

Review

## Phytotherapy in the control of helminthiasis in animal production

Erika Cosendey Toledo de Mello Peixoto<sup>1\*</sup>, Andressa de Andrade<sup>2</sup>, Fillipi Valadares<sup>2</sup>,  
Luciana Pereira da Silva<sup>3</sup> and Regildo Márcio Gonçalves da Silva<sup>3</sup>

<sup>1</sup>Universidade Estadual do Norte do Paraná (UENP/Bandeirantes), BR-369, Km 54, Villa Maria, Caixa Postal 261, CEP 86360-000, Bandeirantes, Paraná, Brasil.

<sup>2</sup>Zootecnista, Rua Acapulco, n 293, Bairro Vila Industrial, CDEP – 85904-120, Toledo, Paraná, Brazil.

<sup>3</sup>Faculdade de Ciências e Letras de Assis, Universidade Estadual Paulista “Júlio de Mesquita Filho”, Laboratório de Fisiologia Vegetal e Fitoterápicos, Avenida Dom Antônio, 2100, CEP: 19806-900, Assis-SP, Brasil.

Accepted 29 March, 2013

Parasitic diseases constitute one of the main problems affecting livestock; however the use of chemical medicaments provides drug resistance residues in animal and environmental contamination. Changes in production concepts require that food must be produced in hygienic conditions, per healthy animals and that are not eliminating antibiotic residues, pesticides or other drugs. This scenario has favored organic production and the use of medicinal plants. For the control of endoparasites, several studies have published the benefits of *Azadirachta indica* A. Juss., *Punica granatum* Linn., *Musa* sp., *Operculina hamiltonii* G. DON., propolis, among others. However, despite the existence *in-vitro* studies that demonstrate the pharmacological properties of phytotherapics, there is still need for clinical trials to determine dosage and its effects *in-vivo*. Investigations of new bioactive natural substances can be of great value for the control of animal health and food safety, which is particularly important for organic production systems in which the use of chemical drugs is a limiting factor for certification.

**Key words:** Agroecology, biological agriculture, medicinal plants, parasitic worms.

### INTRODUCTION

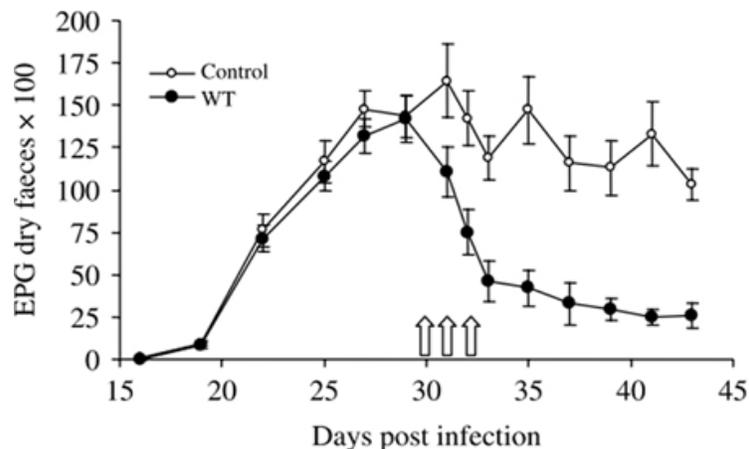
Parasitic diseases are one of the main problems affecting livestock. Losses with lower weight gain, increased mortality, lower carcass yield, lower production of meat and milk, and drug spending are factors that harm the profitability and animal welfare. These losses can be reduced through changes in pasture management, animal nutrition, and the use of chemicals anti-parasitic. Often anthelmintic chemicals, despite showing weak binding ability to the tissues, have high binding to plasma proteins. This results in longer retention of medicaments residue in the serum. Traditionally single doses of anthelmintics are administered, but the use of slow release drugs, also have result in increased risk of residues in food. Moreover, inadequate administration,

use of lower doses, misdiagnosis and lack of rotation of pharmacological bases, have caused drug resistance (Souza et al., 2008; Lima et al., 2010).

Consumers in different countries require more natural foods and higher quality (Casemiro and Trevizan, 2009). Changes in production concepts require that food must be produced in hygienic conditions, by healthy animals and which are not eliminating antibiotic residues, pesticides or other drugs. The use of growth promoters as antibiotics has been banned by the European common market, in poultry and pigs, since January 2006 (Regulation of European Commission no. 1881/2006).

The policy approach of the European Union, in relation to food safety aims assure a high level of safety, health

\*Corresponding author. E-mail: emellopeixoto@uenp.edu.br.



**Figure 1.** FEC (mean and standard deviation 0.2) of BHP sheep receiving WT drench, during experimental infection with mixed nematodes (Expt 2). The three arrows indicate the days of drench administration (Max et al., J. Agric. Sci. (2009), 147:211–218).

and welfare of animals in all its member States. For this, each member introduced control measures from farm to the consumer's table, focusing on proper surveillance (Regulation n° 178/2002 of the European Council, 2002). The protection of consumers to residues of pharmacologically active substances in foodstuffs of animal origin is guaranteed by the National Plan for Waste Management, which is based on Directive n° 96/23/EC of 1996 of the European Council. Thus, all countries wishing to export food products to the European Union must offer products which residue concentration does not exceed the limits, or does not present residues of substances classified as hazardous to consumers' health (Regulation n° 2377/90 of the European Council, 1990).

This scenario has favored the agroecologic and organic products' market. Organic agriculture is practiced over 35 million hectares in 154 countries. Despite the global recession, demand keeps growing. The European market was the most affected, but countries like Portugal and France, showed growth rates above 15% (Sahota, 2012). The North American market surpassed Europe in 2010, although the supply of organic foods continues short of demand. Thus, Latin America has become a major supplier of organic foods.

However, these systems require that the food must be produced without any harmful residues. Thereby advances in the search for natural methods, have been obtained from the exploitation of herbal medicines (Nery et al., 2009). Searches related to natural products are justified by the possibility of avoiding and/or minimizing the use of chemicals, reducing the occurrence of drug resistance, and the presence of residues in animal products. However, although there is a great variability of medicinal plants, their use in veterinary medicine is still not disseminated. Thus, the present study summarizes the

main results obtained from the use of medicinal plants, for the control of helminth infections in production animals.

