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RESEARCH ARTICLE

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Palm wine as a food-based bait for monitoring adult *Ceratitis ditissima* (Munro) (Diptera: Tephritidae) in citrus orchards

John Abraham[1], Carlos Amissah[1], Felix Osei Kuffour[1] and Janice Dwomoh Abraham[2]

[1]*Department of Conservation Biology and Entomology, School of Biological Sciences, College of Agriculture and Natural Sciences, University of Cape Coast, Ghana*
[2]*Department of Biological Sciences Education, Faculty of Science Education, Akenten Appiah-Menka University of Skills Training and Entrepreneurial Development, Mampong-Ashanti, Ghana*

Tephritid fruit flies, including *Ceratitis ditissima*, often invade citrus orchards. These flies cause economic losses to farmers and can prevent farmers from exporting their fruits to foreign markets. To detect the presence of fruit flies in citrus orchards, traps are baited with synthetic lures, which are often expensive for smallholder farmers. Farmers in developing or financially less-endowed countries have to import such synthetic baits, raising the cost of pest monitoring and control. Therefore, we evaluated the potential of palm wine and three other food-based mixtures for trapping *C. ditissima* and the proportion of non-target flies they trap. Transparent deli cup traps were baited with four different food-based baits, namely palm wine, sugarcane spirit-wine mixture, apple cider vinegar and yeast-sugar mixture. The traps were placed within a citrus orchard on fruit-bearing trees. The content of each trap was collected after one week and evaluated. This was repeated for eight consecutive weeks. Traps baited with palm wine captured more *C. ditissima* than those with the other baits. Furthermore, the proportion of non-target insects, *Bactrocera dorsalis* and *Drosophila* spp., in palm wine-baited traps was less than the other baited traps. This study indicates that palm wine, a cheap beverage across Africa, Asia and South America, could be used to monitor the presence of *C. ditissima* in citrus orchards. Smallholder farmers who cannot afford expensive synthetic baits could make use of palm wine to monitor fruit flies in their farms.

CORRESPONDENCE
John Abraham

EMAIL
jabraham@ucc.edu.gh

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INTRODUCTION

Fruit flies belonging to the family Tephritidae are among the most economically important insect pests of horticultural crops (Badii et al. 2015; Ekesi et al. 2016; Yazid et al. 2020). They attack and cause economic damage to a variety of crops including citrus (Lloyd et al. 2010; Vayssieres et al. 2010), mango (Ansari et al. 2012; Gnanvossou et al. 2017; Hanna et al. 2020), guava (Birke et al. 2015), melon (Dhillon et al. 2005; Ansari et al. 2012; Mokam et al. 2018) and papaya (Ansari et al. 2012). About 200 species of known tephritid fruit flies are pests, of which some are polyphagous and may switch host plants depending on their location, season and available host plants (Lloyd et al. 2010; Vayssiéres et al. 2010; Ansari et al. 2012; Kambura et al. 2018). The gravid female fruit fly lays eggs underneath the epicarp of fruits, the eggs hatch and the larvae feed on the pulp, causing fruit damage, while the oviposition wound serves as an entry point for pathogens (Walsh et al. 2011; Thomas et al. 2013; Birke et al. 2015). The losses due to tephritid fruit flies could reach 100% (Dhillon et al. 2005; Thomas et al. 2019). For instance, studies in Papua New Guinea have shown that *Bactrocera cucurbitae* (Coquillett) could damage 95% of bitter gourd fruits and 90% of snake gourd (Hollingsworth et al. 1997). Due to such losses, trade restrictions are often imposed on fruits originating from areas with known fruit fly infestation, leading to fruits being quarantined (Lloyd et al. 2010; Yazid et al. 2020). These restrictions, while necessary, delay the transport of fruits and often results in increased spoilage and further economic losses to farmers.

