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Article

Novel acaricidal efficacy of nine Egyptian plants against the camel tick, *Hyalomma dromedarii* (Ixodida: Ixodidae)

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ABSTRACT

Ticks are very important ectoparasites transmitting diseases to animals and humans and their natural control is an urgent need. The efficacy of nine novel aqueous plant extracts against *Hyalomma dromedarii*, semi-fed males, was evaluated through the adult immersion test; seven concentrations were applied (10, 20, 40, 90, 110, 130, and 170 mg/ml) for each plant extract. Plants could be arranged according to their Mortality (MO) percent, 15 days post-treatment (PT) with 170 mg/ml, as follows: 96% by *Ricinus communis*; followed 84% by *Alchemilla vulgaris*, *Cichorium endivia*, *Quercus cortex*, and *Salvia rosmarinus*. Three, seven, and 12 days PT, the LC_{50} values of the highly effective group including *R. communis* (100.11, 30.34, and 30.22%), *A. vulgaris* (140.11, 60.94, and 50.38%), *C. endivia* (120.56, 60.13, and 30.80%), *Q. cortex* (110.87, 50.05, and 30.13%) and *S. rosmarinus* (110.73, 50.26, and 40.35%) were calculated. The LT_{50} values for the highly effective plants were 1.000, 3.136, 2.282, 4.498, and 2.614 days, PT with 170 mg/ml, for *R. communis*, *A. vulgaris*, *C. endivia*, *Q. cortex*, and *S. rosmarinus* and their LT_{99} values were 9.224, 19.840, 19.296, 22.928, and 29.907 days, respectively. The relative toxicity of the *A. vulgaris*, *Allium sativum*, *C. endivia*, *Cyperus rotundus*, *Lepidium sativum*, *Q. cortex*, *R. communis*, and *S. rosmarinus* were 1.0, 1.6, 1.6, 1.2, 1.3, 1.7, 1.7, and 12 times, more effective than *Lawsonia inermis*, respectively. Phytochemical analyses indicated the presence of phenolic, tannin, and flavonoid compounds. This study recommended using *R. communis* for tick control, followed by *Alc. vulgaris*, *Ci. endivia*, *Q. cortex*, and *S. rosmarinus*.

KEYWORDS: Adult immersion, *Alchemilla*, *Cichorium*, *Quercus*, *Ricinus*, *Salvia*.

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INTRODUCTION

Ticks are important ectoparasites infesting domestic and wild animals leading to blood loss,

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depression of immunity, general stress, irritation, decreased hide value, and transmsion of infectious diseases as babesiosis, anaplasmosis, borreliosis, ehrlichiosis, and rickettsiosis (Adenubi *et al*. 2016).

The camel tick *Hyalomma dromedarii* (Ixodida: Ixodidae) is the predominant species in the Middle East area infesting camels (Perveen *et al*. 2020). Using conventional pesticides has resulted in contamination of the environment and residues in milk and meat products and the development of acaricide-resistant ticks (Adenubi *et al*. 2016; Iqbal *et al*. 2021; Mohammed *et al.* 2022). Alternative control strategies to overcome the drawbacks associated with synthetic acaricides represent an urgent need.

The high cost of developing new chemical acaricides renewed the interest in using safe biorational pesticides (Iqbal *et al*. 2021). Plants contain secondary metabolites used as pesticides because they are biodegradable, less toxic to non-target species, and less likely to have resistance developed against them (Khater 2012) and induce larvicidal, ovicidal, adulticide, repellent, and deterrent effects (Khater *et al*. 2014, 2018; Baz *et al*. 2021, 2022a, b, c).

In Egypt as well as worldwide, there are very few studies which have tested plant extracts against *H. dromedarii* (Abdel-Shafy *et al*. 2007; Habeeb *et al*. 2007; Mahran *et al*. 2020; Abdel-Ghany *et al*. 2021a, b, 2023). Therefore, the aims of this work were to evaluate the in vitro acaricidal efficacy and chemical analyses of locally available and affordable nine plants against *H. dromedarii* and determine their lethal concentration and time values and evaluated their relative toxicities and their photochemical analyses.

MATERIALS AND METHODS

Ticks

Hyalomma dromedarii (males) were collected and identified (Apanaskevich *et al*. 2008) from areas around camels at Toukh (5 to 15 years old) (30° 21′ 11.6″ N and 31° 11′ 31.5″ E), Qalyubia Governorate, Egypt.

Plant extracts preparation

Plants were identified at the National Research Center, Egypt. The applied plants were lady's mantle, *Alchemilla vulgaris* (Potentilleae); garlic, *Allium sativum* (Amaryllidaceae); chicory, *Cichorium endivia* (Asteraceae); tiger nut, *Cyperus rotundus* (Cyperaceae); henna, *Lawsonia inermis* (Lythraceae); garden cress, *Lepidium sativum* (Brassicaceae); oak bark, *Quercus cortex* (Fagaceae); castor, *Ricinus communis* (Euphorbiaceae); and rosemary, *Salvia rosmarinus* [syn.: *Rosmarinus officinalis*] (Lamiaceae). The aqueous extracts of plants were made with minor modifications (Govindarajan *et al*. 2016); 50 g of each plant was added to 300 ml of boiled water and the solution was kept for six hours in the dark, then the extract was sieved and then filtered. Serial dilutions were made from the previously described stock solution (170 mg/ml).

