

Immunohistochemical changes in CD-2 expression in different organs of rats treated with nuclear particles versus hydatidosis

O.A. Najm¹, S.Yakoob Yousif¹, Yasir Yahya Kassim²

¹Department of Biology, ²Department of Physics, College of Education for Pure Science, University of Mosul, Mosul Iraq

Article information

Article history:

Received October 8, 2025
Accepted November 17, 2025
Published July 5, 2026

Keywords:

Echinococcosis
Protoscoleces
Nuclear particles
CD2 expression
Immunomodulation

Correspondence:

O.A. Najm
omaimaadil@uomosul.edu.iq

Abstract

Hydatidosis, a zoonotic disease caused by *Echinococcus granulosus* larvae, is a significant health problem due to cysts localization and drug resistance. Novel approaches such as nuclear particles, alpha particles, may reduce parasite viability and enhance host immunity. This study evaluated the effects of nuclear particles irradiation on protoscoleces isolated from hydatid cysts and its impact on histopathological changes and CD2 expression in the liver, spleen, and intestine of experimentally infected rats aiming of using it as a treatment against hydatid disease. Protoscoleces were isolated from sheep livers and irradiated with nuclear particles at energy (3.3MeV) for either 1-hour or 1.5-hours. Thirty-six male rats were divided into six groups and injected intraperitoneally with irradiated or non-irradiated protoscoleces (groups 1 & 2 protoscoleces irradiated for 1 hour, groups 3 & 4; protoscoleces irradiated for 1.5-hours, groups 5 & 6: protoscoleces non-irradiated). Rats were euthanized for four and five months. Histopathological examinations and CD2 immunohistochemical reactivity were performed to assess tissue integrity and T-cell responses. Rats receiving 1.5-hour irradiated protoscoleces exhibited significantly less histopathological damage in the spleen, intestines and liver with notably stronger CD2 expression (score 3+) at five months, indicating enhanced Natural Killer (NK) cells activation. Tissue architecture was better preserved compared to another groups suggesting a dose-dependent benefit. The present study concluded that nuclear particles irradiation function as a dual parasitocidal and immunomodulatory strategy which could inform future therapies for hydatidosis. Further studies measuring cytokines and other immune markers are recommended to clarify the underlying mechanisms for hydatidosis control.

DOI: [10.33899/ijvs.2025.165936.4551](https://doi.org/10.33899/ijvs.2025.165936.4551), ©Authors, 2026, College of Veterinary Medicine, University of Mosul.

This is an open access article under the CC BY 4.0 license (<http://creativecommons.org/licenses/by/4.0/>).

Introduction

Cystic echinococcosis, caused by the larval stage of *Echinococcus granulosus* remains a serious zoonotic and public health problem particularly in livestock-raising regions (1,2). The liver and lungs are the most affected organs though cysts can appear in any tissue leading to diverse clinical manifestations and chronic inflammatory responses (3-5). Standard treatments primarily include surgery combined with anthelmintic drugs such as

albendazole and mebendazole (6). However, surgical inaccessibility cyst, multiplicity and resistance to medical therapy have driven the search for alternative or adjunctive approaches (7).

Recent evidence suggests that radiation, particularly heavy-ion or alpha particles can disrupt parasite integrity and reduce viability offering a novel approach to parasite control (8-13). Furthermore, these treatments might also modify the host immune response which is pivotal in controlling parasitic infections (14). CD2, a surface glycoprotein on T

cells and natural killer (NK) cells, plays a critical role in T cell activation, adhesion to antigen-presenting cells and modulation of cytokine release (15,16).

Although previous studies have explored the direct cytotoxic effects of radiation on parasites, the potential immunomodulatory role of irradiated protoscoleces has not been comprehensively investigated. Understanding how radiation affects parasite survival and host immune dynamics, especially through markers like CD2 can inform the development of immunoprophylactic or adjunctive therapeutic strategies against hydatidosis (17).

This study investigates the combined histopathological and immunohistochemical effects of pre-irradiated hydatid protoscoleces on rat tissues and hypothesize that radiation will preserve tissue integrity and enhance T cell-mediated immunity as reflected by CD2 expression levels.

