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RESEARCH ARTICLE

PREVALENCE OF DONKEY TRYPANOSOMOSIS IN SELECTED DISTRICTS OF WEST WOLLEGA ZONE, WESTERN OROMIYA

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ABSTRACT

A cross-sectional study was conducted in October 2015 to December 2016 in eleven (11) peasant associations (PAs) randomly selected from three purposively chosen districts (Gimbi, Lalo Assabi and Nedjo) of West Wollega Zone, Oromiya Regional State, Western Ethiopia to estimate the prevalence of donkey trypanosomosis and to identify the trypanosomes species involved and their relationship with the mean PCV. Blood samples were collected from the ear vein of randomly selected 384 donkeys and examined by Buffy coat technique. The overall prevalence of trypanosome infection in donkeys was 18.8% (n=72) and different prevalence rates of donkey trypanosomosis were recorded in Gimbi (11.3%), Lalo Assabi (8.4%) and Nedjo (32.7%) in which highest prevalence was observed. However, the disease was statistically significant among animals from Gimbi (P=0.000, OR=3.807, CI=1.947-7.44) and Lalo Assabi (P=0.000, OR=5.3, CI=2.5-11) districts as compared to those animals sampled from Nedjo district. Highest (43.1%) and lowest (8%) prevalence of the disease was recorded in Waligalte Adda PA of Nedjo district and Gimbi 01, respectively. However, prevalence of donkey trypanosomosis was statistically significant ($p<0.05$) in only two PAs (Werego Arsema and Enango 01) of Lalo Assabi district. Higher prevalence rate was observed in adult (21.1%) than young (11.6%) age groups and in male (27.4%) than in female donkeys (12.3%). Also highest prevalence was seen in animals with poor body condition. Age, sex and body condition showed a significant association with the prevalence of donkey trypanosomosis in the areas ($p<0.05$). *Trypanosoma vivax* (37.5%) was the most predominant trypanosome species encountered during the study period followed by *T. congolense* (26.4%), *T. brucei* (26.1%) and mixed parasites (13.9%). High distribution of different species of Trypanosoma parasites was detected in Nedjo district. The overall mean PCV values were 21.2% and 43.2% for infected and non-infected animals, respectively. Statistically significant difference ($P<0.05$) was observed between the overall mean PCV values of infected and non-infected animals and the mean PCV was found to be dependent on PAs, Sexes, Ages and body conditions of donkeys. In conclusion, the result obtained in this study is an important disease of donkeys in the study areas where donkeys are extensively kept and used. So, further longitudinal study should be conducted in the areas to generate a complete data of the disease in the study areas.

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INTRODUCTION

Trypanosomosis cause significant loss in animal production and it greatly affect people and animal settlement in considerable parts of world (Ermias and Getachew, 2001). Over 10 million KM^2 areas of Africa greatest agricultural potential are infested by tsetse fly, which is the main vector of disease (Dargant *et al*., 2001). It is arguably the most significant disease that occurs across more than a third of Africa, where the keeping of ruminants and equines are crucial (Dhollander *et al*., 2006 and Auty *et al*., 2008).

In Ethiopia, Trypanosomosis is one of the most important disease limiting livestock productivity and agricultural development due to its high prevalence in the most arable and fertile land of south west and north west part of the country following the greater river basins of Abay, Omo, Ghibe, and Baro with a high potential for agricultural development. Currently, about 220,000 KM^2 areas of the above mentioned area infested with five species of tsetse flies including *G. pallidipes, G. morsitans submorsitans, G. fuscipes, G. tachinoides* and *G. longipennis.* Part of the Amhara region is tsetse infested, *G. morsitans sub morsitans* and *G. tachinoides* are reported (Langridge, 1976). Equines play a key role in the agricultural economy of the country where poor infrastructure and very ragged topography in many parts of rural Ethiopia

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have made transportation by vehicle inaccessible. They are used for pack transportation, riding, carting and threshing farm cultivation among others. If equines are not available, women often have to do the same work (Mohammed, 1991 and Abo-Shehada, 1988). Donkey is one of the most important draught animals, exist in millions and serve a key role in the agricultural economy of the world (Soulsby, 1982). The animal constitute 70% of the African equine species and are predominantly found in the arid and semi-arid areas providing a reliable, environmentally friendly and renewable source of power to millions of poor communities of worldwide (Fielding and Pearson, 1991). It is well adapted to arid, semi-arid areas and temperate regions (Oppong, 1979; Fielding, 1987 and Abo-Shehada, 1988). Donkeys are considered as beast of burden in many developing countries including Ethiopia that is the second largest donkey population in the world. They are docile and easy to manage and provide transport among communities in areas with poor terrains or in inaccessible areas with poor infrastructure. They have the ability to withstand harsh conditions in many rural settings in which they are reared but that notwithstanding, the donkey has received least attention especially where they are kept with other livestock. This may be due to the mistaken belief that they are hardy and hardly affected by any disease (Mengistu *et al.,* 2001). The studies of the disease undertaken in the mid to late 1990s show an increased prevalence with values ranging from 43-63% in horses of the Central River Division of the Gambia (Faye *et al*., 2001, Dhollander *et al*., 2006). In general, horses are considered to be highly susceptible to infection, while donkeys are considered to be more resistant, although the reasons for lower prevalence in donkeys could also be due to feeding preferences of the tsetse vector (Gina *et al*., 2008).

