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PHYTOCHEMICAL SCREENING AND *IN-VITRO* ACARICIDAL ACTIVITY OF THREE HERBAL EXTRACTS AGAINST CATTLE TICK *BOOPHILUS DECOLORATUS*

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Keywords:

Azadirachta indica,
Boophilus decoloratus,
Datura stramonium, *Nicotiana glauca*

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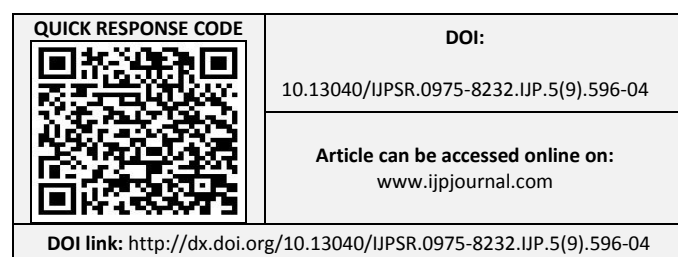
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ABSTRACT: This study was designed to determine the acaricidal effect of *Datura stramonium*, *Nicotiana glauca* and *Azadirachta indica* herbal extracts against the common cattle tick *Boophilus decoloratus*. Qualitative phytochemical analysis using standard techniques was also used to detect secondary metabolites contained in the selected herbs. The percent adult mortality and percentage inhibition of oviposition were studied at different experimental concentrations of 25, 50, 75 and 100 mg/ml to determine the efficacy of leaf extracts. 3% of dimethyl sulfoxide was also used as a negative control. The study determined the presence of secondary metabolites in the herbal leaf extracts, which are able to cause neuro-toxicity in the tick. Inhibition of oviposition at the highest concentration of *Datura stramonium*, *Nicotiana glauca* and *Azadirachta indica* of the treated ticks were 78.68, 86.84 and 52.63%, respectively. At the highest concentration, the adult tick mortality was 90, 100 and 80% for *Datura stramonium*, *Nicotiana glauca* and *Azadirachta indica*, respectively. The results of the current study pointed the potential acaricidal effect of selected medicinal plants with varying potency. Therefore, further study should be done *in-vitro* and *in-vivo* assays to use studied plants as acaricides.

INTRODUCTION: Ethiopia is naturally endowed with different agro-ecological zones and environmental conditions which makes the country suitable for different agricultural production system. The country also possesses the largest livestock population in Africa. However, the rich potential from the livestock sector in general is not efficiently exploited due to several constraints like healthy and nutritional problem, limited genetic potential and other husbandry standards. These limitations results in losses of body weight gains, inedible of organs and carcasses and lower milk yield production¹.

Among the major health concerns disturbing livestock the one is ecto-parasites, particularly tick infestation. Directly, ticks causes skin damage opening up wounds which make the animal susceptible to secondary infection. Indirectly and more significantly, ticks act as vectors of fatal diseases, like babesiosis, cowdrosis and theileriosis². In common practice, these ecto parasites are controlled by commercial chemical acaricides.

However, the use of commercial acaricides has been led to development of widespread resistance, environmental toxicity of chemicals, residuals in animal product and ever increasing cost of acaricides³. One approach to solve the above challenges is to search and integrate the existing conventional methods of health management and ethno veterinary practices, particularly the use of medicinal plants. Medicinal plants are important for healing of human and animal diseases due to the existence of certain specific substances, known



as phytochemicals. Phytochemicals are not nutritious chemical compounds and are naturally found in medicinal plants which results defense mechanisms and protection against various diseases⁴. In Ethiopia, medicinal plants and knowledge of their use provide a vital contribution to human and livestock health care needs throughout the country.

Available literature clearly shows that the contribution of medicinal plant as a primary health care options in the country, where 70% of human and 90% of livestock population depend on traditional medicine⁵.

Among medicinal plants, *Datura stramonium* is stinking and a branched herb that forms a 60 to 150 cm tall shrub, grown in the temperate and tropical region of the globe. The root of the plant is long, fibrous and white, while the stem is erect, smooth and pale yellow green. The leaves of the plant are long, smooth and jagged. The upper surface of the leaves is a darker green and the lower surface is a light green. The plant is used as medicine in different part of the world for asthma symptoms in Ayurveda and as anaesthesia during surgery in china⁶.

