THE EFFECT OF FLOODING AND SODIUM CHLORIDE ON THE DEVELOPMENT OF *AMBLYOMMA VARIEGATUM* (FABRICIUS, 1974) (ACARINA : IXODIDAE).

BY Georgina DIEN EKPENYONG *

FACTOR
OF DISTRIBUTION
FLOODING
AMBLYOMMA
VARIEGATUM

ABSTRACT : The effect of flooding in water and sodium chloride were investigated to establish ecological factors that limit the widespread distribution of *Amblyomma variegatum* in some localities.

Samples of replete adults, eggs, and larvae of *A. variegatum* were immersed in 500 ml of water for different durations (3, 6, 12, 24, 48, 72, and 96h), then dried and incubated at 25°C, 85 % RH. A second batch was immersed in 500 ml of different sodium chloride concentrations (0, 10, 20, 30, 40, 50, 60, 70, 80, 90 and 100 %) for periods as stated earlier and thereafter incubated. They were observed for preoviposition time, ovipositional pattern and time of egg eclosion.

Results showed that replete females died only after 24 h flooding in water. Eggs were most resistant to flooding. Replete females and larvae were more tolerant to Nacl solution than the eggs.

FACTEURS
DE DISTRIBUTION
IMMERSION
AMBLYOMMA
VARIEGATUM

RÉSUMÉ : Les effets du séjour dans l'eau et dans une solution de chlorure de sodium sur *Amblyomma variegatum* ont été recherchés en vue d'établir quels facteurs écologiques pourraient limiter la très large répartition de cette espèce dans certaines localités.

Des adultes gorgés, des œufs et des larves ont été immergés dans 500 ml d'eau pendant des durées variables (3, 6, 12, 24, 48, 72 et 96 h), puis séchés et mis en incubation à 25° et 85 % RH. Pendant les mêmes périodes de temps, un second lot a été immergé dans une solution de chlorure de sodium (500 ml) à différentes concentrations, (0, 10, 20, 30, 40, 50, 60, 70, 80, 90 et 100 %), puis mis en incubation.

Les résultats montrent que les femelles gorgées ne meurent que 24 h après leur séjour dans l'eau ; les œufs sont les plus résistants. Dans la solution de chlorure de sodium, les femelles gorgées et les larves sont plus tolérantes que les œufs.

INTRODUCTION

The " variegated " tick, *Amblyomma variegatum* is an important parasite of bovines grazing pasture in Nigeria. It is the most numerous and the most common tick found in all the ecological zones of Nigeria. It is economically important not only as a vector of mamy protozoan, bacterial and viral diseases but it also destroys the skin of cattle thereby rendering their hides useless as a valuable source of foreign exchange for the country. It is, threfore, important that attempts be made to reduce this ectoparasite of livestock.

To date, direct counts of the numbers of *A. variegatum* attached to specific body regions of

* University of Ibadan, Ibadan, Nigeria.

cattle are used to assess population levels of the parasitic phases while cloth drag over vegetation is used to assess the population of the non-parasitic phases.

Several experiments have been recorded on the effect of temperature and relative humidity on the development of some ticks e.g. *A. variegatum* (DIPEOLU and OGUNJI, 1980a, b), *A. americanum* (L.) (ROBERTSON *et al.*, 1975; LANCASTER and MACMILLEN, 1955; SONENSHINE and TIGNER, 1969), and *Boophilus decoloratus* and *B. geigyi* (AMMO, 1984).

Very little seems to be known about the effect of flooding on terrestrial arthropods such as ticks (HINTON, 1961). MURRAY and VESTJENS (1967) and DANIEL and CERNÝ (1967) attributed the scarcity or complete absence of ixodid ticks from certain habitats to periods of excessive wetness. SUTHERST (1971) observed that the greatest danger to the eggs of ixodid ticks in the field would be the silting-up of egg masses during rains and noted that flooding would have to be prolonged for more than two days to affect the eggs. Besides, flood causes only a temporary reduction in tick numbers and the long-term effect of rainfall is to create pasture conditions that are highly favorable to tick reproduction. THEILER (1964) opined that the tick egg stage is the most suceptible to dessication and that vegetation cover and rainfall determine tick distribution. Several investigators have demonstrated associations between vegetation type and tick distribution within a given biotype (SONESHINE and TIGNER, 1969; SEMTNER, HOWEL and HAIR, 1971). BALASHOV (1972) said that when replete ticks drop from their hosts, their enlarged body prevents extensive movement from an environment that is sometimes harsh and which can limits longevity. Therefore, the quality of the immediate environment as regards moisture and chemical in the soil will influence development.

