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Full Length Research Paper

Comparative assessment of *in-vitro* anthelmintic effects of the aqueous extracts of the seeds and leaves of the African locust bean (*Parkia biglobosa*) on bovine nematode eggs

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The effects of the aqueous extracts of seeds and leaves of the African locust bean (*Parkia biglobosa*) on the viability of the eggs of cattle gastrointestinal helminthes was evaluated in cattle *in vitro*. The faecal egg counts (FEC) was determined using the modified McMaster technique. The seed extracts produced maximum inhibition of the hatchability of the helminth eggs at 0.3, 0.7 and 1.2 ml while the leave extracts produced maximum inhibition at 0.3 and 2.0 ml. The study concluded that the seed extracts has a better potential for use as an anthelmintic agent than the leaf extract.

Key words: *Parkia biglobosa*, anthelmintic, cattle, helminth eggs.

INTRODUCTION

Helminthosis is a disease of animals caused by gastrointestinal nematodes and has been recognized as constituting a major constraint to profitable production of ruminants in Nigeria (Chiejina, 2001). In recent times, some strains of parasites developed resistance to the commonly available anthelmintics (Rolfe, 1997) and this has led to screening of plants for possible anthemintic activities. Helminthosis in ruminants is caused by different nematode worms such as *Haemonchus contortus*, *Trichostrongylus colubriformis*, *Ostertagia circumcincta*, *Oesophagostomum, Bunostomum* and *Cooperia spp* (Soulsby, 1982). Conventional drugs have become expensive and therefore unaffordable to poor and low income farmers and this has led to the use of other cheaper alternatives such as medicinal plants (Maphosa and Masika, 2010). The use of indigenous plant preparations for deworming livestock is becoming popular as one of the alternative and sustainable methods readily adaptable to rural farming communities (Dano and Bogh, 1999).

Research on medicinal plants is one of the leading areas of research globally (Soetan and Aiyelaagbe, 2009). The use of herbal products for medicinal purposes has played an important role in nearly every culture on earth, and knowledge of plants, herbs and spices and their respective and collective roles in promoting health is increasing. If the safety and efficacy of these medicinal plants could be ascertained, they could be an alternative and effectively cheaper approach to the control of helminthosis in animals. Soetan and Aiyelaagbe (2009) reviewed the need for bioactivity-safety evaluation and conservation of medicinal plants. *Parkia biglobosa* is a multipurpose tree of West Africa and is used for nutritional and medicinal purposes (Sabiti and Cobbina, 1992; Mertz et al., 2001). Ibrahim and Nwude (1983) also reported the use of garlic (*Allium sativum*) as a vermifuge, used to inhibit the activity of *Ascarida galli* in chickens and also the hatching of the eggs of *Necator americanus.* Elgamal et al. (1999) reported the anthelmintic effect of saponins extracted from *Zygophylum* species used in traditional medicine. Lasisi et al. (2003) reported the inhibitory effects of condensed saponins on hatching of eggs of bovine gastrointestinal nematodes *in vitro*. Adedapo et al. (2007) also reported the anthelmintic efficacy of the aqueous crude extract of *Vernonia amygdalina*, which significantly reduced the faecal egg count of *Toxocara canis* (ascarids) and *Ancylostoma caninum* (hookworm).

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Nweze and Ngongeh (2007) reported the *in vitro* anthelmintic activity of *Anthocleista djalonensis* root ethanolic extract against the infective larvae stage (L3) of *Heligmosomoides polygyrus*, a parasitic nematode of mice. Other studies have reported the anthelmintic effects of different plant extracts against different helminthes of animals (Bizimenyera et al., 2006; Al-Shaibani et al., 2008; Maphosa and Masika, 2010; Maphosa et al., 2010). There is an increasing desire to reduce the resistance to anthelmintics and this has led to investigation into the possible use of non-chemical means for the control of the nematodes which focuses on preventing the build up of infective larvae on pasture rather than treating the infection (Soetan and Lasisi, 2008). Several reports have been given on the medicinal uses of *P. biglobosa*. Agunu et al. (2005) reported its anti-diarrhoeal properties in mice. Kouadio et al. (2000) showed that the hexane extract from the bark of *P. biglobosa* had some analgesic and anti-inflammatory effects.

