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Medicinal plants as alternative for control of parasites. 3. Arthropods

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ABSTRACT

The control of arthropod (ecto) parasites and disease vectors by use of industrial chemicals being fraught with various problems calls for alternative approaches. Natural products offer world's resource-poor farmers a cheaper, eco-friendly and sustainable alternative. Ethno-veterinary and traditional knowledge has provided a vast range of plants to be evaluated for their repellent, insecticidal and acaricidal properties. Phyto-chemical analysis of medicinal plants has been increasingly generating evidence of presence of bioactive compounds in extracts derived from them. With the current emphasis on reduction (management) of arthropod numbers to acceptable levels, plant products are likely to play a greater role. A large number of efficacy studies in recent years, both in India and abroad, have validated their bioactivity against various target insects and acarines. These studies covered a wide spectrum of arthropod parasites but mosquitoes and ixodid ticks due to their vector significance attracted maximum attention. The widely prevalent cattle tick *Rhipicephalus (Boophilus) microplus* appeared to predominate as the most frequent target species. Plant products have considerable potential, which can possibly be further enhanced by improved technology and fine-tuning of delivery systems such as use of nano-capsules and nano-particles. Plant derived acaricides can also be harnessed as a component of an integrated pest management program. In this article, the current state of knowledge on phyto-chemical sources along with their cidal and inhibitory activity on various stages of target insect and acarine species, the nature of active ingredients and promising advances, has been reviewed.

Key words: Acaricides, Arthropod parasites, Insecticides, Medicinal plants, Plant extracts

Arthropod parasites, apart from their direct ill-effects of discomfort, blood loss and dermatitis to their hosts, are highly important as vectors of disease pathogens affecting man and animals. The ecto-parasitic insects and acarines are widely prevalent throughout the world, particularly in tropical and subtropical countries, which have witnessed a resurgence of epidemic vector-borne infections in the recent past (Atkinson 2010). These can translate to disease, disability and death in humans and enormous production losses in livestock. To combat this menace, mankind had traditionally relied on various naturally occurring products. Prior to 1930, plant-derived insecticides (botanicals) constituted the foremost weapons against such pests. Pyrethrum, rotenone, derris, nicotine, quassia, camphor and turpentine were some such plant products. The era from mid-1930's to around 1960 was marked by the development and dramatic commercial success of synthetic insecticides. At present, there is increasing use of single shot endectocides having high potential for inducing resistance in non-target species. Emergence of resistance combined with concerns over residues in food and environmental issues, have led to the search for safe, economical and sustainable methods of control. As a consequence, there is a reversion towards traditional healthcare and ethno-veterinary knowledge in isolating and evaluating the medicinal plants. Scientists in Mexico found convincing evidence that birds incorporating cigarette ends in their nests, were doing so to ward off insect parasites (IANS 2012). Nictotine deposited in cigarette filter tips is a powerful insecticide which helped birds in keeping ectroparasites at bay. The leaves, stem, flowers, bark and root may be ground or the key components may be extracted by a solvent to prepare a liquid concentrate. The active ingredients of many plants and herbs possess repellent, insecticidal, growth-inhibiting and anti-moulting activities. The several factors, which affect the activity of extract are solvent, extraction time, extract concentration, target anthropod and exposure time. The criteria for anti-parasitic efficacy *in vitro* had ranged from repellent and cidal activity against adults and/or developmental stages to reduction in reproductive index through reduced egg mass and inhibition of hatchability. *In vivo* studies had relied on the reduced counts on skin surface/scrapings as the measure of efficacy. In the Indian context, despite a fairly large count of plant-evaluations for anti-parasitic activity, there is a paucity of dedicated reviews, apart from one on plant acaricides

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(Chhabra and Saxena 1998) and recently on mosquito larvicides (Ghosh *et al.* 2012).