The aim of the present experiments is to investigate the effect of some environmental factors such as flooding (rain) and the effect of sodium chloride solution on the eggs, larvae and replete females of *A. variegatum* which are most of the time on the ground during their life cycle.

MATERIALS AND METHODS

1a. *Effect of flooding on replete females, eggs, and larvae of A. variegatum.*

Replete Females

Adult engorged *A. variegatum* females were cleaned after collection from the Veterinary Control Post, Bodija, Ibadan. Fifty (50) engorged females were immersed in a 500 ml beaker containing 250 ml of water and observed at the following intervals, 3 h, 6 h, 12 h, 24 h, 48 h, 72 h and 96 respectively. After each period, 3 females after the preimmersion period, were wiped dry with filter paper and placed in well labelled universel bottles and incubated at 25°C and 85 % relative humidity. They were observed daily to monitor their preoviposition time, ovipositional pattern and time of egg eclosion.

Eggs

About 4,000 *A. variegatum* eggs were put in clean universal bottles which were then filled with water. After 3 h, 12 h, 24 h, 48 h, 72 h, 96 h and 120 h in water, some eggs were removed, dried on filter paper and placed in clean and dry bijou bottles and plugged with cotton wool.

Larvae

About 4,000 *A. variegatum* larvae were also subject to the same treatment as for eggs to determine their viability after immersion in water for various periods.

1b. *Effect of sodium chloride solution (Brine) on the eggs, larvae and replete females.*

Replete females

Engorged *Amblyomma variegatum* females were detached from cattle stationed at the Veterinary Control Post, Bodija, Ibadan. 500 mls of sodium chloride solution of various concentrations of 10 %, 20 %, 30 %, 40 %, 50 %, 60 %, 70 %, 80 %, 90 %, and 100 % were made in ten 500 ml beakers. Twenty replete females were dropped into each sodium chloride solution. After a period of 1 h, 3 h,

6 h, 12 h, 24 h, 48 h, 72 h and 96 h respectively, 3 females were wiped dry, incubated and observed as in 1a.

Eggs

About 4,000 *Amblyomma variegatum* eggs were immersed in universal bottles containing the same concentrations of sodium chloride solution as mentioned above. They were observed for periods varying from 3 h to 96 h. After each prescribed time, a small amount of the immersed eggs was removed, dried with filter paper and placed in dry bijou bottles plugged with cotton wool. The eggs were incubated at 25°C and 85 % RH and observed as in 1a.

Larvae

About 4,000 *A. variegatum* larvae were given the same treatment as recorded for the eggs. They were observed daily to monitor their activities after immersion for various periods in sodium chloride solution as above.

Results

Effect of flooding

Replete females

Table 1 shows the effect of flooding on the reproduction of *A. Variegatum* replete females. Replete females did not oviposit after twenty-four hours flooding in water. The preoviposition period and oviposition period were not too significantly different from those of the control. After prolonged exposure in water, the incubation (pre-eclosion) period was longer but all the eggs laid hatched into larvae.

Flooding time (H)	Average preoviposition period (days)	Average oviposition period (days)	Average incubation period (days)	Percent eclosion (%)
Control	11.5 ± 3.66	30 ± 7.35	60 ± 5.66	100
3	11.5 ± 1.22	34 ± 1.22	65 ± 2.5	100
6	12 ± 1.22	34 ± 1.22	68 ± 1.58	100
12	12 ± 0.707	37 ± 1.58	82 ± 2.55	100
24	12 ± 0.707	38 ± 1.41	82 ± 1.58	100
48 and above	*	*	*	*

* All died after 24 h flooding.

TABLE 1 : *Effect of flooding on the reproduction of A. variegatum* replete females (n = 50 adult females).

Eggs

Table 2 shows the effect of flooding on the eggs of *A. variegatum*. Eggs flooded for up to 96 h in water still hatched into larvae. After 124 h in water, all the eggs died. Eggs flooded for up to 48 h in water all hatched into larvae but after immersion in water for over 72 h mortality of the eggs progressively increased. Incubation period and duration of hatching increased with increasing flooding time.

