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Meta-analysis of camel trypanosomosis in Ethiopia

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Abstract

Camel is animal of great economic contribution to pastoralists and endowed with prestigious social value. Camels play significant role in the livelihood of the pastoralists and agro-pastoralists living in the fragile environments. The main purpose of this meta-analysis was to compute the pooled prevalence estimate of camel trypanosomosis based on the available studies. Published articles on camel trypanosomosis were searched in PubMed, Google scholar and African journals online (AJOL). The key electronic search words were: camel, trypanosomosis, *Trypanosoma evansi,* and surra.The preliminary screening of the articles was based on the title and abstract. The analysis was based on 11 cross-sectional study reports, which were done between the year 2008 and 2016, and in four administrative regions of Ethiopia. The pooled prevalence estimate in a random effects meta-analysis was 9.2% (95% CI 7.1- 11.8). Enormous heterogeneity were noted among the studies (I^2 = 94.4%). In subgroup and multivariable meta-regression analyses however, only two predictors namely sample size and diagnostic techniques were found to have significant effect ($p < 0.05$). Accordingly, the aforementioned predictor sex explained 51.8% of the explainable proportion of the heterogeneity noted between studies. In this regard, serological and molecular based reports were noted to have higher prevalence compared to wet smear and buffy coat. On the other hand, studies with smaller sample size had less prevalence compared to large sample size. *Trypanosoma evansi* is the only species identified and reported in all the study reports. This analysis results underscores the need for further study that involve more sensitive diagnostic techniques to reveal the precise magnitude of the disease, and to identify the vectors in all camel rearing areas of the country.

Keywords: Camel; Ethiopia; Hetrogenity; Meta-analysis; Pooled prevalence; Trypanosoma evansi

Introduction

The camel population of Ethiopia is estimated to be about 979,318 heads. Only one species of camel that is *Camelus dromedarius* is found in Ethiopia. Its distribution coincided with that of the drylands, which means semi-arid and arid regions of southern, eastern and North eastern parts of the country (Tefera and Abebe, 2012; Simenew *et al*., 2013). Camels played significant role in the livelihood of the pastoralists and agro-pastoralists living in the fragile environments. They are the major sources of milk, meat, transportation and draught power, and income for pastoral community (Tefera and Abebe, 2012).

Inspite of camels' great economic contribution for pastoralists and high prestigious social value, it is not properly utilized due to traditional management systems, pressure of multiple changes in the production, environment and various camel diseases (Mehari *et al*., 2007; Tefera and Abebe, 2012). Among many diseases that affect camels in Ethiopia, trypanosomosis (Surra) is the most important parasitic diseases (Demeke, 1998; Tekle and Abebe, 2001). It is widely distributed throughout camel rearing areas of the country, and causes considerable economic loss mainly due to decline in camel productivity. Therefore, the purpose of this review was to compute a pooled prevalence estimate of camel trypanosomosis at national level, and to identify the most important predictors that could contribute to the heterogeneity between the reports. Moreover, it was to point the information gap on the problem in camel rearing areas of Ethiopia.

Materials and methods

Study protocol

A Systematic reviews and Meta-Analyses guidelines described by Moher *et al*. (2009) were employed during review process. It includes a template for literature search with predefined inclusion and exclusion criteria, in addition to a quality assessment format and data extraction template.

Literature search method

The method employed for the literature search was electronic search, which was done by data bases including, PubMed, Google scholar and African journals online (AJOL). The key words for electronic search include: camel, trypanosomosis, *T. evansi,* and surra. The primary screening was done based on

title and abstract to see the compliance level with review objectives. Those reports that met the primary requirement were subjected to second steps of screening, where reports were fully scanned at a closer range. The specific criteria for the articles to be considered were design of the study, laboratory procedure, analysis of data and presentations of the result.

Inclusion and exclusion criteria

The quality of each article was assessed by reviewing study objective, design, data analysis and presentation, and conclusions. Accordingly, those articles whose qualities were rated moderate to high were accepted for subsequent data extraction, which include data on apparent prevalence, sample size, diagnostic test used and administrative region. The specific inclusion criteria include: publication year after 2000, design of the study, random sampling of the study population, clarity of result presentation and the laboratory methods used to identify infected camels.

