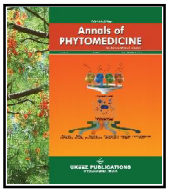
Annals of Phytomedicine 11(2): 725-731, 2022

DOI: http://dx.doi.org/10.54085/ap.2022.11.2.88

Annals of Phytomedicine: An International Journal

http://www.ukaazpublications.com/publications/index.php

Print ISSN : 2278-9839 Online ISSN : 2393-9885



Original Article : Open Access

Phytochemical screening and *in vitro* lousicidal and acaricidal activities of *Justicia schimperiana* (Hochst. ex Nees) T. Anderson leaf in West Showa Zone, Ethiopia

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Article Info

Article history
Received 17 July 2022
Revised 5 September 2022
Accepted 6 September 2022
Published Online 30 December 2022

Keywords
Acaricidal
Amblyomma variegatum
Bovicola ovis
Justicia schimperiana (Hochst. ex Nees) T. Anderson
Phytochemical

Abstract

Ectoparasite infestations, particularly lice and ticks in livestock, are very common in Ethiopia. Even though, *Justicia schimperiana* (Hochst. ex Nees) T. Anderson has been used to treat ectoparasite infestation in Ethiopia, its efficacy has not been validated experimentally in the laboratory. Therefore, the aim of this study was to determine phytochemical constituents as well as lousicidal and acaricidal activity of a methanolic extract of *J. schimperiana* leaf against *Amblyomma variegatum* and lice (*Bovicola ovis*). Fresh and healthy *J. schimperiana* leaves were collected, washed, dried under shade, powdered and extracted with 99.8% methanol using the maceration technique. The phytochemical constituents of the plant were tested using standard laboratory tests. Adult lice and ticks were collected from sheep and cattle, respectively. An experiment was started within 1 h of parasite collection. The extract was checked for its lousicidal and acaricidal activities using adult immersion tests at different time intervals after exposure within 24 h. Diazinon 0.1% and 5% tween 20 were used as positive and negative controls, respectively. All tests were conducted in triplicate. Flavonoids, glycosides, saponins, phenols and tannins were present in the methanolic extract of *J. schimperiana* leaves, but steroids were not. At 24 h post-exposure, 200 and 100 mg/ml concentrations of the plant extract had shown strong lousicidal activities, similar to that of the 0.1% diazinon ($p>0.05$). At 24 h after exposure, the acaricidal activities of 200, 100 and 50 mg/ml were significantly ($p<0.05$) higher than the reference acaricide (0.1% diazinon). Comparatively, all concentrations, the extract showed better activity against lice than ticks. The extract's efficacy increased with increase time after exposure and concentration. The leaf extract of *J. schimperiana* especially at higher concentrations showed a good killing effect against *Bovicola ovis* and *A. variegatum*, suggesting that it could be used as a future alternative to treat lice and ticks infestation.

1. Introduction

Ectoparasites are organisms that live on a host's skin. Ectoparasite infestations are a global problem that creates a serious threat to animal health and productivity (Wall and Shearer, 2001). Ectoparasite infestations cause blood loss, toxicities, irritation and allergic reactions and act as a vector for different diseases (Radostits *et al*., 2000). Ticks and lice are among the common external parasites that are widely distributed in all agro-ecological zones in Ethiopia (Kumsa *et al*., 2012).

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Lice are permanent and host-specific arthropod parasites. It is transmitted from animal-to-animal through contact. Domestic animals, particularly sheep, are infested with lice, which are common ectoparasites. Among lice species, *Bovicola ovis*, biting louse, is an economically important ectoparasite, found throughout most sheep-raising areas of the world. They feed by chewing on the skin surface, which causes irritation and itching with the ultimate outcome of hair loss, downgrading and rejection of skins in tannery industries, as well as decreased production and reproduction (Wall and Shearer, 2001; Kumsa *et al*., 2012).

