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An aqueous extract of *Maerua edulis* (Gilg & Ben) DeWolf tuber is as effective as a commercial synthetic acaricide in controlling ticks on cattle *in vivo*

Emmanuel T. Nyahangare[a,b], Brighton M. Mvumi[c,*], Christopher Magona[a], Jacobus N. Eloff[b]

[a] Department of Animal Science, Faculty of Agriculture, University of Zimbabwe, P O Box MP167, Mt Pleasant, Harare, Zimbabwe
[b] Phytomedicine Programme, Department of Paraclinical Sciences, Faculty of Veterinary Science, University of Pretoria, Private Bag X04, Onderstepoort 0110, South Africa
[c] Department of Soil Science & Agricultural Engineering, Faculty of Agriculture, University of Zimbabwe, P O Box MP167 Mt Pleasant, Harare, Zimbabwe

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ABSTRACT

Farmers in Zimbabwe claim that plant extracts of *Cissus quadrangularis, Aloe vera* and *Maerua edulis* are effective in controlling cattle ticks. On-station experiments were conducted at Henderson Research Station to determine the *in-vivo* efficacy of crude aqueous extracts of *Cissus quadrangularis* (succulent stems), *Aloe vera* (succulent leaves) and *Maerua edulis* (leaves and tuber) at concentrations of 15%, 15% and 10% *w/v* respectively, against cattle ticks. An amitraz-based acaricide and water were used as positive and negative controls, respectively. Thirty Mashona steers were allocated to the six treatments in a completely randomised design experiment where each animal was an experimental unit replicated five times. The animals were each sprayed weekly with 5 L of the test or control solutions using a knapsack sprayer after which full body tick counts were recorded every other day for seven weeks. The experiments were conducted between January and February when conditions are optimal for tick development. The *M. edulis* tuber extract was as effective as the amitraz-based commercial acaricide. The other three plants extracts were, however, as ineffective as the negative control (water). *Maerua edulis* tuber plus soapy water-oil extract is effective against cattle ticks and have potential to be developed into an acaricidal product and thus benefit mostly resource-challenged smallholder farmers who cannot afford commercial synthetic acaricides. *In vivo* studies using acaricidal plants are rare.

1. Introduction

For a long time, researchers and farmers have grappled with the negative effects of ticks and tick-borne diseases particularly on the African continent (Estrada-Peña and Salman, 2013). Ticks are important vectors of various parasites including *Ehrlichia (*Cowdria*) ruminatium, Theileria parva* and *Babesia bigemina* (Bissinger and Roe, 2010; Wanzala et al., 2012; Adenubi et al., 2016; Vudriko et al., 2016). These vectors cause diseases that affect cattle productivity and profitability including *Babesiosis* (Red water), *Theileriosis* (January disease), *Cowdriosis* (Heart water) and *Anaplasmosis* (Gall sickness) if not controlled effectively. Effects of ticks are not limited to diseases alone but they also suck blood and can cause ear and teat damage in cattle. Additionally, they can cause tick "worry" and are generally associated with weight loss and reduced productivity of the animals (Kaaya and Knapp, 2003; Estrada-Peña and Salman, 2013).

Economically, tick control programmes can take up a significant proportion of the national fiscus (Taylor, 2001; Kaaya and Knapp, 2003; Bowman et al., 2004; Rajput et al., 2006; Mapholi et al., 2014). Globally, the cost of controlling ticks is estimated to range from 13.9 to 18.7 billion US dollars (Estrada-Peña and Salman, 2013). There are many examples in Africa where tick and tick-borne disease control programmes have used up millions of US dollars from the fiscus (Moyo and Masika, 2009; Leta et al., 2013; Mapholi et al., 2014). It is estimated that the annual cost in US dollars of importing acaricides in different African countries are: Zimbabwe $9.3 million (Perry et al., 1990), Zambia $10 million (Pegram et al., 1988), Kenya $16 million (Tatchell et al., 1986), Tanzania and Uganda $26 million (Kagaruki, 1997; Okello-Onen and Nsumbuga-Mutaka, 1997), and Nigeria $30 million (Dipeolu and Ndungu, 1991). This is not an African problem alone but other countries also suffer the same fate. Brazil's tick control programmes cost approximately 2 billion US dollars in 2000 (Moyo and Masika, 2013). However, current data on the economic impact of ticks and tick-borne diseases are scarce.