Materials and methods

Animals and ethics

Thirty-six male albino rats aged 6–8 weeks and weighing 200–250 grams, were maintained under standard laboratory conditions (22±2°C, 12-hour light/dark cycle) with ad libitum access to food and water. Ethical approval was obtained from the Institutional Animal Care and Use Committee, University of Mosul (Approval No. UM.VET.2024.099, dated January 2, 2024).

Protoscoleces collection and irradiation

Hydatid cysts were collected from sheep livers slaughtered in Mosul, Iraq. Protoscoleces were isolated, washed, and viability assessed using 0.1% eosin staining. Only samples with ≥99% viability were used (18,19). Protoscoleces were irradiated using nuclear particles americium source (²⁴¹Am) in the activity (1μCi) at energy (3.3MeV) and the distance between the source and the sample was maintained at 2.717cm (20,21) for either 1 hour or 1.5 hours in the Nuclear Laboratory, College of Education for Pure Science, University of Mosul.

Experimental design

36 male rats were divided into six groups (n=6 per group), all rats were intraperitoneally injected with 2000 viable protoscoleces: Groups 1 & 2: Protoscoleces irradiated for 1 hour, euthanized for 4 and 5 months. Groups 3 & 4: Protoscoleces irradiated for 1.5 hours, euthanized for 4 and 5 months. Groups 5 & 6: non-irradiated controls, euthanized for 4 and 5 months.

Histopathology and immunohistochemistry

Liver, spleen and intestine tissues were fixed in 10% neutral buffered formalin, embedded in paraffin and sectioned (5 μm). Hematoxylin-eosin staining was performed for histological examination. CD2

immunohistochemistry involved antigen retrieval (citrate buffer, pH 6.0), incubation with anti-CD2 monoclonal antibody and visualization using DAB chromogen (22,23).

CD2 expression was semi-quantitatively scored: 1+ (weak), 2+ (moderate), 3+ (intense) (24).

Quantitative immunohistochemical analysis

Immunohistochemical slides were digitally evaluated to determine the expression intensity of CD2 in spleen, intestine and liver tissues. Images from each experimental group were captured at 100× magnification using a calibrated light microscope (Leica Microsystems, Germany). Quantitative analysis was performed using Image J software (National Institutes of Health, Bethesda, MD, USA), which enabled the precise measurement of positively stained areas and optical density values. The expression levels were quantified as mean intensity scores (Mean ± SD), which were subsequently summarized and tabulated (see Table 2). Each measurement was independently verified by two blinded pathologists to ensure consistency and to minimize observer bias. This digital approach provided an objective and reproducible assessment of immunohistochemical staining, supporting the statistical rigor of the comparative analysis.

Statistical analysis

Data were analyzed using GraphPad Prism version 10.0 (GraphPad Software, San Diego, CA). Numerical data were presented as mean ± standard deviation (SD). Differences among groups were evaluated using one-way analysis of variance (ANOVA) followed by Tukey's post hoc test for multiple comparisons. A p-value <0.05 was considered statistically significant. CD2 expression scores were converted into semi-quantitative values to allow for statistical comparison.

Results

Statistical findings

Quantitative analysis revealed statistically significant differences among the experimental groups (P<0.05) regarding tissue architecture preservation and CD2 immunohistochemical expression, particularly in rats injected with protoscoleces irradiated for 1.5 hours these groups exhibited markedly superior histological outcomes in the spleen, intestine and liver samples, alongside higher CD2 expression scores, compared to both the 1-hour irradiated and non-irradiated control groups. These differences were corroborated by one-way ANOVA followed by Tukey's post hoc test, underscoring the robust protective and immunomodulatory effects of prolonged irradiation.