To be eligible, the following inclusion criteria were used: a study had to be (i) published in a reputable journal, (ii) written in English, (iii) cross-sectional study and (iv) conducted in Ethiopia (v) number of infected animals, size of study population and test method available (vi) published as of 2000.

Data extraction

The data extracted included: year of publication, study area (i.e. administrative region and district), diagnostic method, sample size, number positive, number negative and prevalence. The 95% confidence intervals of the point estimates were computed.

Data analysis

The statistical software used in the analysis was STATA 12.1 version (Statacorp 4905 Lakeway Drive College Station). Prevalence estimates were logit-transformed using the formula: lp = ln [p/(1 − p)], where lp = the logit event estimate; ln = the natural logarithm; p = study level estimate. The variance of the logit estimate was computed by using the following formula: v(lp) = 1/(np) + 1/[n(1−p)], where v=variance and n=sample size. The standard error of log prevalence (SE) was also calculated using the formula: ln−p = Sqrt (1/sample − n×p×(1−p)). Log-transformation was performed to normalize the prevalence

distribution. Finally, the pooled prevalence estimate was computed using the formula: $p = 1/(1+e-lp) \times 100$, where "e" is the base of natural logarithm.

Random effects meta-analyses of the described outcome were performed using the method of DerSimonian and Laird. The estimate of heterogeneity was taken from the inverse-variance of the random-effect model using the metan command in Stata (Borenstein *et al*., 2009; Dohoo *et al*., 2009; Sterne *et al*., 2009). The metan command in Stata generates an estimate of the Cochran's Q statistic which tests for differences in effect sizes across studies, an estimate of the variance of effect sizes between studies (τ^2), and Higgins I^2 (hereafter denoted I^2) which is an estimate of the proportion of the observed variance that reflects true differences in effect size (Borenstein *et al*., 2009; Sterne *et al*., 2009).

Results

Literature search results

Five hundred eighty nine reports were retrieved using aforementioned key words and databases, of which 545 were rejected due to title and host species. Thirty two of the reports were excluded due to absence of prevalence data, and one article was rejected due to year of publication, before 2000. Finally, a total of 11 artciles were considered for systemic review and meta-analysis of camel trypanosomosis. All these published articles were peer reviewed, and published in 2008-2016. From the 11 eligible articles, 34 animal level reports were extracted at administrative region level, breed of camel and type of the diagnostic methods employed for the study. Regarding the diagnostic tests, seven, fifteen, eight and four of the study reports were based on Giemsa staining, BCT (Buffy Coat Technique), serological tests and molecular (i.e. CATT/*T. evansi* and RoTat 1.2ITL), respectively (Table 1). Flow diagram to show the eligible study reports selection is shown on Figure 1.

