Patho - physiological Effects of Experimental *Trypanosoma congolense* and *Trypanosoma vivax* Infections in the Grasscutter (*Thryonomys swinderianus*,Temminck)

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ABSTRACT: Trypanosome infection might affect grasscutter's productivity while in domestication. The PCV, MCHC, total WBC and Lymphocytes of the grasscutters experimentally infected with *T. congolense* and *T. vivax* decreased ($p<0.05$), while MCV increased ($p<0.05$) 21dpi. Plasma glucose and cholesterol were decreased ($p<0.05$). Body temperature fluctuated between 37.4^{0}C and 39.2^{0}C with a peak on day 12 (39.2^{0}C) in *T. congolense* and 37.5^{0}C to 40.1^{0}C which peaked on day 8 (40.1^{0}C) in *T. vivax*. The livers and kidneys showed vacuolar and tubular epithelial degeneration respectively, with thrombosis in alveolar blood vessels. It was concluded that the grasscutter may serve and might have been playing the role of reservoirs hosts for this economically important disease. Infected grasscutters though did not show clinical signs of this disease, but clearly manifested haematological and tissue changes which could lead to death. [Nature and Science 2010;8(10):88-101]. (ISSN: 1545-0740).
Key words: Patho-physiology, *Trypanosoma congolense, Trypanosoma vivax,* Infections, Grasscutter

INTRODUCTION

Wildlife serves as source of animal protein to Africans, in urban and rural communities (Ajayi, 1975; Fonweban and Njwe, 1990). Preference for bush meat is widely accepted in sub – Saharan Africa.

Among the wild rodents hunted as bush meat, the grasscutter (cane rat, cane cutter) is the most preferred (Ajayi, 1975; Baptist and Mensah, 1986; Fonweban and Njwe, 1990). It is also the most expensive meat in West Africa, where it contributes to the local and export earnings. Consequently, it is hunted aggressively (Baptist and Mensah, 1986; Asibey and Addo, 2000). The collection of grasscutters from wild usually leads to destruction of environment through the setting of bush fires and cutting of the forest by hunters (NRC, 1991; Yeboah and Adamu, 1995; Ntiamoa-Baidu, 1998).

To ameliorate this problem, domestication of grasscutters in the sub-region has recently been introduced and encouraged.

So much research work on the Socio-economic and zoo technical characteristics of the grasscutter (Baptist and Mensah, 1986; Mensah et al, 1986; Holzer et al, 1986; NRC, 1991; Awa-Ndukum et al, 2001) had been studied. In addition, few preliminary studies on parasitic diseases of grasscutters, carried out in Cameroun (Awah-Ndukum et al, 2001), reported the occurrence of ectoparasites such as Fleas (*Xenopsylla sp*) and endoparasites like Cestode (*Hymenolopsis sp*), Nematode (*Heterakis sp*) and in Ghana (Yeboah and Simpson, 2004), 4 species of ticks*;* 6 sp of helminthes comprising 2 Cestodes and 4 Nematodes.

Opara et al (2006) had also reported natural infection of both the captive – reared and wild grasscutters with *Trypanosoma species.* The authors suggested that failure of establishment of clinical trypanosomiasis in these rodents could be attributed to the nature of food varieties the grasscutters consume. However, the effect of this infection on the vital organs of the grasscutters has not been studied and documented. Such information is scarce.

This study therefore, is aimed at investigating and documenting the histopathological effects trypanosome infection in the grasscutter.

MATERIALS AND METHODS

Twenty seven (27) grasscutters were housed in a raised iron cage and fed forages and water for a week. The tails of the grasscutters were snipped at the tip and blood samples from them were collected into sample bottles containing EDTA, to evaluate their haematological values and screening for trypanosomes.

They were later randomly distributed into 3 treatment groups of 9 grasscutters each, such that Treatment "O" (TO) was for control which received forages and water, treatment "V" (TV) was for grasscutters to be infected with *T .vivax* while treatment "C" (TC) were for *T. congolense.*

Acclimatization was observed for two weeks, while a measurement of their temperature was taken daily.

Just 1ml of the prepared diluted blood containing 2 x 104, *T. congolense* and *T. vivax* stock isolated from

grasscutters was administered intra - peritoneally into each of the grasscutters in TV and TC respectively. Treatment "O" (TO) served as control and did not receive any trypanosome organisms.

Measurements of the rectal temperature were taken continuously daily until the experiment was terminated.

Blood smears were made to confirm parasitaemia in the grasscutters, using the dark ground / phase contrast Buffy coat method (Paris et al, 1982) at three days interval.

Twenty one days post infection (21dpi), 5 grasscutters were randomly selected from each of the treatment groups and sacrificed. Blood samples were again collected from the grasscutters for determination of haematological indices, while some organs of the sacrificed animals were also collected and taken to the laboratory for histopathological studies.

Data generated were then subjected to descriptive statistics.

RESULTS

The results of experimental infection of grasscutters using *Trypanosoma congolense* and *T. vivax* on some haematological indices are shown in figures 1, 2, 3, 4 and 5.