Ticks are one of the most widespread and harmful blood-sucking ectoparasites of domestic animals across the world (Radostits *et al*., 2000; Walker, 2003). Tick infestations caused toxicities, blood loss, hide damage and produce wounds that provide a route for secondary infection, cause lameness, reduce weight gain and milk production and act as a vector for a number of pathogenic organisms.

In Ethiopia, *A. variegatum*, the tropical tick, is one of the most prevalent and extensively distributed ticks on livestock (Walker, 2003). The control of ectoparasite infestations is mostly based on the use of chemicals. However, the intensive use of such compounds leads to resistance (Yadav *et al*., 2017). Resistance of the ectoparasites to the existing drugs is a serious issue in the treatment and control of ectoparasites especially lice and tick infestations. Due to this, the search for novel lousicidal and acaricidal drugs from medicinal plants is becoming an active area of research. Traditional medicine is an indigenous medicine that is used to maintain human and animal health around the world (Camejo-Rodrigues *et al*., 2003). Traditional medicine is used in Ethiopia to treat 70% of the human and 90% of the livestock population (Boominathan *et al*., 2009). The medicinal value of ethnomedicinal plants depends on their chemical substances or phytochemical constituents which are naturally found in plants (Boominathan *et al*., 2009; Sindhu *et al*., 2021). Medicinal plants have remarkable amount of free radical scavengers such as flavonoids, glycosides, phenols, saponins and terpenoids, The presence of natural antioxidants in plants, living organisms holds superoxide, hydroxyl radical, peroxynitrite and nitric oxide free radicals are in control of number of diseases such as cancer and other diseases, resulting from inflammation (Tamanna Malik *et al*., 2020). *J. schimperiana* (Dhumuuga in Afaan Oromo), also known the synonym *Adhatoda schimperiana*, is a shrub with branched stems that belongs to the family Acanthaceae. *J. schimperiana* is used to treat a variety of diseases in Ethiopia, including malaria (Silva *et al*., 2017), diabetes mellitus (Tesfaye *et al*., 2016), coccidiosis (Birhanu *et al*., 2015), liver disease (Megersa *et al*., 2010), rabies and blackleg (Amenu, 2007), hepatitis, asthma and jaundice, epilepsy (Umer *et al*., 2010). Antidiarrheal activity of 80% methanolic leaf extract of *J. schimperiana* in mice was reported by Mekonnen *et al*. (2018).

Furthermore, ethnobotanical survey in Ethiopia revealed that the *J. schimperiana* plant has been used to treat lice infestations. However, this is a result, the phytochemical constituents as well as evaluates the lousicidal and acaricidal activities of a methanol extract of *J. schimperiana* leaf against *B. ovis* and *A. variegatum*.

2. Materials and Methods

2.1 Description of plant collection area

J. schimperiana was collected from the Toke Kutaye district of West Showa, Oromia regional state, Ethiopia. The district is located 132 kilometres west Addis Ababa. Geographically, it lies between 8 47’ to 9 21 latitudes and 37 32 to 37 03’E longitude and at elevations ranging from 1580 to 3194 meters above sea level. The area receives an annual rainfall of 800 to 100 mm, while the annual temperature ranges from 10 to 29°C. The district has a climatic condition of 18% Kolla, 55% Woina dega and 27% Dega TLFDO (2016).

2.2 Study design

An experimental study in which unsexed adult lice and ticks were assigned treatment and control groups with three replications was conducted to determine the lousicidal and acaricidal activities of a methanolic extract of *J. schimperiana* leaf *in vitro*.

