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RESEARCH ARTICLE

Exploration and prevalence of gastrointestinal worm in buffalo from West Java, Central Java, East Java and Lombok, Indonesia

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ABSTRACT

The studies of parasites in buffaloes have not been widely explored. The aim of the present study was to explore the prevalence of gastrointestinal worm infection in buffaloes. The fresh faecal samples were collected from 89 buffaloes and observed by a modified McMaster technique. The faecal of buffaloes were collected in Bogor, Demak, East Java, and Lombok. The results of identification on gastrointestinal parasites show that there were one cestode and eight nematode. The total prevalence and infestation of cestodes and nematodes was found highest in Bogor. The prevalence and infestation of differences in geographical conditions were found highest in Bogor. The prevalence of gastrointestinal worm in males were highest than female, whereas for larger was found in females. The prevalence of gastrointestinal worms was found at age <1 year, whereas a larger infestation was found at the age of 1-5 years. The calculation of FEC in West Java was 840 EPG, in Central Java 375 EPG, in East Java 570 EPG and in Lombok 13 EPG. This study informed that there were six genera of gastrointestinal worm found in West Java and Central Java, eight genera in East Java and five genera in Lombok. The totality sample identification of faecal to culture technique used water medium were found six genera i.e *Strongyloides, Haemonchus, Cooperia, Oesophagostomum, Bunostomum* and free living larvae.

Keywords: Buffalo, explorations, gastrointestinal worm

INTRODUCTION

Gastrointestinal worm lives in the digestive system in a larger organism/host (Shah, 2012). There are parasites and non parasites of gastrointestinal worm. The huge losses of the host can be caused by parasitic worms, due to the decrease in productivity and growth disorders, digestive disorders, anemia, even the death (Zajac and Conboy, 2011; Sahinduran, 2012). Gastrointestinal worms that found in the digestive of ruminants were *Trichuris* and *Oesophagostomum* in colon, *Trichostrongylus*, *Cooperia*, *Nematodirus,* Bunostomum, *Strongyloides, Moniezia, Eimeria* and *Cryptosporodium* in small intestine, *Paramphistomum* in rumen, *Haemonchus, Ostertagia*, and *Trichostrongylus* in abomasum (Foreyt, 2001). Whereas gastrointestinal worms that found in buffalo are *Trichostrongylus*, *Oesophagostomum*, *Haemonchous*, *Toxocara*, *Ostertagia*, *Bunostomum*, *Strongyloides*, and *Trichuris* (Afridi *et al*., 2007).

Gastrointestinal worms infestation in ruminant can be affected by geographical condition of the area. The differences of worms and the prevalence of each location will be different due to the habitat and environmental condition. Some areas in Java and other areas have different environmental condition. Areas that have environmental condition that are different with other areas was Bogor district. Bogor is one of the areas in West java which has high rainfall. The highest rainfall will influence the gastrointestinal worm infestation. Gastrointestinal worm infestation in ruminant will be highest in rainy season than summer (Chavhan *et al.,* 2008; Patel *et al*., 2015). In addition to the environmental factor that influenced the gastrointestinal worm, livestock management, differences age and on sex are important factor for gastrointestinal worm infestation (Soulsby, 1982; Levine, 1994).

Gastrointestinal worms are specific host and not specific (live in more one host). Gastrointestinal worm i.e *Toxocara* sp. and *Oesophagostomum* sp. can be found in cattle and buffalo, *Haemonchus* were found in sheep and goats (Raza *et al.,* 2007), cattle and buffalo (Bilal *et al*., 2009; Choubisa and Jaroli, 2012). In the body of the host, the egg of gastrointestinal worm released along with the faecal. Faecal is breeding gastrointestinal worm eggs into larvae. Number of worm eggs in each gram of faecal can be used to determine the level of worm infestation. Identified the worms

based on the morphology of eggs were difficult to recognize the genera, becaused the size overlapping, shape and character are similar. Eggs identified using the relationship between character size (length and width), the shape and characteristic of cell (Foreyt, 2001; Zajac and Conboy, 2011). Identification of the worm larvae can strengthened the introduction for the types of worm up to genera until species. This approach was done by description of head shape, tail, oesophagus types and number cell intestine (Wyk and Mayhew, 2013).

Some research on gastrointestinal worms in buffalo have been conducted in Italy (Rinaldi *et al.,* 2009), Bangladesh (Mamun *et al.,* 2011), India (Choubisa and Jaroli, 2012; Singh *et al.,* 2012) and Pakistan (Bilal *et al.*, 2009; Raza *et al.,* 2013). In Indonesia, the study of gastrointestinal worm has not been widely explored. This is a problem to identification the types of gastrointestinal worm in faecal of buffalo.

MATERIAL AND METHODS

This study was conducted from November 2013 to July 2014. The faecal of buffaloes were collected in several areas, in Bogor West Java Province (Darmaga, Ciampea, and Tenjolaya), in Demak Central Java (Mranggen), East Java Province (Tulungagung, Kepanjen, and Lamongan). Whereas from Lombok at Lamper village. Samples were analyzed at the Laboratory of physiology and behaviour, Departement of Biology, Bogor Agricultural University.

Site selection

The location was selected based on geographical condition. West Java was chosen to represent the location with moist/damp condition. Whereas the dry location represented by Central Java, East Java, and Lombok/NTB.

Sampling buffalo faecal

The faecal of 27 buffaloes were collected in several areas from Bogor, 15 buffaloes from Demak, 37 buffaloes from East Java Province and 10 buffaloes from Lombok. The buffaloes categories based on Raza *et al.*, (2013) (< 1 years i.e 15 buffaloes, 1 – 5 years i.e 35 buffaloes, and > 5 years i.e 39 buffaloes) recorded based on the farmer. The age categories of buffaloes were collected in < 1 years (The faecal of buffalo was taken on the soil in fresh condition, with characteristic damp and green. Faecal were collected with plastic bags and stored in container for identification.

Observation worms eggs and Identification of worms

Worm eggs were calculated by using floatation solution technique modified by Southwell *et al.*, (2008). Eggs in the faecal will be floating in salt solution, becaused the density of eggs were lowest than the flotation solution. Two grams faecal were mixed into 58 ml of saturated NaCl solution, and then stirred to homogeneous. The floating eggs on the top layer were taken with pipetted and drop into a slide counter worm eggs (Mc Master). The worm eggs were observed under the microscope with 100X magnification. Identification of gastrointestinal worm eggs based on size characters i.e length and width, eggs shape, characteristic cell in eggs (Figure1) (Foreyt, 2001; Zajac and Conboy, 2011).

