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Research Article

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A push–pull strategy to suppress stable fly (Diptera: Muscidae) attacks on pasture cattle via a coconut oil fatty acid repellent formulation and traps with *m*-cresol lures

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Abstract

BACKGROUND: Stable flies [*Stomoxys calcitrans* (L.)] are economically important pests of cattle and other livestock. As an alternative to conventional insecticides, we tested a push–pull management strategy using a coconut oil fatty acid repellent formulation and an attractant-added stable fly trap.

RESULTS: In our field trials we found that weekly applications of a push–pull strategy can reduce stable fly populations on cattle as well as a standard insecticide (permethrin). We also found that the efficacy periods of the push–pull and permethrin treatments following on-animal application were equivalent. Traps with an attractant lure used as the pull component of the push–pull strategy captured sufficient numbers of stable flies to reduce on-animal numbers by an estimated 17–21%.

CONCLUSIONS: This is the first proof-of-concept field trial demonstrating the effectiveness of a push–pull strategy using a coconut oil fatty acid-based repellent formulation and traps with an attractant lure to manage stable flies on pasture cattle. Also notable is that the push–pull strategy had an efficacy period equivalent to that of a standard, conventional insecticide under field conditions. © 2023 The Authors. *Pest Management Science* published by John Wiley & Sons Ltd on behalf of Society of Chemical Industry.

Supporting information may be found in the online version of this article.

Keywords: permethrin; integrated pest management; stable fly traps; biopesticide

1 INTRODUCTION

Stable flies [*Stomoxys calcitrans* (L.)] are important pests of pastured cattle[1,2] and stress induced by their biting reduces productivity and causes economic loss.[3] The economic impact of stable flies in the United States is estimated to be US$2.66 billion per year.[4]

For cattle in pasture systems, insecticide use is the most convenient, effective and viable option for stable fly control to date.[5] Although insecticides with different modes of action are registered for stable flies in the United States (https://www.veterinaryentomology.org/vetpestx), on-animal application products for stable flies are mostly pyrethroids categorized as group 3A sodium channel modulators by the Insecticide Resistance Action Committee[6] (https://irac-online.org/). The over-dependence on group 3A insecticides to manage blood-feeding stable flies on cattle increases the risk of pyrethroid resistance development which has been reported from various locations in the United States[7–9] and Brazil.[10] Additionally, pyrethroid-associated kdr and kdr-his alleles have been detected in stable fly populations globally.[11,12]

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Biorational compounds with repellent activity are potential alternatives to pyrethroids to prevent stable fly feeding on pasture cattle.[13–15] Repellent plant essential oils derived from catnip and geranium and fatty acid mixtures have demonstrated activity against biting flies.[13,16,17] A trial with catnip oil in a soybean and paraffin wax solution showed promising spatial repellency against stable flies.[18] Mullens *et al.*[13] observed strong antifeedant and repellent behaviors from both stable and horn flies exposed to a mixture of 8, 9 and 10 carbon saturated fatty acids in bioassays. In a field study, Zhu *et al.*[19] found that on-animal application of an aqueous starch-based formulation containing coconut oil-derived fatty acids (SFCA) was effective for 96 h in the field. In laboratory trials, a mixture of 8, 10 and 12 carbon coconut fatty acids exhibited strong antifeedant properties and their corresponding methyl esters showed toxicity against stable flies.[17] Another study reported that direct application of fatty acid sprays are lethal to horn flies.[14]

The behavioral effects of fatty acid biopesticides on insects have been well-documented, yet the mode-of-action (MoA) of their toxicity to insects remains unclear. An early study[20] hypothesized that fatty acids bind to external and internal insect cuticle and cause lysis of hemolymph and body cells. A trial with German cockroaches (*Blattella germanica* L.) showed that fatty acids exhibit both topical and fumigant toxicity depending on the carbon level.[21] Ware[22] suggested that the MoA of saturated fatty acids is similar to that of insecticidal soaps (alkali salts of fatty acids) that penetrate through the insect cuticle and disrupt cell membranes. Cytotoxicity and cell membrane effects of fatty acids have been demonstrated in microbial and human cell systems.[23,24] A study by Ren *et al.*[25] indicated that fatty acid biopesticides induce apoptosis in AW1 neuronal cell lines of *Helicoverpa zea*. Although further study is needed to fully understand the MoA of fatty acid biopesticides on insects, they offer valuable options for controlling stable flies and other insect pests and for possible integration within insecticide resistance management plans.

Besides direct cattle treatments, various attract-and-kill technologies have been used for stable fly management.[2,26] Traps using visually attractive materials such as Alsynite[27,28] or Coroplast[29] have been used to lure stable flies to panels, cards or cylinders coated with an insect adhesive.[30] More recently, Knight Stick traps (BugJammer, Inc., Stockton, NJ, USA) were found to capture three- to five-fold more flies than Alsynite traps, with the greater attraction attributed to the sticky wraps.[31]

Stable fly trap innovations have included the addition of host-associated attractive odorants to increase capture rates. For example, Alsynite traps modified to release CO_2 caught ≤25-fold more stable flies than traps without CO_2.[32] Baiting blue cloth panel traps with 1-octen-3-ol sachets increased trap captures of stable flies more than two-fold.[33] Phenol and derivatives *m*-cresol and *p*-cresol are associated with animal odors and dung emissions, and are known stable fly attractants.[34–36] Cylindrical alsynite panels, modified with the inclusion of lures containing mixtures of phenol with *m*-cresol or *p*-cresol increased stable fly capture rates more than two-fold compared to traps without lures.[37] In a field trial of stable fly attractants, more stable flies were captured on traps with *m*-cresol than the other odorants.[25] An adhesive tape material impregnated with *m*-cresol was able to reduce stable fly numbers and cattle stress.[38]

