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Research Article

Histopathological and Biochemical Evaluation of Albendazole in the Treatment of Infected Mice with Hydatid Cyst

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ABSTRACT

Introduction: Hydatidosis, caused by the larval stage of *Echinococcus granulosus*, is a prevalent parasitic disease affecting both humans and animals. Albendazole is currently the most effective drug for treating hydatid cysts. This research aimed to investigate the histopathological and biochemical effects of Albendazole on the liver, lung, and kidney of mice experimentally infected by hydatid cysts.
Materials and methods: A total of 20 mice weighing approximately 220 g were used. The rats were randomly divided into the Albendazole group (100 mg/kg/day) and the control group (infected Rats without treatment). At the end of the experiment, tissue samples from the liver, lung, and kidney were collected for histopathological evaluation. Liver blood tests were used to assess liver functions or liver injury (alkaline phosphatase, alanine aminotransferase, and bilirubin).
Results: After 30 days of daily treatment, the total number of cysts, size, and weight of the largest cyst were significantly lower in the Albendazole group, compared to the control group. The study addressed histopathological changes in the liver, kidneys, and lungs caused by hydatid cysts, such as tissue necrosis, hemorrhage, and local inflammation, indicating the potential for serious complications and significant damage to these organs. The group treated with Albendazole showed severe histopathological changes in the liver, kidneys, and lungs, compared to the control group. This suggests that Albendazole may trigger a more aggressive response in these organs to the cysts, leading to increased tissue damage. In addition, alkaline phosphatase, alanine aminotransferase, and bilirubin concentrations revealed a significant increase in the Albendazole group.
Conclusion: While Albendazole is an effective drug for treating hydatidosis, it can also cause severe side effects on various organs in the body. Therefore, alternative treatment strategies need to be developed to minimize these adverse effects.

1. Introduction

Hydatid cyst is one of the common parasitic diseases between humans and animals, which is caused by *Echinococcus* tapeworms[1]. This disease is transmitted by eating water or food containing the eggs of this parasite or through contact with an infected animal[2]. The eggs of this parasite are found in the feces of carnivorous animals infected with this parasite[3]. Dogs, foxes, and wolves are the animals that are usually infected with this parasite. These animals get sick when they eat animal parts infected with this cyst, including sheep or from the rodent family[4]. The type of disease in humans can vary depending on the type of cyst caused by echinococcosis. Therefore, the specific type of echinococcosis responsible for the infection determines the type of disease that occurs[5]. The disease is usually diagnosed through ultrasound, but a CT scan or MRI can also be used [6]. A blood test that checks for antibodies against this infection and imaging can also be effective [7]. There are three primary approaches to treating hydatid cysts, including drug therapy, surgery (extracting the cysts from the body), and the Puncture, Aspiration, Injection, and Re-aspiration (PAIR) method, which involves draining the fluid from the cysts [8,9]. Although surgery or the PAIR method can treat the disease, there is a chance

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of recurrence. Therefore, taking medication for an extended period can serve as a preventive measure [10].

Currently, the most effective and widely used drug for treating hydatid cysts is Albendazole [11]. Based on research studies, this benzimidazole derivative has been extensively used in veterinary medicine and is considered a safe broad-spectrum drug in both humans and animals [12,13]. Albendazole binds to beta-tubulin in the helminth and prevents its polymerization, thereby hindering glucose absorption in the parasite [14,15]. Moreover, its inhibitory effect on tubulin polymerization leads to a reduction in cytoplastic microtubules [16]. Albendazole is commonly used before and after surgery as a routine treatment for hydatid cysts [17,18]. However, it may cause adverse effects, such as headaches, increased pressure in the cerebral ventricles, dizziness, itching, urticaria, Stevens-Johnson syndrome, and more [19]. Additionally, reports have indicated its impact on liver, lung, and kidney organs [20]. Albendazole has been found to cause a significant decrease in hepatic alkaline phosphatase and lactate dehydrogenase enzymes and some pathological lesions, including periportal necrosis, cholestasis, and periportal fibrosis in the liver [21,22]. Furthermore, it can cause hepatocellular necrosis and fibrosis, lymphocytic infiltration in the lungs, and changes in renal function and nephrotoxicity parameters [23]. The present study aimed to examine the effects of Albendazole treatment on hydatid cyst-infected mice through biochemical and histopathological evaluation of the liver, lung, and kidney.

