Evaluation of the Ixodicidal Effect of Neem Essential Oil (Azadirachta Indica) in Domestic Dogs Infested by Ticks (Rhipicephalus Sanguineus) in the Veterinary Clinic Castim from the City of Arenillas Province El Oro

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Abstract

The present investigation was carried out to evaluate the ixodicide effect of Neem Essential Oil (Azadirachta indica) in domestic dogs infested by ticks (Rhipicephalus sanguineus) that were admitted to the CASTIM Veterinary Clinic, through the use of a shampoo based on Neem essential oil. (Azadirachta indica) at two concentration doses of 20% and 30% and with its positive control Derrivante (Ethion¬ + Cypermethrin) at 10%. thus determining: the effectiveness of the concentration of Neem oil, the time range in which the ixodicide effect occurs after application and the comparison of the positive control Derrivante (Ethion + Cypermethrin) at 10% vs. Neem essential oil 20% and 30%. evidencing its effectiveness, promoting the solution to the problem presented by most dogs at the Coastal or Littoral region level. In this research projected a total of 36 dogs male and female patients were analyzed by non-probabilistic consecutive sampling significance level of 0.05% admitted to the Veterinary Clinic CASTIM in a period of four months, classifying them according to size, type of coat (short and long) and their habitat (inside and outside the home) as indicators in which the acaricide efficiency of essential oil of Neem both 20% and 30% with residual effect 15 days were assessed in addition to the commercial product have Derrivante (ethion + Cypermethrin) as a positive control. According to data obtained, Neem Oil 30% had a greater effect in relation to the concentration to 20%, demonstrating the degree of infestation of ticks declined significantly as at 5 observation.

Keywords: Ccypermethrin, Ethion, ticks, Ixodicide, Neem, residual.

1. Introduction

Nowadays, at a global and regional level, the relationship of commitment towards pets, our friends dogs, has been improving, both on the part of: owners in their care and well-being and professionals by making scientific updates, in order to provide detailed information on the various problems that can affect their pets not only through conventional medicine but also through the new non-conventional medicine based on the use of of natural products harmless to both animals and humans. By providing them with this information, we cover the paradigms that often affect the care of pets, knowing that this medicine is proven one hundred percent to be effective with possible short or long-term treatments, thus improving animal and public health indirectly.

The present research was carried out to evaluate the ixodicidal effect of Neem Essential Oil (Azadirachta indica) in domestic dogs infested by ticks (Rhipicephalus sanguineus) that were admitted to the CASTIM Veterinary Clinic, through the use of a shampoo based on Neem essential oil (Azadirachta indica) at two doses of 20% and 30% concentration and with its positive control Derrivante (Ethion + Cypermethrin) at 10%. thus determining: the efficacy of the concentration of Neem oil, the time range in which the ixodicide effect occurs after application and the comparison of the positive control Derrivant (Ethion + Cypermethrin) at 10% vs. Neem essential oil 20% and 30%.

In an in vitro study, they tested the efficacy of Neem against larvae of Rhipicephalus pulchellus, obtaining the oil by compression of cut dried seeds, and preparing dilutions in distilled water at 10, 20, 30 and 40 % (Handule et al., 2002).

Isea - Rodríguez and Hernández, 2013 establish the biocidal potential of Neem, with efficacy in tick control.

In the clinic of small species in warm and tropical climates, canine patients who have tick infestation almost always come, their nutritional and mood deteriorated, and can even lead to death if the disease is not treated in time, with or without the presence of symptoms such as: Anemia, dehydration, fever, vomiting, hemorrhages, bruises, skin problems with inflammation, abscesses, itching in patients and may be facing possible diseases such as: ehrlichiosis, piroplasmosis, borreliosis, among others.

Tick infestation often becomes a problem that can be taken out of the hands of the clinician because a high degree of resistance has been produced in this vector due to the inappropriate application of acaricidal or ixidocidal products such as organophosphates, pyrethroids, amidines, endectocides, phenylpyrazoles, benzoylureas and deltamethrins mainly. The observation and care of the patient by the owner is very important since there are cases in which this arthropod invades even the habitat of the human being, becoming a communicable and harmful disease.

