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Pathophysiological and pro-inflammatory cytokine surveys on livestock normally infected with *Taenia saginata* cysticercosis

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Abstract

Background: Cattle and buffaloes can contract cysticercosis, an infection of the muscles brought on by *Taenia saginata* larvae. Despite having a global spread, cysticercosis is more prevalent in impoverished nations due to impaired hygiene standards. It has been discovered that *Taenia saginata* cysticercosis routine visual diagnosis is not very effective, especially in mild infections. Therefore, a more trustworthy *in vivo* test might be used as an alternative in slaughterhouses and epidemiological studies. Biochemical assays are possibly utilized as an alternative to detect cysticercosis inside a topical environment.

Aim: Investigating serum biochemical alterations in cattle with cysticercosis was the goal of the current research. As a further method of diagnosis, it was also determined how *Cysticercus bovis* affected pro-inflammatory cytokines and histopathology.

Methods: Blood samples from 42 slaughtered cattle (21 healthy and 21 sick animals) were taken from Assiut abattoir. Using an ELISA and spectrophotometer, respectively, their serum's pro-inflammatory cytokines and biochemical profile were evaluated. These cattle were chosen between March 2023 and February 2024.

Results: A percentage of 4.6% of the 455 cattle examined after being slaughtered had *T. saginata* cysticerci infections. All values in the serum biochemistry were considerably different ($p < 0.01$), whereas the majority of biochemical parameters increased significantly ($p < 0.01$) in infected animals. In contrast, there was a substantial ($p < 0.01$) decline in HDL-c, SOD, CAT, and GSH. On the other hand, procytokine inflammatory indices for both TNF- α and IL-1 β indicated a substantial increase ($p < 0.01$) in infected cattle. Additionally, the histological results revealed significant alterations in the tissues of infected livestock.

Conclusion: This has been inferred cysticercosis possesses negative impacts on cattle's plasma biochemical profiles, indicating the field applicability of biochemical measures in outbreaks of bovine cysticercosis. Pro-inflammatory cytokine indices and histological changes could be included as further indicators of *T. saginata* cysticercosis in cattle.

Keywords: *Taenia saginata*, *Cysticercus bovis*, Cattle, Serum enzymes, Pro-inflammatory cytokine.

Introduction

Cysticercosis is a muscle contagion brought on by the maggot phase of *Taenia saginata* (Hancock *et al.*, 1989). *Taenia saginata* is an economically and medically significant cestode parasite (McFadden *et al.*, 2011). *Taenia saginata* cysticercosis is prevalent throughout the world, although it has the paramount financial influence and communal soundness inferences in the tropics and subtropics (Abdo *et al.*, 2009). After consuming *T. saginata* eggs (proglottids) from infected humans, cattle get infected as intermediate hosts. After about 10 weeks, cysticerci develop in cow muscle and become infective to individuals (Flisser *et al.*, 2005; McFadden *et al.*, 2011). The disease is found in animals

and humans worldwide (Minozzo *et al.*, 2002), with varying prevalence (Doyle *et al.*, 1997). Cysticercosis is more common in underdeveloped nations due to poor sanitation and the ingestion of pickled or insufficiently stewed or sun-dried flesh (Frolova, 1982; Symth, 1994; Minozzo *et al.*, 2002). The illness is also an issue in advanced nations where underdone steak is commonly consumed as a repast. It is competence noting that even great-ranking flesh inspection in developed-country butchery has not resulted in the eradication of this scrounger (Frolova, 1982; Symth, 1994; Eddi *et al.*, 2003).

According to (Neva and Brown, 1994; Dural *et al.*, 2015; Symeonidou *et al.*, 2018), intestinal

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blockage, weakness, weight loss, nausea, vomiting, gastroenteritis, and abdominal discomfort are all signs of cysticercosis in humans. Rarely do movable gravid segments cause a major condition like the appendix or biliary tract infestation. Live cattle with cysticercosis do not exhibit any symptoms, but due to a large infestation of the grubs, there may be inflammatory cardiomyopathy or cardiac defect (Gracey and Collins, 1992; Belay and Afera, 2014).

Economic losses in the meat sector are closely related to infection status. If a corpse is determined to have a severe infection or generalized cysticercosis, it must be destroyed. Light cysticercosis results in the confiscation of the diseased-ridden sections; additionally, the cadaver must be held in reserve in a chilly warehouse at a hotness not transcending -70°C for 21 days to incapacitate the spongers (Kandil *et al.*, 2004; Scandrett *et al.*, 2009; Ibrahim and Zerihum, 2012).