Histopathological findings

Spleen

At four months, rats injected with non-irradiated protoscoleces (positive control) exhibited severe necrosis in both the white and red pulp areas, along with marked hemorrhagic splenitis and extensive macrophage infiltration (Figure 1-A). By five months (positive control), these lesions progressed manifesting as diffuse fibrinoid necrosis and prominent atrophy of lymphoid follicles and increased macrophages (Figure1-D). In contrast, rats that received protoscoleces irradiated for 1 hour (Figure1-B) demonstrated moderate architectural preservation at four months with

partial cellular depletion in the white pulp and fewer necrotic foci in the red pulp. However, by five months (Figure 1-E), mild atrophy and moderate necrosis re-emerged. Remarkably, rats treated with protoscoleces irradiated for 1.5 hours showed intact spleen structures at four months (Figure1-C), minimal necrosis and well-preserved lymphoid follicles with only mild atrophy noted at five months (Figure1-F) suggesting a strong protective effect mediated by prolonged irradiation. These findings indicate that 1.5-hour irradiation effectively mitigates parasite-induced splenic damage, via enhanced immune clearance and reduced local inflammation.

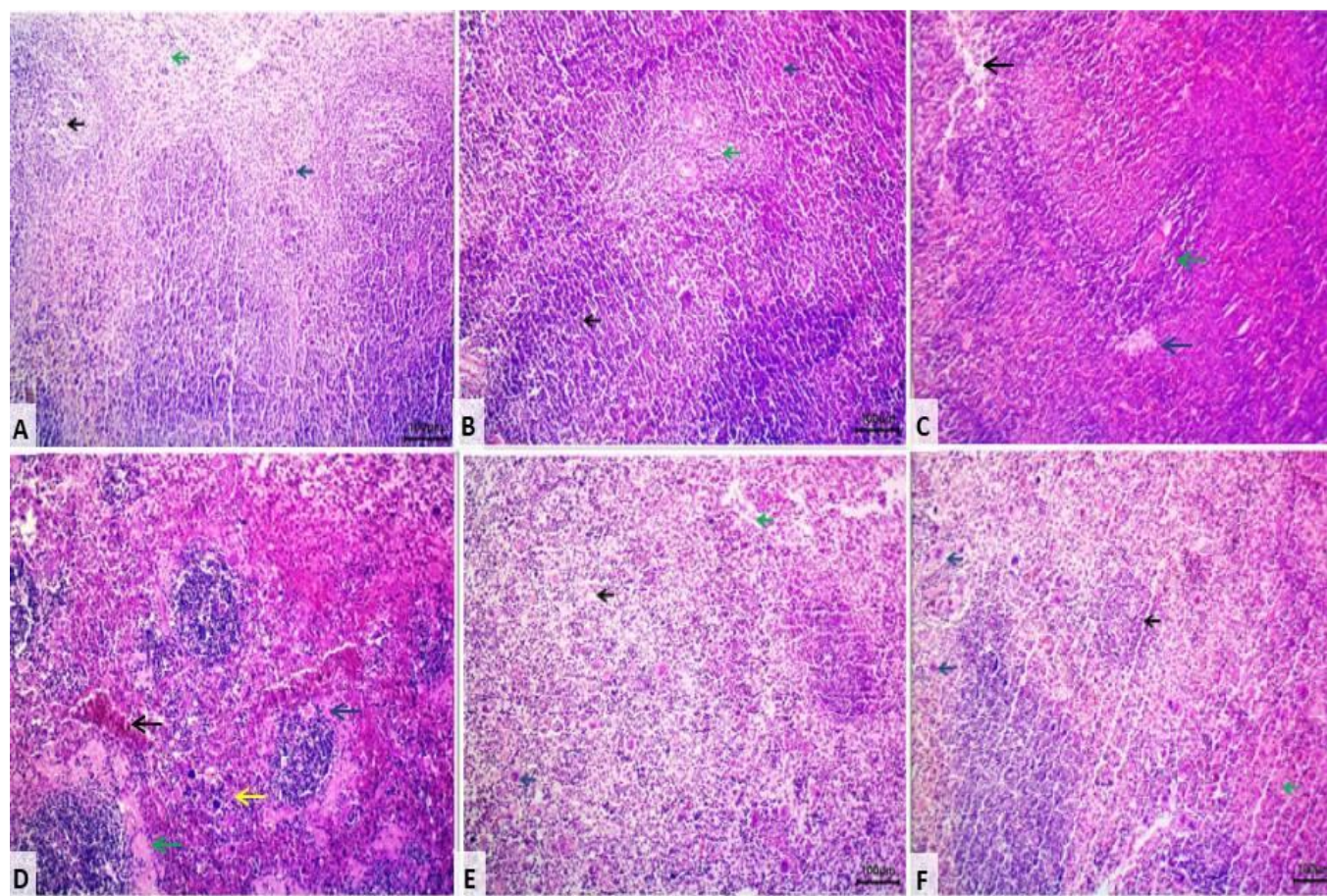


Figure 1: Histological sections of the rat spleen showing tissue architecture across experimental groups [A, B & C]: 4 months and [D, E & F]: 5 months hydatid cyst infected. [A&D]: positive control group without radiation treatment group, [A]: severe necrosis of cells in white pulp (black arrow), and red pulp (green arrow), with increase macrophages in the red pulp (blue arrow). [D]: Severe hemorrhagic splenitis (black arrow), diffuse fibrinoid necrosis of ellipsoidal sheath (green arrow), atrophy and necrosis of cell in white pulp (blue arrow) and increased macrophages (yellow arrow). [B] 1 hour, [E] 1 hour radiation treatment group: intact architecture of the white pulp (black arrow), with