Adult immersion tests

The adult immersion tests (AIT) were made to test plant extracts' efficacy against *H. dromedarii* (Khater and Hendawy 2014). Ten males per replicate were used in each test and each tick group was immersed for 1 min. in 100 ml solution of each concentration. Seven concentrations were applied (10, 20, 40, 90, 110, 130, and 170 mg/ml) for each plant extract and diluted in distilled water. The immersed ticks were kept in a Petri dish containing a filter paper (Whatman n. 1). Three replicates were applied for each concentration and the control group was treated with distilled water. Petri dishes were kept at 27 ± 2 °C and 80 ± 5% relative humidity (RH). Tick mortalities (MO) were recorded up to 15 days PT.

Chemical analysis

Total phenolic contents

The total phenolic contents of the aqueous extracts were determined according to the method described in a previous protocol (Taie *et al.* 2015). The amount of total phenolics was calculated as gallic acid equivalents from a calibration curve. The standard curve was prepared using 500, 400, 350, 325, 300, 250, 225, 200, 150, 125, 100 and 50 mg/L solutions of gallic acid in methanol (y = 0.0009x; R2 = 0.9875). Total phenol values were expressed as gallic acid equivalents (mg GAE/g of dry mass). The test was conducted in triplicates.

Total flavonoid content

Total flavonoid contents were estimated (Taha *et al.* 2015). Total flavonoid values, expressed as quercetin equivalents (mg QE/g of dry mass), are a common reference compound for flavonoids. All tests were performed in triplicates.

Total tannin content

Total tannin was measured by modifying the Folin-Ciocalteu method using polyvinyl polypyrrolidone (PVPP) for separation of tannin phenols from non-tannin phenols. The difference between total and simple phenol values represented the entire tannin content, expressed as mg GAE g D. W (Taha *et al*. 2015).

Data analysis

The data were analyzed using SPSS V23 (IBM, USA), the mortalities were compared via One-Way Analysis of Variance (ANOVA) followed by the Tukey's test. A p-value of > 0.05 is non-significant. The lethal concentration (LC) and time (LT) values were calculated through the Probit analysis. Mortalities were corrected according to the following equation (Abbott 1925):

$$\text{Corrected Mortality\%} = (\text{MT\%} - \text{MC\%}) / (100 - \text{MC\%}) \times 100$$

where MT is the number of dead ticks in the treated group and MC is the number of dead ticks in the control group.

Relative toxicity was calculated (Khater and Geden 2018) according to this formula:

RT = LT50 (LC_{90}, LC_{95}, or LT_{99}) for *Lawsonia inermis* plant/ LT50 (LC_{90}, LC_{95}, or LT_{99}) for their plant extract

RESULTS

Adult immersion tests

The efficacy of nine plant extracts was evaluated against *H. dromedarii*, semi-fed adults. Plants could be arranged according to their Mortality percent (MO%), 15 days PT with 170 mg/ml, as follows: 96% by *Ricinus communis*; followed 84% by *Alc. vulgaris*, *Ci. endivia*, *Q. cortex*, and *S. rosmarinus*; and 80% by *All. sativum*, *Cy. rotundus*, and *Le. sativum*; and finally, 76% by *La. inermis* as the least effective extract (Table 1).

Three, seven, and 12 days PT, the LC_{50} values of the highly effective group were calculated for *R. communis* (100.11, 30.34, and 30.22), *Alc. vulgaris* (140.11, 60.94, and 50.38), *Ci. endivia* (120.56, 60.13, and 30.80), *Q. cortex* (110.87, 50.05, and 30.13), and *S. rosmarinus* (110.73, 50.26, and 40.35). Furthermore, the LC_{99} values of *R. communis* (440.16, 380.61 and 340.55), *Alc. vulgaris* (650.11, 430.87, and 410.21), *Ci. endivia* (480.31, 470.85, and 390.86), *Q. cortex* (460.41, 370.49, and 370.00), and *S. rosmarinus* (530.58, 360.52, and 350.15) indicated a time-dependent effect (Tables 2–4).

Lawsonia inermis was considered the reference substance as it had the highest LC_{50} value 12 days PT and the relative toxicity of the applied *Alc. vulgaris*, *All. sativum*, *Ci. endivia*, *Cy. rotundus*, *Le. sativum*, *Q. cortex*, *R. communis*, and *S. rosmarinus* were 1.0, 1.6, 1.6, 1.2, 1.3, 1.7, 1.7, and 1.2 times, respectively, more effective than *La. inermis* (Table 5). PT with 170 mg/ml, the LT50 values for the highly effective plants were 1.000, 3.136, 2.282, 4.498, and 2.614 days, respectively, for *R. communis*, *Alc. vulgaris*, *Ci. endivia*, *Q. cortex*, and *S. rosmarinus* and their LT_{99} values were 9.224, 19.840, 19.296, 22.928, and 29.907 days, respectively (Table 6).

Table 1. Mortality percentage of the nine plant extracts against *Hyalomma dromedarii*.