The use and abuse of conventional medicine with chemical products used to control the arthropod alter the normal physiological functioning of the patient and may have side effects such as resistance to the product, allergic reactions or antagonisms that increase the lack of control of the disease, for this reason it is of vital importance to reconnect with nature through our ancestral knowledge. by using alternative medicine with products that do not directly require

traditional medicine but natural medicine and why not use the varieties of flora that nature offers us.

WHO in an article "Health for All in the Year 2000" establishes traditional medicine as an alternative for the cure and prevention of diseases, highlighting the benefits of herbal medicines in human health. (Álvarez 1998).

Tabla N° 1 Esquema de Tratamientos utilizados en la Investigación

Producto	Tratamiento	Dosis	Perros	Tamaño	Pelaje		Hábitat		Aplicación	Tiempo de Acción Terapéutico			
					corto	largo	D	F	días	Días			
Ethión + Ciperm.	T1 10 %	1ml x Lt de agua	12	Mediana 8-15 kg	3	3	3	3	1 día	1	5	10	15
Champú Aceite esencial de Neem	T2 20%	80 ml de gel +20 ml de aceite	12	Mediana 8- 15 kg	3	3	3	3	1 día	1	5	10	15
	T3 30%	70 ml de gel +30 ml de aceite	12	Mediana 8-15 kg	3	3	3	3	1 día	1	5	10	15
TOTAL PACIENTES			36 perros		9	9	9	9					

Fuente: Ramos C., Castillo M., Quizhpi R., Barrionuevo A. 1

In original language Spanish

The Ixodicidal effect of Neem essential oil on ticks of the Ixodidae family is due to its active ingredient according to Decree 1345-2007 which regulates the process, registration of medicines defining the active ingredient as "any matter of vegetable, animal, chemical origin that has properties that constitute a medicine". (Rodríguez 2010).

2. Materials and Methods

To know the ixodicidal effectiveness of Neem essential oil, a shampoo based on 20% and 30% Neem essential oil was applied in an ixodical bath carried out on patients infested by ticks (Rhipicephalus sanguineus) according to their breed size (homogeneous patients - medium breed), coat size (short and long) and the habitat of coexistence inside and outside the home. , with groups of 3 patients per treatment with a single dose of application, giving us a total of 36 patients; a control group was added with application only of the DERRIVANT tickicide (Ethion + Cypermethrin) at 10%, at a dose of 1 ml for each liter of water in a spray bath, with 3 replicates each treatment.

After application, direct observation was carried out on a surface of 10 cm2 at the level of the cervico-thoracic area, using the exploratory field, deductive and descriptive method, establishing the number of ticks by the degree of infestation with its increase or reduction after the application of Neem essential oil and its ixodicidal action at days 1, 5, 10 and 15.

In the Modality and type of Research, the experimental statistical design of Complete Randomized Blocks (DBCA) was carried out with 4 replications, in addition to the analysis of variance for the evaluated indicators and Duncan tests at 5 %, which helped in the interpretation of the data collected and the determination of the best concentration of Neem Essential Oil.

3. Results and Discussion

When evaluating the effect of Neem Oil as Ixodicide at different concentration doses in dogs infested with ticks based on the indicators (Patient Size, Coat Size and Habitat), it is stated that in this experiment the data produced did demonstrate the degree of effectiveness of Ixodicide on patients infested by ticks, obtaining that the concentration with the greatest effectiveness of Ixodicide on ticks is 30% and that if it can be compared with a chemically proven ixodicidal drug such as Derrivant without much difference from each other.

According to Table N°1, it can be seen that Treatment 1 corresponding to Derrivant (Ethion + Cypermethrin) in terms of the degree of infestation there is a considerable decrease in the degree of tick infestation from day 5 of observation in patients without difference in indicators, but there is again a great difference to increase to day 15 of observation, which can be said to depend on the environment in which the patient develops and the residual form of the product, so it is recommended to reapply the product from this stage.