Cysticercosis cannot be detected in live animals by antemortem testing, hence postmortem analysis of the carcass is the only method used to make the diagnosis at abattoirs. The existing tests (ELISA) need a lot of time, money, and effort to obtain. Therefore, it is beneficial for diagnostics to create some appropriate tests that can recognize diseased animals before slaughter. The tests help to find the disease's origin and may aid in disease management. In the domestic circumference and to meet the requirements of universal exordium for livestock with a sufficient level of reliance, hematological and biochemical trials were possibly performed as an alternative to the verification of *Cysticercus bovis* (Wrights, 1998; Saeed *et al.*, 2016). Damage to the host (pig) liver tissues by *T. saginata* asiatica cysticerci results in metabolic disorders of glycogen, fats, and protein, and alterations in ferment anabolism (Linghu *et al.*, 2007). Myocarditis and heart failure in cattle may result from a severe infestation of *T. saginata* cysticerci (Gracey and Collins, 1992; Belay and Afera, 2014). The literature on the variations in the serum biochemical factors of livestock infected with *T. saginata* cysticerci is scarce, nevertheless. The objective of the current investigation was to use serum biochemistry to identify *Taenia saginata* cysticercosis in slaughtered cattle. It was also done to look for changes in pro-inflammatory markers and histopathological changes caused by this infection.

Materials and Methods

Samples selection and collection of blood

At the Assiut abattoir in the Assiut governorate, blood samples were collected from 42 slaughtered cattle. Twenty-one healthy cattle and 21 cysticercosis cattle out of 455 detected by post-mortem analysis were used for biochemical analysis. Each animal's jugular vein was used to collect 5 ml of blood, which was then placed in clean, dry tubes and allowed to clot before the serum was centrifuged out and kept at -20°C to be used for biochemical and pro-inflammatory cytokine analysis.

Biochemical determinations

Biochemical analysis was achieved spectrophotometrically as aspartate aminotransferase (AST), lactate dehydrogenase (LDH), and creatine kinase (CK-total) vigors were specified to utilize kits obtained from Human Gesell Schafftur Biochemical und Diagnostic mbH, Germany. Serum total cholesterol, triglycerides, LDL-cholesterol, and HDL-cholesterol grades were specified via kits purchased from DiaSys Diagnostic System GmbH, Germany. Serum glutathione (GSH), lipid peroxidation (Malondialdehyde) (MDA), and nitric oxide (NO) grades, in addition to that superoxide dismutase (SOD), and catalase (CAT) vigors were valued via kits acquired from Biodiagnostic, Dokki, Giza, Egypt.

Determination of pro-inflammatory cytokine

Using ELISA (Dynatech Microplate Reader Model MR 5000, 478 Bay Street, Suite A213 Midland, ON, Canada), serum TNF- α and IL-1 β concentricities were mensurated by mouse Elisa kits obtained from SinoGeneClon Biotech Co., Ltd, No.9 BoYuan Road, YuHang District 311112, Hang Zhou, China.

Histopathological study

Carcasses were meticulously examined at the time of slaughter, and *C. bovis*-infected hearts were taken to the pathology lab. Heart samples were then preserved for an overnight period in neutral buffered formalin (10%). The specimens were subjected to dryness in progressive $\text{C}_2\text{H}_5\text{OH}$, purified in xylene, and implanted in paraffin, and then have been sectioned tissue into thin sections measured at 5 μm . Then, tissues were deparaffinized, and then stained with H&E, finally it was examined by a light microscope (Sethiya *et al.*, 2014).

Statistical analysis

For the datum investigation, the SPSS 25 program was applied. To come across the data, medium \pm SD were listed. To statistically compare mean differences in measured variables between healthy and infected animals, a *T*-test was employed. The current data was tested at a significance level ($p \leq 0.05$).

Ethical approval

This study was licensed by the Scientific Committee of the College of Science - Al-Azhar University (Assiut Branch), Egypt. Certificate reference number: AZHAR 12/2023.

Results

The prevalence of *C. bovis* was 4.6% in the current investigation employing meat inspection, which was recorded among 455 investigated cattle in Egypt.

In Table 1 and Figure 1, the serum cholesterol, triglyceride, LDL, and HDL concentrations of infected and uninfected cattle with cysticercosis are shown. When compared to the control group, *T. saginata* cysticercosis infection caused a significantly higher level of serum cholesterol, triglycerides, and LDL while also causing a significantly lower amount of HDL.

Table 1. Mean \pm SD of lipids profile measured in the serum of healthy and *C. bovis* -infested Cattles ($n = 21$).

Parameters	Cattle		<i>p</i> - value**
	Healthy ($n = 21$)	Infected ($n = 21$)	
Cho mg/dl	82.03817 \pm 0.81649	121.7375 \pm 4.63672	0.000
Trg mg/dl	33.85599 \pm 1.61686	81.16845 \pm 1.56743	0.000
HDL mg/dl	41.52915 \pm 0.86105	36.00173 \pm 0.87883	0.000
LDL mg/dl	37.32101 \pm 0.65916	72.0606 \pm 5.46470	0.000

(**) The medium variance is important at the 0.01 scales.

