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Effect of Sodium Chloride Solution on the Oviposition Capacity of Engorged Adult Females and Hatchability of Eggs of Dog Ticks: Rhipicephalus sanguineus and Haemaphysalis leachi leachi

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Abstract: Effect of various concentrations of sodium chloride (NaCl) solution on the oviposition capacity of engorged adult females and hatchability of eggs of dog ticks: Rhipicephalus sanguineus and Haemaphysalis leachi leachi was investigated under laboratory condition. The concentrations of sodium chloride solution used were 2, 5, 10, 30 and 60% and saturated solution and the duration of times of immersion were 2, 4, 6, 12 and 24 h. The engorged adult females of R. sanguineus and H. leachi leachi did not oviposit after immersion in saturated sodium chloride solution. The number of eggs oviposited after treatment in 2% Nacl solution by R. sanguineus (1272.33±57.73) and H. leachi leachi (1122.31±120.09) were respectively lower than those of their controls 2665.31±42.5 and 1881.00±228. Adult females of H. leachi leachi did not oviposit when immersed in any of the sodium chloride concentrations for 24 h. The time of exposure of the eggs of both species to NaCl solutions did not affect their hatching capability. The eclosion periods of eggs in all the concentrations of NaCl solution were shorter than their controls. The percentage hatchability of eggs after immersion in 2, 5, 10, 30 and 60% NaCl solutions were lower than their controls. The percentage hatchability decreased as the concentration of NaCl solution increased with no hatching of eggs in saturated NaCl solution. I recommend that saturated NaCl solution be integrated into the overall policy of control of dog ticks because of its acaricidal properties causing outright mortality of engorged adult females and eggs of R. sanguineus and H. leachi leachi. This will drastically reduce the overdependence on imported acaricides.

Key words: Dog, sodium chloride, *Rhipicephalus sanguineus*, *Haemaphysalis leachi leachi*, oviposition, hatchability

INTRODUCTION

Ticks have long been regarded as constraints to human welfare and that of domesticated animals (Taylor, 2001). Ticks and the diseases they transmit are widely distributed throughout the world, particularly in tropical and subtropical countries (FAO, 1984; Shaw et al., 2001). Rhipicephalus sanguineus and Haemaphysalis leachi leachi are the two species of ticks infesting dogs in Nigeria (Dipeolu and Akinboade, 1982; Adejinmi, 2005). The importance of ticks as parasites lies in their ability to successfully multiply and establish in their environment and their ability to transmit diseases to their hosts and to maintain their physiological requirements (Akinboade, 1986). Hence, there is need to check the menance of tick infestation through tick control methods.

In companion, animals particularly dogs the main method of control of ticks and in which total reliance has been placed is the use of chemicals referred to as Acaricides. Some of the currently available chemicals in use against ticks infesting dogs are Amitraz®, Diazintol®, Friponil®, Deltamethrin, Asuntol® and Ivermectin. These are either used as tick bath or in form of powder as dust or pouron on dogs and they have been found to be effective against *R. sanguineus* and *H. leachi leachi* (Franc and Cadiergues, 1999; Estrada-Pena and Ascher, 1999; Taylor, 2001). However, acaricides have suffered from a number of drawbacks including the development of resistance and concerns over human and environmental safety (Taylor, 2001). The search for safer technologies has however been hindered by the limited number of target sites present in arthropods and to some degree by the ever-increasing costs of research and development of compounds with novel modes of action (Taylor, 2001).

Only Amoo (1984) had studied the effect of brine on *Boophilus* sp. The researcher reported that the engorged adult females of *B. decoloratus* and *B. geigyi* did not oviposit in brine solution at 30% concentration or more and the numbers of eggs laid by these species after immersion in 10% brine solution were significantly lower than those of controls.

Available literature on the effect of sodium chloride solution on ticks infesting dogs is scanty. This study reports the oviposition capacity of engorged adult females and hatchability of eggs of dog ticks: *R. sanguineus* and *H. leachi leachi* when subjected to treatment with varying concentrations of sodium chloride solution in the laboratory.

The aim is to develop an effective, environmentally friendly, cheap, acceptable and practicable control measures against these ticks.