The PCV, MCHC, total WBC and Lymphocytes of grasscutters experimentally infected with *T. congolense* and *T. vivax* decreased ($p<0.05$), as shown in figures 1, 3, 4 and 5 respectively, while figure 2 shows that the MCV of these animals significantly increased ($p<0.05$) 21dpi.

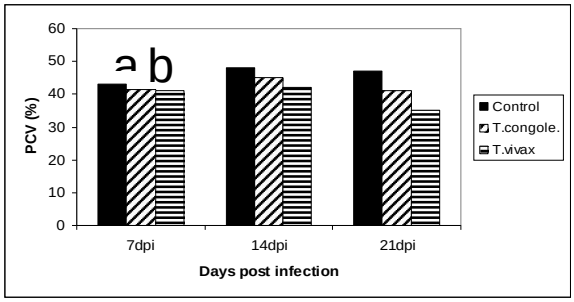


Fig. 1: Packed cell volume (PCV) of grasscutters (n = 27) post infection with *T. vivax* and *T .congolense*.
ab means on the same row with different superscripts are significantly different

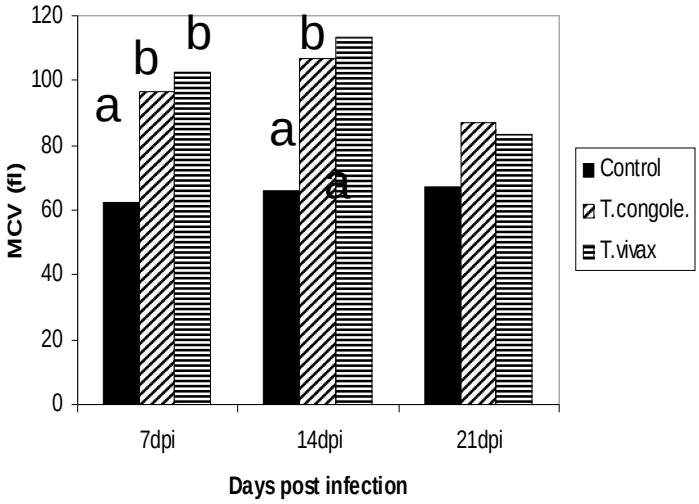
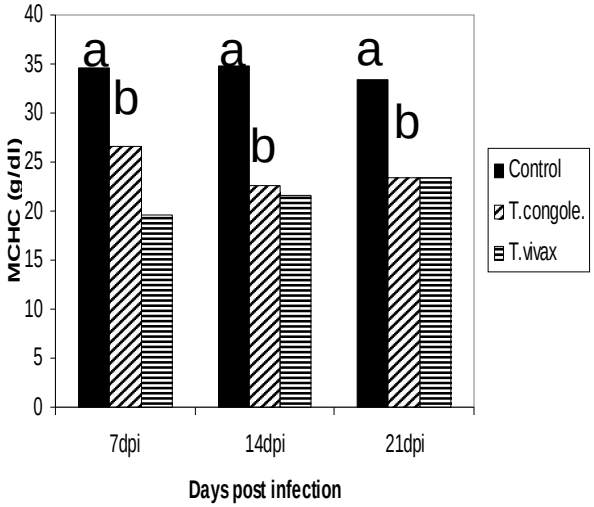


Fig. 2: MCV values of grasscutters experimentally infected with isolates of *T. congolense* and *T. vivax*



. Fig. 3: MCHC values of grasscutters experimentally infected with isolates of *T. congolense* and *T. vivax*

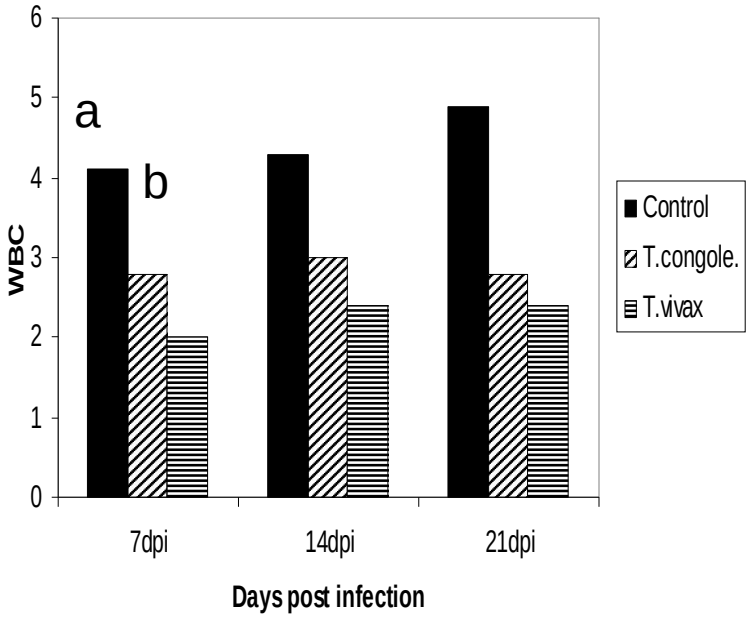


Fig 4: Leukocyte (10^3µL) values of grasscutters experimentally infected with isolates of *T. congolense* and *T. vivax*