The use of attractants and repellents have been demonstrated as promising insect management tools and can be used together in an innovative way. Push–pull strategies were first used to manage insects in cropping systems and are designed to modify insect behaviors, resulting in a lowered pest abundance on the crop.[39,40] The first application of a push–pull strategy to an animal system used novel repellents in an applicator hung on the neck of cattle to successfully reduce tsetse fly transmission of trypanosomes and thereby reduce cattle trypanosomiasis.[41] Extension of this strategy to other blood-feeding flies was identified as a research priority by the United States Department of Agriculture research committee, S1076 Fly Management in Animal Agriculture Systems and Impacts on Animal Health and Food Safety (USDA NIMSS; https://www.nimss.org/projects/18522).

The goal of this research was to determine the efficacy of a push–pull strategy on stable fly infestation levels on pasture cattle. Our objectives were two-fold; (i) to prevent stable flies from visiting cattle treated with a repellent, and (ii) to capture the stable flies on traps with both attractive visual and olfactory cues. The expected outcome is a novel stable fly management program using a design with two management tactics instead of one and that will lessen reliance on conventional insecticides for stable fly management.

2 MATERIALS AND METHODS

2.1 Products and materials used

Permethrin (ProZap Insectrin® ×10%), an insecticide commonly used as an on-animal application to manage stable flies, was compared against a repellent. Both treatments were applied to cattle using garden sprayers (1.9 L, Home & Garden Sprayer, model no. 7215015; ACE Hardware, Oak Brook, IL, USA).

A fatty acid repellent formulation was prepared from natural coconut oil fatty acids (ACME-HARDESTY, Blue Bell, PA, USA) containing an average free fatty acid composition of caprylic acid (C8:0; 6.65 ± 0.71%), capric acid (C10:0; 7.77 ± 0.87%), lauric acid (C12:0; 51.25 ± 1.54%), myristic acid (C14:0; 16.91 ± 0.53%), palmitic acid (C16:0; 9.37 ± 0.89%), stearic acid (C18:0; 1.26 ± 0.11%), oleic acid (C18:1; 6.33 ± 0.40%) and linoleic acid (C18:1; 0.50 ± 0.36%). Other components of the repellent formulation were Genu pectin DD-slow set Z (CP Kelco, Atlanta, GA, USA) and waxy cornstarch (Waxy No. 1, A.E. Staley Mfg. Co., Decatur, IL, USA) with a moisture content of 9.3%.

Stable fly trap components consisted of common 10.8-cm-diameter white plumbing pipe (available from building supply outlets) and Knight Stick® sticky wraps (http://www.bugjammer.com). Red rubber septa (Chemglass, CG-3022-95, 8 mm; https://chemglass.com/septum-stoppers-sleeve-type-septa) treated with *m*-cresol (Sigma-Aldrich, C85727-100G, 99%; https://www.sigmaaldrich.com/US/en/product/sigald/c85727) were used as slow-release lures.

2.2 Repellent preparation

The SCFA repellent was prepared in a similar way to that as described by Zhu *et al.*[19] A detailed description of the preparation is available as Appendix S1.

2.3 2019 and 2020 field trials

Field trials were done in a pasture complex with cattle sorting and handling facilities, and four irrigated (0.04 km^2) and three nonirrigated pastures (0.07 km^2) of similar carrying capacity of 5–6 animals, located at the West Central Research and Extension Center, North Platte, Nebraska. The upland pastures consisted of native grasses, primarily big blue stem (*Andropogon geradii*), little blue stem (*Schrizachyriun scoparium*) and western wheat grass (*Pascopyrum smithii*). The irrigated pastures were seeded with cache meadow brome (*Bromus biebesteinii*), smooth brome (*Bromus inermis*), pennlate orchardgrass (*Dactylis glomerata*, 'Pennlate') and garrison creeping foxtail (*Alopecurus arundinaceus*). Each irrigated pasture had a water tank. The nonirrigated pastures shared a central watering tank with fencing to prevent herds in different pastures from co-mingling.

In 2019, the trial period was 5 weeks starting 10 July and in 2020, the trial ran for 4 weeks starting 21 June. Each week, six of the seven available pastures were used, and one was rested from grazing.

2.3.1 *Test animals and experimental design*

For the 2-year trial, testing was done using a different set of yearling cattle each year (steers and heifers, 5/8 Red Angus × 3/8 Simmental) that were maintained on pasture with *ad libitum* water. Cattle were randomized into herds of five animals that remained together throughout the trial period and treatments were randomly assigned to herds. The grouping of cattle to herds and then treatments to herds was termed 'treatment herd' and was done to avoid effects from possible residue carryover if herds were to receive different treatments each week.

2.3.2 *Treatments*

(1) Push–pull. A two-component treatment:
   (a) SCFA (push component). Applied to individual cattle at a volume of 1000 mL of a 2.5% SCFA – distilled water solution (v/v).
   (b) Stable fly traps (pull component) integrated in pastures with push–pull treatment herds. Trap design and placement are described in Section 2.3.4.
(2) Permethrin. Applied to individual cattle at a volume of 500 mL of a 0.05% v/v distilled water solution.
(3) Untreated Control. 500 mL water applied per animal.

