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# Invitro Treatment of Blue Ticks Using a Phytosubstance

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**Abstract:** Ticks are some of the most devastating ectoparasites of livestock causing several tick borne diseases. Management of ticks is increasing getting difficulty using synthetic acaricides due their rapid tolerance to the synthetic acaricides. Botanical acaricides are now used as an alternative management strategy to curb such problems. The blue tick, *Rhiphcephalus (Boophilus) decolaratus*, is a one-host tick that parasitizes cattle and vectors a debilitating protozoan parasite that causes babesiosis. Acaricidal properties of thyme were investigated on the cattle blue tick, *R.B. decolaratus* using various concentrations to establish the lethal dose concentration. The acaridal activity of thyme on ticks was monitored and recorded at two-hour intervals for 48 hours. Tick mortality was observed at all concentrations except the control. The lowest mortality rate was observed for 10 $\mu$ l/cm<sup>3</sup> while 40 $\mu$ l/cm<sup>3</sup> resulted in total tick mortality. The LC<sub>50</sub> was established as 1,9 $\mu$ l/cm<sup>3</sup>. Significant differences (p<0.01) were observed between concentrations, however the mortality was directly proportional to thyme concentration. The thyme oil dehydrated the ticks leading to death within 48 hours. The study thus concludes that thyme oil can be used as a complimentary or alternative acaricide in an integrated livestock program or where organic means are enforced or to as a way of minimizing tolerance of ticks to conventional acaricides.

**Keywords:** Acaricidal, Mortality, Lethal Dose, Thyme Oil, Bioassay

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## 1. Introduction

For a long time, ticks have negatively impacted livestock producers causing a number of tick-borne diseases [1-2]. Ticks are responsible for causing and contributing to a number of different ailments and failure to control the tick population among the herd can lead to unpleasant health consequences for the herd. Without doubt, tick-borne diseases are the major limitation of cattle production [3] perpetuated by climate change pattern that continues to make an increasing amount of habitats suitable for tick distribution, growth and reproduction globally. Tick infestation in the herd can result in a number of tick-borne diseases such as lyme disease, heart water, gall sickness among others [4-5]. Ticks can cause skin irritation and fur loss in individual animals. In Sub Saharan Africa, livestock producers face high costs of production due to frequent use of acaricides. Many governments have provided highly subsidized regular plunge pool cattle dipping with synthetic acaricides like organophosphates, chlorinated hydrocarbon

acaricides and carbamate acaricides which can be effective although other cattle owners opt to carry out individual spray races to manage ticks.

It is now widely accepted that some plants contain phytosubstances that exhibit properties that can repel, kill or alter certain physiological processes of different types of ticks. Some phytosubstances exhibit acaricidal properties hence can be used as sustainable and green methods of managing ticks. These phytosubstances are used alone or in an integrated program with synthetic acaricides. Synthetic acaricides are effective, however pose environmental and health hazards. Furthermore, ticks and mites can quickly develop tolerance to multiple acaricides increasing incidences of tick-borne diseases. In addition, the synthetic acaricides are costly, a huge limiting factor for many resource poor livestock owners in the tropics. In poor countries the dipping frequency is limited due unavailability of acaricides. Where resources are limited, or where organic methods are a preferred option, acaricidal phytosubstances can be recognized as a complimentary approach to the

management of livestock health and to boost livestock production especially at community level. A number of plants contain phytosubstances that have acaricidal properties and among them is thyme (*Thymus vulgaris*) as well as its relatives that contain similar phytosubstances [6].

In vitro tests were carried out to test the acaricidal effects of *T. vulgaris* on the blue tick, *Rhipicephalus (Boophilus) decoloratus*, (a one-host tick) and the most common and damaging tick in Southern Africa [7]. The objectives of this study were i) to determine the lethal concentration (LC<sub>50</sub>) for *T. vulgaris* oil on *R. (B.) decoloratus* and ii) outline the mode of action for *T. vulgaris* oil on *R. (B.) decolataus*

## 2. Materials and Methods

Adult ticks *R. (B.) decoloratus* were collected from Shrewsbury Farm, Beatrice, Zimbabwe (18° 9' 56" S, 30° 57' 59" E, 1370m) in the dry months when the frequency of dipping is low. In this area, cattle have a communal plunge pool dipping system and a cattle dipping, though enforced by law, is predominantly voluntary with some livestock producers often defaulting the dipping schedule. The ticks were collected using tweezers from the soft parts of cattle such as ears, the inner thighs, flanks, forelegs and abdomen. A total of 500 adult ticks were used. The bioassay was carried out on treated filter paper in ventilated petri dishes for 48 hours in a randomized block design in eight treatments with vegetable oil as a blank (Table 1). Twenty adult ticks were placed on sprayed filter paper in the ventilated petri dishes at 25°C, 60±5%RH and 12: 12 L: D photoperiod. The percentage concentration was converted into log concentration (Log (C%)). Ticks were regarded dead if they did not move their legs upon probing with a pin. The proportion of dead ticks was determined using Eq 1:

$$Proportion, P = \frac{No.of\ dead\ ticks}{No.of\ dead\ ticks + No.of\ live\ ticks} \quad (1)$$

The proportion, *P*, ranged from 0.02 to 1.