Studies carried out in Ethiopia, Uganda and Kenya and other countries have shown that donkeys are afflicted by ailments that affect other livestock species, with helminths being an important constraint to growth and productivity (Ayele *et al*., 2006, Saul *et al*; 1997, Githiori *et al*., 1998). Also of significance are conditions due to neglect and mishandling with harness wounds being common (Blakeway 1994, Saul *et al.,* 1997). Where they are found in tsetse infested areas, trypanosomiasis is an important constraint to performance and productivity of donkeys (Kanchula and Abebe, 1997; Githiori *et al*., 1998). The importance of trypanosomiasis in donkeys was demonstrated in the study by Assefa and Abebe (2001) where more than 93% of infected donkeys were in poor body condition, in contrast to 4.17% in the non-infected ones. According to the study of Abebe and Wolde (2010) in Assosa and Homasha district in Benishangul Gumuz regional state, the prevalence of donkey trypanosomosis was 6.3%. Shelima *et al*., (2006b) stated that the prevalence of donkey trypanosomosis in Welayta zone, southern Ethiopia was 28.5%. In other studies the prevalence of donkey trypanosomosis in the Northern Omo zone, southern Ethiopia was 21% (Kebede and Getachew, 1997). Currently the 6% prevalence was reported from Assosa District, Benishangul Gumuz Regional State, Northwest Ethiopia (Assefa *et al*., 2015). This shows the presence of scanty information on equine trypanosomosis regardless of the fact that these animals play a key role in the agricultural economy of the country. They also serve the community in a very ragged topography in many parts of rural Ethiopia where transportation by vehicle inaccessible. The study areas have also large equine population and widespread practice of equine for multidirectional activities.

But, the trypanosomosis was claimed by farmers to be the leading health constraint of these animals in the area. However, there was no sufficient information on the problems stated above which needs study in the study area. Therefore, objectives of this study were to estimate the prevalence of equine trypanosomosis and to identify the trypanosomes species involved and their relationship with the mean PCV.

MATERIAL AND METHODS

Description of the Study Area: The study was conducted from October 2015-June 2016 in purposively selected three districts of West Wollega, Western Oromiya. West Wollega is one of the 18 Administrative Zones of Oromiya National Regional State. Administratively, the Zone has 21 districts, of which 19 are rural districts and 2 are urban administrations which again subdivided into 533 Peasant Associations (487 rural and 46 urban dwellers associations). Gimbi Town, which is located at a distance of 441 km from Addis Ababa, is the capital of the zone. Astronomically, it is located between $8^{o}12'$-$10^{o}03'$ N latitudes and $34^{o}08'$-$36^{o}10'$E longitudes. It is located in the western part of Oromiya Region, bordered with Benishangul Gumuz Regional State in the Northwest, Northeast & East; Qellem Wollega Zone in the West, East Wollega Zone in the East, Gambella Regional State and Illubabor Zone in the South. The land area of the Zone is estimated to be 14,160.29 km^2. It experiences tropical climate because of the relatively high angular position of the sun. The mean annual temperature is fairly high. Generally, mean annual temperature of the Zone varies from $15^{o}C$ to over $25^{o}C$ (SEADWWZ, 2008/09). As reported by West Wollega Zone Finance and Economic Development Office (WWZFEDO) (2011), the annual rainfall pattern in the Zone generally decreases from East to West following the physiographic nature of the Zone. The mean annual rainfall of the Eastern high lands range from 1800-2000mm, while in the central plateaus range between 1600-1800mm and in the remaining parts of the Zone it becomes between 1200-1600mm and becomes less than 1200mm in the Southwestern parts of the Zone. Livestock population of West Wollega Zone is 1,775,404 Bovines, 385,098 Ovine, 353,385 caprines, 137,926 Equines, 2,066,678 poultry and 620,397 Bee colonies (WWZLDHAO, 2011). From the nineteen (19) rural districts of the Zone, three of them were selected:

Gimbi district: At present it has 32 administrative sub divisions out of which 31 are peasant associations and the reaming 1 is urban center. Gimbi is the Capital of the Zone. The district is located in the eastern part of west wollega Zone and at a distance of 441km from Addis Ababa. It is bounded by East Wollega zone in the East, Beni-Shangul Gumuz Regional State north, Lalo Assabi district in West and Homa district in South and has a total area of 1172 Km^2. Astronomically, the district is located between $9^{0}10^{0}$-$9^{0}17^{0}$ North latitude and $35^{0}44^{0}$-$36^{0}09^{0}$ East longitudes. The mean minimum and maximum annual temperature ranges between $10^{o}C$ and $30^{o}C$. The mean annual rain fall is 1400-1800ml. It lies with on altitudinal range of 1200m-2222 m above sea level (a.s.l.). As reported by Ghimbi District Finance and Economic Development office (GDFEDO) (2001), the district has high livestock potential with 107,334 cattle, 13,476 Ovine, 5124 Caprine, 5211 Equine, and Poultry 44144 and 25600 Bee Colonies.

Lalo Asabi district: having an area of 418 km^2, is located in the eastern part of West Wellega zone at a district of 23 km from Gimbi town (zone capital). It is situated along Gimbi-Dembi Dolo main road. It shares common boundaries with Gimbi, Ayra Guliso, Bodji and Yubo districts, and Benishangul-Gumuz Regional State. Enango town is its capital which is about 23km far away from Capital of the Zone (Gimbi). Alititudinally, it stretchers between 1500 and 1900 m.a.s.l. The district has high livestock potential with 38000 cattle, 4100 Ovine, 4180 Caprine, 5383 Equine, 20,000 Poultry. The district is classified into kola (2.2%) and woinadega (97.8%) agro climatic zones (SEPLAD, 2011).