Flooding time (H)	Average incubation period (days)	Average duration of hatching (days)	Percent eclosion (%)	Mortality
Control	60 ± 5.66	7 ± 0.707	100	—
3	80 ± 1.58	7 ± 0.707	100	—
6	85 ± 2.5	7 ± 0.707	100	—
12	85 ± 1.5	10 ± 0.707	100	—
24	94 ± 1.5	10 ± 0.707	100	—
48	94 ± 1.5	10 ± 0.707	100	—
72	99 ± 1.2	30 ± 6.02	80	20
96	102 ± 1.4	30 ± 2.34	50	50
120 and above	*	*	*	*

* Eggs did not hatch after 96 h flooding.

TABLE 2 : *Effect of flooding on the hatching pattern of the eggs of A. variegatum* (n = 4,000 eggs).

Larvae

Table 3 shows the effect of flooding on the larvae of *Amblyomma* variegatum.

Larvae flooded in water for up to 48 h were still active but when dried and placed in bijou bottles, some mortality recorded. After 72 h flooding, mortality progressively increased and at 96 h, the mortality rate was hundred percent.

Flooding time (H)	Mortality	Comments
Control	0	Very active
3	0	Very active
6	0	Very active
12	10	Death recorded after one month storage
24	20	Death recorded after one month storage in bijou bottles
48	30	Death recorded after two weeks storage in bijou bottles
72	60	Over 60 % died after one week
96	100	All died after 96 h flooding

TABLE 3 : *Effect of flooding on the larvae of A. variegatum* (n = 4,000 larvae).

Effect of sodium chloride solution

Replete females

Table 4 shows the effect of various concentrations of sodium chloride solutions on the oviposition pattern of engorged *A. variegatum* females.

Replete females immersed in 100 % sodium chloride solution swam around for six hours before they died. None oviposited after this treatment.

Solutions between 70 — 100 % sodium chloride were still too toxic to the replete females.

In 60 % sodium chloride solution for one hour, about 50 % of the replete females oviposited. After 1 h in this concentration, all the females died. The average incubation period was 83 ± 2.55 days (control, 60 ± 5.66).

When placed for up to six hours in 50 % sodium chloride solution, about 60 % of the females were still able to lay viable eggs although the preoviposition and incubation periods were longer. In 30-40 % sodium chloride solution oviposition took place when exposed for up to six hours. Preoviposition and incubation periods were longer. 95 % of eggs laid hatched. After 7-12 h of exposure, all females died.

In 10-20 % sodium chloride solution oviposition took place when exposed for up to 24 h. Preoviposition period was long but the incubation period was as in the control. All the eggs laid hatched into larvae.

Normal control without treatment	Sodium chloride concentration (%)	Exposure time (H)	Average preoviposition period (days)	Average oviposition period (days)	Average incubation period (days)	Percent eclosion (%)	Comments
Control	—	—	11.5 ± 3.66	30 ± 7.35	60 ± 5.66	100	
	10	1-24	18 ± 1.22	35 ± 1.0	60 ± 2.55	100	
	20	1-24	18 ± 1.22	35 ± 1.0	60 ± 2.55	100	
	30	1-6	14 ± 1.22	35 ± 0.707	88 ± 2.86	95	Dead
		7-12	—	—	—	0	Dead
	40	1-6	14 ± 1.58	35 ± 1.0	88 ± 2.86	95	Dead
		7-12	—	—	—	0	Dead
	50	1-6	13 ± 1.41	34 ± 1.0	83 ± 2.55	95	Dead
		7-12	—	—	—	0	Dead
	60	1	11 ± 0.707	34 ± 1.22	83 ± 2.55	90	Dead
		2-6	—	—	—	0	Dead
	70-100	1	—	—	—	—	Dead

TABLE 4 : *Effect of sodium chloride solution on A. variegatum replete females* (n = 20 adults per experiment).

Eggs

Table 5 presents the results of the effect of sodium chloride solution on the eggs. In 60-100 % sodium chloride, all the eggs died. Eggs placed for 1h in 50 % sodium chloride solution hatched into larvae. After 1 h they all died.