MATERIALS AND METHODS

The ticks used for this study were adult females in various stages of engorgment collected individually by careful detachment with pairs of forceps from household dogs brought to veterinary clinics in Ibadan. The ticks were collected into glass bottles and conveyed in Kilner jars to Parasitology Laboratory in the Department of Veterinary Microbiology and Parasitology, University of Ibadan, identified and individual weights were determined and recorded using a sensitive Sartorious balance (Type 2472 Germany).

Five engorged female ticks of the same weight each of R. sanguineus and H. leachi leachi were immersed in various concentrations (2, 5, 10, 30 and 60% and saturated solution) of sodium chloride (NaCl) for different durations of time (2, 4, 6, 12 and 24 h) as described by Amoo (1984). After the duration of immersion time, the salt solution was pipetted off and the ticks were returned in clean and dry bijou bottles plugged with cotton wool to the incubator maintained at 25°C and 85% Relative Humidity (RH). Another set of five engorged female ticks of the same weight each of R. sanguineus and H. leachi leachi which were not treated with sodium chloride solution served as controls. The ticks were observed daily for oviposition after the salt treatment. The eggs laid by each tick were removed daily and counted using the method of Dipeolu and Ogunji (1980).

The preoviposition period, duration of oviposition and the number of eggs laid by each tick were recorded and all the eggs laid by the five ticks of each tick species and at the different duration of time were pooled together.

To investigate the effect of sodium chloride on the eggs 0.05 g of freshly laid eggs of *R. sanguineus* and *H. leachi leachi* were immersed in various concentrations 2, 5, 10, 30 and 60% and saturated sodium chloride solution for different durations of time 2, 4, 6, 12 and 24 h, respectively. After this treatment the eggs were harvested by pipetting the sodium chloride solution out of the bottle and immediately returned in another clean, dry and tightly corked bijou bottle to the incubator maintained at 25°C

and 85% RH (Amoo, 1984). Another 0.05 g freshly laid eggs each of R. sanguineus and H. leachi leachi which were not treated with sodium chloride solution served as the controls. The eggs were observed daily till the end of hatching of eggs and two weeks after the last larva (e) have hatched out of the eggs. From the day, hatching started until ended the hatched larvae were separated at 8.00 h every morning from the unhatched eggs. This was possible because before hatching the eggs usually clumped together. Once hatching started the larvae move away from the clump. The separation of larvae from unhatched eggs was therefore achieved by scooping the lump (s) of unhatched eggs into another bijou bottle and covered tightly while 10% formalin was added to the first bijou bottle which then contained only hatched larvae (Dipeolu and Akinboade, 1982).

The number of larvae was ascertained under a dissecting microscope. This was repeated every day until no larva (e) was seen to hatch. The unhatched eggs which existed in small clumps were observed for another two weeks before they were declared unhatched and dead. They were then taken out of bijou bottle and counted through the addition of xylene under a dissecting microscope (Dipeolu and Ogunji, 1980). The eclosion period duration of hatching, day of peak hatching and percentage hatchability of eggs were recorded.

All data were subjected to statistical Analyses of Variance (ANOVA), χ^2 -test and student's t-test using the computer package SPSS version 1.12002.

RESULTS AND DISCUSSION

Table 1 shows the effect of sodium chloride (NaCl) solution on the oviposition capacity of engorged adult females of *R. sanguineus* and *H. leachi leachi*. The engorged adult females of *R. sanguineus* and *H. leachi leachi* did not oviposit in saturated sodium chloride solution in all durations of time. Also the number of eggs oviposited after treatment in 2% NaCl solution was lower than those of controls. *H. leachi leachi* did not oviposit when immersed in any of the concentrations of NaCl solution for 24 h.

