**Draft guidance document on Risk Assessment and Risk Management
of living modified Mosquitoes**

***Prepared by the Ad Hoc Technical Expert Group on
Risk Assessment and Risk Management***

*Version of 7 March 2010*

**OBJECTIVE**

The Ad Hoc Technical Expert Group (AHTEG) on Risk Assessment and Risk Management has developed a Roadmap for Risk Assessment which sets out the necessary steps to conduct a risk assessment in accordance with Annex III to the Cartagena Protocol on Biosafety[[1]](#footnote-1).

The present document[[2]](#footnote-2) aims at complementing the Roadmap on specific issues that may need special consideration for environmental releases of Living Modified Mosquitoes (LMMs). It focuses mainly on Paragraphs 8 (a) and (e) of Annex III.

The present Guidance Document also provides additional information that may contribute to better understanding of the issue, and help regulators to conduct risk assessment and risk management in their particular case of environmental release of LMMs.

Each topic in this document contains reference to the appropriate step in Annex III. Suggestions for supporting bibliographies are also provided through links to web pages in the Biosafety Clearing House.

This is intended to be a “living document” that will be shaped and improved with time as new experience becomes available and new developments in the field of applications of LMOs occur, as and when mandated by the Parties to the Protocol. A similar sentence should be added in the other guidance documents.

**Introduction**

LMMs are being developed to control the population of vectors in order to reduce transmission of vector-borne human diseases, especially malaria, dengue and chikungunya. Control, including eradication, of such diseases is a widely-recognized public health goal.

Various genetic control strategies are being developed to control the population of mosquito vectors by either suppressing their population or reducing their vector competence. These strategies can also be categorized according to the technology involved and the method of implementation. Some are intended to be sterile or self-limiting (i.e. unable to pass the modification on indefinitely through subsequent generations), and thus depend on continued releases of male mosquitoes. Others are self-sustaining or self-propagating (i.e. heritable modifications intended to spread through the target population) and would depend on small and infrequent releases. Thus, the strategy under consideration is an important factor in the risk assessment process.

The biology and ecology of mosquitoes, and their importance to public health as vectors of human disease and morbidity, pose new considerations and challenges to the risk assessment and risk management of LMOs, which have traditionally dealt with LM crop plants mainly. As regulations and experience pertaining to LMMs vary across countries, this guidance document is intended to help Disease Endemic Countries (DECs) interested in evaluating and possibly deploying LMMs to combat mosquito-borne diseases. While various approaches to combat vector borne diseases using LMMs may have many issues in common, it is recognized that there may be different sets of challenges to address the specific strategies

**Scope**

This document focuses on the risk assessment and risk management of LMMs developed for use in vector control of human diseases such as malaria, dengue, chikungunya and yellow fever. There are related technologies, for example, mosquitoes infected by a naturally occurring mutant strain of *Wolbachia* (such as *wMelPop*) to reduce their vector competence, which are not covered by this document because such modifications are not considered paratransgenic. However, DECs interested in release of such mutant strains may still refer to this document on common features between these mutant strains and LMMs such as species-specificity and self-propagation, and their risk implications in the event of open releases into the environment. There are also other dimensions that should be taken into consideration in the decision for environmental releases of LMMs which are not governed by Annex III of the Protocol. They encompass among others: economic, health and social trade-offs associated with the technology application as well as ethical, social and cultural issues that are expected to influence the acceptance of these methods.

**POTENTIAL ADVERSE EFFECTS**

*(see Step 1 of the Roadmap for Risk Assessment)*

A consideration that we frequently encounter (but more in estimating potential risks of, for instance, LM *Plasmodium*, is potential extension of the habitat of a vector due to climate change. Is that relevant here too, e.g. in point to consider (c) in line 101, or in line 156 (ecosystem effects)?

A specific and reasonably comprehensive list should be provided for potential adverse effects of a particular LM mosquito strain, taking into account the species of the mosquito, the LM trait, molecular mechanisms of genetic modification, the intended receiving environment, as well as objective and scale of the intended release. For instance, this list could contain: (a) the kinds of possible adverse effects where there is solid scientific evidence; (b) the protection goals of the country where the LMMs will be introduced; (c) the species and ecological processes that could be affected by the introduction of the LMMs; (d) a conceptual link between the identified environmental protection goals and the introduction of LMMs into the environment; (e) an evaluation of the likelihood and consequences of the identified possible adverse effects, risk management measures that can be put in place to address these effects, and balancing the risks with potential benefits of releasing the LMMs.