low numbers of cells in other white pulp (green arrow), and intact architecture of red pulp (blue arrow). [C] 1.5 hour radiation treatment group: loss and necrosis of cells in white pulp (black arrow), and red pulp (green arrow), with increase macrophages as giant cells (blue arrow). [F] 1.5 hour radiation treatment group: mild atrophy and low numbers of the cells in white pulp (black arrow), intact architecture of the red pulp (green arrow), with increase macrophages as giant cells (blue arrow). H&E stain, 100X; scale-bar=100µm.

Intestine

In the non-irradiated controls (positive control), severe villous epithelial necrosis, crypt degeneration, and marked thinning of the muscular layer were observed after four months (Figure 2-A) and five months (Figure 2-D), particularly prominent at five months. Conversely, the 1-hour irradiation groups displayed mild to moderate epithelial necrosis at four months (Figure 2-B) with preserved muscularis and limited inflammatory infiltrates by five months (Figure 2-E), the overall intestinal architecture appeared intact with only focal immune cell infiltration. Most notably, the 1.5-hour irradiation groups showed intact villi, preserved glandular structures and minimal inflammatory infiltration at both four months (Figure 2-C) and five months (Figure 2-F) with a consistently well-

defined muscular layer. Such histological preservation strongly supports the hypothesis that extended irradiation reduces parasite viability, thereby preventing extensive mucosal damage and secondary inflammation.

Liver

Figure 3 shows liver of rats infected with hydatid cyst (positive control) group without radiation, [A]: after 4 months, [B]: after 5 months shows presents of laminated membrane of protoscoleces (double head arrow), surrounding by increased fibrous tissue (black arrow), infiltration of inflammatory cells in portal area (green arrow) and necrosis of hepatocytes (blue arrow) with increased fibrous tissue between it (yellow arrow).