2.3 Plant selection, identification, collection and preparation

The plant was selected based on a preliminary ethnobotanical survey reported by Meragiaw *et al*. (2016). The *J. schimperiana* plant was identified by a taxonomist from the Forestry Department at Ambo University. *J. schimperiana* leaves were collected in December 2020. To reduce possible contamination, latex gloves were worn during plant collection. The collected leaves were transported to the laboratory of the Veterinary Laboratory Technology Department, Guder Mamo Mezemir Campus, and Ambo University. In the laboratory, the collected leaves were washed with to remove soil and dust. Bandiola (2018) spread out on paper sheets and dried under shade for two weeks. The dried leaves were ground using an electric grinder. The powder was weighed using a sensitive digital balance and kept until needed for the extraction. The *J. schimperiana* plant leaves used in this research were collected from the Toke Kutaye district of West Showa. This plant was identified by Biruk Bedore at the Department of Forestry, Ambo University, Ethiopia. The voucher number given for *J. schimperiana* was AUH/192.

2.4 Ethical approval

In this study, naturally infected sheep and cattle with lice and ticks, respectively, were used to collect the study parasites for the *in vitro* study. The Ambo University Animal Scientific Research Ethical Committee (ASREC) assessed the methodology of this study and provided ethical clearance with Ref. No: ASREC/016/20/10/2020 dated October 20, 2020.

2.5 Plant extraction

A total of 1000 g of *J. schimperiana* leaf powder was macerated in 99.8% methanol (100 mg extract 100 ml of methanol) and shaken for 72 h by an automatic orbital shaker (Bandiola, 2018). The liquid part was separated from the herbal residues through a whatman filter paper 1 using an electrical suction pump. Then, the filtrate was concentrated in a vacuum rotary evaporator and dried using an oven at a temperature of 40°C (Bandiola, 2018; Gul, *et al*., 2017; Demisse, 2021). The resulting extract was then transferred into a vial, labelled and the weight of the dried crude extract was determined and a yield percentage was calculated according to Bandiola (2018). The extract was kept at 4°C Celsiusin a refrigerator until required for tests:

$$\text{Yield (\%)} = \frac{\text{Weight of extracts (g)}}{\text{Weight of the plant material (g)}} \times 100$$

2.6 Phytochemical screening

The methanolic extract of *J. schimperiana* leaves was screened to determine the presence or absence of phytochemical constituents such as saponins, tannins, flavonoids, steroids, glycosides and phenols using standard laboratory tests. The qualitative phytochemical analysis results were expressed as (–) for the absence and (+) for the presence of the constituents.

2.6.1 Test for saponins

A foam test was used to determine the presence of saponins in the plant extract. 0.5 g of *J. schimperiana* extract was diluted with 2 ml of distilled water and shaken vigorously. The presence of saponins was detected by the formation of foam that lasted for 10 min (Pandey *et al*., 2014).

2.6.2 Test for tannins

A ferric chloride test was used to check for tannins in the plant extracts. In 5 ml of distilled water, 50 mg of *J. schimperiana* extract

was dissolved. Thereafter, four drops of 5% ferric chloride was added. The presence of tannins was known by the formation of a dark green colour (Bandiola, 2018).

2.6.3 Test for phenols

The phenol test was done using a ferric chloride test. Four drops of concentrated ferric chloride solution were added to 2 ml of extract. The presence of phenols was indicated by the formation of a bluish-black colour (Pandey *et al*., 2014).

2.6.4 Test for flavonoids

The Bate-Smith and Metcalf test was used to determine the flavonoids present in the extract. Concentrated hydrochloric acid (0.5 ml) was added to the extract, which was then boiled in a water bath for 15 min and observed for an hour. The presence of flavonoids in the plant extract was indicated by the formation of a red or violet colour (Bandiola, 2018; Goli Penchala *et al*., 2021).

2.6.5 Test for glycosides

The Liebermann's test was utilized to detect glycosides in the plant extract. 2 ml extract was mixed with 2 ml chloroform and 2 ml acetic anhydride. The presence of glycosides was indicated by the formation of a violet to blue to green, reddish brown ring (Karthikeyan and Vidya, 2019).

2.6.6 Test for steroids

Salkowski test was performed to detect steroids. 5 ml of the plant extract was mixed with 3 ml of chloroform and concentrated H_2SO_4 acid. The appearance of a reddish-brown colour was an indicator of the presence of steroids (Malik *et al*., 2017).