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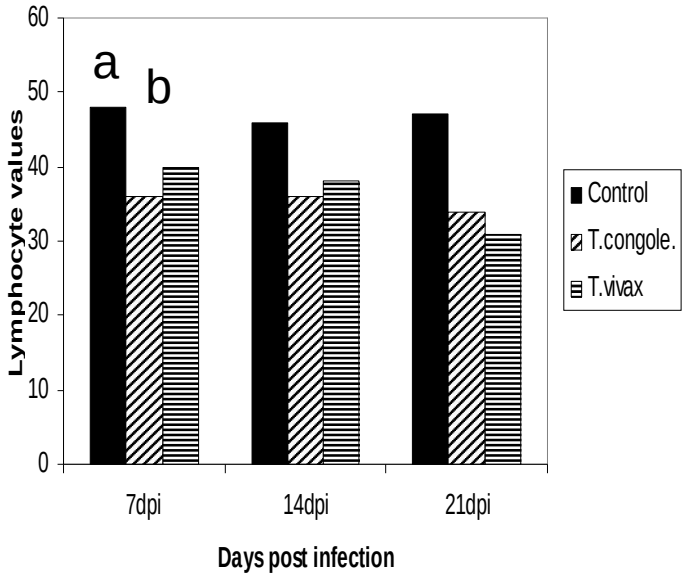

Fig. 5: Lymphocyte (10^3µL) of grasscutters experimentally infected with isolates of *T. congolense* and *T. vivax*

Table 1 shows that significant differences existed in the plasma cholesterol, urea, Creatinine and albumin of grasscutters experimentally infected with trypanosomes. Plasma glucose, sodium, chloride, bicarbonate, total protein levels (table 2) were similar. The plasma enzyme, ALT levels among the three groups of animals were also similar, except AST which was significantly ($p< 0.05$) higher in grasscutters experimentally infected with *T. vivax* (table 3).

Table 1: Plasma biochemical values of grasscutters (n =27) post *T. vivax* and *T. congolense* infections.

Parameters	TO	TV	TC		TO	TV	TC		TO	TV	TC
A) Solutes.											
Glucose	191[a]	184[b]	180[b]		188[a]	180[b]	182[b]		200[a]	176[b]	180[b]
Cholest.	68[a]	66[b]	60[b]		62[a]	53[b]	60[a]		66[a]	57[b]	61[b]
Creatinin.	0.2[b]	0.5[a]	0.2[b]		0.2[b]	0.7[a]	0.3[b]		0.4[b]	1.2[a]	0.5[b]

Total Protein	5.7	5.2	5.1		5.0	4.3	4.5		5.2	4.7	5.1
Albumin	3.5	3.1	3.3		3.6	3.0	3.2		3.6	2.7	3.5
	7dpi				**14dpi**					**21dpi**	

TO=control, TV= *T. vivax*, TC= *T. congolense*

Table 2: Electrolyte values of grasscutters (n =27) post *T. vivax* and *T. congolense* infections.

Parameter	TO	TV	TC		TO	TV	TC			TV	TC
B) Salts											
Potassium (mmol/L)	4.0	3.7	4.0		4	3.2	3.7		4.2	4.4	4.0
Chloride (mmol/L)	96.3	97.2	97		95	101	99		98	102	100
Bicarbonate (mmol/L)	24.5	24.2	25		25.6	25.2	26.1		25.9	26	27
21dpi		**7dpi**		**14dpi**							

Table 3: Enzymes values of grasscutters (n =27) post *T. vivax* and *T. congolense* infections.

Parameter	TO	TV	TC		TO	TV	TC		TO	TV	TC
AST(IU/L)	12.8	13.1	12.6		13.2	14.1	13		14 [b]	16 [a]	13.5 [b]
ALT(IU/L)	7	8	6		6	8	7		7	9	7
	7dpi				**14dpi**				**21dpi**		

The results of the body temperature measurements of grasscutters not infected and those experimentally infected with *T. congolense* and *T. vivax* are reported in figures 6 and 7 respectively.
The body temperatures of the grasscutters not infected with trypanosomes (fig. 6) ranged between 36.3^{O}C and 38.1 ^{O}C, while the temperatures of the grasscutters experimentally infected (fig. 7) fluctuated between 37.4^{0}C and 39.2^{0}C with a peak on day 12 (39.2^{0}C) in *T. congolense* and 37.5^{0}C to 40.1^{0}C which peaked on day 8 (40.1^{0}C) in *T. vivax*.