Cuadro N° 1 Evaluación del efecto Ixodicida del Derrivante (Ethión + Cipermetrina) 10 % (T1) Tiempo y grado de infestación de garrapatas en perros

Indicador	Observación		Conteo de Garrapatas antes de la aplicación	Conteo garrapatas después de la Aplicación Día 1	Conteo garrapatas después de la Aplicación Día 5	Conteo garrapatas después de la Aplicación Día 10	Conteo garrapatas después de la Aplicación Día 15
I PELO CORTO	Pcte 1	FC	17 S	17	0	2	5
	Pcte 2	FC	5 L	5	0	0	0
	Pcte 3	FC	3 L	3	0	0	0
II PELO LARGO	Pcte 1	FC	15 S	15	2	0	0
	Pcte 2	FC	6 M	6	0	0	0
	Pcte 3	FC	8 M	8	0	0	0
III DENTRO	Pcte 1		6 M	6	0	0	0
	Pcte 2		5 L	5	0	0	0
	Pcte 3		10 M	10	5	0	0
IV FUERA	Pcte 1		15 S	15	5	0	0
	Pcte 2		23 MS	22	0	2	2
	Pcte 3		5 L	5	0	0	0
Media			9.8 M	9.8 M	1.0 L	0.3	0.6

FC: Fuera de casa, L: Ligero (1-5), M: Moderado (5-10), S: Severo (10-20), MS: Muy severo + 20

Fuente: Ramos C., Castillo M., Quizhpi R., Barrionuevo A.

In original language Spanish

In Figure N° 1,2,3 and 4 we can observe the ixodicidal effect of Derrivant on the time and degree of tick infestation in patients infested with both short and long hair, inside and outside the home;

there is a considerable decrease in the degree of tick infestation from day 5 of observation in patients without difference in indicators. but it increases again on day 15 of observation, and it can be said that it depends on the environment in which the patient develops and the residual form of the product, so it is recommended to reapply the product from this stage.

Figura N° 1 Efecto Ixodicida de Derrivante sobre el Tiempo y grado de Infestación de garrapatas en pacientes infestados de Pelo Corto

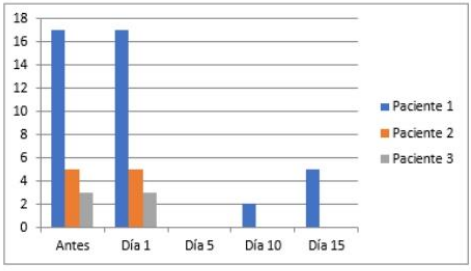


Figura N° 2 Efecto Ixodicida de Derrivante sobre el Tiempo y el grado de Infestación de garrapatas en pacientes infestados de Pelo Largo

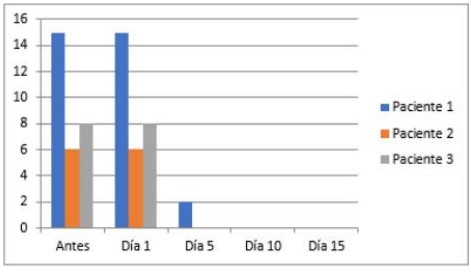


Figura N° 3 Efecto Ixodicida de Derrivante sobre el Tiempo y grado de Infestación de garrapatas en pacientes infestados Hábitat- Dentro de casa

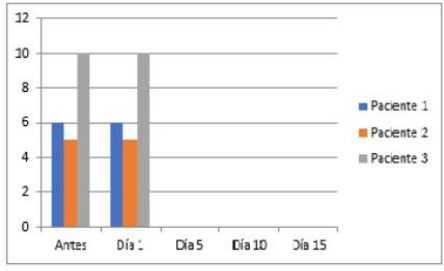
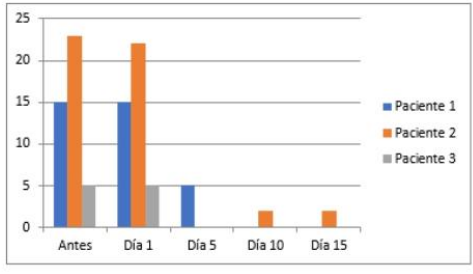


Figura N° 4 Efecto Ixodicida de Derrivante sobre el Tiempo y el grado de Infestación de garrapatas en pacientes infestados Hábitat- Fuera de casa



Fuente: Ramos C., Castillo M., Quizhpi R., Barrionuevo A.