Nejo District: is one of the West Welega Zone which is bordered on the southeast by Boji, on the west by Jarso, on the northwest by Mana Sibu, and on the north and east by the Benishangul-Gumuz Region. The administrative center is Nejo; other towns in Nejo include Gori and Wara Jiru. It is known by gold mine deposited on Tulu Kami and the Laga Gumbi hills, as well as along the Alaltu and Dilla Rivers. Coffee is an important cash crop of Nejo. Over 50 square kilometers is planted with this crop (LHE, 2007). The 2007 national census reported a total population for this district of 130,909 in 25,336 households, of whom 64,654 were men and 66,255 were women; 24,505 or 18.72% of its population were urban dwellers. The majority of the inhabitants observed Protestantism, with 63.72% reporting that as their religion, while 33.69% observed Ethiopian Orthodox Christianity, and were 1.72% Muslim (PHCE, 2007). The district had a total livestock population of 172941 live stocks in 2000. Of these Cattle 126500, Sheep 25276, Goats 12773, Mules 574, Horses 18 and Donkeys 8700. Some of the major live stock diseases common in the district include anthrax, Black legs, FMD, CBPP internal and external parasite, Bacterial infection (SEADWWZ, 2008/09).

Study Animals and Design: The study animals were indigenous breeds of donkeys, all age and sex categories. Animals in the areas were kept under extensive husbandry system together with other livestock around villages. A cross-sectional study was carried out to determine the prevalence of equine trypanosomosis in the study areas.

Sampling Technique and Sample Size Determination

The three districts were purposively selected based on the accessibility, donkey population, lack of scanty information about the disease, presence of tsetse and other biting flies. These districts share similar farming system but there was a little difference in agro-ecological conditions. They have also different ranges of livestock population. Animals included in the study were distributed over the selected districts. Three PAs were randomly selected from each district in consultation with the respective district especially Livestock and Fisheries Development and Resource office expert's. The simple random sampling technique was also being followed to select the animals to be included in the study. A sample size should be determined based on the study type and sampling method for the investigation. The sample size was decided based on formula described by (Thrusfield, 2005) with 95% confidence interval at 5% desired absolute precision and it is achieved by assuming the expected prevalence of 50%. Accordingly, a total of 384 desired sample sizes for the study should be calculated and based on the animal population they have, this sample sizes was divided to the three districts.

$$n = 1.96^2 P_{exp}(1-P_{exp}) / d^2$$

Where: **n** = required sample size; $\mathbf{ex_p}$ = expected prevalence and **d** = desired absolute precision.

$$\text{Therefore, } n = \frac{(1.96)^2 * 0.5 * (1-0.5)}{0.25} = \frac{3.8416 * 0.25}{(0.05)^2} = 384$$

Data Collection Methods: The data were collected to determine the prevalence of donkey trypanosomosis in the area from November 2015 to April 2016. Here some different technical procedures, laboratory procedures and scientific methods were applied for investigation of the parasites (Uilenberge, 1998).

Sample collection

Parasitological and Hematological Survey: To determine the prevalence of donkey trypanosomosis and to estimate the potential risk factors associated with the disease in the study areas, a cross-sectional parasitological survey was used. A total of 384 blood samples were collected directly from the ear veins of the study animals into heparinized capillary tubes, after properly securing the animal and aseptically by puncturing with the tip of lancet on the margin of ear veins of the animals. During bleeding, each lancet was used for each animal to prevent cross contamination of the sample. Subsequently, a heparinized capillary tubes were filled three fourth of their length. The blood samples were examined by the capillary micro-hematocrit centrifugation method to estimate the packed cell volume (PCV) as an indicator of anemia. Consequently the packed cell volume (PCV) or anemia of each sample was determined by using a hawksley micro-haematocrite reader (Murray *et al*., 1977) and animals with PCV value range of 30-46% as a normal (Knottenbelt 2005), which mean below 30 were considered as anemic. After determination of the PCV, the Buffy coat (BC) was examined by dark ground/phase contrast microscope (Murray *et al.,* 1983) for the detection of trypanosomes in the blood.

Study methods and laboratory procedures

Then parasitological examinations were performed according to the following two procedures. First, the capillary tubes were sealed by using cristaseal at one end. After the first 24 samples were taken, the blood in the capillary tubes should immediately be placed in the centrifuge according to their order and centrifuged at 12,000 revolutions per minute (rpm) for five minutes. Second, the capillary tubes were cut 1cm below the Buffy coat intersection to include the top layer of red blood cells (Parries *et al*., 1982). The content of the capillary tubes was then expressed on to a clean microscope slide, mixed and covered with 22x22mm cover slip. Then, the slide will be examined using a microscope with 40x objective to determine the prevalence of trypanosomosis using the buffy coat /phase contrast/ dark ground technique (BCT). Confirmation of trypanosome species by morphological characteristics were done after staining the blood smear with Giemsa and examination with oil immersion microscopy with 100x power of magnification (Murray *et al*., 1977; Uilenberge, 1998).

Statistical Analysis: The data were collected from each study animal and laboratory analyses were coded into appropriate variables and entered in Microsoft Excel spread sheet. Data analysis was made using Statistical Package for Social Science

(SPSS), version 20 software. Descriptive statistical analysis technique was used to analyze the data. The risk of the disease related to the sex and altitude were assessed using Odds ratio (OR). The level of significance for statistical tests was set at less than or equal to 0.05.

