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## ***Assessment of Mosquito Control Sprayers performance based on physical deposition and biological efficacy***

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### **Abstract**

Mosquito Control in France represents about 45 000 ha (domestic and overseas area) and represents a public health concern operated by public authorities. Apart from aerial spraying, mosquito control is mainly achieved by agricultural sprayers (pickup – ATV or manually mounted mist blowers). As spraying applications may lead to variable performance, a specific research program was designed in order to simultaneously assess spray deposition and biological efficacy. This study was a part of the overarching project (Life + Integrated Mosquito Control Management - LIFE08/ENV/F/000488). Four mist blowers were tested (Martignani B748 – Tifone City 300 (ATR- TVI nozzles) – Swingtec Mobilstar – knapsack mist blower with AU 8000 rotary nozzle) as used for larvicide application. 22 tests were conducted including 3 repetitions for each equipment setup. BTi (*Bacillus Thuringiensis israelensis*) was applied with an application volume of 8 to 12.5 L/ha including 1 to 2.5 L/ha of BTi Vectobac WG® or Aquabac XT® respectively. Each tank mix contained about 1g/L of Brilliant SulfloFlavine (BSF) dye tracer. Deposition values at different distances were calculated after tracer content measurement. 3 Petri dishes (f 8 cm) were used to collect ground deposits and placed around a jar including 25 mosquito larvae (*Aedes Aegypti* L.) for biological control. Alive larvae were counted immediately after application, 24h and 48 h after application. Average deposition in 3 Petri dishes was compared with alive larvae at different distances and for each kind of equipment. 30 collecting plots placed on a 10m x 10m grid leading to 5 lines of 4 plots located at 20-30-40-50m from the application path. A central line of collectors placed every 5m was used from 55 to 90m. Applications were realized downwind as advised by mosquito control operators at 8 km/h for vehicle mounted sprayers and 4 km/h for hand operated sprayer. Individual performance of each sprayer (i.e. efficient spray range) was compared to others and to manufacturer indications. A deeper analysis in terms of lethal dosage of Bti was also realized. For example, an application volume of 0.5 L/ha (1/25<sup>th</sup> of legal application volume) was found to generate acceptable mortality in some cases. Tentative of explanation regarding the global variability obtained after crossing physical deposits and biological efficacy was conducted. Among all parameters of influence, droplet size and atmospheric conditions were mainly identified. Indeed, best biological efficacy was obtained with lower application volume when small droplet size were produced (Tifone and rotary nozzle).

**Keywords:** mosquito control, spraying efficacy, biological efficacy, mist blowers

## Introduction

Mosquito Control in France represents about 45 000 ha (domestic and overseas area) and is of a public health concern managed by public authorities [BECKERT et al, 2007]. Apart from aerial spraying, mosquito control is mainly achieved by agricultural sprayers mainly pickup mounted mist blowers, ATV mounted air assisted sprayers or knapsack mist blowers [DERUDNICKI et al, 2012]. Since biological efficacy of active ingredients is largely studied at laboratory scale, the translation of results at field scale is much more difficult. Indeed, the dosage dependence is directly linked to the sprayer technology, to the spray range and to atmospheric conditions [DOUZALS, 2012]. The need for updated references lead to the implementation of this topic to an overarching project (Life + Integrated Mosquito Control Management) [LIFE08/ENV/F/000488].

## 1. Materials and methods

### 1.1. Spraying Techniques

Different spray application techniques are described in following Table1.

Table1. Sprayers characteristics and settings.

Spraying technique	Martignani B 748	Tifone City 300	Swingtec Mobilstar	Ciffarelli Nuova 5 HP
Type	Canon mist blower - pneumatic	Canon mist blower	Cold fogger	Motorized knapsack mist blower
Vehicle	Pickup	Pickup	Pickup	Manually operated
Spray range (m)	25	25		
Dv50 ( $\mu\text{m}$ )	250	150 - 300	20 - 100	40 - 120
Forward speed (km/h)	9	9	9	3
Nozzles type	ULV mode	Hollow cone ATR – TVI	-	Micron AU 8000
Max. Pressure (bar)	20	40	n.c.	n.c.
Air volume	$4500 \text{ m}^3 \cdot \text{h}^{-1}$	n.c.	none	$1200 \text{ m}^3 \cdot \text{h}^{-1}$
Air speed	$80 \text{ m} \cdot \text{s}^{-1}$			$125 \text{ m} \cdot \text{s}^{-1}$
Tank capacity (L)	300	300	80	14
Power (kW)	11.7	19.8	13.2	3.68

*n.c.* : not communicated by manufacturer

Droplet sizes were measured using a Malvern Spraytec device under static conditions at 50 cm from canon/diffuser outlet.