Table 2 and 3 show the effect of various concentrations of NaCl solution on the eggs of *R. sanguineus* and *H. leachi leachi* immersed for different duration of time, respectively. The results show that the time of exposure of the eggs of both species to NaCl solution did not affect their hatching capability. However, the percentage hatchability of eggs of both species after immersion in 2, 5, 10, 30 and 60% NaCl solutions were lower than their respective controls. The percentage hatchability decreased as the concentration of

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Table 1: Effect of various concentrations of sodium chloride solution on the oviposition capacity of engorged adult female of *Rhipicephalus sanguineus* and *Haemaphysalis leachi* leachi

	Exposure time (h)	R. sanguineus			H. leachi leachi		
NaCl Conc.		Mean pre- oviposition (days)	Mean duration of oviposition (days)	Mean no. of eggs laid	Mean pre- oviposition (days)	Mean duration of oviposition (days)	Mean no. of eggs laid
Control	-	4.0±0	10.33±0.58	2565.33±47.52	4.33±0.58	8.0±1.0	1881.00±228.11
0	2	4.0±0	10.33±0.58	1272.33±57.73	5.0±0	7.0 ± 1.0	1122.33±120.09
	4	4.33±0.58	10.0 ± 0	1160.33±47.36	5.33±0.58	7.0 ± 1.0	962.0±125.22
	6	5.0±0	8.33±0.58	1185.67±4.73	5.67±0.58	8.67±0.58	967.0±8.19
2%	12	5.0±1.0	6.0±0	1143.33±10.02	5.33±0.58	8.67±1.53	625.33±78.95
	24	5.33±0.58	6.33±0.58	766.33 ± 23.54	-	•	-
	2	4.0±0	8.0±0	1232.33±5.86	3.0±0	8.67±0.58	1159.67±13.05
	4	5.0±1.0	8.67±0.58	952.33±56.59	3.67 ± 0.58	10.33±1.15	1006.0±15.13
	6	5.67±0.58	7.67±1.15	928.37±59.54	4.0±0	6.33 ± 0.58	625.0±40.04
5%	12	5.67±0.58	6.0±0	848.67±10.97	4.33±0.58	6.0±0	342.67±2.08
	24	6.0±0	5.0±1.0	466.33±7.37	-	•	-
	2	6.0±0	8.67±0.55	1023.0 ± 21.70	4.0±0	5.33±0.58	983.67±9.29
	4	5.67±0.58	7.33 ± 0.58	1015.0±5.20	4.0±0	4.67±0.58	272.33±4.16
	6	5.33±0.58	6.0±0	990.33±8.33	5.33±0.58	7.67 ± 0.58	162.67±3.79
10%	12	5.0±0	5.0 ± 1.0	983.67±5.51	-	-	-
	24	5.33±0.58	5.0±0	336.0±4.0	-	-	-
	2	4.0±0	6.0±0	1024.0±4.58	4.0±0	4.0 ± 0	353.33±4.16
	4	4.67±0.58	6.0±0	1024.0 ± 4.58	4.67±0.58	4.67 ± 0.58	247.67±6.51
	6	4.67±0.58	6.0±0	861.0±36.06	7.0 ± 0	7.0±0	229.0±1.73
30%	12	5.0±0	6.0±0	645.33±5.51	-	-	_
	24	5.0±0	6.0±0	449.0±9.90	-	-	_
	2	5.0±0	6.0±0	587.0±33.15	7.67 ± 0.58	7.67 ± 0.58	1062±32.10
	4	5.0±0	6.0±0	237.33±2.57	8.0±0	8.0±0	693.0±5.57
	6	5.0±0	4.33±0.58	182.0 ± 3.61	-	-	_
60%	12	-	-	-	-	-	_
	24	-	-	-	-	-	_
	2	-	-	-	-	-	_
	4	-	-	-	-	-	_
	6	-	-	-	-	-	-
SAT	12	-	-	-	-	-	-
	24	-	-	-	-	-	_

Table 2: Effects of various concentrations of sodium chloride solution on hatchability of eggs of Rhipicephalus sanguineus

