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**HARD TICK INFESTATION OF CATTLE WITH
SPECIAL REFERENCE TO THEIR TYPING AND
CONTROL BY IVERMECTIN AND NEEM OIL
(ASHOK) IN BENI-SUEF GOVERNORATE**
(With 6 Tables and One Figure)

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**إصابة الأبقار بالقراد الجامد مع إشارة خاصة لتصنيفها ومقاومتها بالايفرمكتين
وزيت النيم (الاشك) في محافظة بني سويف**

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اظهرت النتائج ان نوع القراد الموجود على عدد 28 من الابقار في مزرعه في هذه التجربه كان من نوع بووفيلس انيوليتس. بلغت نسبة التخلص من القراد الجامد في الابقار المصابة حوالى 100% بعد اسبوع من حقنها بعقار الايفومك (ايفرمكتين) 1% بالنسبة للاطوار اليافعة و 81.9% في الحوريات. وانخفضت هذه النسبة الى 98.6% في الاطوار اليافعة وزادت الى 95.8% في الحوريات عند نهاية التجربة (بعد خمسة اسابيع من بداية العلاج). وبلغت نسبة الشفاء من الاطوار اليافعة 95.6% ، 100% و 100% بعد اسبوع من استعمال زيت النيم(الاشك) بالتخفيفات 1.6% ، 302% و 6.4% على الترتيب وكانت في البرقات 96.8% و 95.1% و 100% مع التخفيفات الثلاثة المستعملة من الاسبوع الثانى من العلاج حتى نهاية التجربة (خمسة اسابيع) ولوحظ انخفاض في معدل الشفاء الى 96.8% بعد الاسبوع الثالث في حالة استعمال التخفيف 6.4% لزيت النيم وكان ذلك استثناء من كل النتائج المسجلة طوال التجربة. ولوحظ زيادة معدل التخلص من القراد (اليافع و الحوريات) بزيادة تركيز زيت النيم المستعمل بعد اسبوع واحد من العلاج حيث بدت معدلات الشفاء 95.6% و 98.4% و 100% مع التركيزات : 1.6% و 3.2% و 6.4% على الترتيب (الاطوار اليافعة) و 96.8% و 95.1% و 100% (الحوريات) عند نفس التركيزات. اظهرت النتائج ان تأثير زيت النيم كان اكثر كفاءه ولأطول فتره من الايفومك (الايفرمكتين) 1% حتى نهاية التجربة.

SUMMARY

Typing of ticks among 28 infested cattle revealed that *Boophilus annulatus* was responsible for cattle infestation in a farm in Beni suef. Ivomec injection evidenced curicity percentage of 100% after one week for adult ticks and 81.9% for nymphs. This percentage decreased to 98.6 % for adult ticks and increased to 95.8 % in nymphs at the end of the experiment (5 weeks post treatment). Neem oil curicity percentage after one week for adult ticks was 95.6 %, 98.4 % and 100 % at dilutions 1.6, 3.2 And 6.4 % respectively and for nymphs was 96.8 %, 95.1 % and 100% at the three dilutions respectively from the second week until the end of the experiment (5 weeks) for adult and nymph except the curicity % at the third week post treatment at dilution 6.4% was decreased to 99.8% which considered exception for the whole recorded results and may be due to re-infestation or wrong in application of drug on the animals. Increasing of concentration of Neem oil increased curicity percentage of adult ticks and nymphs after one week of treatment and that revealed in recorded results 95.6%, 98.4% and 100 % at dilution of 1.6, 3.2 and 6.4% respectively for adult and 96, 8, 95.1 and 100% at dilutions of 1.6, 3.2 and 6.4% for nymph respectively. The results revealed that Neem oil was more effective and of longer duration than that of Ivermectin 1% until the end of the experiment.

Key words: *Hard tick, cattle, ivermectin, neem oil*

INTRODUCTION

(*Boophilus microplus* Canestrini, 1887 is one of the most widely distributed tick species and constitutes a major problem for the cattle industry in tropical and subtropical regions of the world. The tick is responsible for severe losses caused by tick worry, blood loss, hide damage, injection of toxins, and diseases transimition (Sabatini *et al.*, 2001; Ducornez *et al.*, 2005). These losses can be minimized by treating the cattle with acaricides. However, continuous use of these agents has led to the problem of resistance in the arthropods (Klafke *et al.*, 2006), while it is also expensive and cause environmental contamination. Therefore, new compounds or original strategies are necessary in order to control this parasite. It was found that extract of some plants have acricidal effects on *Boophilus microplus* and *Rhipicephalus*

appendiculatus (Kaaya *et al.*, 1995). Neem oil 4% was widely studied only for phytophagous pest control (Dimetry and Al Hwary, 1995)

In the present work, we aimed to evaluate the curicity of Neem oil *in vivo* compared with Ivermectin among natural infested cattle with *Boophilus annulatus*.