2. Materials and Methods

2.1. Ethical approval

All procedures were approved by the Animal Care Committee of Veterinary Medicine, Ferdowsi University, Mashhad, Iran. The principles of laboratory animal care were followed, and specific international rules and regulations were observed.

2.2. Collection of protoscoleces

The protoscoleces needed to infect mice were sterilely collected from hydatid cysts in sheep liver prepared at Mashhad slaughterhouses. Protoscoleces were examined by observing the movement of cytoplasm under the light microscope (Olympus, Japan) and staining with 10% eosin × 40 magnification.

2.3. Animals

A total of 20 mice with a maximum age of 2-3 months with an average weight of 220 g were bought from the laboratory animal unit of Ferdowsi University, Mashhad, Iran, and kept in laboratory conditions with free access to water and commercial food daily. Experimental animals were kept in standard cages with a minimum of 50% humidity, 24°C temperature, and 12 hours of dark/light cycle with appropriate ventilation in a particular cage. To infect each mouse, 1500 Protoscolex was injected intraperitoneally into half a milliliter of physiological serum containing penicillin. Five months after infection, the existing mice were randomly divided into two equal groups. Mice were randomly divided into the Albendazole group (100 mg/kg/day) and a control group, in which they were infected without any treatment.

2.4. Histopathological examination

Upon completion of the experimental period, all the mice were euthanized and necropsied to obtain tissue samples. Following a macroscopic examination of the organs, the livers, lungs, and kidneys of each group of mice were placed separately in a 10% formalin solution to ensure proper fixation of the tissue specimens for subsequent histopathological studies were performed at a pathology laboratory. To evaluate tissue damage, 10-12 microscopic fields were observed under a light microscope, with different magnifications used according to the type of lesion. Subsequently, a grading system ranging from - to +++ was used to assign grades to the observed lesions, with - denoting the absence of any lesion and +++ indicating the presence of a severe lesion.

2.5. Biochemical examination

In order to prevent blood clotting, blood was immediately taken from the heart of the mice with a 2 ml syringe and slowly transferred into the tube. To prepare serum, the blood sample without anticoagulant was placed in a centrifuge and centrifuged at 3000 rpm for 10 minutes. After removing the sera with a sampler and placing them inside the microtube, the samples were placed in a freezer (-80°C), and the values of biochemical factors, including alkaline phosphatase (ALP), alanine aminotransferase (ALT), aspartate aminotransferase (AST) and total bilirubin, were immediately measured.

2.6. Statistical analysis

The data from this study were transferred to SPSS software version 23 for statistical analysis and were compared using the Kruskal-Wallis test. The Mann-Whitney test was used to compare the groups. In this study, a p-value less than 0.05 was considered statistically significant.

3. Results

3.1. Parasite treatment

The study evaluated the effectiveness of Albendazole in reducing the growth and size of hydatid cysts in the liver. The results showed that after 30 days of daily treatment, the Albendazole group had a significantly lower number of cysts, smaller cyst size, and lower weight of the largest cyst, compared to the control group (Figure 1). This suggests

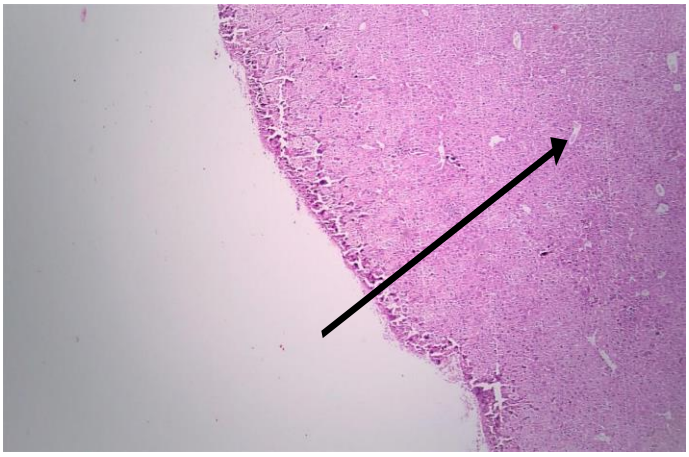

Figure 1. Parasite cyst in liver tissue (arrowhead) of the control group (H&E, x 40)

