



## Review Article

# An Overview of Plants with Acaricidal and Anthelmintic Properties

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## Abstract

Geo-climatic and socio-economic conditions provide a favourable environment for parasitic population of livestock in Pakistan. Hard ticks (Ixodidae) and gastrointestinal nematodes pose most serious threats to livestock industry. Stakeholders rely on synthetic drugs to control these parasites. Emergence of drug resistance in these parasites; however, has provoked interest in alternate of synthetic drugs. Testing of plants used in ethnoveterinary medicine for their antiparasitic activity employing standard procedures has been reported to be promising. Plants have been most frequently used for deworming purposes followed by as acaricides and insecticides; whereas, reports as to their antiprotozoal use are relatively less. Use of plants as antiparasitics is more frequent in developing countries having low accessibility to the modern parasite control practices. Likewise, validation studies on the use of plants as antiparasitics have been more frequently carried out in the developing countries compared with those having livestock farming as commercial enterprise. This article presents an overview of the plants having acaricidal and anthelmintic activity, limitations as to the application of phytotherapy to control parasites and future prospects of the use of plants as antiparasitics. © 2017 Friends Science Publishers

**Keywords:** Anthelmintic; Acaricides; Antiparasitic; Ticks; Gastrointestinal nematodes; Plants

## Introduction

Livestock play a crucial role in human food supplies and economy of Pakistan (Anonymous, 2014–2015). Parasitism is one of the most important constraints in optimum productivity of animals (Solcan *et al.*, 2015). Production losses due to parasitism may vary from one parasite to the other. In tick infestation, for example, losses may be due to loss of blood (Gabr, 2012), tick paralysis (Chand *et al.*, 2016), transmission of different diseases (Duvall and Boireau, 2015; Kmiecik *et al.*, 2016), treatment costs (Gomes *et al.*, 2016), etc. Likewise, gastrointestinal nematodes (GINs) may interfere with food intake and absorption adversely affecting animal productivity (Geurden *et al.*, 2015; Ravinet *et al.*, 2016).

Globally, tick infections are recognized as most devastating ectoparasites (Godfrey and Randolph, 2011; Opara *et al.*, 2016; Demessie and Derso, 2015) resulting in huge economic losses (Zheng *et al.*, 2012; Asmaa *et al.*, 2014; Chen *et al.*, 2014). Likewise, tick associated concerns as to their increasing prevalence, lowered animal productivity; their zoonotic potential and control have been considered as an important area for future research (Ashfaq *et al.*, 2015; Jabbar *et al.*, 2015; Zahid *et al.*, 2016). Helminth infections are also ranked high among the factors leading to low production in animals throughout the world with varying prevalence depending on the climatic conditions, awareness

of farmers on the recommended parasite control practices and accessibility to the animal health care (Khan *et al.*, 2010b; Tasawar *et al.*, 2010; Qamar *et al.*, 2011; Borji *et al.*, 2012; Katoch *et al.*, 2012; Roeber *et al.*, 2013; Singh *et al.*, 2014; Bansal *et al.*, 2015; Dalal *et al.*, 2015; Lashari *et al.*, 2015; Scala *et al.*, 2015; Voigt *et al.*, 2016).

## Control of Parasitism

In general, control of ticks and GINs predominantly depends upon chemotherapy, even after the advancements in genetically, immunological and biotechnological methods (Sorge *et al.*, 2015; McTier *et al.*, 2016; Verma and Singh, 2016). Globally; however, use of synthetic drugs for animal health and production is facing challenges due to a variety of factors, e.g., because of their high costs (Mondal *et al.*, 2013), general toxicity (Patel *et al.*, 2013), drug residual problems in milk and meat (Elmanama and Albayoumi, 2016; Tochi *et al.*, 2016) and development of drug resistance in ticks (Abbas *et al.*, 2014; Coles and Dryden, 2014; Heath and Levot, 2015; Kumar and Partap, 2015; Muyobela *et al.*, 2015; Vudriko *et al.*, 2016) and GIN (Playford *et al.*, 2014; Alonso-Diaz *et al.*, 2008; Borges *et al.*, 2015; Geurden *et al.*, 2015; Muniz-Lagunes *et al.*, 2015; Ramos *et al.*, 2016). In addition, quality of antiparasitic drugs, particularly in developing countries, has led to attention of the stakeholders to find alternatives, may be as a part of drug resistance

management programs (Zaman et al., 2012a,b; Sindhu et al., 2014; Ghosh et al., 2015a; Kumar et al., 2016). Prospects of using plants as alternates to synthetic antiparasitic drugs have been discussed in the following paragraphs.