2.7 Collection, transportation and identification of ticks and lice

Lice were obtained from naturally infested sheep purchased from Guder, the Toke Kutaye and transported in plastic bottles covered with cotton net gauze to the Veterinary Laboratory Department, Guder Mamo Mezemir Campus, Ambo University. Identification of the lice was done using a stereoscopic microscope according to the morphological descriptions of Wall and Shearer (2001). In this study, only adult *B.ovis* lice species were used for the *in vitro* test. Adult ticks were collected using forceps from different sites of naturally infested cattle found at Ambo University's, Guder Mamo Mezemir Campus farm, kept in universal bottles and transported to the laboratory, where they were identified using a stereoscopic microscope according to Walker (2003).

2.8 Preparation of working concentrations

The leaf extracts were diluted with 0.5% tween 20. Both the lousicidal and acaricidal efficacy tests used the same concentrations of the extract (200, 100, 50, 25, 12.5 and 6.25 mg/ml). Tween 20 (0.5%) was used as a negative control while 0.1% diazinon was used as a positive control. Positive control, 0.1% diazinon 60 EC was diluted in water according to Heukel Bach *et al*. (2006), the manufacturer's recommendation (1:1000).

2.9 Adult immersion test

2.9.1 *In vitro* lousicidal activity test

The *in vitro* lousicidal efficacy test was started within one hour of lice collection and identification (Abuand Bekele, 2014). The collected lice were randomly divided into groups, each containing 10 in each group. The entire experiment was done in triplicate (Islam *et al*., 2018). 1 ml of each concentration of the plant extract, 0.5% tween 20 and 0.1% diazinon was applied directly to each petri dish containing lice. After one minute of contact time, the solution was filtered and dried using what man filter paper and incubated at 36ºC and 80% humidity for 24 h. After 30 min, 1 h, 2 h, 3 h, 6 h and 24 h of post-exposure, lice were observed under a stereoscope and the deaths of lice were recorded at each time interval (Sisay *et al*., 2015). The lack of limb movement and failure to respond when touched with a needle tip was used to confirm lice death (Abu *et al*., 2014).The percentage mortality of lice was calculated using the formula (Krishnaveni and Venkatalakshmi, 2014).

$$\text{Mortality } \% = \frac{\text{No. of dead lice}}{\text{Total No. of lice}} \times 100$$

The insecticidal activity was graded by Gemeda *et al*. (2014) as strong (mortality >80%), moderate (mortality 80-60%), weak (mortality less than 60 and greater than 40%) and little or no activity (mortality <40%).

2.9.2 *In vitro* acaricidal activity test

An *in vitro* adult immersion test was performed to determine the plant's activity against *A. varigatum*. The collected ticks were randomly divided into groups, each containing 10 in each group.The entire experiment was done in triplicate. The *in vitro* testing started 1 h after the ticks were collected and identified. The ticks were dipped in the prepared extract concentrations; negative control, 0.5% tween 20, and positive control, 0.1% diazinon, in a separate tube. After 2 min of contact time, the ticks were removed from the solutions, dried using filter paper and placed in a petri dish and incubated for 24 h at 28ºC with a relative humidity of 80% stated (Islam *et al*., 2018). Tick deaths were recorded after 30 min, 1 h, 2 h, 3 h, 6 h and 24 h of post-exposure. Tick mortality was checked with a blunt needle repeatedly and ticks were classified as dead, if no reaction was observed (Kumar *et al*., 2011). The percentage mortality of ticks was calculated by the formula previously used by Krishnaveni and Venkatalakshmi (2014):

$$\text{Mortality } \% = \frac{\text{No. of dead ticks}}{\text{Total No. of ticks}} \times 100$$

2.10 Data analysis

A statistical software package, SPSS Windows Version 20, was used for data analysis. A one-way analysis of variance (ANOVA) with multiple comparison tests (Post Hoc/ Turkey's test) was used to compare the mortality of lice and ticks with different concentrations of the extract and controls at different time intervals. The study's findings were presented as the percentage mortality of the parasite or mean of mortality ± standard error (Mean ± SE). All significant levels are set at $p< 0.05$.

