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COMPARISON OF THE PERFORMANCE OF GARLIC, TOBACCO AND TEPHROSIA AS NATURAL TICK CONTROL BIOCIDES ON MASHONA COWS

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ABSTRACT

In this paper comparison on the use of natural tick control biocides (garlic, tobacco and tephrosia) on Mashona cows using nested design with repeated measures was done. Data was collected for two seasons that is wet season and dry season at Makoholi Research Institute in Masvingo Province in Natural Region IV of Zimbabwe. Model assumptions diagnostic checking was performed and all the assumptions were satisfied. Analysis was done using Genstat version 14 and the results showed that all the biocides perform better than tick-buster with tobacco being the most effective in both seasons when applied after every five days in wet season and ten days in dry season.

Key Words: *Mashona cows, nested design, two-way ANOVA.*

INTRODUCTION

Natural products offer the world's resource-poor farmers a cheaper alternative to synthetic acaricides (Salwa, 2010). These are crude plant material such as leaves, flowers, fruit, seed, stems, wood, bark, roots, rhizomes and other plant parts. The plants are locally available and potentially easy to produce. Farmers are not only the end users of these natural biocides, but they also form the source of the traditional knowledge about the use of these natural products. Their ethno-veterinary and medical knowledge offers a range of herbs to be evaluated for their insecticidal and acaricidal properties. These plants and herbs are known to possess insecticidal, growth inhibiting, ant-moulting and repellent activities.

In Africa livestock is regarded as an "African hoofed bank", whose roles in enhancing gender equity, providing household food security, reducing extreme poverty, maintaining community's cultural life, increasing household income to improve health, education and agriculture, promoting biodiversity, providing material support and mechanical power for farming and many other varied domestic benefits, in rural communities are undoubtedly treasured and indispensable (Smith and Parker, 2010). Most of the Southern African rangelands are inhabited by agro-pastoralists who depend on subsistence production (Masiya, 1996). They are always at the mercy of insects, diseases, predators, drought, floods and other natural disasters. Their livelihoods are mostly dependent on livestock. Among the major constraints to livestock productivity in agro-pastoral areas are ticks and tick borne diseases (TBDs) (Muchenje *et al.*, 2008) and tsetse flies.

The problems of ticks and tick-borne diseases are particularly acute, where acaricides and drugs for managing tick-borne diseases are either unavailable or are far too expensive for the smallholder poor rural livestock farmers. Conventional methods or approaches to livestock tick control have been noted only to confer partial control, poisoning of livestock and non-target living organisms by the acaricides in use, reduction and removal of enzootic stability amongst indigenous livestock species (Mashona cattle) frequently exposed to acaricides, residual environmental pollution resulting from the over use of acaricides, high cost of imported acaricides, lack of professionally trained personnel to supervise the dipping processes and resistance of pertinent parasitic protozoa to drugs used for treating tick-borne diseases, have been reported with no eminent solutions.

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The targeted smallholder poor rural livestock farmers constitute the majority of the livestock farming communities on the African continent (Kariuki, 1996), hence the need to develop new novel methods and approaches suitable for local and native conditions and smallholder poor rural livestock farmers. Under these circumstances therefore, the local communities' anti-tick ethno-knowledge is envisaged to provide a basis for identifying and applying potentially useful, community-driven and specific tick control and management interventions that minimize tick resistance problems, environmentally friendly, affordable, easily accessible and safe when handling and the local end user livestock farmer is the expert personnel (Minja, 1994).

In Zimbabwe, livestock ticks, tick-borne diseases and related secondary infections adversely affect animal health and impede the development of livestock industry, which is the major mainstay of the economy in rural areas. Ticks can be controlled using rotational grazing, controlled burning of grass before the onset of the rains, regular dipping using recommended dips for example Tick Buster, Tritix, and Amitraz and applying tick grease on hidden parts of the animal's body. The methods above can be used for controlling ticks though they have side effects and some of them are not applicable in the rural Zimbabwe set up. For instance, rotational grazing is difficult to practice because of shortage of grazing lands in rural Zimbabwe. Controlled burning of grass in rural Zimbabwe will result in veld fires thereby disturbing the ecosystem. Use of chemicals such as Amitraz is difficult since they are expensive.