### Tanniferous plants

Several authors reported the promising potential of plants rich in tannin on nematode control (Oliveira et al., 2011a). The anthelmintic activity of tannins *in vitro* was characterized by decrease in hatching, reduced development and motility of larvae and of the adults (Joshi et al., 2011). *In vivo*, they caused reductions of eggs per gram of faeces (EPG) and the level of parasitic infestation (Max et al., 2009) (Figure 1). It was postulated that tannins can still exert the action as anthelmintic indirectly by enhancing the immune response. Its protection on the ingested proteins, against rumen degradation, would facilitate greater availability, of these proteins, in the lower gastrointestinal tract (Otero and Hidalgo, 2004). The tannins are not absorbed by the gastrointestinal tract of small ruminants, occurs disposal in feces and it reduces the pasture contamination (Mupeyo et al., 2011).

Oliveira et al. (2011b) evaluated the effects of extracts from the leaf and stem of *Anadenanthera colubrina* (Vell.) Brenan, *Leucaena leucocephala* (Lam.) de Wit and *Mimosa tenuiflora* (Willd.) Poir. On larval exsheathment of *Haemonchus contortus in-vitro* and verified the role of tannins in this process. Third-stage larvae of *H. contortus* were incubated with extracts for 3 h and were exposed to sodium hypochlorite solution. The extracts were tested at 300 µg.mL<sup>-1</sup> and accompanied by controls: phosphate buffer solution (PBS) and polyvinyl polypyrrolidone (PVPP). The larval exsheathment was evaluated for 60 min. The 6 extracts blocked larval exsheathment. After

PVPP addition, a tannin inhibitor, the exsheathment percentage was similar to the PBS ( $p > 0.05$ ), except for *L. leucocephala* and *M. tenuiflora* leaf extracts. However, pre-incubation with PVPP of these 2 extracts significantly changed larval exsheathment when compared to the non-treated extracts ( $p < 0.05$ ). These results suggest that *A. colubrina*, *L. leucocephala* and *M. tenuiflora* could be useful in gastrointestinal nematode control and that tannins are probably the main compounds involved in the observed effects.

Thus, consumption of tanniferous plants can determine important perspectives for the control of nematodes.

### Propolis

Throughout history, man has learned to use natural products and one of the most used has been the propolis. Propolis is characterized resinous and balsamic material, collected by bees from the branches, flowers, pollen, buds and exudates of trees. It has been used in the treatment of various diseases in animals (Coelho et al., 2010). Its antibacterial activity (Orsi et al., 2012), anti-inflammatory and immunostimulant (Fischer, 2008) are particularly important for the supportive therapy of parasitosis.

Ghanem et al. (2009) investigated the gastroenteritis caused by coccidial infection in domestic goats. *Eimeria* oocysts were found in 66% of the fecal samples examined, and 6 species of *Eimeria* were morphologically identified. 4 groups of kids were used: healthy kids used as control (CH), naturally infected kids and left untreated, kids treated with Toltrazuril (20 mg/kg body weight, orally for 2 weeks) and kids treated with Propolis (1 ml of 3% aqueous solution/liter of drinking water for 7 days). The results showed that Toltrazuril was highly effective as an anticoccidial drug (94.33% reduction of OPG) and was more effective than propolis which moderately reduced OPG (54.66%). The efficacy of both drugs was further compared based on antioxidant assays, serum biochemical analysis and histopathological changes. There was a significant reduction ( $p < 0.05$ ) in erythrocyte reduced glutathione, glutathione reductase, superoxide dismutase, catalase activities, serum albumin, calcium, sodium and potassium in *Eimeria*-infected kids compared to control. On the other hand, significant elevations ( $P < 0.05$ ) in serum malondialdehyde, iron and nitrate were recorded in infected animals compared to control. The total protein and phosphorus showed non-significant decrease compared to control. Propolis and Toltrazuril treatment significantly alter the serum biochemical aberrances toward the control values. Heinzen et al. (2012) evaluated holstein calves and their crosses, with 90 days old, with *Trichostrongylus* sp. and *Strongyloides* sp. It was directly administered orally, 10 ml of alcoholic extract of propolis 30%, every 8 h for 4 consecutive days.

It was observed an average reduction of 48.48% of egg fecal output in 83% of animals. These values are not considered effective (Wood et al., 1995), but it is important to emphasize that they represent initial results. Perhaps, from additional studies by assessing different doses, dosage and frequency of administration, best results may be achieved. Preliminary tests *in-vitro* are important early step to validation of medicinal plants. However, in veterinary medicine, unlike human medicine, studies in animals are still scarce (Nery et al., 2009).