### Plants as Anti-parasitics

Plants and/or their products have been used for treatment of different diseases for centuries. There is an extended relationship among the coexistence of herbal remedies, parasites and humans (Matsabisa et al., 2013). It is as old as history of man itself. The plant kingdom is a vast storehouse of chemical substances manufactured and used by plants as defenses against insects, bacteria, fungi and viruses (Rattan, 2010; Mithöfer and Boland, 2012; Aslam et al., 2016). Plants are known to produce a range of secondary metabolites such as terpenoids, alkaloids, polyacetylenes, flavonoids and unusual amino acids and sugars (Chen et al., 2011; Savithramma et al., 2011; Hussain et al., 2012), for their defense from attack by pests. Plants constitute major part of the traditional veterinary practices termed as "ethnoveterinary medicine (EVM)" (Upadhyay et al., 2011; Asadbeigi et al., 2014). These plants may also possess' biological activity against significant parasites of veterinary standpoint, which could effectively be used to control ecto- and endo-parasites post-scientific validation. The efficacy of plant extracts/products against endo- and ecto-parasites of animals have been reported with variable success (e.g., Chen et al., 2011; Maphosa and Masika, 2012; Martinez-Ortiz-de-Montellano et al., 2013; Mbaya and Ogwiji, 2014; Silva et al., 2014; Abbas et al., 2014, 2015, 2017; Kumarasinghe et al., 2016). Most frequently used plants are sown either by the farmers or found self-grown, in the fields. These can also be obtained from the herbal/grocery stores easily. Farmers can use the wild herbs by uprooting from fields. In Pakistan, farmers believe that control of a parasite of a particular region is provided by nature in the form the indigenous plants of the area (Personal Communication). In tropical countries, common-and-economic availability of plants render them to be most viable options as alternates of synthetic antiparasitics drugs (Chander et al., 2013; Neergheen-Bhujun, 2013; Tamboli et al., 2015). Literature on the use of plants as acaricides and anthelmintics has been selectively reviewed as under:

### Plants Used as Acaricides

For the last ten years (2005–2015), 58% increase in citation of plants against ticks per year has been noticed (Bhardwaj et al., 2012; Benelli et al., 2016). However, in Pakistan, only a handful number of plants have been used against *R. microplus* (Zaman et al., 2012a; Sindhu et al., 2012; Nawaz et al., 2015) in contrast to more extensive investigations elsewhere (Chen et al., 2011; dos Santos et al., 2013; Nyangare et al., 2015; Fouche et al., 2016a,b).

Some plants reported for their anti-tick activity (acaricides) have been selectively reviewed and listed in Table 1.

### Tests Used for Evaluation of Anti-tick Activity

**In vitro:** Plants have been mostly evaluated through *in vitro* bioassays for preliminary screening. Three tests have been often used, i.e., Larval immersion test, larval packet test adult immersion test and syringe test.

**Larval immersion test (LIT):** This test takes around six weeks for results and is not recommended by FAO (FAO, 2004). Briefly, fully blood engorged female ticks are immersed in various concentrations of the candidate drugs for 2–4 min. All the ticks weighed together pre-immersion (WPI). The efficacy of the drug is measured on the basis of mortality (up to 14 days post-immersion), weight of the eggs laid by the ticks, reproductive index (RI) calculated by egg weight divided by WPI and oviposition inhibition (OI) ( $RI_{control}-RI_{treated}/RI_{Control} \times 100$ ). A major advantage of LIT is that it does not need any specific solvent rendering it more suitable for plants extracts.

**Larval packet test (LPT):** This is a time-efficient test and fully supported by FAO and have been adapted by many workers (Chagas et al., 2016; Vudriko et al., 2016). Briefly, larvae were inserted into drug impregnated filter paper for 24 hrs at certain temperature (27–29°C) and relative humidity (80–85%). Mortality and/or inhibition of larval motility are the criteria to measure the efficacy of the drugs. Trichloroethylene is used as solvent in LPT. Plant extracts are insoluble in this solvent thus only a limited number of scientists used this test for evaluation of efficacy. However, some workers modified LPT and have used acetone, ethanol and methanol in place of trichloroethylene (de Monteiro et al., 2012; Singh et al., 2015). Only condition of the solvent is that it should not cause mortality of tick's larvae more than 5% in control group.

