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Research Article

Run Catch® A Novel, Early Warning Adult Mosquito Trap for Rapid, Low Cost, Extensive Entomological Surveillance and Risk Assessment of Mosquito-Borne Viruses and Malaria

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Abstract

The collection of a representative number of adult mosquito samples in any relevant territorial unit at risk of disease and malaria is delayed due to the difficulty of acquiring a sufficient number of samples during a given time period by rapid trapping, extensive trapping, etc.; nevertheless, monitoring of adult mosquito populations is vital for subsequent speedy investigations of pathogenic entity. Lack of such timely information prevents implementation of pertinent strategies that are technically sound, viable, economically feasible and helpful to those seeking proper measures and prioritization for control purposes.

In recent years, new vector-borne diseases in many European countries and sporadic incidence of some local transmission of malaria after World War II draw attention to this neglected risk and necessitate implementation of intensive surveillance of vector mosquitoes. A rapid, easy and extensive early monitoring is the key intervention to detect infected mosquitoes by regular monitoring of the adult populations in a given territory. The relatively high cost of monitoring adult mosquito populations and the necessity of repeated sampling discourage this practice. Therefore, monitoring of adult mosquito populations must be low cost and this theme is prefixed in the development of Run Catch.

In many European countries, vast areas of wetlands exist during most of the year providing ideal habitats for the breeding of a number of mosquito species belonging to the genera, *Aedes, Culex*, and *Anopheles*. Adults of these mosquitoes besides being ferocious biters are potential vectors of disease organisms and have the capability of dispersing over vast areas from the breeding sites.

Keywords: Run Catch; Trap; Mosquitoes; Entomological Surveillance; Malaria; Vector-Borne Diseases

Introduction

The present study was conducted to evaluate the efficacy of an in-house developed adult mosquito surveillance system, 'Run Catch' and compare efficacy of this tool with conventional methods, such as Co₂ trap and leg-bite, to gain better knowledge concerning adult mosquito populations and early detection of potential disease vectors. Importantly, the causative agents of many diseases are transferred between hosts by certain species of bloodsucking

Culicidae [1,2]. In a recent study, the following deadly viruses were detected in pooled mosquito samples from northern Italy: West Nile virus in *Culex pipiens*, two orthobunya viruses and Tahyna virus in *Ochlerotatus caspius* and Batai virus in *Aedes maculipennis* [3]. Since different species within these complexes can sustain arboviral outbreaks [4-7], specific diagnosis of the vectors is essential for the development of control strategies.

Materials and Methods Run catch trap®

This is a patented device made from a net mounted on top of a moving car for collecting adult mosquitoes and is linked to a GPS system which collects GIS information for tracking mosquito samples. The intial version of device consists of a wooden board secured on top of a pick-up truck to which four circular (30 cm diameter) 2 meters long nylon nets closed at one end are attached for capturing adult mosquitoes



Figure 1: Wooden structure formed by four plastic rings with one mm pore size net, fixed on the roof of a pick-up van during the experiment. Mosquitoes collected in the nets can be seen in the close up view of the trap.

Study site

Field evaluation of the adult mosquito collection nets was conducted for several days during the months of July and August 2008, 2009 and 2010 at Alluvioni Cambiò, a small town located in Piedmont in the northern part of Alessandria Province, Italy (Figure 2a). Each summer residents of this small town suffer from the attack of adult mosquitoes (see supplementary file, S1) belonging to several species, *Ochlerotatus caspius* in particular. These mosquitoes prevail in large numbers in the area because of the proximity of rice fields which support their heavy populations as well as the presence of Po River which provides ideal habitat for mosquito breeding. (Figure 2)