In original language Spanish

In Figure N°5, 6, 7 and 8 it can be seen that Treatment 2 corresponding to 20% Neem Oil does have an Ixodicidal effect on average after 10 days, and as for the degree of tick infestation if there is a decrease, but gradually considering that it is a product of natural origin without toxic effects for the patient and that it acts at the level of salivary glands in the tick, both on day 5, 10 and 15 without re-infestation, its residual effect being less than 15 days of Severe grade reaches a slight degree of infestation. Table No. 2

Figura N° 5 Efecto Ixodicida del Aceite de Neem al 20 % sobre el Tiempo y el grado de Infestación de garrapatas en pacientes infestados de Pelo Corto

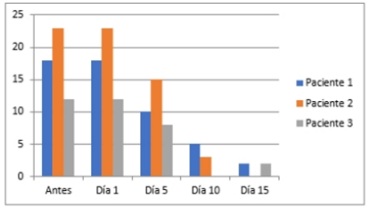


Figura N° 7 Efecto Ixodicida del Aceite de Neem al 20 % sobre el Tiempo y el grado de Infestación de garrapatas en pacientes infestados Hábitat- Dentro de casa

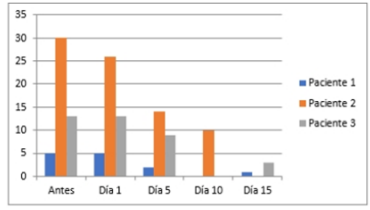


Figura N° 6 Efecto Ixodicida de Aceite de Neem al 20 % sobre el Tiempo y el grado de Infestación de garrapatas en pacientes infestados de Pelo Largo

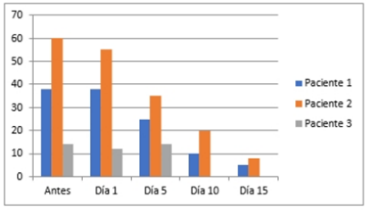
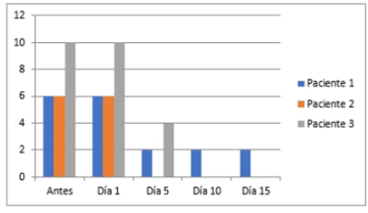


Figura N° 8 Efecto Ixodicida del Aceite de Neem al 20 % sobre el Tiempo y el grado de Infestación de garrapatas en pacientes infestados Hábitat- Fuera de casa



Fuente: Ramos C., Castillo M., Quizhpi R., Barrionuevo A.

In original language Spanish

According to the data obtained in Table N° 3, it can be seen that Treatment 3 corresponding to 30% Neem Oil in terms of the degree of Infestation there is a considerable gradual decrease in the degree of infestation day 15 of observation, being able to say that the Neem Oil with a higher concentration has a greater ixocidal effect on ticks, being able to observe that from Very Severe grade it goes to Light grade and even grade 0 in most experimental units. Figure 9, 10, 11 and 12.

Cuadro N° 3. Evaluación del efecto ixodicida del Aceite de Neem al 30 % (T3) sobre el tiempo y el grado de infestación de garrapatas que infectan a los perros

Indicador	Observación	Conteo de Garrapatas antes de la aplicación	Conteo garrapatas después de la Aplicación Día 1	Conteo garrapatas después de la Aplicación Día 5	Conteo garrapatas después de la Aplicación Día 10	Conteo garrapatas después de la Aplicación Día 15
I PELO CORTO	Pcte 1 FC	48	48	20	0	0
	Pcte 2 FC	4	4	0	0	0
	Pcte 3 FC	39	39	4	2	2
II PELO LARGO	Pcte 1 FC	42	42	30	18	5
	Pcte 2 FC	24	12	6	2	2
	Pcte 3 FC	25	22	6	3	3
III DENTRO	Pcte 1	60	58	9	2	2
	Pcte 2	9	7	2	0	0
	Pcte 3	20	20	11	5	0
IV FUERA	Pcte 1	8	8	3	1	1
	Pcte 2	11	10	4	0	0
	Pcte 3	18	18	12	5	5
Media		25,7	24	8,9	3,2	1,7