**Adult immersion test:** This test takes around four weeks and is recommended by FAO (FAO, 2004). Test needs to be conducted on only healthy ticks, and weight of the group of ticks and egg mass should be proportionate etc.); thus, only a few workers used this test (Parveen et al., 2014; Ghosh et al., 2015b). In this test, ticks are exposed to test product/drug for 30 sec and efficiency is measured by effect on egg laying capacity of the female ticks.

**Syringe test:** This is the most recently introduced test and basically is a modification of LIT. Main difference is use of 14 days old larvae, which are to be exposed to candidate drugs for 30 sec. Special syringes, with cutting nozzle end and withdrawn plunger (2 mL), are prepared. After placing eggs in the cutting end of the syringe, it closes tightly with organza fabric until eggs hatch out in around 14 days. The larvae are immersed for 30 sec and the syringe is placed in fume hood for drying (1 h). The main criterion of efficacy evaluation is larval mortality and inhibition of their motility/walk (Sindhu et al., 2012).

**Table 1:** Plants used as acaricides

Plant	Part used	Plant family	Reference (s)
<i>Acanthus ebracteatus</i>	Leaf	Acanthaceae	Chungsamarnyart <i>et al.</i> , 1988
<i>Acorus calamus</i>	Rhizome	Acoraceae	Pathak <i>et al.</i> , 2004
<i>Aegle marmelos</i>	Leaf	Rutaceae	Kamaraj <i>et al.</i> , 2011
<i>Ageratum houstonianum</i>	Leaf	Asteraceae	Pamo <i>et al.</i> , 2005; Parveen <i>et al.</i> , 2014
<i>Ageratum conyzoides</i>	Whole plant	Asteraceae	Kumar <i>et al.</i> , 2016
<i>Allium sativum</i>	Bulb	Amaryllidaceae	Shyma <i>et al.</i> , 2014
<i>Annona squamosal</i>	Seed	Annonaceae	Ilham <i>et al.</i> , 2014
<i>Artemisia absinthium</i>	Oil	Asteraceae	Thakur <i>et al.</i> , 2007; Godara <i>et al.</i> , 2014
<i>Azadirachta indica</i>	Oil	Meliaceae	Thakur <i>et al.</i> , 2007
	Bark		Pathak <i>et al.</i> , 2004; Maharaj <i>et al.</i> , 2005
	Leaf		Handle <i>et al.</i> , 2002; Pathak <i>et al.</i> , 2004, Nawaz <i>et al.</i> , 2015
	Seed		Chagas <i>et al.</i> , 2016
<i>Citrus spp.</i>	Peel oil	Rutaceae	Ghosh <i>et al.</i> , 2015b
<i>Cymbopogon winterianus</i>	Essential oil	Poaceae	de Mello <i>et al.</i> , 2014
<i>Dahlstedia pentaphylla</i>	Root	Fabaceae	Pereira and Farnadas, 2006
<i>Datura stramonium</i>	Leaves	Solanaceae	Ghosh <i>et al.</i> , 2015a
<i>Drimys brasiliensis</i>	Essential oil of stem/leaf	Winteraceae	Ribeiro <i>et al.</i> , 2007, Ribeiro <i>et al.</i> , 2008
<i>Gynandropsis gynandra</i>	Essential oil	Cleomaceae	Malonza, 1992; Lwande <i>et al.</i> , 1999
<i>Hypericum polyanthemum</i>	Aerial part	Hypericaceae	Ribeiro <i>et al.</i> , 2007
<i>Lavandula officinalis</i>	Essential oil	Lamiaceae	Abdel-Shafy and Soliman, 2004
<i>Lippia gracilis</i>	Essential oil	Verbenaceae	Cruz <i>et al.</i> , 2013
<i>Luffa acutangula</i>	Not Reported	Cucurbitaceae	Chungsamarnyart <i>et al.</i> , 1988
<i>Margaritaria discoidea</i>	Not Reported	Phyllanthaceae	Kaaya <i>et al.</i> , 1995
<i>Marjorana hortensis</i>	Not Reported	Lamiaceae	Abdel-Shafy and Soliman, 2004