Culture and identification of gastrointestinal worm larvae

The water medium culture technique was used in this study. Faecal put into water (Hutchinson, 2009). The small glass (15 ml) was put into the medium glass (35 ml) and then put it in the larger glass (80 ml). These containers are placed in dark. Larvae were harvested on the second days until the seven days (Hutchinson, 2009). Identification of gastrointestinal worm larvae based on the head shape, tail, number intestine cell and esophagus types (Wyk and Mayhew, 2013).

Environmental parameters

The environmental parameters include rainfall, temperature and humidity were obtained from the Meteorology, Climatology and Geophysics (BMKG) Agency. The altitude of this location was measured using the Global Positioning System (GPS).

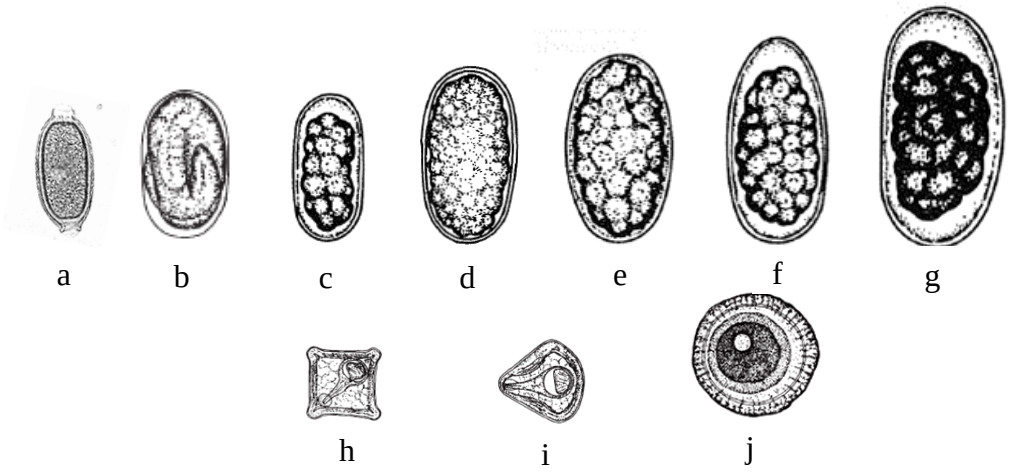

Figure 1. *Capillaria* eggs 45-50x22-30 μm (a), *Strongyloides* eggs 40-60 x 25-40 μm (b), *Cooperia* eggs 77x34 μm (c), *Oesophagostomum* eggs 80x40 μm (d), *Haemonchus* eggs 80x45 μm (e), *Trichostrongylus* eggs 85 x 40 μm (f), *Bunostomum* eggs 95 x 50 μm (g), *Moniezia* eggs (h and i) 60-75 μm in diameter and *Toxocara* eggs 75-95x60-80 μm (j).

Data analysis

The measurement morphometric of eggs was using image J program. The prevalence and infestation of gastrointestinal worm that observed were livestock management and environmental factors. Differences in geographic location, sex and age were analyzed using prevalence method (Rehman *et al.,* 2011). The calculation of faecal Eggs Count (FEC) by (Cringoli *et al.,* 2004; Zajac and Conboy (2011) as follow:

$$\text{Prevalence (\%)} = \frac{\text{Number of infected individual } (n)}{\text{Total number of sample individuals } (N)} \times 100$$

$$\text{FEC} = \text{no. eggs} \times \left(\frac{d}{v} \text{ / 2 g faecal} \right)$$

Where, d = total volume of flotation solution (60 ml), v = volume of solution examined in McMaster (one chamber 0.5 ml x 4 = 2 ml solution total)

RESULTS AND DISCUSSION

Identification gastrointestinal worm eggs

The eggs of gastrointestinal worm that found in this study consist of five orders, six families and nine genera (Table 1). The identification and description of the eggs and larvae of gastrointestinal worm had different shape and morphological. The result showed that the egg of *Moniezia* have diameter ranged between 28.63 μm to 60.79 μm and triangular (Figure 2). *Strongyloides* eggs have length 45.61 - 65.03 μm (r = 50.82 μm); width between 19.76 - 34.21 μm (r = 27.60 μm). The result showed that *Strongyloides* eggs to meaty of larvae and the cell walls are thin (Figure 2).

This result showed that *Capillaria* had two poles the end of egg. *Capillaria* had length 48.05 - 51.26 μm (r = 49.46 μm); width between 21.78 - 23.74 μm (r = 22.76 μm). *Capillaria* eggs elongated shape, yellowish egg cells (Figure 3). *Haemonchus* eggs had length 61.98 - 85.01 μm (r = 68.87 μm); width between 35.61 - 54.29 μm (r = 42.02 μm). *Haemonchus* eggs had oval shape, yellowish cell, three layers eggshell and containing fully developed embryonic cells (Figure 3). *Cooperia* eggs had length 68.11 - 85.37 μm (r = 76.75 μm); width between was 32.68 - 39.81 μm (r = 36.67 μm). Both ends rounded egg, the egg parallel on side and egg cells dark brown (Figure 4). *Trichostrongylus* egg

had length 70.03 - 85.55 µm (r = 81.82 µm); width between 36.37 - 47.23 µm (r = 44.20 µm). *Trichostrongylus* eggs had tapered and blunt end, the outside *Trichostrongylus* have three layers eggshell, yellowish cell and not containing fully developed embryonic cells (Figure 4).

Toxocara eggs had rounded shape, dark yellow egg and thick-walled. The eggs length between 72.55 - 75.43 µm (r = 73.99 µm); width between 67.52 - 69.54 µm (r = 68.53 µm) (Figure 4). *Oesophagostomum* eggs had length 67.52 - 85.60 µm (r = 76.56 µm); width between 35.31 - 48.23 µm (r = 41.77 µm). *Oesophagostomum* eggs had rounded tip, thin walls, dark yellow egg and three layers eggshell. The eggs not containing fully developed embryonic cells (Figure 4). *Bunostomum* eggs length between 87.03 - 128.12 µm (r = 103.45 µm); width ranged between 43.97 - 68.79 µm (r = 54.45 µm). The Shape of eggs blunt rounded, blackish brown egg, and not containing fully developed embryonic cells (Figure 5).