Cuadro N° 2 Evaluación del efecto ixodicida del Aceite de Neem al 20 % (T2) sobre el tiempo y el grado de infestación de garrapatas que infectan a los perros

Indicador	Observación	Conteo de Garrapatas antes de la aplicación	Conteo garrapatas después de la Aplicación Día 1	Conteo garrapatas después de la Aplicación Día 5	Conteo garrapatas después de la Aplicación Día 10	Conteo garrapatas después de la Aplicación Día 15
I PELO CORTO	Pcte1 FC	18	18	10	5	2
	Pcte 2 FC	23	23	15	3	0
	Pcte 3 FC	12	12	8	0	2
II PELO LARGO	Pcte1 FC	38	38	25	10	5
	Pcte 2 FC	60	55	35	20	8
	Pcte 3 FC	14	12	14	0	5
III DENTRO	Pcte 1	5	5	2	0	1
	Pcte 2	30	26	14	10	0
	Pcte 3	13	13	9	0	3
IV FUERA	Pcte 1	6	6	2	2	2
	Pcte 2	6	6	0	0	0
	Pcte 3	10	10	4	0	0
Media		19,6	18,7	11,5	4,2	2,3

Fuente: Ramos C., Castillo M., Quizhpi R., Barrionuevo A.

In original language Spanish

Through the results obtained in this research:¨ Evaluation of the Ixodicidal Effect of Neem Essential Oil (Azadirachta Indica) in domestic dogs infested by ticks (Rhipicephalus sanguineus)¨through the clinical histories and observation before and after the application of the Shampoo based on Neem Oil at 20% and 30% in patients who came with tick infestation to the CASTIM Veterinary Clinic in the city of Arenillas, We can say that it was possible to prove that the hypothesis: Neem essential oil (Azadirachta indica) has a good ixodicidal effect on tick-

infested dogs, if it is condoned because Neem Oil is both 20% and 30%. Table No. 4, if it had a good ixodicidal effect on tick-infested dogs in relation to Derrivante, which had almost 100% effectiveness as it was a product tested within commercial houses as it had this ixodicidal faculty. Figure 13, 14 and 16

Figura N° 9 Efecto Ixodicida del Aceite de Neem al 30 % sobre el Tiempo y el grado de Infestación de garrapatas en pacientes infestados de Pelo Corto

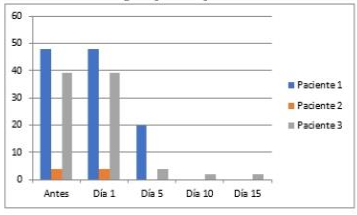


Figura N° 10 Efecto Ixodicida del Aceite de Neem al 30 % sobre el Tiempo y el grado de Infestación de garrapatas en pacientes infestados de Pelo Largo

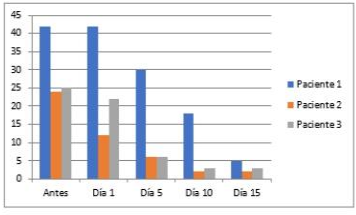


Figura N° 11 Efecto Ixodicida del Aceite de Neem al 30 % sobre el Tiempo el grado de Infestación de garrapatas en pacientes infestados Hábitat- Dentro de casa

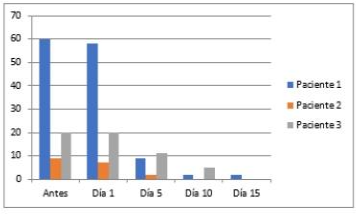
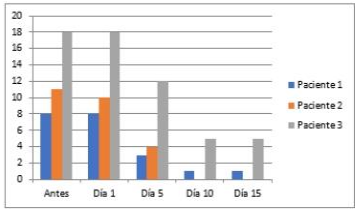


Figura N° 12 Efecto Ixodicida del Aceite de Neem al 30 % sobre el tiempo el grado de Infestación de garrapatas en pacientes infestados Hábitat- Fuera de casa