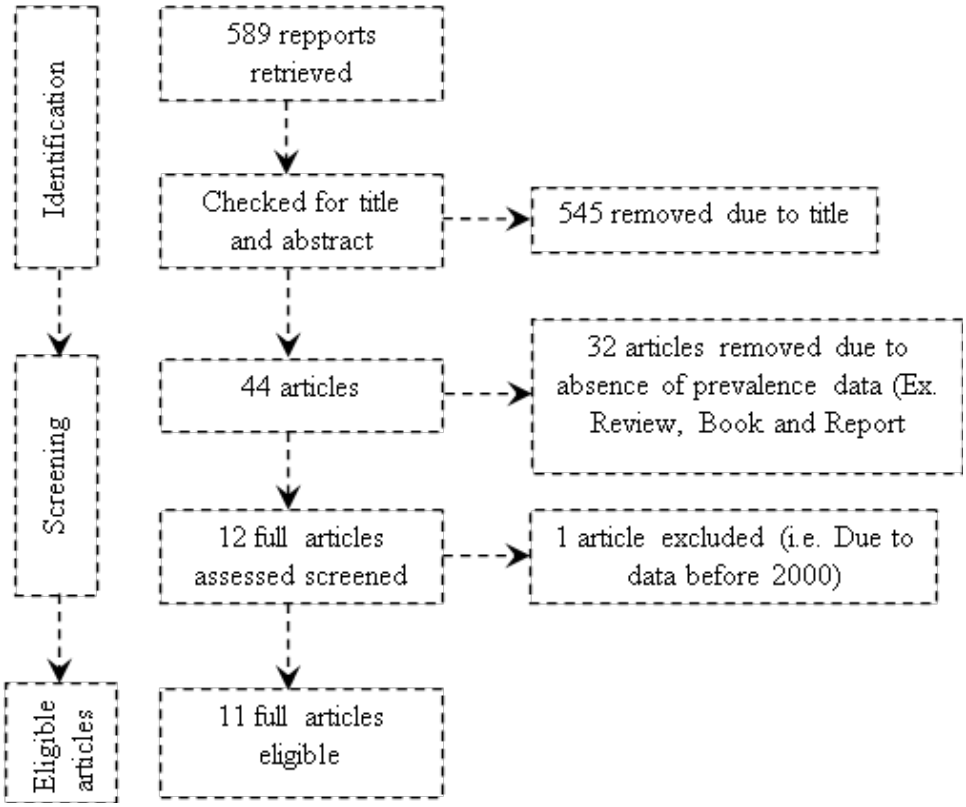

Figure 1. Literature selection flow diagram for systemic review of camel trypanosomosis

Characteristics and quality of the reports

The studies were conducted between 2008 and 2016 in four administrative regions, namely: Afar, Somali, Oromia and Tigray. The study districts are shown on Figure 2. The sample size ranged from 91 to 692 (Table 1). Quantitative data on breed, administrative region, year and diagnostic techniques were extracted from selected articles. From each articles, the estimated prevalence, sample size and number positives were retrieved. Cross-sectional study design was employed by all the studies considered for the review, and sampling was reported to have been done randomly. The diagnostic procedures used were Giemsa staining, BCT, serology and molecular tests. The total number of camels involved in the reviewed articles was 10,992.

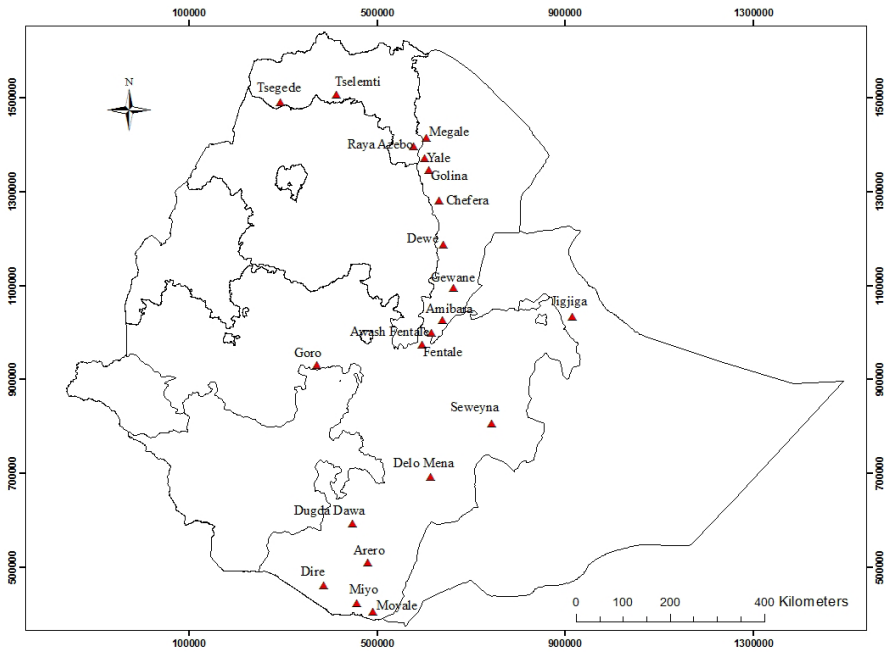

Figure 2. Map of Ethiopia to show areas from where camel trypanosomosis was reported

Table 1. Reports of camel trypanosomosis that used in the review and meta-analysis

Author	Breed	Admin. region	Dx technique	Sample size	App (%)
Abdukadir *et al* (2015)	Somali	Oromia	Gimsa stain	384	10.2
Olani *et al* (2016)	Somali	Oromia	BCT	449	0.2
Olani *et al* (2016)	Somali	Oromia	BCT	462	2.8
Olani *et al* (2016)	Somali	Oromia	BCT	503	4.2
Olani *et al* (2016)	Somali	Oromia	BCT	294	4.4
Olani *et al* (2016)	Somali	Oromia	BCT	692	1.3
Abera *et al* (2014)	Somali	Oromia	BCT	125	15.2
Abera *et al* (2014)	Somali	Oromia	BCT	91	9.9
Abera *et al* (2014)	Somali	Oromia	BCT	176	23.9
Hagos *et al* (2009)	Somali	Oromia	BCT	319	16.3