Nicotiana glauca is a perennial evergreen shrub up to 6 m tall, with strongly branched stems. The leaves are stalked, oval and gray-green. The plant has been widely distributed in many parts of Central America, Africa and the European Mediterranean region⁷. In Ethiopia, the plant was used against ecto parasite of ruminants by farmers located in Eastern Tigray⁸.

Azadirachta indica is fast growing plant commonly found in Asia, Africa and America. It is ever green but in severe dry period it may shed most of its leaves. The branches of the plant are wide and spreading. The fruit, seeds, leaves and almost every part of the tree contain compounds with proven antiviral, anti-inflammatory, anti-ulcer, antifungal, antibacterial and anti parasitic properties⁹.

Therefore the objectives of this study were:

General Objective:

- To determine the acaricidal effect of herbal extracts against the common cattle tick *B. decolratous*.

Specific Objectives:

- To detect the type of secondary metabolites present in crude ethanolic extracts of selected plants using qualitative phytochemical screening methods,
- To determine the efficacious herbal extract and evaluate the effective experimental concentration against *B. decolratous* tick of cattle *in-vitro*.

MATERIALS AND METHODS:

Study Area: The study was conducted at Mekelle University, College of Veterinary Medicine, Mekelle. Mekelle is the capital city of Tigray Regional State and located at about 783 km from Addis Ababa to the north direction. The city is located at 39°33'E longitude and 13°32'N latitude, situated in the extension of the central highlands of Ethiopia. The altitude of the town is between 1665-2220 meters above sea level. The mean annual rainfall is about 619 mm with having bimodal rain fall with short rainy season occurring from March and another from middle September to February. The annual minimum and maximum temperature of the town is 11.8 °C and 29.9 °C, respectively¹⁰.

Study Design: An experimental study was conducted from November 2017 to May, 2018 to determine pytochemical constitutes and an acaricidal activity of three herbal leaves extracts against the common cattle tick *B. decolratous*.

Study Herbs and Collection: The traditionally recognized medicinal plants used by the local communities to treat animals were the study subjects. The herbs used in the present experimental study were *Datura stramonium*, *Nicotiana glauca* and *Azadirachta indica*. The leaves of *Azadirachta indica* specimens was collected from Kalmino Campus, College of Veterinary Medicine while the leaves *Nicotiana glauca* and *Datura stramonium* were harvested from Mekelle City Districts.

Herbal Extraction: The collected plant materials were washed with tap water to remove any traces of soil and other unnecessary particles. Leaves were air dried completely under shade and ground with electric blender into fine powder, sieved and stored in clean stopper bottles until used for extraction. The powdered leaves were macerated using 97% ethanol.

Two hundred fifty grams of each experimental leave powders were weighted using an electronic weighing balance and mixed with 1250 ml of 97% ethanol in each of the flasks and plugged tightly with plastic and sterile gauze. The coarsely powdered materials were kept in contact with selected solvent in the flasks for three days with frequent agitation manually four times per day for 10 min until soluble matter was dissolved.

Then, the liquid part was separated from the herbal residues and filtered through a Whatman filter paper no. 1 using an electrical suction pump. The beaker with liquid then put in a water bath at 42 °C to evaporate the solvent. The weights of the dried crude extracts were determined and a yield percentage was calculated for each experimental herbs.