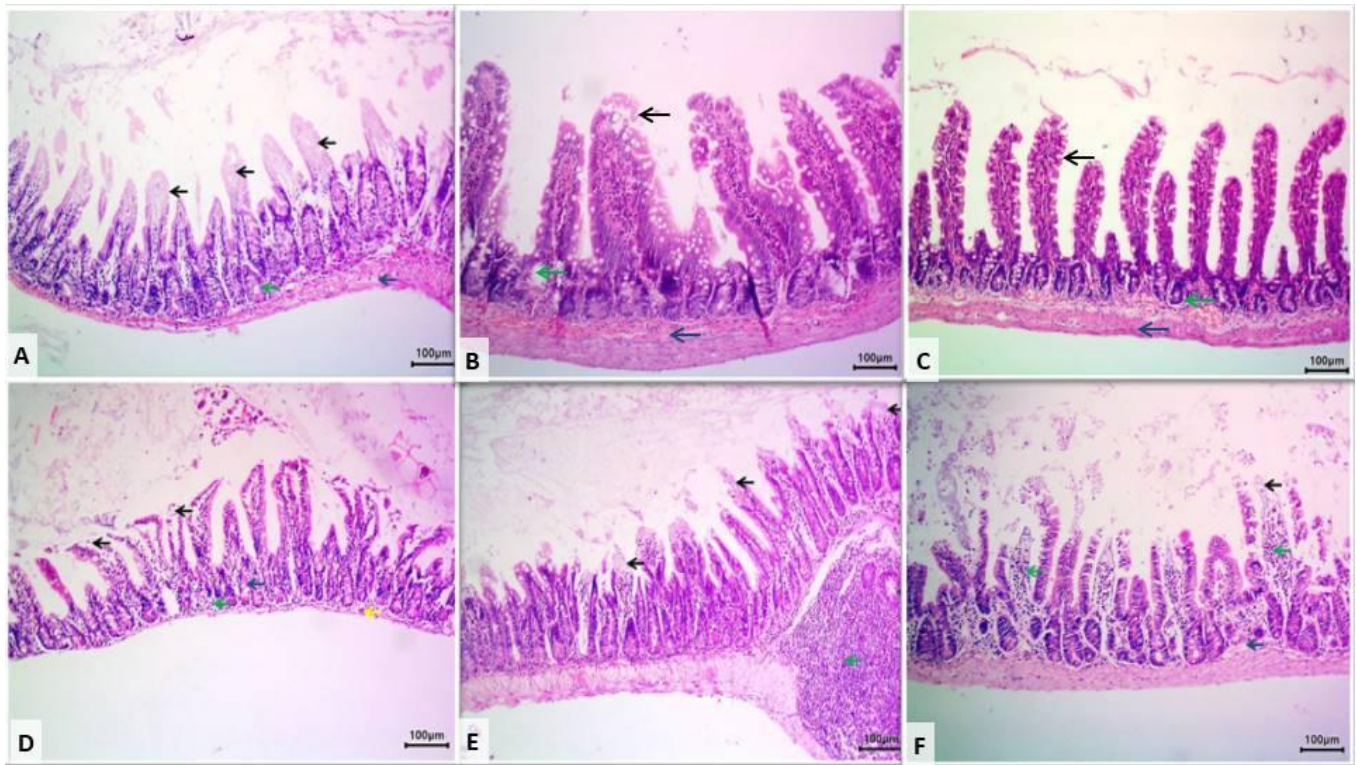


Figure 2: Histological sections of the rat intestine depicting epithelial and glandular integrity among groups [A, B & C]: 4 months and [D, E & F]: 5 months hydatid cyst infected. [A&D]: positive control group without radiation treatment group, [A]: necrosis of the epithelial cells in the top of the villi (black arrow), mild necrosis in the glands (green arrow) and mild necrosis in muscular layer (blue arrow). [D]: Severe necrosis of the epithelial cells in the top of the villi (black arrow), mild increased immune cells (green arrow), mild necrosis of few mucosal glands (blue arrow) and thinning of the muscular layer (yellow arrow) [B] 1 hour radiation treatment group: very mild necrosis of the epithelial cells in the top of the villi (black arrow) and intestinal glands (green arrow), with intact muscular layer (blue arrow). [E] 1 hour radiation treatment group: intact architecture of the villi (black arrow), intestinal glands (green arrow), and muscular layer (blue arrow). [C] 1.5 hour radiation treatment group: mild necrosis of the epithelial cells in the top of the villi (black arrow), focal increased immune cells in the submucosal layer (green arrow), and mild necrosis of few mucosal glands (blue arrow) [F] 1.5 hour radiation treatment group: mild necrosis of the epithelial cells in the top of the villi (black arrow), increased immune cells in the villi (green arrow), and mild necrosis of few mucosal glands (blue arrow). H&E stain, 100X; scale-bar=100µm.

