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Original article

Sensitivity of eggs and larvae of *Amblyomma variegatum* (Acarina: Ixodidae) to *Clausena anisata* essential oil mixed in two vegetable oils as vehicle

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ABSTRACT

Livestock in tropical and subtropical areas is under almost constant threat of ticks, in particular the species *Amblyomma variegatum* Mfr. (Acarina: Ixodidae), especially during the rainy season. The IPM programs against this ectoparasite undoubtedly require knowledge of its bio-ecology and diversification of means of control. Thus, the toxic effects of the essential oil of *Clausena anisata* Hook (Rutaceae) diluted in two vegetable oils were tested on eggs and larvae of *A. variegatum* under the laboratory conditions of the temperature from 22-25°C, relative humidity from 78-91% and a photoperiod of 12 L/12h D. Toxicity tests were carried out in Petri dishes containing Whatman paper on which various test solutions were deposited. In each treated dish, 100 eggs or 40 larvae were released. A dose of 0.124 µL / cm² of a dilution of the essential oil of *C. anisata* prevented the hatching of 95% of the eggs and provoked 60% mortality of the larvae after 24 hours of exposure. Although these results differ significantly from those of the reference acaricide bayticol which is very highly toxic, the essential oil of *C. anisata* could be considered very toxic to the preimaginal stages of the tick *A. variegatum*. However, the cost of

treatment with the essential oil of *C. anisata* should be evaluated to better assess its use in the control of *A. variegatum* by farmers.

1. Introduction

Blood-sucking ticks are obligate arthropod ectoparasites of vertebrates. Various pathogens like protozoa, rickettsia, spirochetes, viruses and helminths are transmitted by ticks more than other group of arthropod vectors to Reptiles, Birds and Mammals (Nejash, 2016; Berggoetz et al., 2014a, 2014b). They are therefore ranked among the most deadly disease vectors of which thirty species were reported (Update news, 2002). Approximately 80 % of global herd is threatened by tick's infestation (Laamri et al., 2012). This seriously affects pastoral activity in many developing countries, especially in Africa (Bisusa et al., 2014; Farougou et al., 2013; Adakal et al., 2013). Amongst about 800 tick species described worldwide (Hoogstraal and Aeschlimann, 1982), *Amblyomma variegatum* (Acarina: Ixodidae) is the most harmful to livestock in West Africa (Mollong et al., 2018c). Anaplasmosis, babesiosis, heartwater, theileriosis are some important parasitic diseases caused by this tick. In addition to the scars left by the bites of adults strongly disabling recovery of hides for leather, this tick produces various toxins that cause irritation, allergies and paralysis observed in their host (Berggoetz et al., 2014a, 2014b). A massive infestation of this tick can provoke a loss of 15-20 kg of wet weight in a young bull in West Africa during the rainy season (Stachurski, 2007). This represents today, a loss of 80 to 100 US dollars and a collapse of the livestock and dairy industry for a population already suffering from malnutrition. The irreparable damage to the teats of cows, especially lactating ones, increases calf mortality by about 6.12 % in sucklings and 20 % in weaned calves (Hounzangbe-Adote et al., 2001; Stachurski et al., 1988).

Different strategies have been developed to cope with the threat of ticks, from removing them by hand to dipping in salt baths. Thereafter, chemical control had imposed itself with the first use of arsenic derivatives (Uilenberg, 1975). These evolved into synthetic products such as organochlorines (DDT, lindane, dieldrin,) and to organophosphates (parathion, parathion ethyl...), carbamates (carbaryl, promacyl...) and pyrethroids such as cyfluthrin, cypermethrin, deltamethrin and flumethrin. Means of control were then extended to the biological and chemical mediators like pheromones (Nawaz et al., 2015; Kosgei et al., 2014; Allan et al., 1998). Vaccination also, has been used to prevent this scourge (Stachurski, 2000; Rutti and Brossard, 1992). Unfortunately, these control methods do not always show sufficient efficacy and while taking care of environmental issues.

