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SCREENING OF INDIGENOUS MEDICINAL PLANTS FOR THEIR ACARICIDAL ACTIVITY AGAINST CATTLE TICKS UNDER *IN VIVO* CONDITION

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ABSTRACT: The acaricidal activity of leaf extracts of *Annona squamosa*, *Azadirachta indica* and *Calotropis procera* were tested against the cattle ticks under *in vivo* condition. Plants were extracted with methanol and water and the extracts were tested against the cattle ticks under *in vivo* condition. The aqueous and alcoholic extracts of *A. indica* showed maximum mortality rate of ticks followed by *A. squamosa* and *C. procera* when tested individually. In combination of plant extracts, hot water extracts of dried leaf powder showed 100% mortality of ticks on 5th day whereas ethanol and methanol extracts showed 83% and 80% of mortality respectively. Based on the above experimental results, it is confirmed that the selected plant materials possess more acaricidal activity against cattle ticks. Plant extracts in combinations are more effective than single drug used.

INTRODUCTION: Plants are used frequently for animal healthcare by different people around the world. Medicinal plants used extensively for primary health care treatment to make domestic animals productive and healthy. Ectoparasitic infestation is one of the major veterinary problems affecting livestock. Among ecto-parasites, ticks have been recognized as the notorious threat due to severe irritation, allergy, toxicosis and cause lowered productivity and mortality. Plants have the ability to synthesize chemical compounds that help them defend against attack from a wide variety of predators such as insects, fungi and herbivorous mammals.

QUICK RESPONSE CODE

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Present investigation deals with the study of acaricidal activity of *Annona squamosa*, *Azadirachta indica* and *Calotropis procera* against cattle ticks under *in vivo* condition.

MATERIALS AND METHODS:

Collection and preparation of plant materials:

The plant materials used for this study are the leaves of *Annona squamosa*, *Azadirachta indica*, *and Calotropis procera.* They are collected, shade dried and mechanically powdered. The leaf powder of selected plants was extracted with solvents like ethanol, methanol and water.

Preparation of extracts:

Cold extract: Both fresh and dried leaf powder were soaked in known volume of distilled water for 24 hours. The extract was filtered through muslin cloth and used for further analysis.

Hot extract: Pre weighed plant powder material was soaked in known volume of distilled water and

kept in hot water bath for 6-7 hours at 70°C and cooled it under room temperature. After 18 hours the extract is filtered using muslin cloth and the filtrate was stored for further use.

Preparation of combination of leaf extracts:

The fresh and dried leaf materials of all the three plants were extracted individually with different solvents. Different concentrations of plant extracts (ranging from 12% -24%) are prepared, combined together in 1:1:1 ratio and used to test the mortality rate of ticks under *in vivo* conditions.

***In vivo* study:**

The crude leaf extracts of *Annona squamosa, Azadirachta indica* and *Calotropis procera* were tested against the ticks infested cows. *In vivo* study was conducted on cow flock of kanjampatti village, pollachi, Coimbatore district, Tamil Nadu. Different concentrations and combination of plant extracts (from 12%- 24%) were tested against the ticks on selected area (Ventral abdomen and neck region) with the help of the hand sprayer. Three animals were used for each test and control group also maintained with 3 animals for evaluating the effect of extracts. Experiment was conducted during the month of March and April. The number of ticks was calculated by summation of the total ticks by counting method. After topical application of sample on the site, mortality was recorded at 1st, 3rd and 5th day respectively. Mortality rate was calculated by counting the number of ticks alive [1].

RESULTS AND DISCUSSIONS: Over the past few decades, plant extracts have been widely used to control pests, mosquitoes, ticks etc. It also possesses various bio-efficacies such as acaricidal, ovicidal and repellent activities. Control of ticks with chemical acaricides has become difficult due to resistance development. Toxicity and resistance problems of insecticides have directed us to find an alternate to use plants as acaricides. Plants produce several secondary metabolites to protect themselves from the continuous attack of naturally occurring pathogens, insects and pests.

The plant extracts of *A.indica, A.squamosa* and *C.procera* were evaluated for its acaricidal activity by spraying the extracts on the animals. The animals were grouped and tested with single plant extract as well as in combination of three plants extracts at the concentration of 24% which was performed well under *in vivo* conditions. The mortality of the ticks occurred after a day but the highest mortality was observed on 5th day. The hot and cold water extracts of leaf powder of *A. indica* showed highest mortality rate than other two plants tested (**Table 1**). Similar studies were carried out in the aqueous extract of *A. india* compared with synthetic drug abamectin and the result showed that 62.5% mortality rate was occurred in plant extracts at the concentration of 20% and 50.5% mortality in synthetic drug after 15 days [2].