Table 1. Classification of Gastrointestinal Worm

Platyhelminthes				
Class	Subclass	Ordo	Family	Genus
Cestode		Cyclophyllida	Anoplocephalidea	*Moniezia*
Nemathelminthes				
Class	Ordo	Family	Super Family	Genus
Nematode	Strongylida	Trichostrongyloidea	Trichostrongylidae	*Haemonchus*
				Bunostomum
				Cooperia
				Trichostronglus
	Rhabditida	Rhabditoidea	Strongyloididae	*Strongyloides*
		Strongyloidea	Trichonematidae	*Oesophagostomum*
	Enoplida	Trichuroidea	Trichuridae	*Capillaria*
	Ascaridida	Ascaridoidea	Ascarididae	*Toxocara*

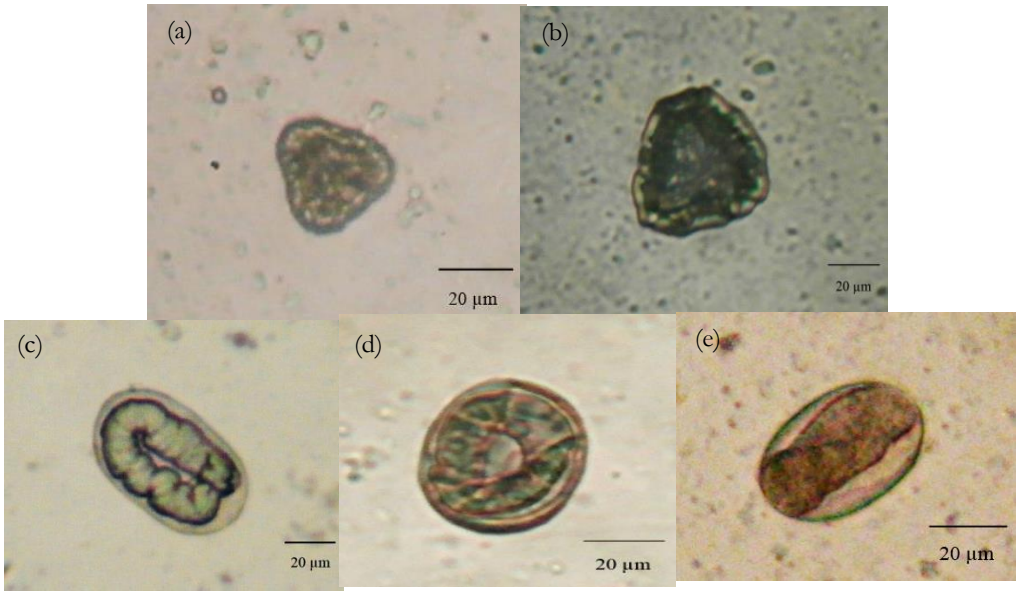

Figure 2. (a) *Moniezia* eggs 28.63 µm in diameter, (b) *Moniezia* eggs 46.32 µm in diameter (c) *Strongyloides* eggs 28.82x46.15 µm, (d) *Strongyloides* eggs 46.88x34.13 µm and (e) *Strongyloides* eggs 46.88x34.13 µm

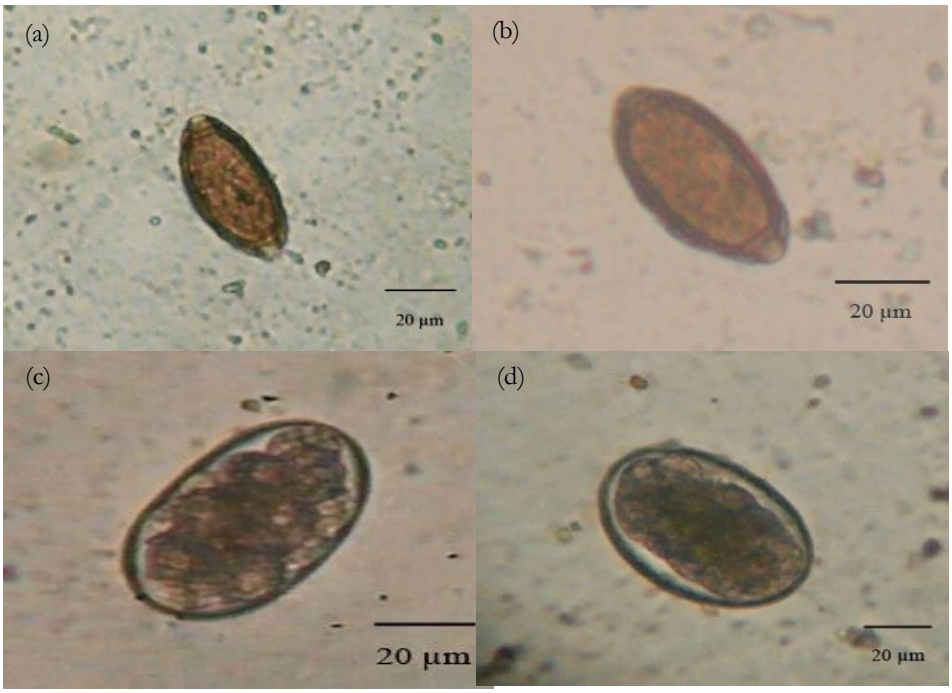

Figure 3. (a)*Capillaria* eggs 48.05x21.78 µm, (b) *Capillaria* eggs 51.76x23.74 µm (c) *Haemonchus* eggs 68.42x47.57 µm and (d) *Haemonchus* eggs 64.98x39.82 µm