Experimental design

During the first year (summer 2008), the effective surface area of trapping net and maximum abundance time of flying mosquitoes in the test area were determined (Figure 2a). For trapping net, a wooden board with four plastic rings of two different diameters (20 and 30 cm) with attached 2 m long nylon nets closed at one end were used to capture adult mosquitoes. The system was fixed on the top of a pick-up truck (Figure 1). The vehicle was driven at a constant speed of 40 km/h for a distance of 1 km (Figure 2b). To maintain the desired constant speed, it was necessary to pro-

vide an initial acceleration to reach the desired speed at the time of departure. In order to avoid accidental capture of adult mosquitoes during the acceleration phase of the vehicle, it was necessary to cover the capturing mouth of each net with a panel, removing it promptly after achieving the desired speed. The catches were repeated (8 times) in the North-South and East- West directions for 10-minute intervals, at 10 minutes before the local sunset time and 40 minutes after the sunset as the experiment's starting and termination points. With the "Run Catch", two standard Co2 traps were simultaneously placed, one at the beginning and one at the end of the collection path (Figure 2b) to evaluate the prevalence of adult mosquito species in the trial area. After the field capture phase, the captured adult mosquitoes in each 10 minutes' interval, were counted. During the following year, the captures with optimized size of trapping net, 30 cm diameter, were made. In addition, to correlate the number of total mosquitoes in a standard trap, four volunteers were placed to perform leg test though uncomfortable. The test area of "leg monitoring" located 800 meters away from the ongoing "Run Catch" experiment path in a football field. Every 10 minutes the number of mosquito bites on one leg (of the 4 volunteers positioned in 4 different areas of the field) was monitored. In the third year of experiment, 'Run Catch' trapping was repeated and the same protocol tested in a different field area in the village of Oviglio, situated in a similar set up to Alluvioni Cambiò, 40 km in the southern part in the vicinity of 300 ha of irrigated rice fields and having heavy infestation of mosquitoes.



Figure 2: The study site and its surrounding areas; 2a) Alluvioni Cambiò, a small town located in Piedmont in the northern part of Alessandria Province, Italy. The large area of nearby rice fields is circled in yellow; 2b) proximity of Po River. The study site for adult mosquito collection was selected between the River and Alluvioni Cambiò. The yellow line shows the one km long path of sampling adult mosquitoes and location of two Co2 traps (indicated by number 1 and 2) placed at the start and the end point of the one km long sampling path.

A version of this trap, has been designed for night use, by installing a micro-reflector at the mouth of each trap for illuminating the trap's net while the truck is running and consequently increasing attraction of mosquito species that are not abundant during the twilight hours; usually, fewer Anopheles mosquitoes, in comparison with Aedes and Culex, are collected. Recently, new vectorborne diseases in many European countries and some local transmission of malaria after the Second World War draw attention to this neglected risk, which imposes, to whom it may concern, the implementation of intensive surveillance. A rapid, easy and extensive monitoring is the key intervention if an imminent presence of infected mosquitoes are reported early by regular monitoring of the adult populations in a given territory. The high cost of adult monitoring is not conducive to widespread adult monitoring and the rather necessary repetition of the same. Therefore, the monitoring should be low cost and this consideration is prefixed in the development and realization of Run Catch Co2 -baited tarps (Talbalaghi, personal observation).

For the evaluation of efficacy of attractiveness of Run Catch Trap to *Anopheles* spp. mosquitoes, the nocturnal version is developed.

Having a low-cost, rapid tool for extensive use and for monitoring where attractive substances such as Co_2 and Lure are not available, greatly facilitates entomological surveillance and above all the early detection.

Data treatment

All field collected mosquito counts (either with 'Run Catch' or Co₂ trap or Leg test) were processed according to mosquito species type, time of collection and type of collection. The polynomial correlation between Run Catch and Co₂ traps was determined.