Plant extract	**Day**	**Concentration (mg/ml)**						
		170	**130**	**110**	**90**	**40**	**20**	**10**
Alchemilla vulgaris	1	20.00a	20.00a	20.00a	16.67ab	10.00b	10.00b	10.00b
	2	40.00a	36.67ab	36.67ab	33.33ab	26.67ab	23.33b	26.67ab
	3	48.29a	44.85a	44.85a	41.40a	37.95a	34.51a	34.51a
	5	57.17a	53.60a	46.47ab	46.47ab	42.90ab	35.76b	32.19b
	7	66.67a	59.26a	55.56a	51.85ab	51.85ab	37.04b	37.04b
	9	74.44a	62.96b	59.26b	51.85bc	51.85bc	51.85bc	40.74c
	12	76.19a	72.22a	68.25a	68.25a	64.29a	52.38b	44.44b
	15	84.41a	76.02ab	76.02ab	76.02ab	72.02abc	56.04bc	52.04c
Allium sativum	1	23.33a	20.00ab	16.67abc	16.67ab	13.33abc	10.00bcd	6.67cd
	2	36.67a	33.33ab	30.00abc	30.00abc	26.67abc	20.00bc	16.67c
	3	55.19a	51.74a	51.74a	48.29ab	44.85ab	34.51bc	24.16c
	5	60.99a	53.90ab	53.90ab	50.35ab	46.81ab	39.72bc	29.08c
	7	63.33a	60.00ab	60.00ab	60.00ab	56.67ab	50.00ab	43.33b
	9	70.37a	66.67ab	66.67ab	62.96ab	59.26abc	51.85bc	44.44c
	12	80.02a	64.03ab	64.03ab	64.03ab	56.04bc	52.04bc	44.04c
	15	80.02a	68.03ab	68.03ab	64.03abc	60.03bc	56.04bc	48.04c
Cichorium endivia	1	20.00a	16.67ab	13.33abc	13.33abc	10.00abc	6.67bcd	3.33cd
	2	36.67a	30.00ab	23.33abc	20.00bc	20.00bc	16.67bc	10.00cd
	3	55.19a	48.29ab	44.85abc	41.40abc	37.95bc	31.06cd	20.72d
	5	60.74a	50.04ab	50.04ab	46.47ab	46.47a b	35.76b	32.19b
	7	66.67a	59.26ab	55.56ab	51.85abc	44.44bc	44.44bc	37.04c
	9	74.07a	66.67ab	62.96ab	59.26b	55.56b	55.56b	40.74c
	12	76.02a	68.03ab	64.03ab	60.03abc	60.03abc	56.04bc	44.04c
	15	84.01a	72.02ab	68.03abc	64.03bc	64.03bc	13.33ab	52.04c
Cyperus rotundus	1	23.33a	16.67ab	13.33abc	13.33abc	13.33ab	23.33ab	6.67bc
	2	36.67a	33.33a	23.33abc	20.00bc	30.00ab	37.95ab	20.00b
	3	48.29a	48.29a	44.85abc	41.40abc	37.95ab	42.90a	31.06b
	5	57.17a	57.17a	50.04ab	46.47ab	39.33a	48.15ab	35.76a
	7	62.96a	59.26ab	55.56ab	51.85abc	48.15ab	48.15cd	40.74b
	9	74.07a	66.67ab	62.96ab	59.26b	55.56bcd	48.04b	44.44d
	12	80.02a	64.03b	64.03ab	60.03abc	56.04b	56.04b	48.04b
	15	80.02a	68.03ab	68.03abc	64.03bc	60.03b	13.33ab	56.04b
Lawsonia inermis	1	20.00a	13.33ab	13.33abc	6.67bc	6.67bc	6.67bc	3.33cd
	2	33.33a	26.67ab	23.33abc	20.00bc	20.00bc	20.00bc	13.33cd
	3	48.29a	44.85ab	41.40ab	41.40abc	34.51bc	27.61cd	20.72b

Mortality percentages followed by the same small letter within the same row were not significantly different (P > 0.05).

Table 1. Continued.

Plant extract	Day	Concentration (mg/ml)						
		170	130	110	90	40	20	10
Lawsonia inermis	5	53.60a	50.04ab	50.04ab	46.47ab	39.33ab	35.76b	28.62d
	7	59.26a	55.56ab	51.85ab	48.15abc	44.44bc	40.74bc	33.33c
	9	66.67a	59.26ab	59.26ab	51.85b	51.85b	48.15b	37.04c
	12	68.03a	64.03ab	60.03ab	52.04abc	52.04abc	52.04bc	36.05c
	15	76.02a	68.03ab	64.03abc	60.03bc	60.03bc	56.04bc	48.04c
Lepidium sativum	1	20.00a	16.67ab	16.67ab	13.33ab	13.33ab	6.67bc	6.67bc
	2	36.67a	30.00ab	30.00ab	30.00ab	30.00ab	16.67bc	13.33dc
	3	51.89a	41.58ab	45.02ab	41.58abc	34.71abc	27.84bc	24.40c
	5	57.17a	53.60ab	53.60ab	46.47abc	46.47abc	35.76bc	28.62c
	7	66.67a	62.96a	59.26ab	55.56abc	48.15bcd	44.44cd	37.04d
	9	74.07a	70.37ab	59.26abc	66.67ab	55.56bcd	48.15cd	40.74d
	12	76.02a	60.03bc	60.03bc	56.04bc	56.04bc	44.04bc	40.05c
	15	80.02a	64.03ab	64.03ab	60.03ab	60.03ab	52.04b	48.04b
Quercus cortex	1	30.00a	23.33ab	20.00abc	16.67bcd	13.33bcd	10.00cde	6.67de
	2	36.67a	36.67a	30.00abc	26.67abc	23.33bcd	16.67cd	13.33d
	3	55.19a	51.74a	48.29ab	44.85abc	34.51bcd	31.06cd	24.16d
	5	64.31a	53.60ab	53.60ab	53.60ab	39.33bc	35.76bc	32.19c
	7	70.37a	66.67ab	66.67ab	62.96ab	51.85bc	44.44c	40.74c
	9	74.07a	70.37ab	66.67ab	66.67ab	62.96ab	59.26ab	48.15b
	12	80.02a	68.03ab	68.03ab	64.03ab	56.04b	60.03ab	48.04b
	15	84.01a	72.02ab	72.02ab	68.03b	64.03bc	60.03bc	52.04c
Ricinus communis	1	26.67a	23.33ab	23.33ab	20.00ab	16.67ab	16.67ab	13.33b
	2	40.00a	36.67ab	36.67ab	30.00abc	23.33bc	23.33bc	20.00c
	3	62.08a	55.19ab	51.74abc	48.29bc	41.40cd	34.51d	34.51d
	5	67.88a	57.17ab	57.17bc	42.90cd	42.90cd	46.47bcd	39.33d
	7	74.07a	70.37ab	66.67ab	62.96ab	55.56ab	51.85bc	37.04c
	9	77.78a	74.07ab	70.37a	66.67a	59.26ab	59.26ab	44.44b
	12	88.01a	72.02ab	72.02ab	64.03bc	60.03bc	60.03bc	52.04c
	15	96.00a	80.02ab	80.02ab	72.02bc	68.03bc	64.03bc	60.03c
Salvia rosmarinus	1	26.67a	20.00ab	20.00ab	13.33bc	13.33bc	13.33bc	6.67cd
	2	43.33a	33.33ab	33.33ab	26.67ab	26.67ab	26.67ab	23.33b
	3	51.74a	51.74a	48.29a	48.29a	41.40ab	34.51b	31.06b
	5	67.88a	53.60ab	53.60ab	46.47ab	46.47ab	39.33ab	32.19b
	7	74.07a	66.67ab	62.96ab	59.26abc	55.56bc	44.44cd	37.04d
	9	74.07a	70.37a	66.67a	62.96a	59.26ab	44.44bc	40.74c
	12	80.02a	68.03ab	68.03ab	60.03bc	60.03bc	52.04c	48.04c
	15	84.01a	72.02ab	72.02ab	68.03bc	68.03bc	56.04d	52.04d