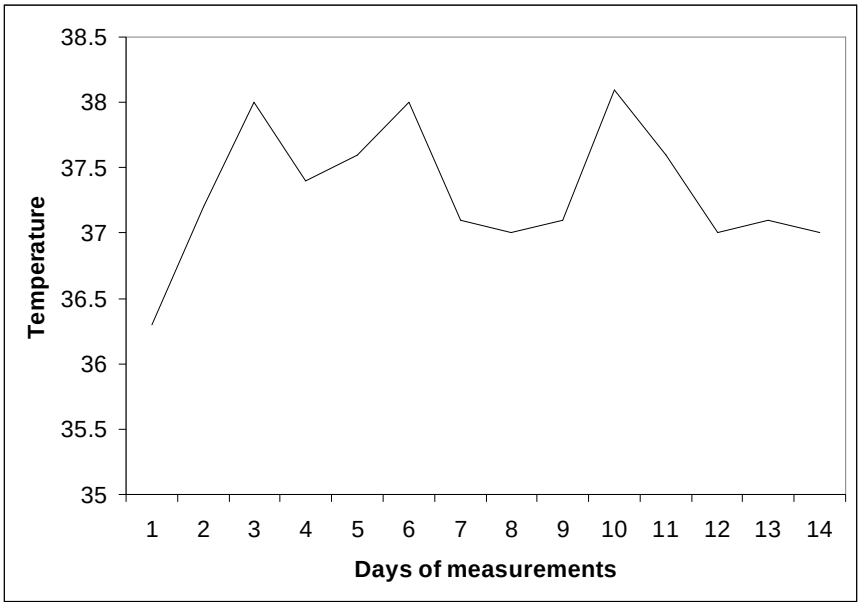

Fig. 6: Temperature chart of captive – reared grasscutters before experimental infection with trypanosomes, showing a normal temperature range of 36.3 ^{0}C to 38.1^{0}C

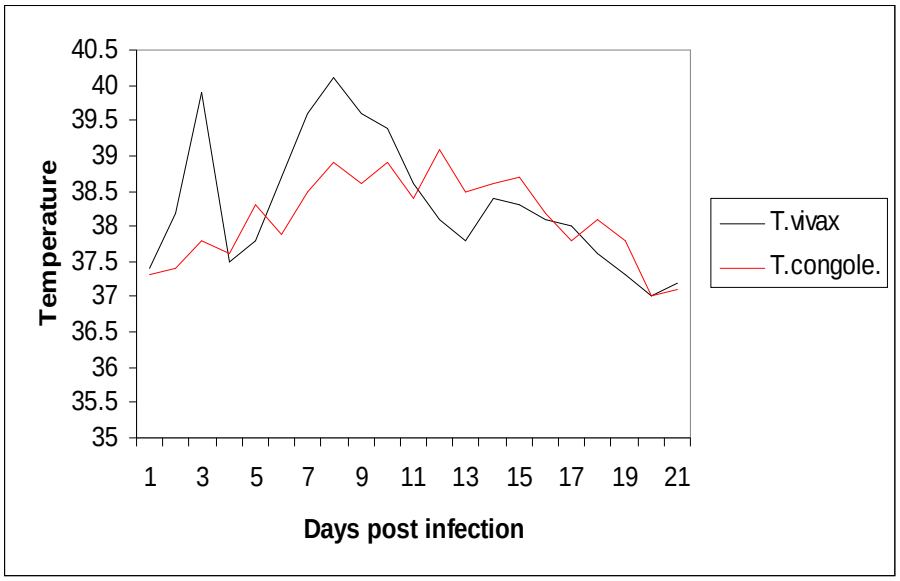

Fig. 7: Temperature chart of captive – reared grasscutters experimentally infected with *T. vivax T. vivax* and *T congolense*..

The histopathological effects of experimental infection of the grasscutters with *T. vivax* and *T congolense* are presented in plates 1 to 12 respectively.

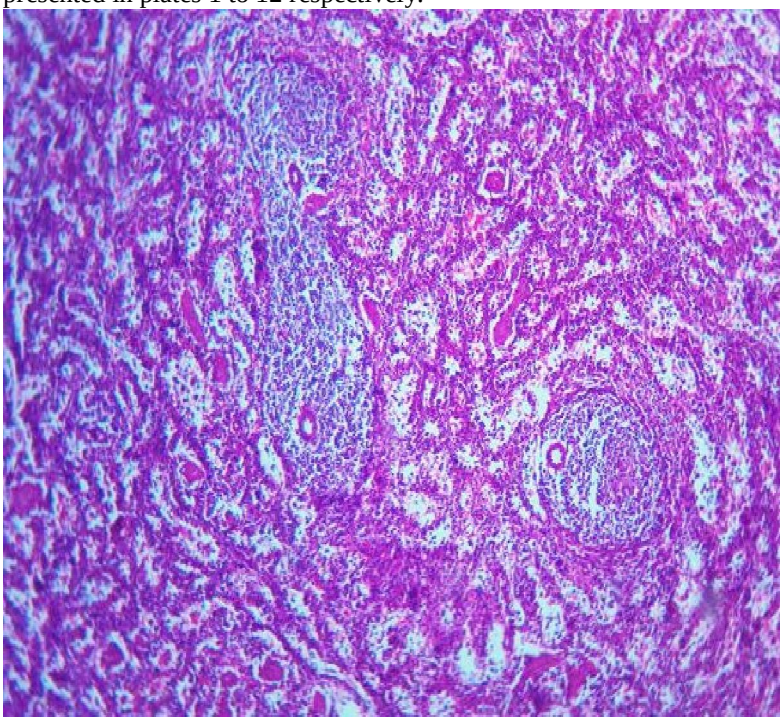

Plate 1: Photomicrograph showing the normal spleen of grasscutter (*Thryonomys swinderianus*)