<i>Matricaria chamomile</i>	Flower	Asteraceae	Pirali-Kheirabadi and Razzaghi-Abyaneh, 2007
<i>Melia azedarach</i>	Leaf	Meliaceae	Matias <i>et al.</i> , 2003
	Fruit		Sousa <i>et al.</i> , 2011
<i>Melinis minutiflora</i>	Whole plant	Poaceae	Muro <i>et al.</i> 2004, Fernandez-Ruvalcaba <i>et al.</i> 2004
<i>Mentha piperita</i>	Whole plant	Lamiaceae	Abdel-Shafy and Soliman, 2004; Chagas <i>et al.</i> 2016
<i>Neoglaziovia variegata</i>	Leaves and aerial part	Bromeliaceae	Dantas <i>et al.</i> , 2015
<i>Nicotiana tabacum</i>	Leaf	Solanaceae	Choudhary <i>et al.</i> , 2004; Maroyi, 2012, Zaman <i>et al.</i> , 2012a, Farooq <i>et al.</i> , 2008
<i>Ocimum basilicum</i>	Leaves	Lamiaceae	Abdel-Shafy and Soliman, 2004; Martinez-Velazquez <i>et al.</i> , 2011, Veeramani <i>et al.</i> , 2014
<i>Ocimum suave</i>	Leaf	Lamiaceae	Mwangi <i>et al.</i> , 1995, Magona <i>et al.</i> , 2011
<i>Pimentadioica dioica</i>	Bark and leaf; Seed	Myrtaceae	Brown <i>et al.</i> , 1998; Martinez-Velazquez <i>et al.</i> , 2011
<i>Pongamia pinnata</i>	Essential Oil, Seed	Fabaceae	Thakur <i>et al.</i> , 2007; Handle <i>et al.</i> , 2002
<i>Cleome hirta</i>	Essential oil	Capparaceae	Ndungu <i>et al.</i> , 1999
<i>Sapindus saponaria</i>	Stem	Sapindaceae	Fernandes <i>et al.</i> 2005
<i>Semecarpus anacardium</i>	Leaves	Anacardiaceae	Ghosh <i>et al.</i> 2015a
<i>Stemonia collinsiae</i>	Rhizomes; Root	Stemonaceae	Chungsamarnyart <i>et al.</i> , 1988; Kongkiatpaiboon <i>et al.</i> , 2014
<i>Stylosanthes hamata</i>	Aerial parts	Fabaceae	Fernandez-Ruvalcaba <i>et al.</i> , 1999, Muro <i>et al.</i> , 2003
<i>Stylosanthes humilis</i>	Aerial parts	Fabaceae	Fernandez-Ruvalcaba <i>et al.</i> , 1999, Muro <i>et al.</i> , 2003
<i>Syzygium aromaticum</i>	Essential oil	Myrtaceae	de Mello <i>et al.</i> , 2014
<i>Tamarindus indicus</i>	Seeds; Fruits	Fabaceae	Guneidy <i>et al.</i> , 2014
<i>Vitex agnus-castus</i>	Seed	Lamiaceae	Mehlhorn <i>et al.</i> , 2005

Ticks used in experimental and/or natural infestations in above cited studies in decreasing order of frequency were *Rhipicephalus microplus*, *R. annulatus*, *R. appendiculatus*, *R. sanguineus*, *R. haemaphysaloides* and *R. pulchellus*.

**In vivo ear bag method:** A muslin cloth bag is fabricated (13 × 17 cm) to facilitate attachment of seed ticks (freshly hatched larvae) on the animals. After successful attachment of the seed ticks, the candidate drugs are applied topically. The evaluation criterion is number of ticks dropping off the animal (Ghosh *et al.*, 2011, 2013, 2015a,b; Zaman *et al.*, 2012a).

### Plants Used as Anthelmintics

Worldwide, a number of medicinal plants have been used to treat gastro-intestinal helminthiasis (Orr, 2015; Habibi *et al.*, 2016; Nosal *et al.*, 2016; Liaqat *et al.*, 2016). An account of the plants used as anthelmintics is given in Table 2.

### Tests Used for Evaluation of Anthelmintic Activity

The tests and test worms used by different workers to

evaluate the efficacy of plants have been presented in Table 2.