Mortality percentages followed by the same small letter within the same raw were not significantly different (P > 0.05).

Table 2. Lethal concentration values (mg/ml) of regulatory bioassays three days post exposure of *Hyalomma dromedarii* semi-fed adults to plant extracts.

Plant	LC_{50} (Upper–Lower)	LC_{90} (Upper–Lower)	LC_{95} (Upper–Lower)	LC_{99} (Upper–Lower)	Regression equation	X^2 (Sig.)	R^2
Alchemilla vulgaris	140.11 (10.17–26.12)	420.20 (28.69–97.93)	500.17 (33.67–18.56)	650.11 (42.99–157.28)	Y = 0.82 + 0.06x	11.45 (0.075a)	0.40
Allium sativum	100.84 (5.26–45.64)	330.36 (20.49–268.29)	390.74 (23.96–332.25)	51.72 (30.39–452.32)	Y = 0.79 + 0.07x	12.80 (0.046a)	0.47

X^2 (Sig.): Chi square (significance).

Table 2. Continued.

Plant	LC_{50} (Upper–Lower)	LC_{90} (Upper–Lower)	LC_{95} (Upper–Lower)	LC_{99} (Upper–Lower)	Regression equation	X^2 (Sig.)	R^2
Cichorium endivia	120.56 (9.83–17.65)	320.25 (24.43–51.97)	370.84 (28.36–61.91)	480.31 (35.69–80.60)	Y = 0.97 + 0.08x	8.93 (0.177a)	0.60
Cyperus rotundus	130.29 (9.67–22.82)	390.83 (27.64–85.02)	470.36 (32.46–102.93)	610.48 (41.45–136.56)	Y = 0.82 + 0.06x	11.84 (0.066a)	0.42
Lawsonia inermis	120.26 (9.24–18.42)	34.90 25.48– 62.53)	410.32 (29.82–75.30)	530.36 (37.91–99.30)	Y = 0.87 + 0.07x	12.26 (0.056a)	0.49
Lepidium sativum	130.11 (10.22–18.86)	330.53 (25.13– 55.57)	390.32 (29.15–66.18)	500.17 (36.67–86.11)	Y = 0.98 + 0.08x	8.00 (0.238a)	0.59
Quercus cortex	110.87 (9.31–16.31)	300.90 (23.65– 48.43)	360.29 (27.49–57.76)	460.41 (34.65–75.31)	Y = 0.96 + 0.08x	8.54 (0.201a)	0.06
Ricinus communis	100.11 (7.70–13.64)	280.87 (22.24– 44.51)	340.19 (26.06–53.56)	440.16 (33.17–70.60)	Y = 0.86 + 0.08x	11.45 (0.106a)	0.57
Salvia rosmarinus	110.73 (8.72–17.66)	340.79 (25.30– 63.10)	410.32 (29.70–76.28)	530.58 (37.90–101.05)	Y = 0.83 + 0.07x	11.79 (0.067a)	0.47

X^2 (Sig.): Chi square (significance).

Table 3. Lethal concentration values (mg/ml) of regulatory bioassays seven days post exposure of *Hyalomma dromedarii* semi-fed adults to plant extracts.