Treatments were replicated twice. See Fig. 1 for a diagrammatic overview of the treatment components and objectives.

2.3.3 *Application procedures and stable fly assessments*

On Day (D)1 of each trial week, herds were moved into separate holding pens before passing through a cattle chute (Fig. 2) where spray applications were made to the legs, belly and flanks of each animal. Separate sprayers were used for each material applied and were cleaned with a 1% concentration of ammonia (38 mL per 3.78 L water), and then rinsed with water following use. Applications were made sequentially starting with untreated control herds, then push–pull herds, and lastly permethrin herds. After treatments were applied, herds were released into randomly assigned pastures.

Stable fly assessments were conducted by visually counting the total number of stable flies on all legs, sides and belly of each animal and the resulting total number of stable flies per animal was termed fly load. Fly counts were made between 13:00 h and 16:00 h on D1 to D4 in 2019 and D1 to D3 in 2020, post-treatment each week of the trial period.

2.3.4 *Stable fly traps*

The stable fly traps consisted of a white, 30-cm-long plastic pipe base with a Knight Stick® sticky wrap secured around the pipe with binder clips. An olfactory lure was placed in a screened holder and suspended in the top center of the pipe ≈3 cm below the top edge (Fig. 3). The red rubber septum lure was prepared by dissolving 1 mg *m*-cresol in 100 mL hexane and applying it to the septum. The septum was placed in a fume hood to let the solvent evaporate. After drying, the lure was wrapped inside aluminum foil and stored in a freezer until use. The pipe was anchored to the ground using steel shepherd hooks. To limit cattle damage to the traps, they were enclosed in a steel wire mesh (0.76 m high, 1.98 m long, 0.58 m diameter). We used this trap design because of its demonstrated effectiveness[31] and stability in windy conditions. Four traps were placed in push–pull treatment pastures in locations which the cattle used as loafing areas. Traps were reset with new sticky wraps and lures weekly.

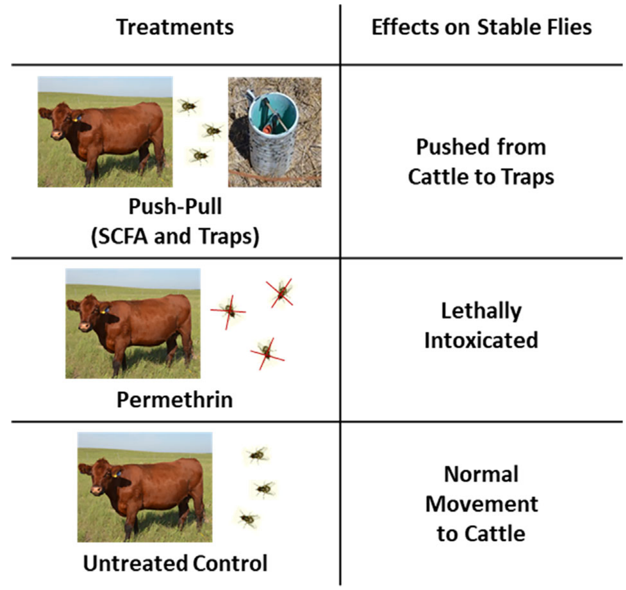

Figure 1. Weekly treatment application being made individually to cattle in a treatment herd. After all cattle were treated, the treatment herd was released into their randomly assigned pasture for the trial week.

Within trial weeks, daily counts of stable flies were made by removing captured flies from the sticky wraps with forceps and transporting them to the laboratory for later counting. Sticky wraps were replaced at the start of each week.

In order to estimate the contributions of traps to reducing on-animal fly loads, we applied the LaBrecque *et al.*[42] ratio of 1:55 of on-animal to environmental stable fly populations to observed fly loads (per animal per week) on cattle in push–pull treatment herds.

2.4 Statistical analyses

Herd was the experimental unit, so the average number of flies calculated for each day by herd was the response variable analyzed. The data analysis for this paper was generated using the GLIMMIX procedure in SAS/STAT software v9.4 of the SAS System for Windows. For all analyses, a Kenward–Rogers degree of freedom adjustment was used. Least squares means (LSM) were determined for all fixed effects in the final model. An alpha level of 0.05 was used for determining significance.

The trial design was a repeated-measures, random complete block, where each herd was randomly assigned to one of three treatments (untreated control, permethrin, push–pull). There were two sets (replicates) of repeated measures. The first was weeks and the second was days nested within week. The initial statistical model included the fixed effects of year, treatment, week, day, and all two-, three and four-way interaction terms, and the random year by herd nested within treatment, year by

Figure 2. Spray Application to cattle in sorting chute.

Figure 3. Views of a stable fly trap consisting of a white plastic pipe surrounded with a clear sticky wrap and with an olfactory lure suspended on the inside. Protective fencing surrounding the trap to minimize cattle damage also is shown.

period by herd nested within treatment, and year by week by herd nested within treatment. The error term for year, treatment and the year by treatment interaction was the random year by herd nested within treatment random effect. The error term for week was the random year by week by herd nested within treatment using an AR(1) covariance structure to account for repeated weeks. The error term for day and all interaction effects including day was the year by day by period by week by herd nested within treatment term using an AR(1) covariance structure to account for repeated days nested within week. Year was involved in several significant interactions. Year 2019 was a high moisture year, which resulted in high fly loads. By contrast, year 2020 was dry with reduced fly loads compared to the previous year.