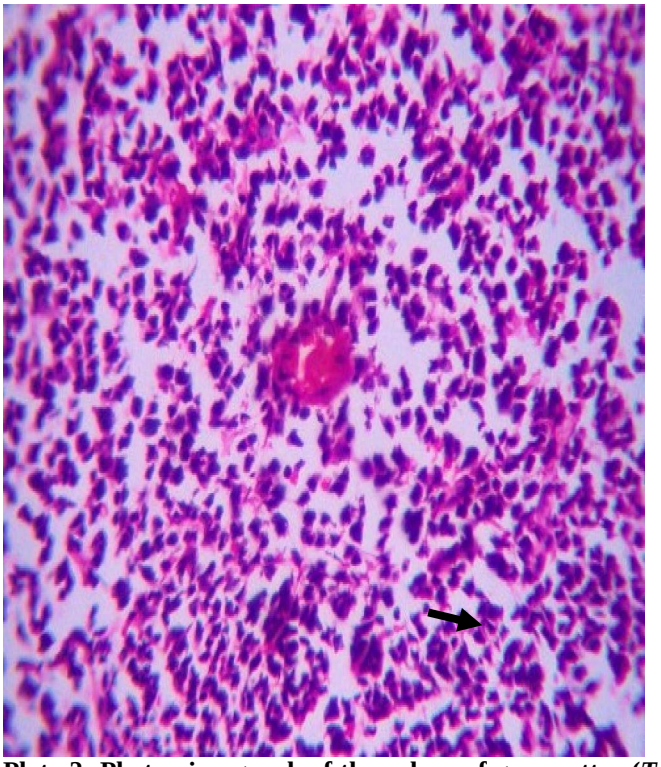

Plate 2: Photomicrograph of the spleen of grasscutter (*T. swinderianus*) infected with *T. congolense* showing lymphoid depopulation of the PALS and presence of numerous macrophage, some of which had phagocytosed lymphocytes (arrows) (H & E; x550)

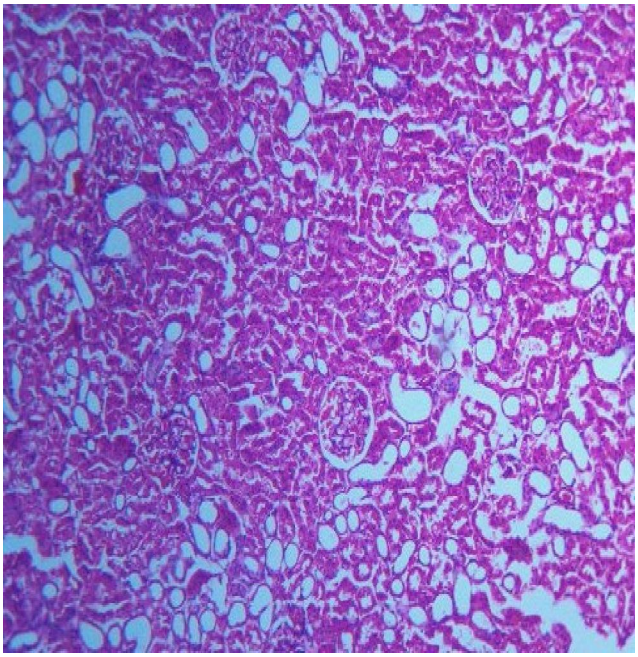

Plate 3: Photomicrograph showing the normal Kidney of grasscutter
(*T. swinderianus)*

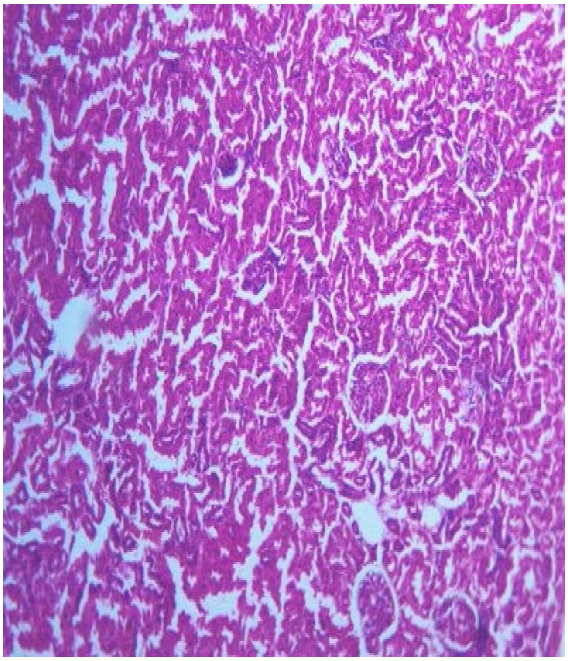

Plate 4: Photomicrograph of the kidney of grasscutter (*T. swinderianus*) infected with *T. vivax* showing a focal area of glomerular degeneration and mild focal interstitial mononuclear cellular infiltration (H & E; x350)