Plant	LC_{50} (Upper–Lower)	LC_{90} (Upper–Lower)	LC_{95} (Upper–Lower)	LC_{99} (Upper–Lower)	Regression equation	X^2 (Sig.)	R^2
Alchemilla vulgaris	60.94 (4.02–9.78)	270.23 (20.65–43.89)	320.99 (24.70–54.22)	430.78 (32.24–73.66)	Y = 0.51 + 0.07x	11.13 (0.085a)	0.593
Allium sativum	60.51 (−50.17–25.90)	30.68 (18.27–1577.23)	370.53 (21.88–2032.81)	500.38 (28.51–2887.53)	Y = 0.42 + 0.06x	13.50 (0.036a)	0.471
Cichorium endivia	60.131 (−94.26–22.05)	290.116 (17.38 –2113.66)	350.632 (20.83–2734.81)	470.854 (27.14–3900.12)	Y = 0.42 + 0.06x	14.904 (0.021a)	0.478
Cyperus rotundus	60.70 (−49.45–31.42)	310.15 (18.50–2031.45)	380.09 (22.14–2614.06)	510.09 (28.84–3707.07)	Y = 0.43 + 0.06x	13.28 (0.039a)	0.471
Lawsonia inermis	50.88 (3.27–8.13)	23.05 (18.19–33.54)	270.92 (21.79–41.37)	370.05 (28.47–56.13)	Y = 0.51 + 0.08x	11.04 (0.087a)	0.662
Lepidium sativum	60.25 (3.25–8.90)	26.04 (19.88–41.18)	310.65 (23.87–51.07)	420.17 (31.28–69.67)	Y = 0.48 + 0.07x	11.57 (0.072a)	0.592
Quercus cortex	50.05 (−4.07–9.82)	220.92 (15.34–69.37)	270.99 (18.43 –88.66)	370.49 (24.07–125.02)	Y = 0.44 + 0.08x	13.15 (0.041a)	0.605
Ricinus communis	30.34 (−51.0 –9.16)	220.77 (14.05–263.32)	280.28 (17.14–350.75)	380.61 (22.70–514.97)	Y = 0.3 + 0.07x	17.69 (0.007a)	0.501
Salvia rosmarinus	50.26 (2.47–7.49)	220.48 (17.71–32.80)	270.36 (21.32–40.69)	360.52 (28.01–55.57)	Y = 0.47 + 0.08x	12.43 (0.053a)	0.636

X^2 (Sig.): Chi square (significance).

Table 4. Lethal concentration values (mg/ml) 12 days post exposure of *Hyalomma dromedarii* semi-fed adults to plant extracts.

Plant	LC_{50} (Upper–Lower)	LC_{90} (Upper–Lower)	LC_{95} (Upper–Lower)	LC_{99} (Upper–Lower)	Regression equation	X^2 (Sig.)	R^2
Alchemilla vulgaris	50.38 (−8.29–11.62)	250.12 (16.04–122.91)	300.71 (19.25–158.15)	410.21 (25.13–224.40)	Y = 0.43 + 0.07x	14.14 (0.028a)	0.556
Allium sativum	30.98 (−36.34–10.12)	230.86 (14.77–239.50)	290.49 (17.91–315.87)	400.06 (23.61–459.33)	Y = 0.33 + 0.07x	16.84 (0.010a)	0.503
Cichorium endivia	30.80 (−362.62–10.64)	230.67 (14.30–2074.95)	290.30 (17.37–2763.94)	390.86 (22.90–4056.59)	Y = 0.32 + 0.07x	19.14 (0.004a)	0.477
Cyperus rotundus	40.64 (−8.53–9.83)	230.29 (15.16–91.90)	280.58 (18.28–118.77)	380.51 (23.96–169.32)	Y = 0.4 + 0.07x	14.22 (0.027a)	0.571
Lawsonia inermis	50.55 (−6.95–11.79)	250.54 (16.38–117.33)	310.21 (19.64–150.60)	410.84 (25.62–213.15)	Y = 0.43 + 0.07x	13.61 (0.034a)	0.553
Lepidium sativum	40.28 (−5.56–8.65)	200.78 (14.03–59.36)	25.46 (16.94–76.38)	340.24 (22.21–108.49)	Y = 0.4 + 0.08x	14.44 (0.025a)	0.617
Quercus cortex	30.13 (−44.03–8.75)	210.79 (13.53–208.47)	270.08 (16.54–278.41)	370.00 (21.93–409.84)	Y = 0.29 + 0.07x	18.47 (0.005a)	0.507
Ricinus communis	30.22 (−25.00–8.47)	200.48 (12.97–122.86)	250.37 (15.84–163.19)	340.55 (20.96–239.09)	Y = 0.31 + 0.08x	19.04 (0.004a)	0.537
Salvia rosmarinus	40.53 (−5.10–9.03)	210.40 (14.40–62.54)	260.18 (17.35–80.30)	350.15 (22.72–113.77)	Y = 0.42 + 0.08x	14.14 (0.028a)	0.614

X^2 (Sig.): Chi square (significance).

Table 5. The relative effect of the applied plant extracts against *Hyalomma dromedarii*.

Plant	12 days post treatments
Alchemilla vulgaris	1.0
Allium sativum	1.6
Cichorium endivia	1.6
Cyperus rotundus	1.2
Lawsonia inermis[*]	1.0
Lepidium sativum	1.3
Quercus cortex	1.7
Ricinus communis	1.7
Salvia rosmarinus	1.2

[*] The reference plant extract is *Lawsonia inermis*.

Table 6. Lethal times (days) of bioassays post exposure of *Hyalomma dromedarii* to plant extracts.

Plant	LT_{50} (Upper–Lower)	LT_{90} (Upper–Lower)	LT_{95} (Upper–Lower)	LT_{99} (Upper–Lower)	Regression equation	X^2 (Sig.)	R^2
Alchemilla vulgaris	3.136 (−1.064–5.328)	20.725 (9.369–12.338)	14.947 (26.188–11.230)	19.840 (36.584–14.569)	Y = 0.41 + 0.13x	907[a]	0.885
Allium sativum	4.928 (7.106–2.005)	13.997 (10.789–22.708)	16.568 (27.794–12.616)	21.391 (15.941–37.437)	Y = 0.72 + 0.14x	16.84 (0.010a)	0.940

X^2 (Sig.): Chi square (significance).

Table 6. Continued.

