Improving strategies for controlling Culicoides spp. with available ready to use adulticides

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Introduction and Background

Culicoides, commonly known as biting midges (Ceratopogonidae), are vectors of several pathogens that pose significant threats to both animal and human health. These vectors are responsible for the transmission of diseases such as bluetongue virus, epizootic hemorrhagic disease, and, more recently, oropouche fever. Oropouche fever stands apart from the other mentioned diseases because it can be transmitted to humans, whereas bluetongue and epizootic hemorrhagic disease primarily affect ruminant animals. *Culicoides* and mosquitoes share similar habitat and can vector diseases, which makes mosquito control operations uniquely placed to develop and respond to the threat posed by *Culicoides.* Effective control of *Culicoides* populations is crucial for reducing the risk of these diseases. However, managing these biting midges remains challenging due to their small size, cryptic habitats, and complex behavioral patterns.

One approach to mitigating *Culicoides*-borne diseases is the use of adulticides to target adult midges. While adulticides have been effective in controlling mosquito populations, their efficacy in managing *Culicoides* populations remains underexplored. Moreover, the timing of adulticide applications is critical to their success, as *Culicoides* species exhibit distinct temporal activity patterns that influence their abundance and behavior.

To improve vector control strategies, this study aims to evaluate the efficacy of two ready-to-use insecticides—Fyfanon ULV (EPA #279-3539, active ingredient: malathion) and Evergreen 5-25 (EPA #1021-1803, active ingredients: pyrethrins and piperonyl butoxide)—in controlling *Culicoides* populations through a Field Cage Test (Stark et al. 2017). In parallel, Rotator traps (John Hock) will be utilized to collect *Culicoides* specimens and analyze their temporal activity patterns, which will inform the optimal timing for adulticide applications. By understanding the relationship between *Culicoides* behavior and adulticide application timing, this study aims to provide evidence-based recommendations for local vector control professionals.

Objectives

1. Evaluate the efficacy of two EPA-registered adulticides
2. Assess the temporal patterns of *Culicoides* spp.
3. Provide evidence-based control strategies
4. Optimize vector management practices

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5. Contribute to the broader understanding of *Culicoides* control techniques

Hypotheses

The effectiveness of adulticides in controlling *Culicoides* populations will depend on understanding their temporal patterns. Fyfanon ULV (malathion) and Evergreen 5-25 (pyrethrins and piperonyl butoxide) will demonstrate different levels of efficacy in controlling adult *Culicoides*, with optimal control achieved when the insecticides are applied in alignment with peak activity periods of *Culicoides* spp.

Contribution to AMCA's *Culicoides* Management Guidance

Through the analysis of field cage test data, Fort Bend County Environmental Health (FBC EH) will be looking to provide valuable information to the AMCA *Culicoides* Management Guidance. FBC EH will focus on two interrelated areas. First, we aim to provide more detailed temporal activity patterns of *Culicoides*, which can be used to optimize control timing. Second, we will evaluate the efficacy of two ready-to-use adulticides—Fyfanon ULV (malathion) and Evergreen 5-25 (pyrethrins and piperonyl butoxide) to contribute to the understanding of their effectiveness in managing *Culicoides* populations.

Materials and Methods, including figures and tables if needed

Adult *Culicoides* Collection

Culicoides specimens for this experiment will be captured as adults using CDC mini light traps (Model 1012) or a Collection Bottle Rotator (Model 1512) both utilizing fine mesh collection nets. CDC light mini traps will be placed in existing as well as newly defined trapping locations around Fort Bend County to sample for the *Culicoides* populations. Once population dynamics have been established, Rotator traps will be deployed at selected sites to assess the temporal distribution of *Culicoides* activity. Large quantities of live specimens will be collected based on these trapping efforts, allowing for same-day field cage testing. Rotator traps will be set up in both urban and exurban areas to ensure a broad understanding of Culicoides distribution across different environments. The traps will be checked every 24 hours to ensure timely collection of specimens, with weekly sampling conducted throughout the study to capture the temporal patterns of *Culicoides* activity.

