This highlight refers to the PhD Thesis by Antoine Cribellier (2021, Cum Laude)

Biomechanics of the capture and escape of malaria mosquitoes

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Haematophagous female mosquitoes need to interact with their hosts in order to get a blood meal necessary for egg production. Many dangerous pathogens such as the malaria parasite and the dengue virus take advantage of this mosquito–vertebrate interaction to spread themselves. This is what makes mosquitoes the most dangerous animal in the world. Malaria alone kills more than 400,000 people – mostly young children – every year (WHO, 2020), and killed many more in the past.

To get a blood meal, female mosquitoes must seek a vertebrate host (see Figure 1). For this, they can detect CO_2 and volatiles that signal the presence of a nearby host upwind (Cardé et al. 2010, Cardé 2015). Then, they will rely on visual cues to find the source of the odour plume (Van Breugel et al. 2015, Vinauger et al. 2019). Up until this point, the host-seeking flight behaviour of mosquitoes has been studied extensively, most often in wind-tunnel experiments (Kennedy 1940, Cooperland and Cardé 2006, Hawkes and Gibson 2016). Finally, mosquitoes will use additional cues, so-called short-range cues such as heat and humidity, to decide whether and where it will land (Hawkes and Gibson 2016, Beeuwkes et al. 2008. The importance of these cues is also well established but the flight dynamics of mosquitoes and the role of airflow – another important cue used by mosquitoes – in close vicinity of their hosts is still relatively poorly understood.

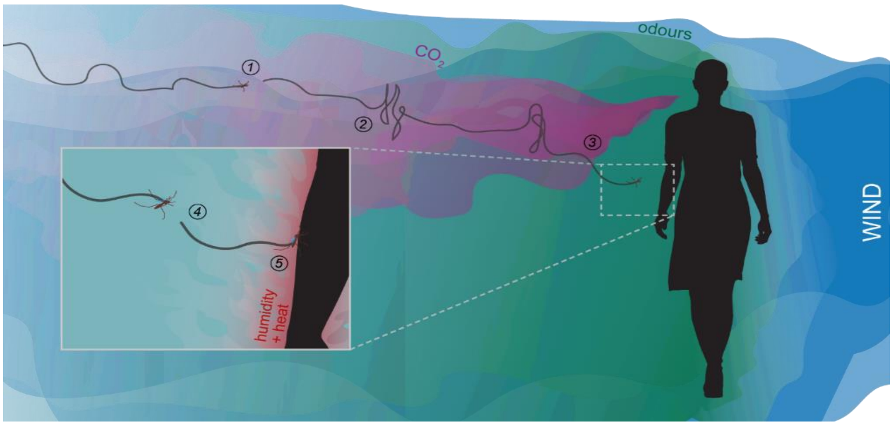

Figure 1: Infographic on how mosquitoes detect and fly towards a blood host (Cribellier 2021). (1) A flying female mosquito encounters CO_2 and odour plumes. (2) The mosquito will surge upwind and cast crosswind to find the source. (3) The mosquito will inspect visually interesting objects, and then (4) will use cues such as heat and humidity to select a spot to land (5).

Counter-flow odour-baited traps have been developed (Kline 2002, Kawada et al. 2007, Bhalala and Arias 2009). These traps use counter-flows generated by a fan to, first, diffuse a human-mimicking odour blend and CO_2, thus attracting host seeking mosquitoes in their vicinity, and second, capture mosquitoes that would have approached it by sucking them inside the trap (Kröckel et al., 2006). Until recently, odour-baited traps were exclusively used as tools for monitoring mosquito populations. However, they are now being considered as insecticide-free vector control tools that could effectively reduce mosquito populations when combined with bed nets (Homan et al., 2016).

