

## 7- Summary

During the period from August 2010 to August 2012 the present study was conducted on 415 cattle and 50 buffaloes. The age of these animals ranged from one day to above five years old. The animals belonged to farms and villages of AL-wady AL-Geded, Assiut and AL-Fayoum Governorates.

The clinical examination of the animals during this study concluded that some of those animals were actually infected and showed different degrees of tick infestation. The clinical Signs recorded on the infected animals from both species, included enlargement of the superficial lymph nodes, fever, various degrees of respiratory affection and ocular lesions as lacrimation or corneal opacity. In addition to, other clinical signs which were less common were recorded in individual cases like nervous manifestations, hemoglobin urea and bloody to tarry like diarrhea. Other animals were chronically infected; carriers and some animals were clinically healthy. The results of the current study concluded that the conventional method of diagnosis such as (blood films and lymph smears) is still recommended for day to day examination in clinically infected cases, especially during the acute stage. It gave 24.58 % and 6% in blood films of cattle and buffaloes respectively if compared to lymph smear which record 68.09% and 30.77% in cattle and buffaloes respectively, thus the use of lymph smears detected more infected cases than using blood films.

The (TaSP) ELISA test was sensitive and specific for detection of *T. annulata* infection in both cattle and buffaloes through detection of specific antibodies. The infection rate in cattle was 73.25% while in buffaloes it was 42%. The ELISA is recommended for use during the epidemiological surveys to evaluate the incidence of the infection.

The results of the present study clearly showed that the qualitative simplex (Tams-1) target based PCR assay was a sensitive test for detection of tropical theileriosis infection. The infection rates in blood taken from cattle and buffaloes were 46.19% and 8.11% respectively.

The infection rates as determined by using the loop-mediated isothermal amplification (LAMP) assay were 65.24% and 43.24% in blood samples taken from cattle and buffaloes, respectively.

The results of the present study confirmed that Reverse Line Blot (RLB) assay was the most sensitive and specific assay for detection of *Theileria* species infection, allowing in addition the determination of mixed infections in both cattle and buffaloes. The infection rates with tropical theileriosis were (65.24% and 51.35%) in blood samples taken from cattle and buffaloes, respectively. Based on its high sensitivity and specificity it was used as the reference test to evaluate the results of conventional, serological and molecular assays.

The infection rate among examined samples were higher in AL-Wady AL-Geded governorate (19.28%) followed by Assiut governorate (16.63%) and AL-Fayoum governorate (11.80%) in cattle, respectively. On the other hand the infection rate in buffaloes was higher in Assiut governorate (20%) if compared with that in AL-Fayoum governorate (18%).

Cattle were more susceptible than buffaloes which usually act as carriers of the infection. Within cattle breeds, Frisian were more sensitive and susceptible than native breed, infection rates were 34.94%, 12.77%, respectively in cattle and 38% in buffaloes.

The male animals in cattle were more susceptible to the disease than female animals (24.10% and 23.61%) in male and female cattle respectively. While in female buffaloes the infection rate was higher than in male buffaloes (20% and 18%), respectively.

Animals less than one year old were more susceptible to infection in both species, if compared with the older animals. The infection rate was 26.51% in cattle and 22% in buffaloes.

The infection with tropical theileriosis is higher during hot months in both cattle and buffalo with infection rates of 33.98% and 38% respectively, while the lowest infection rates were recorded during non-hot months when cattle showed 13.73% while no infection was detected in buffaloes.

The evaluation of the conventional method showed that the sensitivity of this method was 43.80% and 15.80% in cattle and buffaloes, respectively. The specificity was 100% in both cattle and buffaloes.

Serological method used (TaSP) ELISA, had a sensitivity of 80.29% and 57.89% in cattle and buffaloes, respectively while the specificity was 56.17% and 50% in cattle and buffaloes, respectively.

Molecular assays, in case of the qualitative simplex (Tams-1) target-based PCR assay, the sensitivity was 70.80% and 15.79% in cattle and buffaloes, respectively, while the specificity was 100% in both cattle and buffaloes.

On the other hand the sensitivity using the loop-mediated isothermal amplification (LAMP) assay were (78.10% and 47.37%) in cattle and buffaloes, respectively. The specificity was 57.53% and 61.11% in cattle and buffaloes, respectively.

In the current study 11 samples from cattle and buffaloes from the three governorates were subjected to cloning, sequencing and alignment of the translated Tams-1 amino acid sequence. The results showed that the Tams-1 polypeptide is variable. Thus it cannot be recommended for applications of serological diagnosis or/and vaccines preparation against *T. annulata* infection. In addition, 12 samples from cattle and buffaloes from the three governorates were subjected to cloning, sequencing, and alignment of the 18S srRNA gene. Phylogenetic analysis was carried out by using (NJ) method. The results

revealed that in Assiut and AL-Fayoum governorates there were different species of *Theileria* in addition to *Theileria annulata* which infecting both cattle and buffalo. These species included *Theileria lestoquardi*, *T. uilenbergi* and *Theileria ovis*. Interestingly, *Theileria annulata* is the only species founded in the AL-Wady AL-Gaded governorate with respect to the samples analyzed.