**Egg hatch test:** Fresh eggs (unhatched eggs) are incubated for 72 h with various concentrations of candidate drugs. Inhibition of hatching is main criterion for efficacy of the candidate drugs. Inhibition of hatching is main criterion for efficacy of the candidate drugs (Coles *et al.*, 2006). Eggs for this test are either isolated from faeces of donor animals or by triturating the female *H. contortus* in Phosphate Buffer Saline after collection from abomasum of slaughtered animals. Eggs obtained from female *H. contortus* could be of different stages of embryonation so copro-purified eggs generate more reliable results (Taylor *et al.*, 2002; Várady *et al.*, 2007). Tap water has been used for preparation of serial dilutions of the candidate drugs by many researchers, which have been found inappropriate because ions naturally present in the water effect on egg hatching.

**Table 2:** Plants used as anthelmintics

Plant	Part used	Helminth (s)	Family	Reference (s)
<i>Acacia albida</i>	Seed	Mixed infection of GIN	Fabaceae	Nwude & Ibrahim, 1980
<i>Acacia gaumeri</i>	Leaves	<i>Haemonchus contortus</i>	Fabaceae	Alonso-Diaz et al., 2011
<i>Acacia pennatula</i>	Leaves	<i>Haemonchus contortus</i>	Fabaceae	Alonso-Diaz et al., 2008
<i>Acacia nilotica</i>	Fruit	Mixed infection of GIN	Fabaceae	Bachaya et al., 2009
<i>Adhatoda vasica</i>	Root and leaf	Mixed infection of GIN	Acanthaceae	Al-Shaibani et al., 2009b; Somnath et al., 2015
<i>Agrimonia eupatoria</i>	Not Reported	Mixed infection of GIN	Rosaceae	Farnsworth et al., 1985
<i>Albizia anthelmintica</i>	Bark	Mixed infection of GIN	Mimosaceae	Minja, 1989
	Root	<i>Haemonchus contortus</i>		Githiori et al. 2003; Gathuma et al. 2004; Grade et al. 2008
<i>Allium sativum</i>	Bulb	<i>Haemonchus contortus</i> ; Mixed infection of GIN	Amaryllidaceae	Iqbal et al., 2001; Ahmed et al., 2014
<i>Aloe ferox</i>	Leaves	<i>Haemonchus contortus</i>	Asphodelaceae	Maphosa et al., 2010
<i>Amomum aromaticum</i>	Seeds	<i>Haemonchus contortus</i>	Zingiberaceae	Kaushik et al., 1981
<i>Anacardium occidentale</i>	Essential oil	<i>Haemonchus contortus</i>	Anacardiaceae	Ademola & Eloff, 2011
<i>Ananas comosus</i>	Not reported	<i>Haemonchus contortus</i>	Bromeliaceae	Ahmed et al., 2014
<i>Areca catechu</i>	Nut	<i>Haemonchus contortus</i>	Arecaceae	Barbieri et al., 2014
<i>Artemisia brevifolia</i>	Whole plant	<i>Haemonchus contortus</i>	Compositae	Iqbal et al., 2004; Irum et al., 2015
<i>Artemisia herbaalba</i>	Shoot	<i>Haemonchus contortus</i>	Asteraceae	Idris et al., 1982; Seddiq et al., 2011
<i>Azadirachta indica</i>	Leaf	Mixed infection of GIN; <i>Haemonchus contortus</i>	Meliaceae	Radhakrishnan et al., 2007; Jamra et al., 2015
	Seed	<i>Haemonchus contortus</i>		Hördegen et al., 2006; Costa et al., 2008
		Mixed infection of GIN		Iqbal et al., 2010