Plant	LT_{50} (Upper–Lower)	LT_{90} (Upper–Lower)	LT_{95} (Upper–Lower)	LT_{99} (Upper–Lower)	Regression equation	X^2 (Sig.)	R^2
Cichorium endivia	2.282 (−2.963–4.592)	20.318 (8.736–11.655)	14.312 (26.203–10.627)	19.296 (13.989–37.425)	Y = 0.28 + 0.13x	19.14 (0.004a)	0.864
Cyperus rotundus	2.828 (−1.950–5.112)	12.385 (9.314–21.564)	15.094 (11.225–27.510)	20.176 (14.648–38.826)	Y= 0.36 + 0.13x	14.22 (0.027a)	0.752
Lawsonia inermis	3.639 (6.160–1.399)	14.603 (10.790–27.792)	17.712 (12.891–35.278)	23.542 (49.458–16.696)	Y = 0.43 + 0.12x	13.61 (0.034a)	0.942
Lepidium sativum	3.257 (0.070–5.130)	10.923 (17.438–8.420)	21.713 (10.001–13.096)	17.172 (12.841–29.858)	Y= 0.51 + 0.16x	14.44 (0.025a)	0.977
Quercus cortex	4.498 (0.791–6.873)	14.651 (11.061–25.540)	17.529 (13.055–31.749)	22.928 (16.679–43.513)	Y = 0.56 + 0.12x	18.47 (0.005a)	0.955
Ricinus communis	1.000 (−3.229–2.448)	5.530 (4.036–10.148)	6.815 (5.001–13.425)	9.224 (6.629–19.756)	Y = 0.2 + 0.26x	19.04 (0.004a)	0.947
Salvia rosmarinus	2.614 (−1.183–4.583)	12.733 (9.672–21.373)	12.733 (9.672–21.373)	29.907 (12.591–16.926)	Y = 0.38 + 0.15x	14.14 (0.028a)	0.959

X^2 (Sig.): Chi square (significance).

The phytochemical analyses indicated variations in the total phenolic, total flavonoid, and total tannin contents. Among all investigated plant extracts, *R. communis* possessed the highest total phenolic contents (95.50 ± 0.17mg/g), while *Q. cortex* contained the highest flavonoids content (70.78 ± 0.17mg QE/g). On the other hand, *L. inermis* extract contained a high concentration of total tannins reaching 75.38 ± 0.21 (mg /g) (Table 7).

Table 7. Total contents of active compounds in the investigated plants.

Plant Name	Total phenolic (mg GAE/g)	Total flavonoid (mg QE/g)	Total tannin (mg GAE /g)
Alchemilla vulgaris	35.47 ± 0.19	26.1 ± 0.12	24.8 ± 0.60
Allium sativum	35.53 ± 0.27	31.33 ± 9.02	0.093 ± 1.96
Cichorium endivia	5.45 ± 0.17	1.6 ± 0.08	N.D
Cyperus rotundus	0.55 ± 0.009	0.11 ± 0.007	0.40 ± 0.010
Lawsonia inermis	66.13 ± 0.13	48.34 ± 0.22	75.38 ± 0.21
Lepidium sativum	43.67 ± 0.11	19.9 ± 0.14	4.9 ± 0.09
Quercus cortex	55.59 ± 0.33	70.78 ± 0.17	35.26 ± 0.14
Ricinus communis	95.50 ± 0.17	46.34 ± 0.23	15.53 ± 0.17
Salvia rosmarinus	46.54 ± 0.12	10.7 ± 0.17	N.D

N.D: not detected

DISCUSSION

Botanicals effectively control pests (Iqbal *et al*. 2021) and the present study investigated the *in vitro* acaricidal effect of plant extracts against *H. dromedarii* and the results showed that the used extracts induced a considerable acaricidal effect. *Ricinus communis* was the most effective one followed by *Alc. vulgaris*, *Ci. endivia*, *Q. cortex*, and *S. rosmarinus* inducing high tick MO%, while the least effective one was *L. inermis* which caused 76% mortality after 15 days PT with the highest concentration (170 mg/ml).

The lethal effect of the applied materials indicated a dose concentration- and exposure time -

dependent relationship. A similar finding was previously reported (Khater and Hendawy 2014; Eltaly *et al.* 2022; Hegazy *et al.* 2022).

This study showed that *R. communis* was the most effective extract, 96% MO 15 days PT with 170 mg/ml. Similar results were recorded against *Rhipicephalus microplus* for instance 100 mg/ml caused 95% mortality within 14 days PT (Zahir *et al.* 2009). *Ricinus communis* also had an acaricidal effect (73%) against *Rhipicephalus decoloratus* and *Rhipicephalus pulchellus* 24h PT, 100 mg/ml (Kemal *et al.* 2020).

Besides its acaricidal effect, *R. communis* has been shown to possess insecticidal activity against *Callosobruchus maculatus* (Coleoptera: Chrysomelidae: Bruchinae) affecting adult survival 100%, 14 days PT (Hussein *et al.* 2016), and larvicidal activity against mosquito species, *Culex pipiens*, *Aedes caspius*, *Culiseta longiareolata*, and *Anopheles maculipennis* (Aouinty *et al.* 2006).

Ricinus communis, in this study, possessed very high contents of total phenolic and flavonoid compounds (95.5 mg GAE/g and 46.34 mg QE/g, respectively) and a moderate amount of total tannin (15.53 mg GAE/g); such compounds might be attributed to its high acaricidal effect. A similar study showed that the insecticidal activity of *R. communis*is extract might be because of protein, ricins, and alkaloid ricinine, which are lethal at very low concentrations **(**Kodjo *et al*. 2011).

The second most effective plants in this work (84% MO, 15 days PT with 170 mg/ml) were *Alc. vulgaris* and *Q. cortex*, and their LC_{50} values 12 days PT were 50.38 and 30.13%, while their LC_{99} values were 410.21 and 370.00%, respectively. Interestingly, the acaricidal and insecticidal activity of these plants has been reported for the first time, according to our knowledge.

