Effects of *Melastoma malabatricum* extract on nutrient digestibility of local goat infected with gastro intestinal parasites concise and informative

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Abstract The effects of Melastoma malabatricum extract were evaluated on intake and nutrient digestibility of goat infected with gastro intestinal parasites. Before the experiment all goats were dewormed with albendazole to remove gastro intestinal parasites infestation previously, then goats were naturally infected with parasites by allowing the experimental goats to graze on infected pasture under oil palm from 08.00-16.00 daily for 15 days and followed infected orally with 1000 infective larvae. After the infestation, 20 goats were stratified based on live weight and confined individually in wooden pen. Goats divided in 4 treatments namely T0: No extract T1: Aquaoes extract Melastoma malabatricum 250mg/kg BW/2 week, T2: Aquaoes extract Melastoma malabatricum 250mg/kg BW/ 3 week and T3: single dose of Ivermectine (control positive). All experimental goats received 1% feed supplementation (50% Palm Kernel Cake and 50 % rice bran), forage and water were given ad libitum. Result indicated that treatment had sigificantly effect (P <0.05) on dry matter and crude fiber intake, the dry matter intake (gDM/kgLW^{0.75}) was 44.93-49.55 or 2.69-2.91% per live weight. Feed intake of OM, Crude Protein, and Ether Extract did not differ (P>0.05) among the treatments. There were no significant differences (P>0.05) among treatments in dry matter, crude protein, crude fiber, extract ether digestibility. The organic matter digestibility was significantly higher in goat received single dose of Ivermectine compare with the others.

Keywords: Digestibility, goat, Melastoma malabathricum, parasites

Introduction

Gastrointestinal nematode infection predominantly *Haemonchus contortus* being the highest rank in health and productivty problem in ruminant (Waller and Chandrawathani, 2005; Besier *et al.*, 2016; Dutta *et al.*, 2017; Selemon, 2018). Adult *Haemonchus contortus* colonize or nested in the abomasal mucosa of the goat and feed on their blood from capillaries 0.05 ml blood per day. (Wang *et al.*, 2017; Hoberg and Zarlenga, 2016). Dutta *et al.* (2017) observed gross lesions include generalized oedema, anaemia, and presence of fluid in the

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body cavities. Fluid also found in the abomasal and sometimes mixed with free blood with large number of adult *Haemonchus contortus* parasites causing ulcerative haemorrhagic spots in the abomasal mucosa.

Taylor *et al.* (2007) described the lifecycle of *Haemonchus contortus*. Its begin by ingestion of the L3, exsheat in the rumen then penetrates to the mucosa of the abomasum where the transformation into the fourth larval stage (L4) takes place. The final moult into adult stage takes place 10-15 days later and upon reemergence of the parasite into the abomasal lumen. An adult worm could produced up to 5000-10.000 eggs then secreted in the feces, hatch, and are ingested by the goats through the consumption of grasses – especially those that are short and/or covered in dew (Machen *et al.*1998). Therefore the incidence of re-infection is high in grazing goats.

Untreated Haemonchosis, can lead to protein deficiency, anemia, bottle jaw, and death in infected animal (Suteky and Dwatmadji, 2015, 2016). According to Soulsby (1982), the cause of death in Haemonchosis is anaemia accompanied by hypoproteinaemia and oedema. Damages caused by *Haemonchus contortus* can lead to billions of economic losses to the livestock production or breeding industry (Roeber *et al.*, 2013; Emery *et al.*, 2016; McLeod, 2004) especially for young animals. Economic losses are due to the high cost of preventive as well as curative treatment (Miller and Horohov, 2006).

The use of pharmaceutically drugs, such as albendazole, benzimidazole, ivermectin etc is the common method to control of *Haemonchus*. However, the current efficacy of these drugs is becoming less effective due to the development of anthelmintic resistance against most commonly used anthelmintics (Saddiqi *et al.*, 2010; Bowie, 2014; Waller *et al.*, 2004). The existence of anthelmintic resistance in many countries, including United States, Brazil, South Africa, Australia, New Zealand, and European countries have been reviewed by Kaplan and Vidyashankar (2012). Machen *et al.* (1998) describe the actions that cause increased anthelmintic resistance: frequent dosing, underdosing to save money, inappropriate administration, wrong anthelmintic choices, and massive re-exposure to the parasites.

The development of natural plants as anthelmintic is being considered in developing countries, many researcher heve been exploring alternative options for natural dewormers, because of these practices are more accessible, easy to prepare and administer, less toxic, inexpensive and less side effect (Wabo Pon é et al., 2010; Ngeh et al., 2007; Bowie, 2014).