3. Results

In the present study, qualitative phytochemical analysis showed the presence of different groups of secondary metabolites such as flavonoids, glycosides, saponins, phenols and tannins, but not

steroids in the methanolic leaf extract of *J. schimperiana* (Table 1). The 14.5% yield was obtained from *J. schimperiana* leaves extract and the extract has a dark green colour, is semi-solid and sticky in nature.

Table 1: Results of qualitative phytochemical screening of methanolic extract of *J. schimperiana* leaves

Secondary metabolites	*J. schimperiana*
Flavonoids	+
Saponis	+
Phenols	+
Tannins	+
Glycosides	+
Steroids	-

3.1 *In vitro* lousicidal activity of *J. schimperiana* against *B. ovis*

Mortalities of *B. ovis* treated with different concentrations of *J. schimperiana* leaf extract were revealed. At 24 h after exposure, a strong lousicidal activity of the extract at 200 and 100 mg/ml concentrations, a moderate activity at 50 and 25 mg/ml concentrations, and a weak activity at 12.5 and 6.25 mg/ml concentrations were shown. Generally, the percentage mortality of *B. ovis* lice treated with *J. schimperiana* extracts varied from 50% - 100% at 24 h post-exposure (Figure 1).

At 24 h post-exposure, lice mortality was statistically higher with 200 and 100 mg/ml concentrations than with all lower concentrations and the negative control. At 24 h post-exposure, there was a significant difference ($p<0.05$) between the reference drug, diazinon and higher concentrations of the plant extract, 200 and 100 mg/ml, in the mortality of lice. However, there is no significant difference ($p>0.05$) in the effects of the 200 and 100 mg/ml concentrations of the plant extract and 0.1% diazinon at 24 h post-exposure (Table 2).

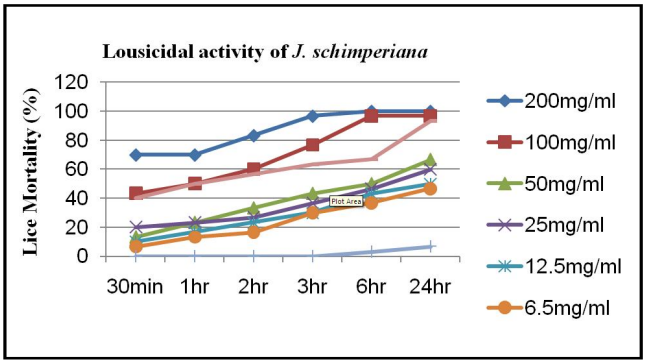

Figure 1: Percentage mortalities of lice treated with methanolic extract of *J. schimperiana*.

Table 2: *In vitro* lice killing effect of methanolic extract of *J. schimperiana* leaves against *B. ovis* for 30 min, 1 h, 2 h, 3 h, 6 h and 24 h of post-exposure, at each time interval