RESULTS

Overall Prevalence of Donkey Trypanosomosis: From a total of 384 examined donkeys, overall of 72 donkeys were found to be infected by different species of trypanosoma parasites and overall prevalence of donkey trypanosomosis (18.8%) was recorded in the study areas. The study comprised peasant associations (PAs), sex, age, Packed cell Volume (PCV), Color, Species of parasites and body conditions of animals as a major factors those play a role for the infection of trypanosoma parasites in the study areas. Comparison was made among the three districts (Gimbi, Lalo Assabi and Nedjo) included in the study. The result of analysis indicated that different prevalence rates of donkey trypanosomosis with 11.3%, 8.4% and 32.7% were recorded in Gimbi, Lalo Assabi and Nedjo districts, respectively. However, the disease was statistically significant among animals sampled from Gimbi and Lalo Assabi districts as compared with those animals sampled from Nedjo district (Table 1).

Arsema and Enango 01) of Lalo Assabi district (Table 1). As analysis of frequency, using descriptive statistics showed that highest numbers of animals sampled were 41 (35.7%), 50 (42%) and 51 (34%) from Were Seyo PA of Gimbi, Tosio PA of Lalo Assabi and Waligalte Adda of Nedjo district. This revealed that, highest frequencies of sampled animals were recorded in Tosio PA of Lalo Assabi district, but highest prevalence rate (43.1%) of the disease was recorded in Waligalte Adda PA of Nedjo district (Table 2 and 3).

Prevalence of Donkey Trypanosomosis Based on Sex, Age, Colour and Body conditions of Animals

Analysis of age wise prevalence of donkey trypanosomosis between two age groups indicated that almost there was a difference in prevalence rate between these age groups that was 21.1% and 11.6% in adult and young, respectively. This result revealed that, higher prevalence rate was observed in adult than young age groups. This result indicated as adult age groups were more likely to be affected by the disease with two times as compared to that of young animals (OR= 2.04; 95% CI:1.03-4.07; P=0.042). Generally, age showed a significant association with the prevalence of donkey trypanosomosis in the areas ($p<0.05$) (Table 3). On the other hand, an association between the disease and sexes of animals (female and male)

Table 1. Association of prevalence of donkey trypanosomosis among the three districts

Districts	Nº. of examined	Nº. of positive	Prevalence Rates (%)	P-value	Odd Ratio	95% CI	
						Lower	Upper
Gimbi	115	13	11.3	0.000	3.807	1.947	7.443
Lalo Assabi	119	10	8.4	0.000	5.288	2.543	10.995
Nedjo	150	49	32.7	-	-	-	-
	384	72	18.8				

Table 2. Association of prevalence of the disease among Peasant Associations

All PAs of the districts	Nº. of examined	Nº. of positive	Prevalence Rates (%)	P-value	Odd Ratio	95% CI	
						Lower	Upper
Gimbi District							
Were Seyo	41	6	14.6	0.16	2.333	0.71	7.69
Choli Michael	29	3	10.3	0.09	3.467	0.81	14.77
Gimbi 01	25	2	8	0.07	4.600	0.87	24.23
Lalo Chirecha	20	2	10	0.13	3.600	0.67	19.22
Lalo Assbi District							
Tosio	50	6	12	0.08	2.933	0.89	9.58
Werego Arsema	39	2	5.1	0.02	7.400	1.43	38.23
Enango 01	30	2	6.7	0.04	5.600	1.07	29.23
Nedjo District							
Waligalte Adda	51	22	43.1	0.20	0.527	0.20	1.42
Nedjo 03	45	12	26.7	0.86	1.100	0.38	3.15
Nedjo 02	26	7	26.9	0.89	1.086	0.33	3.58
Tola Wakayo	28	8	28.6	-	-	-	-
Total	384	72	18.8	-	-	-	-

Prevalence of Donkey Trypanosomosis Based on Peasant Associations

Different prevalence rates of the disease were observed among 11 PAs of the three districts with 14.6%, 10.3%, 8%, 10%, 12%, 5.1%, 6.7%, 43.1%, 26.7%, 26.9% and 28.6%, in Were Seyo, Choli Michael, Gimbi 01, Lalo Chirecha, Tosio, Werego Arsema, Enango 01, WA, Nedjo 03, Nedjo 02 and T/Wataye, respectively. The study revealed that, highest (43.1%) and lowest (8%) prevalence of the disease was recorded in Waligalte Adda (WA) PA of Nedjo district and Gimbi 01, respectively. However, prevalence of donkey trypanosomosis was statistically significant ($p<0.05$) in only two PAs (Werego

was studied and out of examined animals, the majority or 220 (57.3%) were females while about 164 (42.7%) of them were males. But, higher prevalence rate was recorded in male donkeys (27.4%) than in females (12.3%). This result indicated as male animals were more likely to be affected by that diseases with almost three times as compared to that of female donkeys (OR= 2.70; 95% CI: 1.59-4.59; P = 0.000). Generally, sex showed a significant association with the occurrence of major livestock diseases in the areas ($p<0.05$) (Table 3). Based on body condition of the animals, donkeys were grouped into three as poor, medium and good with prevalence of 42 (53.8 %), 13 (8.4 %) and 17 (11.3 %), respectively.