A study was made on how mosquitoes behave around counter-flow odour-baited traps (Cribellier et al., 2018). We recorded thousands of three-dimensional flight tracks of female malaria mosquitoes (*Anopheles coluzzii*) around a well-known trap, the BG-Suna, in opposite orientations to represent two widely used traps. We visualized the average behaviour of mosquitoes and flight dynamics around the traps on two-dimensional heat maps. This allowed us to identify that mosquitoes were following stereotypical behaviours: when approaching the traps, by flying downward, and when close to the inlet of the traps, by accelerating upward. These behaviours led to very different capture dynamics of mosquitoes, and consequently to contrasting short-range attractiveness and capture mechanisms of the two traps. For example, the standing BG-Suna was more attractive than the hanging BG-Suna, while being also the only trap that triggered escape-like responses of mosquitoes.

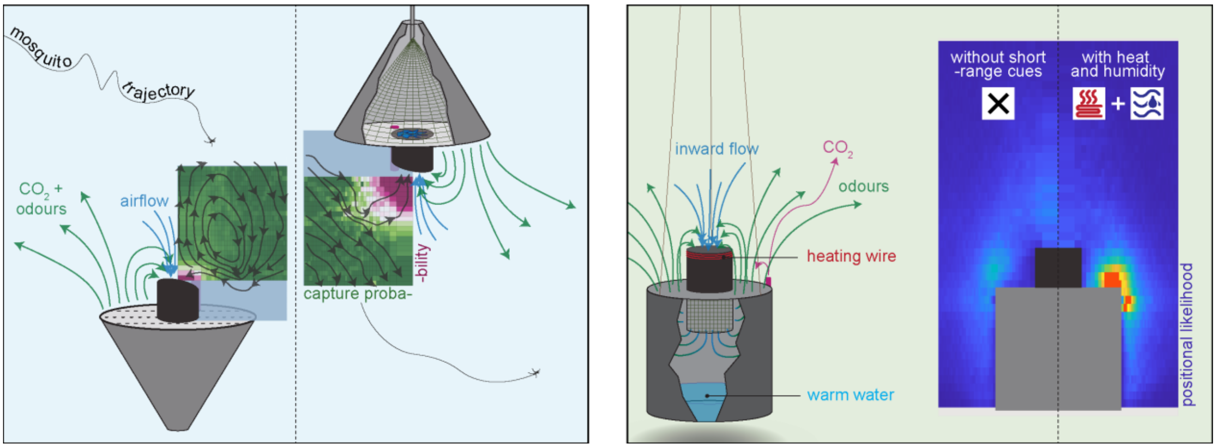

Figure 2: Graphical summary of the first (left) and second (right) studies (Cribellier, 2021). First, we studied how flying mosquitoes behaved around an odour-baited trap either standing or hanging. Then, we tested whether adding heat and humidity to a newly designed trap resulted in improved trapping performance, and how this impacted mosquito flight behaviour.

Based on our findings, integrated with the literature, we developed a new counter-flow odour-baited trap: the M-Tego (PreMal b.v.). One specificity of this trap is that it can generate two additional host cues that have been found to attract mosquitoes at short range, namely heat and humidity. Then, we tested this new trap against the BG-Suna in laboratory and semi-field conditions (Cribellier et al., 2020). In both conditions, the M-Tego without additional short-range cues was found to capture more than twice as many malaria mosquitoes than the standing BG-Suna. And when the M-Tego generated heat and humidity, it captured around 4.5 times as many mosquitoes as the BG-Suna. By recording the flight tracks of mosquitoes around this new trap, we showed that mosquitoes are more attracted to the M-Tego and spent more time close to it when the trap generates heat and/or humidity. Additionally, we did not observe escape-like responses of mosquitoes near the M-Tego, which explains why the M-Tego captures more mosquitoes than the standing BG-Suna even when they produce the same host cues.

Following this, we focused on the escape manoeuvres of flying mosquitoes. To study those, we built a new experimental setup where mosquitoes were tracked in three dimensions and in real time (Cribellier et al., 2022). Based on the real-time position of a mosquito, a mechanical swatter was then automatically triggered to simulate an attack towards it. This allowed us to study the escape performance of day-active and night-active mosquitoes (Aedes aegypti and An. coluzzii, respectively). For that, we recorded the flight behaviour of these two species when attacked by the swatter in four light intensities ranging from dark to overcast daylight conditions. Using Bayesian generalized linear models (B-GLM), we discovered that these mosquitoes exhibited enhanced escape performances in their respective natural light conditions (overcast for Aedes and dark for Anopheles). Furthermore, the high escape performance of Anopheles in the dark was mostly explained by its increased flight unpredictability in this light condition, whereas the higher escape performance of Aedes in overcast daylight compared with sunrise was due to its fast visually induced escapes.