Over the years there have been efforts to look for alternative cost-effective tick control remedies like use of ethnoveterinary plants in response to emerging challenges associated with use of the conventional synthetic acaricides (Isman, 1994; Stevenson et al., 2012;

* Corresponding author.
E-mail addresses: mvumibm@agric.uz.ac.zw, mvumibm@hotmail.com (B.M. Mvumi).

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Grzywacz et al., 2013; Khan and Damalas, 2015). These practices have been slowly gaining popularity in many parts of the world particularly in the developing countries (Samie et al., 2010). Traditional practices, especially acaricidal plants, are locally available, affordable and mostly environmentally-benign and therefore can offer a viable alternative or complementary remedy to conventional synthetic-based tick control programmes (Njoroge and Bussmann, 2006; Isman, 2008; Pirali-Kheirabadi et al., 2009; Fouche et al., 2016).

Ethnoveterinary studies have provided databases of plants with claimed acaricidal properties globally (Adenubi et al., 2016). In a limited number of cases, the efficacy has been examined *in vitro*. In Zimbabwe, several surveys and literature reviews indicate that many plant species have been used with potential acaricidal activities (Maroyi, 2012; Ndhlovu and Masika, 2012; Nyahangare et al., 2015; Marandure, 2016). Despite the wide availability of these plant species, there are no products available on the market largely because the comprehensive documented scientific evidence of their efficacy is lacking. It is therefore critically important to provide scientific evidence of the efficacy and safety of traditionally acclaimed acaricidal plants for the benefit of the livestock industry.

In most cases where efforts have been made to validate the effectiveness of acaricidal plants, research has been limited to *in-vitro* laboratory bioassays at the expense of *in-vivo* trials (Adenubi et al., 2016). This is because *in-vivo* experiments are expensive and logistically challenging to carry out and many institutions do not have facilities to do these experiments (Moyo et al., 2009; Santillán-Velázquez et al., 2013). However, live animal *in-vivo* data are crucial because they provide evidence of the efficacy of the plant extracts under field conditions on the animal.

The other challenge is the lack of clear guidance on the registration process of these products as the current regulatory framework was designed for registration of synthetic pesticides (Sola et al., 2014). In many countries, it is a legal requirement to have evidence of *in-vivo* activity of a particular product and this explains why there are not many plant-based acaricidal products on the formal markets.

In the current study, the acaricidal efficacies of *Cissus quadrangularis* (L.) (Vitaceae), *Maerua edulis* (Gilg & Ben) DeWolf (Capparaceae) and *Aloe vera* (Barbadensis Miller) (L.) Burm.f. (Xanthorrhoeaceae) were tested against cattle ticks *in-vivo*. These plants were selected because they were initially identified by farmers and other stakeholders as acaricidal in a survey conducted in semi-arid cattle producing areas of Zimbabwe. In the survey, the most frequently mentioned plants used against cattle ticks across the surveyed districts, in descending order, were: *C. quadrangularis* (30.1%), *Lippia javanica* (Burm.f.) Spreng. (Verbenaceae) (19.6%), *Psydrax livida* (Hiern) Bridson (Rubiaceae) (14.9%) and *Aloe* sp. (14.9%) (Nyahangare et al., 2015). It was established from farmers that normally, these plants are prepared by crushing and soaking in water overnight and spraying the extract on the animals. *In vitro* preliminary screening of *C. quadrangularis*, *A. vera* and *M. edulis* showed that water extracts of these plants were indeed acaricidal against tick larvae (Chereni, 2014). While *M. edulis* was not ranked highly in the survey, the few respondents who used it, claimed that it was very effective. Literature search also confirmed that in Zambia some preliminary *in-vitro* screening of potentially acaricidal plants showed that *M. edulis* water extract was effective against cattle ticks (Kaposhi, 1992). *Lippia javanica* was not included in the current study because earlier studies confirmed *in-vivo* acaricidal activity of the aqueous extracts (Madzimure et al., 2011). The objective of the current study was therefore to confirm farmer claims of acaricidal efficacy of the selected plants while also validating laboratory efficacy findings (Chereni, 2014), under farm conditions.