The effects of ticks include inflammation, itching and swelling at the bite site, anaemia, irritation and tick worry leading to reduced feed intake, emaciation and development of wounds that may serve as sites for secondary infection (Sanelson, 1975). The immune system of the animal infected with ticks drops making it susceptible to many other diseases. Ticks are a problem in dairy production causing significant economic losses mainly in dairy cows (Latif and Jongejan, 2002). The teats are damaged resulting in a reduction in milk yield, which is a cheaper source of protein to rural resource-poor farmers. The loss is more pronounced in the hot humid areas of tropical regions especially during the wet season. Ticks damage the skin thereby reducing hide quality and creating room for the secondary source of infection (Latif and Jongejan, 2002). Ticks compromise veterinary issues because they transmit diseases, produce toxics and cause physical damage to dairy animals (Rajput *et al*. 2006). Ticks can be carriers of pathogens, which they transmit from host to host during blood sucking thereby causing a number of diseases (Lane and Crosskey, 1996). The diseases include Babeosis, Anaplasimosis, Theleriosis, Heart water, East Cost Fever and other diseases of lesser importance causing severe losses to the livestock industry. Parasites are mainly controlled by the use of conventional drugs and these are expensive, unavailable and associated with high risk for many smallholder cattle producers in Zimbabwe.

Ethno-veterinary biocides are cheap, familiar, locally available and easily accessible, and therefore, can be a better alternative. *Tephrosiavogelii* can be used as an insecticide to reduce tick infestation in cattle (Gaskins *et al.*, 1972). The plant is a potential source of rotenone, an important non-residual insecticide. *Tephrosiavogelii*is a multipurpose leguminous plant, native to the tropical countries and has been introduced to Zimbabwe.

Ethno-veterinary biocides differ from region to region, among and within communities and hence it is less systematic, less formalized and not universally recognized as a valid method of diseases and parasites control in cattle. Much of this valuable ethno-veterinary knowledge is being lost and/or replaced by modern techniques. Sharing this knowledge is vital to ensure that it is used and is preserved for the future (Scoones *et al*, 1994).

The interest in using plant-based repellents is on the rise and some plant species have so far been screened for tick repellency (Palsson and Jaenson 1999). (Van der Merwe, 2000) studied the adverse range of indications for which *A. Marlothii* have been used by Tswana people. He noted some contradictions such as its use for both diarrhoea and constipation. Decoctions and infusions of *A. Marlothii* have been used to treat chickens, ataxia, for blood cleaning, blowfly, dystocia, hasten weaning and for tick control (Hutchings *et al.* 1966).

(Zorloni, 2008) evaluated various plant species from southern Ethiopia for tick toxicity and repellency activities. There were no plant extracts which showed both good repellency and toxicity activities. Plants which have been used as acaricides against *Boophilusmicroplus, Rhipicephalushaemaphysaloides and Hylalommaanalicumanatolicum* include extracts of *Cedrusdeodara, Azadirachtaindica*and *embeliaribes* (Maske *et al.* 1996).

Tephrosiavogelii, a plant which grows wild in much of sub-Saharan Africa, has traditionally been used by Samburu and Maasai pastoralists in Kenya to rid their livestock of ticks. Now, with backing from the botany department of the Kenya Museum among others, use of Tephrosia leaf extract as a low-cost acaricide is spreading to farmers in central Kenya, with impressive results.

The efficacies of several concentrations of rope tobacco against the tick *Boophilusmicroplus* on naturally infested Holstein dairy cattle were investigated. The treatments used were Amitraz at 0.025%; rope tobacco aqueous extract (RT) at 1.25%+neutral detergent at 0.5% (3 sprayings with 24 h intervals between them), RT at 1.75%+whitewash at 0.5% (3 sprayings); RT at 5.0%+whitewash at 2.0% (3 sprayings); RT at 3.75%+neutral detergent at 0.5% (one spraying) and the untreated control group. Engorged ticks with a length superior to 4.0 mm were evaluated, before and on days 1, 2, 3, 4, 5, 6, 7 and 14 day after treatment. The mean efficacy was 100.0; 77.5; 22.0; 63.8; 25.3 and 0.0%, respectively. Differences (P<0.05) between treatment with Amitraz and rope tobacco formulations were significant (Olivo, 2009).