Alchemilla vulgaris, in this study, contained a reasonable amount of total phenolic, tannin, and flavonoid contents: 35.47, 24.8 mg GAE/g, and 26.1 mg QE/g, respectively. Likewise, the methanol extract of *Alc. vulgaris* contains four flavonoid glycosides and its known pharmacological properties of flavonoids could explain its usage in popular medicine (D'Agostino *et al.* 1998). In the same way, *Alc. vulgaris* exert several biological activities as it contains numerous compounds such as phenols, tannins, flavonoids, and phenolcarboxylic acids (Tadić *et al*. 2020). *Quercus cortex*, in this work, contained very high contents of total phenolic, tannin, and flavonoid compounds (55.59, 35.26 GAE /g, and 70.78 QE/g respectively), which might be attributed to its lethal effects.

The present work demonstrated that *S. rosmarinus* caused 84% MO against adult ticks, 15 days PT with 170 mg/ml concentration. A similar finding was recorded as over 85% mortality of *R. microplus* larvae was recorded at concentrations 10 and 20% which decreased to 40% at a concentration of 5%, and zero mortality at concentrations 2.5 and 1.25% (Martinez-Velazquez *et al*. 2011). Rosemary was also effective against mosquito larvae (Shalaby and Khater 2005). In contrast to our result, rosemary had no acaricidal effect against *Dermatophagoides farinae* and *D. pteronyssinus* (0.1 μl/cm2 for 5 min.) (Rim and Jee 2006). It worth mentioning that rosemary oils controlled the buffalo louse, *Haematopinus tuberculatus* (Khater *et al*. 2009) and *Lucilia sericata* larvae (Khater *et al*. 2011).

Lepidium sativum was less effective against ticks in this study, 80% mortality PT with 170 mg/ml. Similarly, its oil revealed moderate larvicidal activity against *Culex pipiens* larvae and its major compounds were β-caryophyllene, eugenol, eucalyptol, α-terpinyl acetate, and (E)-anethole (Kimbaris *et al*. 2012). Comparably, relatively high repellency (76.68%) of *Le. sativum* oil on two-spotted spider mite, *Tetranychus urticae* Koch infesting mulberry trees was recorded (El-Yamani *et al*. 2017). Chemical analysis of *Le. sativum* in the present work indicated the presence of total phenolic, tannin, and flavonoid compounds (43.67, 4.9 GAE/g, and 19.9 QE/g, respectively). In contrast, another study revealed that *Le. sativum* seeds contained lower amounts of alkaloids (0.40%), flavonoids (0.42%), saponins (2.8%), tannins (0.61%), and phenols (0.004%) (Al-Snafi, 2019b).

Allium sativum induced high mortality 80.02%, at a concentration of 170 mg/ml, 15 days PT in this investigation. LC_{50} values were 100.480, 6.51, and 3.98% after three, nine, and 12 days PT, respectively. Equivalently, higher mortality of engorged females *Boophilus annulatus* reached

100% after application of 10 and 20% concentrations of garlic oil within 48 and 24 h, respectively (Aboelhadid *et al*. 2013).

In another study, *All. sativum* had high acaricidal (100% MO) bioactivity against *Hyalomma marginatum rufipes* in less than an hour (Nchu *et al*. 2005) and reduced the severity of the *R. microplus* infestation (Costa-Júnior and Furlong 2011). High mortalities of *Riphicephalus microplus* larvae reached 90% PT with 2.5% (Martinez-Velazquez *et al*. 2011). Also, *All. sativum* and *Zingiber officinale* extracts induced 100% larvicidal effect at 2.5% concentration against the brown dog tick, *Rhipicephalus sanguineus* (Jeyathilakan *et al*. 2019).

Similar insecticidal activity of garlic oil was exhibited with 100% mortality against *Cephalopina titillator*, 24h PT (Khater 2014) and more toxicity to *Callosobruchus maculatus* (Denloye 2010). *Allium sativum* in this study contained high content of total phenolic (35.53, 93.13 mg GAE/g) and flavonoid and tannin compounds (0.31 mg QE/g and 0.93 mg GAE/g, respectively) which could cause acaricidal effects. In contrast, another study revealed that *All. sativum* contains lower amounts of alkaloids (7.20 ± 0.05), saponins (4.30 ± 0.02); and tannins (4.80 ± 0.03) (Ali and Ibrahim 2019).

Cichorium endivia induced 84% mortality, 15 days PT with 170 mg/ml in this work. It also had insecticidal activity against mosquitoes and houseflies (Mansour *et al*. 2014) and exhibited strong larvicidal activity against larvae of *Anopheles stephensi*, *Aedes aegypti*, and *Culex quinquefasciatus* (Ali *et al*. 2018).

Cyperus rotundus, in this study, exhibited 80% MO, 15 days PT with 170 mg/ml, and contained a large amount of active ingredients. Likewise, it was effective against *Blattella germanica* (L.) (Chang *et al*. 2012). Another study showed that the oil of *Cy. rotundus* has major compounds like monoterpenoids 1,8-cineole, *p*-cymene, α-Cyperone, α-pinene, β-pinene, caryophyllene oxide, terpinen-4-ol, β-pinene, and myrtenal (Lawal and Oyedeji 2009). *Cyperus rotundus* essential oil contains (62%) cyperene, *α*-cyperone, isolongifolen-5-one, retention, and cyperorotundene which exhibit antioxidant, cytotoxic, and apoptotic properties (Kilani *et al*. 2008).