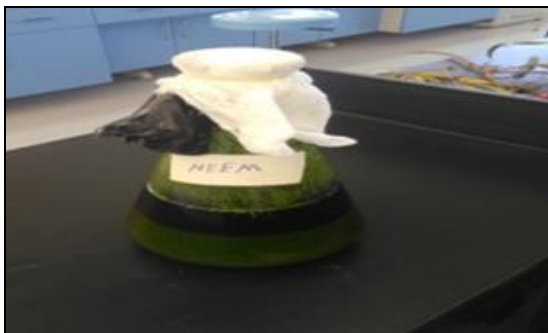
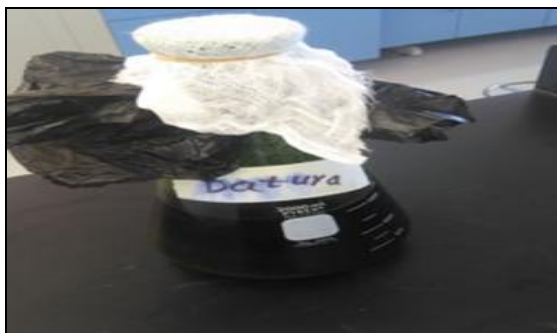
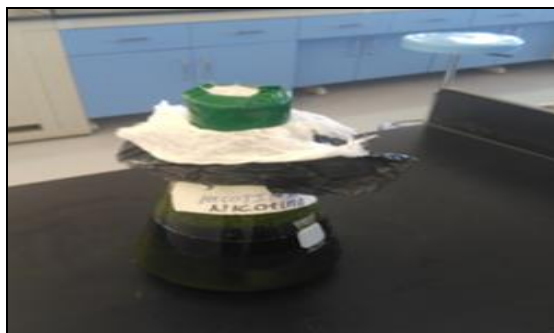


FIG. 1: IMAGE DURING EXTRACTION



FIG. 2: LIQUID FILTER USING AN ELECTRICAL SUCTION PUMP

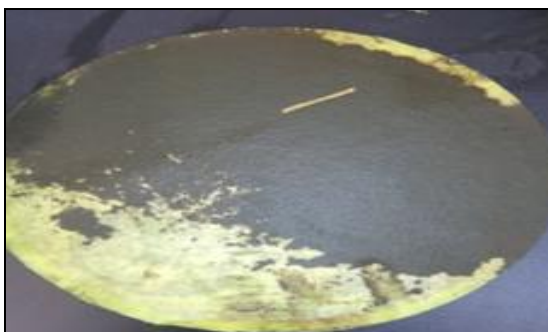


FIG. 3: RESIDUES OF FILTERED LIQUID

Phytochemical Screening: The crude extract of *A. indica*, *N. glauca* and *D. stramonium* were subjected to phytochemical screening using the generally accepted laboratory techniques for qualitative determinations. Screening was carried out for crude extracts to identify the active chemical constituents¹¹.

Test for Saponins:

Frothing Test: This test was performed by mixing 1 gm of crude extract with 5 ml of distilled water in

a test tube and it was shaken vigorously for 15 min. The formation of stable foam that remains for 10-15 min indicated the presence of saponins.

Test for Flavonoids:

Shinoda Test: This test was performed by mixing 0.1 g of the extracts with few drops of 1% NaOH. An intense yellow color was produced in the extract which became colorless on addition of a few drops of diluted HCl acids indicates the presence of flavonoids.

Test for Tannins:

Ferric Chloride Test: About 0.5 g of the extract in 10 ml of water in a test tube in a beaker was boiled on hot plate stirrer for 5 min and then filtered with filter paper. Two drops of 0.1% FeCl₃ solution was added and observed for brownish or blue black coloration indicating the presence of tannins.

Test for alkaloids:

Mayer's Test: This test was performed by mixing 1g of the extract with 2 ml of HCl and heated on water bath for 5 min and 3 drops of Mayer's reagent was added and checked for creamy white precipitate.

Test for Phlobatannins:

Hydrochloric Acid (HCl) Test: About 0.2 g of extracts were added to 2 ml of 1% HCl in test tube in beaker and the mixture was boiled on hot plate stirrer and cooled for 5 min. Deposition of a red precipitate was taken as evidence for the presence of phlobatannins.

Test for Phenolic Compounds:

Ferric Chloride Test: in this test 0.2 g of extract was dissolved in 5 ml of distilled water, and 2 ml of 5% ferric chloride solution was added. The formation of bluish green color indicated presence of phenolic compounds.

Test for Glycosides:

Sulfuric Acid (H₂SO₄) Test: In this test 2 ml of concentrated H₂SO₄ was added carefully to 1g of each crude extract in test tube and shaken gently. A reddish brown color was observed for all extracts. This indicated the presence of glycone portion of the glycoside.

Preparation of Experimental Concentrations:

There were four different experimental concentrations prepared to test the efficacy of three experimental herbal plants against the *Boophilus decoloratus* tick. The experimental concentrations were 100, 75, 50 and 25 mg/ml. 3% dimethyl sulfoxide (DMSO) used as solvent and control group.