The increasing use of chemical acaricides faces the emergence by tick's resistance (Adehan et al., 2016; Abbas et al., 2014; Guerrero et al., 2012; Chevillon et al., 2007). These pesticides have further negative consequences for the environment and the health of farmers, livestock and consumers (De Meneghi et al., 2016). The use of pheromones and vaccines requires significant prior efforts to achieve high efficiency. This fact has motivated the search for new and more effective means of control that cattle breeders in developing countries could afford easily.

The goal of the present work is to strengthen chemical control strategies against *A. variegatum* Mfr. (Acarina: Ixodidae) through diversification involving botanical acaricides in a biologically, economically and environmentally acceptable way. *Clausena anisata* Hook (Rutaceae) essential oil has been shown to be effective on the tick *A. variegatum* on sheep (Nuto et al., 2008), but a suitable solvent carrier of this essential oil must be identified. In this work, we evaluated *C. anisata* essential oil toxicity using vegetable oil of *Jatropha curcas* Linn. (Euphorbiaceae) and that of the kernels of *Elaeis guineensis* Jacq. (Arecaceae) as solvents on the eggs and larvae of *A. variegatum*. The results were compared with those of bayticol, a reference pyrethroid acaricide used worldwide.

2. Materials and methods

2.1. Collection and rearing of the tick

Females of *A. variegatum*, engorged with blood, were collected from four sites located in ecological zone V of Togo: Lome Gbossimé (06 ° 09'45 '' N, 01 ° 12'38 '' E), Lome Campus (06 ° 10 '49 '' N, 01 ° 12'66 '' E), Adétikopé (06 ° 18'85 '' N, 01 ° 12'42 '' E) and Koudassi (02 ° 63'52 '' N, 07 ° 33'97 '' E). Cattle and sheep were the hosts of *A.*

variegatum at these sites. The ticks were collected from the ground early in the morning before the departure of the hosts for grazing or in the afternoon on their return. The engorged females were kept in plastic jars sterilized with absolute alcohol and containing cotton moistened with water. The jars were covered with a muslin cloth for aeration. Ticks were collected at least two weeks after anti-tick treatment of host animals. The female ticks were kept under ambient laboratory conditions for spawning from April to October 2010. The eggs were kept in the same laboratory conditions until the larvae hatched. The development of the larvae also took place in the same conditions. During the experiment, temperatures varied between 21 and 29 °C and relative humidity between 75 % and 100 %. Photoperiod was about 12 L/12h D.

2.2. Test chemicals and solvents

The essential oil of *C. anisata* was obtained from ''Les Distilleries d'Huiles Essentielles'' (3MRG AROMATIQUE, 04 b. p. 623 Lome-Adidogome) at Ahlon Sassanou in the Dayes district (Republic of Togo). The chemical screening of this essential oil was obtained by Moudachirou et al. (1997) and showing that its major constituents were Limonene, β-phellandrene and garmacrene D/ γ²-cardinene in Togo. According to same authors, the minor constituents of essential oil of *C. anisata* belonging to the group of bicycles (sabinene and pinene) and some sesquiterpenes (caryophylene and cadinene). (E)-anethole and estragol are other major constituents of this essential oil (Addae-Mensah et al., 1996). Chemical analysis of the essential oil use was carried out by Pyrenessences Analysis on 01/04/2017.

The bayticol manufactured by Bayer CropScience, 5th Avenue Vorterskroom Nigel, 1490 and contained 1% (m/v) flumethrin was the reference product used as positive control to assess the toxicity of the essential oil of *C. anisata*.

Vegetable oils used as solvents for the test chemicals were palm kernel oil purchased at the Adidogomé (Lomé, Togo) market and *J. curcas* oil obtained by pressing the crushed seeds steamed for 15 to 20 minutes. Each test material was mixed with an equal volume of one or the other vegetable oils to prepare a stock solution. Each stock solution was successively diluted 1:1 (v/v) to prepare test solutions.