	Cake and Leaf	Mixed infection of GIN		Gowda, 1997; Mostofa et al., 1996
<i>Boswellia dalzielii</i>	Bark	Mixed infection of GIN	Burseraceae	Nwude and Ibrahim, 1980
<i>Butea Spp.</i>	Various parts	<i>Haemonchus contortus</i> ; Mixed infection of GIN	Fabaceae	Singh et al., 2015; Lateef et al., 2006b; Iqbal et al., 2006
<i>Caesalpinia crista</i>	Seed	Mixed infection of GIN; <i>Haemonchus contortus</i>	Fabaceae	Jabbar et al., 2007; Bhardwaj et al., 2015
	Fruit	Mixed infection of GIN		
<i>Calliandra calothyrsus</i>	Legume	<i>Haemonchus contortus</i>	Fabaceae	Cresswell, 2007; Florence & Mbida, 2011,
<i>Calotropis procera</i>	Flower	Mixed infection of GIN	Apocynaceae	Iqbal et al., 2005
	Latex	<i>Haemonchus contortus</i>		Murti et al., 2015; Cavalcante et al., 2016
<i>Carum copiticum</i>	Seed	Mixed infection of GIN	Apiaceae	Lateef et al., 2006a; Boskabady et al., 2014
<i>Carissa edulis</i>	Root	Mixed infection of GIN	Apocynaceae	Mishra et al., 2012
<i>Cassia spectabilis</i>	Root	<i>Haemonchus contortus</i>	Fabaceae	Moraes-Costa et al., 2015
<i>Chenopodium album</i>	Whole plant	Mixed infection of GIN	Amaranthaceae	Jabbar et al., 2007; Nayak et al., 2010
<i>Chenopodium ambrosioides</i>	Leaf	<i>Haemonchus contortus</i> ; Mixed infection of GIN	Amaranthaceae	Eguale & Giday, 2009; Salifou et al., 2013
	Essential oil	<i>Haemonchus contortus</i> ; Mixed infection of GIN		Ketzis et al., 2002; Macdonald et al., 2004
<i>Chrysophyllum cainito</i>	Stem	<i>Haemonchus contortus</i>	Sapotaceae	Fernandez et al., 2013
<i>Coriandrum sativum</i>	Seeds	<i>Haemonchus contortus</i>	Apiaceae	Eguale et al., 2007
<i>Cucurbita Mexicana</i>	Seeds	<i>Haemonchus contortus</i>	Cucurbitaceae	Iqbal et al., 2001
<i>Cymbopogon nardus</i>	Whole plant	<i>Haemonchus contortus</i>	Poaceae	Jeyathilakan et al., 2010
<i>Dalbergia latifolia</i>	Bark and Stem	<i>Haemonchus contortus</i>	Fabaceae	Daryatmo et al., 2010
<i>Elephantorrhiza elephantina</i>	Roots	<i>Haemonchus contortus</i>	Fabaceae	Maphosa et al., 2010
<i>Embelia ribes</i>	Seed	<i>Haemonchus contortus</i>	Myrsinaceae	Swarnkar et al., 2009
<i>Erythrina senegalensis</i>	Bark	<i>Haemonchus contortus</i>	Fabaceae	Williams et al., 2016
<i>Eucalyptus globulus</i>	Leaves	<i>Haemonchus contortus</i>	Myrtaceae	Kanojia et al., 2016
<i>Fagara heitzii</i>	Leaves	Mix infection of GIN	Rutaceae	Hounzangbe et al., 2005
<i>Ferula foetidissima</i>	Not Reported	<i>Haemonchus contortus</i>	Apiaceae	Pustovoi, 1968
<i>Ficus religiosa</i>	Bark	<i>Haemonchus contortus</i>	Urticaceae	Iqbal et al., 2001
<i>Fumaria parviflora</i>	Whole plant	Mixed infection of GIN	Fumariaceae	Al-Shaibani et al., 2009a
<i>Hagenia abyssinica</i>	Fruit	Mixed infection of GIN	Rosaceae	ITDG and IIRR, 1996
<i>Heracleum sosnowskyi</i>	Not Reported	Mixed infection of GIN	Apiaceae	Gadzhiev and Eminova, 1986a, b
