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Prevalence Study of Bovine Tuberculosis and Genus Typing of its Causative Agents in Cattle Slaughtered at Dilla Municipal Abattoir, Southern Ethiopia

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Abstract: A cross-sectional study was conducted at Dilla Municipal abattoir from December, 2012 to June 2013 to investigate the prevalence, associated risk factors and to identify the causative agents of Bovine tuberculosis (bTB) in Gedeo (Dilla Municipal abattoir), Southern Ethiopia. Postmortem examination, mycobacterial culturing and Multiplex genus typing techniques were employed. An overall prevalence of 2.6% (20/768) was recorded upon detailed post mortem examination with the major tuberculous lesions (75%) localized in the thoracic cavity (respiratory tract). Among the different risk factors analyzed, age ($x2$=12.4; P =0.002) and body condition ($x2$=35.7; P<0.001) were associated with bTB infection. Animals that have medium (OR=0.08; 95% CI: 0.02-0.43) and good body condition (OR=0.03; 95% CI: 0.01- 0.13) were less likely to have tubeculous lesions than poor body conditioned animals. Mycobacterial culture result revealed that growth was seen in 15 % (3/20) of the bTB suggestive lesions. Further Genus typing of the three culture positive isolates showed a band size of 1030bp for the two that represent Non Tuberculosis *Mycobacterium* species (NTM). While one isolate did not show a signal at all to the genus *Mycobacterium*. The study signified the importance of NTM in the formation of tuberculous granulomatous lesions in cattle and their importance in the epidemiology of bTB. In conclusion, characterization of these isolates to specific species level and further investigation aiming at identification of the source of NTM infections, transmission routes, pathogenicity and their public health significance in the study area is recommended.

Key words: Abattoir · Bovine tuberculosis · Gedeo · Dilla · Genus typing · Risk factors · Ethiopia · Postmortem examination · Prevalence

INTRODUCTION

Bovine tuberculosis (bTB) is an infectious zoonotic disease [1] caused by *M.bovis*, member of the *Mycobacterium tuberculosis* complex (MTBC), which affects humans and many vertebrate animals and characterized by progressive development of granulomas in tissues and organs [2, 3].

bTB has been widely distributed worldwide, causing great economic loss in animal production and the most frequent cause of zoonotic TB in man [4]. Particularly in Africa, it represents a potential health hazard to both animals and humans, as nearly 85% of cattle and 82% of the human population live in areas where the disease is prevalent or only partially controlled [5]. Its epidemiology and public health significance remains largely unknown due to several factors [6].

bTB is considered to be a prevalent disease in cattle populations in Ethiopia. Abattoir inspection studies [7-10] indicated the widely occurrence of the disease in different regions of the country. Few studies have also indicated the zoonotic transmission of the disease to humans [11-14].

However, the status of the disease is not well established in livestock and most studies focused mainly on urban areas in central Ethiopia. In order to embark in a future national bTB control program, the epidemiology of the disease has to be assessed widely in different regions

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of the country. As epidemiological data of bTB related to the prevalence, principal risk factors for infection in cattle are lacking in Gedeo Zone, SNNPR and southern part of Ethiopia, this cross-sectional study was done in Dilla Municipal abattoir, Gedeo Zone of the Southern Ethiopia to investigate the causative agents, prevalence and associated risk factors of bTB in slaughtered cattle.

MATERIALS AND METHODS

Study Area and Animals: The study was conducted from December 2012 to June 2013 in Dilla municipal abattoir, Gedeo, Southern Nations Nationalities and Peoples Regional State (SNNPR) in Ethiopia. According to the available logistics and time, a total of 768 apparently normal animals slaughtered in the abattoir were included in the present study period. The animals used for the study were mainly originated from Gedeo Zone and other areas of SNNPR (Sidama and Wolaita-Sedo) and neighboring oromia region (Borena, Arsi).

Study Design, Sample Size and Sampling Strategy: A cross-sectional study was designed and a simple random sampling method was employed to sample cattle in the slaughter house. A total of 768 cattle were investigated for post mortem examination based on the available logistics and duration study period. All epidemiological data of 768 cattle including body condition, sex, breed and origin of the cattle were recorded during the antemortem examination.