2.3. Toxicity tests

The effect of the solvent oils was first evaluated on eggs of *A. variegatum* at different ages. Then, seven different concentrations of the essential oil of *C. anisata* and bayticol diluted in the vegetable oils were tested on eggs and larvae. Two hundred microlitres of each test solution were poured on a filter paper placed in a glass Petri dish of 8 cm diameter. After uniformly spreading of the test solution on the entire surface of each filter paper disc, 100 eggs or 40 larvae were introduced in each Petri dish. Distilled water was used as control. Eggs were tested at three age points, 14, 35 and 49 day-old choices related to the developmental stages of the eggs of *A. variegatum*. Larvae were tested when 15-day old. Mortalities of the eggs and the larvae were recorded respectively after eggs hatching and 24 hours of exposure. Three and five repetitions respectively for eggs and larvae were performed per dose.

2.4. Presentation of the results

Concentrations of the essential oil of *C. anisata* and of bayticol were expressed as the volume of the stock solution applied per area of the filter paper disc (µL/cm^2). The effective mortality caused by the tested compound was calculated using Abbott (1987) formula. The LD_{90}, LD_{50} and LD_{10} for larvae after 24 hours of exposure were determined using the Windl 23 Version 2.0 CIRAD-CA/MABIS software.

2.5. Statistical data analysis

Each trial was analyzed as repeated measures analysis of variance (ANOVA) at 5 % with SPSS.v.16.0 software. Averages were discriminated with the Post Hoc Test (LSD) when the analysis of variance revealed statistically significant differences.

3. Results and discussion

3.1. Chemical components of *C. anisata* essential oil

Chemical analysis results of *C. anisata* leaves essential oil showed that Trans-anethole and Estragol are the major components (Table 1).

Table 1
C. anisata leaf essential oil compounds and their proportion.

Pic	TR (min)	Chemical compounds	Proportion (%)
1	7.7	α-Pinene	1.07
2	10.0	β-Pinene	0.17
3	11.6	β-Myrcene	0.25
4	12.8	Limonene	0.29
5	14.3	γ-Terpinene	0.23
6	15.2	p-Cymene	2.31
7	21.6	Citronellal	0.14
8	23.3	Linalool	0.14
9	25.2	β- caryophyllene	0.22
10	27.1	Estragol	57.06
11	27.3	α-Humulene	0.20
12	28.4	Z- β-Farnesene	0.32
13	28.5	Neral	0.34
14	29.0	Citronellol	0.19
15	29.1	Cis-Anethole	0.10
16	29.6	α-Curcumene	0.22
17	31.0	Trans-Anethole	29.88
18	34.8	Caryophyllene oxide	0.40
19	35.6	Anisaldehyde	2.67
20	36.1	Epoxy-6,7- Humulene	0.23
21	38.2	Anicetone	0.19
22	46.4	3-Methoxycinnamaldehyde	0.32
23	49.7	Hydroxyphenyl butanone Mw=164	0.30
		Total	97.24

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3.2. Effects of solvent vegetable oils, essential oil and bayticol on eggs

Sensitivity of the eggs of *A. variegatum* to the vegetable oils decreases with their age. The 14 and 35 day-old eggs were much more sensitive to the vegetable oils. Mortality in these eggs was 70.85 ± 6.29 % to 83.39 ± 4.33 % compared with about 10 % in the control (Table 2). Mortality varied from only 11.44 ± 7.2 % to 15.50 ± 7.78 % in the oldest eggs of 49 days. Tests with the stock solutions of *C. anisata* essential oil and bayticol caused 100 % destruction of eggs regardless of their age (Table 2).

Table 2
Effects of raw extract of vegetable oils, essential oil and bayticol on eggs (n = 100 eggs).

Tests	Quantity of water or test crude product	Mean mortality (%) in the eggs in relation to their ages (in days)*		
		14	35	49
Control	200µL	9.67 ± 2.08c	9.67 ± 2.09c	9.67 ± 2.10b
Palm kernel oil	200µL	81.55 ± 3.89b	70.85 ± 6.29b	11.44 ± 7.26b
J. curcas oil	200µL	83.39 ± 4.42b	74.91 ± 4.99b	15.50 ± 7.78b
Essential oil of *C. anisata*	200µL	100 ± 00a	100 ± 00a	100 ± 00a
Bayticol	200µL	100 ± 00a	100 ± 00a	100 ± 00a

* Means with different letters in the same eggs group are different statistically at 5%.