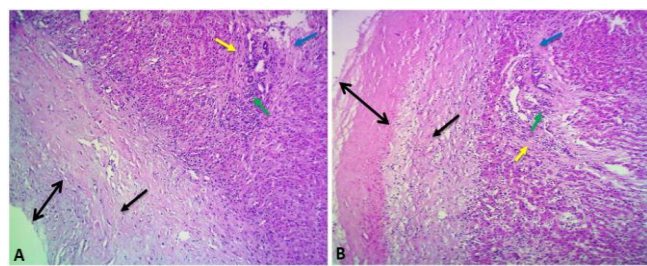


Figure 3: photomicrograph of rat's liver of hydatid cyst infected (positive control) group, [A]: after 4 months and [B]: after 5 months shows presents of laminated membrane of hydatid cyst (double head arrow), surrounding by increased fibrous tissue (black arrow), infiltration of inflammatory cells in portal area (green arrow) and necrosis of hepatocytes (blue arrow) with increased fibrous tissue between it (yellow arrow). 100X.

In figure (4-A), mild necrosis and vacuolar degeneration of the hepatocytes, mild infiltration of inflammatory cells and stenosis of sinusoids were recorded after 4 months of infected rats with protoscoleces exposure to nuclear particles for 1hour. On the other hand, dilatation of sinusoids and mild necrosis of the hepatocytes were observed after 4 months of infected rats with protoscoleces exposure to nuclear particles for 1.5hours (figure 4-B).

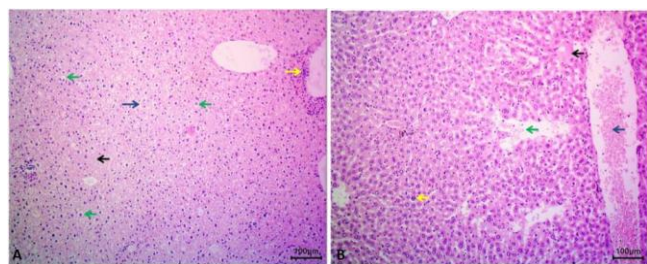


Figure 4: photomicrograph of rat's liver. [A]: After 4 months of infected rats with protoscoleces exposure to nuclear particles for 1hour: mild necrosis (black arrow) and vacuolar degeneration of the hepatocytes (green arrow), mild infiltration of inflammatory cells (yellow arrow) and stenosis of sinusoids (blue arrow). [B]: after 4 months of infected rats with protoscoleces exposure to nuclear particles for 1.5hours: dilatation of sinusoids (green arrow) and mild necrosis of the hepatocytes (black arrow).

Figure 5 after 5 months of infected rats with protoscoleces exposure to nuclear particles for 1hour, congestion of central vein and very mild vacuolar degeneration of hepatocytes were observed [A]. On the other hand, normal architecture of hepatic tissue, central vein and portal area were seen after 5 months of infected rats with protoscoleces exposure to nuclear particles for 1.5hours [B].

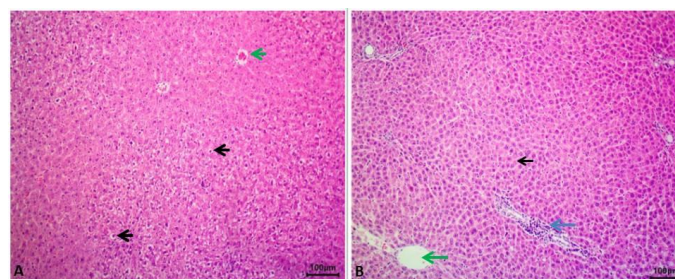


Figure 5: photomicrograph of rat's liver. [A]: after 5 months of infected rats with protoscoleces exposure to nuclear particles for 60minute: very mild vacuolar degeneration of hepatocytes (black arrow), and congestion of central vein (green arrow). [B]: after 5 months of infected rats with protoscoleces exposure to nuclear particles for 90minutes: intact architecture of hepatocytes (black arrow), central vein (green arrow) and portal area (blue arrow). 100X.