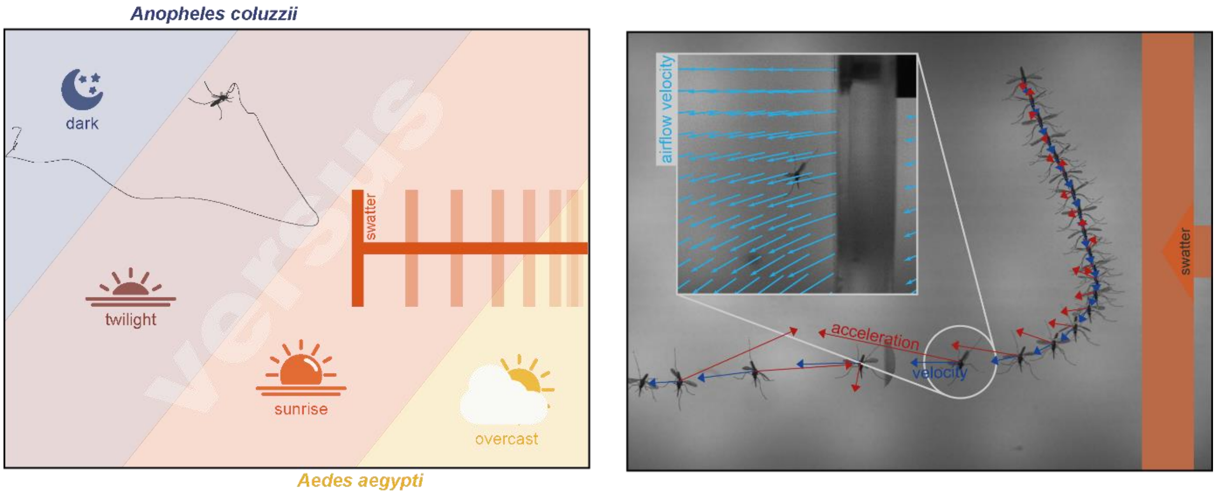

Figure 3: Graphical summary of the third (left) and forth (right) studies (Cribellier, 2021). We investigated how the escape performance of diurnal (Aedes) and nocturnal (Anopheles) mosquitoes varied with light conditions, as well as what were their respective escaping strategies. We also examined how mosquitoes performed escape manoeuvres and what was the role of the airflow induced by the attacker in their escape success.

Finally, we zoomed in on the escape manoeuvres of *An. coluzzii* mosquitoes with the aim to understand whether they rely on the airflow induced by the attacker to escape successfully. First, we compared the escape performance of mosquitoes when attacked by either a solid or a perforated swatter, in both dark and low-light conditions. We showed that the faster the air movements induced by the attack, the fewer mosquitoes that were hit by the swatter. This demonstrates that airflow plays an important role in the escapes of these mosquitoes. However, at this point, it was still unknown whether mosquitoes are using the airflow passively or actively. Then, using high-speed video cameras (12500 fps) and a newly developed neural-network-based tracker, we recorded the kinematics of the body and wings of flying mosquitoes when escaping. By combining this with results from a CFD simulation of the airflow induced by the attack, we estimated the aerodynamic forces involved during mosquito manoeuvres. We discovered that these mosquitoes, although seemingly moving passively with the airflow, were actively moving with the bow wave induced by the swatter. Moreover, we found that, despite that mosquitoes contributed actively to most of their escape acceleration, the passive effect of the airflow also significantly contributed to their success.

Competing interests

A.C. and F.M. are part of the scientific supervisory board of PreMal b.v., but have no financial competing interests.

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