Garlic is believed by many pet owners and experts to be an effective means of repelling fleas and ticks, but not all experts agree. Some vets do not believe garlic is a viable means of flea and tick control. Many vets who do believe it is effective recommend using garlic in combination with other natural treatments (such as dietary brewer's yeast and herbal flea powders), oral medications, topical treatments and/or tick collars under the consultation of a veterinarian to avoid the possibility of overdosing. (Elizabeth, 2001)

In this paper we compare the effectiveness of garlic, tobacco and tephrosia as natural tick control biocides on Mashona cows using nested design with repeated measures.

METHODOLOGY

Simple random sampling was used to obtain a representative sample of sixteen (16) cows from a population of 100 cows at Makoholi Research Institute. In order to evaluate the effectiveness of the treatments, and to determine the proper time for application of the treatments, Nested design with repeated measures is used. In this design four (4) different Mashona cows (experimental units) are randomly allocated to each of the four (4) different dip types (treatments) under investigation. Thus there are four replicates which constitute the whole experiment for comparing treatment means. Replication was done to allow the accurate estimation of the experimental error as well as improve the sensitivity of statistical tests for comparing treatment means and also improve the reliability of the estimates of the treatment means. Then twenty-six (26) counts of ticks observed are taken across time on each cow. Table 1 shows the diagrammatic layout of the design.

	TICKBUSTER(1)	GARLIC(2)	TOBACCO(3)	TEPHROSIA(4)	
PERIOD	COW 1 2 3 4	COW 1 2 3 4	COW 1 2 3 4	COW 1 2 3 4	T0TAL
1	Y_{111} … … Y_{141}	Y_{211} … … Y_{241}	Y_{311} … … Y_{341}	Y_{411} … … Y_{441}	$Y_{\bullet\bullet 1}$
2	Y_{112} … … Y_{142}	Y_{212} … … Y_{242}	Y_{312} … … Y_{342}	Y_{412} … … Y_{442}	$Y_{\bullet\bullet 2}$
3	Y_{113} … … Y_{143}	Y_{213} … … Y_{243}	Y_{313} … … Y_{343}	Y_{413} … … Y_{443}	$Y_{\bullet\bullet 3}$
…….	…. …. …. …..	…. …. …. …..	…. …. …. …..	…. …. …. …..	……
K	Y_{11k} … … Y_{14k}	Y_{21k} … … Y_{24k}	Y_{31k} … … Y_{34k}	Y_{41k} … … Y_{44k}	$Y_{\bullet\bullet k}$
TOTAL	$Y_{11\bullet}$ … … $Y_{14\bullet}$	$Y_{21\bullet}$ … … $Y_{24\bullet}$	$Y_{31\bullet}$ … … $Y_{34\bullet}$	$Y_{41\bullet}$ … … $Y_{44\bullet}$	$Y_{\bullet\bullet\bullet}$
MEAN	$\bar{Y}_{11\bullet}$ … … $\bar{Y}_{14\bullet}$	$\bar{Y}_{21\bullet}$ … … $\bar{Y}_{24\bullet}$	$\bar{Y}_{31\bullet}$ … … $\bar{Y}_{34\bullet}$	$\bar{Y}_{41\bullet}$ … … $\bar{Y}_{44\bullet}$	$\bar{Y}_{\bullet\bullet\bullet}$

Table 1: Nested design with repeated measures DIP TYPES

The model is given as: $Y_{ijk} = \mu + \alpha_i + \beta_{j(i)} + T_k + (\alpha T)_{ik} + \mathcal{E}_{ijk}$ where,

- Y_{ijk} –is the % death of ticks at time k for the j^{th} cow assigned the i^{th} dip type.
- μ –the overall population mean and α_i– is the effect of the i^{th} dip type.
- $\beta_{j(i)}$–is the effect of the j^{th} cow in the i^{th} dip type and T_k–is the effect of the k^{th} period.
- $(\alpha T)_{ik}$ – is the interaction effect of the i^{th} dip type and the k^{th} period.
- $\mathcal{E}_{ijk}$ – are the random errors which are assumed to be independent and normally distributed with mean zero (0) and variance (σ^2).
- i–dip type (1, 2, 3, and 4), j–cows (1, 2, 3, and 4) and k–period/counts (1, 2, 3, 4, … ,26)