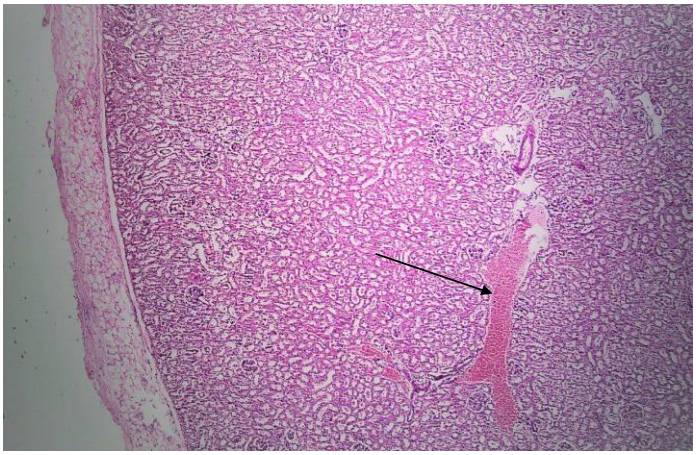

Figure 3. Kidney tissue hyperemia (arrowhead) in the control group (H&E, x 40)

that Albendazole effectively reduced the growth and size of hydatid cysts in the liver.

3.2. Histopathological findings

3.2.1. Control group

The study also addressed histopathological changes in the liver, including tissue necrosis, hemorrhage, and local inflammation caused by hydatid cysts (Figure 2). The histopathological changes observed in the liver highlight the potential for serious complications associated with hydatid cysts, which can cause significant damage to liver tissue and other organs.

The investigation also documented histopathological alterations in the kidneys, such as tissue necrosis, hemorrhage, and local inflammation, induced by hydatid cysts (Figures 3-6). These histopathological changes underscore the potential for grave complications associated with hydatid cysts, as they can substantially harm kidney tissue and other organs.

In the histopathological study of the lungs, no changes were seen.

3.2.2. Albendazole group

Histopathological changes in the liver, including tissue necrosis, hemorrhage, and local inflammation, were significantly more severe in the Albendazole group, compared to the control group (Figure 7). This suggests that Albendazole may cause a more aggressive response in the liver to the cysts, leading to increased tissue damage.

The findings of this study demonstrate that the Albendazole group exhibited significant histopathological alterations in the kidneys, characterized by tissue necrosis, hemorrhage, and localized inflammation, compared to the control group (Figure 8). These results suggest that Albendazole may have induced a more robust response in the kidneys to the cysts, culminating in heightened tissue damage.

The current investigation indicated significant histopathological modifications in the lungs, including tissue necrosis, hemorrhage, and local inflammation, in the Albendazole group, compared to the control group (Figures 9, 10). These findings suggest that Albendazole may provoke a more aggressive response in the lungs to the cysts, resulting in increased tissue damage.

3.3. Biochemical findings

Elevated levels of ALT, ALP, AST, and bilirubin were detected in the Albendazole group, indicating the

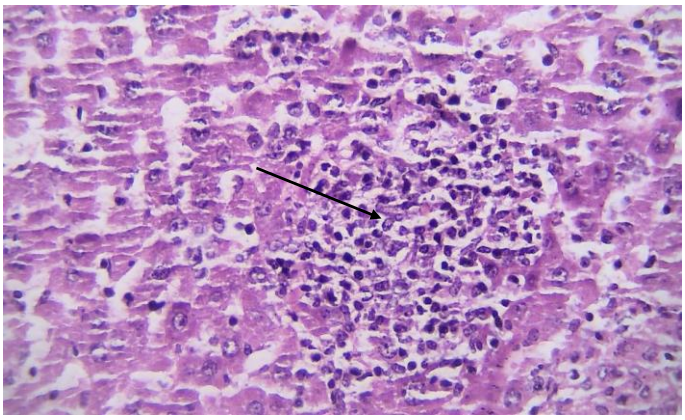

Figure 2. Inflammation in the liver tissue of the control group (H&E, x 400)