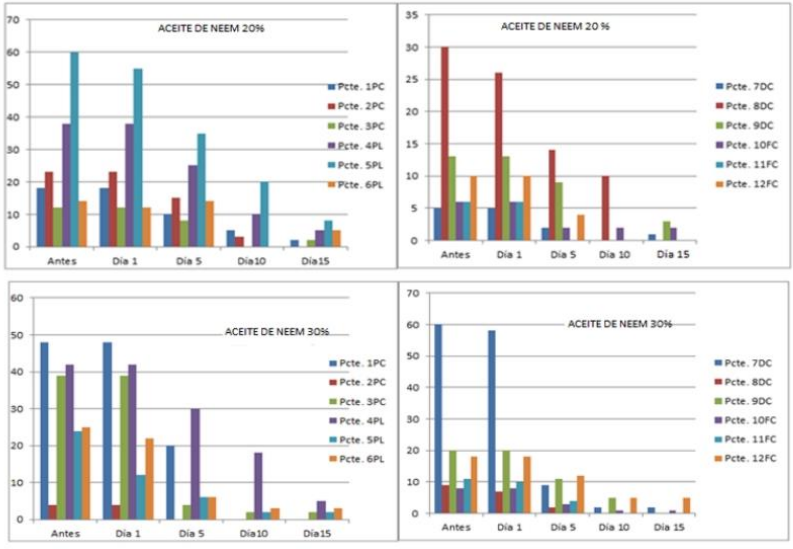
Cuadro N° 4 Evaluación de la concentración de Aceite de Neem (Azadirachta indica) al 20 % vs. Aceite de Neem al 30 % como Ixodicida en perros domésticos infestados por garrapatas

Indicador	Obs.	Garrapatas antes de la aplicación	Día 1	Día 5	Día 10	Día 15	Indicador	Obs.	Garrapatas antes de la aplicación	Día 1	Día 5	Día 10	Día 15
	Pcte 1	18	18	10	5	2		Pcte 1	48	48	20	0	0
I PELO CORTO	Pcte 2	23	23	15	3	0	I PELO CORTO	Pcte 2	4	4	0	0	0
	Pcte 3	12	12	8	0	2		Pcte 3	39	39	4	2	2
	Pcte 1	38	38	25	10	5		Pcte 1	42	42	30	18	5
II PELO LARGO	Pcte 2	60	55	35	20	8	II PELO LARGO	Pcte 2	24	12	6	2	2
	Pcte 3	14	12	14	0	5		Pcte 3	25	22	6	3	3
	Pcte 1	5	5	2	0	1		Pcte 1	60	58	9	2	2
III DENTRO	Pcte 2	30	26	14	10	0	III DENTRO	Pcte 2	9	7	2	0	0
	Pcte 3	13	13	9	0	3		Pcte 3	20	20	11	5	0
	Pcte 1	6	6	2	2	2		Pcte 1	8	8	3	1	1
IV FUERA	Pcte 2	6	6	0	0	0	IV FUERA	Pcte 2	11	10	4	0	0
	Pcte 3	10	10	4	0	0		Pcte 3	18	18	12	5	5
Media		19,6	18,7	11,5	4,2	2,3	Media		25,7	24	8,9	3,2	1,7
		EFECTO IXODICIDA - CONCENTRACIÓN DE NEEM AL 20 %							EFECTO IXODICIDA - CONCENTRACIÓN DE NEEM AL 30 %				

Fuente: Ramos C., Castillo M., Quizhpi R., Barrionuevo A.

In original language Spanish

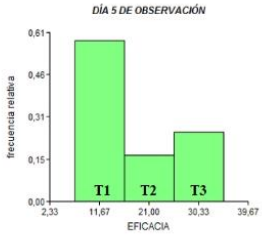
Figura N°13. Evaluación de la concentración de Aceite de Neem (*Azadirachta indica*) al 20 % vs. Aceite de Neem al 30 % como Ixodicida en perros domésticos infestados por garrapatas



Fuente: Ramos C., Castillo M., Quizhpi R., Barrionuevo A.