Analysis of data will be done while basing on the following assumptions

- α_i's, T_k's and (αT_{ik}'s) are assumed to be fixed real constants satisfying the constraints that $\sum_{i=1}^{4} \alpha_i = \sum_{k=1}^{26} T_k = \sum_{i=1}^{4} \alpha T_{ik} = 0$.
- $\beta_{j(i)}$'s – are assumed to be random variables which are independent and normally distributed with mean zero (0) and variance σ_β^2 .
- ε_{ijk}'s – are independent random variables.

Hypotheses to be tested are given below:

1. Dip type by Period Interaction effect.
- H_0: All the $(\alpha T)_{ik}$'s are equal to zero *versus* H_1: Some $(\alpha T)_{ik}$'s are significantly different from zero.
2. Dip type effect.
- H_0: All the α_i's are equal to zero *versus* H_1: Some α_i's are significantly different from zero.
3. Period effect
- H_0: All the T_k's are equal to zero *versus* H_1: Some T_k's are significantly different from zero.

Table 2: Structure of the ANOVA.

Source	SS	$d.f$	MS	F
Dip type	*SS Dip type*	*a - 1*	*MS Dip type*	$\frac{MS\ Dip\ type}{MS\ Error}$
Period	*SS Period*	*t - 1*	*MS Period*	$\frac{MS\ Period}{MS\ Error}$
Period X Dip type	*SS Period X Dip type*	*(a - 1)(t - 1)*	*MSPeriod X Dip type*	$\frac{MSPeriod\ X\ Dip\ type}{MS\ Error}$
Error	*SS Error*	*a(n - 1)(t - 1)*	*MS Error*	…………………….
Total	*SSTotal*	*N - 1*	………………………..	……………………….

Pair-wise comparisons are done and the Hypotheses to be tested are

H_0: $\mu_{i.} = \mu_{i\square.}$ *versus* H_1: $\mu_{i.} \neq \mu_{i.\square}$ for $i \neq i\square$.

The means $\mu_{i.}$ and $\mu_{i\square.}$ are estimated by $\bar{Y}_{i..}$ and $\bar{Y}_{i\square..}$ respectively. Since the model is a mixed effect model, the standard error of $\bar{Y}_{i..}$-$\bar{Y}_{i\square..}$ is given by $\sqrt{2MSCow(Dip\ type)/4k}$ and the Least Significant Difference (LSD) for the pairwise comparisons is given by:

LSD $=t_{a(b-1)}^{\alpha/2}\sqrt{2MSCow(Dip\ type)/bk}$,

where a = 4(treatments), b = 4(cows) and k = 26 (periods).

Thus, the means $\mu_{i.}$ and $\mu_{i\square}$ are significantly different if: $|\bar{Y}i.. - \bar{Y}i\square..| > LSD$

RESULTS AND ANALYSIS

The data obtained was continuous in nature as it measured the number of ticks obtained after applying each of the treatments which include tephrosia, tobacco and garlic using tick buster as a control at each of the standard and reduced intervals. Genstat version 14 was used for data analysis which includeddiagnostic checking of model assumptions (normality, homogeneity of variance and independence of residuals), two-way ANOVA and pair-wise comparison of treatments.

Diagnostic checking for model assumptions

The following results were obtained for diagnostic checking of model assumptions:

Normality assumption

From the histograms of residuals and the normal and half-normal plots in fig. 2 and 3 of data for standard and reduced intervals we can conclude that the residuals are normally distributed since the histograms of residuals arebell-shaped and non-skewedand the normal and half-normal plots are approximately straight lines.