Insecta: Neem (*Azadirachta indica*) because of its known medicinal and pesticidal properties (Schmutterer 1990) attracted attention. Field evaluation of repellent action of neem oil against sandfly (Sharma and Dhiman 1993) and anopheline/culicine mosquitoes (Rajnikant and Bhatt 1994, Sharma *et al*. 1995, Prakash *et al*. 2000) was reported. The neem oil was used at 1, 2 and 3 % concentration in mustard oil. The repellent action of lemon grass (*Cymbopogon citratus*) and citronella (C. *nardus*) against mosquitoes was also documented (Ansari and Razdan1995). There are several studies on screening of plant extracts/ compounds for larvicidal activity against mosquitoes: *Annona squamosa* and *Pongamia glabra* (George and Vincent 2005), *Solanum villosum* (Chowdhuri and Chandra 2007), *Capsicum annum* (Madhumathy *et al*. 2007), day jasmine (Ghosh *et al*. 2008) and *Jatropha curcas* (Kalimuthu *et al.* 2011). Larvicidal and smoke repellency effect of *Toddalia asiatica* and *Aegle marmelos* and similar bioefficacy of *Mentha piperita* essential oils (Kumar *et al.* 2011) against the dengue vector *Aedes aegypti* were also reported (Vineetha and Murugan 2009). Bagavan *et al*. (2009) investigated the adulticidal and larvicidal effects of several indigenous plant extracts against the fourth instar larvae of malaria vector *Anopheles subpictus* and Japanese encephalitis vector *Culex tritaeniorhynchus*. Highest mortality was found in leaf hexane extract of *A. squamosa,* methanol extracts of *Centella asiatica* and *Pergularia daemia* against *A. subpictus*. Methanol extract of *Phyllanthus emblica* (Aamala) against *C. tritaeniorhynchus*, was also promising. Adult emergence inhibition (EI) and adulticidal activities of several medicial plant extracts against *Anopheles stephensi* were studied (Abduz Zahir *et al.* 2010a). The highest EI activity was found in ethyl acetate extract of *Anisomeles malabarica* while the most effective adulticidal activity was observed in chloroform / acetone extract of *Gloria superba, Tridax procumbens,* and in ethyl acetate extract of *Ocimum basilicum.* Mosquitoes, due to their enormous vector significance for life-threatening diseases, had attracted a lot of attention for alternative control studies involving plant extracts. Ghosh *et al.* (2012) listed many such plants which possess target-specific activities, predominantly larvicidal against different species of vector mosquitoes. Against the related nematoceran *Simulium* spp. in India, Hazarika *et al.* (2012) observed good repellency potential of essential oils extracted from *Homalomena aromatica, Vitex negundo* and *Ageratum conizoides* with protection for >2h at 5% concentration and >5 h at 10% concentration in human volunteers. *In vitro* effect of 10% aqueous solution of *Annona squamosa* (custard apple) seed preparation on adult houseflies *Musca domestica* (Desai1997) was found toxic. Khater *et al*. (2011) reported that Egyptian plant oils of lettuce, chamomile possessed insecticidal effect against *Lucilia sericata.* Essential oils of *Mentha piperita* achieved housefly larval LC_{50} of 0.54 µl/cm^2 and *M. citrata* 48.4 µl/ cm^2 by contact toxicity (Kumar *et al.* 2012a). By fumigation and contact, 100% suppression was observed. Significant activity of *Citrus sinensis* essential oil against larvae and pupae of housefly, by contact toxicity and fumigation (Kumar *et al.* 2012b) suggested its use as an ecofriendly houseful control measure. Sinthusiri and Soonwera (2012) reported good susceptibility (knock down effect) of housefly *M. domestica* to essential oils of *Cymbopogon citratus* (Lemon grass), *Mentha piperita* (peppermint) and *Lavendula augustifolia* (lavender) at 10% concentration. Evaluation of lousicidal and repellency properties (Kumar *et al*. 2002) revealed that extracts from flowers of *Adhatoda vassica (Vasinja)* and leaves of *Azadirachta indica* caused 80% mortality to tropical hen louse *Lipeurus lawrensi tropicalis* while that of the leaves of *Argemone mexicana (Peeli kateli)* remained at 73%. Leaf extracts of *A. indica* imparted 77% repellence against the said louse while percentage of repellence of peel of *Citrus vulgaris* (orange), seeds and fruit pulp of *Cassia fistula* (Amaltas) was recorded as 60, 69 and 65% respectively. Later, the lousicidal potential of Himalayan shrub *Zanthoxylum alatum* (Timm) against the same louse species *L. lawrensi tropicalis* was tested (Kumar *et al*. 2003). The undiluted seed extract and its 1: 1 dilution caused 100% mortality within 12 h, while 1: 5 and 1: 10 dilutions could cause 93.3 and 83.3% mortality in 48h. Khan *et al*. (2008) tested crude mentha oil (*Mentha piperita*) diluted with methanol at different concentrations for mortality and repellence tests against tropical hen louse and found highly effective *in vitro* (100% mortality in 48h at 8% concentration). When tried *in vivo* on 10 highly infested birds, it was capable of controlling the lice after the fourth day. *In vivo* treatment of birds naturally infested with *Menopon gallinae* and *L. tropicalis* with neem oil spray at 5ml/ bird, pungan oil at 5ml / bird, comparative to cypermethrin (0. 2%) in a third group (Ponnudurai *et al*. 2011) revealed that lice intensity came down to 4 and 5, 2 and 3, and 3 and 2 on 7th, 14th and 21st day post-treatment (p.t.) in pungan and neem oil groups, respectively. In cypermethrin treated group, the lice count came down to zero on seventh day p.t. No reinfestation was observed up to 60 days in treated groups. Essential oils from camphor (*Cinnamomum camphora),* onion (*Alium cepa*), peppermint (*Mentha piperita*), camomile (*Matricaria chamomilla*) and rosemary (*Rosmarinus officinalis*) were evaluated in Egypt (Khater *et al.* 2009) for lousicidal and repellent effects. Both *in vitro* (contact bioassay) and *in vivo* treatments against buffalo louse *Haematopinus tuberculatus* and infesting flies such as *M. domestica, Stomoxys calcitrans, Haematobia irritans* and *Hippobosca equina* indicated the potential of these oils for further development. *In vitro* efficacy of extracts of dried leaf and flower of *Rhinacanthus nasutus* and *Cassia auriculata* in various solvents like acetone, chloroform, ethyl acetate, hexane and methanol (Kamaraj *et al*. 2010) were tested *in vitro* against the haematophagous fly *Hippobosca maculata* and nymph of goat louse *Damalinia caprae*. Formulations containing azadirachtin fraction of neem, viz. Neemrich I 80 EC and Neemrich II