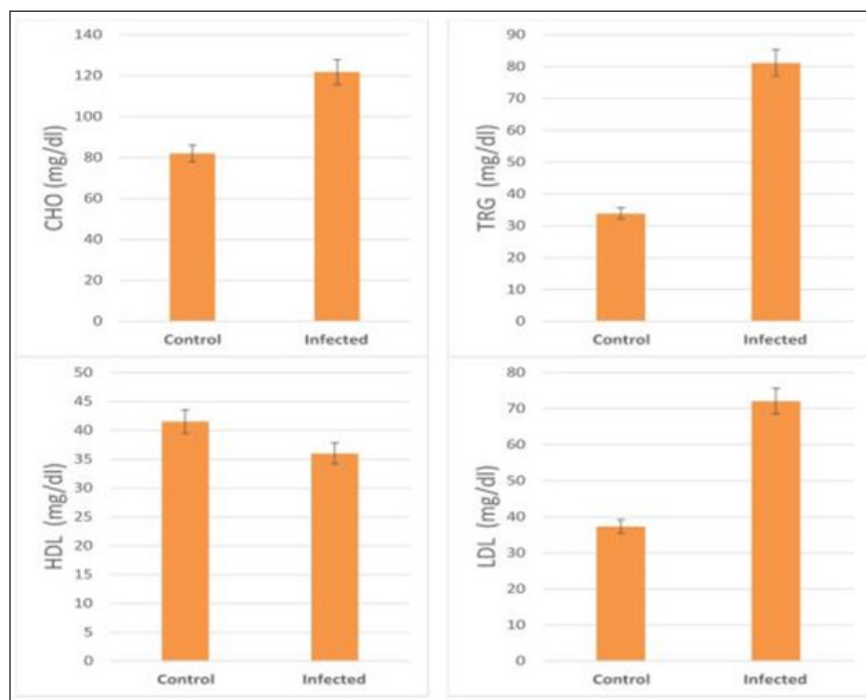


Fig. 1. Serum lipids profile in the control and infected livestock with *C. bovis*. (Mean \pm SD, $N = 21$).

Table 2. Mean \pm SD of enzymes measured in the serum of healthy and *C. bovis*-infested cattle ($n = 21$).

Parameters	Cattle		<i>p</i> - value**
	Healthy ($n = 21$)	Infected ($n = 21$)	
LDH u/l	1188.198 \pm 80.42384	1865.019 \pm 168.639	0.000
Ck-total u/l	356.6247 \pm 5.13652	407.597 \pm 24.1935	0.000
AST IU/L	68.20345 \pm 3.16010	94.98898 \pm 2.70363	0.000

(**) The medium variance is important at the 0.01 scales.

The activity of AST, CK-total, and LDH in the serum of infected cattle showed a substantial rise ($p < 0.01$, Table 2, and Fig. 2).

In cow serum infected with cysticercosis, there is a statistically significant decrease in antioxidant enzymes (SOD, CAT, and GSH). In contrast, there is a statistically significant rise in MDA and NO levels

compared to control samples, which is a biomarker of lipid peroxidation (Table 3 and Fig. 3).

Our findings demonstrated the influence of *T. saginata* cysticercosis on particular immunological markers in cattle serum. In rapprochement to the controller set, the grades of TNF- α , and IL1 β , were observed to be