**Effects on biological diversity (species, habitats and ecosystem services)**

*Rationale:*

The release of LMMs may have a negative impact on the target and other species, such as:

*New or more vigorous pests, especially those that have adverse effects on human health:* (i) The released LMMs may not function as expected. Gene silencing or production failures could result in the release of non-sterile or competent mosquitoes and thus increase the vector population or disease transmission. (ii) The released LMMs could transmit another disease more efficiently. Such diseases might include yellow fever, chikungunya, etc. (iii) Suppression of the target mosquito might lead to niche replacement, i.e. enable another vector species to fill the empty niche, which may in turn result in higher levels of the target disease or a new disease in humans. Rigorous monitoring prior to, during and after the trials can help determine if replacement with a different vector species could be realised. (iv) The released LMMs might become nuisance pests. (v) The released LMMs might cause other pest problems to become more serious, including agricultural pests and other pests that affect other valued human activities.

*Harm to or loss of other species*. The released LMMs might cause other valued non-pest species (for instance fishes the mechanism how this could come about is not immediately clear) to become less abundant. These include species of economic, cultural, and/or social importance such as wild foods, iconic species and endangered species. Ecological effects might result from competitive release if the target mosquito is reduced, or from trophic consequences of species that rely on mosquitoes for food during some specific time of the year. Effects might also occur if (i) the target mosquito was also transmitting a disease to another animal species, (ii) the released LMMs transmit a disease of another animal species more efficiently, or (iii) a vector of an animal disease was released from ecological control by the reduction of the target mosquito. Sterile inter-specific matings between the released LMMs and other mosquito species could disrupt the population dynamics of these other species leading to harm or loss of valued ecological species. The released LMMs might also degrade valued ecosystem processes such as pollination.

*Disruption of ecological communities and ecosystem processes*. The ecological communities in the ephemeral, small aquatic habitats occupied by the vector mosquitoes targeted with LMMs are unlikely to be greatly disrupted beyond the possibilities already addressed above under “harm to or loss of other species.”

*Points to consider:*

1. What is the impact of the strategy under consideration on the target mosquitoes?
2. May the LMMs have an adverse effect on other species becoming agricultural, aquacultural, public health, or environmental pests, or produce nuisances or health hazards?
3. What is the habitat range of the target species (see above, line 50)?
4. Is the target species native / invasive in a given area?
5. Will the release affect mosquito species that are pollinators or otherwise are known to participate in valued ecosystem processes?
6. What species do the target mosquitoes typically interact with in the environment?
7. May the LMMs have an adverse effect on other interacting organisms?

**Gene Flow**

*Rationale:*

Gene flow in regard to biosafety refers to the transfer of transgenes or modified genetic elements from the LMO to non-modified organisms. It can occur via cross-hybridization or independent movement of the transgenes or genetic elements. Whether gene flow occurs and what adverse effects it might have depend on various factors such as the LM technology used, the trait or traits carried by the mosquitoes, the receiving environment, etc.

Based on the existing knowledge on the ecology and biology of mosquito species that transmit malaria, dengue and chikungunya, it may not be likely that other host species will be affected by LMMs. Depending on the nature and scale of the LM technology to be deployed, additional studies may be required on gene flow among vectors, their mating behaviour, interactions between vectors sharing the same habitat, how parasites and pathogens respond to the introduction of LMMs, etc. It is important that such studies are designed with specific, measurable, environmentally-significant end-points in order to arrive at a timely decision, especially if the objective of deployment is reduction of disease burden.

*Gene flow through cross-hybridization:* Some LMMs are being designed to spread a trait rapidly through the target mosquito population. For instance, for *Anopheles gambiae*, the trait may be expected to spread throughout the *A. gambiae* species complex. Other LM mosquito technologies are designed to be self-limiting and, thus, spread of the transgenes or genetic elements in the target mosquito population is not expected. For such self-limiting technologies, the potential for an unexpected spread of the transgenic trait should be considered by focusing on the ways that any management strategy to limit the spread could fail. Gene flow between different species should be considered for all of the LM mosquito technologies. Mosquitoes, like other insects, typically have strong reproductive isolating mechanisms that do not allow inter-specific gene flow. In addition, the fitness conferred by the transgenic trait and the size and frequency of the introduction of the LMMs into the environment will also determine the likelihood and rate of spread of the transgenes or genetic elements.

*Independent movement of the transgenes or genetic elements:* This is commonly referred to as “horizontal gene flow”, which is the movement of genetic information from one organism to another through means other than sexual transmission. Gene drive systems for moving genes into wild populations should be one of the initial foci of the risk assessment. The risk of horizontal gene flow in LM mosquitoes that do not contain a gene drive system is likely to be smaller but should nevertheless be assessed on a case-by-case basis.