Citrus is considered as a major cash crop in many countries including Ghana where this study was conducted (Anno-Nyarko et al. 1998; Ofosu-Budu et al. 2007). However, citrus orchards are affected by fruit fly infestation from *Ceratitis ditissima* (Munro) (Diptera: Tephritidae) (Foba et al. 2012). The *Ceratitis* genera contains several multivoltine species (Chen et al. 2006; Ansari et al. 2012), of which one is *C. ditissima*. *Ceratitis ditissima* produces several generations per year making them a major concern in citrus orchards. Although *C. ditissima* has been identified as an abundant tephritid fruit fly species in citrus orchards in Ghana (Foba et al. 2012), it has also been sighted in mango orchards, albeit in smaller numbers (Hanna et al. 2020; Zida et al. 2020; J. Abraham, unpubl.).

Olfaction in insects enables them to navigate to their host plants (Robacker & Heath 1996; Siderhurst & Jang 2010; Abraham et al. 2014, 2015, 2022). Many studies have demonstrated that insects orientate toward odours they perceive as food (e.g. Kimbokota et al. 2013; Cha et al. 2014; Epsky 2015; Knight et al. 2016; Cloonan et al. 2018). In *Drosophila* spp. it is known that fermentation products are particularly attractive and have been used to monitor their presence in fruit farms (Landolt et al. 2012; Cha et al. 2014; Cloonan et al. 2018). Among tephritid fruit flies, it is known that the oriental fruit fly, *Bactrocera dorsalis* Drew, Tsura & White (Diptera: Tephritidae) is attracted to host fruit odours (Kimbokota et al. 2013). Such food odours could potentially be harnessed to monitor fruit flies and subsequently optimised for control (Biasazin et al. 2018). Generally, fruit flies are controlled with insecticides, with some farmers following strict calendar-based spraying, which may impact negatively on the environment (Beers et al. 2011; Badii et al. 2012; Abraham et al. 2015).

Moreover, it is expensive for many smallholder farmers to buy synthetic insecticides to apply in their farms regularly on a calendar-basis. It is therefore necessary to find cheap and environmentally friendly ways of monitoring the presence of *C. ditissima* in citrus orchards. This could help farmers to detect the insect pest early and subsequently facilitate its control (Ekesi et al. 2006; Suszkiw 2014; Abraham et al. 2015; Marakhan et al. 2017). The volume of synthetic insecticides to be applied per year could be reduced if monitoring is done to know when and where to apply insecticides (Zehnder et al. 1995; Stewart et al. 2002).

In this study, we evaluated the potential of palm wine and three other food-based mixtures for monitoring the presence of *C. ditissima* in citrus orchards. In addition, we investigated the percentage capture of non-target flies compared to the target fly, *C. ditissima*. The tested baits, based largely on locally available materials were palm wine, sugarcane spirit-wine mixture, apple cider vinegar and yeast-sugar mixture. These were tested in an attempt to provide other viable alternative baits to commercial baits, which are mostly imported.

MATERIALS and METHODS

Study area

The study was conducted in a 3.68 ha citrus orchard (*Citrus sinensis* (L.) Osbeck cv. Valencia late) located in the Central Region of Ghana (5.085607°, –1.558507°). The citrus trees were planted in rows. The distance between the trees in the rows was 4 m. Furthermore, the study orchard was surrounded by other citrus orchards (Figure 1). The study was conducted over an 8-week period between November through to January, which coincides with the major fruiting season of citrus in Ghana (Lawson et al. 2017). The study commenced two weeks before the harvesting period (i.e. 21 November) and ended three weeks after harvesting (i.e. 9 January) when *C. ditissima* populations had reduced drastically.