Because of these differences, years were analyzed separately. The final model for each year included treatment, week, day, and all two- and three-way interaction terms and the random herd nested within treatment and period by herd nested within treatment effects. Treatment was tested over the random herd nested within treatment random effect and period and treatment by period were tested over the random period by herd nested within treatment random effect. The rest of the effects were tested over the residual error.

3 RESULTS

We had anticipated beginning the study in 2018 but because of drought stress to pastures we were limited to a comparison of the effects of the repellent SCFA and permethrin on fly loads. The results (Table S1) suggested that SCFA could limit on-animal stable fly loads in pasture conditions and encouraged further testing.

3.1 2019 results

The treatment means and SEM were 42.7 (0.747), 28.7 (0.788) and 29.9 (0.706) for untreated control, permethrin and push–pull herds. Stable fly infestation levels remained relatively constant from D1 to D4 after treatment in the untreated control herd, whereas they increased daily after D1 in herds treated with SCFA and permethrin [$P < 0.01$; Table 1, Fig. 4(A)].

Stable fly numbers began declining after study Week (W)2 and by W5 the fly loads of untreated control herds and treatment herds were similar. However, for W1 to W4, fly loads on treatment herds were lower than those for the untreated control herd [Table 2, Fig. 5(A)].

3.2 2020 results

The treatment means and SEM were 12.1 (0.405), 8.5 (0.452) and 9.5 (0.407) for untreated control, permethrin and push–pull herds. As in 2019, we again saw stable fly load response over days being low in the push–pull and permethrin treatments early in weeks, and then converging later in the week to levels equivalent to the untreated check value [$P = 0.172$; Table 1 and Fig. 4(B)]. However, fly loads were higher on untreated control herds than on treatment herds and permethrin and push–pull fly loads were similar over all days [Table 1 and Fig. 4(B)].

Except for W1, where the push–pull treatment herds had a higher fly load than the permethrin treatment herds, weekly fly load responses of push–pull and permethrin herds were similar during the trial period [$P = 0.1742$; Table 2 and Fig. 5(B)].

3.3 Pull effect

The traps used in the push–pull treatment herds captured large numbers of stable flies in both trial years. In 2019, four traps per push–pull pasture caught a total of 13 638 stable flies and in 2020, when stable fly populations were lower on untreated control herd cattle (Table 3), a total of 4286 flies were captured.

Using the LaBrecque ratio,[42] we estimated that the stable fly traps reduced fly loads in push–pull herds by 17% and 21% in 2019 and 2020, respectively. Without traps, we estimate that observed fly loads would have increased in 2019 from the observed 29.9 to 34.9 and in 2020 from 9.5 to 11.4 (Table 3).

4 DISCUSSION

We conducted 5- and 4-week trials in 2019 and 2020 with three treatments using a push–pull design. Fly loads on untreated control cattle were on average five-fold higher in 2019 than in 2020, and therefore the 2 years were analyzed separately. However, despite the difference in average fly load between years, the 2019 and 2020 results were consistent, and the push–pull and permethrin treatments significantly reduced fly loads compared to the untreated control values. We also found that the push–pull and permethrin treatments did not differ significantly from each other in both years.

Campbell *et al.*[43] investigated the relationship between fly load and average daily weight gain in pastured yearling steers in Nebraska and found that 2.79 stable flies per leg were sufficient to reduce average daily weight gain of grazing cattle by ≈0.2 kg per day. Thus, treatments reducing fly loads by ≈3 or more per leg provides measurable weight gain protection and economic benefit to producers. Furthermore, they found that when cattle were transferred from pastures to feed lots with high nutrition

Table 1. Simple effect comparisons of fly load differences

Comparison	Day	Fly load mean Difference* (±SEM)	*P*-value
2019			
Untreated control *versus* push–pull	1	19.0 (2.88)	<0.0001
	2	12.6 (2.88)	0.0002
	3	9.1 (2.88)	0.0076
	4	8.6 (2.88)	<0.0119
Untreated control *versus* permethrin	1	18.7 (2.88)	<0.0001
	2	13.6 (2.88)	<0.0001
	3	10.2 (2.88)	0.0027
	4	8.9 (2.88)	<0.0096
Permethrin *versus* push–pull	1	0.3 (2.88)	0.9943
	2	−1.0 (2.88)	0.7325
	3	−1.1 (2.88)	0.9281
	4	−0.2 (2.88)	0.9962
2020			
Untreated control *versus* push–pull	1	6.7 (0.83)	<0.0001
	2	2.4 (0.83)	0.0164
	3	1.3 (0.83)	0.2586
Untreated control *versus* permethrin	1	8.5 (0.83)	<0.0001
	2	4.1 (0.83)	<0.0001
	3	2.1 (0.83)	0.0367
Permethrin *versus* push–pull	1	−1.8 (0.83)	0.9170
	2	−1.7 (0.83)	0.1090
	3	−0.8 (0.83)	0.6014

Note: Treatment by day least squares means (LSM) by period adjustment for multiple comparisons: Tukey's honestly significant difference.
Note: push–pull is a two-component treatment, cattle were treated weekly with SCFA and stable fly traps were integrated into the pasture.
*Difference in LSM of the indicated comparison.

