

Laboratory Evaluation of Crude Sweet Orange Peel Oil for Acaricidal Effect on Cattle Ticks

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Abstract

The acaricidal effects of crude peel oil from green and ripe sweet orange (*Citrus sinensis*) on cattle ticks were investigated in-vitro. Parameter measured was the mortality rate of the ticks treated with these extracts at six different levels and at 24 hours after treatment. Dilution level had significant effect ($P < 0.05$) on the ticks mortality rate among the investigated extracts except for the Asuntol (the commercial acaricide used as control). The extracts can therefore be used effectively against cattle ticks within the dilution rate of 1:10 to 1:1000.

Keywords: acaricide, cattle ticks, orange peel oil extract, dilution levels, mortality rate.

Introduction

Ticks which are blood sucking arthropods are ectoparasites of domestic and wild animals and are widely distributed in the world. These arthropods parasitize man and animals causing several economic loss and serving as vectors of many infectious diseases.

Ticks affect about 800 million cattle and a similar number of sheep (Robert *et al.* 1982). Hunter (1996) described ticks as wingless acarine group of arthropods with jointed legs, but do not have such clearly defined body components as insects. Moreover, they can be subdivided into hard and soft ticks based on the presence or absence of the hard protective shell on their dorsal surface.

Members such as *Ixodes ricinus* and *Argas spp* are active blood sucking types leading to the anaemic state of the host (Wharton, 1976). A number of ticks are involved in the transmission of protozoal, bacterial, viral and rickettsial diseases. *Boophilus microplus* for example transmits babesis. *Ixodes ricinus* transmits staphylococcus aureus; a bacterium causing tick pyaemia and rhipcephalus transmits the *rickesttia spp* causing tick-borne fever in cattle (Graham and Hourrigan, 1977). The adaptation and mode of life of ticks enable their enormous population to be fairly maintained and their continuous adverse effects in livestock production can not be overlooked.

varieties of chemicals have been used worldwide to control tick. However, Nolan(1979) reported that complete eradication is extremely difficult and the development of resistance against these chemicals(acaricides) has led to the involvement of other ways of tick control. Thompson *et al.* (1978) reported that *Melinis minutiflora* (molasses grass) and *Andropogon gayanus* (gamba grass) can be used for tick control within a marginal tick zone.

Measures such as pasture burning and rotational grazing are equally being used for tick control. In order to safeguard successful tick management for the future, the development of novel active ingredient from local materials is inevitable.

Leaves, barks and root components of many plants have been reported to have medicinal attributes if well prepared and applied so also crude oil from fruits and seeds of many plants (Schnaubelt, 1999)

Young and Foster (1996) reported that the oils of both bitter orange and sweet orange are used in the formulation of herbal teas due to the peel's flavor profiles, their digestive and carminative effects and for the production of emollient, cathartic, carminative and laxative products. Sweet orange peel oil can be used in treating prolapse of the uterus and dysuria, diarrhoea and blood in the faeces and also used as an expectorant and sedative.

Orange peel oil is aromatic but is seldom used except to cover the taste of disagreeable medicines or to lessen their tendency to nausea, and for these

purposes it is frequently added to bitter tinctures and infusions (Blumenthal, 2005).

McHale (2002) reported that *Citrus sinensis* peel oil are readily available and that the oil can be cold pressed from the peel. He further reported that D-Limonene, N-decyllic aldehyde, Linalol, Terpineol and β -carotene are active substances responsible for the unique colour of this orange peel oil.

Wheaton and Stewart (1970) reported that the major chemical in the *citrus sinensis* peel oil is Synephrine and this was reported to be created in the fruits growth in a chemical pathway involving tyramine and N-methyltyramine. Synephrine is alkaloid with similar structure to ephedrine and appear to be present in slightly higher quantities in unripe fruit than in the ripe fruit (Hosda *et al.*, 1990).

The objective of this study therefore was to investigate the acaricidal effect of green (unripe) and ripe sweet orange (*citrus sinensis*) peel oil at different concentrations on cattle ticks in-vitro.

Materials and Methods

Unripe *citrus sinensis* peel oil, ripe *citrus sinensis* peel oil and Asuntol (commercial acaricide) were tested on 1320 fresh live cattle ticks; *Amblyomma variegatum* for 24 hours.

Oil was extracted mechanically from the unripe and ripe *citrus sinensis* by hand squeezing of the peel gotten fresh from the fruits.

Preliminary test was carried out in the first instance as described by Nolan(1979) to know whether there is need for serial dilution or not. This was carried out on

the two crude peel oils and the Asuntol which served as control. 10ml of each extract was pipetted into a bottle each containing 60 fresh live ticks. The bottles were shaken, drained and 20 ticks were transferred into each of 3 Petri-dishes per extract (3 replicates per extract and Asuntol). A strip of filter paper was put inside each Petri-dish to drain the excess extract. All Petri-dishes were tightly covered and observations were taken 24 hours later.