Postmortem Examination: Detailed post-mortem inspection of each carcass was carried out according to [15, 16]. In the slaughterhouse, carcasses and offals were identified using the butcher number. All the lobes of the lung and associated lymph nodes were inspected, palpated and incision were made on the suspected organs. The animal was classified as lesioned (infected) when tuberculous lesion was found and if not as non lesioned (not infected). The lymph nodes and other tissue specimens such as the lungs, liver and kidneys were examined in detail during post-mortem under a bright light source. The lobes of the two lungs were inspected and palpated externally. Then, each lobe was sectioned into about 2-cm-thick slices to facilitate the detection of lesions. Similarly, lymph nodes were sliced into thin sections (about 2 mm thick) and were inspected for the presence of visible lesions.

Sample Collection and Transportation: Specimens were collected from the slaughtered animals under aseptic conditions by carefully removing from the carcass and placed in a 50 ml capacity of universal bottle with screw caps containing 5 ml of sterile 0.85% saline water and kept at -20^{0}C (frozen) in Dilla University Laboratory until it is transported to Aklilu Lemma Institute of Pathobiology (ALIPB) TB laboratory.

Isolation and Identification of Mycobacteria

Mycobacterial Culturing: All specimens collected from the abattoir were processed and prepared for mycobacterial culture. The specimens were digested and decontaminated, using 4% NaOH in order to initiate the release of mycobacteria organisms from body fluids and cells and reduce bacterial contaminants [17]. The procedure used for culturing of the tissues was as recommended by OIE. The tissue was first defatted and homogenised using a mortar and pestle, followed by decontamination with 4% NaOH, centrifugation at 3000 rpm for 15 minutes at room temperature. After the supernatant was discarded then neutralized using 10% HCl. Finally, the sediment was inoculated on to two Lowenstein-Jensen, one contain either pyruvate which favoring growth *M. bovis* or glycerol which favoring growth *M. tuberculosis*. Cultures were incubated at 37°C and followed up for a minimum of 8 weeks [18-20].

Genus Typing of Isolates: AFB positive isolates were heat-killed by mixing approximately 2 loopful of colonies in 200 ìl distilled H_2O followed by incubation at 80°C for 45 min. Following a standard procedure, multiplex Polymerase Chain Reaction (m-PCR) was used to confirm the presence of genus Mycobacterium in the isolate and to differentiate MTBC from *M. avium complex* and other mycobacterial species [21].

Data Collection, Management and Statistical Analysis: Data related to age, breed, body condition and origin of each animal were recorded on a data sheet during the ante mortem examination. Presence or absence of tuberculous lesions and affected tissue(s) were recorded on postmortem examination. The recorded data were entered into Microsoft Excel data sheets and analyzed using STATA 11 statistical software [22]. Descriptive statistics was used to determine the proportion of cattle carcass harboring tuberculous lesions. The range and frequency of anatomical sites with tuberculous lesions were recorded for each carcass examined. Logistic regression analysis was used to assess the association between

prevalence and animal risk factors using two stage processes; primarily; the analysis of all variable of interest and on the second stage to multivariable analysis using multiple regressions. The difference between the effects of different risk factors on prevalence was analyzed using the Pearson chi-square ($x2$) test. The odds ratio (OR) was calculated to assess the strength of association of different factors with the prevalence of bTB. A statistically significant association between variables was said to exist if the calculated P<0.05 and if 95% confidence interval (CI) in OR did not include 1.

RESULT

Prevalence and Associated Risk Factors Based on Detailed Post Mortem Inspection: Upon detailed post mortem examination of 768 animals, an overall 2.6% (20/768) prevalence of bTB was found in the study area; out of which 1.95% (15/768) of the cases were detected in male animals and 0.65% (5/768) of the cases were detected in female animals. The vast majorities were zebu (*Bos indicus*) breeds and the remaining 20% were their crosses. Of the risk factors considered (Table 1) age group ($x2$=12.4; P =0.002) and body condition ($x2$=35.7; P<0.001) was found significantly associated with bTB infection. Multivariable analysis showed that animals that have medium and good body condition (OR=0.1; 95% CI: 0.02-0.4; OR=0.03; 95% CI: 0.01- 0.1) were less likely to have tubeculous lesions than those with poor body conditioned animals respectively (Table 2).