Tested food-based baits

We tested the potential of the following food-based baits: palm wine (PW), sugarcane spirit + wine mixture (SCS–W), apple cider vinegar (ACV) (Heinz, Heinz North America, Pittsburgh, PA) and yeast + sugar mixture (Y–S) as attractants of *C. ditissima*. Fresh PW was obtained from the sap of the inflorescence of the oil palm tree (*Elaeis guineensis* Jacq.; Arecaceae: Cocoeae) (Stringini *et al.* 2009; Karamoko *et al.* 2012). Palm wine was chosen because it is easily available in Ghana, several other African countries, Asia and South America (Uzochukwu et al. 2007; Ukwuru & Awah 2013, Shet & Belur 2015). The sugarcane spirit (ca 50% v/v) + wine mixture (13% v/v) was prepared by modifying the recipe for Droskidrink (Grassi et al. 2014; Burrack et al. 2015) by mixing 675 ml of sugarcane spirit with 230 ml of red wine (Rey Don Garcia, Rioja, Spain) and 18 g of white sugar (CSR sugar, Sugar Australia, Yarraville, VIC) to obtain ~900 ml of liquid. The sugarcane spirit was obtained from a local brewer who distilled fermented sugarcane juice. The Y–S mixture was prepared following the guidelines published in Walsh et al. (2011) and Burrack et al. (2015) by mixing 10.14 g of yeast (*Saccharomyces cerevisiae*; S.I. Lesaffre, Marcq., France), 50.70 g of white sugar (CSR sugar) and 900 ml of water. Apple cider vinegar (Heinz) was obtained from a grocery store (Sonturk Supermarket, Cape Coast, Ghana). These particular food-based baits were chosen because similar baits have previously been used in monitoring other fruit flies e.g. *Bactrocera minax* (Enderlein), *Ceratitis capitata* Wiedemann, *Drosophila suzukii* (Matsumura) (Beers et al. 2011; Landolt et al. 2012; Zhou et al. 2012; Lee et al. 2013; Burrack et al. 2015; Guillemain et al. 2021). Palm wine and sugarcane spirit were particularly chosen because they are common and relatively cheap in most parts of Africa, Asia and South America (Alcarde et al. 2014; Bortoletto & Alcarde 2015). Moreover, fruit flies are generally known to be attracted to fermented products (Cha et al. 2015; Piñero et al. 2015; Candia et al. 2018).

Field trapping

One hundred transparent deli cup traps were constructed from 370 ml disposable cups (Everpack, Accra, Ghana) by drilling five holes (diameter ~12 mm) equidistant to each other and ~3 cm from the top of the cup (Figure 2). The traps were baited with the four food-based baits, so that there were 25 cups each baited with 30 ml of PW, SCS–W mixture, ACV or Y–S mixture. These were deployed systematically in the orchard by hanging them singly at the distal ends of branches of the citrus trees so that same baits do not repeat directly after each other. The distance

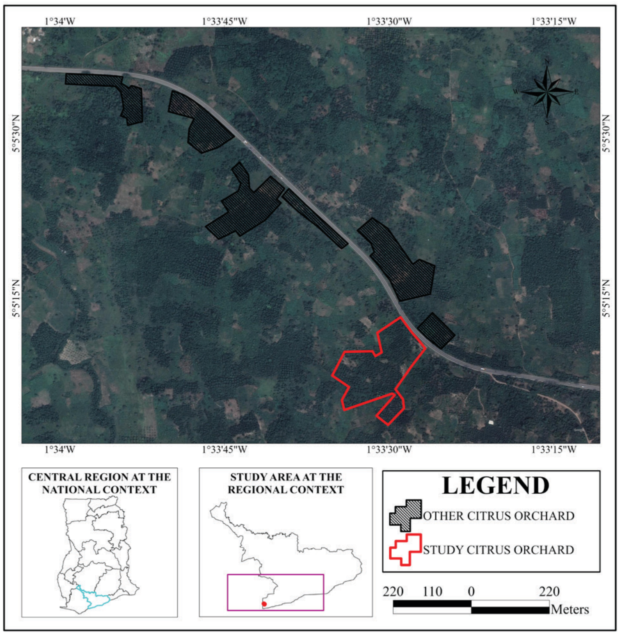

Figure 1. Study area showing the study citrus orchard marked with red borderlines and the other citrus orchards in the area marked with black borderlines and shaded

Figure 2. Deli cup trap constructed from transparent 370 ml disposable cup (Everpack, Accra, Ghana) by drilling five holes (diameter ~12 mm) equidistant to each other and ~3 cm from the top of the cup

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between traps was 12 m × 24 m and they covered ~2 ha of the citrus orchard (Figure 3). Contents of baited traps were checked weekly.