Author	Breed	Admin. region	Dx technique	Sample size	App (%)
Hagos *et al* (2009)	Somali	Oromia	Serology	319	30.7
Hagos *et al* (2009)	Somali	Oromia	BCT	300	7.7
Hagos *et al* (2009)	Somali	Oromia	Serology	300	18.7
Bogale *et al* (2012)	Somali	Oromia	Gimsa stain	395	18.2
Lemecha *et al* (2008)	Afar	Oromia	Gimsa stain	637	5.0
Lemecha *et al* (2008)	Afar	Afar	Gimsa stain	376	5.6
Kassa *et al* (2011)	Afar	Oromia	BCT	150	4.7
Kassa *et al* (2011)	Afar	Oromia	Gimsa stain	383	4.4
Tadesse *et al* (2012)	Somali	Somali	BCT	362	3.9
Fikru *et al* (2015)	Afar	Afar	Gimsa stain	199	2.0
Fikru *et al* (2015)	Afar	Afar	Serology	199	21.1
Fikru *et al* (2015)	Afar	Afar	Molecular	199	20.6
Fikru *et al* (2015)	Afar	Afar	Gimsa stain	200	2.0
Fikru *et al* (2015)	Afar	Afar	Serology	200	27.5
Fikru *et al* (2015)	Afar	Afar	Molecular	200	21.5
Birhanu *et al* (2015)	Afar	Afar	BCT	411	4.6
Birhanu *et al* (2015)	Afar	Afar	Serology	411	15.6
Birhanu *et al* (2015)	Afar	Afar	Molecular	411	13.4
Birhanu *et al* (2015)	Afar	Tigray	BCT	343	3.2
Birhanu *et al* (2015)	Afar	Tigray	Serology	343	11.4
Birhanu *et al* (2015)	Afar	Tigray	Molecular	343	9.6
Weldegebrial *et al* (2015)	Afar	Afar	BCT	208	2.9
Weldegebrial *et al* (2015)	Afar	Afar	BCT	200	7.5
Weldegebrial *et al* (2015)	Afar	Afar	Serology	208	17.8
Weldegebrial *et al* (2015)	Afar	Afar	Serology	200	30.0
Overall					9.2

Dx=Diagnostic, APP=Aparent prevalence

Trypanosoma species and vectors

Trypanosoma evansi is the only species reported as a cause for camel trypanosomosis in all study reports. The following mechanical vectors were reported from two districts, namely: Fentale and Gewane from Oromia and Afar regions,

respectively: *Stomoxys*, *Tabanus*, *Lyperosia* and *Hippobosca* (Lemecha *et al*., 2008; Kassa *et al*., 2011). It is reported from different areas that *Stomoxys* and *Tabanus* are the most important haematophagus flies that can transmit *T. evansi* (Taylor and Authié, 2004; Radostits *et al*., 2007).

Meta-analysis

The logit-transformed data of the reports were used to run meta-analysis in a random effect model. The raw and logit-transformed effect size distribution is shown on Figure 3. Accordingly, the estimated pooled prevalence of trypanosoma infection in camels was 9.2% (95% CI: 7.1, 11.8). In a random effect model, the calculated Cochran value (Q) was 590.59 (df=33 and p=0.000). The effect size and respective weight of each eligible study report in the review is also presented on the forest plot (Figure 3). The estimate of between study variance (τ^2) was 0.64. The variation in effect size attributed to heterogeneity (ι^2) was 94.4%. The 95% CI of the prevalence estimates along with measure of heterogeneities between study reports for each region, breed and diagnostic test are shown in Table 2.