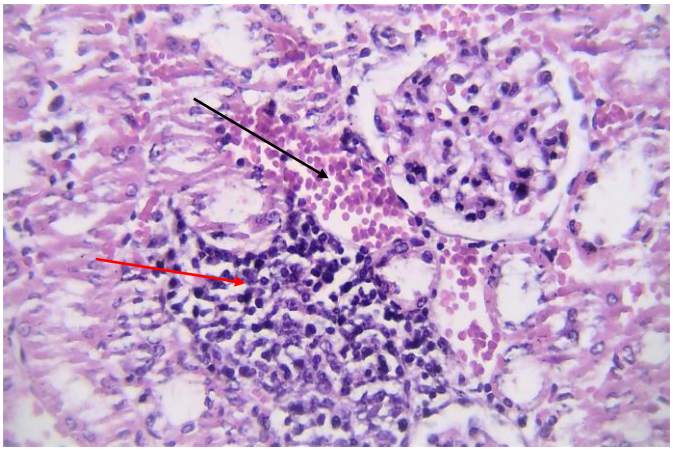

Figure 4. Bleeding (black arrow) and inflammation (red arrow) in the kidney tissue of the control group (H&E, x 400)

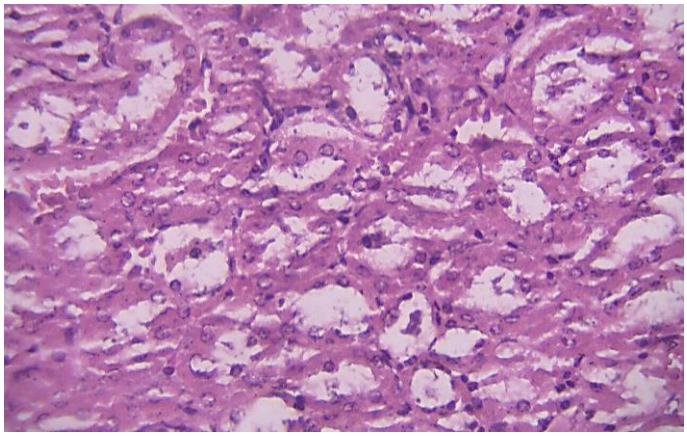

Figure 5. Cell swelling in the epithelial cells of the tubes in the kidney tissue (arrowhead) of the control group (H&E, x 400)

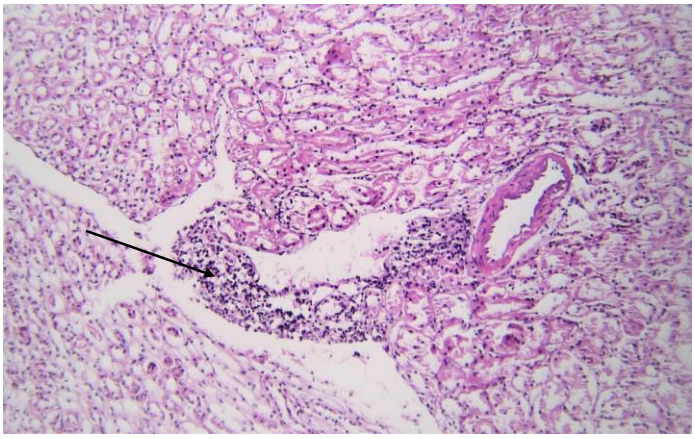

Figure 8. Inflammation (arrowhead) of kidney tissue of Albendazole 100 mg/kg group (H&E, x 100)

possibility of Albendazole-induced liver damage or dysfunction (Table 1). No significant differences were observed between albendazole and control groups for ALT activity and bilirubin concentration. The ALP activity showed no significant difference in Albendazole and the control group. The ALT and ALP are enzymes produced by liver cells and are released into the bloodstream when liver cells are damaged. Bilirubin, a byproduct of red blood cell breakdown, is usually processed and eliminated by the liver, but its accumulation in the bloodstream indicates liver damage. It is important to note that while Albendazole effectively reduces the growth and size of hydatid cysts, it may also cause liver damage or dysfunction. Therefore, close monitoring of liver function is necessary during Albendazole treatment.