20 EC (CSIR, Pune) at 0.5–1.0% concentrations were 79.0 to 85.7% effective in control of natural infestations of lice and fleas in sheep and goats (Muraleedharan *et al*. 2003). A similar neem-based commercial formulation 'Nimbitor' EC containing tetranotis terpinoid as active component, at 2 - 8% concentration (Maske *et al*. 2000) resulted in 100% killing of lice *Linognathus vituli* and fly *Hippobosca maculata* of cattle *in vitro* at 4% concentration in 10–20 min. Ponnudurai *et al*. (2007) recorded 100% reduction of lice in birds that were treated with herbal formulation containing ingredients of *A. indica, Cedrus deodara, Brassica campestris* and *Ocimum sanctum.* All the lice species died within 30 min of spraying the drug and p.t. lice count fell to zero. Additionally, fly repellent activity against house flies with 90–96% fly reduction lasted for 4h. In another study (Sonule *et al*. 2012), 4 herbal products viz. Karanj oil, eucalyptus oil, Neemazal (neem oil), each diluted in1% Teepol solution and emulsified, and a commercial 'Topicure' (*Pinus longifolia*, *Eucalyptus* and *C. deodara* oils) were tested against buffalo louse *Haematopinus tuberculatus* and goat louse *in vitro* by dip in concentrations of 4, 5 and 10%. A standard chemical Butox (1%) was also included. The trial was supplemented with *in vivo* trial in naturally infested buffaloes and goats. The results indicated efficacy levels of Karanj oil 83–100%, eucalyptus oil 85–95%, Neemazal 86–100% and Topicure 100%. An ayurvedic proprietary liniment 'Arlin', containing ingredients of proven antiparasitic properties, viz. eucalyptus oil, turpentine, deodar oil and camphor, was tried in goat kids (Muraleedharan and Sahadev 2012) carrying concurrent infestations with lice (*Linognathus*), fleas (*Ctenocephalides felis orientis*) and ticks. Arlin 0.4% aqueous emulsion was sprayed on the whole body as a single application. It resulted in 42.22%, 44.58% and 75.0% reduction of ticks, lice and flea infestations respectively on day 21p.t. Later when 0.5% Arlin was applied by swabbing on ticks attached to ears of adult sheep and goats, reductions of 65.15% and 75.54% respectively were achieved on day 21, indicating that efficacy can be augmented by either increasing the number of applications or concentration of the product. Recently, Umadevi and Umakanthan (2012) reported the complete eradication of the poultry flea *Echidnophaga* sp. from 9 backyard poultry farms by keeping leaves of *Jatropha* sp. (purging tree) at different places of the floor of poultry yard in the evening. The fleas were found clinging (stuck) to leaves on the subsequent morning, and most of them were dead.