Asuzu and Harvey (2003) reported that the methanolic extract of *P. biglobosa* shows significant protection against the neurotoxic and cytotoxic effects of venoms of poisonous snakes. Gronhaug et al. (2008) conducted a survey which reported that *P. biglobosa* is used as a remedy to treat different types of wounds and pain from fungal infection and that the stem bark was used most frequently (56.8%) followed by the leaves of the plant (21.6%). This study was necessitated by the fact that literatures are scarce on the use of *P. biglobosa* as an anthelmintic agent and also that the seed is not usually used for medicinal purposes. Therefore, this study was carried out to investigate the anthelmintic effects of the seeds and leaves of the African locust bean (*P. biglobosa*).

MATERIALS AND METHODS

Fresh seeds and leaves of *P. biglobosa* were collected from Ajibode in Ibadan, a human settlement in Akinyele Local Government area of Oyo State, located South West of Nigeria, and about 2 km from the University of Ibadan, Ibadan, Oyo State, Nigeria and were identified at the herbarium of the Department of Botany and Microbiology, University of Ibadan, Nigeria. The seeds and leaves were sun dried and later ground into powder form.

Aqueous extraction of *P. biglobosa* seeds and leaves

200 g of the ground seeds of *P. biglobosa* was weighed using a sensitive weighing balance and was poured inside a bowl, then 2 L of distilled water was added and stirred immediately. Stirring was done every 30 min and after 20 h, the supernatant was filtered and stored in capped bottle and preserved inside the refrigerator at 4°C. The same procedure was done for the leaves of *P. biglobosa*.

Phytochemical analyses of *P. biglobosa* seeds and leaves

The phytochemical analysis was done according to the method of Trease and Evans (1989).

Screening of helminth eggs in faecal samples

Faecal samples were collected directly from the rectum of 10 different cattle and were screened for the presence of helminthes by the modified McMaster technique (Roespstorff and Nansen, 1998). Faecal samples with the highest number of eggs per gram (800 epg) were selected. Some of the bovine nematode eggs encountered were *Haemonchus*, *Trichostrongylus*, *Oesophagostomum* and *Bunostomum* species. 1 g of each of the faecal samples was placed into 9 Petri dishes. To these were added different volumes of the seeds and leaf extracts of *P. biglobosa*. The different volumes used were 0.3, 0.7, 1.2, 2.0 ml and to the 9th Petri dish which served as the control, was added 5.0 ml of distilled water. The faecal culture and the extracts were left for 7 days inside the Petri dishes at room temperature, and examined for the presence or absence of larvae using a light microscope at X40 magnification.

RESULTS

Phytochemical screening of the seeds of *P. biglobosa* shows presence of alkaloids, cardenolides and saponins while the leaves of *P. bloglobosa* shows presence of alkaloids, cardenolides, saponins and tannins. The 4 Petri dishes containing the extract of the leaves of *P. biglobosa* all recorded deaths of mature larvae of the helminthes at the different volumes used while only 2 of the Petri dishes containing the extract of the seeds of *P. biglobosa* recorded the deaths of mature larvae of the helminthes at higher volumes of 1.2 and 2.0 ml (Table 1).

DISCUSSION

The results show that almost all the volumes of the seed extract of *P. biglobosa* inhibited the hatching of the bovine nematode eggs (+++). This observation is quite similar to the earlier study of Soetan and Lasisi (2008), in which Pearl millet (*Pennisetum glaucum*) saponin extract was shown to inhibit bovine nematode eggs *in vitro* at concentrations of 25 to 500 µg/ml. Bizimenyera et al. (2006) also reported the *in vitro* activity of *Peltophorum africanum* Sond (Fabaceae) extracts on the egg hatching and larval development of the parasitic nematode (*T. colubriformis*). The results show that the seed extract of *P. biglobosa* has more anthelmintic potential than the leave extract of *P. biglobosa*. These anthelmintic effects could probably be due to the presence of saponins. The seed extracts of *P. biglobosa* exhibited maximum inhibition of the bovine helminth eggs (+++) at 0.3, 0.7 and 1.2 ml, while the leave extract exhibited maximum inhibition of the bovine helminth eggs (+++) at volumes of 0.3 and 2.0 ml (Table 1). The presence of tannins in the leaves of *P. biglobosa* could also be responsible for the seeming anthelmintic effects. Molan et al. (2000a, b, 2002) reported the inhibitory effects of tannins against gastrointestinal nematodes and deer lungworms.

Table 1. Comparative inhibitory effects of different volumes of seed and leave extracts of *P. biglobosa* on bovine nematode eggs.

Seed extract (0.3 ml)	Leave extract (0.3 ml)	Seed extract (0.7 ml)	Leave extract (0.7 ml)	Seed extract (1.2 ml)	Leave extract (1.2 ml)	Seed extract (2.0 ml)	Leave extract (2.0 ml)
E +++	E +++	E +++	E -	E +++	E -	E -	E +++
L -	L +++	L -	L +++	L +	L +++	L +	L +++

E +++: Presence of several helminth eggs; L +++: presence of several helminth larvae; L +: presence of one helminth larva; E - : absence of helminth eggs; L -: absence of helminth larva.