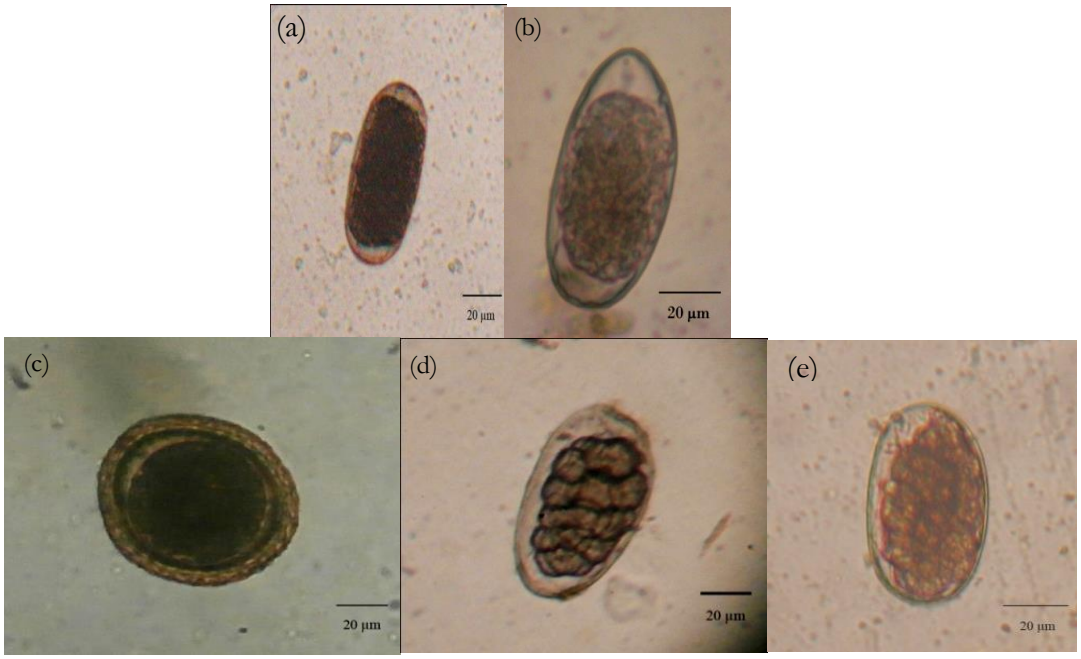

Figure 4. (a) *Cooperia* eggs 80.67x39.52 µm, (b) *Trichostrongylus* eggs 78.42x35.62 µm (c) *Toxocara* eggs 75.43x67.52 µm, (d) *Oesophagostomum* eggs 85.61x48.23 µm and (e) *Oesophagostomum* eggs 67.52x35.31 µm

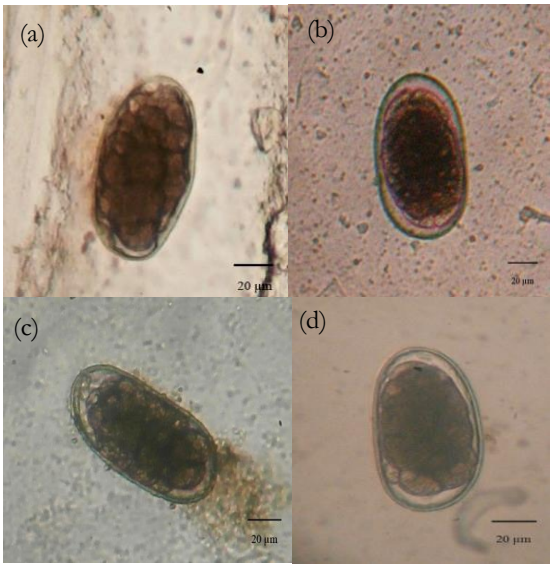

Figure 5. (a) *Bunostomum* eggs 107.33x55.02 µm, (b) *Bunostomum* eggs 119.36x68.77 µm (c) *Bunostomum* eggs 100.37x47.64 µm and (d) *Bunostomum* eggs 108.42x55.33 µm

Eggs size of gastrointestinal worms

The results showed that there were differences between length and width which compared to some literatures (Table 2). Three genus worm eggs had length between 30-50 µm, five genus had length 70-100 µm and only one genus more than 100 µm. The diameter of eggs in *Moniezia* was 47.96 µm. Grifiths (1978), Foreyt (2001) and (Zajac and Conboy, 2011) reported that *Moniezia* had differences size of the diameter with the result (between 12.04 - 27.04 µm). Genus *Strongyloides* had an average size with length and width 50.82 µm and 27.60 µm. The identification based on Grifiths (1978), Foreyt (2001) and Zajac and Conboy (2011), showed that the length of the egg was still in the size range, while the ratio of the width of the egg with the identification of Grifiths (1978) and Foreyt (2001) were higher in the results of this study with the margin was 2.6 µm and 5.6 µm, the ratio of the width to the identification of eggs by Zajac and Conboy (2011) were higher in the literature.

Table 2. Differences length size and width gastrointestinal worm eggs

No	Gastrointestinal Worm	Average (µm)	Size (µm)		
			Griffiths (1978)	Foreyt (2001)	Zajac and Conboy (2011)
30 – 50 µm					
1	*Moniezia*	Diameter 47.96	Diameter 60	Diameter 60	Diameter 65-75
2	*Strongyloides*	50.82x27.60	40-60x 25	50x22	40-60x32-40
3	*Capillaria*	51.26x22.76		50x30	45-50x22-25
70 – 100 µm					
1	*Haemonchus*	68.87x42.02	65-80x40-45	80x45	
2	*Cooperia*	76.65x36.67	70-90x40	77x34	
3	*Trichostrongylus*	81.82x44.20	85x35	85x40	
4	*Oesophagostomum*	76.56x41.77	80x40	80x40	
5	*Toxocara*	73.99x68.53	90x80		75-95x60-75
> 100 µm					
1	*Bunostomum*	103.45x54.45	105x40	95x50	

The measurement results of the genus *Capillaria* had an average size with length and width (51.26 µm and 22.76 µm). The identification results according to Zajac and Conboy (2011) reported that the length of the larger eggs were found in the research, the differences between the maximum length in the literature was 1:26 µm, while the width of the egg was still in the size range with literature (22 – 25 µm). The results of Foreyt (2001) were higher in the results of this study with the margin 1.26 µm, while the widths of the eggs were higher in the literature with the margin 7.24 µm.

The measurement results against *Haemonchus* had an average size with the length and width (68.87 µm and 42.02 µm). The identification results according to Grifiths (1978) said that the length and width of the eggs were still in the size range with literature, while the morphological egg size compared with the results of Foreyt (2001) showed that in the literature were higher with the margin 11.13 µm of length and width 2.98 µm. The result of measurement in genus *Cooperia* had an eggs average with length (76.65 µm) and width (36.67 µm) (Griffiths (1978) and Foreyt (2001) reported that the length of the egg was still in the size range with the literature, while the width of the eggs were different, (3.33 µm and 2.67 µm).