### *Azadirachta indica* A. Juss

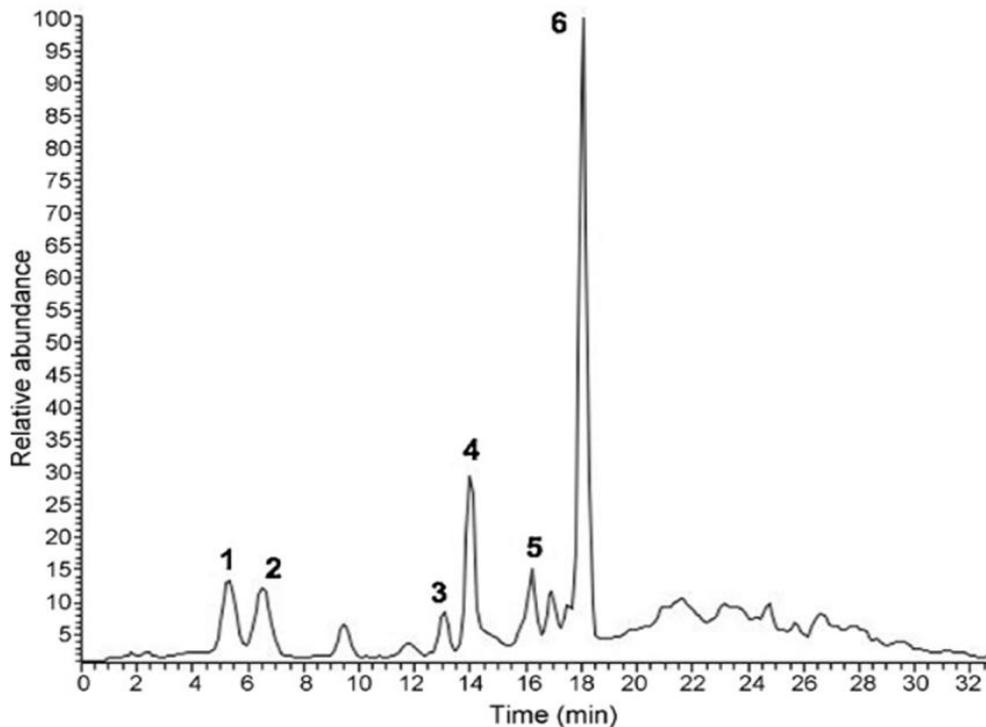
The Neem (*Azadirachta indica* A. Juss.) is a tree of the family Meliaceae, originally from India, that can reach up to 30m tall and live up to 200 years. The inhabitants of India and Asian countries use the extract of the leaves and the oil extracted from the seeds for more than 2,000 years as fertilizer of soil and pest control in agricultural and livestock. Its main active principle, azadirachtin, is responsible for pesticidal activity (Forim et al., 2010). It has over 50 terpenoids compounds and most of them, act on insects (Deleito and Borja, 2008; Maciel et al., 2010), mites (Broglio-Micheletti et al., 2010) and helminths (Chagas and Vieira, 2007; Lipinski et al., 2011).

Chagas and Vieira (2007) evaluated aqueous extract of dried leaves of Neem in sheep. The concentration of 240.000 part per million (ppm) *in-vitro* has inhibited eclosion of larvae of gastrointestinal nematodes in 89%. However, tests *in-vivo* with smaller doses (30g of dried leaves per animal per day for five days) did not promote reduction of egg fecal output. Chagas and Vieira (2007) recommended that research studies should be directed to the biological action of the oil extracted directly from the seed, which has a higher amount of azadirachtin in comparison with the leaves. Moreover, the administration of seed oil is also easier, since the leaves have bitter taste and are avoided by the animals.

The anthelmintic effect of Neem and garlic was evaluated by administering in buffaloes of 2 g of Neem and 2 g of dehydrated garlic (Lipinski et al., 2011). The animals were treated daily during 6 months. Feces and blood were collected in the beginning of the test and 2 more times later. Helminth eggs per gram of feces counting technique (EPG), blood chemistry and hematologic tests were utilized. The animals had a significant reduction of EPG ( $p < 0.01$ ) in comparison with the beginning of the test. Furthermore, there were no hematological and biochemical indicatives of toxicity to the animals.

### *Punica granatum* Linn.

*Punica granatum* Linn. belongs to the Punicaceae family, is a woody shrub that has small leaves, orange-red



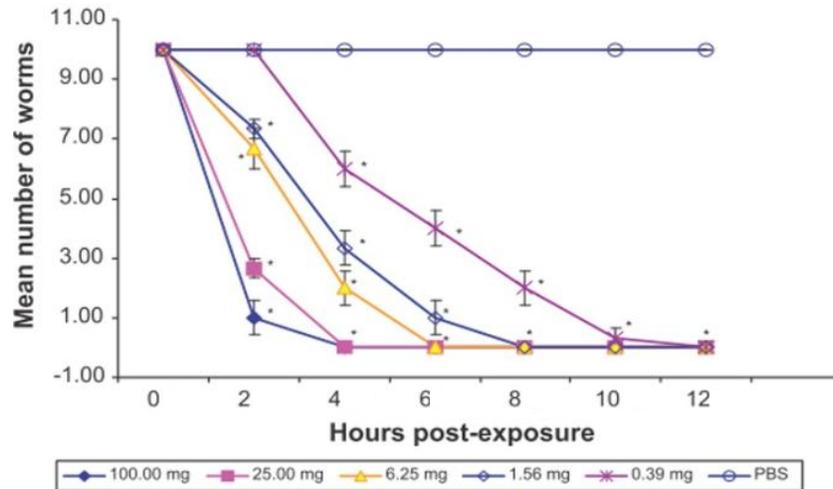
**Figure 2.** TIC chromatographic profile of *Pg*-FET. Peaks 1 to 2: punicalin isomers; peaks 3 to 4: punicalagin isomers; peak 5: ellagic acid exoside; Peak 6: ellagic acid. (Dell'Agli et al., J. Ethnopharmacol. 125 (2009) 279–285).

flowers, and fruits spherical at the end of its branches. Its main constituents are alkaloids (pelletierine, isopeletierine, metilpeletierine), tannins, phenolic compounds (anthocyanins, quercetin, phenolic acids) and flavonoids. Among its chemical constituents, it was observed that the punicalgina is probably a major contributor to the antimicrobial effect observed (Werkman et al., 2008).

In Brazil, *Punica granatum* Linn. is popularly used after boiling the peel of the fruit in water, against pharyngitis, tonsillitis and other infections (Silva et al., 2008). By studying the therapeutic applications of pomegranate, Werkman et al. (2008) stressed the possibility of employment in nosocomial infections due to bacterial resistance to conventional antibiotics. They still highlighted that the use of pomegranate can be easily prepared without compromising biological properties.