Distribution and Location of Tubercular Lesions: The distribution and frequency of suspicious TB lesions in tissues of positive animals is presented in Table 3. Of the total lesions observed; 40% were localized in lung,

Table 1: The association of risk factors of animals with tuberculous lesions in cattle slaughtered in Dilla municipal abattoir, Gedeo zone, Southern Ethiopia using Chi-square test

Variable	Number examined	Number positive (%)	$x2$ Value	P-Value
Sex			0.03	0.8
Female	179	5(2.8)		
Male	589	15(2.6)		
Breed			2.6	0.1
Local	695	16(80)		
Cross	73	4(20)		
Age			12.4	0.002*
<4years	65	1(1.5)		
4-6years	669	15(2.2)		
>6years	33	4(12.12)		
Body condition			35.7	<0.001*
Poor	10	3(15)		
Medium	164	8(40)		
Good	594	9(45)		

* Statistically significant ($P < 0.05$)

Table 2: Logistic regression analysis of risk factors of animals with tuberculous lesions in cattle slaughtered in Dilla municipal abattoir, Gedeo zone, Southern Ethiopia

Variable	Number examined	Number positive (%)	Crude OR(95%CI)	Adjusted OR (95%CI)
Sex				
Female	179	5(2.8)	1	1
Male	589	15(2.6)	0.9 (0.3-2.5)	1.2(0.4- 3.6)
Breed				
Local	695	16(80)	1	1
Cross	73	4(20)	2.46 (0.8- 7.5)	2.1(0.6-7.2)
Age (in years)				
<4years	65	1(1.5)	1	1
4-6years	669	15(2.2)	1.47 (0.2-11.3)	1.6 (0.2- 14.2)
>6years	33	4(12.1)	8.83(0.9- 82.5)	10.6(0.9-119.4)
Body condition				
Poor	10	3(15)	1	1
Medium	164	8(40)	0.12 (0.03- 0.6)	0.1(0.02-0.4)
Good	594	9(45)	0.04 (0.01-0.2)	0.03(0.01- 0.1)

Table 3: Distribution and frequency of tuberculous lesions in the tissues of infected animals in Dilla manucipital abattoir, Gedeo Zone

Anatomic site	Organ affected	Frequency
Head	Retropharyngeal LN	1
Thorax	Bronchial LN	4
	Lung	8
	Mediastinal LN	3
Abdomen	Mesenteric LN	2
Carcass	Prescapular LN	2
	Total	20

LN=lymph nodes

20% in bronchial lymph nodes and 15% in mediastenal lymph nodes. Mesenteric lymph nodes and prescapular lymph nodes account 10% each and retropharyngeal lymphnode (5%) of the total lesions detected. With respect to location to anatomical site, 75% and 10% of the lesions were located in thoracic cavity and the head area respectively. The remaining lesions were found in abdomen (Mesentric lymphnode) region (10%) and the carcass (10%).

Mycobacteriology: Out of the total 20 tuberculous lesions that were mycobacteriologically processed and cultured, growth was observed in 3 (15%) of the lesioned samples on culture.

m-PCR genus typing of AFB isolates: Two isolates out of the three culture positive showed a PCR product size of 1030bp up on Genus specific m-PCR typing which is specific for NTM. While one isolate did not show a signal at all to the genus *Mycobacterium*.

DISCUSSION

Based on the detailed abattoir inspection procedure in cattle slaughter in Dilla municipal abattoir, the proportion of carcasses with bTB suggestive lesions was found to be 2.6%. The 2.6% prevalence of bTB recorded in this study was in parallel with other results obtained by other researchers [23, 24]. However, the finding was lower than findings reported from different abattoirs [8, 10, 25, 26-31]. On the contrary, our finding was relatively higher than a study finding conducted at Hawassa Municipal abattoir with a prevalence rate of 1.1% [32]. These variations in prevalence could be due to the differences in the epidemiology of the disease in the animal populations and/or interventions taken to control it.

The best evidence of the transmission route of *M. bovis* to cattle is the pattern of lesions observed in slaughtered animals [33]. Majority of TB suggestive lesions were detected in the lung and its associated lymph nodes (75%).This was consistent with the previous reports [25, 26, 29] which indicated that respiratory route is the primary route of transmission and infection [25, 26, 33, 34-39]. It is also stated that the usual and common transmission of Mycobacteria from humans to cattle is direct and by the respiratory route [36, 40].

The post mortem examination result indicated statistically significant difference among the age groups ($P< 0.05$). As the age of the animal increases, owing to increased chances of exposure and infection with bTB. This is due to the fact that stressors, malnutrition and immunosuppressants increase with age [41]. It has been suggested that increased incidence of bTB in older animals can be by a declining of protective capability in aging animals [36]. The occurrence of lesions in medium and good conditioned animals was less likely as depicted in Table 2. It is known fact that animals under good body condition are with good immune status that can respond to any foreign protein better than those with poor body condition.

bTB suggestive pathologic lesions were further cultured on growth media. However, culture positivity of suspicious tissues was 15% which is much lower than previous reports from cattle [10, 15, 42] and camel [30, 31, 43, 44]. This could be also due to miscategorization of non tuberculous lesions as tuberculous lesions [26] that are caused by other granuloma-causing organisms [40, 45]. Tuberculous gross visible lesions may be caused by an array of pathogens [46, 47].