Arachnida (acarina): The potential of neem seed extract for developing products as bio-acaricides was revalidated by Gupta *et al*. (2000) who evaluated its petroleum fractions against the cattle tick *Boophilus microplus in vitro*. Confirmatory studies on the *in vitro* efficacy of neem seed oil obtaining up to 100% mortality depending on concentration tried, against larvae of *Rhipicephalus sanguineus* (Choudhury 2001) came forth. Somewhat contrary to these observations, Kala Kumar *et al*. (2000) reported only 60–75% success with neem seed oil in *in vitro* trials against *B. microplus*, *Hyalomma anatolicum anatolicum* and *Rhipicephalus haemaphysaloides* on naturally infested buffaloes and cattle, whereas seed oil of custard apple (*Annona squamosa*) and pyrethrum (reference standard) were 100% efficacious. The active ingredient of custard apple seed oil 'annonaine' an alkaloid was considered as possible factor for its acaricidal efficacy. Further, they observed inhibition of oviposition in females of all 3 tick species after *in vitro* treatment with the custard apple seed oil. The repellent activity of some forage plants against ixodid ticks keep ticks away from pasture, thereby reducing their breeding and attachment to grazing animals. Utilizing this potential in the forage legume *Stylosanthes scabra* for biocontrol of ticks was reported (Khudrathulla and Jagannath 1998). Later, they (Khudrathulla and Jagannath 2000) reported 100% larvicidal activity in extracts of the fodder grass against *B. microplus* at 3% and above concentrations and against *R. sanguineus* at 6.0% and above concentrations. Aqueous extract gave better acaricidal effect than methanol extract. Efficacy of *karanj* oil alone (Shrivastava and Das 2003) and as 50: 50 mixture with neem oil (Kumar *et al*. 2005) as spray against *B. microplus* natural infestation in cattle was tried. In the latter study, daily spray with oily mixture for 7 days achieved 97.95% freedom from ticks on day 8 p.t. with ivermectin (200μg kg^{-1} b. wt.) as the reference standard. Methanolic extracts of neem leaves and bark, *nochi* (*Vitex negundo*) leaves, *vashambu* (*Acorus calamus*) rhizome and *pungu* (*Pongamia pinnata*) leaves were evaluated for *in vitro* acaricidal efficacy (Pathak *et al*. 2004). Neem bark extract was observed to be the most effective followed by vashambu. Tobacco leaf (*Nicotiana tabacum*) aqueous extract, undiluted and 50% dilution on engorged adult *R. haemaphysaloides* ticks (Choudhary *et al.* 2004) had an immediate effect on mortality and long term effect on fecundity parameters, viz. adversely affected egg-laying and hatching of larvae. Hydro-distillate of *Hyptis suaveolens* leaves, an essential oil with a strong aroma, was 100 and 98% effective *in vitro* and *in vivo*, respectively, on a single application (Kala Kumar *et al*. 2006). The adults and nymphal stages of *Hyalomma* sp., *Rhipicephalus* sp. and *Haemaphysalis* sp. were highly susceptible. Moreover, engorged females failed to oviposit on treatment. In another trial (Vatsya *et al*. 2006), hydro-distilled extracts of medicinal plants, *Artemesia annua*, *A. vulgaris* and *Ocimum kilimandscharicum* and oil seeds of *Pongamia glabra* were tested *in vitro* for efficacy against *B. microplus*. Of these, O. *kilimandscharicum* exhibited highest efficacy (98.34%) followed by *P. glabra* (96.67%), *A. annua* (95.0%) and *A. vulgaris* (93.34%). The weight of egg mass and per cent hatchability were also severely affected p.t. Muraleedharan *et al*. (2008) screened 8 aqueous herbal extracts for acaricidal effects, 2 in goats and 6 in sheep. The leaves of *Clerodendron inerma* and *Croton bonplandianum* in goats reduced infestation to 68.5 and 64.19%, respectively, on day 9. In sheep, the maximum reduction percentage recorded with the flowers of *Tagetus erecta* was 30.08 on