Research on developing natural plant as an anthelmintic can be promoted as alternative choices because farmer can used their own resources for the prevention and treatment of livestock diseases (Lem *et al.*, 2014). In their study

Lem *et al.* (2014) found that *in vitro* Hot water extract (HWE), Methanol (MET) and Cold water extract (CWE) of stem bark of *Terminalia glaucescens* inhibited embryonic development by 98.1%, 96% and 86.5% respectively at 5000 μg/ml, compare to Albendazole had a 77.9% inhibition. Several plant reported have capability as ovicidal or larvicidal activity such as water extract of *Manihot esculenta* (Al-Rofaai *et al.*, 2010) *Rhus glutinosa* (Alemu *et al.*, 2014), *Azadirachta indica* (Tomar and Preet (2017).

The used of *Melastoma malabatricum* extract as an alternative anthelmintic was reported by Suteky and Dwatmadji (2011, 2015, 2016). Our findings showed that the crude aqueous and ethanolic extracts of *Melastoma malabatricum* extract have ability as ovicidal activity 84.44% and 86.05% respectively, our study also found that ethanolic extract showed larvicidal effect (79.08%) at concentration 50mg/ml, which is not significantly (P<0.05) different with positive control (Oxfendazole 40 mg/ml) (Suteky and Dwatmadji, 2011). Crude aqueous extract of *Melastoma malabatricum* could decreased Fecal Egg Counts and reduce mortality rate in infected goats (Suteky and Dwatmadji, 2015, 2016).

As *Haemonchus contortus* colonized in abomasum, therefore this parasites will reduce the utilization of absorbed nutrients (Dynes *et al.*, 1998). Preston and Leng (1987) said that infection of parasites affects the absorption of amino acids from the small intestine, and consequently will decrease voluntary feed intake. A reduction in voluntary feed intake of up to 50 % is commonly observed during infection with gastro intestinal nematodes (Sykes and Greer, 2003).

The objectives were to evaluate the effects of *Melastoma malabatricum* extract on feed intake and nutrient digestibility of goat infected with gastro intestinal parasites.

Materials and methods

The Study Site

The study was performed in 2015-2016 at the Small Oil Palm plantationt in Central Bengkulu, Bengkulu Indonesia for the natural infection. Experimental research was done in Animal Science Out Door Laboratory Faculty of Agriculture University of Bengkulu, and proximate analysis conducted in Integrated Laboratory Bogor Agriculture University, Bogor Indonesia.

Plant Extract preparation

The leaves of *Melastoma malabathricum* were harvested from their natural habitat around University of Bengkulu. The leaves were dried at room temperature for 5-7 days and milled to powder using an electrical blender and stored in air-tight plastic bags at room temperature for further use in the laboratory. Aquaous extract of *Melastoma malabathricum* were prepared according to Suteky and Dwatmadji (2011).

Experimental Animal

Twenty local female goats of 6-8 months, with initial BW about 8 kg, were drenched with Albendazole within 2 weeks before the natural infection with nematode dominated by Haemonchus contortus. The experimental goats were subjected to graze on contaminated pastures from 08.00-16.00 for 15 days, following grazing animals were kept in a shed at night. The goats also infected orally with 1000 infective larvae. After the infestation, 20 goats were stratified based on live weight and were confined individually in wooden pen over period of 14 days for diet adaptation, 14 days treatments and 7 days for data collection. All goats then were devided in 4 treatments consisting 5 goats for replication and kept under same condition. The treatments were as follows: T0: No extract T1: Aquaoes extract Melastoma malabatricum 250mg/kg BW/3 week, T2: Aquaoes extract Melastoma malabatricum 250mg/kg BW/2 week and T3: single dose of Ivermectine (control positive). During the experiment the health status of goats were observed.

Experimental Design

The experiment was designed as Randomized complete Block Design with four treatments and or replications.

Sampling

The concentrate (1%/BW) contained 50% rice brand and 50% Palm cernel cake and forage was weighed using electronic scale and offered ad libitum at 08:00 in the morning. The amount of forage offered about 10% greater than feed intake during the animals' adaptation period. The refused feed were collected and weighed before morning feeding. In the same time, feces were also collected every 24 hours for 7 days, harness equipped was used for fecal collection to prevent urine mix with feces. Approximately 10% of sample

feces were dried and the composite were made per goat and ground for later analysis. Analyses of dry matter (DM), organic matter (OM), crude protein (CP), Ether Extract and Crude Fibrous were set using the methods recommended by AOAC (1990), these analyses were performed in integrated laboratory, Bogor Agricultural University.

Statistical Analysis.

All data were analysed using ANOVA. Treatment means were compared by Duncant Multiple Range Test (DMRT), and significance was declared at 5% probability level. All statistical analyses were performed by SPSS for Window 16.