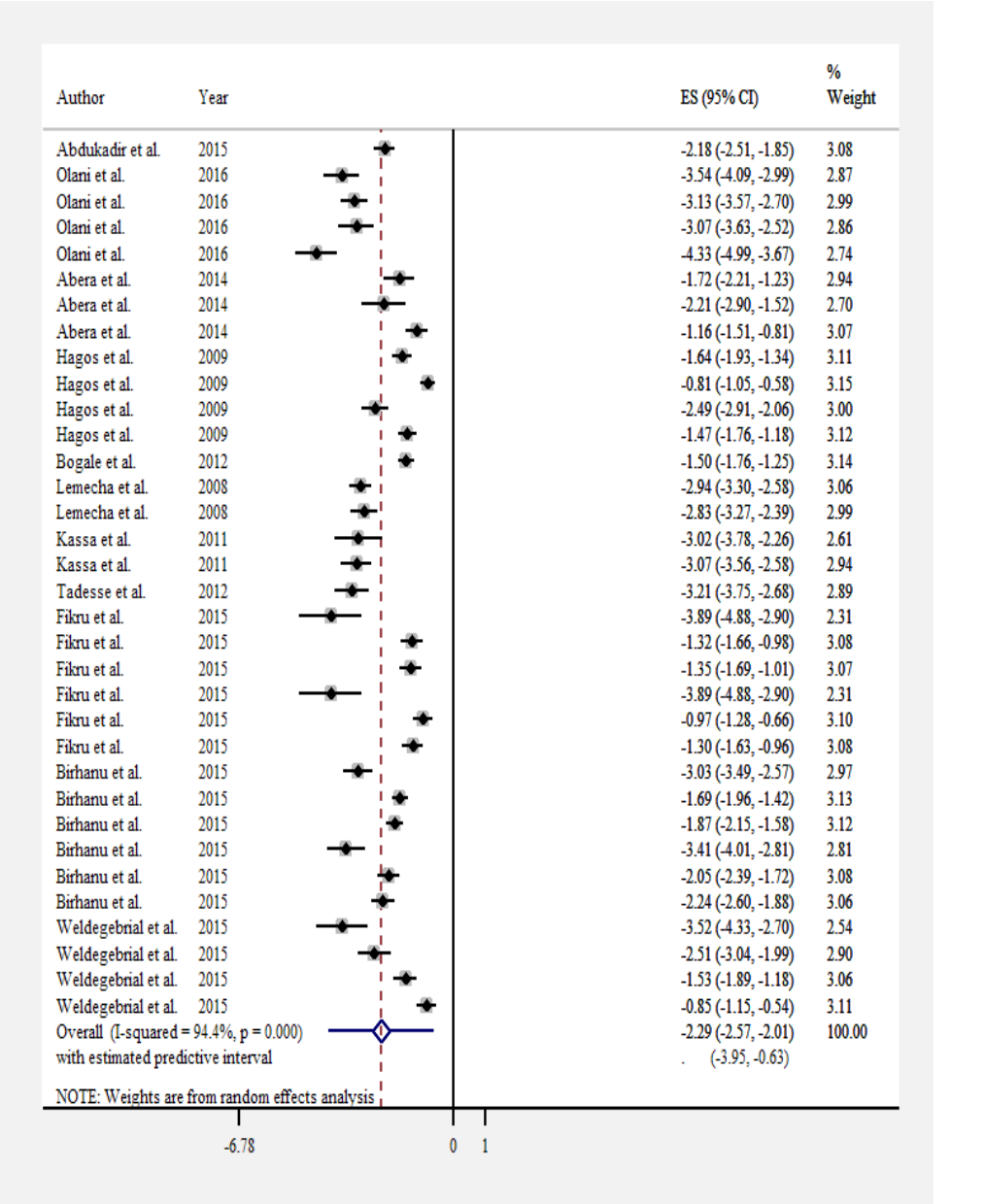

Figure 3. Forest plot of logit-prevalence estimates of camel trypanosome infection in Ethiopia

Table 2. Proportion of the between study variance explained (R^2) by each variable considered in meta-regression on prevalence of camel trypanosomosis in Ethiopia.