They reported that condensed tannins (CT) extracted from different forages have the ability to inhibit the development of *T. columbriformis* eggs at larva stage 1 (L1) to larva stage 3 (L3) and to reduce larva motility. They suggested that these CT present in forages may have the ability to break the life cycle of sheep nematodes and reduce the contamination of pasture with infective larva and this may reduce dependence on anthelmintic drugs as the main method of controlling helminthes. It was observed that at increasing level of seed extracts, the helminth eggs show inconsistent rate of hatching while at increasing level of the leaf extract, there was consistent rate of hatching of eggs into larvae.

The pharmacological basis of the treatment of helminthes possibly involves disruption of the energy processes of the helminthes. The mechanism of interference apparently occurred through reactions necessary for the generation of metabolic energy and subsequent paralysis of the parasite or neuromuscular coordination that is depression of muscular activity, which leads to paralysis of the parasite and their subsequent expulsion from the intestine (Bueding, 1969). Interference with the neuromuscular coordination in the parasite may occur by inhibiting the breakdown of excitatory neurotransmitters or by acting like excitatory neurotransmitters resulting in spastic paralysis of the parasite (Nweze and Ngongeh, 2007). The other mechanisms involve acting like an inhibitory neurotransmitters or causing hyperpolarization resulting in flaccid paralysis of the parasite (Clarence et al., 1986). The spastic or flaccid paralysis of an intestinal helminth allows for the normal peristaltic actions of the host to expel the parasite and levamisole and piperazine are examples of drugs with this mechanism of action (Clarence et al., 1986). Niezen et al. (2002) reported that the diet of the host can have a significant impact on egg hatching and the subsequent development of *T. colubriformis* larvae in the laboratory and in the field. They observed that *Dorycnium rectum*, a herbage having moderate to high concentrations of condensed tannin consistently reduced the development of *T. colubriformis* larvae.

In conclusion, the seed extracts of *P. biglobosa* has a better anthelmintic potential more than the leaf extracts and so it could be recommended to animals as an anthelmintic agent as the seed is consumed by humans. More study is required on the anthelmintic effects of *P. biglobosa in vivo* and to also evaluate its biosafety and clinical potentials. Further research is also required to isolate and structurally characterize the active anthelmintic compounds and to elucidate its mechanism of action. There is also a need to discover the cheapest methods of extraction for the effective anthelmintic components that will be easily adaptable for use by rural communities against helminthosis.

REFERENCES

Adedapo AA, Otesile AT, Soetan KO (2007). Assessment of the Anthelmintic efficacy of the aqueous crude extract of *Vernonia amygdalina*. Pharmaceut. Biol., 45(7): 564-568.

Agunu A, Yusuf S, Andrew GO, Zezi AU, Abdurahman EM (2005). Evaluation of five medicinal plants used in diarrhoea treatment in Nigeria. J. Ethnopharmacol., 101: 27-30.

Al-Shaibani IRM, Phulan MS, Arijo A (2008). Ovicidal and larvicidal properties of *Adhatoda vasica* (L) extracts against gastrointestinal nematodes of sheep *in vitro*. Pak. Vet. J., 28: 79-88.

Asuzu IU, Harvey AL (2003). The anti-snake venom activities of *Parkia biglobosa* (*Mimosaceae*) stem bark extract. Toxicon, 42: 763-768.

Bizimenyera ES, Githiori JB, Eloff JN, Swan GE (2006). *In vitro* activity of *Peltophorum africanum* Sond (Fabaceae) extracts on the egg hatching and larval development of the parasitic nematode *Trichostrongylus colubriformis*. Vet. Parasitol., 142: 336-343.

Bueding E (1969). Some biochemical effects of anthelmintic drugs. Biochem. Pharmacol., 18: 1541-1547.

Chiejina SN (2001). The epidemiology of helminthes infection of domesticated animals in the tropics with emphasis on fascioliasis and parasitic gastroenteritis, In: Chowdhury, N., Tada, I. (Ed): Perspectives on Helmintology. Science Publishers Inc., Enfield, pp. 41-87.

Clarence MF, Asa M, Huebner RA (1986). Eds. The Merck Veterinary Manual, 6th Ed. Merck and Co. Inc., New Jersey, USA, pp. 1568-1580.