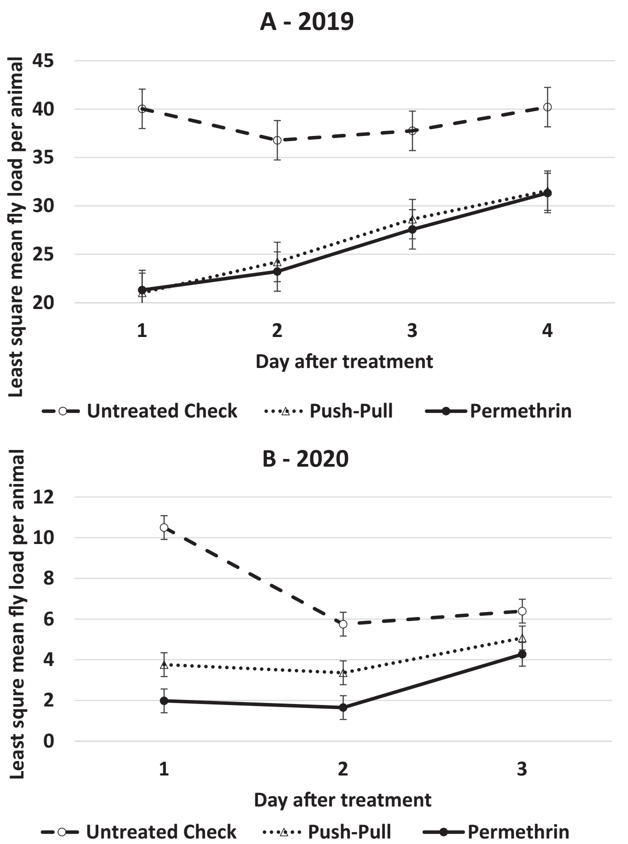

Figure 4. Least squares mean (LSM, +SE) fly load per treatment by day in 2019 and 2020.

Table 2. Simple effect comparisons of fly load differences

Comparison	Week	Fly load mean Difference* (± SEM)	*P*-value
2019			
Untreated control *versus* push–pull 1	1	13.79 (3.27)	0.0031
	2	13.94 (3.27)	0.0029
	3	16.57 (3.27)	0.0007
	4	12.87 (3.27)	0.0051
	5	4.54 (3.27)	0.3763
Untreated control *versus* permethrin	1	17.70 (3.27)	0.0004
	2	12.52 (3.27)	0.0024
	3	16.07 (3.27)	0.0009
	4	10.48 (3.27)	0.0190
	5	7.44 (3.27)	0.0979
Permethrin *versus* push–pull	1	−3.91 (3.27)	0.476
	2	1.42 (3.27)	0.9012
	3	0.50 (3.27)	0.987
	4	2.39 (3.27)	0.749
	5	−2.90 (3.27)	0.657
2020			
Untreated control *versus* push–pull 1	1	1.28 (0.924)	0.381
	2	3.88 (0.924)	0.003
	3	3.64 (0.924)	0.005
	4	3.66 (0.924)	0.004
Untreated control *versus* permethrin	1	3.89 (0.924)	0.003
	2	5.58 (0.924)	0.0001
	3	4.75 (0.924)	0.0006
	4	3.99 (0.924)	0.002
Permethrin *versus* push–pull	1	−2.61 (0.924)	0.037
	2	−1.70 (0.924)	0.198
	3	−1.11 (0.924)	0.473
	4	−0.325 (0.924)	0.934

Note: Treatment by week least squares means (LSM) by period adjustment for multiple comparisons: Tukey's honestly significant difference.
Note: push–pull is a two-component treatment, cattle were treated weekly with SCFA and stable fly traps were integrated into the pasture.
*Difference in LSM of the indicated comparison.

diets for finishing, compensatory weight gains did not occur. Their results emphasize the importance of protecting pasture cattle from stable fly stress.

Average fly loads of push–pull and permethrin herds in 2019 were 12.9 lower than those on cattle in untreated control herds, well above the 2.79 threshold of Campbell *et al.*[43] for detecting measurable reductions in cattle daily average weight gain. However, in 2020, although the push–pull and permethrin treatments again reduced fly load relative to that of untreated control herds, the average difference was small, at 1.4, and probably would not have resulted in a measurable reduction in daily weight gain.

The SCFA formulation tested probably is the first natural product to demonstrate a 4-day efficacy period under field conditions against a biting fly. We further demonstrated that SCFA was as effective as permethrin in reducing fly loads and presumably in maintaining normal weight gain. However, biopesticides, especially repellents, often are considered ineffective because of a perceived short residual efficacy period. For example, Rehman *et al.*[44] found that frequent reapplications of repellent plant-based materials were needed to retain effective mosquito control. In our trials, we found that the medium chain fatty acids present in the SCFA formulation had an effective longevity period equivalent to permethrin. This is likely to be due to a combination of the encapsulation of the fatty acids by the starch matrix and the ability of the starch to adhere to the cattle's hair. In addition, SCFA has benefits requested by producers such as an expected low cost and environmental safety: the fatty acid active ingredients and starch components in SCFA are listed as 'generally regarded as safe' (GRAS) by the US Environmental Protection Agency.