The result of measurement in genus *Trichostrongylus* had an eggs average with length (81.82 µm) and width (44.20 µm). The differences of morphology with the identification by Grifiths (1978) and Foreyt (2001) reported that the length and width of the eggs were different. The longer egg according to the literature was 3.18 µm, while the width between 9.2 µm and 4.2 µm. Genus *Oesophagostomum* had an average size with length and width (76.56 µm and 41.77 µm). The measurement results according to Grifiths (1978) and Foreyt (2001) showed that the length of *Oesophagostomum* was 80 µm and the width was 40 µm. These results indicate that the size of the eggs were higher in the literature with the margin 3.44 µm, while the width of the eggs were higher in the results of this study with the margin 1.77µm. The measurement results of *Toxocara* had an average size with length and width (73.99 µm and 68.53 µm). The identification results according to Zajac and Conboy (2011) showed that a greater length range eggs were found in the literature between 75 – 95 µm, while the width of the egg was still in the range (60 – 75 µm). Grifiths (1978) reported that in the literature had the same length and width (11.47 µm and 16:01 µm).

Genus *Bunostomum* had long size compared to other Nematode eggs. The average length and width of eggs were 103.45 µm and 54.45 µm. Differences of the morphology with the identification by Grifiths (1978) reported that the length of the egg was almost the same, with the difference was 1.55 µm, while the width of the eggs have a large size in the results of this study, the difference was 14.45. The identification results according to Foreyt (2001) with the results of observations had differences of the morphology. The larger size of the egg was found in the results of this study, the difference in length was 8.45 µm and width was 4.45 µm.

Identification of gastrointestinal worm larvae

Six types of worms have been identified i.e *Strongyloides*, *Haemonchus*, *Cooperia*, *Oesophagostomum*, *Bunostomum* and free living of nematode larvae. The head of *Strongyloides* larvae have slightly rounded and the tail not have a sheath. Larvae of *Strongyloides* have a very long esophagus, almost fully the body length (Figure 6). The larvae free living nematodes the types of Rhabditiform esophageal there are the body (corpus), flat/straight (isthmus) and spherical gibbosity (bulb) (Figure 6). The larvae of *Haemonchus* have the tapered head as bullet and the tail sheath is not length (Figure 7). The head of *Cooperia* larvae have rounded and tapered tail sheath. There are pair of refractile bodies on the head and tail sheath not length (Figure 8). The head of *Oesophagostomum* larvae have squared and the tail sheath is very long (Figure 9). The tail sheath to have was length ± 18 – 20 % from totality body length. The head of *Bunostomum* larvae have tapered as bullet and long tail sheath. The tail sheath to have was length ± 13 % from totality body length. The worm larvae also have a bubble or spherical on the esophagus (bulb) (Figure 10).

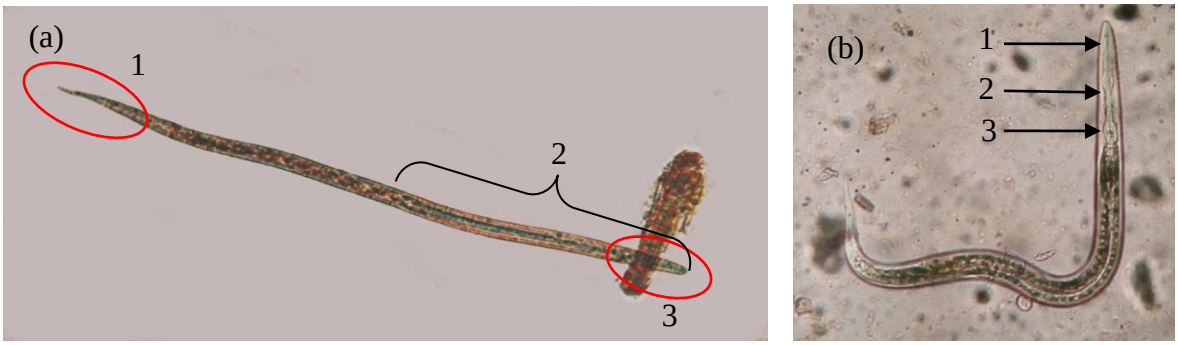

Figure 6. (a) Tail sheath (1), esophagus (2) and head (3) *Strongyloides* larvae
(b) *Corpus* (1), *isthmus* (2) and *bulb* (3) Rhabditiform larvae types esophagus nematode free living

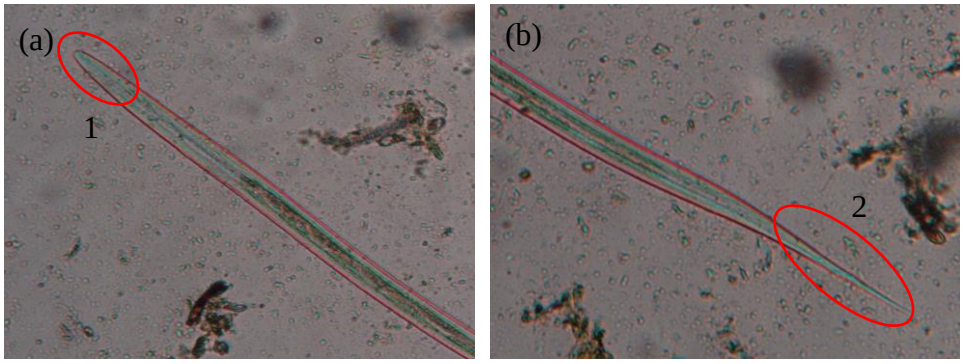

Figure 7. (a) Head (a) and tail sheath (b) *Haemonchus* larvae

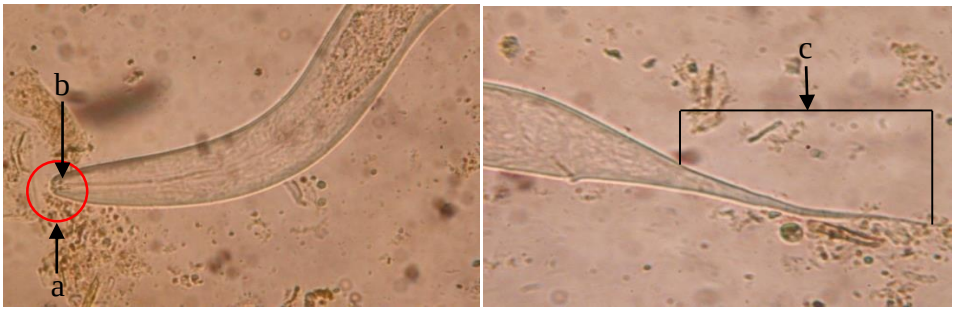

Figure 8. Head (a), *refractile bodies* (b) and tail sheath (c) *Cooperia* larvae