	Time of	Eclosion period (days)	Duration of	Day of peak	Remaining days	
	immersion	(n; %) larvae on	hatching	of hatching	of hatching and	No. of eggs
NaCl conc. (%)	(h)	1st day	(days)	(n; %) larvae	(n; %) larvae	(hatchability %)
Contol	-	21 (187;29.5)	4	2 (285;44.9)	2 (162;24.66)	634 (96.5)
	2	12 (2;0.3)	10	7 (130;19.8)	8 (319;48.6)	518 (78.8)
	4	14 (11;1.7)	8	5 (19;28.9)	6 (248;37.8)	449 (68.3)
2	6	10 (4;0.)	10	4 (129;19.6)	8 (313;47.6)	446 (67.9)
	12	11 (1;0.2)	9	4 (138;21.0)	7 (302;45.9)	441 (67.1)
	24	14 (35;5.3)	8	3 (159;24.2)	6 (129,19.6)	323 (49.2)
	2	12 (35;5.3)	10	4 (120;18.3)	8 (62,9.4)	217 (33.0)
	4	13 (46,7.0)	8	1 (46;7.0)	6 (92;14.0)	184 (28.0)
5	6	14 (12,1.8)	9	3 (56,8.5)	7 (60,9.1)	128 (19.5)
	12	14 (9,1.4)	8	2 (37,5.6)	6 (78;11.9)	124 (18.9)
	24	14 (65,9.9)	7	1 (65,9.9)	5 (49,7.5)	114 (17.4)
	2	13 (23,3.5)	10	3 (87;3.2)	8 (86;13.1)	196 (29.8)
	4	13 (11,1.7)	8	8 (73;11.1)	6 (57;8.7)	141 (21.5)
10	6	14 (13,2.0)	9	5 (68;10.4)	7 (38,6.8)	119 (18.1)
	12	16 (5,0.8)	8	4 (51;7.8)	6 (50,7.6)	106 (16.1)
	24	16 (2,0.3)	6	3 (29,4.4)	4 (40,6.1)	71 (10.8)
	2	10 (14;2.1)	8	2 (28,4.3)	6 (36,5.5)	78 (11.9)
	4	10 (8,1.2)	6	3 (23,3.5)	4 (35;5.3)	66 (10.1)
30	6	12 (18;2.7)	8	1 (18;2.7)	6 (23,3.5)	59 (9.0)
	12	14 (4,0.61)	6	2 (21,3.2)	4 (18,2.7)	43 (6.6)
	24	16 (10;1.5)	4	2 (23,3.5)	2 (7;1.1)	40 (6.1)
	2	14 (3;0.5)	10	6 (7,1.1)	8 (13;1.9)	23 (3.5)
	4	16 (1;0.2)	8	3 (4,0.6)	6 (8;1.2)	13 (2.0s)
60	6	· <u>-</u>	-	-	-	-
	12	-	-	-	-	-

Table 2: Continue

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NaCl conc. (%)	Time of Immersion (h)	Eclosion period (days) (n; %) larvae on 1st day	Duration of hatching (days) (n; %) larvae	Day of peak of hatching (n; %) larvae	Remaining days of hatching and (n; %) larvae	No. of eggs (hatchability %)
	24		` _		<u>-</u>	
	27	=	_	_	-	-
	2	-	-	-	-	-
	4	-	-	-	-	-
Saturated	6	-	-	-	-	-
	12	-	-	-	-	-
	24	-	-	-	<u>-</u>	-

The % hatchability of control, tested against the hatchabilities by Chi-square (χ^2)

Table 3: Effects of various concentrations of sodium chloride solution on hatchability of eggs of Haemaphysalis leachi leachi