Producers also are concerned about possible insecticide resistant stable fly populations. Because the MoA of fatty acid insecticides is likely to be different from that of the commonly used insecticides[20,22,25] and because SCFA was shown to be effective within a push–pull strategy (and perhaps as a standalone product) SCFA could be used in rotation with conventional insecticides in an insecticide resistance management plan.

As the pull component of the push–pull treatment, we found that cylindrical sticky traps with *m*-cresol lures, similar to a sticky trap found effective by Hogsette and Kline,[31] captured numerous stable flies. By applying the LaBrecque ratio[42] to our data, we estimated that trap effect reductions of fly loads for push–pull herds were 17% in 2019 and 21% in 2020. Our estimates are lower than the 40% trapping rate modeled as needed for single tactic insect control.[45] However, when used in a combination strategy such as push–pull, fly load reductions of 40% are not needed. Our results also suggest that increases in trap numbers may be needed in

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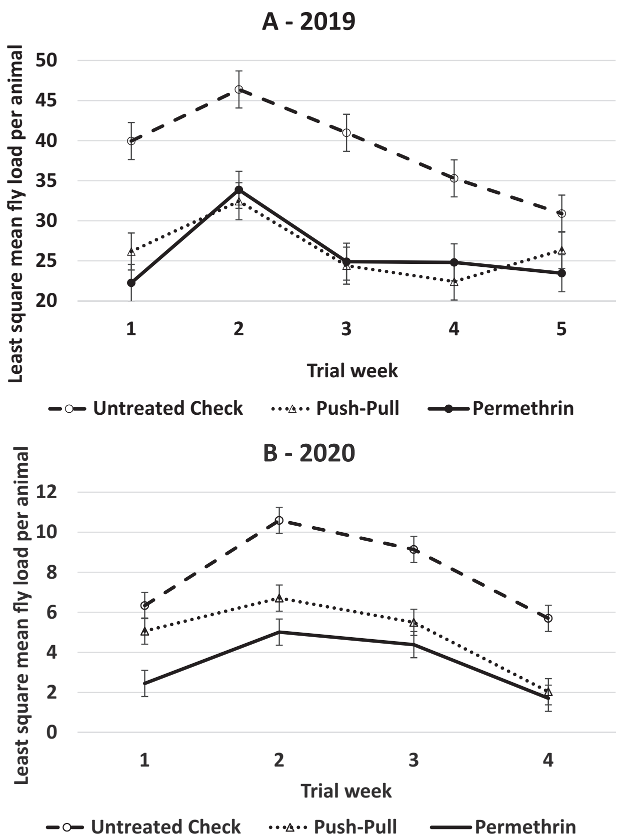

Figure 5. Least squares mean (LSM, +SE) fly load per treatment by week in 2019 and 2020.

Table 3. Application of the LaBrecque ratio* to stable fly trap captures in 2109 and 2020 to estimate potential fly loads if traps had been absent

2019	
(A) Total stable flies captured on traps	13 638
(B) Stable flies captured by traps per week per animal	272.8
(C) Reduction in on-animal flies by trapping (B × LaB1*)	4.96
(D) Observed weekly mean fly load per animal	29.9
(E) Potential fly load (C + D) and % increase (C/D) without traps	34.9 & 17%
2020	
(A) Total stable flies captured on traps	4286
(B) Stable flies captured by traps per week per animal	107.2
(C) Reduction in on-animal flies by trapping (B × LaB*)	1.9
(D) Observed weekly mean fly load per animal	9.458
(E) Potential fly load (C + D) and % increase (C/D) without traps	11.4 & 21%

Note: The trial period in 2019 was 5 weeks and 4 weeks in 2020. In both years there were ten cattle total in push–pull herds (two replicates of five cattle per herd). Each week, four traps were placed in each pasture with a push–pull treatment herd.
*LaB = Labrecque *et al.*[42] ratio of 1 on-animal to 55 environmental stable flies.

years when stable fly populations are high as in 2019 relative to years such as 2020 when populations are lower. In summary, sticky traps with *m*-cresol as an attractant lure complemented the use of SCFA in a stable fly push-management strategy. However, improvements of trap deployment and design (numbers, placement and attractiveness) remain as goals.

Our trial demonstrated that a push–pull strategy can be as effective as a conventional insecticide application program in managing stable flies on pasture cattle. However, practical considerations remain to improve acceptability to producers. Weekly gathering of cattle for retreatment with biopesticides (or conventional insecticides) can stress cattle, increase producer management costs, and thereby lessen the benefit of stable fly population reduction[43] in pasture livestock production systems. Mist blower or automated spray systems that cattle pass through as part of their routine behavior could be, after cattle acclimation, less stressful alternatives for some production systems. Increases in trap capture rate would enhance the impact of the pull component of the push–pull tactic.

5 CONCLUSIONS

This paper is the first case study reporting the use of a push–pull integrated pest management strategy to manage stable flies and only the second application of a push–pull strategy to a fly in the family Muscidae.[46] As such, it represents a low-impact alternative to conventional insecticides for management of stable flies on pastured cattle. However, more research is needed to refine the system and reduce the need for frequent retreatment.

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DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available from the corresponding author upon reasonable request.

SUPPORTING INFORMATION

Supporting information may be found in the online version of this article.