Experimental tick Collection and Identification:

Adult male and female (engorged and partially engorged) *B. decoloratus* ticks were collected from intensive and semi intensive farms in Mekelle town. Cattle were restrained and the entire body

surfaces of the animals were examined thoroughly and the ticks were collected from neck/dewlap, shoulder, belly, udder, anus and legs. The collected ticks were placed in labelled plastic flasks which were aerated; free of any acaricide and maintained with a humidity using moisten cotton as per standards³. The ticks were transported to the veterinary parasitological laboratory and examined under stereomicroscope for identification according to the standard identification keys¹².

In-vitro Test:

Adult Immersion Test (AIT): The investigational procedure suggested by¹³ and modified by (FAO) was used to carry out the experiment. The engorged female ticks collected from the field were washed thoroughly with tap water and kept for drying on filter paper. Ten viable ticks were put in group and weighed; an effort was made to obtain groups with similar weights. Three replicates of five different test tubes were made ready each containing the different crude extract concentrations (100, 75, 50 and 25 mg/ml) and the control (3% DMSO). The ticks on each group were immersed into each experimental concentration for 5 min.

The ticks were removed out of the extract solutions and gently dried on filter paper. Then these ticks were kept separately in petridishes with filter paper at bottom. The petridishes were covered with sterile gauze and put in a biochemical oxygen demand (BOD) incubator at 28 ± 2 °C with a relative humidity of 80% for 7 days. After 7 days of incubation, the ticks on each petridishes were observed for egg laying and eggs produced by each group were weighed. Then the efficacy of the crude extracts was evaluated using percent inhibition of oviposition (IO %) and which was calculated by;

$$IO\% = \frac{MEC - MET}{MEC} \times 100$$

Where, MEC and MET are mass of eggs laid by control ticks and treated ticks, respectively and IO (%) percent inhibition of oviposition.

Filter Paper Impregnation Method (FPIM): The experimental procedure adopted by FAO (1984)¹⁴ was used to investigate adult male / female (partially engorged) tick mortality. Whatman filter papers of the same diameter as that of the petridishes were impregnated with one ml of each

experimental concentration (100 mg/ml, 75 mg/ml, 50 mg/ml and 25 mg/ml) and control (3% DMSO). Then these filter papers were placed in the petri dishes and ten adult ticks were put onto these filter papers of each petridishes.

A similar filter paper impregnated with the same strength of the extract was placed on top of the ticks. Three replications were done for each concentration. The petri dishes were then closed with sterile gauze and rubber bands to prevent ticks from crawling out. Tick mortality was then recorded after 24 h of exposure to crude extracts and the control. The criteria for death of ticks were determined by observing signs of the movement and by pricking it with needle. The ticks were judged as dead, if there are no signs of movements. Finally, the mortality of the adult tick was calculated by using the formula:

$$\text{Mortality (\%)} = \frac{\text{Dead tick count}}{\text{Total tick count}} \times 100$$

Data Management and Analysis: Data of the variables of interest such as concentrations of the experimental extracts, tick egg weight and mortality percentages were entered into Microsoft excel spread sheet and descriptive statistics of the collected data was calculated. Percent inhibition of oviposition and mortality percentage for each leaf extract was used to determine its effectiveness.

RESULTS: In the present study four different experimental concentrations (100, 75, 50 and 25mg/ml) of three selected herbal extracts were tested against the cattle tick *Boophilus decoloratus*. DMSO 3% was used as a control in the experiment.



Yield percentages of the herbal extracts, phytochemical screening, percentage inhibition of oviposition and percentages of adult tick mortality of each extracts were determined.

Yield Percentages of the Herbal Extracts: In the present study, 250 gm of herbal material was macerated with 1.25 litre of 97% ethanol to produce a measured amount of dried crude extracts. Therefore, the yield percentages of each of the three experimental herbs are calculated and given in **Table 1**. Highest yield was detected for *D. stramonium* and lowest yield for *A. indica*.