Concentration of sodium chloride (%)	Exposure time (H)	Incubation (days)	Eclosion (%)	Mortality (%)
Control	—	60 ± 5.66	100	0
10	1-6	250 ± 14.14	20	80
	7-12	340 ± 15.81	5	95
20	1- 6	250 ± 7.07	15	85
	7-72	290 ± 8.98	5	95
30	1-6	100 ± 1.58	25	75
	7-72	200 ± 3.16	5	95
40	1-24	80 ± 1.11	100	0
	25-72	90 ± 0.707	50	50
50	1	100 ± 0.81	50	50
	2-6	*	*	*
60-100	1	*	*	*

* All died before eclosion.

TABLE 5 : *Effect of sodium chloride solution on the hatching pattern of the eggs of A. variegatum* (n = 4,000 eggs per experiment).

In 40 % sodium solution for up to 24 h all the eggs hatched into larvae but the incubation period was longer than in the control. Up to 72 h, the eggs still hatched but there was some mortality.

In 10-30 % sodium chloride solution for up to 72 h the eggs still hatched into larvae but for some reason mortality rate of larvae was very high. The incubation period was very long. At any given exposure time the number of eggs that hatched was very low.

Larvae

Table 6 presents the results of the effect of sodium chloride solution on the larvae. In 70 %-100 % sodium chloride solution, all the larvae died even after 1 h of exposure. In 50 %-60 % sodium

Concentration of sodium chloride %	Exposure time (H)	Mortality (%)	Comments
Control	—	0	Very active
10	1-6	0	Very active
	7-12	50	Activity reduced
	13-72	100	All dead
20-40	7-6	10	Activity reduced slightly
	7-72	100	All dead
50-60	1-6	50	Reduced activity
	7-24	100	All dead
70-100	1-6	100	All dead

TABLE 6 : *Effect of sodium chloride solution on the larvae of A. variegatum* (n = 4,000 larvae per experiment).

chloride solution for up to 6 h, some larvae still remained active but mortality rate was fifty percent. After 12 h in sodium chloride solution all larvae died.

In 20-40 % sodium chloride solution for up to 6 h, a few larvae died but most were active. After 6 h, mortality was high and after 72 h mortality was hundred percent.

In 10 % saline for up to 6 h, all the larvae were alive and active. At 12 h mortality was 50 % and at 72 h they all died.

DISCUSSION

Amblyomma variegatum is a seasonal tick and the replete females, eggs and larvae are more numerous during the rainy season. These stages of the life cycle are therefore at risk of being washed away or destroyed if they drop on or develop in waterlogged soil or soil impregnated with water-soluble chemicals such as sodium chloride. BALASHOV (1972) had shown that replete female ticks are not capable of extensive movements after dropping from their hosts because of their enlarged body. Therefore, the quality of the immediate environment in terms of vegetation cover and moisture will influence the duration of their oviposition and the viability of the

eggs. The implication of this is that if the soil on which they fall is impregnated with sodium chloride or flooded, the replete females will not live long enough to lay their eggs and the eggs may not hatch.

The results obtained from the effect of flooding on *A. variegatum* replete females show that flooding for up to 24 h had no adverse effect on the oviposition of the females. It was observed that after 24 h flooding in water, all females died. This means that if the replete females drop in a waterlogged soil where the water cannot be drained before 24 h, they will not survive. The implication of this is that *A. variegatum* though the most numerous tick in Nigeria, will not be found in some micro-ecological zones though the temperature, relative humidity and rainfall may be adequate for their abundance. DANIEL and CERNÝ (1967), attributed the scarcity or complete absence of *Ixodes ricinus* from certain habitats to periods of excessive wetness. Eggs laid by females submerged in water took a longer time to hatch into larvae. The larvae of *A. variegatum* were observed to be able to withstand flooding for up to twenty-four hours but after this period, mortality gradually increased.

The eggs were observed to be most resistant stage to flooding. They were viable even after flooding in water for ninety-six hours ! Eclosion of eggs into larvae was one hundred percent after 48 h flooding but thereafter mortality progressively increased. Flooding was observed to prolong the incubation (preeclosion) period and the duration of hatching. The result of this experiment confirms SUTHERST's (1971) opinion that flooding would have to be prolonged for at least three days to affect the eggs of *Boophilus microplus*.

