusion

The Target Malaria Mali team's work on the non gene drive sterile male mosquitoes is now completed and Target Malaria is no longer present in Mali. The team is proud to have been the first allowed to import genetically modified mosquitoes into Mali. The team has grown greatly during this period, advancing their skills in how to maintain genetically modified mosquitoes, as well as working under a regulatory permit and maintaining compliance with the conditions issued with it, including submission of a final report to the National Biosecurity Committee on the work performed. The MRTC team is looking forward to using the new skills and knowledge in molecular engineering to promote the training of African scientists.