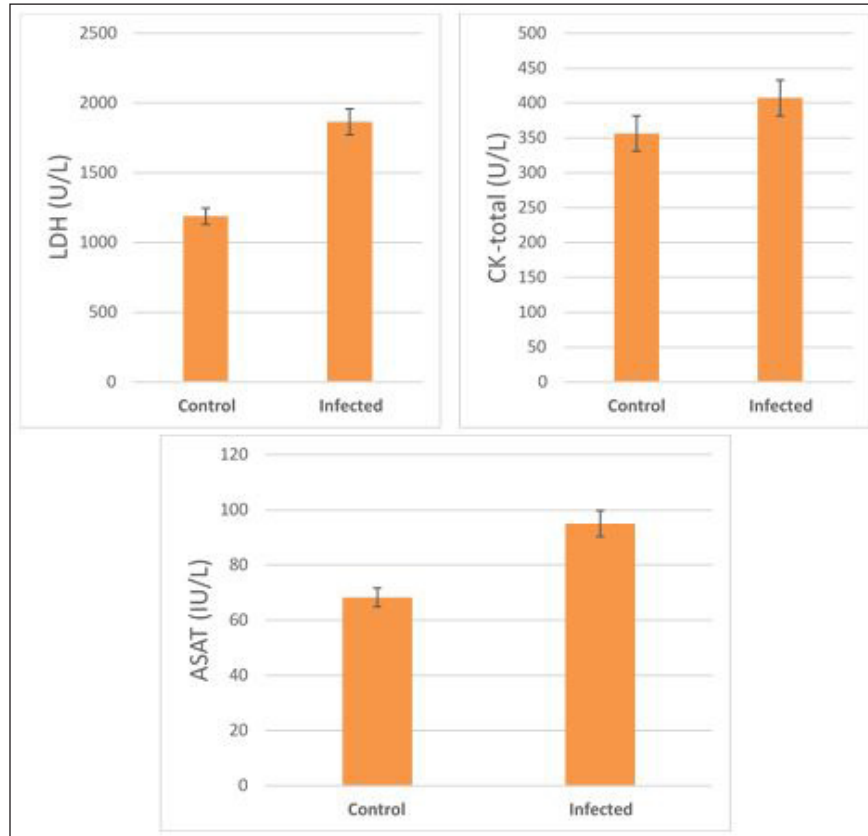


Fig. 2. Serum enzymes in the control and infected livestock with *C. bovis*. (Mean ±SD, N = 21).

Table 3. Mean ±SD of oxidative stress markers measured in the serum of healthy and *C. bovis* -infested Cattles (n = 21).

Parameters	Cattle		p- value**
	Healthy (n = 21)	Infected (n = 21)	
NO mmol/g	43.61225 ± 1.52916	56.21797 ± 2.46296	0.000
MDA mmol/g	5.469721 ± 0.66880	9.53065 ± 1.46651	0.000
SOD u/g	409.1913 ± 35.4137	150.3647 ± 15.8811	0.000
CAT u/g	1649.439 ± 225.335	1175.664 ± 86.6494	0.000
GSH mmol/g	9.069354 ± 0.77639	7.956358 ± 0.71893	0.000

(**) The medium variance is important at the 0.01.

considerably higher in the infected group (Table 4 and Fig. 4).

The histological structure of the cardiac myocytes was normal, with central oval nuclei and acidophilic sarcoplasm. In the intercellular gaps, a few tiny blood capillaries could be seen (Figs. 5 and 6). A cross-section of a proglottid of *T. saginata* revealing thick outer tegument, degenerating myocardiocytes surrounding it, and altered histological architecture was visible in the cardiac muscle tissue of infected calves. Mononuclear

infiltration, cardiac oedema, and hemorrhage were all signs of a mild inflammatory reaction (Figs. 7 and 8).

Discussion

In this study, 4.6% of cattle were infected with *C. bovis*. Our outcome is deemed upper than formerly logged by Haridy *et al.* (1999) (0. 23%), Rodriguez- Hidalgo *et al.* (2003), (0. 37%), Abdo *et al.* (2009) (1. 65%), Kandil *et al.* (2012a,b) (4%) and (4.4%), respectively, Yassien *et al.* (2013), (0.58%), El-Alfy *et al.* (2017) (0.51%),

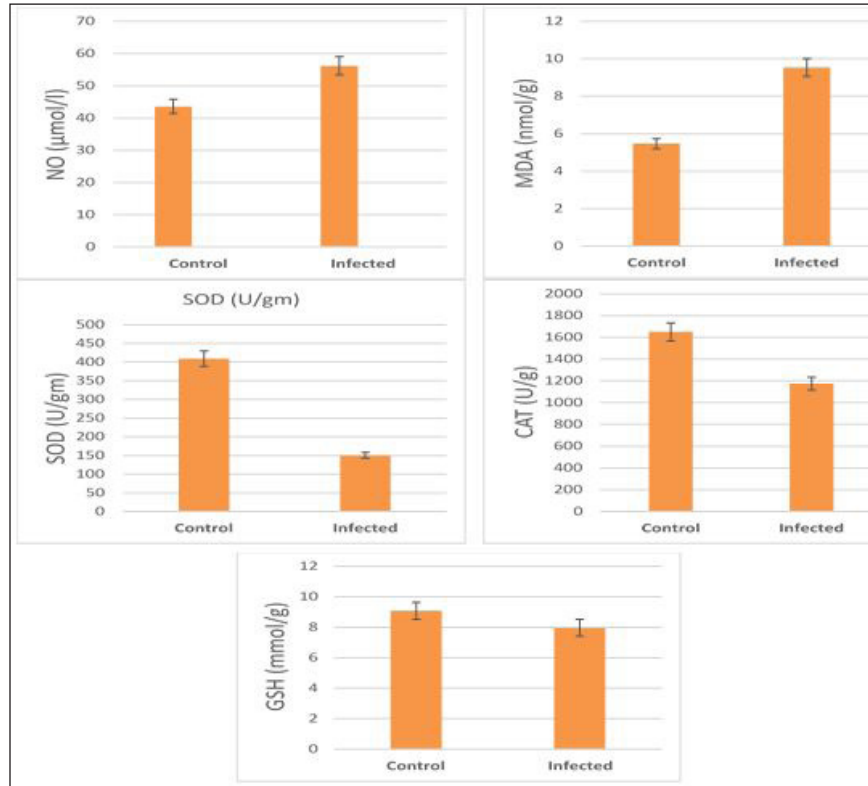


Fig. 3. Serum oxidative stress in the controller and infected livestock with *C. bovis*. (Mean \pm SD, $N = 21$).