From our study, NTM species was isolated from the bTB suggestive lesions. Previous studies also showed the association of NTM with granulomatous lesions in cattle and humans [9, 10, 30, 31, 44, 48].

In conclusion, Mycobacteria other than *M. tuberculosis* complex members were isolated from the suspicious lesions in our study although *M. bovis* has been known to be the major cause of bTB in cattle. The isolation of NTM from tuberculous lesions highlights their importance in causing granulomatous lesions and the epidemiology of the disease in cattle. Characterization of these isolates to specific species level and further investigation aiming at identification of the source of

NTM infections, transmission routes, pathogenicity in animals and their public health significance in the study area is recommended.

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REFERENCES

1. Buncic, S., 2006. Integrated Food Safety and Veterinary Public Health. 1st ed. CABI, London, UK., pp: 61.
2. Amanfu, W., 2006. The situation of tuberculosis and tuberculosis control in animals of economic interest. Tuberculosis, 86: 330-335.
3. OIE, 2009. Bovine Tuberculosis. OIE Terresterial Manual,Paris, pp: 683-698.
4. Tenguria, K.R., F.N. Khan, S. Quereshi and A. Pandey, 2011. Epidemiological Study of Zoonotic Tuberculosis Complex (ZTBC). World Journal of Science and Technology, 1(3): 2231-2587.
5. Anonymous, 1994. Zoonotic tuberculosis (mycobacterium infection): a memorandum from WHO (with participation of FAO). Bulletin of World Health Organization, 72: 851-857.
6. Ayele, W.Y., S.D. Neill, J. Zinsstag, M.G. Weiss and I. Pavlik, 2004. Bovine tuberculosis: an old disease but a new threat to Africa. International Journal of Tuberculosis and Lung Disease, 8: 924-937.
7. Bogale, A., E. Lemma, K. Tadele and S. Britton, 2000. Bovine Tuberculosis: A cross-sectional and epidemiological study in and around Addis Ababa. Bulletin of Animal Health and Production in Africa, 48: 71-80.
8. Shitaye, J.E., B. Getahun, T. Alemayehu, M. Skoric, F. Treml, P. Fictum, V. Vrbas and I. Pavlik, 2006. A prevalence study of bovine tuberculosis by using abattoir meat inspection and tuberculin skin testing data, histopathological and IS6110 PCR examination of tissues with tuberculous lesions in cattle in Ethiopia. Veterinarni Medicina, 11: 512-522.
9. Berg, S., R. Firdessa, M. Habtamu, E. Gadisa, A. Mengistu, L. Yamuah, G. Ameni, M.H. Vordermeier, D.B. Robertson, H.N. Smith, H. Engers, D. Young, R. G. Hewinson, A. Aseffa and V.S. Gordon, 2009. The Burden of Mycobacterial Disease in Ethiopian Cattle: Implications for Public Health. PLoS ONE, 4: 5068.
10. Nemomsa, B., G. Gebrezgabiher, T. Birhanu, G. Tadesse, H. Tadelle and B. Getaneh, 2014. Epidemiology of bovine tuberculosis in Butajira, Southern Ethiopia: A cross-sectional abattoir-based study. African Journal of Microbiology Research, 8(33): 3112-3117.
11. Kiros, T., 1998. Epidemiology and zoonotic importance of bovine tuberculosis in selected sites of Eastern Shewa Ethiopia. Faculty of Veterinary Medicine, Addis Ababa University and Freie Universitat, Berlin, Germany. MSc Thesis.
12. Regassa, A., 2005. Study on Mycobacterium bovis in animals and human in and around Fiche, North Shewa zone, Ethiopia. MSc. Thesis. Faculty of Veterinary Medicine, Addis Ababa University, Debre-Zeit, Ethiopia.
13. Ameni, G., G. Hewinson, A. Aseffa, D. Young and M.H. Vordermeier, 2008. Appraisal of interpretation criteria for the comparative intradermal tuberculin test for diagnosis of tuberculosis in cattle in Central Ethiopia. Clinical Vaccine Immunology, 15: 1272-1276.
14. Fetene, T., N. Kebede and G. Alem, 2009. Tuberculosis infection in animal and human populations in three districts of Western Gojam, Ethiopia. Zoonoses and Public Health, 58(2011): 47-53.
15. Ameni, G., A. Aseffa, H. Engers, D. Young, S. Gordon, G. Hewinson and M.H. Vordermeier, 2007. High prevalence and increased severity of pathology of bovine tuberculosis in Holsteins compared to Zebu breeds under field cattle husbandry in central Ethiopia. Clinical and Vaccine Immunology, 14: 1356-1361.
16. Biffa, D., A. Bogale and E. Skjerve, 2010. Diagnostic efficiency of abattoir meat inspection service in Ethiopia to detect carcasses infected with Mycobacterium bovis: Implications for public health. Biomedical Center Public Health, 10: 462.
17. WHO, 1998. Laboratory service in tuberculosis control part III culture. Geneva, pp: 1-95.

