Use of Transgenic Arthropods in Genetic Control Programmes:

Strain evaluation and risk assessment at the International Atomic Energy Agency

Hervé C. Bossin, Gerald Franz, and Alan S. Robinson

Joint FAO/IAEA Programme of Nuclear Techniques in Food and Agriculture
International Atomic Energy Agency, Vienna, Austria
Contact Person: H.Bossin@iaea.org

The IAEA is involved in the assessment and use of modern biotechnology likely to enhance the peaceful use of nuclear energy and to improve the delivery of the Agency's programmes to Member States.

Probably the most suitable pest control strategy for the integration of transgenic technology is the Sterile Insect Technique (SIT), where the presence of the transgene in the environment is limited to the sterile insects that are released. Germline transformation technology is functional in a growing number of arthropods species including species of medical, agricultural and veterinary importance for which SIT is already implemented (fruit flies, tsetse flies, moths) or is under development (mosquitoes). Potential improvements include the development of strains that (1) produce only male insects for sterilization and release and (2) carry a marker that distinguishes them from wild insects, thus increasing the efficiency of these SIT programmes.

However, major technical challenges remain that concern the genotypic and phenotypic stability of the transgenic strains and their ability to express the transgene in a reliable and predictable manner in operational programmes where many millions of insects have to be reared for sterilization and release. A second area of concern is the biological fitness of transgenic strains when they have to compete with individuals in the field. Finally, of much broader significance to the use of transgenic arthropods is the development of an appropriate regulatory framework. Insects that are currently released in SIT programmes are not subject to significant regulatory constraints, but this will change if the released insects are transgenic, even if they are sterile. The release of genetically modified arthropods will require a thorough risk assessment protocol, moving from the laboratory through contained field cages to open field release.

Activities conducted at the IAEA on transgenic insects are designed to better understand their potential and limitations in relation to SIT and to improve decision-making capability in Member States through capacity building in the fields of biosafety and risk assessment.

Technical support

The IAEA conducts adaptive R&D at its laboratories located in Seibersdorf, Austria. Mediterranean fruit fly *Ceratitis capitata* (medfly) SIT programmes in Member States have benefited tremendously through the introduction of genetic sexing strains constructed using conventional breeding techniques. This technology took 15 years to develop and cannot be transferred to other species where SIT programmes are being carried out. However, the potential exists that "generic DNA genetic sexing constructs" can be developed which, through transgenesis, can be introduced into the genome of many different pest species, without the need to develop a separate system for each individual species.

Research on transgenesis essentially focuses on two pest insects: the medfly, a major agricultural pest and the mosquito *Anopheles arabiensis*, one of the three main malaria vectors in Africa. For mosquitoes, it is essential to develop a system to eliminate females, either based on classical breeding techniques or transgenesis, in order to implement an SIT programme. The first germline transformation of *Anopheles arabiensis* was recently achieved at the Entomology Unit using a *piggybac* transformation vector. This represents a significant achievement which opens the

prospect for developing transgenic-based sexing systems potentially allowing the stringent elimination of females required for the release of sterile male mosquitoes.

In all SIT programmes it is essential to mark sterile insects before they are released so as to be able to differentiate them from their wild conspecifics during the evaluation of the programme. This is currently achieved using a fluorescent powder that has major disadvantages for worker health, accuracy and field monitoring costs. Using transgenesis it is possible to mark released insects with a molecular tag that will make recognition unequivocal and easy to implement. Several strains are now being evaluated which carry a molecular tag.

Biosafety and risk assessment of transgenic fruit flies and mosquitoes are being carried out in contained environments to provide credible, independent technical information to organisations in Member States who may wish to consider the use of this technology in their SIT programmes. This information can be used to make science-based decisions as to the risks and benefits associated with the technology. The Agency has considerable expertise and credibility in the field of genetic control of insect pests and the activities on transgenic insects improves considerably the quality of advice for Member States. The evaluation of transgenic strains includes the assessment of transgene stability and consistency of transgene expression under various (mass)-rearing regimes. The insect rearing facility provides an ideal infrastructure to conduct this work. Fitness evaluation of transgenic insect strains in large field cages will be conducted in a fully contained 250m^2 greenhouse currently under construction.

Over the past fifteen years, the Agency has contributed, through the organization of Coordinated Research Projects (CRP) and consultancy meetings, to the development of knowledge in the field of insect transgenesis with direct applications for SIT. The latest CRP specifically addresses the use of transgenic arthropods to improve the effectiveness of the SIT. This CRP initiated in 2004 for a period of 5 years involves more than twenty partners and observers from both developed and developing countries. CRPs thus contribute in building relevant research capacity in developing countries.

Regulatory support

Of much broader significance to the use of transgenic arthropods is the development of a regulatory framework. Current national regulatory processes, including the availability of suitable risk analysis protocols, may be insufficient to address any eventual release of transgenic arthropods. Likewise, the current negative public perception of transgenic technology in general will make the development and use of transgenic arthropods in pest control potentially difficult. Policy making in regard to transgenic arthropods lags far behind that for transgenic plants and micro-organisms, where the latter are now being extensively used in agriculture and human health. However, USDA through APHIS has developed a set of guidelines that were used to issue a permit for the first field cage evaluation of a transgenic plant pest in 2001. More recently, the Joint FAO/IAEA Division of Nuclear Techniques in Food and Agriculture, IAEA, Vienna and the International Plant Protection Convention (IPPC) secretariat, Rome organized a meeting[1] to address the biosafety and regulatory issues described above. This meeting brought together both scientists involved in transgenic technology and experts in the field of risk assessment and regulatory procedures and led to the development of a preliminary set of risk assessment protocols related to the use of transgenic arthropods in agriculture.

The IAEA will continue its efforts in the development of an international regulatory framework including aspects of risk identification, risk assessment and risk management.

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[1] IAEA technical documentation on the Status and risk assessment of the use of transgenic arthropods in plant protection. http://www-pub.iaea.org/MTCD/publications/PDF/te_1483_web.pdf