Eggs observation with binocular microscope revealed that the treatments with both vegetable oils provoked morphological damages to the eggs. For instance, the 14 day-old eggs became wrinkled and translucent (Fig. 1 B) when compared to the control (Fig. 1 A). In this case embryogenesis continued in a few eggs only. The eggs aged

14 days and treated with the essential oil or bayticol stock solution did not show any sign of embryogenesis. They became completely wrinkled, dark, dehydrated, empty and without any mucilaginous protective substance after the period of exposure (Fig. 1 C).

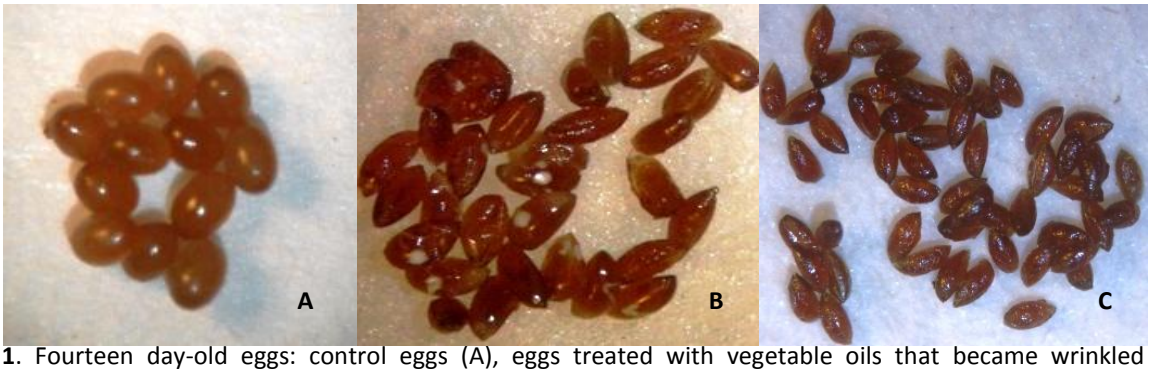

Fig. 1. Fourteen day-old eggs: control eggs (A), eggs treated with vegetable oils that became wrinkled and translucid (B) and the eggs treated with essential oil or bayticol that became more wrinkled and black (C) (x 320).

3.3. *C. anisata* essential oil and bayticol dilutions toxicity on the older 49 days eggs

The concentration of 0.016 µL/cm^2 of the essential oil caused 49.45 % and 58.30 % mortality in the eggs when diluted respectively in palm kernel and *J. curcas* oils (Table 3). The same dilution of bayticol diluted in palm kernel and *J. curcas* oils provoked respectively 80.44 % and 85.24 % mortalities in these eggs (Table 3). Statistical analysis of the results revealed that, our essential oil toxicity did not vary significantly with the diluents (F = 0.818, df = 1 and p = 0.41687). From 0.031 µL/cm^2 and for the same concentration of the other treatments, mortalities in the dilutions of the essential oil and of bayticol were no more significantly different statistically after 24 h of exposure (F = 1.374, df = 1 and P = 0.30617). Bayticol was highly toxic; even its very low concentration of 0.001 µL/cm^2 in palm kernel and *J. curcas* oils killed 45.39 % and 52.77 % of the eggs respectively (Table 3).

Table 3
Average mortality of the older 49 days eggs treated with different dilutions (n = 100 eggs).