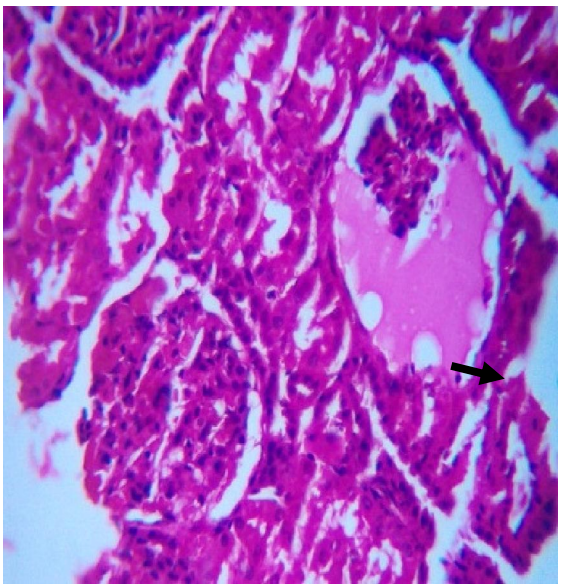

Plate 5: Photomicrograph of the kidney of grasscutter (*T. swinderianus*) infected with *T. congolense* showing glomerular and tubular degeneration and necrosis and presence of proteinaceous cast (arrow) within Bowman's capsule (H & E; x550)

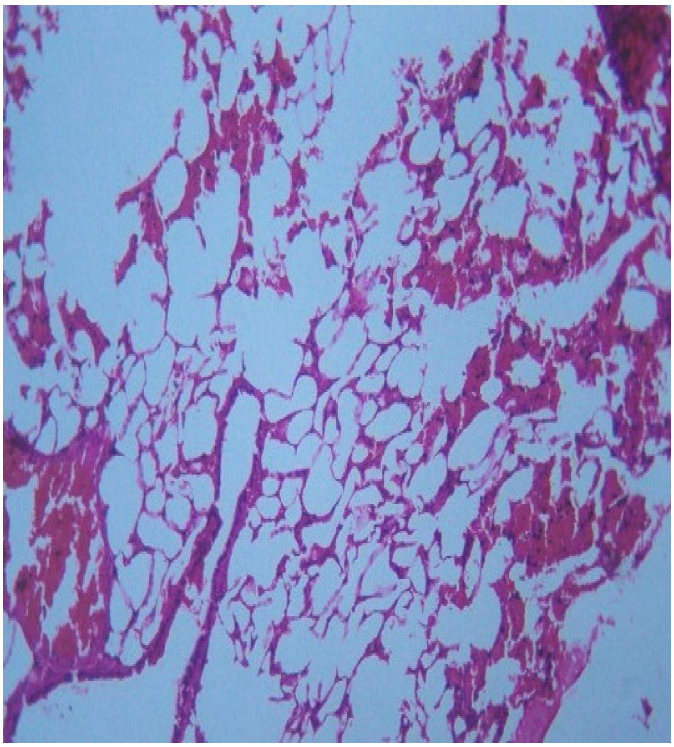

Plate 6: Photomicrograph showing the normal lung of grasscutter (*T. swinderianus*)

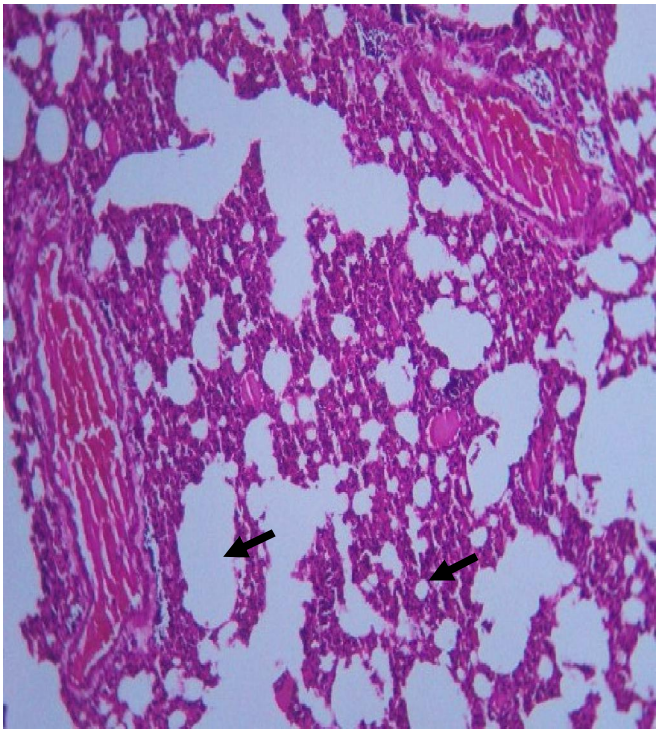

Plate 7: Photomicrograph of the lung of grasscutter (*T. swinderianus*) infected with *T. vivax* showing moderate pulmonary congestion, thickened interalveolar septa and mild perivascular mononuclear cell aggregations. Note the presence of thrombi in venules (arrows) (H & E; x350)