Pickup mounted equipments were used at  $9 \text{ km} \cdot \text{h}^{-1}$  and knapsack sprayer was  $3 \text{ km} \cdot \text{h}^{-1}$ .

All applications were achieved in the downwind direction accordingly to good application practices for mosquito control in France.

For Pickup mounted equipments, different orientations were given to the canon from  $0^\circ$  (horizontal position),  $25^\circ$  upward without obstacle and  $25^\circ$  with obstacle (greenhouse of  $17 \times 20 \text{ m}$  and  $3 \text{ m}$  height). These last two orientations were tested to simulate eventual difference in biological efficacy under different sprayer settings.

Forward speed, flow rates and GPS positions were recorded by using the TICSAD® traceability system developed at IRSTEA Montpellier [TICSAD].

Spray mixes were composed of BTi (*Bacillus Thuringiensis var. Israelensis*) under two different formulations: Vectobac WG ( $3000 \text{ UTI/mg} - 1 \text{ kg} \cdot \text{ha}^{-1}$ ) and Aquabac XT ( $1200 \text{ UTI/mg} - 2.5 \text{ L} \cdot \text{ha}^{-1}$ ) applied at a volume rate of  $8 \text{ L} \cdot \text{ha}^{-1}$  and  $12.5 \text{ L} \cdot \text{ha}^{-1}$  respectively. A dye tracer (Brilliant SulfloFlavine) at  $1 \text{ g/L}$  was added in each tank mix. By using a calibration curve, the fluorescence value was related to a dye concentration and subsequently to the applica-

tion volume.

Meteorological conditions such as air temperature, relative humidity, wind speed and direction were recorded with a frequency of 10Hz and processed as a fantail showed in Fig. 2.

## 1.2. Experimental Setup

Larvicide applications were done on a line of 80 m and sprayer was oriented downwind. Collectors (3 Petri dishes per plot as shown in Fig.1) were located according to a sampling grid of 10 x 10 m and 50 m long and 40 m wide. A central line of collectors were located every 5m up to 85m (Fig. 2). During these experiments a rotary impeger [Clayson et al, 2010] was also tested.



*Figure 1: Range of collectors used : (left) Rotary impeger – (right) Petri dishes – Water Sensitive Papers*