	Mean mortality/concentrations (µL/cm²)**											
Treatments*	0.001	0.002	0.004	0.008	0.016	0.031	0.062	0.124	0.249	0.498	0.995	1.99
Eo + opk	-	-	-	-	49.44 ± 25.47a	73.79 ± 14.61bc	85.97 ± 6.67cde	94.46 ± 3.32de	95.2 ± 3.88e	97.04 ± 1.69e	100 ± 00e	100 ±00e
Eo + oj	-	-	-	-	58.3 ± 21.50ab	76.75 ± 11.66bcd	83.39 ± 8.78cde	95.2 ± 3.38de	97.41 ± 1.69e	98.15 ± 1.27e	100 ± 00e	100 ± 00e
B + opk	45.38 ± 10.28a	63.09 ± 13.30bc	71.21 ± 2.93bcd	74.54 ± 9.65cd	80.44 ± 12.78de	89.66 ± 4.99ef	95.20 ± 3.88f	100 ± 00f	100 ± 00f	100 ± 00f	100 ± 00f	100 ± 00f
B + oj	56.27 ± 3.91ab	73.43 ± 7.75bcd	75.27 ± 10.52bcd	83.02 ± 8.00def	85.23 ± 9.41def	91.51 ± 4.47ef	96.68 ±2.92f	100 ±00f	100 ± 00f	100 ± 00f	100 ± 00f	100 ± 00f

*Eo + opk = essential oil of *C. anisata* + palm kernel oil; Eo + oj = essential oil of *C. anisata* + *J. curcas* oil; B + opk = bayticol + palm kernel oil; B + oj = bayticol + *J. curcas* oil; ** Means with different letters in the same dilutions for essential oil or bayticol are statistically different at 5%.

Eggs treated with essential oil or bayticol exhibited morphological deformations that varied more or less according to the dose that was applied. Embryonic development was blocked in doses above 0.498 µL/cm^2. No hatching of eggs occurred with those doses. With doses lower or equal to 0.498 µL/cm^2, eggs showed hatching slots; however, the larvae could not get rid of their chorion. Those of them that emerged died (Fig. 2).

Fig. 2. Different aspects of eggs treated with dilutions of the essential oil (concentrations in µL/cm^2 ≤ 0.498) showing the appearance of slots of emergence (arrows) (A) and hatched dead larvae that could not get rid of their chorion (B) (arrows) (x320).

3.4. Effects of the vegetable oils, essential oil and bayticol on larvae behaviour

The larvae hatched and kept in the laboratory for two weeks under our experimental conditions moved normally in search of a host after two weeks. It means that they could forage for food on their host. They did not die when exposed to water (control) or to palm kernel or *J.curcas* oils for 24 or 72 hours. But they were all killed in less than 15 minutes when exposed to the essential oil or bayticol. The dead larvae appeared wrinkled under magnifying glass.

The control larvae and those treated with low doses of the essential oil and bayticol moved relatively well in all directions in the Petri dishes. On the contrary, the larvae treated with relatively high concentrations of the essential oil and bayticol became very agitated. They fell down on their backs, stood up again and tried to get out of the dishes. They gathered in the corners of the dishes, on the bottoms or under the lids and were finally knocked down and died.

3.5. Effects of different dilutions of essential oil and bayticol on 15 days larvae

Different dilutions of this essential oil with both vegetable oils were toxic to the larvae with more than 96 % mortality with a dose of 0.50 µL/cm^2 after 24 hours of exposure. The same dilutions of bayticol in palm kernel or *J. curcas* oils killed all the larvae within the same time period of exposure. Statistical analysis didn't show significant difference in toxicity of the essential oil in relation to the vegetable oil used as solvent on one hand and between the concentrations of both pesticides on the other hand (F = 0.400, df = 1 and P = 0.54473).

Essential oil concentrations less than or equal to 0.062 µL/cm^2 caused only a limited mortality of below 50 % in 24 hours of exposure. Concentrations above 0.062 µL/cm^2 caused greater mortalities than 50 % regardless of the solvent used (Table 4).

Statistically, there is no significant difference between the dilutions made with each of the two vegetable oils (F = 1.040, df = 1 and P = 0.33771). Similarly, the toxicity of bayticol was not influenced by the vegetable oils (Table 4); (F = 1.915, df = 1 and P = 0.20378). With a dose of 0.249 µL/ cm^2, mortalities caused by bayticol were greater, even if they were statistically similar to those of the essential oil (Table 4).