Pomegranate shows properties as anthelmintic, antibacterial, anti-inflammatory and antioxidant (Menezes et al., 2008). Its antioxidant action is relevant by the sequestration of free radicals and other oxidants, responsible for several degenerative cellular changes, often associated with chronic conditions such as the case of parasitoses. Flavonoids exhibit ideal conformation structural for scavenging free radicals, they are found in all plant organs, mainly in the fruits. Because it is rich in tannins, the *P. granatum* Linn. has been considered for the control of helminths in livestock. Dell'Agli et al. (2009)

studied *in vitro* the antiplasmodial activity of the methanolic extract of *P. granatum* Linn. by determining the activity of crude extracts, purified fractions and isolated compounds on asexual blood stages of *Plasmodium falciparum*. Additionally these authors assessed *in-vivo* the activity of the asexual and sexual stages developing in the vertebrate and mosquito host, using the rodent malaria model *Plasmodium berghei*. Urolithins, the ellagitannin metabolites, were also investigated for antiplasmodial activity. Chloroquine susceptible and resistant strains of *Plasmodium falciparum* were used for *in vitro* studies and the rodent malaria model *Plasmodium berghei*—BALB/c mice was used for *in vivo* assessments. Recombinant plasmepsins 2 and 4 were used to investigate the interference of *Punica granatum* Linn. compounds with the metabolism of haemoglobin by malaria parasites. The *The Punica granatum* Linn. methanolic extract inhibited parasite growth *in vitro* with a  $IC_{50}$  of 4.5 and 2.8 g/ml, for Chloroquine susceptible and resistant strains, respectively. The activity was found to be associated to the fraction enriched with tannins (*P. granatum*-FET,  $IC_{50}$  2.9 and 1.5 g/ml) in which punicalagins (29.1%), punicalins, ellagic acid (13.4%) and its glycoside could be identified (Figure 2). Plasmepsin 2 was inhibited by *P. granatum* - extract and by *P. granatum* -FET ( $IC_{50}$  7.3 and 3.0 g/ml), which could partly explain the antiparasitic effect. On the contrary, urolithins were inactive. Both



**Figure 3.** Graph showing the time and dose-dependent *in vitro* anthelmintic activity of *Musa paradisiaca* L. crude aqueous methanol extracts at 100.0–0.39 mg/ml concentrations in comparison with positive control levamisole (0.5 mg/ml), on mature live *Haemonchus contortus* of sheep. The inhibition of motility and/or mortality of the worms were used as the criterion for anthelmintic activity. Values shown are means, asterisk (\*) indicates significantly different from previous value at  $P < 0.05$  (Hussain et al., *Veterinary Parasitology* 179 (2011) 92–99).

*P. granatum*-MeOH extract and *P. granatum*-FET did not show any *in-vivo* efficacy in the murine model. The *in-vitro* studies support the use of *P. granatum* Linn. as antimalarial remedy.

Anjos et al. (2012) evaluated the anthelmintic effect of extract of the stalk bark of *Punica granatum* Linn. and the extract of *Musa paradisiaca* Linn. leaves (*Musa paradisiaca*). Using feces of naturally infected cattle. Anjos et al. (2012) evaluated aqueous extracts made from dried plant material and fresh. Better results were obtained by the extract of the dried bark of the stem of the pomegranate, which showed efficacy of 92.29% for *haemonchus*, and 96.97% for *Cooperia*. The aqueous extracts of banana leaves, were not effective, although several species of banana leaves exhibit tannins in its constitution.

### ***Musa* sp.**

The banana may represent an important perspective to the natural control of helminths, not only due to the tannins in its constitution, but also for being one of the most consumed fruits. It was reported that its world's production exceeded 91 million tones (FAO, 2012).

Batatinha et al. (2004) had already examined *in-vitro* the anthelmintic efficacy of the aqueous extract of the banana leaves (*Musa cavendishii* Lin.), and extract of papaya (*Carica papaya* Lin.). Higher efficacy was observed for treatment derived from banana leaves. These tests were performed *in vitro*, and they detected 95% reduction of the superfamily Strongyloidea, using the concentration of 130.6 mg/ml of extract of banana

leaves. Regarding the papaya seed extract, this result was only achieved using concentrations between 464 and 290 mg/ml. They concluded that 97.9% inhibition of larval development of *Haemonchus* spp. may represent an important perspective for the use of extract of banana leaves *in-vivo* tests.

Oliveira et al. (2010) evaluated the *in-vitro* efficacy of extracts of banana crop residues on the inhibition of larval gastrointestinal nematodes in sheep, among them *Haemonchus* spp. They have rated aqueous extracts of leaves, hearts and pseudostems of the banana Silver dwarf cultivar. They observed that all 3 extracts in concentrations equal to or greater than 75 mg/ml, reduced larval development with effectively above 96.9%.

Altaf et al. (2011) evaluated of anthelmintic effects of *Trianthema portulacastrum* Linn. whole plant and *Musa paradisiaca* Linn. leaves against prevalent gastrointestinal worms of sheep. *In-vitro*, it was determined using mature female *H. contortus* and their eggs in adult motility assay and egg hatch test, respectively. *In vivo* anthelmintic activity of crude powder and crude aqueous methanolic extract in increasing doses (1.0-8.0 g kg<sup>-1</sup>) was determined in sheep naturally infected with mixed species of nematodes using fecal egg count reduction test and larval counts from coprocultures. These test were performed pre and post-treatments. Crude aqueous methanolic extracts showed a strong *in-vitro* anthelmintic activity and pronounced inhibitory effects on *H. contortus* egg hatching. Both plants exhibited dose and time dependent anthelmintic effects on live worms as well as egg hatching. *M. paradisiaca* (LC<sub>50</sub> = 2.13 µg ml<sup>-1</sup>) was found to be more potent than *T. portulacastrum* (LC<sub>50</sub> = 2.41 µg ml<sup>-1</sup>) in egg hatch test (Figure 3). However, *in-*

*vivo*, maximum reduction in eggs per gram of feces was recorded as 85.6 and 80.7% with crude aqueous methanolic extract of *T. portulacastrum* and *M. paradisiaca* at 8.0 g kg<sup>-1</sup> on 15th day post-treatment, respectively as compared to that of Levamisole (7.5 mg kg<sup>-1</sup>) that caused 97.0% reduction in eggs per gram. All the species of gastrointestinal nematodes, *H. contortus*, *Trichostrongylus spp.*, *Oesophagostomum columbianum* and *Trichuris ovis* which were prevalent, found susceptible ( $P < 0.01$ ) to the different doses of both plants.