18. Ameni, G., A. Aseffa, H. Engers, D.B. Young, G.R. Hewinson and M.H. Vordermeier, 2006. Cattle husbandry is a predominant factor affecting the pathology of bovine tuberculosis and Gamma Interferon responses to mycobacterial antigens. Clinical and Vaccine Immunology, 13: 1030-1036.
19. Cadmus, S., S. Palmer, M. Okker, J. Dale, K. Gover, N. Smith, K. Jahans, R.G. Hewinson and S.V. Gordon, 2006. Molecular analysis of human and bovine tubercle bacilli from a local setting in Nigeria. Journal of Clinical Microbiology, 44: 29-34.
20. OIE, 2010. Bovine tuberculosis. In: Manual of Diagnostic Tests and Vaccines for Terrestrial Animals. Part 2, Section 2.4. Chapter 2.4.7. World Organization for Animal Health, pp: 683-698.
21. Wilton, S. and D. Cousins, 1992. Detection and identification of multiple mycobacterial pathogens by DNA amplification in a single tube. PCR Methods and Applications, 1(4): 269-273.
22. STATA Corp., 2009. STATA Statistical Software. Release 11.0, College station, Texas.
23. Jemale, M., 2005. Evaluation of Meat Inspection Procedures for the Diagnosis of Bovine Tuberculosis in Awassa Municipality Abattoir. DVM thesis, Faculty Veterinary Medicine, Addis Ababa University, Debre Zeit.
24. Mihreteab, B. and B. Indris, 2011. Evaluation of Routine Meat Inspection Procedure to Detect Bovine Tuberculosis Suggestive Lesions in Jimma Municipal Abattoir, South West Ethiopia. Global Veterinaria, 6(2): 172-179.
25. Ameni, G. and A. Wudie, 2003. Preliminary study on bovine tuberculosis in Nazareth municipality abattoir of central Ethiopia. Bulletin of Animal Health and Production in Africa, 51: 125-132.
26. Teklu, A., B. Asseged, E. Yimer, M. Gebeyehu and Z. Woldesenbet, 2004. Tuberculous lesions not detected by routine abattoir inspection: the experience of the Hossana municipal abattoir, Southern Ethiopia. Review of Science and Technology, 23(3): 957-964.
27. Mamo, B., 2007. Efficiency of Abattoir inspection to detect Tuberculosis Lesions in Cattle Slaughtered at Adama Municipality Abattoir, Central Ethiopia. DVM thesis, Faculty of Veterinary Medicine, Addis Ababa University, Debre Zeit.
28. Biffa, D., F. Inangolet, J. Oloya, B. Asseged, M. Badaso, A. Yilkal and E. Skjerve, 2009. Prevalence of bovine tuberculosis in Ethiopian slaughter cattle based on post-mortem examination. Tropical Animal Health and Production, 41: 755-65.
29. Tigre, W., G. Alemayehu, T. Abetu and G. Ameni, 2012. Preliminary study on the epidemiology of bovine tuberculosis in Jimma town and its surroundings, Southwestern Ethiopia. African Journal of Microbiology Research, 6(11): 2591-2597.
30. Romha, G., G. Ameni, G. Berhe and G. Mamo, 2013. Epidemiology of mycobacterial infections in cattle in two districts of Western Tigray Zone, northern Ethiopia. African Journal of Microbiology Research, 7(31): 4031-4038.
31. Zeru, F., G. Romha and G. Ameni, 2013. Gross and molecular characterization of mycobacterium tuberculosis complex in Mekelle Town municipal Abattoir, Northern Ethiopia. Global Veterinaria, 11(5): 541-546.
32. Regassa, A., A. Tassew, K. Amenu, B. Megersa, F. Abunna, B. Mekibib, T. Macrotty and G. Ameni, 2010. A cross-sectional study on bovine tuberculosis in Hawassa town and its surroundings, Southern Ethiopia. Tropical Animal Health and Production, 42: 915-920.
33. Phillips, C.J.C., C.R.W. Foster, P.A. Morris and R. Teverson, 2003. The transmission of Mycobacterium bovis infection to cattle. Research in Veterinary Science, 74: 1-15.
34. Corner, L.A., 1994. Post-mortem diagnosis of Mycobacterium bovis infection in cattle. Veterinary Microbiology, 40: 53-63.
35. Neill, S.D., J.M. Pollock, D.B. Bryson and J. Hanna, 1994. Pathogenesis of Mycobacterium bovis infection in cattle. Veterinary Microbiology, 40: 41-52.
36. O'Reilly, L.M. and C.J. Daborn, 1995. The epidemiology of Mycobacterium bovis infections in animals and man. A review. Tubercle and Lung Disease, 1: 1-46.
37. Daborn, C.J., J.M. Grange and R.R. Kazwala, 1996. Bovine tuberculosis cycle: an African prospective. Journal of Applied Bacteriology, 81: 275-325.
38. Menzies, F.D. and S.D. Neill, 2000. Cattle-to-cattle transmission of bovine tuberculosis. Veterinary Journal, 160: 92-106.
39. Goodchild, A.V. And R.S. Clifton-Hadley, 2001. Cattle-to-cattle transmission of Mycobacterium bovis. Tuberculosis, 81: 23-41.
40. Radostits, O.M., C.C. Gay, K.W. Hinchcliff and P.D. Constable, 2006. Diseases associated with Mycobacterium species. In: Veterinary Medicine, A Textbook of the Diseases of Cattle, Horses, Sheep, Pigs and Goats. 10th ed. Edinburgh, London, New York, Oxford, Philadelphia, St Louis Sydney, Toronto, pp: 1008-1017.