Table 3. Analysis of frequency and its percentage in different PAs with other risk factors using descriptive statistics

Respective Variables	Gimbi (n=115)				Lalo Assabi (n=119)			Nedjo (n=150)				Total
	W/Seyo (%)	C/ Michael (%)	Gimbi 01(%)	L/Chirecha (%)	Tosio (%)	W/Arsema (%)	Enango 01 (%)	WA (%)	Nedjo 03 (%)	Nedjo 02 (%)	T/Wataye (%)	11
No. of sampled	41(35.7)	29 (25.2)	25(21.7)	20 (7.4)	50(42)	39(32.8)	30(25.2)	51(34)	45(30)	26(17.3)	28(18.7)	384
Age												
Adult	29(25.2)	23(20)	23(20)	18 (15.7)	40(33.6)	32(27)	19(16)	37(24.7)	26(17.3)	23(15.3)	19(12.7)	289(75.3)
Young	12 (10.4)	6(5.2)	2(1.7)	2(1.7)	10(8.4)	7(5.9)	11(9.2)	14(9.3)	19(12.7)	3(2)	9(6)	95(24.7)
Sex												
Male	16(13.9)	8(7)	20(7.4)	5 (4.3)	16(13.4)	21(17.6)	16(13.4)	21(14)	16(10.7)	16(10.7)	9(6)	164(42.7)
Female	25(21.7)	21(18.3)	5(4.3)	15(13)	34(28.8)	18(15.2)	14(11.8)	30(20)	29(19.3)	10(6.7)	19(12.7)	220(57.3)
Body condition												
Poor	9(7.8)	6(5.2)	6(5.2)	4(3.5)	-	-	-	21(14)	20(13.3)	3(2)	9(6)	78(20.3)
Medium	26(22.6)	19(16.5)	17(14.8)	15(13)	25(21)	35(29.4)	18(15.2)	-	-	-	-	155(40.4)
Good	6(5.2)	4(3.5)	2(1.7)	1(0.9)	25(21)	4(3.4)	12(10.1)	30(20)	25(16.7)	23(15.3)	19(12.7)	151(39.3)
PCV												
Anaemic	5(4.3)	4(3.5)	5(4.3)	3(2.6)	1(0.8)	-	-	10(6.7)	8(5.3)	6(4)	9(6)	51(13.3)
Non-anaemic	36 (31.3)	25(21.7)	20(7.4)	17(14.8)	49(41.2)	39(32.8)	30(25.2)	41(27.3)	37(24.7)	20(13.3)	19(12.7)	333(86.7)
Colour												
Black	15(13)	9(7.8)	9(7.8)	5(4.3)	26(21.8)	9(7.6)	14(11.8)	8(5.3)	-	-	5(3.3)	100(26)
Whitish	26(22.6)	20(7.4)	16(13.9)	15(13)	24(20.2)	30(25.2)	16(13.4)	43(28.7)	45(30)	26(17.3)	23(15.3)	284(74)
Species of trypanomes												
None	35 (30.4)	26(22.6)	23 (20)	18 (15.7)	44(37)	37(31.1)	28(23.5)	29(19.3)	33(22)	18(12)	20(13.3)	311(81)
Trypanoma congolense	-	1(0.9)	-	1(0.9)	-	-	-	7(4.7)	3(2)	4(2.7)	3(2)	19(26.4)
T. vivax	5 (4.3)	1(0.9)	2 (1.7)	-	5(4.2)	2(1.7)	1(0.8)	2(1.3)	5(3.3)	1(0.7)	3(2)	27(37.5)
T. brucei	-	-	-	1(0.9)	1(0.8)	-	1(0.8)	6(4)	3(2)	3(2)	2(1.3)	17(26.1)
Mixed	1(0.9)	1(0.9)	-	-	-	-	-	7(4.7)	1(0.7)	-	-	10(13.9)
Result												
Negative	35	26 (22.6)	23(20)	18 (15.7)	44(37)	37(31.1)	28(23.5)	29(19.3)	33(22)	19(12.7)	20(13.3)	312(81.3)
Positive	6(5.2)	3(2.6)	2(1.7)	2(1.7)	6(5)	2(1.7)	2(1.7)	22(14.7)	12(8)	7(4.7)	8(5.3)	72(18.8)

NB: *W/Seyo = Were Seyo, C/ Michael = Choli Michael, L/chirecha = Lalo Chirecha, W/Arsema = Werego Arsema, WA =Waligalte Adda, T/W = Tola Wakayo*

Table 4. Prevalence of donkey trypanosomosis based on sex, age, colour and body conditions of animals

Respective Variables	N°. of examined	N°. of positive	Prevalence Rates (%)	P-value	Odd Ratio	95% CI	
						Lower	Upper
Age							
Adult	289	61	21.1	0.042	2.04	1.03	4.07
Young	95	11	11.6	-	-	-	-
Sex							
Male	164	45	27.4	0.000	2.70	1.593	4.588
Female	220	27	12.3	-	-	-	-
Body condition							
Poor	78	42	53.8	0.000	1.11	1.06	2.23
Medium	155	13	8.4	0.400	1.39	.65	2.96
Good	151	17	11.3	-	-	-	-
PCV							
Anaemic	51	27	52.9	0.000	7.20	3.82	13.56
Non-anaemic	333	45	13.5				
Colour							
Black	100	15	15	0.27	1.42	0.77	2.65
Dalecha	284	57	20. 1	-	-	-	-
Total	384	72	18.8	-	-	-	-

However highest infection was seen in animals with poor body condition. The analysis revealed that, there was a significant association was observed between the disease and poor body conditioned donkeys in the study areas (p<0.05) (Table 3).