day 6, with cloves of *Allium sativum* 30.02 on day 7, whereas with the leaves of *Leucas aspera* it was 29.52 on day 8.0 p.t. Ethanol extracts of leaf, bark and seed of neem, of leaf and seed of peach (*Prunus persica*), bark of mango (*Mangifera indica*), and leaf of guava (*Tsidium guajava*) were evaluated *in vitro* against *B. microplus* (Srivastava *et al*. 2008). Of these, 8% extract from *A. indica* seed showed high level of efficacy (80%) after 5h of treatment. The direct effect of reference standard cypermethrin was 92.44%. Besides the immediate effect on adult ticks, the egg laying property of surviving ticks was significantly reduced. Also significant reduction in the reproductive index of ticks fed on animals treated with *A. indica* seed extract was noted. The authors postulated that the antifeedant and antiovipositional properties of neem seed extract make it a valuable component for developing strategy for integrated pest management (IPM). A subsequent study (Magadum *et al*. 2009) from the same centre was largely confirmatory, with the observation that *A. indica* was more efficacious than the extracts of *Annona squamosa*. Certain neem preparations were also found efficacious in substantial reduction of tick counts (*Haemaphysalis* and *Hyalomma* sp.) on sheep and goats on topical applications (Muraleedharan *et al*. 2009). Of these, undiluted neem oil gave the best results. Comparative evaluation of herbal essential oils, lemon grass (*Cymbopogon citratus*) and citronella (*C. nardus*) on goats naturally infested with 3 genera of ticks (John *et al*. 2009) indicated better performance of lemon grass. In a study to evaluate *in vitro* effect of several indigenous plant extracts (Bagavan *et al*. 2009), the highest mortality against the adult cattle tick *Haemaphysalis bispinosa* was found in leaf hexane extract of *Annona squamosa*, methanol extracts of *Gloria superba* and *Phyllanthus emblica* (LC_{50}=145.39, 225.57 and 256.68 ppm). Abduz Zahir *et al.* (2009) determined the efficacies of leaf, flower and seed extracts of *Achyranthes aspera, Anisomeles malabarica, Gloriosa superba, Psidium guajava*, *Ricinus communis* and *Solanum trilobatum* against the larvae of cattle tick *Rhipicephalus (Boophilus) microplus.* The results indicated that the leaf ethyl acetate extract of *A. aspera,* leaf acetone and chloroform extracts of *A.malabarica,* flower methanol extract of *G.superba* and leaf methanol extract of *R.communis* have the potential to be used as eco-friendly approach for the control of *R.microplus.* Further evidence of anti-parasitic activities of the plant extracts from Southern India was produced in another report (Abduz Zahir *et al.* 2010b). It suggested that the leaf methanol extract of *R. communis,* chloroform extract of *A.malabarica* and chloroform extract of *G. superba* have the potential to be effectively used for the control of *Haemaphysalis bispinosa* ticks and the fly *Hippobosca maculata.* Phytotherapeutic agents like neem seed oil, *karanj* (*Pongamia glabra*) seed oil, a mixture (1: 1) of neem and *karanj* oils, methanolic extract of neem leaves and *karanj* leaves individually and mixture of two were evaluated for *in vitro* effect on *Rhipicephalus* (*B.*) *microplus* by single and double dipping (Bisen *et al*. 2011). Karanj seed oil exhibited highest efficacy in single (70%) as well as double treatment (100%) in terms of per cent mortality of treated female ticks. The acaricidal action of Karanj leaf extract was also encouraging. The mixture provided a slightly lower efficacy. The mean weight of egg mass laid by the surviving female ticks in different treatment groups, showed the lowest values in *karanj* oil treatment group. Muraleedharan and Sahadev (2011) tested an ayurvedic liniment with constituent oils of deodar, turpentine and eucalyptus proved fairly effective in controlling natural infestation with *Hyalomma* and *Haemaphysalis* sp. ticks on sheep and goats. Crude ethanolic extracts of aerial parts (stem and leaves) of *Leucas aspera* produced concentration-dependent adult mortality and complete failure of inclusion of eggs from the treated *R. (B.) annulatus* (Ravindran *et al.* 2011). Amongst 34 extracts prepared from medicinal plants and tested against laboratory reared *R.(B.) microplus* (Ghosh *et al.* 2011), 8 extracts were effective. Of these, the extract prepared from rhizome of *Acorus calamus* highly efficacious and 100% mortality within 14 days p.t. was recorded. The efficacy of the extracts was confirmed *in vivo.* Juliet *et al.* (2012) reported that the ethanolic extract of leaves of *Jatropha curcas* caused significant blocking of hatching of the laid ova of *R. (B.) annulatus* indicating promise for the plant's utilization for control of the tick population. As plant extracts have low water solubility, various organic solvents are used in extraction of active components. To identify suitable solvents to be used in preparation of anti-tick herbal formulations, a comparative experimental assessment (Sharma *et al*. 2012) of 4 solvents (methanol, ethanol, acetone, n-butanol) and 2 surfactants (dimethyl sulphoxide, tween 20) on *R.* (*B.*) *microplus* was made. The study indicated that for developing anti-tick formulation, ethanol, methanol and acetone can be used at a specific concentration in the process of extraction while DMSO is a better option for making working concentration. Summary of eminent studies abroad on acaricidal activity of plants for control of ticks is appended in Table 1.