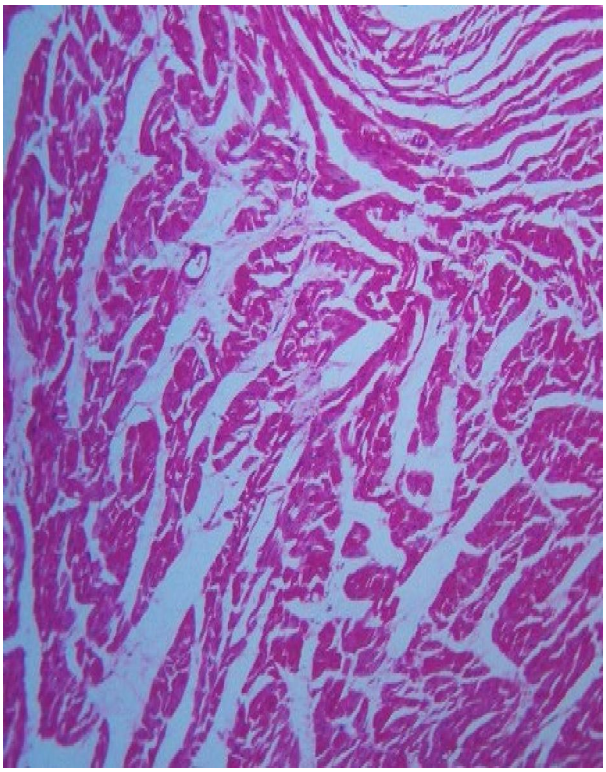

Plate 8: Photomicrograph showing the normal heart of grasscutter (*T. swinderianus*)

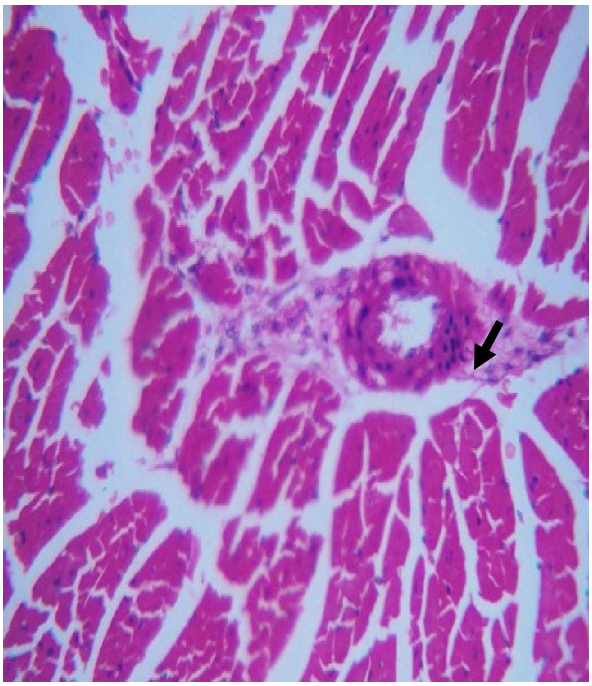

Plate 9: Photomicrograph of the heart of grasscutter (*T. swinderianus*) infected with *T. congolense* showing moderate myocardiac degeneration and atrophy, and fibrinoid degeneration and vasculitis of a vasa vasorum (arrow) (H & E; x550)

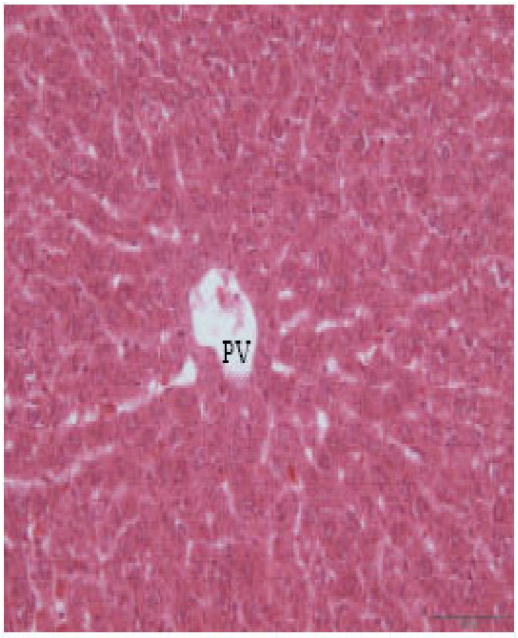

Plate 10: Photomicrograph of normal liver of grasscutter (*T. swinderianus*)
PV: Perivenular region (H & E; X 200)