Parasitological Findings

A total of 384 donkeys were examined and 72 (18.8%) of them were found to be positive for different species of Trypanoma parasites like *Trypanosoma congolense, T. vivax, T. brucei* and some mixed parasites were detected. Out of these infected animals, 19 (26.4%) of them were found to be infected by *T. congolense*, 27 (37.5%) donkeys by *T. vivax,* 17 (26.1%) donkeys by *T. brucei* and 10 (13.9%) of them were found to be infected by mixed parasites. This shows that, *T. vivax* was the predominant species encountered Trypanoma parasites and followed by *T. congolense.* The distribution of different species of Trypanosoma parasites were detected in which highest number was recorded in Waligalte Adda PA of Nedjo district where as it was lower in Gimbi 01 and Lalo Chirecha PAs of Gimbi district and Werego Arsema and Enango 01 PAs Lalo Assabi district.

The following figure indicates the distribution of trypanosome species in the area.

Hematological Findings

Out of the observed animals, 72 of them were positive and overall mean PCV values were 21.2% and 43.2 for infected and non-infected animals, respectively. From the obtained results of overall mean PCV values of infected and non-infected animals, there is statistically significant difference (P<0.05). In this study, mean PCV were found to be statistically significant (P<0.05) in Gimbi and Lalo Assabi districts as compared with Nedjo district. In some of the tested PAs (Werego Arsema and Enango 01), there was statistically significant difference (P<0.05) as compared with other PAs.
Also, the result showed that there was statistically significant difference (P<0.05) between sexes and ages of animals in which male donkeys were more anaemic. The mean PCV was also found to be dependent on body condition of the donkeys in which poor and medium body conditioned donkeys have significantly lower (P<0.05) PCV value than body conditioned donkeys.

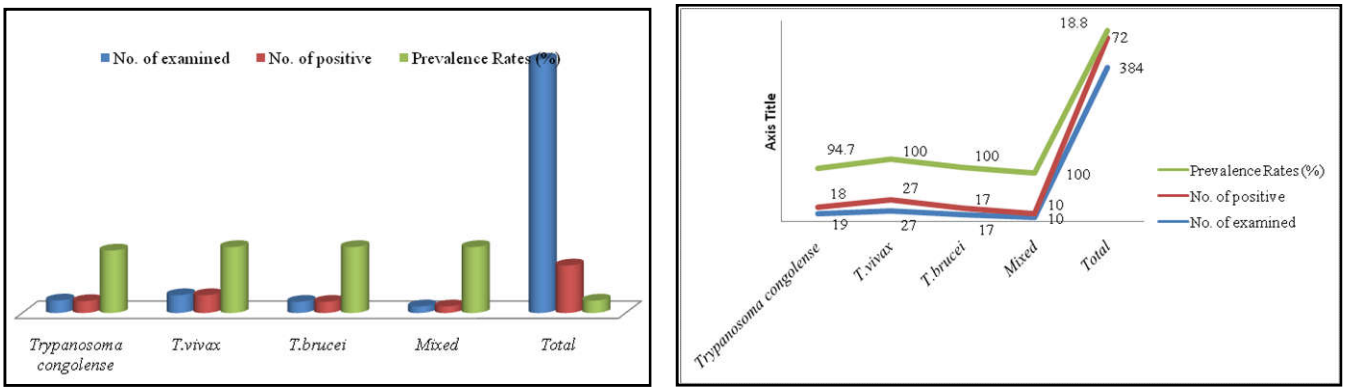

Figure 5. Parasitological findings and species identification

Table 5. Hematological Findings based on different Respective Variables

Respective Variables		N°. of examined	N°. of positive	Prevalence Rates (%)	Mean PCV (%)	P-value
Districts	Gimbi	115	13	11.3	36.8	.000
	Lalo Assabi	119	10	8.4	37.4	.000
	Nedjo	150	49	32.7	45.5	-
Peasant Associations,	Were Seyo	41	6	14.6	40.6	0.16
	Choli Michael	29	3	10.3	34.1	0.09
	Gimbi 01	25	2	8	37.2	0.07
	Lalo Chirecha	20	2	10	34.3	0.13
	Tosio	50	6	12	38.8	0.08
	Werego Arsema	39	2	5.1	37.5	0.02
	Enango 01	30	2	6.7	35.2	0.04
	WA	51	22	43.1	45. 9	0.20
	Nedjo 03	45	12	26.7	48.3	0.86
	Nedjo 02	26	7	26.9	44.2	0.89
	Tola Wakayo	28	8	28.6	41.5	-
Age	Adult	289	61	21.1	51.1	0.042
	Young	95	11	11.6	40.5	-
Sex	Male	164	45	27.4	38.6	0.000
	Female	220	27	12.3	41.9	-
Body condition	Poor	78	42	53.8	34.6	0.000
	Medium	155	13	8.4	37.2	0.400
	Good	151	17	11.3	47.0	-
Trypanosome infection	Infected	51	27	52.9	32.6	0.000
	Non-infected	333	45	13.5	42.3	-
Colour	Black	100	15	15	36.2	0.27
	Dalecha	284	57	20. 1	42.0	-
Overall PCV	Anaemic	51	27	52.9	21.2	0.000
	Non-anaemic	333	45	13.5	43.2	-
	Total	384	72	18.8		