Table 4. Mean \pm SD of pro-inflammatory cytokines measured in the serum of healthy and *C. bovis*-infested Cattles ($n = 21$).

Parameters	Cattle		<i>p</i> -value**
	Healthy ($n = 21$)	Infected ($n = 21$)	
TNF- α ng/ml	29.2788 \pm 1.42620	42.93221 \pm 2.23084	0.000
IL1 β pg/ml	90.1426 \pm 4.60334	118.3686 \pm 4.45166	0.000

(**) The medium variance is important at the 0.01 scale.

and Abdel Aziz *et al.* (2022) (0.47%). Contrarily, our study found fewer instances of *C. bovis* than did Oryan *et al.* (1995) (7.7%), Abdel-Hafeez *et al.* (2015) (20%), and Dyab *et al.* (2017) (7.5%). The variance in infection rates may be brought on by epidemiological elements such as climate, various sanitation practices, the number of samples taken, and the use of control methods and programmes (Usip *et al.*, 2011). Increased values of cholesterol, triglycerides, and LDL of infected cattle with decreased HDL were seen in this study. These findings conflict with those of (Kandil *et al.*, 2012b; Saeed *et al.*, 2016), who discovered that total cholesterol was present in the sera of infected cattle in low concentrations. Decreases in cholesterol may result from the function that cholesterol moves in aetiology by allowing the grubs to survive in steward

tissues, as suggested by Kandil *et al.* (2012b), or they may result from commotions in hepatic enzymes and changes in hormonal exudation triggered by parasite existence. Furthermore, it was shown that adding cholesterol to the RPMI-1640 culture medium increased the survival of ascariasis larval growth, and it may be that certain laborers or ferments enable the parasite to break down and ingest fats/cholesterol (Urban *et al.*, 1984; Wiedermann *et al.*, 1991). In contrast, Scare *et al.*, (2019) found that the addition of cholesterol did not improve worm longevity or viability. The activity of enzymes in the serum of infected cattle showed values higher than uninfected ones. Hepatic impairment may result from an abundance of *C. bovis* (Scandrett *et al.*, 2009). Our discoveries are regular with those of (Pathak and Gaur 1981; Bamorovat

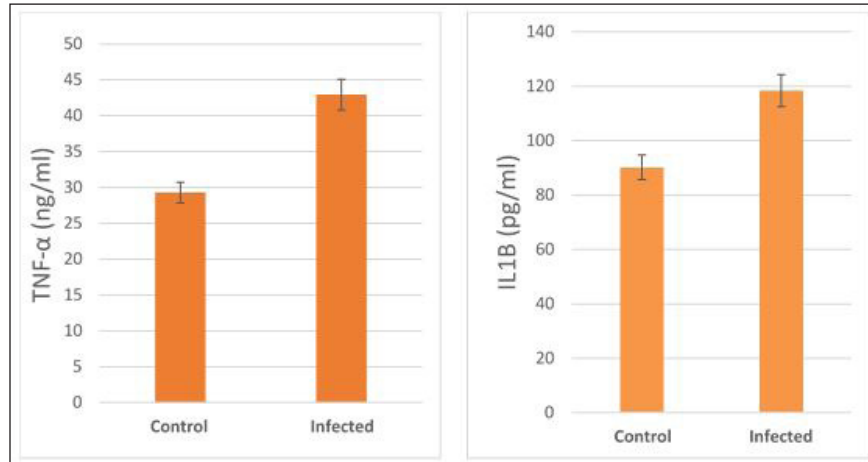


Fig. 4. Serum pro-inflammatory cytokines in the control and infected livestock with *C. bovis*. (Mean ±SE, $N = 21$).