*Points to consider:*

1. Does the release of the LM mosquitoes have the potential to pass their modified traits to non-related organisms? If so, what may be the undesirable consequences?
2. Will the LMMs induce undesirable functions or behaviors within target species, other wild related species or non-related organisms?
3. What mechanisms, if any, are available to recall a trait which has spread unexpectedly?

**Evolutionary responses (especially in vector or pathogen)**

*Rationale:*

Any strong ecological effect also exerts an evolutionary selection pressure. The main evolutionary effects are those that could result in a breakdown in the technology and the resumption of previous disease levels. Other evolutionary effects could be hypothesized but they would first require the occurrence of some adverse effect on a species, community or ecosystem effect (see line 50 above). Therefore, consideration of secondary evolutionary effects can be postponed until such effects are identified and found to be significant.

*Points to consider:*

1. Does the mosquito vector have the potential to evolve to avoid population suppression, regain vector competency or acquire new or enhanced competency of another disease agent? If so, what may be the undesirable consequences?
2. Does the trait have the potential to evolve to lose effectiveness or the pathogen to overcome the limitation posed by the genetic modification? If so, what may be the undesirable consequences?

**Persistence of the transgene in the environment**

*Rationale:*

Inserted transgene(s) may spread and persist in natural populations. Some of the transgenes in LMMs are designed not to persist whereas others are expected to spread rapidly through wild population. In cases where the LMMs have been found through the risk assessment process to have the potential to cause adverse effects to the biological diversity, taking also into account human health, methods may be needed to reduce the persistence of the transgene in the environment or to mitigate the expression of the transgene. Monitoring during and after the environmental release of the LMMs to address prompt detection of unexpected adverse effects may be recommended (see additional considerations on monitoring below).

**Risk Management strategies**

*(see Step 5 of the Roadmap for Risk Assessment)*

Risk assessors may want to consider the following risk management strategies for the release of LMMs into the environment:

1. Monitoring monitoring is NOT a risk management strategy! during and after the environmental release of LM mosquitoes to follow the spread of the trait in the local population.. Operational management processes should carefully follow the design criteria for implementation of the risk management strategies laid out in the risk assessment;
2. Monitoring the potential evolutionary breakdown of the mosquito technology (monitoring for transgene intactness and proper function over time),
3. Monitoring the efficacy and effectiveness of mosquito technology;
4. Monitoring strategies for managing the dispersal and to ensure that the LMMs do not establish themselves beyond the intended receiving environment;
5. Halting the releases if unanticipated effects occur; and/or
6. Mitigations, such as an alternative set of control measures should a problem occur.

**Other Issues**

Somewhere we need a sentence that evaluation leading to deployment should be done with *transparency* (i.e. without burying/suppressing data), and in a *methodical* and *stepwise* manner (e.g. lab trials, cage/semi-field trials, limited open release, full-scale open release, pilot programme, roll-out). Each step should be preceded by risk assessment and risk management (within the scope of this document) as well as other considerations (e.g. ESC) which are outside the scope of this document.

**BIBLIOGRAPHIC REFERENCES**

*See references relevant to the “[Guidance Document on Risk Assessment and Risk Management of LM Mosquitoes](http://bch.cbd.int/onlineconferences/mosquitoesref_ahteg_ra.shtml)”.*

Add

[Female-specific flightless phenotype for mosquito control.](http://www.ncbi.nlm.nih.gov/pubmed/20176967)

Fu G, Lees RS, Nimmo D, Aw D, Jin L, Gray P, Berendonk TU, White-Cooper H, Scaife S, Kim Phuc H, Marinotti O, Jasinskiene N, James AA, Alphey L.

Proc Natl Acad Sci U S A. 2010 Mar 9;107(10):4550-4.

Also, a more recent version of the WHO report stemming from the May, 2009, meeting is now available from Dr. Yeya Toure.

Number of papers in this special issue on transgenic insects:

http://www.msmbb.org.my/apjmbb/html173/173cont.htm

1. The Parties to the Cartagena Protocol on Biosafety have mandated the AHTEG to ‘develop a “roadmap”, such as a flowchart, on the necessary steps to conduct a risk assessment in accordance with Annex III to the Protocol and, for each of these steps, provide examples of relevant guidance documents’. The Roadmap is meant to provide reasoned guidance on how, in practice, to apply the necessary steps for environmental risk assessment as set out in Annex III of the Protocol. The Roadmap also demonstrates how these steps are interlinked. [↑](#footnote-ref-1)
2. The present document has been prepared by the AHTEG and its Sub-Working Group (SWG) on Living Modified Mosquitoes (LMMs) incorporating comments received via Open-Ended Online Forum. [↑](#footnote-ref-2)