The bioactive plant oils/extracts are particularly effective in skin affections including those due to Acari or mange mites. There are several reports on miticidal/repellent plants which alone or in judicious combination with other plant/ active constituent, were successfully used for control of mange (Chhabra and Saxena 1998). Some of the recurrently used entities like oils of *Pongamia* spp. (Karanj), *Cedrus deodara* (Indian cedar or deodar), *Azadirachta indica* (neem) and *Eucalyptus globulus*, *Acorus calamus*, *Allium sativum* (garlic) *Juniperus communis* (Juniper) are utilized in commercial preparations against all forms of mange in livestock and companion animals (Pathak and Shukla 1997, Bhilegaonkar and Maske 1998). Herbal cream/ointment containing one or more of these ingredients gave clinical and parasitological cure against sarcoptic mange in camel and buffalo (Chhabra *et al*. 1994), in buffalo calves (Khurana *et al*. 2001) and in sheep (Sharma *et al*. 1997). Alternative therapy for control of sarcoptic mange in sheep

Table 1. Summary of some studies (abroad) on plants having acaricidal activity against ticks

Name of plant	Part used	Preparation	Test applied	Target species	Efficacy criteria	Reference(s)
Stemona collinsae	Root	Extract	*In vitro* and *in vivo*	*Boophilus microplus*	Mortaility of nymph and adult	Jansawan *et al.* (1993)
(i) *Aganonerion polymorphum*	Leaf and stem	Crude ethanolic extract	*In vitro* (Dippling)	*B. microplus*	Mortality	Chungsamarnyart *et al.* (1994)
(ii) *Calotropis gigantea*	Leaf and stem	Crude ethanolic extract	*In vitro* (Dippling)	*B. microplus*	Mortality	Chungsamarnyart *et al.* (1994)
(iii) *C. procera*	Leaf and stem	Crude ethanolic extract	*In vitro* (Dippling)	*B. microplus*	Mortality	Chungsamarnyart *et al.* (1994)
[Combination of (i) and (iii) and i) and (ii) exhibited enhanced (synergistic) activity]						
Margaritaria discoidea	Leaf and stem (bark more acaricidal)	Hexane extract	*In vitro* and *in vivo*	*Rhipicephalus appendiculatus*	Mortality (Nymphs)	Kaaya *et al.* (1995)
Ocimum suave (East African Tulsi)	Aerial parts	Oil extractedby steam distillation	*In vitro* and *in vivo*	*R. appendiculatus*	Mortality (Larvae)	Mwangi *et al.* (1995)
Pimenta dioica (tropical tree)	Leaf	Hexane extract / Essential oil	*In vitro*	*B. microplus*	Mortality and inhibition of oviposition	Brown *et al.* (1998)
Azadirachta indica	Seed	Oil extract	*In vitro*	*Amblyomma variegatum*	Mortality (Larvae)	Ndumu *et al.* (1999)
Capsicum spp. *Euphorbia obovalifolia* *Ficus brachypoda*	Aerial parts of all 3 combined	Crude extract (ethno-vet. in Ethiopia)	*In vitro* and *in vivo*	*Rhipicephalus decolaratus*	Inhibitory effect on all stages	Regasa (2000)
Azadirachta indica	Seed and stem	Ether extract	*In vitro*	*Boophilus microplus*	100% control of reproduction	Benavides *et al.* (2001)
Tamarindus indicus	Mature fruit	Aqueous (and 10% ethanol extracts)	*In vitro*	*B. microplus* (Engorged females)	56-99% mortality (time related 24h-7 days)	Chungsamarnyart and Jansawan (2001)
Azadirachta indica (Ethiopian neem)	Oil	Extract	*In vitro*	*Rhipicephalus pulchellus*	Larvicidal	Ismail *et al.* (2002)

(Contd...)