Serial Dilution

The *citrus sinensis* peel extracts and asuntol were diluted serially in ten-fold to give the following dilution ratios: 1:10, 1:100, 1:1000, 1:10,000, 1:100,000, and 1:1,000,000. Asuntol being a common acaricide served as control in this experiment. As done in the preliminary test, 10ml of each of the extracts at different dilution level was pipetted into a bottle containing 60 fresh live ticks. The bottles were shaken, drained and 20 ticks transferred into each of the 3 Petri-dishes per each dilution level of each extract and asuntol (i.e., 3 replicates per extract and Asuntol). A strip of filter paper was put inside to drain excess extract, and were covered and observations made 24 hours later.

Statistical Analysis

All data collected were subjected to statistical analysis using analysis of variance procedure of statistical analysis software (SAS 1999). The treatment means were separated using Duncan option of the same software. Meanwhile,

the experimental model used was *randomized complete Block (RCB)*

Results and Discussion

Table 1 shows the mean mortality of the cattle ticks at 24 hours after treatment with undiluted crude unripe (green) *citrus sinensis* peel oil, ripe *citrus sinensis* peel oil and asuntol. All the ticks in each of the undiluted extracts died after 24 hours thereby confirming the acaricidal properties of citrus oil, and hence the need for serial dilution into different levels.

Table 2 shows the mean mortality of the ticks at 24 hours after treatment with the orange peel oil and asuntol at different concentration. For the unripe *citrus sinensis* peel oil, no significant difference ($P > 0.05$) was observed between the mean mortality of the ticks at concentration levels of 1:10, 1:100, and 1:1000, but observed significantly ($P < 0.05$) with further dilution of 1:10,000, 1:100,000 and 1:1,000,000. However, dilution levels of 1:10, 1:100 and 1:1000 gave the highest mean tick mortality percentage of 95.

Significant difference ($P < 0.05$) was observed within the mean ticks mortality for the ripe *Citrus sinensis* peel oil at different concentrations. Dilution level of 1:10 gave the highest mean mortality percentage of 92.5. Asuntol which served as the control gave the best result closely followed by unripe *citrus sinensis* peel oil while ripe *citrus sinensis* peel oil gave the least results. Significant differences ($P < 0.05$) were observed in the mean tick mortalities between the three extracts

stigated and this cut across the six dilution levels investigated.

pe *citrus sinensis* peel oil has more acaricidal effect on cattle ticks than the other one. This agrees with Blumenthal (1955) who reported that synephrine is more concentrated in unripe *citrus sinensis* peel oil than ripe peel oil.

However, the two orange peel crude oil extracts at the six dilution level gave a ticks mortality rate well above 50% 24 hours after application thereby agreeing with the lethal dose 50 (LD50) concept as earlier recommended by Uilenberg (1970) that effective plant extracts against ectoparasites must lead to at least 50% mortality after 24 hours of application. Asuntol gave the best results at all dilution levels being a refined and tested acaricide, unlike the oil from unripe and ripe *citrus sinensis* peel which is crude and not yet refined.

Conclusion

The results of this in-vitro investigation revealed that oil extract from unripe (green) and ripe *citrus sinensis* peel can be used effectively against cattle ticks when diluted. However, further investigation should be carried out in-vivo to ascertain the effect on ticks on host (cattle) and likely effect on performance characteristics and physiological responses of such cattles.

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Table 1: Preliminary evaluation of unripe (green) and ripe *Citrus sinensis* peel oil for their acaricidal effects on cattle ticks

Extract	% mortality @ 24 hours
Unripe <i>C. sinensis</i> peel oil	100.00
Ripe <i>C. sinensis</i> peel oil	100.00
Asuntol (control)	100.00

Table 2: Effect of crude unripe and ripe Citrus peel oil on cattle ticks at 24 hours different dilution level

Extract	Mean % Tick-mortality						SEM
	1:10	1:100	1:1000	1:10,000	1:100,000	1:1000,000	
Unripe <i>C. sinensis</i>	95.00 ^{ay}	95.00 ^{ay}	95.00 ^{ay}	90.45 ^{by}	90.00 ^{by}	87.55 ^{cy}	2.05
Ripe <i>C. sinensis</i> peel oil	92.50 ^{az}	92.00 ^{az}	91.50 ^{az}	87.50 ^{bz}	84.00 ^{cz}	82.40 ^{dz}	1.20
Asuntol (control)	100.00 ^{ax}	100.00 ^{ax}	100.00 ^{ax}	100.00 ^{ax}	98.50 ^{bx}	94.25 ^{cx}	0.95
SEM	2.05	1.95	3.25	2.10	4.35	3.20	

abcd = means along the same row with the same superscript are not significantly different (P>0.05)
 xyz = means along the same column with the same superscripts are not significantly different (P>0.05)