The effect of sodium chloride on replete females, eggs and larvae of *A. variegatum* is interesting. It is surprising that this species is so tolerant to sodium chloride solution. The adults and larvae seem to be more tolerant to high concentrations of sodium chloride than the eggs if submerged for short periods. Replete females still lay viable eggs even when placed in sixty percent sodium chloride solution for 1 h. It was observed that forty percent sodium chloride solution was still a good concentration for the eggs (Table 5). It is rather surprising that the highest mortality for the eggs was recorded in 10 and 20 percent saline respectively.

AMOO (1984) has suggested the use of sodium chloride solution as a cheap acaricide for the extermination of *Boophilus* species since the eggs and adult of this species could not survive in sodium chloride solution of 30 % or more. In the present study it was found that sodium chloride solution will not be suitable as an acaricide for *Amblyomma variegatum* since the concentration needed to kill the replete females and immatures will be high enough to be toxic to vegetation and livestock as well. Other new methods of integrated control will have to be investigated.

REFERENCES

AMOO (A. O. J.), 1984. — Field and laboratory studies on the bionomics of *Boophilus decoloratus* (Koch, 1884) and *B. geigyi* (Aeschlimann and Morel, 1965). — Ph. D. Thesis, University of Ibadan.

BALASHOV (Y. S.), 1972. — Blood-sucking ticks (Ixodoidea) : vectors of diseases of man and animals. — Medskaya Parasit., **29** : 313-320.

DANIEL (M.) and CERNÝ (V.), 1967. — To the methods of studying the environmental temperature of the tick *Ixodes ricinus*. — Folia Parasit., **14** : 177-183.

DIPEOLU (O. O.) and OGUNJI (F. O.), 1980a. — Laboratory studies of factors influencing the oviposition and eclosion patterns of *Amblyomma variegatum* (Fabricuis, 1974) females. — Parasitol., **27** : 257-264.

DIPEOLU (O. O.) and OGUNJI (F. O.), 1980b. — Studies on ticks of veterinary importance in Nigeria 11 : Laboratory studies on the oviposition and eclosion patterns of *Amblyomma variegatum* in relation to their state of engorgement and maintenance temperature. — Bull. Anim. Hlth. Prod. Afr., **28** : 260-280.

HINTON (H. E.), 1961. — How some insects especially the egg stages avoid drowning when it rains. — Proc. S. Lond. Ent. Nat. Hist. Soc., 1960 : 138-154.

HUNTER (W. D.) and HOOKER (W. A.), 1907. — Information concerning the North American fever tick with notes on other species. — US. Dept. Agric. Bur. Entomol. Bull., (72).

LANCASTER (J. J.) and MACMILLEN (H. L.), 1955. — The effects of relative humidity on the loan star tick. — J. Econ. Entomol., **48** : 338-339.

MURRAY (M. D.) and VESTJENS (W. J. M.), 1967. — Studies on the ectoparasites of seals and penguins III : The distribution of the tick *Ixodes uriae*. White and the flea *P. megallionicus* Harch. — Aust. J. Zool., **15** : 715-725.

ROBERTSON (A. S.), PATRICK (C. D.), SEMTNER (P. J.) and HAIR (J. A.), 1975. — The ecology and behaviour of the lone star tick (Acarina : Ixodidae) VI : Response of unfed adults to certain environmental parameters. — J. Med. Entomol., **12** : 525-529.

SEMTNER (P. J.), HOWELL (D. E.) and HAIR (J. A.), 1971. — The ecology and behaviour of the lone star tick : The relationship between vegetative habitat type and tick abundance and distribution in Cherokee. — J. Med. Entomol., **8** : 329-335.

SONENSHINE (D. E.) and TIGNER (J. A.), 1969. — Oviposition and hatching in two species of ticks in relation to moisture. — Am. Entomol. Soc., **62** : 628-640.

SUTHERST (R. W.), 1971. — An experimental investigation into the effect of flooding on the ixodid tick *Boophilus microplus* — Oecologia, **6** : 208-222.

THEILER (G.), 1964. — Ecological aspects of tick distribution, vol. 21. — *In* D. H. S. DAVIS ed. *Ecological studies in Southern Africa* ; Monographics Biol., **14** : 284-300.

Paru en Avril 1993.