<i>Lagenaria siceraria</i>	Seed	<i>Haemonchus contortus</i> ; <i>Pheretima posthuma</i> (Earthworm)	Cucurbitaceae	Khan et al., 2010a
<i>Lawsonia inermis</i>	Flower and seed	<i>Eicinia fetida</i> (Red californian earthworm)	Lythraceae	Wadekar et al., 2016
<i>Leonotis leonurus</i>	Leaves	Mix infection of GIN	Lamiaceae	Maphosa et al., 2010
<i>Leucaena leucocephala</i>	Leaves	<i>Haemonchus contortus</i>	Fabaceae	Alonso-Diaz et al., 2008
<i>Lippia sidoides</i>	Essential oil	Mix infection of GIN	Verbenaceae	Rashid et al., 2016
<i>Lysimila latisliliquum</i>	Leaves	<i>Haemonchus contortus</i> ; Mix infection of GIN	Fabaceae	Camurça-Vasconcelos et al., 2008
<i>Mallotus philippensis</i>	Fruit, powder	Mix infection of GIN	Euphorbiaceae	Alonso-Diaz et al., 2008; Brunet et al., 2008
<i>Melia azedarach</i>	Fruit	Mixed infection of GIN	Meliaceae	Gangwar et al., 2013
<i>Momordica charantia</i>	Fruits	<i>Haemonchus contortus</i>	Cucurbitaceae	Cala et al., 2012
<i>Moringa oleifera</i>	Root	Mixed infection of GIN	Moringaceae	Rashid et al., 2016
<i>Musa paradisiaca</i>	Leaves	Mix infection of GIN	Musaceae	Salles et al., 2014
<i>Myracrodruon urundeuva</i>	Leaves	<i>Haemonchus contortus</i>	Anacardiaceae	Hussain et al., 2010, 2011, Marie-Magdeleine et al., 2014
<i>Myrsine Africana</i>	Leaves	<i>Haemonchus contortus</i>	Myrsinaceae	de Oliveira et al., 2011
<i>Nicotiana tabacum</i>	Leaves	<i>Haemonchus contortus</i>	Solanaceae	Getachew et al., 2012
				Iqbal et al., 2006, Worku et al., 2009, Epperson, 2013, Hamad et al., 2013
<i>Nigella sativa</i>	Seed	<i>Haemonchus contortus</i>	Ranunculaceae	Burke et al., 2009, Shalaby et al., 2012
<i>Piscidia piscipula</i>	Leaves	<i>Haemonchus contortus</i>	Fabaceae	Alonso-Diaz et al., 2008
<i>Rapanea melanophloeos</i>	Fruits	<i>Haemonchus contortus</i>	Myrsinaceae	Githiori et al., 2002
<i>Scutia myrtina</i>	Roots	<i>Haemonchus contortus</i>	Rhamnaceae	Ayers et al., 2007
<i>Semecarpus anacardium</i>	Nut	Mixed infection of GIN; <i>Haemonchus contortus</i>	Anacardiaceae	Pal et al., 2008; Tandon et al., 2011
<i>Spigelia anthelmia</i>	Aerial parts	<i>Haemonchus contortus</i>	Loganiaceae	Ademola et al., 2007
<i>Thymus capitatus</i>	Aerial parts	Mixed infection of GIN	Lamiaceae	Elandalousi et al., 2013
<i>Trachyspermum ammi</i>	Seed	<i>Haemonchus contortus</i>	Apiaceae	Jabbar et al., 2006, Lateef et al., 2006c
<i>Trianthema portulacastrum</i>	Whole plant	Mix infection of GIN; <i>Haemonchus contortus</i>	Aizoaceae	Hussain et al., 2011, de Mello et al., 2013
<i>Vernonia anthelmintica</i>	Seed	<i>Haemonchus contortus</i>	Asteraceae	
<i>Zingiber officinale</i>	Rhizome	Mixed infection of GIN	Zingiberaceae	Peachey et al., 2015
<i>Ziziphus nummularia</i>	Bark	Mixed infection of GIN	Rhamnaceae	Bachaya et al., 2009