Variables	Without sample size		With sample size	
	R^2	*P*-value	R^2	*P*-value
Year	0	0.494	20.11%	0.267
Sample size	19.53%	0.012	-	-
Breed	0	0.960		0.655
Diagnostic technique	35.11%	0.000	51.82%	0.000
Administrative region	0	0.967	16.95%	0.672

Sub-group analysis

The sub-group analysis was made for three potential predictor categories, which include: breed, diagnostic techniques and regional states. The pooled prevalence estimate for both Afar and Somali breed was 9.2%. In both breeds and regional state the observed difference between the respective categories were statistically not significant ($p > 0.05$). But statistically significant difference was observed between the categories of the diagnostic techniques employed for the studies ($p < 0.05$). The diagnostic techniques encountered in the reports were Giemsa staining, BCT, serology and molecular tests with prevalence estimates of 93%, 92.4%, 88.3% and 84.7%, respectively (Figure 4). On the other hand breed was dropped due to multi-collinearity with regional states (gamma = 0.87) and diagnostic techniques (0.66). Since, the number of reports from Somali regional state was few; the report was not included in the region based subgroup analysis.

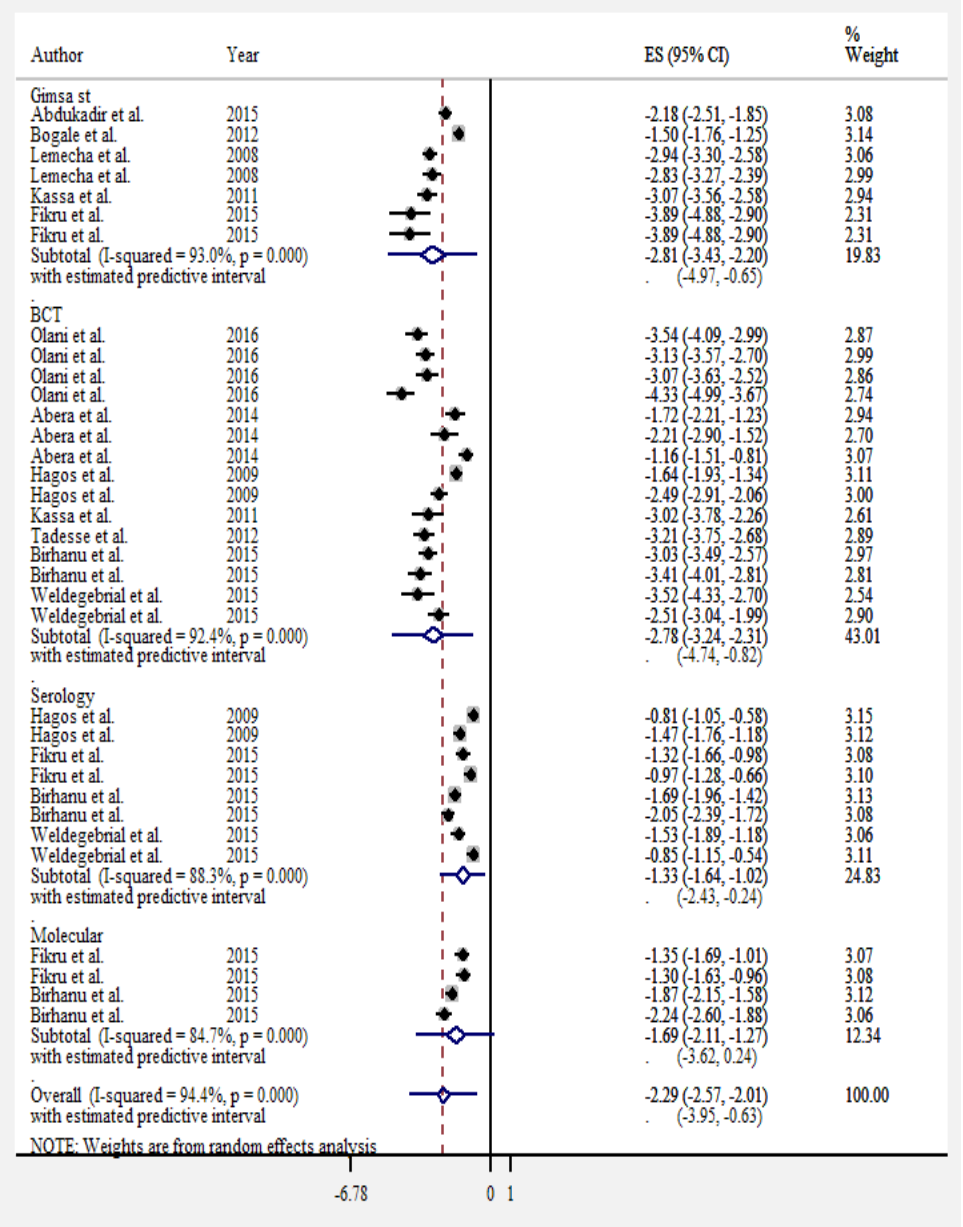

Figure 4. Forest plot for diagnostic test based logit-prevalence estimate of camel trypanosomosis