Extract concentrations (mg/ml)	Mean number of dead lice (mean of mortality ± SE) post exposure					
	30 min	1 h	2 h	3 h	6 h	24 h
200	7.00 ± 0.58^{a}	7.00 ± 0.58^{a}	8.33 ± 0.33^{a}	9.67 ± 0.33^{a}	10.0 ± 0.00^{a}	10.0 ± 0.00^{a}
100	4.33 ± 1.02^{ab}	5.00 ± 1.00^{a}	6.00 ± 0.58^{ab}	7.67 ± 0.88^{ab}	9.67 ± 0.58^{a}	9.67 ± 0.33^{a}
50	1.33 ± 0.67^{b}	2.33 ± 0.33^{b}	3.33 ± 0.88^{c}	4.33 ± 0.67^{c}	4.67 ± 0.88^{b}	6.67 ± 0.33^{b}
25	2.00 ± 0.58^{cb}	2.33 ± 0.33^{bc}	2.67 ± 0.67^{cd}	3.67 ± 0.33^{cd}	5.00 ± 0.58^{bc}	6.00 ± 0.58^{bc}
12.5	1.00 ± 0.58^{dbc}	1.67 ± 0.33^{bcd}	2.33 ± 0.33^{cde}	3.00 ± 0.00^{cde}	4.33 ± 0.33^{bcd}	5.00 ± 0.58^{bcd}
6.25	0.67 ± 0.33^{ecd}	1.33 ± 0.33^{bcde}	1.67 ± 0.33^{cdef}	3.00 ± 0.58^{de}	3.67 ± 0.33^{bcd}	5.00 ± 0.58^{bcd}
0.1% Diazinon	4.00 ± 1.00^{abcdef}	5.00 ± 0.58^{a}	5.67 ± 0.33^{bc}	6.33 ± 0.67^{bc}	6.67 ± 0.33^{c}	9.33 ± 0.33^{a}
0.5 % Tween 20	0.00 ± 0.00^{bcde}	0.00 ± 0.00^{bcde}	0.00 ± 0.00^{ef}	0.00 ± 0.00^{f}	0.00 ± 0.00^{f}	0.67 ± 0.58^{f}

Mean values with different letters in the same column shows the mean difference significance at ($p<0.05$).

3.2 *In vitro* acaricidal activity of *J. schimperiana* against *A. variegatum*

Mortalities of *A. variegatum* treated with different concentrations of *J. schimperiana* leaf extract were carried out in this study. At 24 h post-exposure, the mortality percentage of *A. variegatum* ticks treated with *J. schimperiana* extract ranged from 23.3% to 86.7% (Figure 2). Before 3 h post-exposure, all concentrations of the extract had week to low acaricidal activity at higher and lower concentrations, respectively. At 24 h post-exposure, the three higher concentrations of the extract, 200, 100 and 50 mg/ml, had significantly ($p<0.05$) higher acaricidal activities than the reference acaricide (0.1% diazinon). Tick mortality with diazinon has no significant difference with lower concentrations of extract <25 mg/ml at 24 h post-exposure (Table 3).

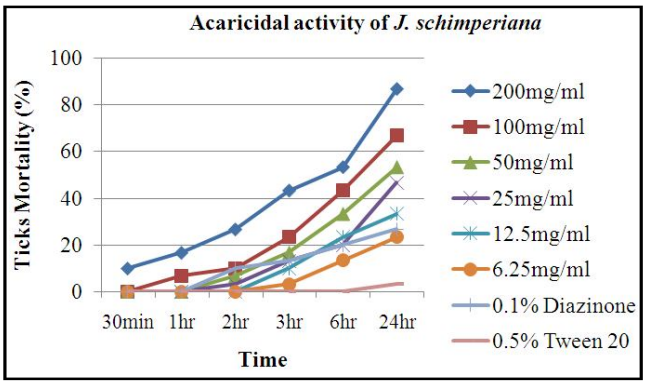

Figure 2: Mortalities percentage of ticks treated with methanolic extract of *J. schimperiana*.

Table 3: ***In vitro*** **ticks killing effect of methanolic extract of** ***J. schimperiana*** **leaf against** ***A. variegatum*** **at 30 min, 1 h, 2 h, 3 h, 6 h and 24 h of post-exposure, at each time interval**