2. Materials and methods

2.1. Study site

The study was carried out at Henderson Research Station (17° 35′ S, 30° 58′ E) in Mazowe district about 32 km north east of Harare. The station is in natural farming region II which receives an average annual rainfall of 750–1000 mm. Peak tick infestation occurs during the wet summer months between January and March and the trial was conducted in January and February of 2016. The most common tick species found in the area include *Rhipicephalus (Boophilus) microplus* Canestrini (Acari: Ixodidae), *Rhipicephalus evertsi* Neuman (Acari: Ixodidae), *Rhipicephalus appendiculatus* Neuman (Acari: Ixodidae), *Hyalomma* spp. and *Amblyomma* spp. (Madzimure et al., 2011).

2.2. Plant collection and preparation of treatments

Cissus quadrangularis stems and *M. edulis* leaves and tubers were collected from Chiredzi district about 430 km south-east of Harare, while *A. vera* succulent leaves were collected at Henderson Research Station located about 30 km north of Harare. The plants were positively identified by a qualified botanist, Mr Christopher Chapano and voucher specimens deposited at the National Herbarium and Botanic Gardens of Zimbabwe. The voucher specimen records are: *C. quadrangularis* (Nyahangare E6), *M. edulis* (Nyahangare E5) and *A. vera* (Nyahangare E37). The leaves and tubers of *M. edulis* and fleshy stems of *C. quadrangularis* and *A. vera,* were separately crushed and mixed with water containing a 1% w/v detergent (green bar soap) for 24 h to create a 25% g/100 mL stock solution. The green bar soap (Sunlight produced by Unilever Pvt Ltd) is widely available in shops in southern Africa and was added to reduce surface tension of water when applied on the animal bodies. The soap was first pulverized and dissolved in 1 L of the stock solution and then added back to the parent solution. After 24 h, each mixture was filtered through a mutton cloth and sufficient water added to yield 10% extracts v/v of *M. edulis* leaves and tubers and 15% v/v of *C. quadrangularis* and *A. vera*. Vegetable cooking oil (Olivine brand, Olivine Industries Pvt Ltd, Harare, Zimbabwe) was added to each preparation at 2% w/v. The vegetable oil was used as a low-cost measure of maintaining and preserving the acaricidal properties of the plant extracts and to aid in penetrating the tick cuticle. Olive oil is a better product but is not affordable to the intended beneficiaries of these technologies. The concentrations (10% and 15%) were optimal recommendations from earlier laboratory bioassays (Chereni, 2014). The plant-based treatments were compared to a positive control of Triatix® spray (12.5% EC amitraz-based compound manufactured by Ecomed Manufacturing, Belmont, Zimbabwe for Coopers Zimbabwe Private Ltd), applied at the prescribed (label) dilution rate of 0.2% v/v

Table 1
Summary of experimental treatments.

Treatment	Description
1	10% w/v *Maerua edulis* leaves water extract + 1% w/v surfactant + 2% w/v vegetable cooking oil
2	10% w/v *Maerua edulis* tubers water extract + 1% w/v surfactant + 2% w/v vegetable cooking oil
3	15% w/v *Aloe vera* water extract + 1% w/v surfactant + 2% w/v vegetable cooking oil
4	15% w/v *Cissus quadrangularis* water extract + 1% w/v surfactant + 2% w/v vegetable cooking oil
5	Water with 1% w/v surfactant and 2% w/v vegetable cooking oil (Negative control)
6	Triatix® (Positive control; 12.5% EC amitraz-based compound 0.2% v/v

The surfactant was a commercial washing solid soap pulverized and dissolved in 1 L of the stock solution and then added back to the parent solution. The vegetable oil was a commercial brand.