Immunohistochemical Analysis (CD2 Expression) Spleen

During the four months, the non-irradiated group showed moderate CD2 expression (score 2+), whereas the 1-hour group exhibited strong expression (score 3+) and the 1.5-hour group presented with strong expression (score 3+) indicating robust T-cell activation. At five months, CD2 expression in the non-irradiated group declined to weak (score 1+), suggesting progressive immunosuppression. Meanwhile, the 1-hour and 1.5-hour groups maintained moderate to strong expression (scores 2+ to 3+), reflecting sustained local immune activity (Figures 6 and 7; Table 1). This temporal pattern reinforces the immunomodulatory role of prolonged irradiation, potentially mitigating T-cell exhaustion commonly observed in chronic parasitic infections.

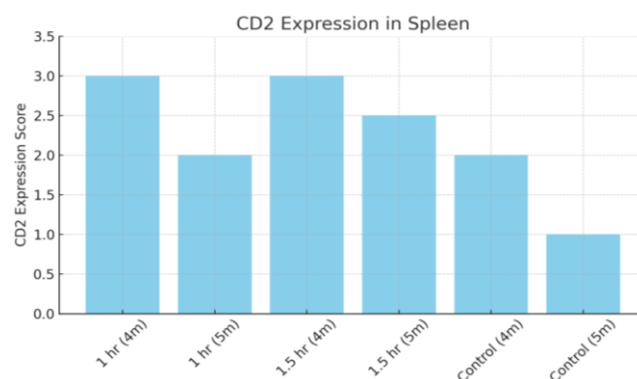


Figure 6: Quantitative distribution of CD2 expression scores in spleen samples among experimental groups (box plot).

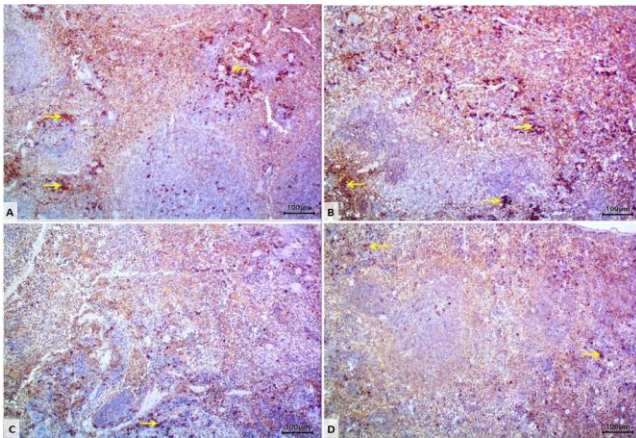


Figure 7: Immunohistochemical expression of CD2 in the Rat's spleen. [A& B]: 4 months and [C& D]: 5 months. [A] 1 hour radiation treatment group: showing intense expression (score 3+) (arrows) as brown stain. [B]: 1.5-hour radiation treatment group showing very intense expression (score 3+) (arrows). [C]: 1 hour radiation treatment group showing moderate expression (score 2+) (arrows). [D]: 1.5-hour radiation treatment group showing moderate expression (score 2+) (arrows). (brown DAB staining; 100×; scale bar = 100 μm).

Intestine

At four months, moderate CD2 expression was noted in the non-irradiated group (score 2+), strong expression in the 1-hour group (score 3+), and strong expression in the 1.5-hour group (score 3+). By five months, CD2 expression in the non-irradiated group declined to weak (score 1+), while the 1-hour group maintained strong expression (score 3+), and the 1.5-hour group decreased slightly to moderate expression (score 2+) (Figures 8 and 9; Table 1). These data highlight that 1.5-hour irradiation supports robust early immune activation but maintains a more regulated response later, potentially reducing chronic inflammation and tissue damage.

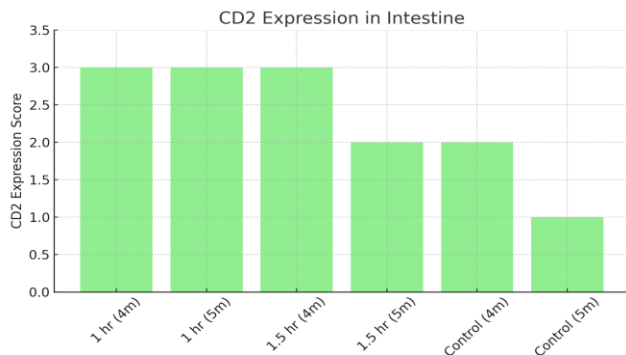


Figure 8: Quantitative distribution of CD2 expression scores in intestinal samples among experimental groups (box plot).