Collection of trapped insects

At the end of each week, the traps were taken down and captured flies were emptied into a labelled vial. This was done so that all insects trapped by the respective baits were placed in separate vials, thus, there were separate vials for PW, SCS–W, ACV and Y–S. The deli cup traps were cleaned and fresh baits were placed in them before hanging on the trees for the following week. Vials containing the captured flies were transported to the laboratory where flies were confirmed as *C. ditissima* or other flies by viewing their features under an optical microscope (Leica, Leica Microsystems Ltd, Heerbrugg, Switzerland) and comparing them with those described in Ekesi & Billah (2007). This was repeated for eight consecutive weeks.

Statistical analysis

Captured *C. ditissima* were sorted by sex and counted. The count of all *C. ditissima* for each treatment (bait) per week was treated as one data set (n = 1/per week). The non-target flies *B. dorsalis* and *Drosophila* spp. were also counted but not separated by sex. One-way analysis of variance (Minitab 17, Minitab Inc., State College, PA) was performed on the number of insects captured. The significance level, α, was 0.05 at a confidence interval of 95%. Where statistical differences occurred, Tukey's test was performed (Minitab).

RESULTS

A total of 2942 adult *C. ditissima* were captured over the entire 8-week period of monitoring with the four different baits in deli cup traps. There were significant differences in the capture of *C. ditissima* by the different baited traps (F = 6.21; df = 3; p = 0.002; Table 1). The average capture of adult *C. ditissima* per week by PW-baited traps was significantly higher than the captures by ACV- and Y–S-baited traps. The capture by PW-baited traps was numerically higher than the capture by SCS–W-baited traps but not statistically higher. A similar observation was made between the numbers of *C. ditissima* captured by ACV- and Y–S-baited traps (Table 1).

Total adult female *C. ditissima* captured over the monitoring period was 1540. There were significant differences in the number of adult females that were captured by the different baited traps (F = 5.72; df = 3; p = 0.003; Table 1). Significantly more females were captured by PW-baited traps than ACV- and Y–S-baited traps. The number of females captured by PW-baited traps was also higher than that captured by SCS–W-baited traps but not statistically different. Almost equal numbers were captured by ACV and Y–S-baited traps (Table 1).

The total number of adult male *C. ditissima* captured over the 8-week study period was 1402. The different baited traps captured significantly different numbers of adult males (F = 6.01; df = 3; p = 0.003; Table 1). The number of males captured by PW-baited traps was significantly higher than those captured by ACV- and Y–S-baited traps. Moreover, the number of *C. ditissima* captured by PW-baited traps was higher than that of SCS–W-baited traps but not statistically different. Almost the same number of males was captured by ACV- and Y–S-baited traps (Table 1).