TABLE 1: YIELD PERCENTAGE OF ETHANOLIC EXTRACTS OF THE EXPERIMENTAL HERBS

| Plants | Weight of the fine powder | Amount of dried crud extract | Yield % |
|----------------------|---------------------------|------------------------------|---------|
| <i>D. stramonium</i> | 250g | 33.2g | 13.28% |
| <i>N. glauca</i> | 250g | 29.19g | 11.68% |
| <i>A. indica</i> | 250g | 27.19g | 10.87% |

Phytochemical Screening: In this study, three experimental ethanolic herbal extracts were subjected for qualitative phytochemical screening and indicated in **Table 2**. The crude extract of all selected plants showed the presence alkaloid.

TABLE 2: PHYTOCHEMICAL ANALYSIS OF ETHANOLIC EXTRACTS OF THE EXPERIMENTAL HERBS

| Variable | <i>D. Stramonium</i> | <i>N. glauca</i> | <i>A. indica</i> |
|--------------------|----------------------|------------------|------------------|
| Alkaloids | + | + | + |
| Tannins | - | + | + |
| Flavonoids | - | - | - |
| Saponins | + | - | - |
| phenolic compounds | - | + | + |
| phlobatannins | - | - | - |
| Glycosides | - | + | + |

Note: + is present, - is absent



FIG. 4: TANNINS POSITIVE AND FLAVONIDS NEGATIVE RESPECTIVELY DURING PHYTO CHEMICAL ANALYSIS

Percentage Inhibition of Oviposition: The results of adult immersion test using the experimental ethanolic extracts of *N. glauca*, *D. stramonium* and *A. indica* were shown in **Table 3**. The efficacy of

crude extract of three experimental herbs against *B. decoloratus* was determined by calculating the percentage inhibition of oviposition. The percentage inhibition of oviposition of the cattle

tick *B. decoloratus* due to *D. stramonium* at a concentration of 100 mg/ml was 78.68% with the least at 25 mg/ml (IO = 65.78%). Similarly, the percentage inhibition of oviposition due to *N. glauca* at a concentration of 100 mg/ml was 86.84% with least at 25 mg/ml (IO = 78.15%). In *A. indica*, the percentage inhibition of oviposition at a concentration of 100 mg/ml was 52.63% with least at 25 mg/ml (IO = 31.57%).



FIG. 5: EGG MASS LIED BY ENGORGED *B. DECOLORATOUS*

Percentages of Adult Tick Mortality: The results of adult tick mortality of *B. decoloratus* by different experimental concentrations (100, 75, 50 and 25 mg/ml) of three selected herbal extracts and control were shown in the following Table. The effectiveness of leaf extract against *B. decoloratus* was determined by calculating the percent adult mortality using by filter paper impregnation method.

Among the three plants *N. glauca* shows effective mortality to the adult tick then followed with *D. stramonium* and *A. indica*. Based on concentration, *N. glauca* extract was showed 100% mortality at 50 mg/ml, 75 mg/ml and 100 mg/ml concentration with the exception of 25 mg/ml (80%). The extract of *D. stramonium* was showed mortality of 90% at 100 mg/ml with least 60% at 25 mg/ml. similarly; *A. indica* was shown 80% of mortality at 100mg/ml with least 50% at 25 mg/ml.

TABLE 3: THE RESULTS OF ADULT IMMERSION TEST OF ETHANOLIC LEAF EXTRACTS

| Plant | Concentration mg/ml | N | M2 | IO |
|----------------------|---------------------|-----------|-------|--------|
| <i>D. stramonium</i> | Control | 3 | 0.38 | 0.00 |
| | 100 mg/ml | 3 | 0.081 | 78.68% |
| | 75 mg/ml | 3 | 0.094 | 75.26% |
| | 50 mg/ml | 3 | 0.11 | 71.05% |
| | 25 mg/ml | 3 | 0.13 | 65.78% |
| <i>N. glauca</i> | 100 mg/ml | 3 | 0.05 | 86.84% |
| | 75 mg/ml | 3 | 0.062 | 83.68% |
| | 50 mg/ml | 3 | 0.069 | 81.84% |
| | 25 mg/ml | 3 | 0.083 | 78.15% |
| | <i>A. indica</i> | 100 mg/ml | 3 | 0.18 |
| 75 mg/ml | | 3 | 0.21 | 44.73% |
| 50 mg/ml | | 3 | 0.22 | 42.10% |
| 25 mg/ml | | 3 | 0.26 | 31.57% |