REFERENCES

1 Hall RD, Thomas GD and Morgan CE, Stable fly, *Stomoxys calcitrans* (L.), breeding in large round hay bales: initial associations (Diptera: Muscidae). *J Kansas Entomol Soc* **55**:617–620 (1982).
2 Rochon K, Hogsette JA, Kaufman PE, Olafson PU, Swiger SL and Taylor DB, Stable fly (Diptera: Muscidae)—biology, management, and research needs. *J Integr Pest Manage* **12**:38 (2021).
3 Mullens BA, Lii KS, Mao Y, Meyer JA, Peterson NG and Szijj CE, Behavioural responses of dairy cattle to the stable fly, *Stomoxys calcitrans*, in an open field environment. *Med Vet Entomol* **20**:122–137 (2006).
4 Taylor DB, Stable fly (*Stomoxys calcitrans*, Muscidae), in *Encyclopedia of Infection and Immunity*, Vol. **2**, ed. by Rezaei N. Amsterdam, Elsevier, pp. 903–913 (2022).
5 Foil LD and Hogsette JA, Biology and control of tabanids, stable flies and horn flies. *Rev Sci Tech* **13**:1125–1158 (1994).
6 Sparks TC and Nauen R, IRAC: mode of action classification and insecticide resistance management. *Pestic Biochem Physiol* **121**:122–128 (2015).