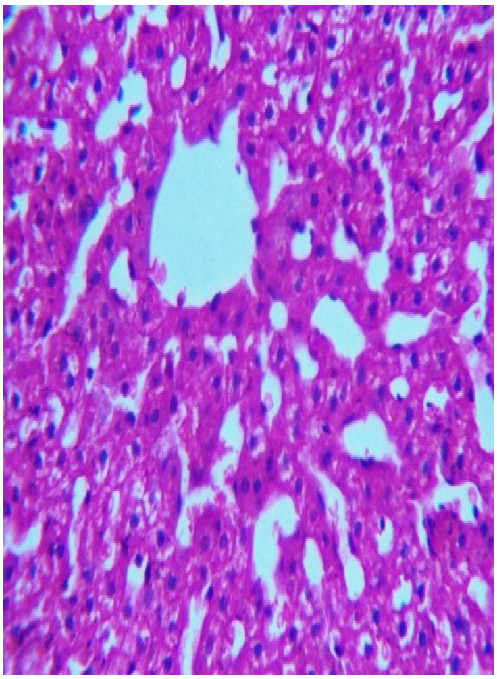

Plate 11: Photomicrograph of the liver of grasscutter (*T. swinderianus*) infected with *T. vivax* showing moderate centrilobular vacuolar degeneration and necrosis of hepatocytes and thinning of Bilroth cords (H & E; x550)

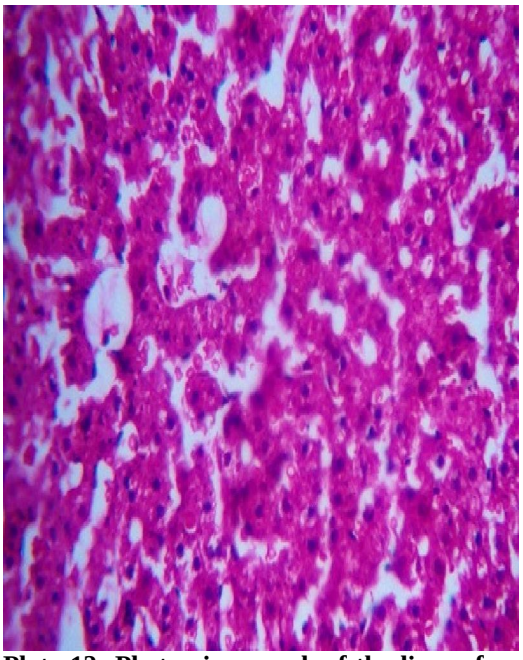

Plate 12: Photomicrograph of the liver of grasscutter (*T. swinderianus*) infected with *T. congolense* showing widespread vacuolar degeneration of hepatocytes and thinning of Bilroth cords (H & E; x550)

DISCUSSION

There were no significant differences (p> 0.05) in the PCV of the grasscutters, 7 and 14 days post infection with *Trypanosoma congolense* and *T. vivax* but (p< 0.05) by 21 days post infection (dpi). This result suggests that infection with any of these species of trypanosomes, does not significantly decrease the PCV, but at about 21dpi.

The MCV of the trypanosome infected grasscutters did not change more than for the uninfected ones, while the MCHC significantly decreased (p, 0.05) suggesting a macrocytic hypochromic anaemia.

Grasscutters experimentally infected with *Trypanosoma congolense* and *T. vivax* suffered leucopenia at 7dpi, while the uninfected ones did not. Thus, this result indicates that trypanosomiasis in grasscutters also leads to leucopenia (Seifert, 1996), which further reduces the animals' immunity and thereby exposing them to other infections.

The significant decrease in plasma glucose among the infected grasscutters agrees with Soulsby (1982), who reported that blood form of trypanosomes absorbs nutrients such as glucose by mediated mechanism of membrane transport.

He also reported that blood protozoa increase the long chain fatty acids of plasma membrane of red blood cells. However, this did not agree with the result of the present study, probably because the grasscutter always has very low body fat content (Adu et al, 1999).

Grasscutters experimentally infected with *T. vivax* (table 41c), had significantly (p< 0.05) higher AST. Serum AST is however not a specific enzyme for the liver, as high levels can also be found in skeletal and cardiac muscles as well as red blood cells (Bush, 1991). Thus, increase in AST may indicate an on – going liver disease (Duncan et al., 1994) as observed at histopathology.

Temperature of grasscutters infected with *T. vivax* and *T. congolense* in this study, showed dramatic fluctuation. Soulsby (1982) had reported that undulating temperature is a clinical feature in animals infected with trypanosomes.

The pathological lesions observed in some of the tissues of the infected grasscutters are in agreement with Soulsby, (1982) and Shah – Fischer and Say (1989), who also reported organ degenerative changes in animal trypanosomiasis. Our findings suggest that trypanosome infected grasscutters, though may appear clinically healthy, the lesions as evidenced in the histopathology could compromise the functions of the affected organs. This might ultimately lead to death.

CONCLUSION

The observation of the causative agents of trypanosomiasis, suggests that the grasscutter may serve and might have been playing the role of reservoirs hosts for this economically important disease. Infected grasscutters though do not show clinical signs of this disease, but clearly manifest

haematological and tissue changes which could lead to death.

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