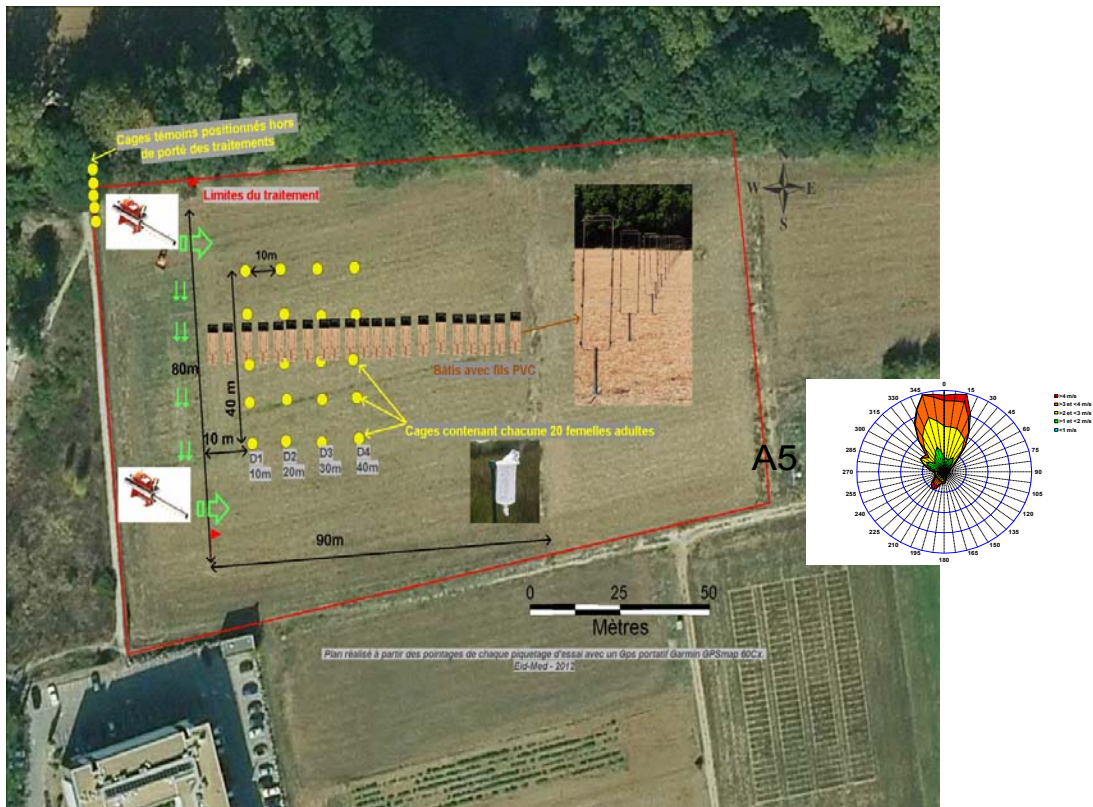


Figure 2: Experimental setup for larvicide application

### 1.3. Biological material

Mosquito larvae were produced into EID Méditerranée laboratories. Species were *Aedes (Stegomyia) Aegypti* L. sensible strain *Bora*. 25 alive larvae were placed into nalgen containers including 200 mL of distilled water with sugar. A test includes 27 containers at different distances (cf §1.2). Alive larvae are counted before spraying, 20min. after spraying when containers are collected, 24h after spray application and 48h after application. A set of 5 containers were placed out the spraying area (upwind) as non-treated samples. Mortality is given by the Abbott's formula (Eq. 1) that takes into account the mortality of treated and non-treated samples:

$$\frac{\text{mortality of treated samples}(\%) - \text{mortality of non-treated samples}(\%)}{100 - \text{mortality of non-treated samples}(\%)} \quad (\text{Eq 1})$$

Each modality (sprayer type and settings) was repeated 3 times.

## 2. Results

### 2.1. Canon mist blowers

The distance where a biological efficacy (mortality %) of 95 % is obtained is introduced in Table 2.



Table 2. Maximum distances where mortality is above 95%.

Settings	Martignani - Aquabac XT		Martignani - Vectobac WG		Tifone - Aquabac XT	
	Test	Distance Eff > 95%	Test	Distance Eff > 95%	Test	Distance Eff > 95%
horizontal	A1	30	V1	20	TA1	85
horizontal	A2	40	V2	20	TA2	85
horizontal	A5	40	V3	40	TA3	85
25° w/o obst	A3	20	V4		TA4	85
25° w/o obst	A4	30	V5		TA5	
25° w obst	A6	20	V6	20	TA6	25
25° w obst	A7	20	V7	20	TA7	17

Different trends were identified. (i) In all cases, the orientation of the canon (25° upward) involved a decrease in the max efficient distance. When spraying over the greenhouse, set as an obstacle, the max. efficient distance decreased additionally. (ii) the effect of sprayer is visible when comparing Martignani and Tifone with Aquabac XT. In the case of the Tifone fitted with ATR (small droplet size nozzles), a max distance efficiency of 85 m is obtained compared to 40 for the Martignani due to coarser droplet sizes. (iii) Vectobac WG formulation was less efficient compared to Aquabac XT due to a lower concentration of active ingredients and a lower application volume.

## 2.2. Knapsack with rotary nozzle

Tests were performed on a shorter sampling area with 3 lines of collectors and containers of 12m (collectors and containers placed every m). Results are shown in Fig 2.