Field Cage Testing

The effectiveness of both chemicals will be assessed with a field cage test with a design as described in Stark et, al 2017. Specimens will be collected the day of each test so that the captured adults are in suitable condition. The Field Cage test will be conducted at a county park

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(Fig 1), where weather conditions will be monitored using a Hobo RX2105-900 weather station. Key environmental factors, such as temperature, wind speed, and humidity, will be continuously recorded during the application process to ensure optimal spraying conditions. Additionally, temperature inversion will be checked before the application begins. The application of each pesticide will take place with a ultra-low volume (ULV) sprayer mounted in the bed of a Utility Terrain Vehicle, to allow a more targeted application to cryptic and hard to reach areas. A DC-IV Droplet Sampler will be used to ensure that appropriate application measures are taking place as chemicals will be switched out between tests using the same spray unit.

Evaluation

Following pesticide application, the *Culicoides* specimens will be left in the field cages for 5 minutes post-application before being collected. The cages will then be returned to a controlled environment to ensure accurate observation of mortality and reduce environmental stress factors, where they will be suspended and provided with 10% sucrose solution ad libitum. Mortality rates will be recorded at 1, 12 and 24-hours (12 hours for permethrin, 24 hours for malathion) post-application. Data from each field test will be recorded and analyzed using appropriate statistical methods. Mortality rates will be calculated for each treatment group, with the natural mortality observed in the control cages used to correct the results. Simple regression analysis will be employed to evaluate the influence of variables such as insecticide type, test date, wind speed, and wind direction on mortality outcomes. Additionally, more advanced multivariate statistical techniques will be utilized to assess the interactions among these variables and identify the key factors that significantly affect the efficacy of the insecticides.

Milestones and Timeline

March 6, 2025

Receive Initial Quotes from all Vendors for Supplies, Equipment and Contract Personnel

March 7, 2025

Submit Proposal to Fort Bend County Commissioners Court for Approval

March 21, 2025

Submit 4 Page Proposal to AMCA

April 30, 2025

Have established surveillance locations identified, and equipment purchased, pending the release of funds

May 15,2025

Field Cage test First Attempt

DRAFT

May 30, 2025

Field Cage Test Second Attempt

June 15,2025

Field Cage Test Third Attempt

June 25, 2025

Complete Data Analysis of Project

June 30, 2025

Submit Findings and Documentation to AMCA

Anticipated Challenges and Solutions

During these experiments several challenges are anticipated. These include the identification of quality trapping sites with existing *Culicoides* populations, collection of ample live adult specimens for field cage test and testing them in a timely manner. FBC EH's entomologists plan to model the experimental design largely based on work previously done in other areas of vector control to limit the number of unique challenges to those directly concerning *Culicoides* insects.

While the focus of this experiment is aimed at control, some efforts must be made in the successful surveillance of *Culicoides* so that specimens can be caught in large enough numbers to be used in field cage trials. Similar work on mosquitoes typically involves the collection of mosquito eggs and rearing them to adult for the purpose of testing, however larval rearing is not seen to be feasible at this time. The limitation of facility, time constraints and the difficulty of rearing a viable number of *Culicoides* in captivity necessitate the collection of live adult specimens.

We intend to utilize use our existing mosquito trapping events in conjunction with additional sites to aid in quicker identification of quality trapping locations across Fort Bend County. The usage of existing trapping events gives a larger subsample of potential *Culicoides* population locations without an exponential increase in required manpower. Additional work will be done to augment target areas with suitable habitat.

In addition to the aforementioned operational constraints, the current availability of personnel is an expected challenge. To combat this limitation, our proposed budget allows for the contract employment of a subject matter expert. The employment of this contractor will give relief and strategic insight to trapping, site surveys, field cage testing and morphological identification.

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