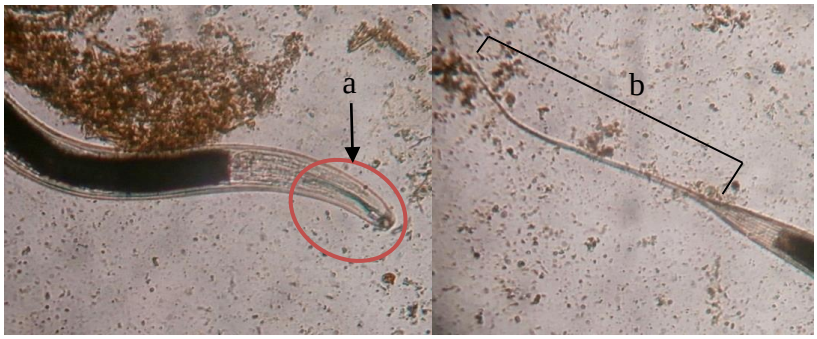

Figure 9. Head (a) and tail sheath (b) *Oesophagostomum* larvae

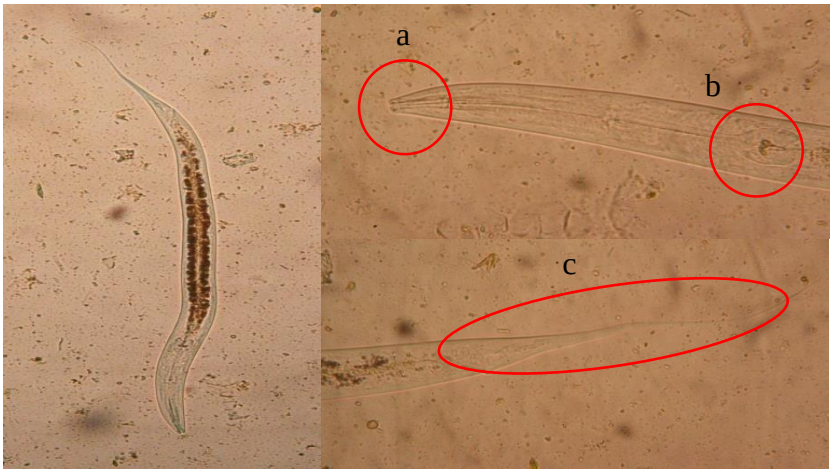

Figure 10. Head (a), *bulb* (b) and tail sheath (c) *Bunostomum* larvae

The *Strongyloides* are also called threadworm. They were found in the small intestine of sheeps, goats, cattle and other ruminants. Larvae of *Strongyloides* can caused intestine inflammation (Levine, 1994). The esophagus types of the larvae are very different i.e the length of esophagus have the same size with body length. This is according to Wyk and Mayhew (2013), said that the length of esophagus reached 40% from the body length. *Haemonchus* are the most influenced class in ruminants, especially in sheep, goats, cattle and other ruminants (Levine, 1994). *Haemonchus* worms caused anemia, depression, weight loss, and death (Miller and Horohov, 2006). The morphology of these larvae are tapered head and the tail sheath is not length. According Wyk and Mayhew (2013), said that the head of larvae are tapered i.e bullet and the tail sheath is not length.

Cooperia are intestinal nematodes in ruminants and nonpathogenic. These larvae were found in small intestine of sheep, goats, cows and usually found in bison. The morphology of larvae have two points on the head. According to Wyk and Mayhew (2013) with Zajac and Conboy (2011), said that the larvae of *Cooperia* have to the pair of refractile bodies on the head. *Oesophagostomum* also called as a nodule worms. These larvae were found in large intestine of sheep, goats, cattle, zebu, buffalo river. These larvae causing inflammation of intestinal wall edema, anemia, diarrhea and decreased appetite (Levine, 1994). The larvae of *Oesophagostomum* have a very long tail sheath (Wyk and Mayhew, 2013).

Bunostomum are hookworms were found in small intestine of sheep, goat and cow. The larvae causing an anemia. The larvae have spherical or bubble on the esophagus. According to, Wyk and Mayhew (2013), said that the larvae of *Bunostoumum* have bulb or jutty on this esophagus types. Free living nematode larvae in this study have the types of Rhabditiform esophagus, that there is corpus, isthmus and bulb. The larvae were dominant in this study. According to, Wyk and Mayhew (2013), that the free living nematode larvae dominate in every cultured larvae in faecal of ruminants.

Livestock management and environmental data of location

The presence of gastrointestinal worms in buffalo was influenced by some factors i.e livestock management and environmental factors (Table 3). Livestock management in this study was semi-intensive. The altitude of location will influenced against the geographical condition. The differences of environmental conditions will have an impact on the types and number of gastrointestinal worms that found.

Table 3. Livestock management and environmental factor

Location	Livestock Management	Environmental factor			
		Elevation (masl.)	Rainfall (mm)	Temperature (°C)	Humidity (%)
Bogor (West Java)				Nov 2013 – Jan 2014	
1. Tenjolaya	Semi intensive	220			
2. Ciampea	Semi intensive	154	432.2	25.1	84
3. Darmaga	Semi intensive	207			
Demak (Central Java)					
4. Mranggen	Semi intensive	44	121	28.3	81.7
Lombok (NTB)				Mei 2014	
5. Lamper	Semi intensive	15	30.09	32.10	
East Java				June – July 2014	
6.Tulungagung	Semi intensive	85			
7. Kepanjen	Semi intensive	139	166	24	75.5
8. Lamongan	Semi intensive	22			

In Indonesia, livestock in buffaloes are conducted with extensive, semi-intensive and intensive. The livestock management of buffaloes in the study as semi-intensive (Table 3). The buffalo which released in the grazing at the morning will be face the high risk of infection from gastrointestinal worm. Larvae move to the top of grass when intensity of light is low and the leaves still wet (Bukhari and Sanyal, 2011).