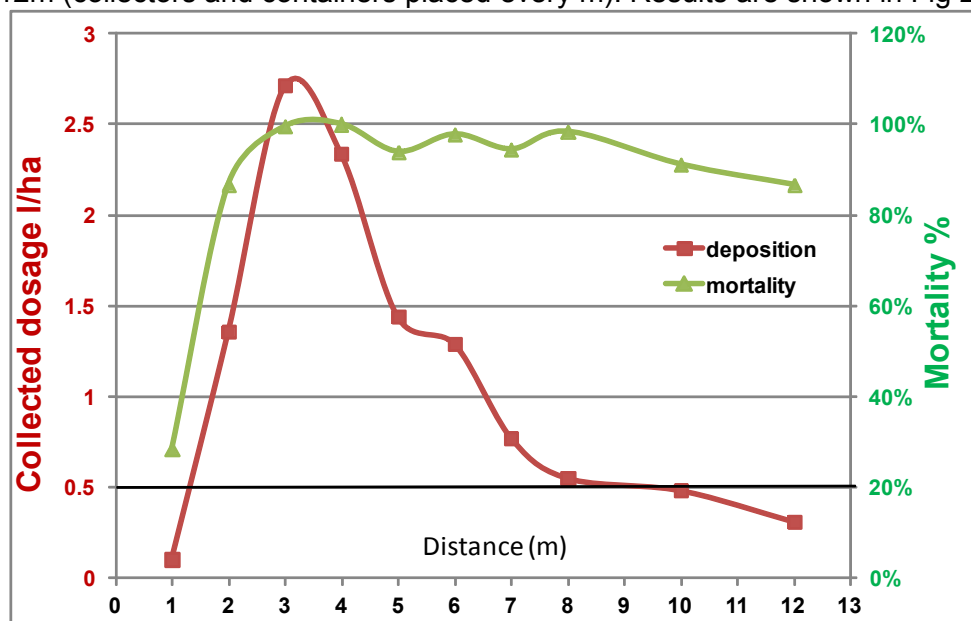


Figure 2. Comparative values of BTi deposition and mortality for different distances. Average of 3 replicates.

Acceptable biological efficacy (> 95%) is shown from 2m to 8m that is a current spray range for this kind of equipment.

## 2.3. Discussion on BTi dosage

Mortality vs. collected doses were plotted for canon mist blowers and the motorized knapsack mist blower. Altogether 579 values were obtained and introduced in Fig 3.