Univariable and multivariable meta-regression

The logit-transformed data of the reports were used for univariable meta-regression to appreciate the proportion of individual predictor's effect on the heterogeneity between groups. The analysis was done with and without controlling for sample size in line with each predictor and the negative R^2 values were set to zero (Borenstein *et al*., 2009). Among the predictors considered in the univariable meta-regression analysis; sample size and diagnostic techniques had significant effects on the observed variation between the studies (Table 2). Then, the second step was multivariable meta-regression to estimate in between study variance explained by significant predictors fitted to the model. The two qualifying predictors, namely: sample size and diagnostic tests were fitted into the multivariable meta-regression model (Table 3). The sample size was inversely related to the prevalence of camel trypanosomosis as shown on the buble plot (Figure 5). The proportions of predictor's effect size on the heterogeneity between study reports recorded for diagnostic tests and sample size were R^2=35.1% and 17.0%, respectively. Finally, the diagnostic techniques and sample size fitted to the model were found to be significant ($p<0.05$), and accounted for 57.18% of the explainable proportion of the heterogeneity (R^2=94.4%). The observed between predictor's category, τ^2 unexplained was 0.3654, whereas, the τ^2 total value was 0.6437.

Table 3. Coefficients and *p*-values of the association on camel trypanosomosis prevalence in meta-regression model (R^2=57.18%, n=11 reports)

Variable	Predictors category/ range	n	Coeff.	*P*-value	Overall *P*-value
Sample size	91 – 692		-0.0023	0.017	0.012
Diagnostic test	BCT	16		Reference	
	Giemsa	7	0.16	0.625	
	Serology	8	1.34	<0.001	
	Molecular	4	1.02	0.010	<0.001

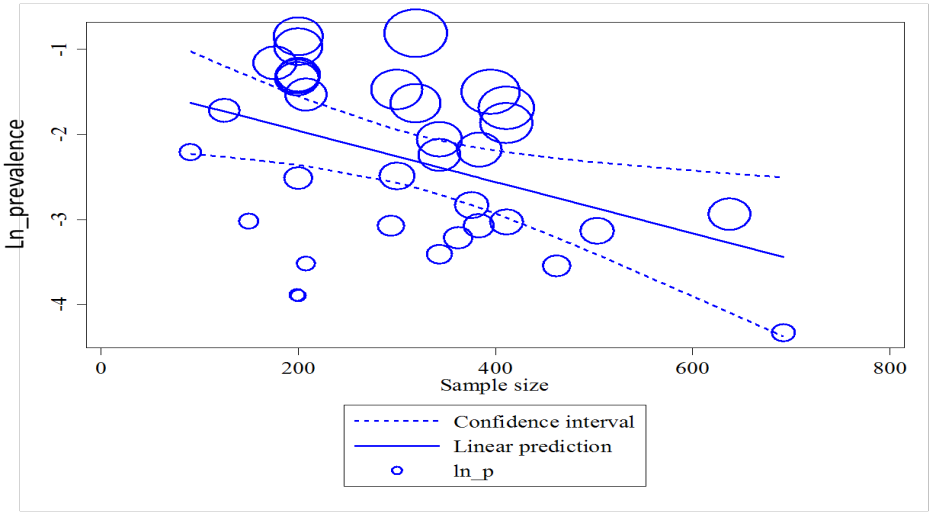

Figure 5. Buble plot to show the relationship between sample size and camel trypanosomosis

Bias assessment and sensitivity analysis

The depicted funnel plot (Figure 6) along with Begg's ($p=0.000$) and Egger's statistics ($p = 0.000$) revealed the presence of bias, which could be due to estimate precision linked to small sample size with large standard error. However, no single study was noted to influence the validity of the summary effect estimate (Influential plot not shown).