The location of grazing is often used to release the ruminant livestock breeders. Grazing buffaloes can get the infection of gastrointestinal worms when infective larvae swallowed (Saverwyns, 2008). The buffaloes have a wide area for grazing and moving from one place to other. Place it makes more risky to the gastrointestinal worm infection (Waller, 2003).

Stock farm which located at altitude have highest prevalence of gastrointestinal worm infection. The locations at altitude areas have suitable climatic conditions for the development life cycle of gastrointestinal worm (Kantzoura *et al.*, 2012). The different altitude with the sea will be caused the differences of temperature and humidity (Roza, 1991). Temperature, humidity and rainfall are important factors of environment, because they have great influence on worm infection in buffaloes. Gastrointestinal worms required suitable climatic conditions for live. The differences the prevalence of gastrointestinal worms in buffalo can be caused by the differences of environmental conditions such as temperature and rainfall (Bilal *et al.,* 2009; Dijk *et al.,* 2010).

Prevalence of gastrointestinal worm on the location

Gastrointestinal worm infections in every location showed that there were differences in prevalence. The highest prevalence of Cestode (*Moniezia*) and nematodes found in Bogor (Table 4). Total prevalence of gastrointestinal worms in Buffalo based on different location, were found highest in Bogor compared in Demak, East Java and Lombok (Table 4). Bogor has a high humidity and temperature that suitable for the Cestode to develop. Cestode required an intermediate host to reach the larvae stage infective such as genus grass mites. They need climate condition was 12-28°C, with humidity 25°C (Narsapur, 1988). The buffaloes will be infected with Cestode larvae caused by mites swallowed when looking at pasture. The Cestode eggs can be develop to the larvae with warm temperature and high humidity. Temperature condition that suitable for the eggs of Cestode develop on the larvae was 25 °C (Narsapur, 1988). Nematodes are gastrointestinal worm that do not required an intermediate host. Their live cycle more quickly (Southwell, 2008). Nematode can grow in optimum rainfall above 55 mm with temperature 18°C (Levine, 1994). This influenced high level of intensity on gastrointestinal worm nematode infection in Bogor (Table 2). The gastrointestinal worm can develop in the rainy season than summer (Chiejina and Fakae, 1984; Pfukenyi *et al.*, 2007).

Table 4. Prevalence gastrointestinal worm based study location

Gastrointestinal Worm	Location			
	Bogor (n=2) %	Demak (n=15) %	East Java (n=37) %	Lombok (n=10) %
Cestode				
Moniezia	17.2	6.7	2.7	10
Nematode				
Haemonchus	24.1	13.3	10.8	0
Bunostomum	24.1	33.3	8.1	20
Cooperia	10.3	13.3	5.4	20
Trichostrongylus	0	0	2.7	10
Strongyloides	20.7	6.7	8.1	10
Oesophagostomum	3.5	6.7	0	0
Capillaria	0	0	2.7	0
Toxocara	0	0	2.7	0
Total Nematode	82.7	73.3	40.5	60

The highest total prevalence of gastrointestinal worms in Buffalo based on the location was found in Bogor (Table 4). The highest prevalence in Bogor related to environmental conditions. Bogor has a high humidity and rainfall that suitable for the gastrointestinal worms to develop. Gastrointestinal worms will be develop to infective larvae in buffaloes to influenced temperature, high humidity and rainfall. The gastrointestinal worms not develop to infective larvae without the favourable environmental conditions. These gastrointestinal worms have a direct life cycle consisting of free-living stages on pasture (egg to infective larvae) and after ingestion, gastrointestinal worm stages (adult) in the host. The low prevalence in Central Java, East Java and Lombok (NTB) were caused by the differences of environmental factor and geographical condition. The third location to have low rainfall and humidity. This is influenced to develop gastrointestinal worms to infective larvae. Differences in the prevalence of ruminants were caused by the differences in environmental conditions such as temperature and rainfall (Bilal *et al.,* 2009; Dijk *et al.,* 2010). The favorable environmental conditions can be influenced the development of gastrointestinal worms infection rates.

Prevalence and infestation of gastrointestinal worm in moist and dry area

Total prevalence and infestation of gastrointestinal worm in moist area (West Java) was the highest (62.1%), compared to the dry areas (Central Java was 53.3%, followed by Lombok 50% and East Java 35.1%). The calculation of Faecal Eggs Count (FEC) in West Java was the highest i.e 840 Eggs Per Gram (EPG), followed by East Java (570 EPG), in Central Java (EPG 375) and Lombok (135 EPG). Based on the prevalence data and gastrointestinal worm infestation in the wet/moist and dry, showed that in Bogor (West Java) had the highest prevalence and worm infestation, compared with dry area in Demak (Central Java), East Java and Lombok (NTB). Prevalence and infestation in Bogor areas (62.1% and 840 EPG), in Central Java (53.3% and 375 EPG), in East Java (35.1% and 570 EPG) and in Lombok (50% and 135 EPG) (Table 5). This was caused by the differences of environmental factor and geographical condition.

Data obtained by Darmaga BMKG (2014), said that the monthly rainfall Bogor between November 2013 until January 2015 were 186.9 mm, 407.7 mm and 702 mm, with average of rainfall 432.2 mm and temperature 25.1 ° C with the average of humidity 84% (Table 3). This monthly rainfall category classified as extremely high rainfall. The rainy season influenced infection of gastrointestinal worm. On the other hand, the altitude factor also influenced the gastrointestinal worms (Kantzoura *et al.,* 2012). Bogor had an average altitude 193.7 masl, while the other areas

only 100 masl (Table 3). In this study, the altitude data showed that in Bogor had different altitude with other areas. This altitude influenced to the environmental condition, this matter will have an impact on the develop of gastrointestinal worms.

Table 5. Total prevalence and gastrointestinal worm infestation areas damp and dry

No	Location	Prevalence (%)	Faecal Eggs Count (FEC) Unit Eggs per Gram (EPG)
1	Bogor (West Java)	62.1	840
2	Demak (Central Java)	53.3	375
3	Tulungagung, Kepanjen, and lamongan (East Java)	35.1	570
4	Lombok (NTB)	50	135

Prevalence of gastrointestinal worms according to sex and age

The prevalence of gastrointestinal worm eggs based on sex were found highest in males than females, *Moniezia* was 10.5% and nematodes was 68.4%, the age factor were found highest at age <1 years (*Moniezia* 13.3% and nematodes 66.6%) (Table 5).