(Table 1 Continues)

Name of plant	Part used	Preparation	Test applied	Target species	Efficacy criteria	Reference(s)
Dahlstedtia pentaphylla	Root	Ethanol extract (terpenoids)	*In vivo* (Spray on 30 bovines)	*B. microplus*	Adulticide	Pereira and Famadas (2006)
Copaifera reticulate	Leaf and stem	Extract (terpenoids)	*In vitro*	*Rhipicephalus (B.) microplus*	Larvidial	Fernandes and Freitas (2007)
Tephrosia vogelii	Leaf	Methanol, aqueous and other extracts	*In vitro*	*Various gener of ixodid ticks*	Cidal (nymph and adult)	Matovu and Olila (2007)
Hypericum polyanthemum	Aerial parts	Crude methanolic extracts	*In vitro*	*R.(B.) microplus*	Larvicidal (100% at higher cone)	Ribeiro *et al.* (2007)
Magonia pubescens (Soap berry)	Stem, bark	Crude ethanolic extract	*In vitro*	*Rhipicephalus sanguineus*	Larvicidal	Fernandes *et al.* (2008)
Calea serrata	Leaf and stem	Hexane extract (precocene)	*In vitro*	*B.microplus*	Inhibition of egg hatching	Ribeiro *et al.* (2008)
Aloe ferox and *Ptaeroxylon obliquum*	Fresh leaves (crushed)	Soaked overnight in water	*In vivo* (spray on cattle). Ethnoveterinary in South Africa	*B. microplus*	High degree of control as per respondent farmers	Moyo and Masika (2009)
Pelargonium roseum and *Ecucalyptus globulis*		Essential oil	*In vitro*	*R.(B.) annulatus*	Adulticidal	Pirali-Kheirabadi *et al.* (2009)
Lippia graveolens and *Allium sativum*		Essential oil	*In vitro*	*R.(B.) annulatus*	Acaricidal	Martinez-Velasquez *et al.* (2010)
Piper spp.	Aerial parts	Essential oil	*In vitro*	*R.(B.) microplus*	Acaricidal	Ferraz *et al.* (2010)
Lavendula augustifolia	Leaf	Essential oil	*In vitro*	*R.(B.) microplus*	Acaricidal	Pirali-Kheirabadi and da Silva (2010)
Hesperozygis ringens	Leaf	Essential oil (pulegone constituent of the oil)	*In vitro*	*R.(B.) microplus*	Inhibition of egg hatching of engorged females	Ribeiro *et al.* (2010)
Petiveria alliacea	Leaf and stem	Crude extract	*In vitro*	*R.(B.) microplus*	Acaricidal	Rosado-Aguilar *et al.* (2010)
Artemisia annua	Leaf	Crude extract (artemisinin)	*In vitro*	*R.(B.) microplus*	Action on engorged females	de Souza Chagas *et al.* (2011)

(Concluded Table 1)

Name of plant	Part used	Preparation	Test applied	Target species	Efficacy criteria	Reference(s)
Tetradenia riparia	Leaf	Essential oil	*In vitro* and *in vivo*	*R.(B.) microplus*	Adulticide	Gazim *et al.* (2011)
Lippia javanica	Leaf	Aqueous extract (Phenolic glycosides flavanoids)	*In vitro*	*R.(B.) microplus*	Adulticide	Madzimure *et al.* (2011)
Calea serrata	Leaf	Essential oil and Precocene II isolated from it	*In vitro*	*R.(B.) microplus*	Toxic to the larvae	Ribeiro *et al.* (2011)
Cymbopogon martini	Leaf	Essential oil (geraniol)	*In vitro*	*R.(B.) microplus*	Activity against engorged females	de Souza Chagas *et al.* (2012)
Azadirachta indica	Leaf					
Nicotiana tabacum	Leaf	Aqueous extracts in combination ethno-vet./Traditional system of Pakistan	*In vitro*	*R.(B.) microplus*	All developmental stages (dose and time dependent response)	Zaman *et al.* (2012)
Calatropis procera	Flower					
Trachyspernum ammi	Seed					
Tagetes minuta	Aerial parts	Essential oil (dihydrotagetone, limonene, tagetone)	*In vivo* (cattle pen trial)	*R.microplus*	Reduction in no. of ticks, average wt. of ticks, egg wt. per engorged female, larval viability	Andreotti *et al.* (2013)
Strychnos spinosa and *Solanum incanum*	Fruit	Extracts	*In vitro* and *in vivo* (Cattle pen trial keeping amitraz as reference control)	*R.(B.) decoloratus*	Effect on all biological parameters (all stages)Efficacy of *S.incanum* fruit extract higher	Madzimure *et al.* (2013)
Lippia triplinervis	Aerial parts	Essential oil (monoterpenes thymol, carvacrol and p. cymene)	*In vitro*	*R. microplus*	Cidal for larvae and engorged females	de Assis *et al.* (2013)