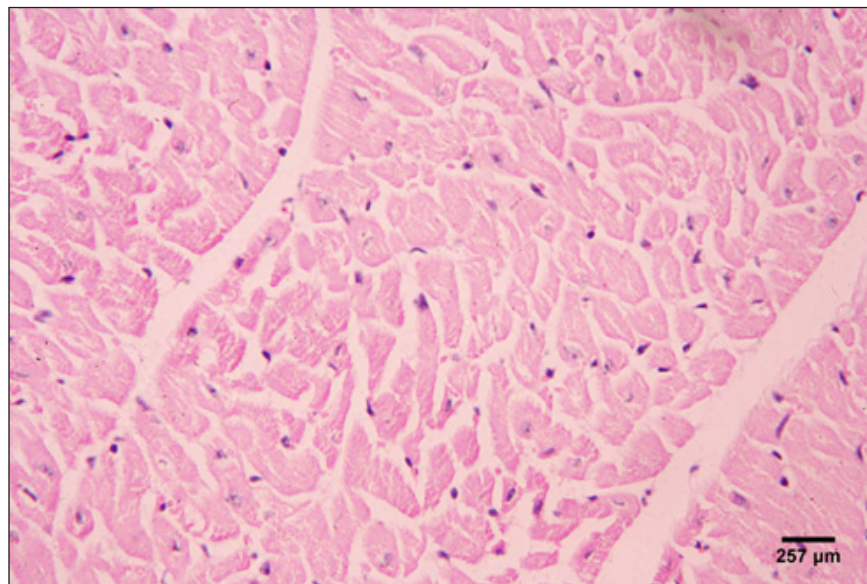


Fig. 5. Photomicrograph of the ventricle of control cattle demonstration of a regular arrangement of cardiac muscle fibers, acidophilic sarcoplasm and central oval vesicular nuclei of the cardiac myocytes (H&E, low power).

et al., 2014), who found that goats empirically disease-ridden with *Cysticercus tenuicollis* had elevated AST, ALT, and ALP levels. They claimed that the significant loss of liver parenchymal cells brought on by grubs transmigration may be the reason for this increase. ALT, AST, and ALP levels rose in *C. tenuicollis*-infective pigs, according to Pathak *et al.* (1984), who also linked these alterations to the metacestode's migration. Infected swine with *C. cellulosae* had significantly higher levels of ALT, AST, ALP, and LD, according to Pathak and Gaur (1985), who speculated that these alterations might be the result of muscle tissue being

harmed by this metacestode. In addition to that ALT, AST, ALP, and GGT levels rose in adult goats infected with liver flukes, according to Aslam *et al.* (2020), who also linked these alterations to the larval migration. Also, Singh *et al.* (2004) reported an increase of AST levels 2 weeks post-infection synchronizing with the migratory phase of *Fasciola hepatica* in the liver parenchyma. They reported that liver damage is the most important cause of the increase in serum ALT activity in the infected sheep. While Radfar *et al.* (2014) noted a notable rise in ALT, AST, and ALP in goats with *C. tenuicollis* infection. Calves that had

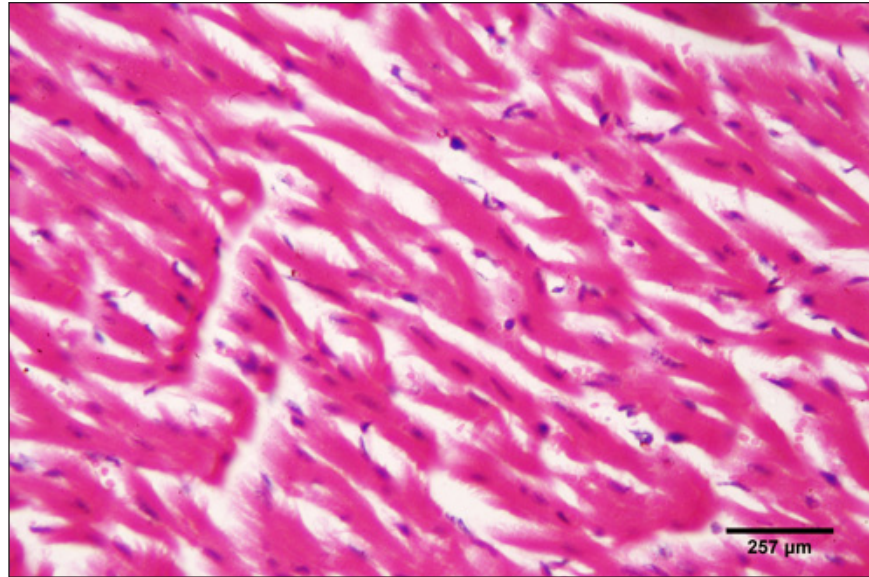


Fig. 6. Photomicrograph of the ventricle of control cattle demonstration of normal cardiac myocytes with acidophilic sarcoplasm and central oval vesicular nuclei and normal histological appearance (H&E, high power).