Dano AR, Bogh H (1999). Use of herbal medicine against helminthes in livestock-renaissance of an old tradition. World Anim. Rev., 93: 60-67.

Elgamal MHA, Shaker KH, Pollman K, Seifert K (1999). (Author should please provide title here). Phytochemistry 40(4): 1233-1236.

Gronhaug TE, Glaeserud S, Skogsrud M, Ballo N, Bah S, Diallo D, Paulsen B (2008). Ethnopharmacological survey of six medicinal plants from Mali, West Africa. J. Ethnobiol. Ethnomed., 4(26): 1-10.

Ibrahim MA, Nwude N (1983). Screening of West African Plants for Anthelmintic Activity. Presented at the Fifth International Symposium on Medicinal Plants, University of Ife, Ife, Nigeria, 13 to 15 July, 1983, p. 19.

Kouadio F, Kanko C, Juge M, Grimaud N, Jean A, N'Guessan YT, Petit JY (2000). Analgesic and anti-inflammatory activities of an extract from *Parkia biglobosa* used in traditional medicine in the Ivory Coast. Phytother. Res., 14: 635-637.

Lasisi OT, Soetan KO, Fafunso MA, Aiyelaagbe OO (2003). Effect of condensed saponins on egg hatching of bovine gastrointestinal nematodes *in vitro*. Nig. Vet. J., 24(3): 30-34.

Maphosa V, Masika PJ (2010). Ethnoveterinary uses of medicinal plants: A survey of plants used in the ethnoveterinaary control of gastrointestinal parasites of goats in the Eastern Cape Province, South Africa. Pharmaceut. Biol., 48(6): 697-702.

Maphosa V, Masika PJ, Bizimenyera ES, Eloff JN (2010). *In vitro* anthelmintic activity of crude aqueous extracts of *Aloe ferox*, *Leonotis leonurus* and *Elephantorrhiza elephantina* against *Haemonchus contortus*. Trop. Anim. Health Prod., 42(2): 301-317.

Mertz O, Lykke AM, Reenberg A (2001). Importance and seasonality of vegetable consumption and marketing in Burkina Faso. Econ. Bot., 55(2): 276-289.

Molan AL, Duncan A, Barry TN, McNabb WC (2000a). Effects of condensed tannins and sesquiterpen lactones extracted from chicory on the viability of deer lungworm larvae. Proc. New Zealand Soc. Anim. Prod., 60: 25-29.

Molan AL, Hoskin SO, Barry TN, McNabb WC (2000b). Effect of condensed tannins extracted from four forages on the viability of the deer lungworms and gastrointestinal nematodes. Vet. Rec., 147: 44-48.

Molan AL, Waghorn GC, McNabb WC (2002). Effect of condensed tannins on egg hatching and larval development of *Trichostrongylus colubriformis in vitro*. Vet. Rec., 150: 65-69.

Niezen JH, Waghorn GC, Graham T, Carter JL, Leathwick DM (2002). The effect of diet fed to lambs on subsequent development of *Trichostrongylus colubriformis* larvae *in vitro* and on pasture. Vet. Parasitol., 105(4): 269-283.

Nweze NE, Ngongeh LA (2007). *In vitro* Anthelmintic activity of *Anthocleista djalonensis*, Niger. Vet. J., 28(1): 9-13.

Roespstorff A, Nansen P (1998). Epidemiology, diagnosis and control of helminthes parasites of swine. Food and Agricultural Organization (FAO) Animal Health Manual, No. 3, FAO, Rome, Italy.

Rolfe PF (1997). Anthelmintic resistance in Australia, its development and management. In: Proc. of the 4th Intn'l Congress for sheep veterinarians. Austr. Sheep Vet. Soc., Indooroopilly, pp. 51-58.

Sabiti EN, Cobbina J (1992). *Parkia biglobosa*: A potential multipurpose fodder tree legume in West Africa. Int. Tree Crops J., 7: 113-139.

Soetan KO, Aiyelaagbe OO (2009). The Need for Bioactivity-Safety Evaluation and Conservation of Medicinal Plants- A Review. Available online at http://www.academicjournals.org/JMPR J. Med. Plants Res., 3(5): 324-328.

Soetan KO, Lasisi OT (2008). Studies on the anthelmintic activities of saponins from Pearl millet (*Pennisetum glaucum*). Trop. Vet., 26(1): 14-19.

Soulsby EJL (1982). Helminths, Arthropods and Protozoa of Domesticated Animals, 7th Ed. Bailliere Tindall, London, pp. 212-218.

Trease GE, Evans WC (1989). A textbook of Pharmacognosy, 13th Edition, Baillere Tindall Ltd., London.