Extract concentrations (mg/ml)	Mean number of dead ticks (mean of mortality ± SE) at min/h post-exposure					
	30 min	1 h	2 h	3 h	6 h	24 h
200	1.00 ± 0.58^{a}	1.67 ± 0.33^{a}	2.67 ± 0.33a	4.33 ± 0.33^{a}	5.33 ± 0.88^{a}	8.67 ± 0.67^{a}
100	0.00 ± 0.00^{b}	0.67 ± 0.33^{b}	1.00 ± 0.00b	2.33 ± 0.33^{b}	4.33 ± 0.33^{ab}	6.67 ± 0.33^{ab}
50	0.00 ± 0.00bc	0.00 ± 0.00^{bc}	0.67 ± 0.33^{bc}	1.67 ± 0.33^{bc}	3.33 ± 0.33^{bc}	5.33 ± 0.67^{bc}
25	0.00 ± 0.00^{bcd}	0.00 ± 0.00^{bcd}	0.33 ± 0.33^{bcd}	1.33 ± 0.33^{bcd}	2.00 ± 0.00^{cd}	4.67 ± 0.33^{bcd}
12.5	0.00 ± 0.00^{bcde}	0.00 ± 0.00^{bcde}	0.00 ± 0.00^{bcde}	1.00 ± 0.00^{bcde}	2.33 ± 0.33^{cde}	3.33 ± 0.33^{cde}
6.25	0.00 ± 0.00^{bcdef}	0.00 ± 0.00^{bcdef}	0.00 ± 0.00^{bcdef}	0.33 ± 0.33^{cdef}	1.33 ± 0.33^{def}	2.33 ± 0.33^{ef}
0.1% Diazinon	0.00 ± 0.00^{bcdefg}	0.00 ± 0.00^{bcdefg}	1.00 ± 0.58^{bcdefg}	1.67 ± 0.33^{bcdefg}	2.00 ± 0.00^{cdef}	2.67 ± 0.33^{def}
0.5 % Tween 20	0.00 ± 0.00^{bcdefg}	0.00 ± 0.00^{bcdefg}	0.00 ± 0.00^{bcdefg}	0.00 ± 0.00^{def}	0.00 ± 0.00^{f}	0.33 ± 0.58^{f}

3.3 Comparison of the plant activities between the parasites

The activities of different concentrations of *J. schimperiana* leaf extract against lice and ticks were compared at 24 h post-exposure. Accordingly, the effect of two concentrations (100 and 6.25 mg/ml) of plant extract showed significantly ($p<0.05$) better activity against lice than ticks. Although, statistically not significant, the others concentrations of the extract, showed relatively better activity against lice than ticks (Table 4).

Table 4: Comparisons of lousicidal and acaricidal activities of different concentrations of ***J. schimperiana*** **extract at 24 h post-exposure leaf**

Concentrations (µ1/ml)	Mortality percentage (%)		*p*-value
	Lice	Ticks	
200	100	86.7	0.116
100	96.7	66.7	0.003
50	66.7	53.3	0.148
25	60	46.7	0.116
12.5	50	33.3	0.067
6.25	50	23.3	0.016

4. Discussion

The percentage yield of *J. schimperiana* extract in the current study was 14.5%, which is in line with the previous study by Mekonnen *et al.* (2018) who reported a 15.70% yield from *J. schimperiana* leaf extracted with 80% methanol extract. In qualitative phytochemical screening tests, flavonoids, glycosides, saponins, phenols and tannins were detected in the methanolic extract of *J. schimperiana* leaves. The present finding is in line with the previous findings by Mekonnen *et al.* (2018) and Tesera *et al.* (2021) who reported the presence of phenols, tannins, flavonoids and saponins in methanolic extract of *J. schimperiana* leaves. Mekonnen *et al.* (2018) and Tesera *et al.* (2021) also, found steroids in the extract of *J. schimperiana* leaves extracted with 80% methanol, which is inconsistence with the present finding in which steroids were not found in the *J. schimperiana* leaf extracted with 99.8% methanol. The variances could be attributable to differences in the concentrations of the solvent. According to Pandey and Tripathi (2014), the solubility of various plant components is affected by the concentration of the solvent utilized. The previous study utilized hydromethanolic (80% methanol), which is more likely to extract a wider range of phytochemicals than absolute methanol, which was utilized in the current study (Bandiola, 2018; Truong *et al.*,2019). The present study found that a methanol extract of *J. schimperiana* leaf had strong lousicidal activity at concentrations of 200 and 100 mg/ml at 24 h post-exposure, with an impact comparable to that of the commercial drug, diazinon. As far as our literature search is concerned, no study has been reported so far on the lousicidal activity of *J. schimperiana* leaf extract. The presence of tannins and saponins in the methanolic extract of *J. schimperiana* leaf, which have been reported to have antiparasite activities (Hrckova and Velebny, 2012), may explain this plant's lousicidal activity. Tannins restrict the energy generation of the parasite by binding glycoprotein to the cuticle of the parasite, which leads to the death of the parasite (Hrckova and Velebny, 2012; Abdalla *et al.*, 2020). Saponins are reported to disrupt the cell membrane of the parasites, thereby changing the morphology of the cells in the cuticle. The disintegration of the cuticle results in the parasite's drying out.