TABLE 1: *IN VIVO* EFFECT OF AQUEOUS EXTRACTS OF FRESH AND DRIED LEAVES AGAINST CATTLE TICKS

S.no	Name of the plants extracts used	No of animals treated	Mortality rate in %					
			Day 1		Day 3		Day 5	
			Fresh leaf	Leaf powder	Fresh leaf	Leaf powder	Fresh leaf	Leaf powder
	Hot water extract							
1.	*Annona squamosa*	3	21	11	46	31	76	54
2.	*Azadirachta indica*	3	21	43	47	56	78	70
3.	*Calotropis procera*	3	11	7	29	28	51	42
	Cold water extract							
4.	*Annona squamosa*	3	21	7	44	22	64	39
5.	*Azadirachta indica*	3	22	31	38	42	72	72
6.	*Calotropis procera*	3	16	12	30	22	53	32
7.	Control	3	0	0	0	0	0	0

The hot water extracts of fresh leaves of *A.indica* and *A.squamosa* showed maximum mortality rate of 78% and 76% respectively on 5th day followed by cold water extracts. Among the three plants tested, *C. procera* showed minimum mortality effect in both cold and hot water extracts (**Fig. 1**).

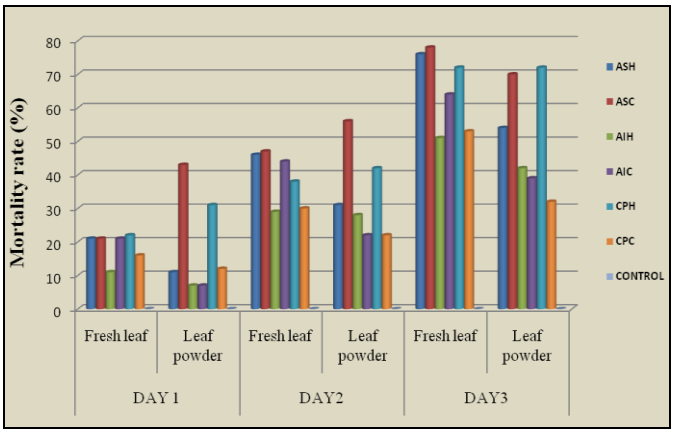

FIG.1: *IN VIVO* EFFECT OF AQUEOUS EXTRACTS OF FRESH AND DRIED LEAVES AGAINST CATTLE TICKS

ASH- *Annona squamosa* Hot water extract
ASC- *Annona squamosa* Cold water extract
AIH-*Azadirachta indica* Hot water extract
AIC-*Azadirachta indica* Cold water extract
CPH- *Calotropis procera* Hot water extract
CPC- *Calotropis procera* Cold water extract

Studies were conducted to findout the efficacy of alcoholic extracts of the selected plants. The mortality rate of ticks was found to be maximum in hot methanol (88%) and ethanol (86%) extracts of *A. indica* on 5th day (**Table 2**). The high mortality rate of *A. india* is due to the presence of azadirachtin compound present in the leaf extract (**Fig.2**). *A.indica* contains Azadirachtin, Nimbin and Nimbinin compounds possess acaricidal and insecticidal activity [3, 4, 5].

TABLE 2: *IN VIVO* EFFECT OF ALCOHOLIC EXTRACT OF LEAF POWDER AGAINST CATTLE TICKS

S.no	Name of the plants extracts used	No of animals treated	Mortality rate in %		
			Day 1	Day 3	Day 5
	Ethanol extract				
1.	*Annona squamosa*	3	28	64	80
2.	*Azadirachta indica*	3	29	68	86
3.	*Calotropis procera*	3	15	38	55
4.	**Methanol extract**	3	20	49	78
5.	*Annona squamosa*	3	29	65	88
6.	*Azadirachta indica* *Calotropis procera*	3	21	46	69

The combination of three plant extracts showed significant result in controlling ticks. The hot water extracts of dried leaf powder showed 100% mortality of ticks whereas ethanol and methanol extracts showed 83% and 80% of mortality respectively (**Table 3, Fig. 3**). These three plants posses alkaloids, glycoside and phenols which are important chemicals to initiate the mechanism of action in *in vivo* causing mortality of ticks. The herbal extracts of *Ricinus communis*, *Thevetia peruviana* and *Mentha piperita* and synthetic drug pyrethroids were tested against the ticks in the flock of goats. The result showed that herbal extracts was more effective in eliminating the adult ticks than synthetic drug tested [1].