with oil of *C deodara*, *Pongamia glabra*, *Jatropha curcas* in combination, as topical application was found satisfactory, safe and cheap (Dimri and Sharma 2004). In goats, Mukherjee and Dasgupta (2001) and Shekhar *et al*. (2007) tried sulphur and Karanj oil mixture. The latter report used a paste of sulphur (10g) and Karanj oil (100ml) applied on alternate days in the first week and thereafter at weekly intervals. Further, it was regarded as low cost and effective alternative to ivermectin injection or amitraz external application. Lundh *et al*. (2005) reported use of azadirachtin-impregnated traps for the control of the poultry mite *Dermanyssus gallinae*. Efficacy of leaf extracts of 2 plants (code named AAU-NW-EVM-1 and AAU-NW-EVM-2 against *Sarcoptes scabiei* var *caprae* in clinically affected goats, applied as 5% ointment (w/w) was evaluated in comparison to ivermectin as standard (Barua *et al*. 2008). Based on mite counts / sq. cm. AAU-NW-EVM-2 gave promising results. For sarcoptic mange in pigs, a herbal mineral preparation (50 ml neem oil, 50ml *karanj* oil and 10g camphor) applied locally was found 100% effective (Sinha *et al*. 2004, Kumar *et al*. 2005, Bariyar and Prasad 2007) although recovery was slower than ivermectin. In dogs, a herbal topical miticidal ointment "Dermanol" along with herbal Anbioflam capsules was efficacious (Pal *et al*. 2002). Seed powder of *A. squamosa* (custard apple, active constituent Iso - squamosin) mixed with equal proportion of petroleum jelly was successfully used topically on 12 dogs naturally infested with *S. scabiei* var *canis* (Bagherwal *et al*. 2008). Topical application of *Argemone mexicana* paste 4–5 times weekly on dogs suffering from demodicosis, gave 100% clinical and parasitological cure (Thakur *et al*. 1995). *Withania somnifera (Ashwagandha)* revealed adjuvant activity with the miticidal *Jatropha curcas* oil, enhanced antioxidant defense parameters and hastened clinical recovery in canine demodicosis (Singh and Dimri 2010). Demodicosis produces immunosuppression and oxidation stress in canines and for better recovery, the combination of amitraz dipping along with oral *Asparagus racemosus* root extract elicited better recovery (Sarkar *et al*. 2012) due to immunomodulatory and antioxidant potential of the herb. In camels, sarcoptic mange is a serious chronic and debilitating disease. As such, various indigenous and plant-based formulations were tried as alternatives to toxic chemicals. A formulation prepared from the juices of onion, garlic and lemon mixed manually with camphor and turmeric, and applied on the affected parts, after bathing the animals with decoction of babool (*Melia azedarach*) bark was found effective (Dixit *et a*l. 2002) in relieving the symptoms as well as making the animals negative for mites. Further, the miticidal properties were proven (Dixit *et al*. 2004) in all clinical and haematobiochemical parameters. Later, an improved version of the formulation was developed (Dixit *et al*. 2005) and therapeutic efficacy was revalidated (Rathod *et al*. 2006, Dixit *et al*. 2009). Kapoor and Sharma (2012) found essential oil of *Cedrus deodar*a effective against *Psoroptes cuniculi* in rabbits.

Despite numerous plant extracts being reported as promising candidates for acaricidal and insecticidal activity both in India and abroad, the feasibility of their usage on infested animals in the field conditions, is yet to be explored adequately. While the desirability of continuing such studies with enlarged efficacy-parameters is obvious, possible adverse effects on live host animal should also be critically monitored. Apart from being a safe, sustainable alternative control option, plant products may also provide synergistic enhancement in judicious combinations or even help prolong the efficacy lifespan of commercial agents in rotational usage. The potential uses and benefits of nanotechnology enhancement involving nanocapsules for vector management is another innovative approach in the domain of biorational insecticides. Velayuthan *et al.* (2012) initiated this trend by synthesizing titanium dioxide (t.d.) nanoparticles mediated with *Catharanthus roseus* leaf extract against the adults of haematophagous fly *Hippobosca maculata* and sheep biting louse *Bovicola ovis.* Synthesized t.d. nanoparticles using *Calotropis gigantea* showed acaricidal activity against *R. microplus* and *Haemaphysalis bispinosa* (Marimuthu *et al*. 2013). Bioactive natural products had improved properties with nickel nanoparticles against the larvae of cattle ticks and mosquito larvae (Rajakumar *et al.* 2013). However, this technology is still in research and development stage.

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