Table 4
Average mortality rate (%) in larvae aged 15 days exposure to different dilutions of *C. anisata* essential oil and bayticol after 24 hours (n = 40 larvae).

	Mean mortality /concentrations (µL/cm^2)**									
Treatments*	**0.001**	**0.002**	**0.004**	**0.008**	**0.016**	**0.031**	**0.062**	**0.124**	**0.249**	**0.498**
Eo + opk	-	-	-	-	5.5 ± 5.41a	12.5 ± 11.72ab	43.5 ± 14.20c	59.5 ± 16.52de	84.5 ± 9.25f	100 ± 00f
Eo + oj	-	-	-	-	8.5 ± 8.02ab	19.5 ± 9.90b	47 ± 8.36cd	61.5 ± 14.85e	83.5 ± 11.53f	100 ± 00f
B + opk	2 ± 3.25a	12.5 ± 8.47b	72.5 ± 10.3c	99 ± 2.23e	100 ± 00e	100 ± 00e	100 ± 00e	100 ± 00e	100 ± 00e	100 ± 00e
B + oj	3.5 ± 4.18ab	12 ± 7.98b	82.5 ± 11.37d	100 ± 00e	100 ± 00e	100 ± 00e	100 ± 00e	100 ± 00e	100 ± 00e	100 ± 00e

*Eo + opk = essential oil of *C. anisata* + palm kernel oil; Eo + oj = essential oil of *C. anisata* + *J. curcas* oil; B + opk = bayticol + palm kernel oil; B + oj = bayticol + *J. curcas* oil; **Means with different letters in the same dilutions for essential oil or bayticol are statistically different at 5%.

The LD50s of *C. anisata* mixtures in palm kernel and *J. curcas* oils were respectively 0.09 ± 0.06 µL/ cm^2 and 0.08 ± 0.10 µL/ cm^2 (Table 5). Those of bayticol were 0.003 ± 0.03 µL/ cm^2 and 0.003 ± 0.03 µl/ cm^2 respectively (Table 5). However, batycol was three thousand times and two thousand and six hundred times more toxic than the essential oil when they are diluted respectively in palm kernel and *J. curcas* oil (Table 5).

Table 5
Mean lethal doses of *C. anisata* essential oil and bayticol diluted in vegetable oils after 24 hours of exposure.

	Treatments with essential oil of *C. anisata*		**Treatments with bayticol**	
Lethal doses	***C. anisata* + palm kernel oil**	***C. anisata* + *J. curcas* oil**	**Bayticol + palm kernel oil**	**Bayticol + *J. curcas* oil**
DL_{10}	0.02 ± 0.15	0.02 ± 0.21	0.002 ± 0.04	0.002 ± 0.04
DL_{50}	0.09 ± 0.08	0.08 ± 0.10	0.003 ± 0.03	0.003 ± 0.03
DL_{90}	0.30 ± 0.07	0.30 ± 0.07	0.005 ± 0.04	0.005 ± 0.04
Equation of the regression line	y = -2.20 + 2.34x	y = -1.90 + 2.16x	y = -2.55 + 5.27x	y = -2.64 + 5.81x

The tests showed that vegetable oils: palm kernel oil and *J. curcas* oil had a toxic effect on eggs of *A. variegatum*, especially, the eggs younger than 49 days. The eggs aged 49 days were less sensitive to the vegetable oils used. The eggs that survived the vegetable oil treatments and the control eggs hatched at the same time. But, unlike the control eggs, the larvae of the treated eggs couldn't leave their chorion easily during hatching and they finally died. The toxicity of the vegetable oils to the eggs younger than 49 days may be due to physical action. The oils may have obstructed the micropyles of the eggs, disrupt breathing and inhibit embryogenesis by oxygen deprivation (Nuto et al., 2008). However, the greater survivability of the 49 day-old eggs in which embryogenesis is more advanced may be explained by the appearance of the emergence slot that could favor breathing of the larvae.