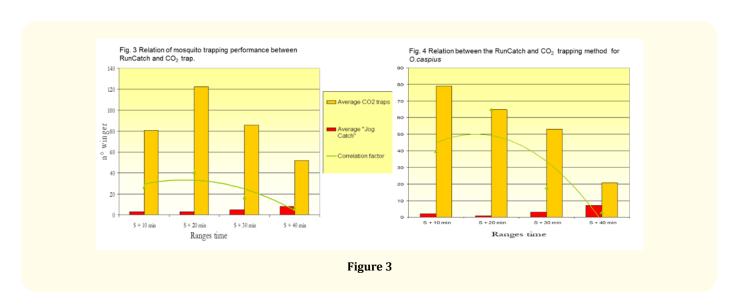
Results

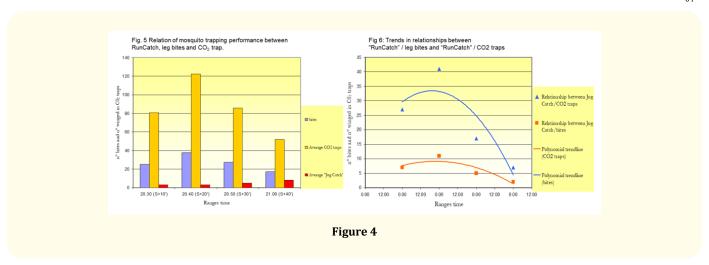
At first, the abundance of mosquito species in the experimental area by standard method (Co₂ trap) and test method (Run Catch)

(Supplementary Figure S1) was determined. Both methods showed similar proportions of the relative abundance of mosquito species, confirming the validity of new experimental approach for mosquito collection. As can be seen that the mosquito species collected were predominated by *Oc. caspius*, (60%), followed by another typical rice field inhabitant, *Culex modestus* and the ubiquitous *Cx. pipiens*. The catch data values are in agreement with the trapping studies carried out over the years through the use of Co_2 traps (S1).

Secondly, the mosquito collection experiment was divided into 4 significant time intervals from 10 minutes starting from sunset minus 10 minutes (S - 10), and then the total average of the catches of all the mosquito species obtained were calculated with both Co₂ traps and with the Run Catch, in each of the 4 time intervals (Figure 3). The catches with the Run Catch started at $10\ minutes$ after sunset and the catch values increased until the last interval "S + 40 minutes", while the Co₂ traps had a peak catch in the second interval "S + 20 minutes" and declining thereafter. Before sunset and 40 minutes after sunset the results are equal to zero. In fact, Co₂ traps are attractive devices so they have a faster effect than the Run Catch, starting to collect as soon as the intensity of the sunlight changes at sunset, while the "Run Catch" increases its effectiveness when mosquitoes are widespread in the environment. The values of the relationships between the two methods have a linear trend over time. The relationships between these values are the correlation factors, which can be used as multipliers to convert the catch data with the "Run Catch" in data of abundance of catches with Co, traps.

The correlation was defined only for the most abundant and aggressive species, *O. caspius*, considering that its presence in the territory is in itself significant (over 60% of the total) (Figure 3). However, the trend obtained is always comparable to the previous ones (Figure 4).





Leg monitoring was carried out to better assess the abundance and nuisance of mosquitoes; the data of the Co_2 traps and the "RunCatch" are compared with those of the leg pricks in the leg monitoring. Figure 5 shows the tendency of catches with Co_2 traps and leg bites is similar, the ratio is 4:1 in "S + 10 min" and "S + 20 min", 3: 1 in "S + 30 min" and "S + 40 min"; A peak can be seen in

the period "S + 20 min" which then declines. While the tendency of the "Run Catch" is inversely proportional to the others, showing a growth from "S + 10 min" (value 3) up to the maximum (value 8) of "S + 40 min". Finally, the relationship between "Run Catch" - leg monitoring was compared with "Run Catch" - Co_2 traps (Figure 5).