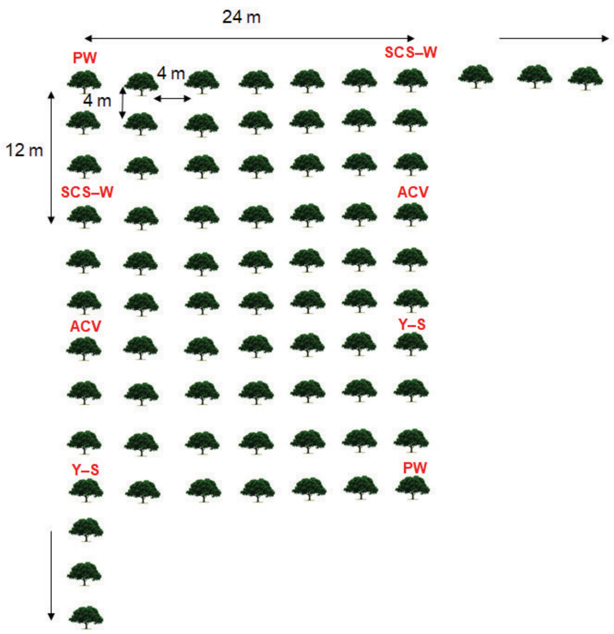

Figure 3. Schematic layout of the study citrus orchard

The "tree" image used here was created by George Hodan and released under Public Domain license CCO (https://creativecommons.org/publicdomain/zero/1.0/). The image is available at https://www.publicdomainpictures.net/en/view-image.php?image=173260&picture=tree. PW = palm wine; CSC–W = sugarcane spirit + wine mixture; ACV = apple cider vinegar; Y–S = yeast + sugar mixture

Besides *C. ditissima*, non-target flies were also captured in the baited traps. A total of 136 adult *B. dorsalis* were captured among all four baits. There were significant differences in the overall capture of *B. dorsalis* by the different baited traps (F = 6.19; df = 2; p = 0.002; Table 1). The number of *B. dorsalis* captured by SCS–W and PW-baited traps were similar and both were significantly higher than the number captured by Y–S- and ACV-baited traps. Yeast + sugar-baited traps captured an average of one per week and the total capture of *B. dorsalis* by ACV-baited traps for the entire 8-week study period was one individual.

A total of 3296 *Drosophila* spp. were captured as non-target flies in the traps. The number of *Drosophila* spp. captured by the different baited traps were significantly different (F = 5.36; df = 3; p = 0.005; Table 1). The number of *Drosophila* spp. captured by both SCS–W and PW-baited traps were significantly higher than that captured by Y–S-baited traps.

Table 1. Capture of adult *Ceratitis ditissima* and the non-target insects, *Bactrocera dorsalis* and *Drosophila* spp., in traps baited with four different food-based baits

Bait	Total capture	Mean ± SE capture/week	Target insect capture				Non-target insects capture			
			Female		Male		*Bactrocera dorsalis*		*Drosophila* spp.	
			Total	Mean ± SE / week	Total	Mean ± SE / week	Total	Mean ± SE / week	Total	Mean ± SE / week
PW	1409	176 ± 47^{a}	749	93 ± 27^{a}	660	83 ± 20^{a}	66	8 ± 3^{a}	1257	157 ± 36^{a}
SCS–W	1041	130 ± 36^{ab}	525	65 ± 16^{ab}	516	65 ± 20^{ab}	64	8 ± 3^{a}	1252	157 ± 36^{a}
ACV	385	48 ± 12^{bc}	200	25 ± 7^{b}	185	23 ± 5^{bc}	1	$\sim 0^{b}$	478	60 ± 12^{ab}
Y–S	107	13 ± 2^{c}	66	8 ± 1^{b}	41	5 ± 1^{c}	5	$\sim 1^{b}$	309	39 ± 13^{b}
Grand Total	2942		1540		1402		136		3296	

Mean ± SE with different letters attached in columns are statistically different at $p \leq 0.05$ based on analysis of variance followed by Tukey's test. PW: palm wine; SCS–W: sugarcane spirit + wine mixture; ACV: apple cider vinegar; Y–S: yeast + sugar mixture.

Though numerically higher, captures by SCS–W and PW-baited traps were not significantly higher than those of ACV-baited traps (Table 1).

When the captures of each bait were evaluated based on the percentage of target and non-target flies captured, SCS–W and Y–S mixtures captured significantly more *Drosophila* spp. than *C. ditissima* (Table 2). The percentage of *Drosophila* spp. captured by ACV-baited traps was about 12% higher than the *C. ditissima* captured but this was not significantly different. Furthermore, PW-baited traps captured only 2% more *Drosophila* spp. than *C. ditissima*, the target fly. In all cases, the tested baits captured more *C. ditissima* than *B. dorsalis* (Table 2).