Due to an uncertainty of a small number of dead ticks in the control and a small number of live ticks at high concentration, only values in the middle of the range were used to calculate the correction factor, Eq 2, to cater for those ticks that could have died independent of thyme oil:

$$Correction\ factor = \frac{Proportion\ in\ treatment - Proportion\ in\ Control}{1 - Proportion\ in\ Control} \quad (2)$$

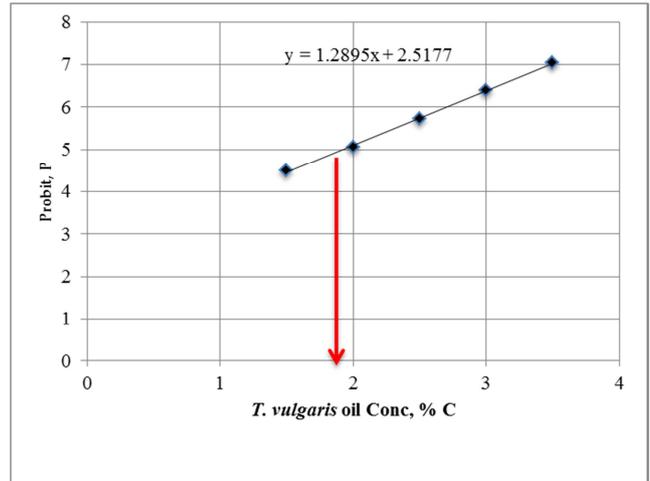
### 2.1. Data Analysis

The LC<sub>50</sub> was calculated using Probit analysis in excel while analysis of variance using R was used to estimate the differences in mean tick mortality.

### 2.2. Results

#### 2.2.1. Lethal Concentration

Within 48 hours, 50% of the ticks were dead as a result of 1.9% concentration of thyme oil (19µl/cm<sup>3</sup>) (Figure 1).



**Figure 1.** Determination of the lethal dose (LC<sub>50</sub>) for *Thymus vulgaris* oil on *Rhipicephalus (Boophilus) decoloratus* at various concentrations. The red arrow indicates the concentration that 1.9% *T. vulgaris* kills 50% of the ticks.

#### 2.2.2. Mean Acaricidal Activity of Thymus Vulgaris Oil

There were significant differences in tick mortality between treatments (*p*<0.001) (Table 1). Tick mortality was evident 15 hours post treatment. The lowest number of ticks was observed in the 10% concentration (10µl/cm<sup>3</sup>) while there was total mortality in the 40% concentration (40µl/cm<sup>3</sup>) 48 hours post treatment.

#### 2.2.3. Effect of Thyme Oil on Ticks

After two hours, ticks in all thyme treatments exhibited a haemorrhagic appearance losing their fluids. Eventually, all ticks lost their fluids and looked dehydrated and did not move when probed with a pin.

**Table 1.** Acaricidal activity of thyme oil (*Thymus vulgaris*) in vegetable oil on *Rhipicephalus (Boophilus) decoloratus* ticks (*n*=100).

Concentrations (µl/cm <sup>3</sup> )	Percent mortality (±SD)	LC <sub>50</sub> (µl/cm <sup>3</sup> )
0	1±0.0	
1.0	25±0.54	
1.5	32±0.58	
2.0	53±0.69	1.88
2.5	77±1.2	
3.0	92±1.3	
3.5	98±1.34	
4.0	100±0.0	

### 2.3. Discussion

The results of this study are similar to the findings of [8-9] while [10] used a number of essential on *Tetranychus cinnabarinus* Boisduval, and achieved remarkable acaricidal effects of the essential oils from medicinal plants. Several authors have documented the use of essentials containing phytosubstances exhibiting acaricidal effects on a number of arthropod pests, for example [10- 20]. This is in response to an increasing tolerance of ticks to various synthetic acaricides [21]. It is imperative therefore to utilize such traditional methods of tick management to maintain livestock production.

The use of vegetable oil to necessitate proper blending in with thyme oil stabilized the thyme oil so it could stay on the

filter paper at least up to the end of the experiment. Vegetable oils have been used in synthetic pesticides such as neem, azadiractin, among others [22-23]. Vegetable oils are ecofriendly posing no health hazards and additionally are easily available affordable and easy to handle.

The use of essential oils such as thyme oil as effective alternative or complimentary acaricides come at a time when livestock production is highly encouraged especially in the African continent where several natural plants containing acaricidal phytosubstances are found. This can improve livestock production by incorporating traditional means to supplement or to work parallel with synthetic acaricides. The mechanisms of acaricide resistance are yet to be well understood. However, the method by which thyme causes tick mortality through dehydration may be difficult for the arthropod pests to tolerate

### 3. Conclusion(s)

This study indicated high acaricidal activity of commercially made thyme oil mixed with vegetable oil. Thyme oil showed high acaricidal activity on *R. (B) decoloratus* ticks and can thus be used as a complimentary or alternative tick control management tool. Thyme oil therefore contains substances that are acaricidal to ticks and possibly to other parasitic arachnids. However it is recommended that further studies be carried to establish other compounds in thyme other than thymol that contribute to the acaricidal nature of the phytosubstance.

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### Conflict of Interest

The authors declare that they have no competing interests.

### Notes/Thanks/Other Declarations

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