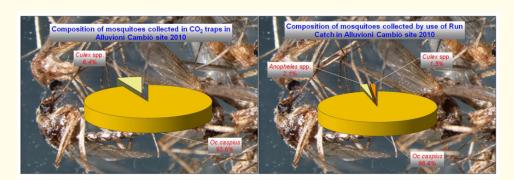


Figure 5: Comparison of simultaneous mosquito collection by Co₂ Trap (left) and by RunCatch Trap in Alluvioni Cambiò location at southern shore of Po River, a few km away far from the 100,000 ha ricefield area simultaneous with same procedure in Oviglio Location 40 Km far from Alluvioni Cambiò.

Speed	Collection Time			
40 km/h	S+10 min	S+20 min	S+30 min	S+40 min
All mosquito species -with trap co ₂	N × 27	N × 41	N × 17	N × 7
o. Caspius -with trap co2	N × 40	N × 65	N × 18	N × 3
All species with legbite	N × 7	N × 11	N×6	N×2

Table 1: Application of correlation factor to determine the relative mosquito abundance as could be predicted through Co₂ trap and/or leg bite estimation.

Transformation of the correlation factor values from "Run Catch" (N) to those with ${\rm Co_2}$ traps or leg bites.

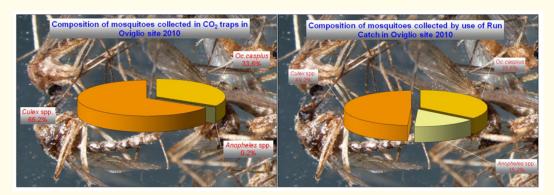


Figure 6: Comparison of simultaneous mosquito collection by Co₂ Trap (left) and by RunCatch Trap (right) in Oviglio location in southern part of first location (40 km away) with 300 ha of ricefields simultaneous with the same procedure in Alluvioni Cambiò location.

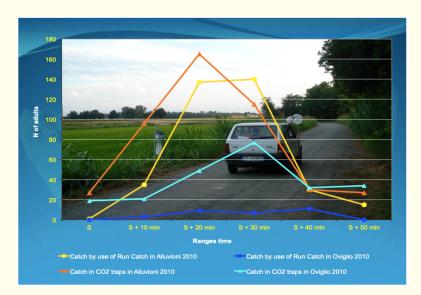


Figure 7: Comparison of simultaneous mosquito collection by Co₂ Trap RunCatch Trap in two locations, 40 Km far from each other of Oviglio and Alluvioni Cambiò in the Province of Alessandria.



Figure 8: First experimental Trap.



Figure 9: Nocturnal version.



Figure 10: Leg monitoring for mosquito landing simultaneously with the Co₂ Trap and RunCach eveluataion.



Figure 11: The final version of Run Catch, called Run Catch TV.



Figure 12: Different version of Rucn Catch.



Figure 13: Fixed version and rotating of Run Catch (R3P).

Discussion

The variation in the attractiveness of mosquitoes can be modified and therefore some factors, such as the dispersal capacity of the mosquitoes of the genus Aedes vary widely, especially the species, Aedes albopictus and Aedes aegypti. The final version of this trap (Figure 13) is not suitable to trap the mosquito species with low dispersal capacity and therefore, some other versions are being studied, taking into account the introduction of color of the car and heat for greater attraction for the species mentioned above, therefore, several versions of Run catch are in the process of perfecting and studying the materials used and the projected names of the versions follow: RunCatch, RunCatch, RunCatch, RunCatch, RunCatch

Conclusion

The objective of this invention and methodology is to acquire rapid, low cost, and extensive adult monitoring device for early surveillance of the infestation and risk assessment of viral-borne diseases and malaria by individuation of infected mosquitoes, rapidly collected and, screened even in the field by Rapid Diagnostic Test. This would arrive in area of first occurrence which, usually far from human habitation centres where the local mosquitoes could get infected first by blood feeding on these infected birds. This novel tool will also improve the early operational activity and the rapid countermeasures to be adopted and also for the evaluation of the effectiveness of taken measures.

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This observation confirms that the method of Co2 traps and the leg monitoring are appealing (attractive to mosquito), instead the "Run Catch" follows the tendency of diffusion of the mosquitoes.

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