		Eclosion period				
	Time of	(days) and (n; %)	Duration of	Day of peak	Remaining days	
	Immersion	(n; %) larvae	hatching	hatching and	of hatching and	No. of eggs
NaCl conc.	(h)	on 1st day	(days)	(n; %) larvae	(n; %) larvae	(hatchability %
Control	-	22 (80; 12.2)	5	2 (248;37.8)	3 (241; 36.7)	569 (86.6)
	2	10 (4; 0.6)	10	5 (130; 19.8)	8 (319; 48.6)	453 (69.0)
	4	9 (120; 18.3)	7	1 (120; 18.3)	5 (78, 11.9)	198 (30.1)
2	6	9 (142; 21.6)	4	1 (142; 21.6)	2 (56; 8.5)	198 (30.1)
	12	11 (2; 0.3)	9	5 (53; 8.1)	7 (98; 14.9)	153 (23.3)
	24	14 (13; 2.0)	1	1 (13; 2.0)	-	13 (2.0)
	2	10 (4; 0.6)	8	3 (66; 10.1)	6 (124; 18.9)	194 (29.5)
	4	10 (1; 0.2)	8	3 (62; 9.4)	6 (142; 21.6)	205 (31.2)
5	6	12 (2; 0.3)	7	7 (43, 6.5)	5 (145; 22.1)	190 (28.9)
	12	12 (7; 1.1)	7	4 (57, 8.7)	5 (96; 14.6)	160 (24.4)
	24	12 (1; 0.2)	8	5 (27; 4.1)	6 (93; 14.2)	121 (18.4)
	2	14 (14; 2.1)	8	5 (85; 12.9)	6 (111; 16.9)	210 (32.0)
	4	10 (2; 0.3)	6	4 (27, 4.1)	4 (33; 5.0)	62 (9.4)
10	6	13 (4; 0.6)	4	3 (26l; 3.9)	2 (27; 4.1)	57 (8.7)
	12	14 (4; 0.6)	4	3 (12; 1.8)	2 (22; 3.4)	38 (5.8)
	24	14 (1; 0.2)	2	1(1, 0.2)	1 (1; 0.2)	2(0.3)
	2	15 (3; 0.5)	5	3 (25; 3.8)	3 (25; 3.8)	53 (8.1)
	4	12 (21; 3.2)	4	1 (21; 3.2)	3 (18; 2.7)	39 (5.9)
30	6	15 (6; 0.9)	3	3 (10, 1.5)	1 (7; 1.1)	23 (3.5)
	12	13 (3; 0.5)	5	45 (5; 0.8)	3 (4; 0.6)	12 (1.8)
	24	16 (9; 1.4)	1	1 (9; 1.4)	-	9 (1.3)
	2	11 (14; 2.1)	4	1 (14; 2.1)	2 (15; 2.3)	29 (4.4)
	4	11 (11; 1.7)	6	1 (11, 1.7)	4 (14; 2.1)	25 (3.8)
60	6	12 (2; 0.3)	2	2 (8, 1.2)	-	10 (1.5)
	12	12 (2; 0.3)	1	1(2; 0.3)	-	2(0.3)
	24	-	-	-	-	-
	2	-	-	-	-	-
	4	-	-	-	-	-
Saturated	6	-	-	-	-	-
	12	-	-	-	-	-
	24	-	-	-	-	-

The % hatchability of control, tested against the hatchabilities by Chi-square (χ^2)

NaCl solution increased with no hatching of eggs in saturated NaCl solution. The eclosion periods were lower than those of the controls. The engorged adult females and eggs of *Rhipicephalus sanguineus* and *Haemaphysalis leachi leachi* did not survive in 60% and saturated sodium chloride solutions.

These findings agree with the observations of Amoo (1984) who reported that adult females and eggs of *Boophilus decoloratus* and *B. geigyi* did not survive in brine solution of 30% and more. Acaricides have two kinds of effect on ticks. Apart from exhibiting a prescriptive toxicity, peculiar to the acaricides being used like cholinesterase inhibition, they cause ticks to discharge their mouthparts and this increases their

vulnerability to grooming or in their impaired physical conditions make them to fall off the host (Wharton and Norris, 1980; Taylor, 2001).

CONCLUSION

Since sodium chloride has hygroscopic properties, it draws water out of engorged female ticks and eggs through osmotic pressure keeping the female ticks from ovipositing and the eggs from hatching. It is therefore, reasonable in this study to suggest that saturated sodium chloride solution has acaricidal properties causing outright mortality of engorged adult females and eggs of *R. sanguineus* and *H. leachi leachi*. However,

further study on the mechanism of action of sodium chloride on these ticks is ongoing in the laboratory.

RECOMMENDATIONS

The implication of this findings is that saturated sodium chloride solution could be an effective mechanism of controling these ticks. To achieve this concentrated Nacl solution could be sprayed into cracks, crevices and surroundings of kernels in place of the usual method of acaricidal spraying of dog kernels. The cost benefit of this method of control is yet to be determined in order to justify its practical use on a large scale. However, it is clear that over-dependence on imported acaricides will drastically reduce in Nigeria and other developing countries if this method is integrated into the overall policy of control of ticks infesting dogs.

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