DISCUSSIONS

The study was conducted from October 2015-September 2016 in purposively selected three districts (Gimbi, Lalo Assabi and Nedjo) of West Wollega Zone, Western Oromiya Regional State. The aims of this study were to estimate the prevalence of donkey trypanosomosis and to identify the trypanosomes species involved and their relationship with the mean PCV. From a total of 384 examined donkeys, overall of 72 (18.8%) donkeys were found to be infected by different species of trypanosoma parasites in the study areas. The study comprised peasant associations (PAs), sex, age, packed cell volume (PCV), Color, Species of parasites and body conditions of animals as a major factors those plays a role for the infection of trypanosoma parasites in the study areas. The overall prevalence rate of donkey Trypanosomosis during this study was 18.8% and different prevalence rates were also recorded as 11.3%, 8.4% and 32.7% in Gimbi, Lalo Assabi and Nedjo districts, respectively. The result revealed that, highest prevalence of trypanosoma infection was recorded in Nedjo district. It was in agreement with the finding of Abebe and Wolde (2010), which was a statistically significant difference in prevalence of trypanosoma infection between the two districts. This might be attributed due to donkeys are the major type of pack animals and often travel long distances crossing a high tsetse and other mechanical vector's challenge areas. When these animals are required for transportation, especially during the day time when fly activity is high, they are more exposed to either tsetse or other blood sucker flies. The overall prevalence rate of the disease during this study was supported by the previous reports of 18.2%-28.5% in different district of southern Ethiopia (Birhanu *et al*., 2006).

The result, especially that of Lalo Assabi district, was comparatively in line with the prevalence of donkey Trypanosomosis (6%) in Assosa district, Benishangul Gumuz Regional State, Northwest Ethiopia which was reported by Assefa *et al.,* (2015) and 6.30% in Assosa and Homosha districts of Benishangul Gumuz, Northwest Ethiopia (Abebe and Wolde, 2010). This might be due to the approach of agro-ecological borders of the areas in which the infected animals might have a chance to get in to this district. It is also supported by the report of Solomon *et al.,* (2010), who conducted a cross-sectional study of equine trypanosomosis and its vectors in Wolayta Zone, Southern Ethiopia, in which out of the 214 donkeys examined, 23 animals (10.7%) were found to be infected with different *trypanosome* species. But, the result was higher than the report of Hailegebrael and Shimelis, (2012) with a prevalence rate of 1.16%, 3.42%, 1.15% and 0% in Abadera, Jawi, Chara, and Dangla districts respectively. The prevalence of trypanosome infection in equine is low because they are not often the preferred host for tsetse as a result less exposed to the challenge as compared to cattle that normally graze over long distance (Radostits *et al*., 2007). Also different prevalence rates of the disease were observed among 11 PAs of the three districts with 14.6%, 10.3%, 8%, 10%, 12%, 5.1%, 6.7%, 43.1%, 26.7%, 26.9% and 28.6%, in Were Seyo, Choli Michael, Gimbi 01, Lalo Chirecha, Tosio, Werego Arsema, Enango 01, Waligalte Adda, Nedjo 03, Nedjo 02 and Tola Wakayo, respectively. The study revealed that, highest (43.1%) and lowest (8%) prevalence of the disease was recorded in Waligalte Adda PA of Nedjo district and Gimbi 01, respectively. However, prevalence of donkey trypanosomosis was statistically significant ($p<0.05$) in only two PAs (Werego Arsema and Enango 01) of Lalo Assabi district (Table 1). The observed differences in prevalence among 11 PAs of the three districts cannot be methodological differences but might be due to mechanical transmission of the disease in the areas. This might be most likely associated with the absence of tsetse flies in most PAs of this study. But, the result of this finding is not supported by the study of Sinshaw *et al.,* (2006) in three districts bordering Lake Tana in Amhara Regional State where no trypanosomes were detected. Analysis of age wise prevalence of donkey trypanosomosis between two age groups indicated that almost there was a difference in prevalence rate between these age groups that was 21.1% and 11.6% in adult and young, respectively. This result revealed that, higher prevalence rate was observed in adult than young age groups. This result indicated as adult age groups were more likely to be affected by the disease with two times as compared to that of young animals (OR= 2.04; 95% CI:1.03-4.07; P=0.042). Generally, age showed a significant association with the prevalence of donkey trypanosomosis in the areas ($p<0.05$). This finding was also in line with the report of Abebe and Wolde, (2010) in which all age of the donkeys above 2 years were infected.

The prevalence rate of the disease on the sex basis (female and male) was studied in which the majority were females (57.3%) while about (42.7%) of them were males. But, comparatively higher prevalence rate was recorded in male donkeys (27.4%) than in females (12.3%). This result indicated as male animals were more likely to be affected by donkey trypanosomosis with almost three times as compared to that of female donkeys (OR= 2.70; 95% CI: 1.59-4.59; P = 0.000). Generally, sex showed a significant association with the occurrence of major livestock diseases in the areas ($p<0.05$). This result is supported by the finding of Frehiwot and Samson, (2010) who reported male donkeys were more prevalent (6.9%) than females (4.9%). The result of this finding also in agreement with the previous studies at different altitudes in North Omo administrative region, conducted by Wondale (1993). But, this finding was not supported by the report of Abebe and Wolde (2010) who reported the absence of significant ($P>0.05$) variation was seen between male and female donkeys. Based on body condition scores (BCS) of the animals, donkeys were grouped into three as poor, medium and good body conditioned animals. Animals with poor BCS had the highest prevalence (53.8%) as compared with those with medium (8.4%) and good (11.3%). The analysis revealed, a significant association was observed between the occurrence of disease and poor body conditioned donkeys in the study areas ($p<0.05$). The result of this finding is also supported by the report of Solomon *et al.,* (2010), who conducted a cross-sectional study of equine trypanosomosis and its vectors in Wolayta Zone, Southern Ethiopia and also in line with the finding of Abebe and Wolde (2010).