DISCUSSION

All the tested baits were food-based and so were attractive to *C. ditissima*. Food is often incorporated in many commercially available baits because of the importance of nutrients in the survival of insects. Baits that incorporate food include CeraTrap, Mazoferm E802, Torula yeast, GF-120, Hymlure and Nulure meant for monitoring and capture of tephritid fruit flies (Epsky et al. 2011; Ekesi et al. 2014; Perea-Castellanos et al. 2015; Piñero et al. 2015; Hanna et al. 2020). Recent findings indicate that locally available materials such as beer waste (Piñero et al. 2015) and poultry manure (Filgueiras et al. 2016) are effective in monitoring tephritid fruit flies in orchards. This fact, coupled with the relatively high cost of commercially available baits to smallholder farmers (Ekesi et al. 2014; Filgueiras et al. 2016; Candia et al. 2018) has made it necessary to test other locally available materials that could help detect and control *C. ditissima* is citrus orchards.

Our findings show that PW is effective in detecting *C. ditissima* in citrus orchards. Moreover, PW-baited traps captured relatively less non-target flies, particularly *Drosophila* spp. This makes it a better bait than the others tested. In fact, ACV is said to be a non-selective bait because it captured only 26–31% of the target fly in a study (Lee et al. 2012). In this study, PW-baited traps captured 42–54% of the target fly, *C. ditissima*. Moreover, it captured 1–3% of *B. dorsalis*. This indicates that, palm wine is relatively selective in attracting *C. ditissima* but could also detect *B. dorsalis* which was not a target for PW-baited traps. This is a plus because it is known that *B. dorsalis* is a serious pest in citrus orchards elsewhere (Li et al. 2019; Faye et al. 2021).

Palm wine emits several volatile organic compounds including acetic acid, benzyl alcohol, ethyl hexanoate, ethyl octanoate, 3-methylbutan-1-ol and nonanal (Uzochukwu et al. 1997). These compounds are known to elicit attractive responses in several tephritid fruit flies. For instance, acetic acid, one of the first volatiles emitted from PW as it ferments (Uzochukwu et al. 1997) elicits antennal responses from *B. cucurbitae* and could be used as one of the components in an attractive bait for monitoring it in cucumber cultivations (Siderhurst & Jang 2010). Likewise, benzyl alcohol and nonanal elicit antennal responses from *B. cucurbitae* with acetic acid and nonanal acting in synergy with other volatiles as a bait for *B. cucurbitae* (Siderhurst & Jang 2010). Moreover, ethyl hexanoate and ethyl octanoate in combination with hexanol and 1,8-cineole attracts *Anastrepha ludens* Leow in citrus orchards (Robacker & Heath 1996). In addition, ethyl octanoate elicits antennal responses from both male and female *C. capitata* (Cossé et al. 1995). The antennal response to these chemicals is very important in the process of discovering and formulating baits for fruit fly monitoring. Another important physiologically active compound in PW, 3-methylbutan-1-ol, has been found to act in synergy with other volatiles such as 4,8-dimethyl-1,3(*E*),7-nonatriene, butyl hexanoate, and dihydro-β-ionone to elicit attraction response from *Rhagoletis pomonella* (Nojima et al. 2003). Palm wine may have been very attractive to *C. ditissima* probably because of the presence of these compounds in the volatiles emitted and a possible synergy with other compounds present in it.

In previous studies, wine has either been used alone or in combination with other baits to monitor fruit flies (Landolt et al. 2012; Burrack 2015; Beers et al. 2021). Fruit fly catch was improved by the combination of wine and other attractants. The combination of sugarcane spirit and wine likely accounts for the relatively good performance of this bait. This bait captured more *C. ditissima* than ACV and Y–S mixture. The ACV and Y–S mixture-baited traps on the other hand captured more *Drosophila* spp. reaching 46–66% and 60–72%, respectively. This is not surprising because *Drosophila* spp. are normally attracted to vinegar and yeast which are products associated to fermentation and rotten fruits (Epsky et al. 2015; Keesey et al. 2015; Cloonan et al. 2018). The highest proportion of *Drosophila* spp. captured in the Y–S-baited traps is in agreement with an earlier study that showed that adding the yeasts, *Saccharomyces cerevisiae* or *Aureobasidium pullulans,* and sugar to insecticides in the class diamides and spinosyns, increased the proportion of *Drosophila suzukii* that were killed significantly by the insecticides (Knight et al. 2016).