Results

Chemical composition of the feeds

Data on chemical composition of the feeds are presented in Table 1. Forage used in this experiment had lower crude protein content and much higher organic matter and crude fiber than those of the chemical composition in concentrate. Inadequate protein content could not meet the daily requirment. Feed supplementation therefore is required for goats to perform without serious health problems. However, during adaption and experimental period all goats refuse to consume the feed supplementation (concentrate). It was also found that during the collection period 4 goats found died in each group due to the infestation of gastro intestinal parasites.

Table 1. Nutrient composition of the feeds

	Forage	Concentrate	
Dry matter	20.49	86.88	
% DM basis			
Organic Matter	11.90	3.84	
Crude Protein	8.01	10.79	
Ether Extract	1.25	5.05	
Crude Fiber	31.12	16.59	

Feed Intake and Digestibility

The mean values for fresh and dry matter intake are shown in Table 2. Result showed that there was significantly differences (P<0.05) on fresh and dry matter intake between the treatments. Either fresh and dry matter intake in T3 (single dose of Ivermectine (control positive) differ significantly with T0 (control negative) and T 2 (Aquaoes extract *Melastoma malabatricum* 250mg/kg BW/3 week. Data on Table 2 clearly indicated that percentage of dry matter intake based on LW were almost similar (P>0.05).

Table 2. Mean values for intake of fresh forage, dry matter, %LW, faecal production and digestibility (%) of the feeds

Dry Matter	Т0	T1	T2	Т3	P
Feed intake					
Fresh Forage (g·d ⁻¹)	1055.57 ^a	1153.58 ab	1095.00 ^a	1244.70 ^b	< 0.05
Dry Matter (g·d ⁻¹)	211.96 ^a	229.12 ^{ab}	217.13 ^a	244.79 ^b	< 0.05
Intake (% LW)	2.69	2.89	2.67	2.91	>0.05
Intake (g/kg LW ^{0.75}					
/day)	44.93	48.41	45.09	49.55	>0.05
E 1 D 1 2 (1 ⁻ 1)	37.03	37.50	36.64	32.76	0.05
Faecal Production (g·d ⁻¹)	37.03	37.30	30.04	32.70	>0.05
Digestibility (%)	82.42	83.57	82.96	86.58	>0.05

T0: No extract T1: Aquaoes extract *Melastoma malabatricum* 250mg/kg BW/2 week, T2: Aquaoes extract *Melastoma malabatricum* 250mg/kg BW/3 week and T3: single dose of Ivermectine (control positive). a-b Means within a row with different superscripts differ (p < 0.05).

The intake values of feed were also done based on dry matter intake per unit metabolic weight to eliminated the effect of differences in live weight and body size. The metabolic weight is the 0.75 power of live weight in kilo grams. Result showed that the mean voluntary intake is not significantly different (P>0.05) among the treatments is about 44.93-49.55 (g/kg LW^{0.75}/day).

Table 2 indicated that there is no effect (P>0.05) of treatment on faecal excretion (32.76-37.50) gram a day and also the apparent digestibility of dry matter during the experimental period (82.42-86.58%).

Table 3. Mean values for intake of fresh forage, faecal production and digestibility (%) of organic matter, crude protein, ether extract and crude fiber

Organic Matter	Т0	T1	T2	Т3	P
Intake $(g \cdot d^{-1})$	15.97 ^a	17.28 ^a	16.33 ^a	17.64 ^a	>0.05
Faecal Production (g·d ⁻¹)	5.43 ^{ab}	6.11 ^b	5.20 ^{ab}	2.66 ^a	< 0.05
Digestibility (%)	63.48 ^a	62.22 ^a	66.67 ^a	85.17 ^b	< 0.05
Crude Protein					
Intake $(g \cdot d^{-1})$	17.05 ^a	18.01 ^a	17.53 ^a	20.02 ^b	< 0.05
Faecal Production $(g \cdot d^{-1})$	4.31	4.35	3.93	3.69	>0.05
Digestibility (%)	74.5	75.64	77.37	81.47	>0.05
Ether Extract					
Intake $(g \cdot d^{-1})$	2.48	2.52	2.55	2.63	>0.05
Faecal Production $(g \cdot d^{-1})$	0.81	0.85	0.75	0.81	>0.05
Digestibility (%)	66.30	66.37	68.45	69.10	>0.05
Crude Fiber					
Intake $(g \cdot d^{-1})$	65.79 ^a	71.87^{ab}	67.59 ^a	75.89 ^b	< 0.05
Faecal Production	8.22	8.46	8.08	7.85	
$(g \cdot d^{-1})$					>0.05
Digestibility (%)	87.43	88.16	87.4	89.64	>0.05

T0: No extract T1: Aquaoes extract *Melastoma malabatricum* 250mg/kg BW/2 week, T2: Aquaoes extract *Melastoma malabatricum* 250mg/kg BW/3 week and T3: single dose of Ivermectine (control positive). a-b Means within a row with different superscripts differ (p < 0.05).