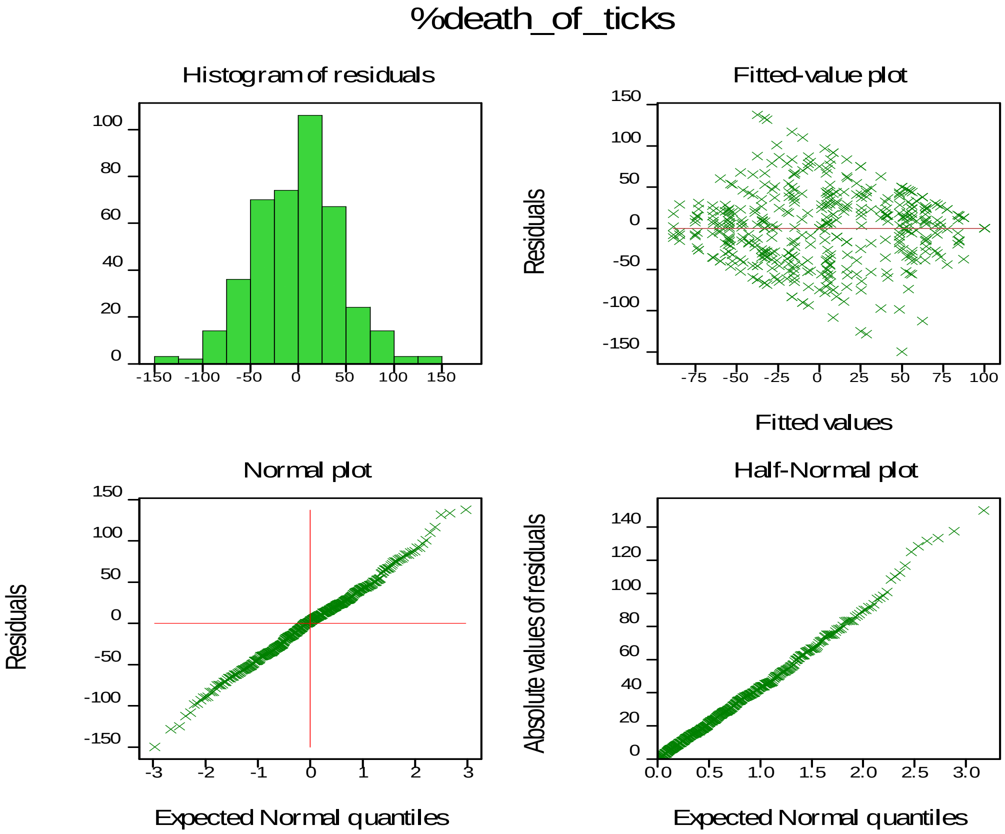

Figure 1: Histogram of residuals and normal plots for data obtained weekly in wet and fortnightly in dry seasons (standard intervals).