Lawsonia inermis was the least effective with plants in the present study, 76% MO 15 days PT. In contrast, similar larvicidal activity of *L. inermis* was recorded against the mosquitoes, *Culex* spp. (Adam 2015), the red flour beetles (Biswas *et al*. 2016), and the Egyptian cotton leafworm, *Spodoptera littoralis*, with enzyme alterations in phenoloxidase, chitinase, protease, alkaline, and acid phosphatases and disturbances in protein levels (Abd-El razzik *et al*. 2018). *Lawsonia inermis*, in the present work, contained high contents of total phenolic, tannin, and flavonoid compounds (66.13, 75.38 GAE/g, and 48.34 QE/g, respectively) which could explain its acaricidal effect. In the same way, *L. inermis* contains phenols, flavanoids, tannins, saponins, alkaloids, terpenoids, quinones, coumarins, and xanthones (Al-Snafi 2019a).

Some other plants such as *Araucaria heterophylla* and *Commiphora molmol* (Baz *et al.* 2022b), *Vitex castus* and *Zingiber officinale* (Eltaly *et al.* 2022); and *Saussurea costus* (Hegazy *et al.* 2022) had effect on *Hyalomma dromedarii*. Moreover, *Taraxacum officinale* (40 mg/mL) exhibit larvicidal activity (96.6%) against the *R. microplus* larvae (Khan *et al*. 2019). Different findings between this study and the other studies might be attributed to the use of different pests, plant extracts, and locality. It is proved that phenols, flavonoids, and tannins could be used for pest management (Rattan 2010).

Similarly, it is recorded that plant compounds like flavonoids, esters, alkaloids, glycosides, and fatty acids control insects in various ways (Baz *et al*. 2021, 2022b) as toxicants, feeding deterrents/antifeedants, growth retardants, repellents, chemosterilants, and attractants (Khater 2012; Khater and Geden 2018; Hegazy *et al.* 2022; Khater *et al.* 2022; Mohammed *et al.* 2022).

CONCLUSION

Because of the global efforts for reducing the reliance on the synthetic pesticides, this study was conducted and revealed novel and potential acaricidal effect of nine aqueous extracts against *H. dromedarii* as effective, environmentally acceptable, inexpensive alternative approaches for controlling ticks. The present study recommends using *R. communis* for tick control, followed by *Alc. vulgaris*, *Ci. endivia*, *Q. cortex*, and *S. rosmarinus*. The ecotoxological studies of improved formulations of the extracts are recommended for future studies.

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اثر کنه‌کشی جدید نه گیاه مصری در برابـر کنـهٔ شـتر، *Hyalomma dromedarii* (Ixodida: Ixodidae)

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چکیده

کنه‌ها انگل‌های خارجی بسیار مهمی هستند که بیماری‌ها را به حیوانات و انسان‌ها منتقل می‌کنند و مهار طبیعی آنها نیازی فوری است. اثربخشی نه عصاره آبی جدید گیاهی در برابر کنه‌های کامل نیمه تغذیه شدهٔ *Hyalomma dromedarii* ، از طریق آزمون غوطه‌ورسازی کنهٔ کامل بررسی شد و هفت غلظت (۱۰، ۲۰، ٤۰، ۹۰، ۱۱۰، ۱۳، و ۱۷۰ میلی‌گرم در میلی‌لیتر) برای هر کدام اعمال شد. عصاره گیاهی گیاهان را می‌توان بر اساس درصد مرگ، ۱۵ روز پس از تیمار (PT) با مقدار ۱۷۰ میلی‌گرم در میلی‌لیتر، به شرح زیر مرتب کرد: ۹٦٪ توسط *Ricinus communis*؛ به دنبال آن ۸٤٪ *Alchemilla vulgaris*، *Cichorium endivia*، *Quercus cortex* و *Salvia rosmarinus* قرار گرفتند. سه، هفت و ۱۲ روز پس از تیمار، مقادیر LC50 گروه بسیار موثر شامل *R. communis* (۱۰۰/۱۱، ۳۰/۳٤ و ۳۰/۲۲٪)، *A. vulgaris* (۱٤۰/۱۱، ٦۰/۹٤ و ٥۰/۳۸٪)، *C. endivia* (۱۲۰/٥٦، ٦۰/۱۳ و ۳۰/۸۰ درصد)، *Q. cortex* (۱۱۰/۸۷، ٥۰/۰٥ و ۳۰/۱۳ درصد) و *S. rosmarinus* (۱۱۰/۷۳، ٥۰/۲٦ و ٤۰/۳٥ درصد) محاسبه شدند. مقادیر LT50 برای گیاهان بسیار موثر ۱/۰۰۰، ۳/۱۳٦، ۲/۲۸۲، ٤/٤۹۸ و ۲/٦۱٤ روز، پس از تیمار با ۱۷۰ میلی گرم در میلی لیتر، برای *R. communis*، *A. vulgaris*، *C. endivia*، *Q. cortex*، و *S. rosmarinus* و مقادیر LT99 آنها به ترتیب ۹/۲۲٤، ۱۹/۸٤۰، ۱۹/۲۹٦، ۲۲/۹۲۸ و ۹۰۷ روز بود. سمیت نسبی *A. vulgaris*، *Allium sativum*، *C. endivia*، *Cyperus rotundus* ، *Lepidium sativum*، *Q. cortex*، *R. communis* و *S. rosmarinus* به ترتیب ۱/۰، ۱/٦، ۱/٦، ۱/۲، ۱/۳، ۱/۷، ۱/۷ و ۱۲ برابر موثرتر از *Lawsonia inermis* بودند. آنالیزهای فیتوشیمیایی وجود ترکیبات فنلی، تانن و فلاونوئیدی را نشان داد. این بررسی استفاده از *R. communis* و پس از آن *Alc. vulgaris* ، *C. endivia*، *Q. cortex*، و *S. rosmarinus* را برای مهار کنه توصیه می‌کند.

واژگان کلیدی: غوطه‌ورسازی کنهٔ کامل، *Alchemilla*، *Cichorium*، *Quercus*، *Ricinus*، *Salvia*.

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