Table 6. Prevalence of gastrointestinal worm based sex and age

Gastrointestinal Worm	Sex (%)		Age (%)		
	Male (n=38)	Female (n=53)	< 1 years (n=15)	1-5 years (n=35)	> 5 years (n=41)
Cestode					
Moniezia	10.5	7.5	13.3	2.9	9.8
Nematode					
Haemonchus	15.8	11.3	13.3	17.1	2.4
Bunostomum	18.4	17	33.3	11.4	14.6
Cooperia	7.9	13.2	20	8.6	12.2
Trichostrongylus	5.3	1.9	0	2.9	4.9
Strongyloides	18.4	9.4	0	14.2	14.6
Oesophagostomum	2.6	1.9	0	2.9	2.4
Capillaria	2.6	0	0	0	2.4
Toxocara	0	1.9	0	0	2.4
Total Nematode	68.4	56.6	66.6	57.1	53.5
Total Prevalence	57.9	43.4	66.7	54.3	41

Infestation level of gastrointestinal worm based on faecal eggs count (FEC)

The infestation of gastrointestinal worm based on the calculation of Faecal Eggs Count (FEC) on sex were found highest in females i.e 1020 Eggs Per Gram (EPG) (r = 113.3) and males 900 EPG (r = 100). The calculation in the age group, were found highest of 1-5 years were 735 EPG (r = 81.7), followed by > 5 years were 615 EPG (r = 68.3 and <1 year were 570 EPG (r = 63.3).

Table 7. Gastrointestinal worm infestation based calculation faecal eggs count (FEC)

No	Gastrointestinal worm	Faecal Eggs Count (FEC) Unit Eggs per Gram (EPG)				
		Sex		Age		
		Male	Female	< 1 years	1-5 years	> 5 years
1	*Moniezia*	90	90	30	105	45
2	*Haemonchus*	450	165	285	270	60
3	*Bunostomum*	135	315	105	105	240
4	*Cooperia*	60	120	45	75	60
5	*Trichostrongylus*	30	45	0	45	30
6	*Strongyloides*	120	120	105	75	60
7	*Oesophagostomum*	15	15	0	0	30
8	*Capillaria*	0	60	0	60	0
9	*Toxocara*	0	90	0	0	90
	Total	900	1020	570	735	615
	Average	100	113.3	63.3	81.7	68.3

Prevalence of gastrointestinal worms based on sex factor

Based on observation showed that *Moniezia* (Cestoda) and Nematodes have the highest prevalence in male buffalo (Table 6). Previous study reported that infection *Moniezia* (Cestoda) and Nematodes were found highest in males than female buffalo (Bilal *et al.,* 2009; Rafiullah *et al.,* 2011; Singh *et al.,* 2012). Totality prevalence of gastrointestinal worms in this study were found highest in males (57.9%) than females (43.4%). Some studies have found that there is none relationship between the sex with gastrointestinal worm infection. The differences in sex prevalence were more influenced by diet (Raza *et al.,* 2013).

Prevalence of gastrointestinal worms based on age factor

The highest prevalence of gastrointestinal worms in *Moniezia* (Cestoda) and the nematodes was found in aged <1 year (Table 6). The highest prevalence *Moniezia* (Cestoda) and nematodes were found in young group age or ≤ 1 years (Pfukenyi *et al*., 2007; Dong *et al.* 2012). Total prevalence of gastrointestinal worms in age group of buffalo were found highest in age <1 year, compared to 1-5 years and > 5 years. Prevalence 66.7%, followed 54.3%, and 39% (Table 6).

Gastrointestinal worm is one of the main problems that can be affecting directly to the productivity of the buffaloes. The age factor of buffaloes was greatly affecting the risk of occurrence the gastrointestinal parasite (Awraris *et al.,* 2012). The buffalo age group had level of immunity were different to gastrointestinal worms infestation, caused by the immune system. The Highest infestation rate was in young ruminant compared with the old ruminant. This is due to young ruminants had the lower gastrointestinal antibodies against worms (Colditz *et al.,* 1996; Khan *et al.,* 2010). The ages of animals are the important factor in the prevalence of gastrointestinal worms infection. The higher prevalence was found in young buffaloes compared to the older (Patel *et al.,* 2015). The infestation of gastrointestinal worm of buffalo in young age can caused the infection by other buffaloes, poor hygienic conditions of the farms and livestock in one farm with the older buffalo (Nugraha, 2015). All the gastrointestinal parasite groups have different transmission patterns, ecological requirements and deployment strategies (Dijk *et al.,* 2010).

Faecal eggs count (FEC)

The infestation of gastrointestinal worm rate based on the calculation of faecal Eggs Count (FEC) showed that the highest were in females (r = 113.3 EPG) than males (r = 100 EPG). The infestation rate for the age factor were found highest in age 1-5 years (r = 81.7 EPG), followed by age > 5 years (r = 68.3 EPG) and < 1 years (63.3 EPG) (Table 7). This is different to results reported by Mamun *et al.,* (2011), where the male buffaloes had the highest EPG compared to

females. Whereas for the age factor were found higher in 6 months age until 2 years, compared to the age of 2-5 years and > 5 years.

CONCLUSIONS

The types of gastrointestinal worm eggs found in the faecal of buffalo in West Java and Central Java, there were six genera i.e *Moniezia, Haemonchus, Bunostomum, Cooperia, Strongyloides* and *Oesophagostomum,* in East Java were found eight genera i.e *Moniezia, Haemonchus, Bunostomum, Cooperia, Strongyloides, Trichostrongylus, Capillaria* and *Toxocara*, while in Lombok were found five genera i.e *Moniezia, Bunostomum, Cooperia, Strongyloides, Trichostrongylus*. The totality sample identification of faecal to culture technique used water medium were found six genera i.e *Strongyloides, Haemonchus, Cooperia, Oesophagostomum, Bunostomum* and free living larvae.

The differences size of morphology from gastrointestinal worm eggs the still very few is reported, this is guess were caused environmental condition, geographycal site and woof. Prevalence and infestation of differences in geographycal conditions were found highest in Bogor. The prevalence of gastrointestinal worms in males were highest than females, whereas for larger infestation was found in females. The prevalence of gastrointestinal worms was found at age <1 year, whereas a larger infestation was found at the age of 1-5 years.

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