Impact of irradiation on vector competence of *Aedes aegypti* and *Aedes albopictus* for dengue and chikungunya viruses

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INTRODUCTION

The sterile Insect Technique (SIT) is considered one of the most innovative approaches that can lead to vector suppression and containment of vector-borne diseases. This technique is based on the releases of large numbers of sterile males which will compete with wild males for mating with the wild female population, leading to a reduction of progenies. Although mechanical sex separation techniques are of high efficiency, a small percentage of females could be accidentally processed together with the males and then released in the field. These females are sterile but can bite and therefore cause risk of disease transmission. The aim of this study was to evaluate the effect of irradiation on dissemination and transmission of Dengue (DENV) and Chikungunya (CHIKV) viruses in *Ae. albopictus* and *Ae. aegypti* females.

METHODS

MOSQUITOES	Ae. *albopictus* strain Rimini (Italy); *Ae. aegypti* strain Juazeiro (Brazil)
IRRADIATION	Female pupae irradiated in water with TrueBeam linear accelerator
	Dose 40 Gy; dose rate 6.2 Gy/min; photons energy 6.0 MV
VIRUSES	Dengue type 2 virus (DENV-2); Chikungunya 06.21 (CHIKV)
INFECTION	Virus RNA load CHIKV $1*10^8$ PFU/ml (17.2 Ct); DENV $1*10^6$ PFU/ml l (21.21 Ct)
	Seven-day-old females fed with 3 ml of infectious blood for 30 min.
	Incubation period of 7 days (CHIKV) and 14 days (DENV)
DISSECTION	legs&wings and saliva samples from each female were obtained
VIRAL RNA	Dissemination quantified by rRT-PCR in legs&wings sample pools (n=8)
	Transmissibility analyzed from saliva samples (pools and individual)

RESULTS

DISSEMINATION (POOLS)

All the pools of legs&wings from both irradiated and unirradiated females infected with CHIKV were positive while for DENV the percentage of positive pools ranged between 64 and 100% with no differences between irradiated and unirradiated females.

Higher infectivity was detected among disseminated female infected with CHIKV most likely due to the initial infectious dose which was higher for CHIKV ($1*10^8$ PFU/ml) then DENV ($1*10^6$ PFU/ml).

TRANSMISSION (POOLS)

All saliva pools obtained from females infected with CHIKV were positive, while only 8 to 27% of the saliva pools from females fed on DENV were positive with 1 to 2 log lower PFU range regardless of radiation exposure (Fig. 1 b, e).

Radiation seems to reduce the transmission, with a significant interaction between the virus type and the radiation status in *Ae. aegypti* (Figure 1 b) but not in *Ae. albopictus* (Fig. 1 e). Overall, irradiation had no effect on the viral load measured in both species with either DENV or CHIKV (Fig. 1 b, e).

TRANSMISSION (INDIVIDUAL FEMALES)

Radiation had no effect on the viral load measured in both species with either DENV or CHIKV (Fig. 1 c, f) without an interaction between the virus type and the radiation status in both species. The reduced transmissibility of DENV in irradiated *Ae. aegypti* females in comparison with unirradiated females was not confirmed in individual saliva samples analysis (Fig. 1 c).

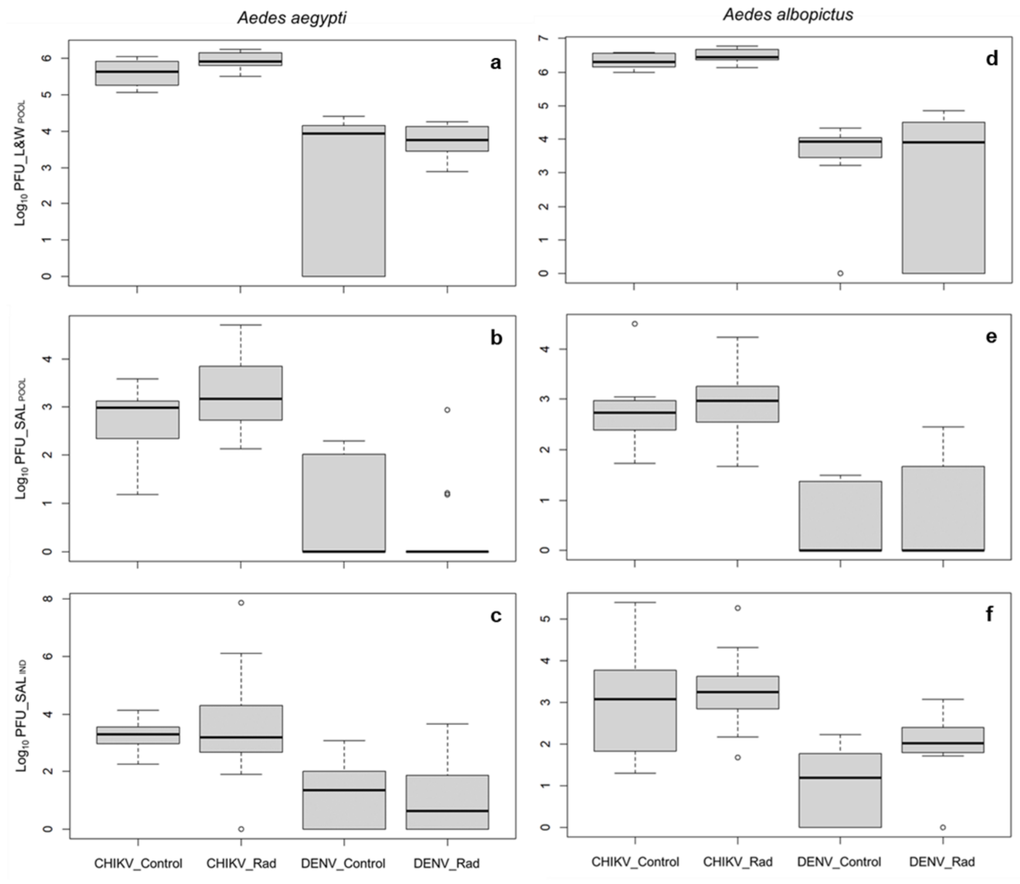

Fig. 1 - Boxplots of the viral load (Log10 PFU) measured in pooled samples (POOL) of legs&wings (L&W) and saliva (SAL) from irradiated (Rad) and unirradiated (Control) *Ae. aegypti* (a, b) and *Ae. albopictus* (d, e) mosquitoes infected with CHIKV or DENV. Each pool sample consisted of homogenates of legs&wings and saliva from 8 individual females. Viral load (Log10 PFU) measured in saliva samples from individual females (IND) are also reported from both irradiated and not-irradiated Ae. aegypti (c) and Ae. albopictus (f) mosquitoes.

CONCLUSIONS

Irradiation of female *Ae. aegypti* and *Ae. albopictus* as pupae with a dose of 40 Gy had no significant effect on the dissemination and transmission rate of DENV and CHIKV. One of the most common effects of radiation-induced somatic damages is a dose-dependent reduced longevity [1] which may decrease vectorial capacity [2]. Moreover, the reduced blood feeding frequency observed in irradiated Aedes females [1], and the increased mortality resulting after the ingestion of an infected blood meal [3], can have a further negative impact on the overall vectorial capacity of irradiated Aedes females accidentally released in Aedes vector SIT control program. This study showed that irradiation did not significantly impact the vector competence of irradiated Aedes females, thus allowing an evidence-based risk assessment of females release together with the sterile males.

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