MATERIALS and METHODS

Chemicals: Ivomec (Ivermectin 1%; product of Merk company, subcutaneous injection at dose rate of 0.2 mg/Kg B.W.

Neem oil 10% (Ashok) made in Germany and examined in the central laboratory insecticides, Ministry. of Agriculture. Egypt and diluted by water to 1.6, 3.2, and 6.4 % for application on infested cattle by spray diluted preparation on animals.

Twenty eight cattle of 2-5 years old (150-300 Kg B.W.) infested with *Boophilus annulatus* ticks were selected in a farm in Beni suef Governorate. The animals were allocated into 5 groups, the first (8 animals) for treatment by injection of ivermectin, the second group (5 animals) for application of Neem oil 1.6 % spray, third group (5 animals) for application of Neem oil 3.2 % spray, the fourth group (5 animals) for application of Neem oil 6.4 % spray and the fifth group (5 animals) kept as control without treatment. and all animals of the experiment were the total number of the farm.

Ticks sp. was identified according to Soulsby (1982) (adult and nymphs) were counted in zero day before treatment as well as at 7th, 14th, 21st and 28th days after treatment.

The stages count was done from the breast, in between the thighs, under the tail and on both sides of the ear and around the eye – these areas were identified accurately to still fixed along the experimental period.

The results were tabulated and the clearance of the animals from ticks under investigation was considered as guide for the drug efficacy.

Curicity percentage was as (mean number of ticks/animals which removed after application of acaricide multiplied by 100) divided on mean number of ticks befor application of acaricide.

The animals in the experiment were kept under observation during the first 6 hours after treatment where any abnormalities in the

site of application or in the general condition of the animals were recorded.

RESULTS

Typing of ticks revealed that *Boophilus annulatus* sp. was responsible for cattle infestations, Fig. 1.

Ivomec injection evidenced curicity percentage of 100% after one week for adult ticks and 81.9% for nymphs. This percentage decreased to 98.6 % for adult ticks and increased to 95.8 % for nymphs at the end of the experiment (5 weeks post treatment) (Table 1).

Neem oil curicity percentage after one week for adult ticks was 95.6 %, 100 % and 100 % at dilutions 1.6. 3.2 And 6.4 % respectively and for nymphs was 96.8 %. 95.1 % and 100 % at the three dilutions respectively. In post treatment from the second week until the end of the experiment (5 weeks) the curicity % for adult and nymph were 100% except the curicity % at the third week post treatment at dilution 6.4% was decreased to 99.6% which was considered exception for the whole recorded results. Increasing of concentration of Neem oil increased curicity percentage of adult ticks and nymphs after one week of treatment and that revealed in recorded results 95.6%. 98.4 And 100 % at dilution of 1.6, 3.2 and 6.4% respectively for adult and 96, 8, 95.1 and 100% at dilutions of 1.6, 3.2 and 6.4% for nymph respectively (Tables 2, 3 and 4).

The results revealed that curicity percent of Neem oil was more effective and of longer duration than that of Ivermectin 1% until the end of the experiment (Tables 2, 3 and 4).

It is worthily to mention that no side effects could be detected on all treated animals either in general health condition or on skin of treated animals, except after further studies on physiological functions of animals under exiperment .in other study.



Fig. 1: *B. annulatus* ticks.

Table 1: Results of using Ivermectin against ticks infestations of cattle
A=adult, N=nymph.

Table 2: Results of using neem oil 1.6% topical application against ticks

Animal No.	Mean No. of ticks/animal after injection of ivermectin											
	Zero Day		7 days		14 days		21 days		28 days		35 days	
	A	N	A	N	A	N	A	N	A	N	A	N
	1	185	30	0	0	0	0	0	0	0	0	0
2	95	45	0	25	0	20	3	15	9	10	10	0
3	280	65	0	0	4	0	0	0	0	0	0	0
4	120	35	0	20	0	25	0	20	0	5	0	10
5	180	5	0	0	0	0	0	0	0	0	0	0
6	290	9	0	0	0	0	0	0	0	0	0	0
7	80	30	0	0	0	0	0	0	0	0	5	0
8	112	28	0	0	0	0	0	15	5	0	0	0
Mean	135.1	30.9	0	5.6	0.5	5.6	0.4	6.3	1.8	1.9	1.9	1.3
Control	170	20	190	30	200	15	225	25	200	24	240	30
Mean % of curcity			100	81.9	99.6	81.9	99.7	79.6	98.7	30.9	98.6	95.8

infestation of cattle.