N = Number of immersed engorged ticks; M2 = mean egg mass per replicate (g); IO = percent inhibition of oviposition

TABLE 4: PERCENTAGES OF MORTALITY OF *B. DECOLORATOUS* TICK BY EXPERIMENTAL ETHANOLIC EXTRACTS

| Plant | Concentration (mg/ml) | N | T | Mortality % |
|----------------------|-----------------------|-----------|-----|-------------|
| <i>D. stramonium</i> | control | 10 | 24h | 0.00% |
| | 100 mg/ml | 10 | 24h | 90% |
| | 75 mg/ml | 10 | 24h | 80% |
| | 50 mg/ml | 10 | 24h | 80% |
| | 25 mg/ml | 10 | 24h | 60% |
| <i>N. glauca</i> | 100 mg/ml | 10 | 24h | 100% |
| | 75 mg/ml | 10 | 24h | 100% |
| | 50 mg/ml | 10 | 24h | 100% |
| | 25 mg/ml | 10 | 24h | 80% |
| | <i>A. indica</i> | 100 mg/ml | 10 | 24h |
| 75 mg/ml | | 10 | 24h | 70% |
| 50 mg/ml | | 10 | 24h | 60% |
| 25 mg/ml | | 10 | 24h | 50% |

N = numbers of tick, T = time exposure

DISCUSSION: Highest percentage yield was detected for *D. stramonium* with 13.28% and the smallest one with *A. indica*, which was 10.87%.

This study verified the presence of biologically active compounds in the experimental ethanolic leaf extracts of *N. glauca*, *D. stramonium* and *A.*

indica. Due to the existence of different bioactive compounds in leaf extracts, antibacterial, antifungal, antiparasitic and anti-inflammatory properties have been attributed to it¹⁵. In previous studies it was reported that phenolic compounds and Tannins were absent in ethanolic extract of *N. glauca*¹⁶.

However, the result of the present studies support that both secondary metabolites were present in the ethanolic leaf extract. Also the previous study shows that saponins and flavonoids are present in ethanolic extracts of *A. Indica*¹⁷. But the current finding of this study shows both bioactive compounds were found to be absent. The outcome of this studies and earlier research studies results were unlike so it might be due to the change in location and genetic variation due to cross pollination, so their genetic makeup were changed and that is why both studies shows different results. Without precise chemical investigations and accurate biological tests it is difficult to verify which of the bioactive compounds contained in traditionally used plants exhibit efficacy against ticks. However, having a closer look at the plants, plant containing neurotoxic substances such as alkaloids, phenolic compounds, tannins and glycosides are able cause acute toxicity in tick¹⁸.

This study revealed the presence of secondary metabolite like alkaloids in all leaf extracts. Also it shows the presence of biologically active compounds such as tannins, phenolic compounds and glycosides in selected plants except *D. stramonium*. This phyto constituents' of medicinal plants are considered to be the chemical components that are so potent and are reported to cause mortality and inhibition of oviposition of adult ticks¹⁹.

The cattle tick *B. decolrtus*, is an economically important ecto parasite of livestock and generates major problem for livestock producers in tropical and subtropical countries. In order to control this ectoparasite, different types of chemicals are used widely²⁰. However, broad spread uses of these chemicals cause serious problems like soil and water contamination, killing non-target organisms and threaten human health due to the toxic residues in milk and meat²¹. These problems had stimulated a great deal of research to look for alternative

treatment from natural products. For this reason, the current study evaluated activity of *N. glauca*, *D. stramonium* and *A. indica* medicinal plants for alternative management of cattle ticks. Hence, natural botanical compounds are residue less, flora and fauna friendly, biodegradable, hold a range of chemically active ingredients which can intervene in all biological processes of the ticks, thus interrupting their life cycle and are accepted as an incorporated part of ethno-veterinary practices.