The fact that the 49 day-old eggs are not sensitive to the vegetable oils justifies their use as solvents. The tests performed with different dilutions of the essential oil in both vegetable oils have proven that our essential oil induced death in the eggs of *A. variegatum* aged 49 days compared to control eggs treated with distilled water. The mortalities are dose-dependent; the concentration of 0.016 µL/cm^2 being the lowest biological effective dose. The vegetable oil of *J. curcas* tended to increase slightly the toxicity of the essential oil against that observed in the dilutions with palm kernel oil. This confirms the argument of Nuto et al. (2008) who suggested that an appropriate vehicle for this essential oil be found. Although bayticol is more toxic than the essential oil, the eggs treated with both products became completely black, wrinkled, dried and did not show any sign of embryogenesis nor any mucilaginous substance that covers eggs and sticks them to the substrate. They also became invaginated longitudinally. In fact, *C. anisata* and bayticol disable the larvae to pull down their chorion. According to Dossa et al. (1996), the effects this essential oil is similar to those of systemic acaricides. These authors observed that the pupae of ticks that feed on hosts treated with systemic acaricides could not molt easily.

Different dilutions of essential oil showed a dose-dependent toxic effect on the larvae of *A. variegatum*. The results indicated mortalities slightly higher with the dilutions made with *J. curcas* oil than those made with palm kernel oil. But statistical analysis did not reveal any significant difference. The essential oil probably affects the larvae through inhalation or contact. Either on eggs or on larvae, bayticol was much more toxic than the essential oil. The vegetable oils used alone did not have any significant impact on the 15 day-old larvae. They survived beyond three months after being exposed to these vegetable oils for 24 or 72 hours.

Natural biological effects of essential oils are not well known. One of the reasons limiting their effectiveness is their volatility. So, Rani and Osmani (1984) argued that their used as contact insecticides is not possible. Ketoh et al. (2000) suggested *C. anisata* essential oil to be a good material for fumigation. According to Chiasson et al. (2004a and b) contact between the essential oil and the target organism is necessary to induce mortality. This was corroborated by the work of Mollong et al. (2018a, 2018b), Alitonou et al. (2004), on adult tick and larvae of *A. variegatum*. To make the control of ticks with essential oils more effective, Nuto et al. (2008) suggested their incorporation into an appropriate relatively heavy oil to reduce their evaporation. This can allow an effective application, as long as their action is rapid. The results of the present work show that palm kernel oil or *J. curcas* oil can be used as vehicles for *C. anisata* leaves essential oil. The mixture of the latter with each of the vegetable oils is homogeneous; indicating good incorporation of these ingredients. In addition, statistical analyses of our results show clearly that there was no antagonistic action of the vegetable oils on the toxicity of the essential oil in our experimental conditions. Though the use of palm kernel oil as the carrier of the essential oil can be envisaged, *J. curcas,* being considered a biofuel, may be less suitable on economic reasons.

The interest of our study on the diversification of acaricides using the essential oil of *C. anisata* is its strong toxicity on pre-imaginal forms of the tick *A. variegatum*, the most harmful in West Africa (Stachurski, 2007) on one hand and the ready availability of its raw material locally and its wide distribution in the West African sub-region (Moudachirou et al., 1997) on the other hand. Because of the wide distribution of the plant, toxicity of its essential oil may vary according to the local ecological conditions, the season and the time period of harvest of the raw material as indicated by (Isman and Grieneisen, 2014). Another advantage of this essential oil is the fact that its raw material has been used against intestinal parasites and some dermatoses in humans (Avlessi et al., 2004; Gundidza et al., 1994). So, the treatments with that essential oil will not probably expose the users and the environment to major risks. Similarly, Tapondjou et al. (2005), Bouda et al. (2001) also showed that essential oils toxicity on mammals were less and caused little damage on the environment because they are biodegradable. *C. anisata* essential oil is an environmentally and economically acceptable alternative for enhancing control strategies for cattle ticks in Africa.

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