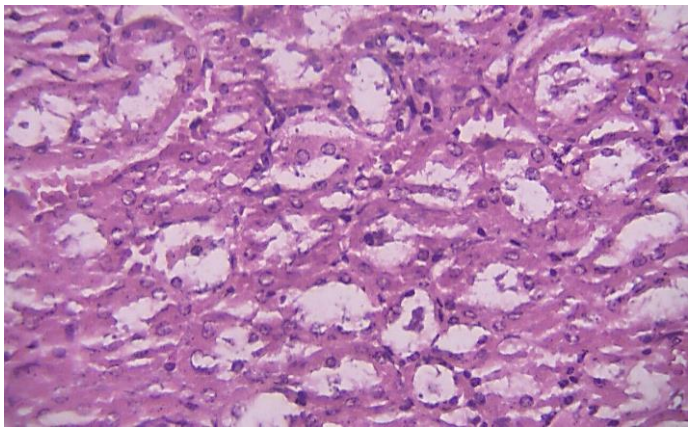

Figure 6. Necrosis of tubular epithelial cells in kidney tissue of the control group (H&E, x 400)

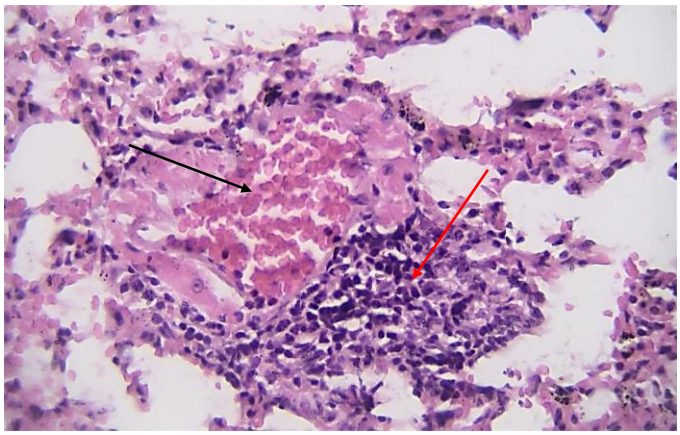

Figure 9. Hyperemia (black arrow) and inflammation (red arrow) in lung tissue of Albendazole 100 mg/kg group (H&E, x 400)

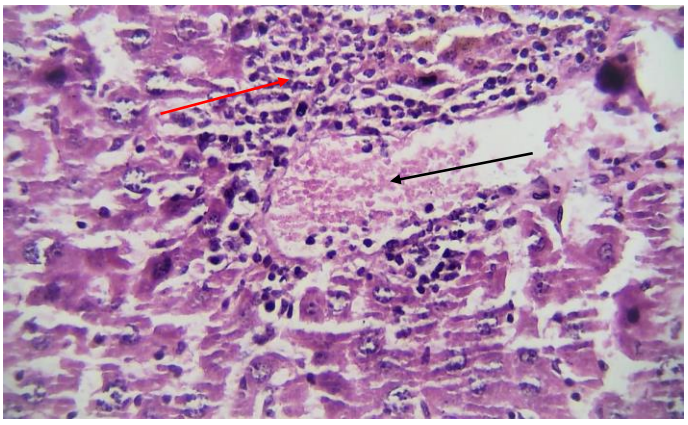

Figure 7. Hyperemia (black arrow) and inflammation (red arrow) in liver tissue of Albendazole 100 mg/kg group (H&E, x 400)

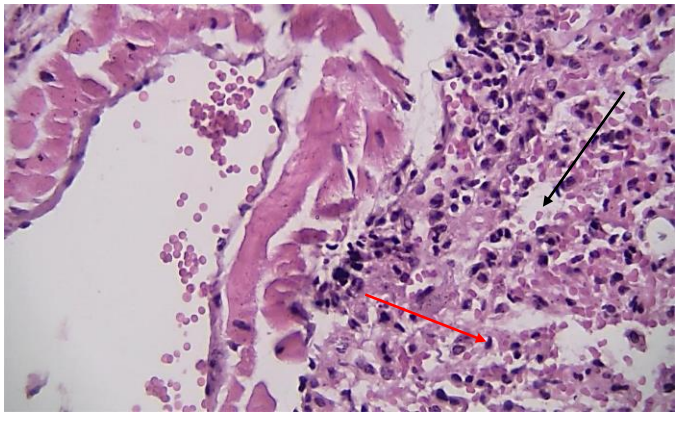

Figure 10. Bleeding (black arrow) and the presence of inflammatory cells (red arrow) in the lung tissue of Albendazole 100 mg/kg group (H&E, x 400)