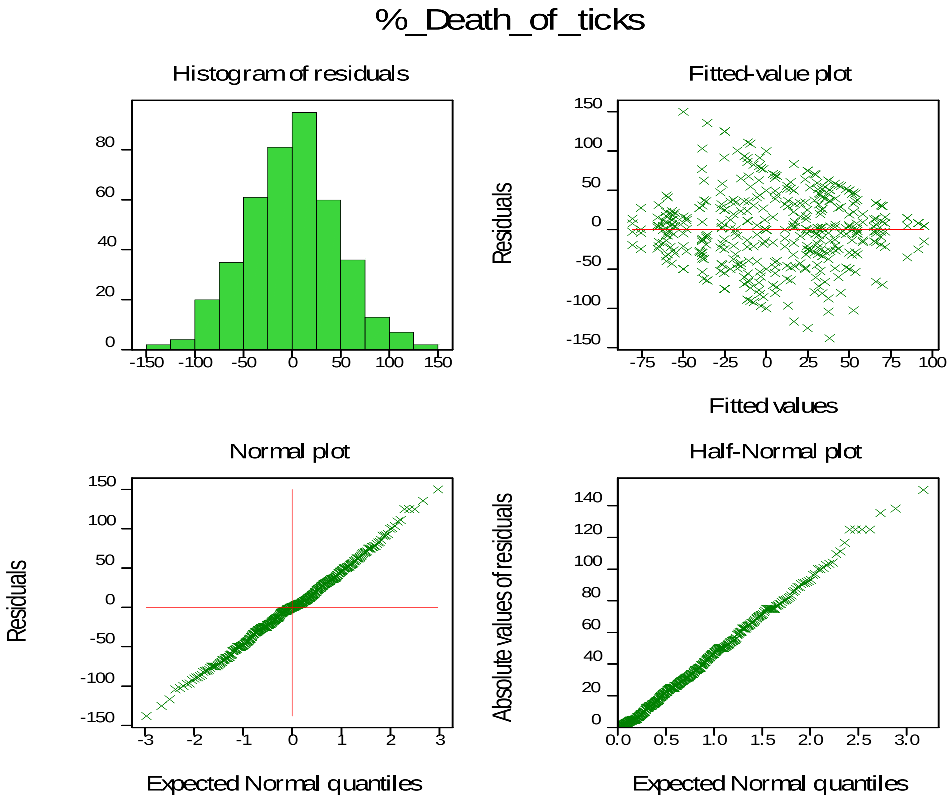

Figure 2: Histogram of residuals and plots for data obtained in the intervals of five days in wet season and in the intervals of ten days in dry season (reduced intervals).

Homogeneity of variance

Based on the plots of residuals against fitted values, the error variance is equal since the plots are pattern-less and the residuals lie within a uniform band. Thus the residuals are random variables with mean zero and a constant varianceσ^2, this is true for both sets of data asillustrated in fig 2 and 3. Thus there is no need for data transformations since the model assumptions are not violated.

Independence of error terms assumption

The plots of residuals against time are pattern-less so we can conclude that the error terms are independent and hence the response variables for both the standard and reduced intervals.

Since the model error assumptions are satisfied by both sets of data, this means that both data sets are adequately described by the specified model.

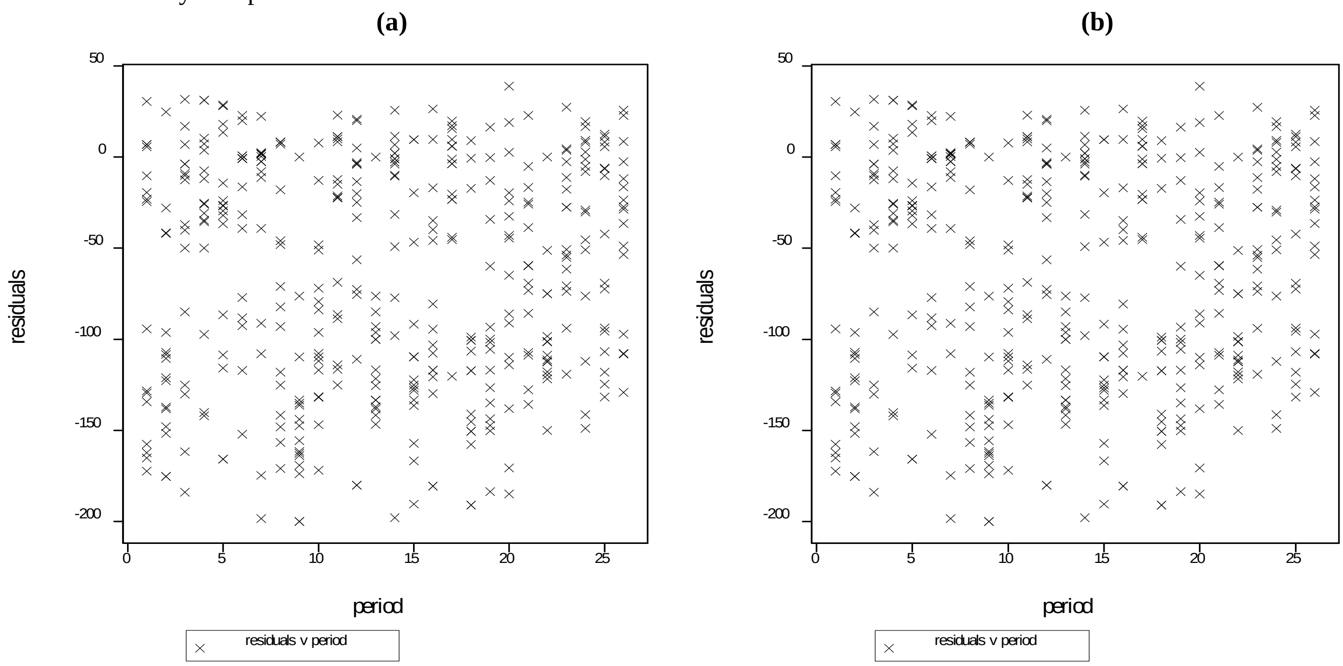

Figure 3: Plot of residuals against time for data obtained (a) weekly in wet season and fortnightly in dry season (b) five days in wet season and ten days in dry season.