### ***Operculina hamiltonii* (G. Don) D.F. Austin & Staples (1983)**

*Operculina hamiltonii* G. Don. is a well-known plant in northeastern of Brazil. Its anthelmintic activity was evaluated on gastrointestinal nematodes of goats naturally infected. Additionally, the effects were observed associated or not by extract *Typha domingensis* Pers. (Silva et al. 2010). 30 caprine of Moxotó breed were submitted to the rhizome of *Typha domingensis* Pers. and to the root of *Operculina hamiltonii* G Don. The treatments were performed orally for 3 consecutive days, at a dose of 10 g of *Typha domingensis* pers. and 9 g of *Operculina hamiltonii* G Don. for 20 kg body weight. *O. hamiltonii* G Don. has determined percentage of reduction of egg fecal output corresponding at 84% and 70% on the seventh day and twenty-fifth day after treatment, respectively. Regarding the *Typha domingensis* Per, this plant showed reduction of 48% and 46% for the same evaluated periods.

Another experiment *in-vitro* was realized to evaluate the action of the ethanolic extract of *O. hamiltonii* G Don. on eggs and larvae of gastrointestinal nematodes of goats (Araujo et al., 2011). The extract was used at concentrations of 50, 25, 12.5, 6.25 and 3.12 mg.ml<sup>-1</sup> in both tests. The percentage of viable eggs decreased as the concentration of the extract increased. When extract concentration reached 25%, the percentage of viable eggs decreased to 53.07%, and decreased further to 29.57% at concentration of 50%. The concentration of the extract of *O. hamiltonii* G Don. was responsible for the reduction of the percentage of viable larvae, where it was observed that the concentration of 50% affected negatively 66.87% of the larvae.

Brito-Junior et al. (2011) evaluated alcoholic extract of *Operculina hamiltonii* G Don. and *Marmodica charantia* Linn. against helminths of caprine naturally infected. The treatments were performed orally for 3 consecutive days, and the results were evaluated by the egg fecal output and larviculture. The extract *Operculina hamiltonii* G Don. has determined average reduction 63% of egg fecal output in 30 days, and 90% at 60 days after treatment, while *Marmodica charantia* Lin. did not decreased egg fecal output significantly.

However, larvicidal and ovicidal effect of the ethanol

extract of leaves of *Marmodica charantia* Linn. were demonstrated by Cordeiro et al. (2010). This extract was evaluated at the concentrations of 50, 25, 12.5, 6.25 and 3.12% on feces of caprine naturally infected with gastrointestinal nematodes. The plates were examined under optical microscope to count the eggs in development and mobile larvae after 24, 48 and 72 h of incubation. The ovicidal and larvicidal activities, for larval motility test were observed in the concentrations above 12%.

### ***Prosopis sp.***

*Prosopis juliflora* Sw. was also assessed on larvae of gastrointestinal nematodes of caprine animals (Batatinha et al., 2011). Using their leaves, these researchers conducted 2 types of extracts methanol and aqueous. 6 different concentrations of the methanol extract (724.5; 557.3; 428.7; 329.8; 253.7 and 195.1 mg/ml) and 1 of the aqueous extracts (110.0 mg/ml) were used for the treatment of larvae cultures, in triple assays. Distilled water and doramectin were used to treat cultures considered to be negative and positive control, respectively. The results revealed a reduction of more than 90% of the infective larvae between the concentrations of 724.5 up to 253.7 mg/ml for the methanol extract and a low percentage of reduction (59.87%) for the aqueous extract. Only the methanol extract of *Prosopis juliflora* Sw. was effective in the *in-vitro* treatment of gastrointestinal nematodes of goat.

The anthelmintic effect of *Prosopis laevigata* (Willd.) MC Johnst (mezquite) n-hexanic extract was evaluated against *H. contortus* endoparasitic stages in artificially infected gerbils (De Jesu's-Gabino et al., 2010). The *in-vivo* effect of the plant extract was evaluated in gerbils artificially infected with *H. contortus*. Plant extract concentration was 40 mg/ml. 3 groups of gerbils were as follows: group 1, *P. laevigata* extract at 100 ml intraperitoneally; group 2, control – Tween 20 in water at a single dose of 100 ml IP; group 3 also served as a control, receiving water only, to determine the mortality due to causes other than the plant extract. An additional group (group 4) was administered fenbendazole, as a positive control. 5 days later the animals were euthanized and stomach and mucosa removed to quantify the nematodes. The parasite population in the plant extract treated group 1 was reduced by 42.5% ( $P < 0.05$ ) with respect to the control group 2; and when control group 3 was used for comparison the parasitic reduction was estimated as 53.11%. This study shows the *in-vivo* anthelmintic effect of *P. laevigata* n-hexane extract for the first time, using gerbils as an *in-vivo* model, with potential use in sheep.

### ***Lippia sidoides* Cham.**

*Lippia sidoides* Cham. is a plant that has been often used

**Table 1.** Plant species evaluated for antiparasitic activity for production animals.

| Family         | Plant                                | Action   | References  |
|----------------|--------------------------------------|--|---|
| Aizoaceae      | <i>Trianthema portulacastrum</i> L.  | Inhibitory effects on egg hatching; reduction EPG  | Altaf et al. (2011)   |
| Convolvulaceae | <i>Operculina hamiltonii</i>         | Reduction EPG; percentage of viable eggs decreased   | Silva et al. (2010), Araujo et al. (2011), Brito-Junior et al. (2011) |
| Cucurbitaceae  | <i>Marmodica charantia</i> L.        | Did not decreased EPG; larvicidal and ovicidal   | Brito-Junior et al. (2011), Cordeiro et al. (2010)                    |
|                | <i>Anadenanthera colubrina</i>       |  |   |
| Fabaceae       | <i>Leucaena leucocephala</i>         | Larval exsheathment  | Oliveira et al. (2011b)   |
|                | <i>Mimosa tenuiflora</i>             |  |   |
|                | <i>Prosopis juliflora</i> Sw         | Reduction of the infective larvae  | Batatinha et al. (2011)   |
|                | <i>Prosopis laevigata</i> (mesquite) | Stomach and mucosa removed to quantify the nematodes; The parasite population in the plant extract treated group 1 was reduced           | De Jesu´s-Gabino et al. (2010)  |
| Meliaceae      | <i>Azadirachta indica</i>            | Inhibited eclosion of larvae<br>Reduction of EPG   | Chagas and Vieira (2007)<br>Lipinski et al. (2011)                    |
|                | <i>Carapa guianensis</i>             | Reduction of the total number of larvae  | Farias et al. (2010)  |
| Musaceae       | <i>Musa cavendishii</i> L.           | Inhibition of larval development   | Batatinha et al. (2004)   |
|                | <i>Musa paradisiaca</i>              | Inhibition of larval development; larval exsheathment<br>Reduced larval development<br>Inhibitory effects on egg hatching; reduction EPG | Anjos et al. (2012)<br>Oliveira et al. (2010)<br>Altaf et al. (2011)  |
| Myrtaceae      | <i>Eucalyptus globulus</i>           | Ovicidal and larvicidal activity   | Macedo et al. (2009)  |
| Punicaceae     | <i>Punica granatum</i> L.            | Antiplasmodial activity  | Dell'Agli et al. (2009)   |
|                |                                      | Decrease of larval viability   | Anjos et al. (2012)   |
| Typhaceae      | <i>Typha domingensis</i>             | Reduction EPG  | Silva et al. (2010)   |
| Verbenaceae    | <i>Lippia sidoides</i>               | Lower egg development  | Souza et al. (2010)   |