Animal No.	Mean No. of ticks/animal after application of Neem oil 1.6 %											
	Zero day		7 days		14 days		21 days		28 days		35 days	
	A	N	A	N	A	N	A	N	A	N	A	N
9	110	45	0	9	0	0	0	0	0	0	0	0
10	180	60	15	5	0	0	0	0	0	0	0	0
11	155	33	0	0	0	0	0	0	0	0	0	0
12	36	18	10	8	0	0	0	0	0	0	0	0
13	20	11	0	0	0	0	0	0	0	0	0	0
mean	114.2	33.4	5	4.4	0	0	0	0	0	0	0	0
Control mean	170	20	190	30	200	15	225	25	200	24	240	30
% of curicity		95.6	96.8	100	100	100	100	100	100	100	100	100

Table 3: Results of using Neem oil 3.2 % as a topical application against tick's infestation of cattle.

Animal No.	Mean No. of ticks/animal after application of Neem oil 3.2%											
	Zero day		7 days		14 days		21 days		28 days		35 days	
	A	N	A	N	A	N	A	N	A	N	A	N
14	280	70	0	0	0	0	0	0	0	0	0	0
15	39	20	0	10	0	0	0	0	0	0	0	0
16	196	88	0	0	0	0	0	0	0	0	0	0
17	55	3	10	0	0	0	0	0	0	0	0	0
18	66	25	0	5	0	0	0	0	0	0	0	0
mean	127.2	41.2	2	2	0	0	0	0	0	0	0	0
Control mean	170	20	190	30	200	15	225	25	200	24	240	30
% of curicity		98.4	95.1	100	100	100	100	100	100	100	100	100

Table 4: Results of using Neem oil 6.4 % as a topical application against tick's infestation of cattle

Animal No.	Mean No. of ticks/animal after application of Neem oil 6.4%											
	Zero day		7 days		14 days		21 days		28 days		35 days	
	A	N	A	N	A	N	A	N	A	N	A	N
19	270	45	0	0	0	0	0	0	0	0	0	0
20	110	50	0	0	0	0	0	0	0	0	0	0
21	48	26	0	0	0	0	0	0	0	0	0	0
22	96	22	0	0	0	0	2	0	0	0	0	0
23	100	35	0	0	0	0	0	0	0	0	0	0
mean	105	35.6	0	0	0	0	0	0	0	0	0	0
Control mean	170	20	150	30	200	15	225	25	200	24	240	30
%of curicity			100	100	100	100	99.4	100	100	100	100	100

Table 5: Comparison of curicity means of ivermectin and neem 1.6% to the control

Statistics values	Control		Ivermectin		Neem 1.6%	
	A	N	A	N	A	N
mean	204.1	24.5	23.8	8.6	19.7	6.3
±S.E.	10	±10	±22	±4.5	±18.8	±5.4
P			0.001	0.01	0.001	0.01
significance			***	***	***	***

Table 6: Comparison of curicity means of different dilutions of neem oil to the ivermectin.

Statistics values	Ivermectin		Neem 1.6%		Neem 3.2%		Neem 6.4%	
	A	N	A	N	A	N	A	N
mean	22.3	8.6	19.9	6.3	21.5	7.2	17.6	5.9
± S.E.	±22.3	±5.4	±18.8	±5.4	±21	±6.8	±17	±5.9
t-test			0.12	0.3	0.06	0.87	0.2	0.35
P			0.9	0.7	0.46	0.17	0.89	0.73
significance			N.S.	N.S.	N.S.	N.S.	N.S.	N.S.

N.S. =not significance

DISCUSSION

In this study evaluation of the efficacy of Neem oil at different dilutions comparatively with the efficacy of Ivermectin injection which is considered the mostly used against monohost ticks infestation among cattle in vivo (Miller *et al.*, 1999). Some recent studies induced that many plant extracts were widely used against mosquitoes (Arnason *et al.*, 1987; Chaven and Nikam, 1988; Balandrin *et al.*, 1995). Neem oil found to be effective against *Boophilus micropolus* (Willams *et al.*, 1996) and against camels ticks, *Hyalomma anatolicam excavatum* (Abdel-Shafy and Zayed, 2002).

In this study we found that the curicity percentages after one week in case of adult ticks were 100 % and 81.9 % in nymph with Ivomec injection, and this percentage of curicity decreased at the end of experiment (after 35 days) 98.6 % in adult and increased in nymph to 95.8%. Similar results in vitro were recorded by (Fahmy *et al.*, 1996).