7 Cilek JE and Greene GL, Stable fly (Diptera: Muscidae) insecticide resistance in Kansas cattle feedlots. *J Econ Entomol* **87**:275–279 (1994).
8 Pitzer JB, Kaufman PE and Tenbroeck SH, Assessing permethrin resistance in the stable fly (Diptera: Muscidae) in Florida by using laboratory selections and field evaluations. *J Econ Entomol* **103**:2258–2263 (2010).
9 Maron PCRG, Thomas GD, Siegfried BD and Campbell JB, Susceptibility of stable flies (Diptera: Muscidae) from southeastern Nebraska beef cattle feedlots to selected insecticides and comparison of 3 bioassay techniques. *J Econ Entomol* **90**:293–298 (1997).
10 Barros ATM, Rodrigues VD, Cançado PHD and Domingues LN, Resistance of the stable fly, *Stomoxys calcitrans* (Diptera: Muscidae), to cypermethrin in outbreak areas in midwestern Brazil. *Rev Bras Parasitol Vet* **12**:1823–1828 (2019).
11 Olafson PU, Pitzer JB and Kaufman PE, Identification of a mutation associated with permethrin resistance in the Para-type sodium channel of the stable fly (Diptera: Muscidae). *J Econ Entomol* **104**: 250–257 (2011).
12 Olafson PU, Kaufman PE, Duvallet G, Solórzano J-A, Taylor DB and Fryxell RT, Frequency of kdr and kdr-his alleles in stable fly (Diptera: Muscidae) populations from the United States, Costa Rica, France, and Thailand. *J Med Entomol* **56**:1145–1149 (2019).
13 Mullens BA, Reifenrath WG and Butler SM, Laboratory trials of fatty acids as repellents or antifeedants against houseflies, horn flies and stable flies (Diptera: Muscidae). *Pest Manag Sci* **65**:1360–1366 (2009).
14 Mullens BA, Watson DW, Gerry AC, Sandelin BA, Soto D, Rawls D *et al.*, Field trials of fatty acids and geraniol applied to cattle for suppression of horn flies, *Haematobia irritans* (Diptera: Muscidae), with observations on fly defensive behaviors. *Vet Parasitol* **245**:14–28 (2017).
15 Mullens BA, Soto D, Gerry AC, Fowler FE and Diniz AN, Effects of fatty acid and geraniol repellent-oil mixtures applied to cattle on blood feeding and reproductive parameters in field populations of *Haematobia irritans* (Diptera: Muscidae). *J Med Entomol* **55**:408–416 (2018).
16 Zhu JJ, Brewer GJ, Boxler DJ, Friesen K and Taylor DB, Comparisons of antifeedancy and spatial repellency of three natural product repellents against horn flies, *Haematobia irritans* (Diptera: Muscidae). *Pest Manag Sci* **71**:1553–1560 (2015).
17 Roh GH, Zhou X, Wang Y, Cermak SC, Kenar JA, Lehmann A *et al.*, Spatial repellency, antifeedant activity and toxicity of three medium chain fatty acids and their methyl esters of coconut fatty acid against stable flies. *Pest Manag Sci* **76**:405–414 (2020).
18 Zhu JJ, Dunlap CA, Behle RW, Berkebile DR and Wienhold B, Repellency of a wax-based catnip-oil formulation against stable flies. *J Agric Food Chem* **58**:12320–12326 (2010).
19 Zhu JJ, Cermak SC, Kenar JA, Brewer G, Haynes KF, Boxler D *et al.*, Better than DEET repellent compounds derived from coconut oil. *Sci Rep* **8**: 1–12 (2018).
20 Siegler EH and Popenoe CH, The fatty acids as contact insecticides. *J Econ Entomol* **18**:292–299 (1925).
21 Sims SR, Balusu RR, Ngumbi EN and Appel AG, Topical and vapor toxicity of saturated fatty acids to the German cockroach (Dictyoptera: Blattellidae). *J Econ Entomol* **107**:758–763 (2004).
22 Ware GW, Biorationals-21st century pesticides, in *The Pesticide Book*, fifth edn. Thomson Publications, Fresno, CA, pp. 269–287 (2000).
23 Kabara JJ and Marshall DL, 11 medium-chain fatty acids and esters, in *Antimicrobials in Food*, ed. by Davidson PM, Sofos JN and Branen AL. Marcel Dekker, New York, pp. 327–360 (2005).
24 Martins de Lima T, Cury-Boaventura MF, Giannocco G, Nunes MT and Curi R, Comparative toxicity of fatty acids on a macrophage cell line (J774). *Clin Sci* **13**:307–317 (2006).
25 Ren Y, Shi J, Mu Y, Tao K, Jin H and Hou T, AW1 neuronal cell cytotoxicity: the mode of action of insecticidal fatty acids. *J Agric Food Chem* **67**:12129–12136 (2019).
26 Cook D, A historical review of management options used against the stable fly (Diptera: Muscidae). *Insects* **11**:313 (2020). https://doi.org/10.3390/insects11050313.
27 Williams DF, Sticky traps for sampling populations of *Stomoxys calcitrans*. *J Econ Entomol* **66**:1279–1280 (1973).
28 Broce AB, An improved Alsynite trap for stable flies, *Stomoxys calcitrans* (Diptera: Muscidae). *J Med Entomol* **25**:406–409 (1988).
29 Beresford DV and Sutcliffe JF, Studies on the effectiveness of coroplast sticky traps for sampling stable flies (Diptera: Muscidae), including a comparison to Alsynite. *J Econ Entomol* **99**:1025–1035 (2006).
30 Zhu JJ, Zhang Q, Taylor DB and Friesen KA, Visual and olfactory enhancement of stable fly trapping. *Pest Manag Sci* **72**:1765–1771 (2016).
31 Hogsette JA and Kline DL, The knight stick trap and knight stick sticky wraps: new tools for stable fly (Diptera: Muscidae) management. *J Econ Entomol* **110**:1384–1389 (2017).
32 Cilek JE, Evaluation of various substances to increase adult *Stomoxys calcitrans* (Diptera: Muscidae) collections on Alsynite cylinder traps in North Florida. *J Med Entomol* **36**:605–609 (1999).
33 Holloway MTP and Phelps RJ, The responses of *Stomoxys* spp. (Diptera: Muscidae) to traps and artificial host odours in the field. *Bull Entomol Res* **81**:51–56 (1991).
34 Jeanbourquin P and Guerin PM, Chemostimuli implicated in selection of oviposition substrates by the stable fly *Stomoxys calcitrans*. *Med Vet Entomol* **21**:209–216 (2007).
35 Jeanbourquin P and Guerin PM, Sensory and behavioural responses of the stable fly *Stomoxys calcitrans* to rumen volatiles. *Med Vet Entomol* **21**:217–224 (2007).
36 Tangtrakulwanich K, Chen H, Baxendale F, Brewer G and Zhu JJ, Characterization of olfactory sensilla of *Stomoxys calcitrans* and electrophysiological responses to odorant compounds associated with hosts and oviposition media. *Med Vet Entomol* **25**:327–336 (2011).
37 Tangtrakulwanich K, Albuquerque TA, Brewer GJ, Baxendale FP, Zurek L, Miller DN *et al.*, Behavioural responses of stable flies to cattle manure slurry associated odourants. *Med Vet Entomol* **29**:82–87 (2015).
38 Zhu JJ, Roh G-H, Asamoto Y, Bizati K, Liu J-C, Lehmann A *et al.*, Development and first evaluation of an attractant impregnated adhesive tape against blood-sucking flies. *Insect Sci* **29**:603–612 (2021).
39 Pyke B, Rice M, Sabine B and Zalucki M, The push-pull strategy-behavioural control of *Heliothis*. *Aust Cotton Grower* **9**:7–9 (1987).
40 Cook SM, Khan ZR and Pickett JA, The use of push-pull strategies in integrated pest management. *Annu Rev Entomol* **52**:375–400 (2007).
41 Saini RK, Orindi BO, Mbahin N, Andoke JA, Muasa PN, Mbuvi DM *et al.*, Protecting cows in small holder farms in East Africa from tsetse flies by mimicking the odor profile of a non-host bovid. *PLoS Negl Trop Dis* **11**:e0005977 (2017).
42 LaBrecque GC, Bailey DL, Meifert DW and Weidhaas DE, Density estimates and daily mortality rate evaluations of stable fly (*Stomoxys calcitrans* (Diptera: Muscidae)) populations in field cages. *Can Entomol* **107**:597–600 (1975).
43 Campbell JB, Skoda SR, Berkebile DR, Boxler DJ, Thomas GD, Adams DC *et al.*, Effects of stable flies (Diptera: Muscidae) on weight gains of grazing yearling cattle. *J Econ Entomol* **94**:780–783 (2001).
44 Rehman JU, Ali A and Khan IA, Plant based products: use and development as repellents against mosquitoes: a review. *Fitoterapia* **95**:65–74 (2014).
45 Weidhaas DE and Haile DG, A theoretical model to determine the degree of trapping required for insect population control. *ESA Bull* **24**:18–20 (1978).
46 Olaide OY, Tchouassi DP, Yusuf AA, Pirk CW, Masiga DK, Saini RK *et al.*, Zebra skin odor repels the savannah tsetse fly, *Glossina pallidipes* (Diptera: Glossinidae). *PLoS Negl Trop Dis* **13**:e0007460 (2019).