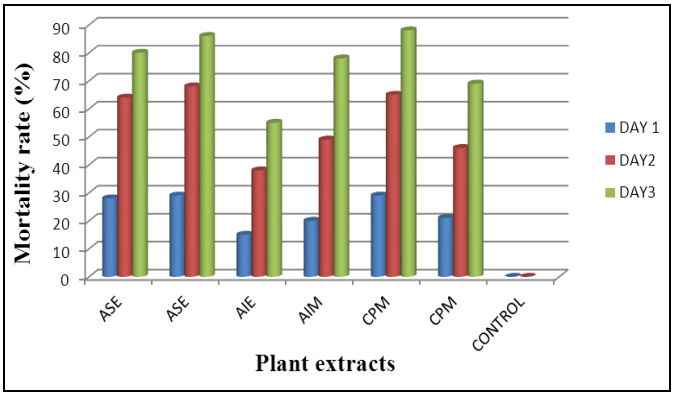

FIG.2: *IN VIVO* EFFECT OF ALCOHOLIC EXTRACT OF LEAF POWDER AGAINST CATTLE TICKS

TABLE 3: COMBINED *IN VIVO* EFFECT OF LEAF EXTRACTS OF *A SQUAMOSA, A. INDICA* AND *C. PROCERA* ON CATTLE TICKS

S.no	Extracts used	No of animals treated	Mortality rate in %		
			Day 1	Day 3	Day 5
1.	Powder +hot water extract	3	41	76	100
		3	38	73	98
2.	Powder+ cold water extract	3	32	54	74
		3	35	53	78
3.	Fresh leaves+ hot water extract	3	48	73	83
		3	44	70	80
4.	Fresh leaves+cold water extract				
5.	Powder+Ethanol extract				
6.	Powder+Methanol extract				

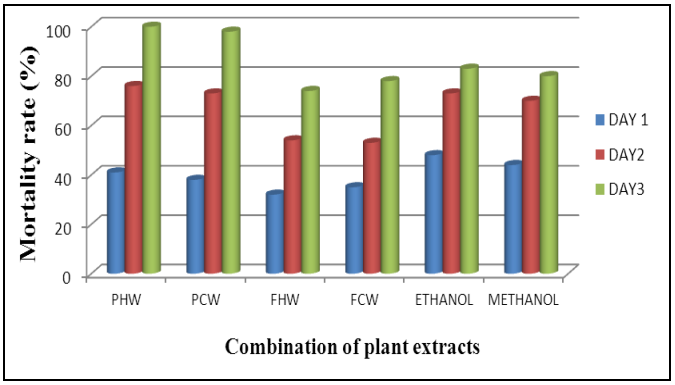

FIG.2: *COMBINED IN VIVO* EFFECT OF LEAF EXTRACTS OF *A. SQUAMOSA, A. INDICA* AND *C. PROCERA* ON CATTLE TICKS

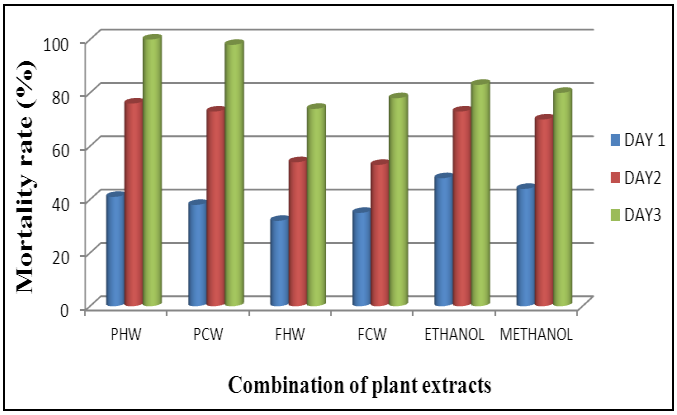

FIG.3: *COMBINED IN VIVO* EFFECT OF LEAF EXTRACTS OF *A. SQUAMOSA, A. INDICA* AND *C. PROCERA* ON CATTLE TICKS

PHW-Powder hot water extract, PCW-Powder cold water extract, FHW-Fresh leaves hot water extract,
FCW- Fresh leaves cold water extract,

CONCLUSION: The selected plants *Annona squamosa*, *Azadirachta indica* and *Calotropis procera* were tested against the cattle ticks under *in vivo* conditions. Both aqueous and alcoholic extracts of *A. indica* showed maximum mortality rate of ticks followed by *A. squamosa* and *C. procera* when tested individually but in combination of extracts of these three plants showed 100% mortality. Hence it is confirmed that the selected plant materials possess more acaricidal activity against cattle ticks. Since the herbal drugs are cost effective and easy to process, it can be used as an alternative medicine to control cattle ticks.

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