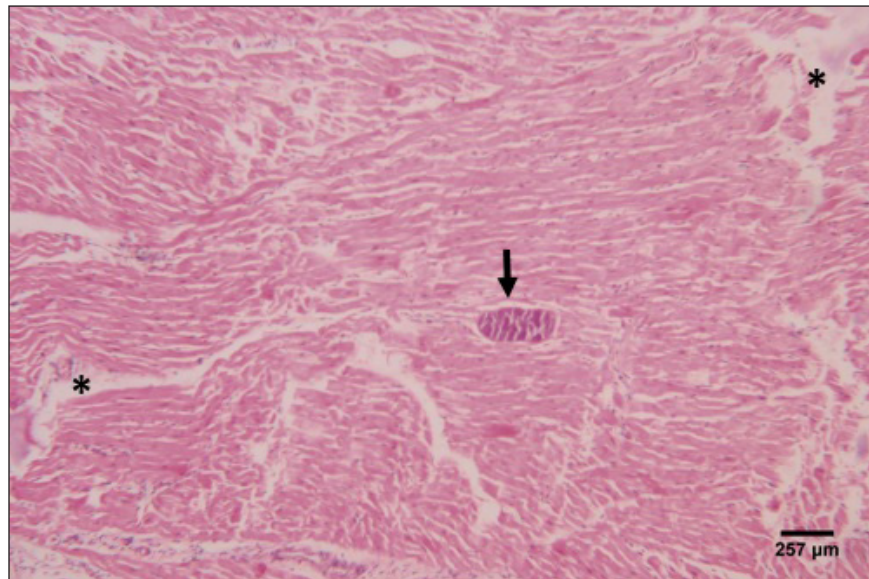


Fig. 7. Photomicrograph of the ventricle of infected cattle demonstration irregular and disturbed arrangement of cardiac muscle fibers with mild cardiac edema (*). cross-section of a proglottid of *Taenia sp.* showing thick outer tegument (short arrow) (H&E, low power).

been empirically infected with 85 thousand and 200 thousand *T. saginata* ova showed elevated CK activity, according to (Blazek *et al.*, 1985; Kursa *et al.*, 1986). In addition to that, Anwar *et al.* (2024) showed a significant increase in LDH activity in infected animals. On the other hand, the current findings conflict with (Kandil *et al.* 2012b; Saeed *et al.*, 2016). According to (Pinzani and Rombonts, 2004; Otto *et al.*, 2010),

some large digits of cysts and consequent continuing deterioration of the liver parenchyma may be to blame for the drop in AST activity in infected cattle.

In our results, cattle serum infected with cysticercosis shows a decrease in antioxidant enzymes, while oxidative stress levels rise. These findings concur with (Atteya *et al.*, 2015). When parasites are present, host cells typically suffer an oxidative assault. According to

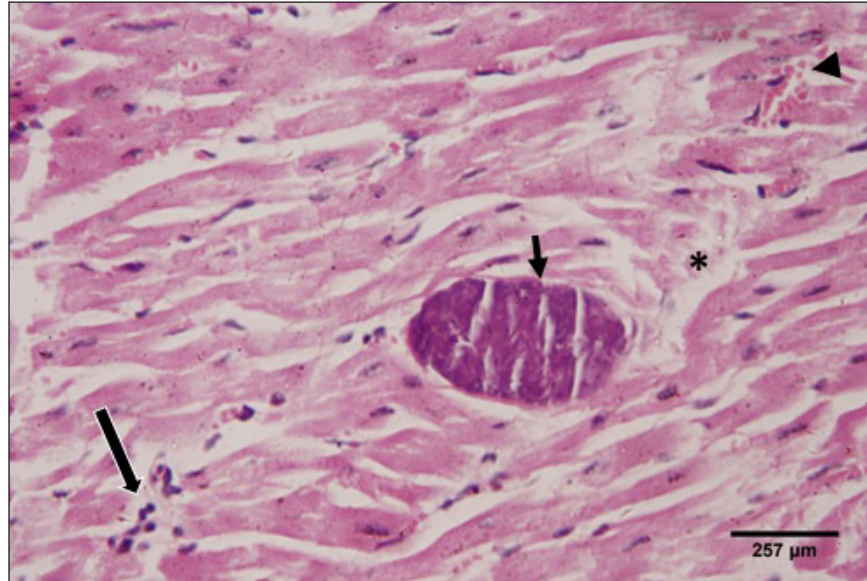


Fig. 8. Photomicrograph of the ventricle of infected cattle demonstration irregular and disturbed arrangement of cardiac muscle fibers. Few myocytes represent degenerated nuclei and minimal vacuolization of the cytoplasm with a disarrangement pattern (*) around the proglottid of *Taenia sp.* showing thick outer integument (short arrow). Mild haemorrhage was noticed (arrow-head) and infiltration of inflammatory cells (long arrow) (H&E, high power).

Kataria *et al.* (2010), alterations in antioxidant capacity (oxidative stress) contribute to parasite persistence in host tissues and are followed by pathological changes and damage that result in the development of various clinical disorders.