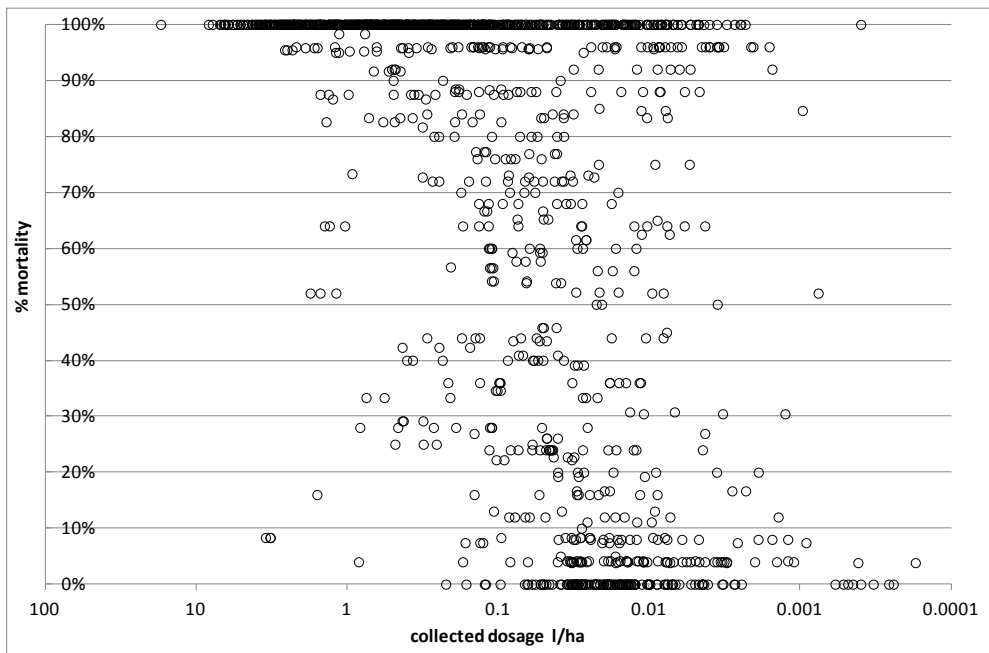


Figure 3: Distribution of mortality % with according collected dosages

Values situated in the range of 0-10% and 90-100% of mortality correspond to 75 % of the total number of values. Mortality curve as a function of recovered dosage % follows a inverse log trend with only 25 % of the total number of values situated between 10% and 90% of mortality. Considering the respective concentration in active ingredients (UTI/mg) for both Aquabac XT and Vectobac WG, data shown in Fig 4 show the dosage efficacy (10<sup>th</sup>, 50<sup>th</sup> and 90<sup>th</sup> percentile) for each range of mortality.

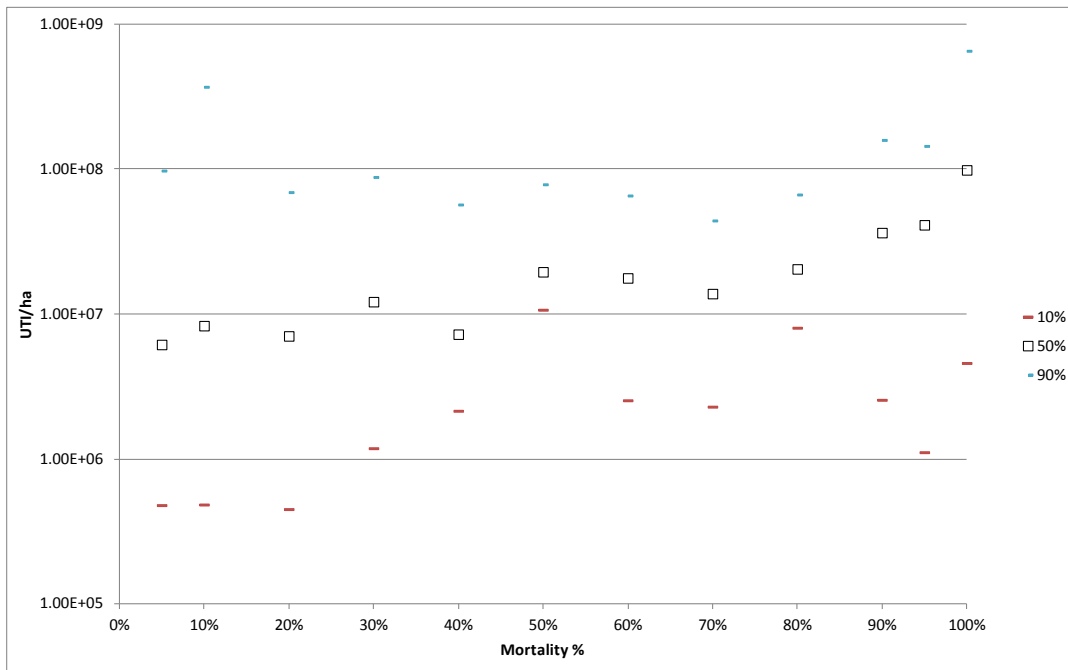


Figure 4: Evolution of different percentiles of the effective dosage (UTI/ha) for different ranges of mortality

As a result, the average BTi dose necessary to obtain a mortality of 95% is 0.1 L/ha for Aquabac XT and 0.5 L/ha for Vectobac WG i.e. 0.8% and 6% of applied dose respectively.

## Conclusions

A comparative study of ground deposition and biological efficacy was achieved for different types of sprayers used for mosquito larvae control. Both effective spray and mortality ranges appeared highly dependent on the quality of spraying (fine droplets are better) and atmospheric conditions (wind speed and direction). Deeper investigation on the effective dosage at local level showed a high variability depending on the BTi formulation. Average effective dosage (95 % mortality) appeared to reach 0.1 to 0.5 L/ha corresponding to 0.8% and 6% respectively of the applied volume. Potential developments are foreseen in the optimization of mosquito control spray applications as well as real time and in-field evaluation indicators of the quality of spraying.

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