Table 1. The serological findings of mice on day 30 of the study

Group	Parameter			
	ALT (U/L)	ALP (U/L)	AST (U/L)	Bilirubin (mg/dl)
Control	125	250	332	0.48
Albendazole 100 mg/kg	139	285	273	0.43

ALP: Alkaline phosphatase, ALT: Alanine aminotransferase, AST: Aspartate aminotransferase

4. Discussion

Albendazole is a widely used drug for treating echinococcosis, but its efficacy and safety are limited by various factors. One of the main limitations of Albendazole is its poor absorption from the gastrointestinal tract, which leads to low hepatic concentrations of the drug [24]. This poor drug absorption results in lower efficacy in treating echinococcosis, leading to the recurrence of cysts after Albendazole treatment. Another significant limitation of Albendazole is its high cost, which makes it inaccessible to many patients in resource-limited settings [25]. Moreover, the drug can cause severe side effects, such as hepatic toxicity, which can be life-threatening in some cases [26]. Additionally, Albendazole resistance has been reported in some cases of echinococcosis treatment, which makes it less effective in treating the infection [27].

To overcome the limitations of Albendazole, it is essential to explore new treatment strategies for echinococcosis. Given the limitations of Albendazole, herbal medicines have gained increasing attention as alternative treatment options for echinococcosis. One approach is to investigate the potential of herbal medicines and natural products for the treatment of the disease [28]. These alternatives could offer a safe and cost-effective treatment option, especially in resource-limited settings where Albendazole may not be affordable or readily available [29]. In addition to the limitations and adverse effects discussed above, there are other factors that affect the effectiveness of Albendazole in the treatment of echinococcosis. For instance, the size, location, and number of cysts, as well as the patient's immune status, can all influence the response to treatment [30]. Furthermore, the duration and dosage of Albendazole treatment may also affect its efficacy [31]. Moreover, there are concerns about the safety of Albendazole in pregnant women, as the drug has been shown to be teratogenic in animal studies. Therefore, caution is advised when using Albendazole in pregnant women, and alternative treatment options should be considered [32]. Another issue related to Albendazole use is the potential for drug interactions, which can affect the pharmacokinetics and efficacy of the drug. For example, Albendazole can interact with drugs that induce or inhibit the cytochrome P450 system, leading to altered drug metabolism and potentially suboptimal therapeutic outcomes [33].

Given the complex nature of echinococcosis and the limitations of Albendazole, it is crucial to explore alternative treatment options for this disease. One promising approach is the use of combination therapy, which involves the simultaneous or sequential administration of two or more drugs with different mechanisms of action [34]. This strategy has been shown to improve the efficacy of Albendazole in some studies, and it may overcome the limitations of the drug [35]. Moreover, further studies are necessary to explore the mechanisms underlying the adverse effects of Albendazole on liver tissue and to identify new therapeutic targets for echinococcosis. The development of novel drugs or drug combinations that are more effective, safer, and affordable could significantly impact the management of echinococcosis and improve the quality of life of affected individuals.

5. Conclusion

In conclusion, this study sheds light on the efficacy of Albendazole in treating hydatidosis, a parasitic disease that affects both humans and animals. The research findings suggest that Albendazole effectively reduces the size and weight of hydatid cysts, but also induces significant histopathological alterations and biochemical changes in the liver, lungs, and kidneys of mice. The study highlights the potential for serious complications and damage to these vital organs caused by hydatid cysts and the need for alternative treatment strategies that minimize adverse effects. Further research is required to determine the long-term effects of Albendazole treatment and to develop new drugs that are effective and safe for treating hydatidosis.

Declarations

Competing interests

The authors have declared no conflicts of interest.

Authors' contributions

The authors of this manuscript contributed to various aspects of the research project. Zahra Mousavi was responsible for the conceptualization of the study, while Mohammad Heidarpour and Hassan Borji contributed to the methodology and formal analysis. Zahra Mousavi provided supervision throughout the research project. All authors were involved in the writing of the original draft, as well as the review and editing of the manuscript. Additionally, all authors have read and approved the final version of the manuscript for publication in the present journal.

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Ethical considerations

The authors declare that this manuscript is original and

has not been submitted elsewhere for possible publication. The authors also declare that the data used/presented in this manuscript has not been fabricated.

Availability of data and materials

The data presented in this study are available on request from the corresponding author.

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None.

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