41. Humblet, M.F., M.L. Boschiroli and C. Saegerman, 2009. Classification of worldwide bovine tuberculosis risk factors in cattle: a stratified approach. Review article. Veterinary Research, 40(50): 1-24.
42. Ameni, G., F. Desta and R. Firdessa, 2010. Molecular typing of Mycobacterium bovis isolated from tuberculosis lesions of cattle in north eastern Ethiopia. Veterinary Record, 167: 138-141.
43. Mamo, G., G. Bayleyegn, T. Sisay, M. Legesse and G. Medhin, 2011. Pathology of Camel Tuberculosis and Molecular Characterization of its Causative Agents in Pastoral Regions of Ethiopia. PLoS ONE, 6: e15862.
44. Ayana, T., K. Tafesse, G. Mamo, T. Sisay and G. Ameni, 2013. Isolation and molecular characterization of non tuberculosis mycobacteria from skin positive reactors and pathological lesions of cattle at Bako, Western Ethiopia. African Journal of Microbiology Research, 7(20): 2190-2197.
45. Grist, A., 2008. Bovine Meat Inspection : Anatomy, Physiology and Disease Conditions. 2nd ed. Nottingham University Press, Nottingham, UK, pp: 296.
46. Diguimbaye-Djaibé, C., V. Vincent, E. Schelling, M. Hilty, R. Ngandolo, H.H. Mahamat, G. Pfyffer, F. Baggi, M. Tanner and J. Zinsstag, 2006. Species identification of non-tuberculous Mycobacteria from humans and cattle of Chad. Schweiz. Arch. Tierheilkd, 148: 251-256.
47. Oloya, J., J. Opuda-Asibo, B. Djønne, J. Muma, G. Matope, R. Kazwala and E. Skjerve, 2006. Responses to tuberculin among Zebu cattle in the transhumance regions of Karamoja and Nakasongola district of Uganda. Tropical Animal Health and Production, 38(4): 275-283.
48. Kazwala, R., D. Kambarage, C. Daborn, J. Nyange, S. Jiwa and J. Sharp, 2001. Risk Factors Associated with the Occurrence of Bovine Tuberculosis in Cattle in the Southern Highlands of Tanzania. Veterinary Research Communications, 25(8): 609-614.