It is desirable to have a selective bait when monitoring a particular insect pest. In our study, though PW was the most selective among the tested baits, it still had a relatively high proportion of *Drosophila* spp. The progressive fermentation of PW may account for the proportion of *Drosophila* spp. it captured (Epsky et al. 2015). In spite of the relatively high proportion of non-target flies in the catches of the PW, it is a promising bait which could be optimised. For example, a 5-component bait for *D. suzukii* was optimised to perform better in a 4-component bait (Cha et al. 2014). Today, that 4-component bait is the basis for the commercial bait Pherocon SWD (Suszkiw 2014).

It is worth noting two important findings that have come out of this study, thus 1) locally available materials could be used as baits in traps to monitor *C. ditissima* and that PW, a relatively cheap beverage, was the most effective among tested materials (SCS–W mixture, Y–S mixture and ACV); 2). Palm wine attracted relatively less non-target flies i.e., *Drosophila* spp., compared to the other baits tested. The overall effectiveness of the baits for monitoring *C. ditissima* could therefore be ranked in descending order as: PW ≥ SCS–W mixture ≥ ACV ≥ Y–S mixture.

In conclusion, this study has shown that PW is attractive to *C. ditissima* and that, when placed in simple homemade deli cup traps, it could attract and trap adult *C. ditissima*. Therefore, raw PW could be utilised to monitor the presence of *C. ditissima* in citrus orchards. The advantage of using PW is that it is relatively

Table 2. Mean percentage catch of *Ceratitis ditissima* and the non-target flies, *Bactrocera dorsalis* and *Drosophila* spp., for each bait per week over an 8-week study period.

Fly	Fly status	Percentage catch per bait			
		PW	SCS–W mixture	ACV	Y–S mixture
Ceratitis ditissima	Target	48 ± 6^{a}	43 ± 3^{b}	44 ± 10^{a}	33 ± 6^{b}
Bactrocera dorsalis	Non-target	2 ± 1^{b}	3 ± 1^{c}	0^{b}	1 ± 1^{c}
Drosophila spp.	Non-target	50 ± 6^{a}	55 ± 3^{a}	56 ± 10^{a}	66 ± 6^{a}

Different letters attached to mean ± SE in columns indicate significant differences at $p \leq 0.05$. PW: $F = 28.26$; df = 2, 21; $p < 0.001$; SCS–W mixture: $F = 11.14$; df = 2, 21; $p < 0.001$; ACV: $F = 13.29$; df = 2, 21; $p < 0.001$. Y–S mixture:
$F = 43.38$; df = 2, 21;$p < 0.001$. PW: palm wine; SCS–W: sugarcane spirit + wine mixture; ACV: apple cider vinegar; Y–S: yeast + sugar mixture.

inexpensive and abundantly available in several countries across Africa, Asia and South America, where citrus is cultivated. Smallholder farmers could therefore access it easily. For large commercial orchards, the use of PW for monitoring could indicate which parts of the orchard are under threat of tephritid fly infestation for remedial action.

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AUTHORS' CONTRIBUTIONS

Conceptualisation – JA; Data curation – JA, CA, FOK; Formal analysis – JA; Funding acquisition – JA, CA, FOK, JDA; Investigation – JA, CA, FOK; Methodology – JA; Project administration – JA, JDA; Resources: JA, CA, FOK, JDA; Supervision – JA, JDA; Validation – JA; Visualisation – JA; Writing original draft: JA, CA; Writing review and editing – JA, CA, FOK, JDA.

ORCID IDs

John Abraham – https://orcid.org/0000-0001-6049-6042
Janice Dwomoh Abraham – https://orcid.org/0000-0002-9908-275X
Carlos Amissah – https://orcid.org/0000-0002-3971-0005

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