This might be a donkeys having poor BCS shows as they are not in good nutritional management and a relatively high probability to be infected with chronic hemoparasites those includes Trypanosoma species. In this study, both the occurrence of trypanosome infection and mean PCV were found to be dependent on the body condition score of the animals ($P>0.05$) in which poor and medium body conditioned donkeys have significantly lower ($P<0.05$) with the PCV value than body conditioned donkeys. It was observed that poor body conditioned donkeys had significantly higher prevalence and lower mean PCV than those in good body condition. The result of this finding is supported by reports recorded in other equine

trypanosomosis related studies conducted by different authors. It has been also indicated that trypanosome infection causes a progressive loss of condition, weight loss, and the animals become easily exhaustive (Seifert, 1996). A total of 72 (18.8%) donkeys were found to be positive for different species of Trypanoma parasites like *Trypanosoma congolense, T. vivax, T. brucei* and some mixed parasites. About 19 (26.4%) of them were found to be infected by *T. congolense*, 27 (37.5%) donkeys by *T. vivax,* 17 (26.1%) donkeys by *T. brucei* and 10 (13.9%) of them were found to be infected by mixed parasites. This shows that, *T. vivax* was the predominant species encountered Trypanoma parasites and followed by *T. congolense*. This might be due to the presence of high population of biting flies (Tabanus, Stomoxys) those responsible to mechanically transmit these parasites from infected to the normal during blood sucking. The distribution of different species of Trypanosoma parasites were detected in which highest number was recorded in WA PA of Nedjo district where as it was lower in Gimbi 01 and Lalo Chirecha PAs of Gimbi district and Werego Arsema and Enango 01 PAs Lalo Assabi district. This might be increased due to exposing the donkeys to these biting flies, especially, at a day time in which the flies become active and awareness of the owners to treat their animals. It is in agreement with previous reports of Kanchula and Abebe (1997) and Yimam (1993) in which *Trypanosoma vivax* was reported to be the predominant species. However, the result was not in line with the finding of Frehiwot and Samson, (2010) who reported *T. congolense* (68.75%) was the most prevalent species followed by *T. brucei* (31.25%) and with that of Assefa and Abebe (2001) and Shelima *et al*., (2006b). Out of the observed animals, 72 of them were positive and overall mean PCV values were 21.2% and 43.2% for infected and non-infected animals, respectively. From the obtained results of overall mean PCV values of infected and non-infected animals, there was statistically significant difference ($P<0.05$). In this study, mean PCV was found to be statistically significant ($P< 0.05$) in Gimbi and Lalo Assabi districts as compared with Nedjo district. It was supported by the prevalence of trypanosome infection which was significantly higher in Asosa than Homosha district. In some of the tested PAs (Werego Arsema and Enango 01), there was statistically significant difference ($P<0.05$) as compared with other PAs. Also, the result showed that there was statistically significant difference ($P<0.05$) between sexes and ages of animals in which male donkeys were more anaemic. This finding is consistent with previous reports of Abebe and Wolde, (2010), from Gambia (Dhollander *et al*., 2006) and many other literatures (Urquhart *et al*., 1996; Secka 2003). Different researchers have been reported detection of anemia (lowered PCV) in trypanosome infected donkeys around the globe (Dhollander *et al*., 2006; Shelima *et al*., 2006a; Pinchbeck *et al.,* 2008).

However, the fact that other diseases of parasitic origin could also produce anemia posed difficulty to associate the low PCV observed in this study with trypanosomosis. Moreover, the study donkeys were not screened for gastrointestinal or hemoparasites during the study period. It is, therefore, essential that other anemia producing parasites are identified and their effects known in order to assess the net effect of trypanosomosis on PCV. Contrary to other studies (Dhollander *et al*., 2006) that found no age specificity, in this study trypanosome infection was observed exclusively in donkeys above 2 years of age. The reason for the absence of trypanosomes in the younger donkeys is not clearly known whether related to natural immunity or some other factors and this require further investigation.

Conclusion and Recommendations

The study was conducted in Gimbi, Lalo Assabi and Nedjo districts of West Wollega Zone; to estimate the prevalence of equine trypanosomosis and identify the trypanosomes species involved and their relationship with the mean PCV. The result of analysis indicated that different prevalence rate with 18.8% overall prevalence of donkey trypanosomosis was recorded in the study areas. Generally, the results suggested that trypanosome is an important disease of donkeys in the study areas where donkeys are extensively kept and used. The prevalence of donkey trypanosomosis in Nedjo district was higher than the prevalence of the disease in Gimbi and Lalo Assabi districts. Different species of Trypanoma parasites like *Trypanosoma congolense, T. vivax, T. brucei* and some mixed parasites were observed. *Trypanosoma vivax*, which can be transmitted mechanically, was the predominant species Trypanoma parasites and followed by *T. congolense*. The distributions of different species of these parasites were detected in which highest number was recorded in WA, but lower in Gimbi 01 and Lalo Chirecha, Werego Arsema and Enango 01 Peasant Associations. The overall mean PCV values were 21.2% and 43.2% for infected and non-infected animals, respectively. As obtained from the result, PCV is negatively impacted by the disease which resulted in reduction of working ability of the donkeys in the study areas.

Based on the above conclusion, the following recommendations were forwarded:

- The medication equipments should be sterilized before use for each animal.
- The working performance of the donkeys should be improved by minimizing the burden of disease by treating the patient animals.
- Donkey owners should not expose their animals to the biting flies, especially, at a day time in which the flies become active
- Entomological survey needs to be conducted in different seasons and agro-ecological zones in the areas to generate a complete data of the disease.
- More sensitive techniques such as serology or PCR should be used for the effective diagnosis of the disease.

Conflict of Interests

All authors have declared that no competing interests exist.

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