At 200 and 100 mg/ml concentrations, *J. schimperiana* leaf extract outperformed the commercial acaricide diazinon in terms of acaricidal efficacy against *A. variegatum* at 24 h post-exposure. The acaricidal activity of this plant may be due to the presence of tannins and saponins in the suspicion methanolic extract of *J.*

schimperiana leaves, which were reported to possess antiparasite activities (Hrckova and Velebny, 2012). The extract of the plant had much higher efficacy compared to the commercial compound, diazinon, putting the latter under suspicion for acaricidal resistance. The extract was significantly more effective against *B. ovis* (lice) species than *A. variegatum* ticks. This might be attributed to thickness of the integument in ticks as compared to lice which interferes with absorption of the active ingredients present in the extracts of the study medicinal plants.

The overall results of this study indicated that the mortality of both lice and ticks caused by the extract increased with time after exposure and concentration. This indicated that time and concentration played an important role in influencing the viability of the lice and ticks. This result is in line with the findings of Alemu (2015) and Amante *et al.* (2019) who reported time and concentration-dependent effects of botanicals.

5. Conclusion

This study aimed to demonstrate the relative efficacy of *J. schimperiana* leaf extract against *B. ovis* and *A. varigatum*. *J. schimperiana* contains flavonoids, glycosides, saponins, tannins and phenols. The findings revealed that the methanolic extract of *J. schimperiana* leaf had strong lousicidal activities at 200 and 100 mg/ml concentrations, similar to diazinon. In addition, the plant had shown strong acaricidal activity at a 200 mg/ml which was higher than diazinon at 24 h post-exposure. The extract was more effective against lice than against ticks. As a result, the present study stated that *J. schimperiana* could be used as an alternative in the control of *B. ovis* and *A. variegatum* infestations. Based on the results obtained in the present study, it can be concluded that the methanolic extracts of *J. schimperiana* were rich in phytochemicals; this may be attributed to the presence of bioactive molecules that may serve as effective compounds. Thus, further research may be warranted to isolate and characterize the bioactive compounds, solubility properties and the presence of certain key functional groups to confer in drug discovery.

Acknowledgments

The authors would like to thank the Department of Veterinary Laboratory Technology, School of Veterinary Medicine, Ambo University for using the laboratory facilities to entire research work. The authors would also to convey the special thanks to Mr. Biruk Bedore for plant identification and authentication.

Conflict of interest

The authors declare no conflicts of interest relevant to this article.

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Citation **Askale Gizaw, Banti Kena, Dagmawit Atalel Babele, Bizunesh Mideksa Borena, Galma Boneya, Mohammed Siraj, Solomon Shiferaw, Waktole Terfa, Miressa Tadesse, Morka Dandecha Bayu, Nega Desalegn Tadese, Edilu Jorga Sarba and Chandra Sekhar Singh Bhaludra (2022). Phytochemical screening and *in vitro* lousicidal and acaricidal activities of *Justicia schimperiana* (Hochst. ex Nees) T. Anderson leaf in West Showa Zone, Ethiopia. Ann. Phytomed., 11(2):725-731. http://dx.doi.org/10.54085/ap.2022.11.2.88.**