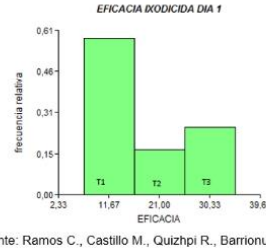
In original language Spanish

Figura N°15 Eficacia Ixodicida del Aceite de Neem al 20 % y 30 % vs. Derrivante



Fuente: Fuente: Ramos C., Castillo M., Quizhpi R., Barrionuevo A. (2016)

Figura N°14 Eficacia Ixodicida del Aceite de Neem al 20 % y 30 % vs. Derrivante



Fuente: Fuente: Ramos C., Castillo M., Quizhpi R., Barrionuevo A. (2016)

In original language Spanish

According to Isea, Rodríguez and Hernández in 2013, in one of their investigations, they applied a spray with an aqueous preparation to 15% of leaves, to calves between 5 and 6 months of age, where he showed that Neem was 68% effective. The positive control (subcutaneous ivermectin, 200 µg/kg) achieved 100% efficacy. Here he counted the number of ticks in the lumbar region (30 cm2) on days 7, 14, 21 and 28, after treatment.

Similarly, in this research, similar results were observed when having a positive control with Derrivant (Ethion + Cypermethrin) with 100% effectiveness in relation to 20% and 30% Neem Oil, obtaining the highest degree of effectiveness with 30% Neem Oil, at the level of the 10 cm2 Cervico-thoracic region with observation days 1, 5, 10 and 15 after treatment

Ernesto Antonio Hurtado, in his research on EVALUATION OF THE AQUEOUS EXTRACT OF NEEM SEED (Azadirachta indica) AS A TICKICIDE IN CATTLE, uses the aqueous extract at concentrations of 4%, 5% and 6% obtaining better results at concentrations of 5% and 6%, where its ixodicidal effectiveness was 87.97% and 88.20% respectively, it is worth mentioning that compared to this research the concentration was much higher and the individual of the study and the species were changed of tick that affects the dog in this case, but nevertheless the results were similar.

Mendoza, N. and Velásquez, J. (2015) who used concentrations of 5%, 10% and 15% of the aqueous extract of Neem seed in cattle with results of 80.48%, 85.21% and 86.99% of tick mortality respectively, similar results were obtained in the present research.

4. Conclusions

When evaluating the Ixodicidal Effect of Neem Essential Oil (Azadirachta Indica) in domestic dogs infested by ticks (Rhipicephalus sanguineus) at the CASTIM Veterinary Clinic, it was determined that, if there is an Ixodicidal effect of Neem essential oil within the patients subjected to the experiment with the gradual decrease in the number of ticks, as well as the change in the degree of infestation of Neem Essential Oil, it was determined that there is an Ixodicidal effect of Neem essential oil within the patients subjected to the experiment with the gradual decrease in the number of ticks, as well as the change in the degree of infestation of the Essential Oil of Neem in the São Paulo Essential Oil. some cases to slight and even reaching grade 0.

When performing the experiment, it was possible to evidence and determine that the 30% concentration Neem essential oil was the most effective with the highest ixodicidal effect unlike the 20% Neem Oil that had its ixodicidal effect, but did not present the expected results, nor its desired effectiveness time.

When assessing the range of time of effectiveness of the treatments, it was obtained that the best treatment within the time range was Neem essential oil at 30% on day 5 of observation, which reduced by 50% the total number of ticks of the patients at the level of the cervico-thoracic area, in addition to presenting a residual effect of 15 days.

The size, coat and habitat of the patients did not influence the effectiveness of the Ixodicidal Neem Essential Oil, because it had its effectiveness gradually, although habitat is an important factor in this research was almost imperceptible in relation to the ixodicidal effect of the product.

When carrying out this research, it was established that when comparing the ixodicidal effect of Derrivante vs. Neem Essential Oil 20%-30% is established that the three treatments have an ixodicidal effect, Derrivant 100%, Neem Oil 30% of 60% effectiveness and Neem Essential Oil 20% of 40% effectiveness on average within the values analyzed.

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