GIN = gastrointestinal nematodes

Only those plants were selected which showed significant reduction (35%, 5&gt;) in fecal egg counts as compared to control

Thus, protocol of this test has been revolutionized by using distilled deionized water as solvent of drugs in lieu of tap water (von Samson-Himmelstjerna *et al.*, 2009).

**Larval migration inhibition assay:** Young larvae, less than two weeks, are kept with various concentrations of candidate drugs in dark. Post 24 h incubation, contents are sieved and larvae are allowed to migrate for 24 h. The sieve containing un-migrated larvae is washed carefully in separate wells. Success of the test is measured by inability of larvae to migrate through sieve due to paralysis (Alonso-Díaz *et al.*, 2011; Kamaraj and Rahuman, 2011).

**Larval development assay:** This test measure shifts of first larval stages of Trichostrongylids to infective stage (Third larval stage) and takes 6–7 days to be completed (Demeler *et al.*, 2010). Briefly, eggs are collected from faeces of donor animals. At least five serial dilutions (10 µL) of candidate extract dissolved in DMSO/Distilled are dispensed in different wells of 96 well micro titration plate having 190 µL Agar (2%), nutritive media (Earle's balanced salt solution, Bacteria and yeast extract) and amphotericin B. About 100 eggs (10 µL) are dispensed in these wells containing plant extract and incubated at 25°C for 6 days (to obtain L3). At the end of incubation period Lugol's iodine solution is added in each well and number of eggs (un-hatched), L1, L2 and L3 are counted.

**Adult motility assay:** The basic theology of this test is similar as described in larval migration inhibition test. Adult worms collected from freshly slaughtered animals are subjected to various drug concentrations for 6 h. Observance of motility for 30 min in PBS post-treatment is main criteria. Briefly, adult worms collected from freshly slaughtered animals are subjected to various drug concentrations for 6 h. Dead and alive worms are counted in each test concentration at hourly basis. To confirm death of the worm non-motile worms are exposed to luke warm water for ~ 1 min. This test is used to obtain time dependent as well as dose dependent data on mortality of worms (Ferreira *et al.*, 2016; Raza *et al.*, 2016; Uppala *et al.*, 2016).

**In vivo:** Fecal egg count reduction test and post-mortem examination are the two tests to perform *in vivo* anthelmintic assessment trials. Former test is adopted most due to ease of conduction and being inexpensive (Lone *et al.*, 2012, 2013; Mondal *et al.*, 2015; Grzybek *et al.*, 2016).

**Fecal egg count reduction test:** Efficacy is measured through reduction of number of eggs in fecal samples post-treatment with candidate drug (Khan *et al.*, 2015; Meenakshisundaram *et al.*, 2016). Fecundity of *H. contortus* is high. So, it yields best reliable results in case of *H. contortus* due to strong positive correlation between magnitudes of infection with number of eggs in feces. Animals of about same age, either sex and approximately equal weight are randomly divided in treatment groups. Animals are administered with different levels of plant extracts at day 0 and faecal egg counts are performed at day 0, 7 and 14 post-treatment. Efficacy is

**Table 3:** Plant families (%) studied as acaricidal and anthelmintics

Family	Acaridcidal	Anthelmintic
Acanthaceae	0.41	1.41
Aizoaceae	0	1.41
Amaryllidaceae	0.41	2.87
Anacardiaceae	0.41	4.23
Apiaceae	0	7.04
Apocynaceae	0	2.87
Arecaceae	0	1.41
Asphodelaceae	0	1.41
Asteraceae	1.29	2.84
Bromeliaceae	0.43	0.71
Burseraceae	0	0.71
Compositae	0	0.71
Cucurbitaceae	0.43	2.84
Ebenaceae	0	0.71
Euphorbiaceae	0	0.71
Fabaceae	2.15	16
Fumariaceae	0	0.71
Lamiaceae	2.58	2.28
Loganiaceae	0	0.71
Lythraceae	0	0.71
Meliaceae	0.86	2.28
Mimosaceae	0	0.71
Moringaceae	0	0.71
Musaceae	0	0.71
Myrsinaceae	0	2.13
Myrtaceae	0.86	1.42
Poaceae	0.86	0.71
Ranunculaceae	0	0.71
Rhamnaceae	0	1.42
Rosaceae	0	1.42
Sapotaceae	0	0.71
Solanaceae	0.86	0.71
Verbenaceae	0.43	0.71
Woodsiaceae	0	0.71
Zingiberaceae	0	1.42

measured through reduction of number of eggs in fecal samples post-treatment with candidate drug. Fecundity of *H. contortus* is high. So, it yields best reliable results in case of *H. contortus* due to strong positive correlation between magnitudes of infection with number of eggs in feces (Rowe *et al.*, 2008).

## Conclusion

Empirical and scientific evidence suggest several plants are moderately effective against ticks and helminthes in animals. Though there are several papers published and databases developed on the use of plants in animal health, yet lot has to be done to document the rich indigenous knowledge existing in different cultures around the globe. There is strong need to document the plants with their botanical and local names, indications of the usage both in technical and local languages, and standardization of plant based products as to their formulation and dosage. There is also need to develop more sensitive, convenient and reliable screening assays for scientific validation of the plants.

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