The mean values for intake of organic matter, crude protein, ether extract and crude fiber are shown in Table 3. It seems that organic matter and ether extract intake did not differ between treatments, organic matter intake was not affected by extract *Melastoma malabatricum* or Ivermectin. Single injection with Ivermectine (group of control positive) causing higher significantly (P<0.05) for crude protein and fiber intake.

Faecal production of did not differ significantly (P>0.05) except for faecal priduction in organis matter. The percentage of apparent organic matter digestibility was significantly different (P<0.05) range from 62.22-86.17%. Tabel 3 shown that no significance effect for digestibility of crude protein, ether extract and crude fiber.

Discussion

During collection day, one goat in each group found death due to Helminthiasis. Animals infected with parasite especially *Haemonchus contortus* can show a series of symptoms, including anemia, emaciation, diarrhea or even death under heavy burden (Bowman, 2009, Roeber *et al.*, 2013; Emery *et al.*, 2016; Suteky and Dwatmadji, 2015, 2016). Sutherland and Scott, 2010), reported that worm burdens can range from 2,000 to 20,000 worms, as a result blood losses can be considerable. In our previous research Suteky and Dwatmadji (2011) found that goats graze in infected pasture the mortality rate could reach 67% starting 3 week post infection. In high level of parasite infestation, the mortality rate as high as 80% was recorded in goats either treated with aquaous extract *Melastoma malabatricum* or single dose ivermectin. It seem that ivermectine could not always decrease the mortality rate due to helminthiosis, and the degree of goats response depent on the level of infestation.

In others experiment, Suteky and Dwatmadji (2015) found that no mortality rate of goats with low-moderate infestation of *Hamemonchus contortus*, goats were suplemented with combination palm kernel cake, rice brand and cassava leaf meal. Acording to Zaralis *et al.* (2008) the manifestation of gastrointestinal parasitism infestation is dependent on immunity stage, physiological condition, time of infection and nutritional status. Our findings showed that crude protein on the forage is 8.1%, considerably similar those reported by other workers. They reported that crude protein of tropical grasses in different part of the world ranges from 2-27 % of dry matter (Minson, 1990). During the adaptation and experimental period all goats were refuse concentrate (PKC and rice brand), this result is not in line with the finding by Chanjula *et al.* (2011) PKC very palatable for goats.

Bambou *et al.* (2009) stated that only a few published researches are documented about the effects of gastrointestinal parasitism on feed intake and digestibility. In this experiment result indicated that treatment had sigificantly effect (P <0.05) on dry matter, crude protein and crude fiber intake. The mean DM and crude fiber intakes were found to be similar (P >0.05) between goats treated with aquaoes extract *Melastoma malabatricum* 250mg/kg BW/3 week and ivermectine, indicating that thus extract 250mg/kg BW/3 week had similat effect with ivermectine on DM and crude fiber intake of growing goats.

The coefficient digestibility of DM was numerically higher in T3 (control positive) compared with others. According to Zhang *et al.* (2010) cited by Xu *et al.* (2017) DMI was an important index in ruminant nutrition, which was affected by several factors such as dietary nutrient levels, live-weight,

health condition. As a control positive T3 have less faecal egg counts than other treatments. Apparent digestibility coefficient of organic matter in T3 (control positive) higher significantly (P<0.05) than other treatments in the range of 62.22-85.17%. This result is better than finding reported by Romero *et al.* (2018) in vivo apparent digestibility of organic matter on goat infected with parasites around 52.1-60.8% depending on the type of feed supplementation.

The crude protein (CP) intake in group of goat given ivermectine (T3) were significantly higher (P<0.05) than the other treatments. Apparent digestibility of CP not significantly (P>0.05) different but numerically higher in ivermectine groups. Preston and Leng (1987) strongly emphasized that CP intake important determinat on liverstock performance. In this experiment effect of parasites more severe in groups of control negative (T0).

The mean value of EE intake and digestibility can be seen in Table 3, eventhought no significantly different it seem that increasing EE intake coincide with the increasing EE digestibility. Luginbuhl *et al.* (2000) reported an increased EE digestibility match with increased EE consumtion in goats. The intake of crude fiber in T3 differ significantly (P<0.05) with T0 and T2 but did not different with T1. It means that the used of aquaeous extract Melastoma given every 2 weeks have similar effect with positive control. According to Lu *et al.* (2005) dietary fibre play an important role to balancing of nutrient requirement and goat production throught the combination with the intake and digestion of nutrients.

The used of *Melastoma malabatricum* extract as a natural anthelmintic for goat infected with gastrointestinal parasite showed similar effect with positive control (single dose ivermectine) in dry matter intake and digestibility of dry matter, crude protein, EE and crude fiber. Futher research should determine the optimal doses and frequency of *Melastoma malabatricum* extract and the development of consumable feed supplementation to reduce mortality rate of the animals.

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