When compared to healthy animals, cows and buffalo with fascioliasis and cysticercosis had lower antioxidant values. According to El-Moghazy (2011) and Atteya *et al.* (2015), while MDA levels are rising, CAT, SOD, GSH, and GST activity is declining. Infected sheep with distomatosis had lower levels of non-enzymatic antioxidants and antioxidant ferment actions in their livers than the non-infected group (Kolodziejczyk *et al.*, 2005; Değer *et al.*, 2008). Total antioxidant status decreased, indicating a reduction in the antioxidant capability of the *Fasciola hepatica*-infected rat liver tissue. Increased oxidative alterations of lipids and proteins were the outcome of diminished antioxidant capacities (Siemieniuk *et al.*, 2008). Additionally, there is a statistically considerable rise in the action of the antioxidant ferment SOD in the *T. saginata*-diseased skeletal muscles compared to the non-infected cattle's control value (Luszczak *et al.*, 2011).

Reactive oxygen species (ROS) are released as a result of altered liver and heart parasites' antioxidant capacities, causing damage to cell membranes and other components that ultimately result in cell death. Once the pathogen has been eradicated, the cell is equipped with several antioxidant defense enzymes

that protect the cell from oxidative damage (Belkaid, 2007). To defend their tissues from inflaming damage, hosts have an administrative web built on cytokines that control the resolve of inflaming (Belkaid, 2007).

Our findings demonstrated that $\text{TNF-}\alpha$ and $\text{IL1}\beta$ were higher in the infected group. The current returns are compatible with Palma *et al.* (2019), who examined the variations in the oncosphere growth of *Taenia solium* and *T. saginata*. Analyze the manifestation of cytokines and MMPs in human peripheral blood mononuclear cells (PBMCs) after these activated oncospheres (AO) and postoncospherical (PO) antigens stimulated them. Rats received intracranial inoculations of the AO and PO styles of either *T. solium* or *T. saginata*. Neurocysticercosis (NCC) was manifested in rats infected with the AO and PO shapes of *T. solium* but not in those infected with *T. saginata*. Antigens of the AO and PO shapes of both species were used to stimulate human PMBCs, and the amount of cytokines and MMPs produced was assessed. In comparison to *T. saginata* AO, the *T. solium* AO antigen triggered a greater generation of IL-4, IL-5, IL-13, IFN- γ , and IL-2 cytokines. When contrasted to *T. solium*, the PO antigen from *T. saginata* boosted the construction of the cytokines IL-4, IL-5, IL-13, IFN- γ , IL-1 β , IL-6, IL-10, TNF- α , and IL-12.

Due to the early inflammatory response in cardiac muscle and the increased visibility of macroscopic lesions, the cardiac was validated as the ideal location

for the diagnosis of cysticercosis (Scandrett *et al.*, 2009). Heart inflammation brought on by the emergence of a pseudoepithelial boundary and a granulation tissue zone was the heart's reaction to the presence of *C. bovis*. Later, when the *Cysticercus* began to experience necrotic alterations, a new type of inflammatory response began to emerge (Sterba *et al.*, 1979; Kumar *et al.*, 2013). In the current investigation, routine H&E staining and histological analysis of the control group's heart muscles revealed a regular arrangement of cardiac muscle fibers with what appeared to be normal striations and branching. The histological results revealed significant alterations in the tissues of infected livestock. These findings are consistent with those of (Atteya *et al.*, 2015), who demonstrated that microscopic lesions in the heart included cellular infiltration of lymphocytes, eosinophils, plasma cells and macrophages, necrosis of the tissue, proliferation of fibroblasts and development of granulation tissue along with the disintegration of cysts. Hashemnia *et al.* (2016) examined the ovine cysticercosis pathological lesions in sheep that had been killed. Severe degenerative and necrotic changes in the muscle fibers and also the sections of cysticerci in the affected organs were surrounded by a zone of degenerative, necrotic changes and polymorphonuclear leucocytes, lymphocytes, and fibroblasts were the predominant histopathological abnormalities. Abdel Aziz *et al.* (2022) showed that, severe fibrosis among cardiac muscles, fibrous tissue infiltrated with mononuclear inflammatory cells, and necrosis and atrophy in cardiac myocytes.

Conclusion

In the epilogue, cysticercosis has detrimental influences on the serum biochemical profile in livestock, and trialing these factors can be used as a backup experiment to diagnose *T. saginata* cysticercosis in lifelike livestock and assess the precision of the flesh-checking transaction for detecting cysticercosis in livestock cadavers.

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Conflict of interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Authors' contribution

AA provided the study's concept, while the methodology, validation, and formal analysis were created by NA. The investigations, preparation of the original draft, revision of the manuscript, and review and editing of the paper were carried out by AA and

AMK. The version that was presented and written by all authors has been confirmed.

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Data availability

Every dataset is included in the publication.

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