Eggs per gram of faeces (EPG).

as herbal medicine in northeastern of Brazil. The hydroalcoholic extract of *Lippia sidoides* Cham. was evaluated on embryonated eggs from feces of naturally infected goats (Souza et al., 2010). Embryonated eggs were obtained from feces of

goats naturally infected with Trichostrongylidae nematodes and the fecal egg count was determined by using the modified McMaster technique. 50 ml of the suspension containing 40 eggs were transferred to polystyrene plates and

incubated with 12 different concentrations, and evaluations were performed during 72 h at room temperature. The results demonstrated different efficacy of extracts, with lower egg development rate at 500 mg ml<sup>-1</sup>. In conclusion, the

hydroalcoholic extract of *L. sidoides* may play an important role on the *in-vitro* development of gastrointestinal nematode eggs, indicating ovicidal activity.

### ***Eucalyptus globules* Wood.**

*E. globulus* Wood. belongs to the family Myrtaceae. Despite being from Australia, this plant is now spread worldwide. The essential oil from the leaves presents important pharmaceutical activity due to properties anti-inflammatory, analgesic, antioxidant, antibacterial, antifungal, insecticide and miticide against *Boophilus microplus*. Its effect on the emergence and development of larvae of *H. contortus* has been evaluated by using different concentrations (Macedo et al., 2009). The chemical composition determination of *Eucalyptus* Wood. essential oil (EGEO) was through gas chromatography and mass spectrometry. Egg hatch test (EHT) was *globulus* performed in concentrations 21.75; 17.4; 8.7; 5.43 e 2.71 mg ml<sup>-1</sup>. In larval development test (LDT) they used the concentrations 43.5; 21.75; 10.87; 5.43 e 2.71 mg ml<sup>-1</sup>. Each trial was conducted by negative control with Tween 80 (3%) and positive control, 0.02 mg ml<sup>-1</sup> of thiabendazole in EHT and 0.008 mg ml<sup>-1</sup> of ivermectin in LDT. The maximum effectiveness of eucalyptus essential oil on eggs was 99.3% in concentration of 21.75 mg.ml<sup>-1</sup> and on larvae was 98.7% in concentration 43.5 mg ml<sup>-1</sup>. The concentration of eucalyptus essential oil that inhibits 50% of the eggs and larvae was 8.3 and 6.92 mg ml<sup>-1</sup>, respectively. The oil chemical analysis identified as main component of the monoterpen 1,8-cineol. Thus, eucalyptus oil showed ovicidal and larvicidal activity *in vitro* over *H. contortus*, determining its possible use for the control of gastrointestinal nematodes in sheep and goats.

### ***Carapa guianensis* Aubl.**

The *Carapa guianensis* Aubl. family Meliaceae, is economically important due to the value of its timber. Its seed can determine a yield of 70% of medicinal oil, which is used as antirheumatic, antibacterial and insect repellent. In order to evaluate the action of the seed oil of *C. guianensis* Aubl. in larvae of gastrointestinal nematodes of goats and sheep. Farias et al. (2010) examined different concentrations: 100, 50, 30, 25 and 10%. Regarding at ewes, the fecal culture identified, the presence of larvae of *Haemonchus*, *Oesophagostomum*, *Strongyloides* and *Trichostrongylus*, noting predominance of *Haemonchus*. The percentage of the reduction of the total number of larvae was highly effective, except for the *Strongyloides*. Farias et al. (2010) observed that the oil of *C. guianensis* Aubl. at concentrations of 100, 50 and 30%, was similar to the action of Doramectin. Table 1

presents the plant species evaluated for antiparasitic activity for production animals.

## **CONCLUSION**

Investigations of new bioactive natural substance may be of great value for the control of animal health and food safety due to the possibility to decrease the quantity and frequency of use chemicals. This consideration is particularly important for agroecological production systems, organic or biological-dynamical, which use of chemical drugs is considered a limiting factor.