Comparing the performance of tobacco, garlic and tephrosia extracts

The two-way ANOVA was used to compare the effect of the four dip types (tick buster, garlic extract, tobacco extract and tephrosia extracts)

Table 3: Analysis of variance (ANOVA): Wet and dry seasons data for the standard dipping intervals.

Variate - (% Death of ticks)

Source of variation	d.f	S.S	M.S	v.r	F pr
Period	25	463582	18543	7.00	<.001
Dip type	3	21087	7029	2.65	0.049
Period*Dip type	75	385355	5138	1.94	<.001
Residual	312	826629	2649		
Total	415	1696654			

Table 4: Analysis of variance (ANOVA): Wet and dry season data for the reduced intervals.

Variate (% Death of ticks)

Source of variation	d.f	S.S	M.S	v.r	F pr
Period	25	398036	15921	5.50	<.001
Dip	3	54750	18250	6.30	<.001
Period * Dip type	75	254157	3389	1.17	0.181
Residual	312	903883	2897		
Total	415	1610826			

The ANOVA results in Tables 3 and 4indicate that the dip effects are significantly different with a p-value of 0.049 for the standard interval and <0.001 for the reduced interval. Pair-wisecomparison of the treatment means was used to determine which dip type should be used on Mashona cows. Table 5 shows the treatment means for the two time intervals.

Table 5: Treatment (dip type) means for standard and reduced dipping intervals.

Dip type	Standard Interval Means	Reduced Interval Means
Tickbuster	14.4	20.6
Garlic	-2.7	2.7
Tobacco	-2.6	9.9
Tephrosia	-0.1	-11.0

Table 6: Absolute difference of the means of the dip types for standard and reduced dipping intervals.

$\lvert\bar{Y}i..-\bar{Y}i'..\rvert$	$\lvert\bar{Y}1..-\bar{Y}2'..\rvert$	$\lvert\bar{Y}1..-\bar{Y}3'..\rvert$	$\lvert\bar{Y}1..-\bar{Y}4'..\rvert$	$\lvert\bar{Y}2..-\bar{Y}3'..\rvert$	$\lvert\bar{Y}2..-\bar{Y}4'..\rvert$	$\lvert\bar{Y}3..-\bar{Y}4'..\rvert$
Standard Interval	17.1	17	14.1	0.1	2.6	2.5
Reduced Interval	17.9	10.7	31.6	7.2	13.7	20.9

The least significant difference (LSD) for dip types for the standard dipping interval is 14.04. The means $\mu_{i.}$ and $\mu_{i'.}$ are significantly different if$\lvert\bar{Y}i..-\bar{Y}i'..\rvert > LSD,$ comparing each of the absolute differences between each of the three biocides and the tick-buster, we conclude that natural biocides are significantly different to the performance of the control (tick-buster). However the performance of natural biocides is the same, since the absolute differences of means between them is less than the LSD.

The least significant difference (LSD) for dip types for the reduced interval is 14.69.The performance of tobacco and tick-buster is the same since 10.7 = $\lvert\bar{Y}1..-\bar{Y}3'..\rvert <$ L.S.D = 14.69 whereas garlic and tephrosia are significantly different to the performance of the control (tick-buster). Thus tobacco can be used as an alternative to tick- buster when applied after every five days in wet season and ten days in dry season.

Testing whether period has an effect in the experiment

One biocide may perform better in wet season as compared to dry season or vice-versa. This knowledge is very useful to farmers so that they are able to use the right biocide at the right time of the year.

From the results in table 3 and 4, the p-value of <0.001 indicates that period effects are significant for both the standard and reduced intervals.

The significance of period effects leads us to test for interaction effects of period and the dip types.

Interaction effect between period (season) and dip type

For the standard interval (Table 3), the dip type and period interaction effects are significantly different as indicated by a p-value of <0.001, while for the reduced interval, the interaction effects are the same since the p-value is 0.181(Table 4).