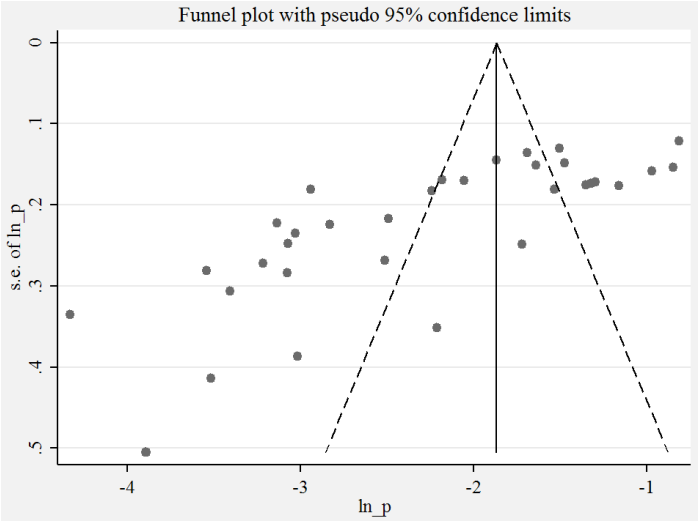

Figure 6. Funnel plot for logit-prevalence estimate of camel trypanosomosis

Discussion

This is the first systematic review and meta-analysis, quantitative review, attempt made on camel trypanosomosis in Ethiopia. All the studies of selected articles were done in pastoral areas of the country. In this analysis, a total of 11 cross-sectional studies that met the inclusion criteria were used, and resulted in 34 animal's level data set to calculate the prevalence of camel trypanosomosis. Moreover, it was used to investigate the level of heterogeneity among the available reports. The national level pooled prevalence estimate of camel trypanosomosis was found to be 9.2% (95% CI: 7.1-11.8). This pooled estimate is computed in consideration of the entire eligible studies, with prevalence range of 0.2 (Olani *et al*., 2016) to 30.7% (Hagos *et al*., 2009). Analytical approach for pooled estimate of the different study reports consider the existing of variance (ι^2) between the studies. Hence, the inverse variance square (ι^2) of this review was 94.4%, which revealed the presence of variation that attributed to real and high heterogeneity. Among the captured predictors, sample size and type of diagnostic test used in the studies resulted in a significant variation in pooled prevalence estimate. Of the total 94.4% heterogeneity, 57.2% was explained by sample size and diagnostic tests.

The diagnostic tests explained 35.1% of the explainable proportion of the heterogeneity that was observed between the study reports. The pooled estimate prevalence of serological (p <0.05, 95% CI=16.2-26.5) and molecular (p *<0.05*, 95% CI=10.8-22.0) tests were significantly higher (p *<0.05*) than Giemsa staining (95% CI=3.1-10.0) and BCT (95% CI=3.8-9.0). This observation is in agreement with the reports from various areas (Pathak *et al*., 1997; Ngaira *et al*., 2003; Njiru 2004; Singh *et al*., 2004; Abdel-Rady, 2006; Abdel-Rady, 2008). It is due to the higher sensitivity of serological and molecular tests compared to Giemsa staining and BCT. Serological and molecular tests able to detect low parasitaemic and chronic *T. evansi* infections of camel, which could not be detected by Giemsa staining and BCT methods. This is a confirmation and evidences that Giemsa staining and BCT methods under-estimate the prevailing prevalence of camel trypanosomosis in the areas.

About 19.5% of the proportion of between study variance was explained by the sample size. This is mainly due to the fact that in some of the studies, especially of sample size less than or equal to 200, the study animals and sites were selected purposively based on convenience and camel owners willingness. The prevalence in most of these reports is relatively higher. Generally, the higher the sample size, the lower was the prevalence of camel trypanosomosis. Studies used in this analysis did not cover all the camel rearing areas of the country. Moreover, they lack full information on the vector density, age and sex of the host animal, as well as, lack of consistent and sufficient data on some important factors like season and size.

Conclusion

The pooled estimate prevalence of camel trypanosomosis was higher; however, some degrees of variability seen between diagnostic methods used in the studies as well as sample size. Serological and molecular tests were found highly sensitive for the diagnosis of camel trypanosomosis. Therefore, these diagnostic methods could be adopted for the diagnosis of camel trypanosomosis. Finally, further study with more sensitive diagnostic tests to know the precise magnitude of the disease; and identification and quantification of the vectors in camel rearing, pastoral, areas of the country are recommended.

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Conflict of interest

The author declares that there is no conflict of interest.

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