## **REFERENCES**

- Anjos C., Matsumoto LS, Fertoni LH. S, Silva RMG, Silva CS, Silva BT, Mello-Peixoto ECT (2012). Extracts of *Punica granatum* L. and *Musa paradisiaca* on larval inhibition of *Haemonchus* spp. and *Cooperia* spp. from bovines. VI Symposium Iberoamerican on Medicinal Plants. Proceedings... Ponta Grossa. 15-19 Jun. CD-ROM.
- Altaf H, Nisar KM, Iqbal Z, Sohail Sajid M, Kasib Khan M (2011). Anthelmintic activity of *Trianthema portulacastrum* 1 L. and *Musa paradisiaca* L. against gastrointestinal nematodes of sheep. Vet. Parasitol. 179:92-99.
- Araujo MM, Vilela VLR, Silva WA, Sousa RVR, Feitosa TF, Athayde ACR (2011). *In vitro* anthelmintic effectiveness of ethanolic extracts of *Operculina hamiltonii* (G. DON) D.F. Austin and Staples (1983) - Batata de Purga. Ars. Vet. 27(3):192-196.
- Brito-Junior L, Silva MLCR, Lima FH, Athayde ACR, Silva WW, Rodrigues RG (2011). Comparative study of anthelmintic action of purge potato (*Operculina hamiltonii*) and São Caetano Melon (*Momordica charantia*) in goats (*Capra hircus*) naturally infected. Sci. Agrotec. 35(4):797-802.
- Batatinha MJM, Santos MM, Botura MB, Almeida GM, Domingues LF, Almeida MAO (2004). *In vitro* effects of extracts of leaves of *Musa cavendishii* Linn. and seed of *Carica papaya* Linn. on cultures of larvae of gastrointestinal nematodes of goats. Braz. J. Med. Plants. 7:11-15.
- Batatinha MJM, Almeida GN, Domingues LF, Simas MMS, Botura MB, Cruz ACFG, Almeida MAO (2011). Effects of aqueous and methanol extracts of algarroba on cultures of larvae of gastrointestinal nematodes in Goats. Braz. Anim. Sci. 12(3):514-519.
- Brito-Junior L, Silva MLCR, Lima FH, Athayde ACR, Silva WW, Rodrigues RG (2011). Comparative study of the anti-helminthic action of the potato of purges (*Operculina hamiltonii*) and the cantaloups of São Caetano (*Momordica charantia*) in naturally infected goats (*Capra hircus*). Ciênc. Agrotec. 35(4):797-802.
- Broglio-Micheletti SMF, Dias NS, Valente ECN, Souza LA, Lopes DOP, Santos JM (2010). Action of extract and oil neem in the control of *Rhipicephalus* (*Boophilus*) *microplus* (Canestrini, 1887) (Acari: Ixodidae) in laboratory. Braz. J. Vet. Parasitol. 19:44-48.
- Casemiro AD, Trevizan SDP (2009). Organic Food: Challenges for the Public Domain of a Concept International [Workshop advances in cleaner production]. São Paulo; pp. 1-9. São Paulo – Brazil – May 20th-22nd – 2009.
- Chagas ACS, Vieira LS (2007). Ação de *Azadirachta indica* (Neem) em nematódeos gastrintestinais de caprinos. Braz. J. Vet. Res. Anim. Sci. 4(1):49-5.
- Coelho MS, Silva JhV, Oliveira ERA, Amâncio ALL, Silva NV, Lima RMB (2010). Propolis and its use in production animals. Arch. Anim. Sci. 59:95-112.
- Cordeiro LN, Athayde ACR, Vilela VLR, Costa JGM, Silva WA, Araujo MM, Rodrigues OG (2010). *In vitro* effect of ethanol extract of leaves São Caetano Melon (*Momordica charantia* L.) on eggs and larvae of gastrointestinal nematodes of goats. Braz. J. Medic. Plants 12(4):421-426.