The explanation of this result is that period or the seasons have something to do with the performance of the biocides. So we would like to know the biocide that can be applied in wet season and that can be applied in dry season for the standard interval. Thus we compare the dip type effect for data from wet season and dry season separately and then use pair-wise comparison to compare the biocides effect in each case.

Comparing the performance of tobacco, garlic and tephrosia extracts using data for wet season.

This helps us to be able to determine the biocide to be used in wet season considering the two cases that is treating weekly or after every five days. The hypotheses to be tested are the null hypothesis that the dip type effects are the same versus the alternative that some dip type effects are significantly different.

Resultsfor the standard interval data in wet season

Table 7: Analysis of variance (ANOVA): Wet season data for the standard dipping intervals.

Variate (% Death of ticks)

Source of variation	d.f	S.S	M.S	v.r	F pr
Period	12	246541	20545	7.31	<.001
Dip type	3	45235	15078	5.36	0.002
Period*Dip type	36	223210	6200	2.21	<.001
Residual	156	438468	2811		
Total	207	953454			

From table 9 above the p-value of 0.002 indicates that in the wet season the biocides are significantly different, thus we need to perform pair-wise comparison in order to come up with a biocide that can be used amongst the three natural biocides in the wet season for the standard dipping intervals.

Table 8: Treatment (dip type) means in wet season.

Dip type	1	2	3	4
Means	29.6	-3.7	-1.1	-7.4

Table 9: Absolute difference of the means of the dip types for standard interval data in the wet season

$\lvert\bar{Y}i..-\bar{Y}i□..\rvert$	$\lvert\bar{Y}1..-\bar{Y}2□..\rvert$	$\lvert\bar{Y}1..-\bar{Y}3□..\rvert$	$\lvert\bar{Y}1..-\bar{Y}4□..\rvert$	$\lvert\bar{Y}2..-\bar{Y}3□..\rvert$	$\lvert\bar{Y}2..-\bar{Y}4□..\rvert$	$\lvert\bar{Y}3..-\bar{Y}4□..\rvert$
$\lvert difference\rvert$	33.3	30.7	37	2.6	3.7	6.3

From the results in Table 11 above, we can conclude that the biocide effects are significantly different to the effect of the control (Tick-buster) by using pair-wise comparison. This is because the absolute differences between the means of biocides and that of the control (Tick-buster) are greater than that of the least significant difference (LSD) which is equal to 20.5. However, the absolute difference of the means of biocides amongst themselves is less than the LSD so we can conclude that the performance of the three biocides is the same.

Comparing the performance of biocides using data for dry season

Comparison of the performance of the extracts will help us determine the biocide to be used in dry season considering the two cases that is treating fortnightly or after every ten days.

Results for standard interval data in dry season

Table 10: Analysis of variance (ANOVA): Dry season data for the standard dipping intervals.

Variate (% Death of ticks)

Source of variation	d.f	S.S	M.S	v.r	F pr
Period	12	215215	17935	7.21	<.001
Dip type	3	3798	1266	0.51	0.677
Period*Dip type	36	134200	3728	1.50	0.049
Residual	156	388161	2488		
Total	207	741374			

Since the p-valueis 0,677 (table 15), we conclude that there is insufficient evidence to support that in dry season the biocides are significantly different. Thus all the three biocides can be used for controlling ticks fortnightly in dry season.

CONCLUSION AND RECOMMENDATIONS

Conclusion

Based on the results of the study we can conclude that natural tick control biocides are not very effective when applied weekly in summer and fortnightly in winter when controlling ticks on Mashona cows. However, they proved to be effective when applied after every five days in wet season and every ten days in dry season with tobacco being the most effective amongst the three biocides in both seasons. In the dry season all the biocides when applied after every fortnight qualify to be useful in controlling ticks.

Recommendations

Farmers in rural areas of Zimbabwe and resettlement areas are encouraged to overcome the problem of ticks attack by using Tobacco extract. When controlling ticks using tobacco they are supposed to apply the extract after every five days in wet season and ten days in dry season. Alternatively in wet season they can also use any of the three biocides since they proved to be equally effective.

Further research can be done on the chemical composition of the medicines since this can also be a contributing factor to the effectiveness of the medicines, these medicines can be affected by temperatures and others can be quickly removed by rain water.

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