In previous study, ethanolic leaf and seed extracts of *D. stramonium* were reported for acaricidal, repellent, and reproductive inhibitory properties against two-spotted spider mites, *in-vitro*. The leaf and seed extracts, which were applied in 167, 250 and 145, 750 mg/ml concentrations, respectively, resulted 98 and 25 % mortality among spider mite²². Entire part of *D. stramonium* has got several importances and toxicity to a greater extent. The alkaloids isolated from the plants are a found to be toxic to the animals. But, leaves extracts of *D. stramonium* are reported with significant acaricidal activities against *B. microplus*²³. Some study also investigated toxic effect of the leaf extracts of *Datura species* plants at various concentrations on grasshoppers and red ants²⁴.

In current study, *D. stramonium* was evaluated against *B. decolrtus* tick and shows inhibition of oviposition of engorged female ticks from 65.78% to 78.68%, when tested at concentrations ranging from 25 to 100 mg/ml. The highest inhibition of ovipostione (78.68%) was observed at the higher concentration (100 mg/ml). The percent of adult tick mortality caused by the crude ethanolic extract of this herb is varied from 60% to 80% when tested at concentrations ranging from 25 to 100 mg/ml at exposure time of 24 h. This results were slightly in agreement with the study made by Srikanta *et al.*, (2015)²⁵ on ethanolic extract of *D. stramonium* in India, who reported 60% mortality of adult tick and 83% percentage inhibition of oviposition at 100mg/ml.

Azadirachta indica is one of commonly grown plant in different parts of Africa and Asia. In previous study, it has been verified to have effect against mange and mosquito larvae²⁶. The petroleum fractions of *Azadirachata indica* seed also evaluated *in-vitro* test and found effective

against *B. microplus* tick²⁷. In the present study, the ethanolic leaf extract of *A. indica* were evaluated against *B. decolrtus* and imparted inhibition of oviposition ranging from 52.63% to 31.57% at different experimental concentrations. The percent mortality caused by the ethanolic extracts *A. indica* is varied from 50% to 80% at concentrations ranging from 25 to 100 mg/ml. The highest mortality and inhibition of oviposition was observed at 100 mg/ml.

This finding is in agreement with Kalakumar *et al.*, (2000)²⁸ also assessed the effect of *A. indica* and have shown 60-75% efficacy against buffalo ticks but failed to inhibit oviposition in female ticks; but, the present finding, in contrary to this, reported up to 52.63% inhibition in oviposition. Related results were reported also with Rahul *et al.*, 2008,²⁹ in which 65% mortality of adult *Bopilus* tick was recorded within 24 h of treatment with extract *A. indica* at 100 mg/ml.

In current study, the leaf extracts of *N. glauca*, shows good acaricidal activity from the other selected herbal extracts. It shows the highest percentage of inhibition of oviposition and mortality of adult ticks at all concentration when compared to *D. stramonium* and *A. indica*. At concentration of 100 mg/ml, the leaf extract of *N. glauca* exhibited 86.84% of inhibition of oviposition and 100% of mortality rate in the present study is in contrast with Flávia *et al.*, 2014³⁰ in Brazil, who reported 22.64% of inhibition of oviposition and 46.46 mortality rates at the same concentration against *B. microplus*.

The result of this study and previous research studies results were different so it might be due to the variation in phyto-constituents of the plant from the place to the place. Further investigations are recommended in this regard.

CONCLUSION AND RECOMMENDATIONS:

The findings from this study determine that the crude ethanolic extracts of *N. glauca*, *D. stramonium* and *A. indica* contain secondary metabolites with *in-vitro* acaricidal activities against adult *B. decolratous* tick and showed mortality and inhibition of oviposition effect especially *N. glauca* and *D. stramonium* showed high efficacy. The results obtained, therefore,

suggest that these extracts could have good candidates for developing potentially useful and effective acaricides for the control of ticks in livestock in the future. In addition, tests with animals can be performed with *N. glauca* and *D. stramonium* to evaluate the toxicity and to confirm their acaricidal activity in target species.

Based on the above conclusion, the following recommendations are forwarded:

- Further *in-vivo* tests on live animal should be done before deciding to use studied plants as acaricides, especially those showed high efficacy.
- Further investigation of isolated fractions at different dose levels on larva stage of *B. decolratous* tick should be practiced.
- Further acaricidal activity tests on other ectoparasites need to be done to know if the plants have broad spectrum activity.

ACKNOWLEDGEMENT: Nil

CONFLICT OF INTEREST: Nil

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