- Council Regulation nº 2377/90 of European Council, 26 of June of 1990, laying down a Community procedure for the establishment of maximum residue limits of veterinary medicinal products in foodstuffs of animal origin. Official J. L 224:1-8.
- Council Regulation nº 178/2002 of Parliament and European Council, 28 of January of 2002, laying down the general principles and requirements of food law, establishing the European Food Safety Authority and laying down procedures in matters of food safety. Official J. Eur. Comm. 31:1-9.
- Council Regulation nº1881/2006 of European Council, de 19 of December of 2006, that setting maximum levels for certain contaminants in foodstuffs. Official J. Eur. Comm. L364:5-24.
- Deleito CSR, Borja GEM (2008). Neem (*Azadirachta indica*): an alternative in the control of flies in livestock. Braz. J. Vet. Res. 28:293-298.
- Dell'Agli M, Galli GV, Corbett Y, Taramellib D, Lucantonio L, Habluetzel A, Maschia O, Caruso D, Giavarini F, Romeod S, Bhattacharyae D, Bosisio E. Antiplasmodial activity of *Punica granatum* L. fruit rind. (2009). J. Ethnopharm. 125:279-285.
- Directive 96/23/CE of European Council, de 29 de Abril de 1996, concerning measures to control certain substances and residues thereof in live animals and animal products and repealing Directives 85/358/CEE e 86/358/CEE and the decisions 89/187/CEE e 91/664/CEE. Offic. J. L302:1-17.
- FAO (2012) Food and agriculture Organization of United Nations. Agriculture data base ProdSTAT. On line <http://faostat.fao.org/site/567/default.aspx#ancor> (August 07 2012).
- Farias MPO, Teixeira WC, Wanderley AG, Alves LC, Faustino MAG (2010). *In vitro* evaluation of the effects of seed oil *Carapa guianensis* Aubl. on larvae of gastrointestinal nematodes of goats and sheep. Braz. J. Med. Plants 12(2):220-226.
- Fischer G, Hübner SO, Vargas GD, Vidor T (2008). Immunomodulation induced by propolis. J. Biolog. Instit. 75(2):247-253.
- Forim MR, Matos AP, Silvia MFGF, Cass QB, Vieira PC, Fernandes JB (2010). The use of HPLC in the control of neem commercial products quality: Reproduction of the insecticide action. Quim. Nova. 33:1082-1087.
- De Jesu's-Gabino AF, Mendoza-de Give P, Salinas-Sa'nchez DO, Lopez-Arellano ME, Liebano-Hernandez E, Hernandez-Velazquez VM, Valladares-Cisneros G (2010). Anthelmintic effects of *Prosopis laevigata* n-hexanic extract against *Haemonchus contortus* in artificially infected gerbils (*Meriones unguiculatus*). J. Helminthol. 84:71-75.
- Ghanem MM, Mohamed ADA, Ramadan MY (2009). Clinical, biochemical and histopathological study on parasitic gastroenteritis associated with caprine coccidiosis: comparative effect of Toltrazuril and Propolis. Ion Ionescu de la Brad Iasi 11(1):565-580.
- Heinzen EL, Mello-Peixoto ECT, Jardim JG, Garcia RC, Oliveira NTE, Orsi RO (2012). Extract of propolis in the control of helminthiasis in calves. Acta Vet. Brasil. 6(1):40-44.
- Joshi BR, Kommuru DS, Terrill TH, Mosjidis JA, Burke JM, Shakya KP, Miller JE (2011). Effect of feeding sericea lespedeza leaf meal in goats experimentally infected with *Haemonchus contortus*. Vet. Parasitol. 178:192-197.
- Lima WC, Athayde ACR, Medeiros, GR, Lima DASD, Borburema JB, Santos EM, Vilela VLR, Azevedo SS (2010). Nematode resistant to some anthelmintics in dairy goats in Cariri Paraibano, Brazil. Braz. Vet. Res. 12:1003-1009.
- Lipinski LC, Martinez JL, Santos MVR, Ferreira JN, Pfau DR (2011). Evaluation of the anthelmintic effect and metabolic changes in buffaloes (*Bubalus bubalis*) with the administration of neem cake and dehydrated garlic in the Southern of Parana State Res. Braz. J. Agroecol. 6(3):168-175.
- Macedo ITF, Bevilaqua CML, Oliveira LMB, Camurça-Vasconcelos ALF, Vieira LS, Oliveira FR, Queiroz-Junior EM, Portela BG, Barros RS, Chagas ACS (2009). *Eucalyptus globulus* sobre *Haemonchus contortus* Ovicidal and larvicidal activity *in vitro* of *Eucalyptus globulus* essential oils on *Haemonchus contortus*. Braz. J. Vet. Parasitol. 18(3):62-66.
- Maciel MV, Morais SM, Bevilaqua CML, Amóra SSA (2010). *In vitro* insecticidal activity of seed neem oil on *Lutzomyia longipalpis* (Diptera: Psychodidae). Braz. J. Parasitol. 12:105-112.
- Max RA, Kassuku AA, Kimambo AE, Mtenga LA, WAKELIN D, BUTTERY PJ (2009). The effect of wattle tannin drenches on gastrointestinal nematodes of tropical sheep and goats during experimental and natural infections. J. Agric. Sci. 147:211-218.
- Menezes SMS, Pinto DN, Cordeiro LN (2008). Biologics activities *in vitro* and *in vivo* of *Punica granatum* L. (pomegranate). Braz. J. Med. 65(11):388-391.
- Mupeyo B, Barrya TN, Pomroya WE, Ramirez-Restrepo CA, Lopez-Villalobosa N, Pernthanerc A (2011). Effects of feeding willow (*Salix* spp.) upon death of established parasites and parasite fecundity. Anim. Feed Sci. Technol. 164:8-20.
- Nery PS, Duarte ER, Martins ER (2009). Effectiveness of plants for the control of gastrointestinal nematodes of small ruminants: A review of published studies. Braz. J. Med. Plants. 11(3):330-338.
- Oliveira NL, Duarte ER, Nogueira FA, Silva RB, Filho, DEF, Geraseev LC (2010). Efficacy of banana crop residues on the inhibition of larval development in *Haemonchus* spp. from sheep. Rural Sci. 40(2):488-490.
- Oliveira LMB, Bevilaqua CML, Morais S M, Camurça-Vasconcelos ALF, Macedo ITF (2011a). Plantas taniníferas e o controle de nematóides gastrintestinais de pequenos ruminantes. Rural Sci. 41(11):1967-1974.
- Oliveira LMB, Bevilaqua CML, Macedo ITF, Morais SM, Monteiro MVB, Campello CC, Ribeiro WLC, Batista EKF (2011b). Effect of six tropical tanniferous plant extracts on larval exsheathment of *Haemonchus contortus*. Rev. Bras. Parasitol. Vet. 20(2):155-160.
- Orsi RO, Fernandes A, Bankova V, Sforcin JM (2012). The effects of Brazilian and Bulgarian propolis *in vitro* against *Salmonella typhi* and their synergism with antibiotics acting on the ribosome. Natural Product Research: Formerly Natural Product Lett. 5:430-437.
- Otero MJ, Hidalgo LG (2004). Tanniferous plants and control of gastrointestinal nematodes of small ruminants. Livest. Res. Rural Dev. 16(2):1-9.
- Sahota A (2012). Global Organic Food and Drink Market Presented at: BioFach Congresss 2012, Messezentrum Nürnberg, Germany, February 15.
- Silva MAR, Higino JS, Pereira JV, Siqueira-Júnior JP, Pereira MSV (2008). Antibiotic activity of the extract of *Punica granatum* Linn. over bovine strains of *Staphylococcus aureus*. Braz. J. Pharmacogn. 18(2):209-212.
- Silva CF, Athayde ACR, Silva WW, Rodrigues, OG, Vilela VLR, Marinho PVT (2010). Evaluation of effectiveness of cattail (*Typha domingensis* Pers.) and potato-de-purge [*Operculina hamiltonii* (G. Don) DF Austin and Staples] fresh on gastrointestinal nematodes of goats naturally infected in semi-arid climate. Braz. J. Med. Plants 12(4):466-47.
- Souza AP, Ramos CI, Bellato V, Sartor AA, Schelbauer CA (2008). Resistance of bovine gastrointestinal helminths to anthelmintics in the Planalto Catarinense. Rural Sci. 38(5):1363-1367.
- Souza WMA, Ramos RAN, Alves LC, Coelho MCOC, Maia, MBS (2010). *In vitro* evaluation of "alecrim pimenta" (*Lippia sidoides* Cham.) hydroalcoholic extract (HAE) on the development of gastrointestinal nematode (Trichostrongylidae) eggs. Braz. J. Med. Plants 12(3):278-281.
- Werkman C, Granato DC, Kerbauy WD, Sampaio FC, Brandão AAH, Rode SM (2008). Therapeutic use of *Punica granatum* L. (pomegranate). Braz. J. Med. Plants 10(3):104-111.
- Wood IB, Amaral NK, Bairden K, Duncan JL, Kassai T, Malone JB, Pankavich JÁ, Reinecke RK, Slocombe O, Taylor SM (1995). Vercruyse, J. World Assoc. Advan. Vet. Parasitol. (W.A.A.V.P.) second edition of guidelines for evaluating the efficacy of anthelmintics in ruminants (bovine, ovine, caprine). Vet. Parasitol. 58:181-213.