Neem oil curicity % after one week was 95.6 %, 96.4 and 100% at dilutions 1.6, 3.2 and 6.4 % Neem oil respectively. In the post treatment, the curicity percent for adult or nymph was 100 % for the three dilutions (from the second week until end of the experiment) except with dilution 6.4 % at the third week the curicity percent

decreased to 99.4 % then became 100 %. These results to some what extent agreed with those of (Abel-Shafy, and Zayed, 2002). They found that significant effect on hatching larvae, unfed larvae and adult ticks to reaching 100% after 15th, 3rd and 15th day post treatment respectively, but no significant effect on moulting rate of fed nymph.

From the above results it could be concluded that Neem oil has high significant effect for the control (animals not treated) as the effect of ivermectin but was not found significant difference between effect of different dilutions of neem oil and ivermectin on *boophilus* ticks and found the duration of neem effect was extended until the end of experiment (five weeks), finally we can use neem as natural and safe insecticide.

REFERENCES

- Abdel-Shafy, S.O. and Zayed, A.A. (2002):* Short communication in vitro acaricidal effect of plant extract of neem seed oil (*Azadirachta indica*) on egg, immature and adult stages of *Hayaloma anatolicom excavatum* (Ixoddoidea: Ixodidae). *Veterinary Parasitology* 106: 89-96.
- Arnason, J.J. Philogene, B.J.R.; Donko, N. and Kubo, I. (1987):* Limonoids from the Meliaceae and Rutaceae reduce feeding growth and Development of *Ostrinia nubilulis*. *Entomol. Exp. Appl.* 43: 22-26.
- Balandrin, M.F.; Klocke, J.A.; Wurtele, S. and Bollinger, W.H. (1995):* Natural plants chemicals source of industrial and medicinal materials. *Science* 228: 1154-1160.
- Chavan, S.R. and Nikam, S. (1989):* Investigation of alkanes from neem leaves larvicidal activity. *J. Pest Sci.* 13: 1-12.
- Chungsamryat, N.; Ratanakreetakul, C.; Jansawan, W.; Naron, C.; Chainarong, R. and Weerapol, J. (1994):* Acricidal activity of plant crude extract to Tropical cattle ticks *Kaset Sort J. Nat. Sci.* 28: 649-660.
- Dimetry, N. and Al Hwary, F.M.A. (1995):* Neem Azal flowers as an inhibitor growth and reproduction in cowpea aphid *Aphis Crccivora Koch.* *J. Appl. Entomol.* 119: 649- 660.
- Ducornez, S.; Barré, N.; Miller, R.J. and de Garine-Wichatisky, M. (2005):* Diagnosis of amitraz resistance in *Boophilus microplus* in New Caledonia with Modified larval packet test. *Vet Parasitol* 130: 285-292.

- Fahmy, M.M.; Mousa, W.M. and El-Ghysh, A. (1996):* Application of new Formulation of Ivermectin (Ivomec)-pour-on) to control of external parasites of farm animals. 4th Sci. Cong. Proc., April 3-6, 1996 Vet. Med. J., Giza. 44, 2: 339-349.
- Kaaya, G.P.; Mwangi, E.N. and Malanza, M.M. (1995):* Acricidal activity of Margaritaria discodea (Ephorbiaceae) plant extracts Against ticks Rhipicephalus appendiculatus and amblyomma variegatum (Ixodidae). Int. J. Acarol. 21: 123-129.
- Klafke, GM.; Sabatini, GA.; Albuquerque, TA.; Martins, JR.; Kemp, DH.; Miller, RJ. and Schumaker, TTS. (2006):* Larval immersion tests with ivermectin in populations of the cattle tick Rhipicephalus (Boophilus) microplus (Acari: Ixodidae) from state of Sao Paulo, Brazil. Vet. Parasitol. 142: 386-390.
- Miller, J.A.; Davery, R.B.; Oehler, D.D.; Pound, J.M.; George, J.I. and Ahrens, E.H. (1999):* Control of Boophilus annulatus (Acari: Ixodidae) on cattle using injectable microspheres containing ivermectin. J. Eco / Entomo. 92: 1142-1146.
- Sabatini, GA.; Kemp, DH.; Hughes, S.; Nari, A. and Hansen, J. (2001):* Tests to determine LC50 and discriminating doses formacrocyclic lactones against the cattle tick, Boophilus microplus. Vet. Parasitol. 95: 53-62.
- Soulsby, E.J.L. (1982):* Helminths, Arthropods and Protozoa of Domesticated Animals, 7th Ed. Bailliere, Tindall and Cassell LTD., London. P. 765.
- Williams, L.A.D. and Ajai-Mansingh, A. (1996):* The Insecticidal and acricidal action of compounds from Azadirachta indica (A. Jun) and their use in tropical pest management. Inter. Pest Manage, Rev., 1, 133-145.