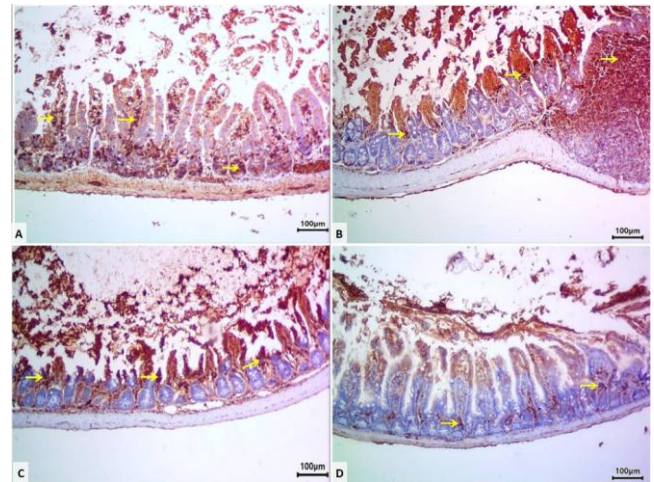


Figure 9: Immunohistochemical expression of CD2 in the Rat's intestine. [A& B]: 4 months and [C& D]: 5 months hydatid cyst infected. [A] 1 hour radiation treatment group: showing intense expression (score 3+) (arrows) as brown stain. [B]: 1.5-hour radiation treatment group showing very intense expression (score 3+) (arrows). [C]: 1 hour radiation treatment group showing intense expression (score 3+) (arrows). [D]: 1.5-hour radiation treatment group showing moderate expression (score 2+) (arrows) (brown DAB staining; 100×; scale bar = 100 μm).

Liver

At four months, the non-irradiated group showed weak to moderate CD2 expression (scores 1+ to 2+), while the 1-hour group showed moderate expression (score 2+), and the 1.5-hour group showed strong expression (score 3+). At five months, CD2 expression in the non-irradiated group dropped to weak (score 1+), while both irradiated groups retained moderate expression (score 2+), indicating sustained hepatic immune responsiveness even in late infection stages (Figures 10 and 11; Table 1). This finding suggests a possible long-term protective hepatic effect through T-cell mediated immunity, potentially important for preventing parasite dissemination.

Interpretation

The data collectively indicate that pre-irradiation of protoscoleces for 1.5 hours provided the highest level of tissue protection and the most robust CD2-mediated immune activation at four months. Although CD2 expression decreased slightly in five months, it remained higher in irradiated groups than controls, suggesting a sustained but moderated immune response.

Histopathological analyses consistently demonstrated that irradiation reduced tissue damage and inflammatory infiltration, supporting the hypothesis that nuclear particles can function as effective immunomodulatory agents.

Table 1: Summary of histopathological findings and semi-quantitative CD2 expression scores among experimental groups

Group	Treatment	Time (months)	Spleen Histology	Intestine Histology	Liver Histology	CD2 Expression (Spleen)	CD2 Expression (Intestine)	CD2 Expression (Liver)
1	1-hr irradiated protoscoleces	4	Moderate preservation	Mild necrosis	Mild changes	3+	3+	2+
2	1-hr irradiated protoscoleces	5	Mild atrophy	Preserved	Mild changes	2+	3+	2+
3	1.5-hr irradiated protoscoleces	4	Well preserved	Intact	Minimal changes	3+	3+	3+
4	1.5-hr irradiated protoscoleces	5	Mild atrophy	Mild necrosis	Mild changes	2+/3+	2+	2+
5	Non-irradiated controls	4	Severe necrosis	Severe necrosis	Moderate changes	2+	2+	1+/2+
6	Non-irradiated controls	5	Severe necrosis and atrophy	Severe necrosis	Severe changes	1+	1+	1+

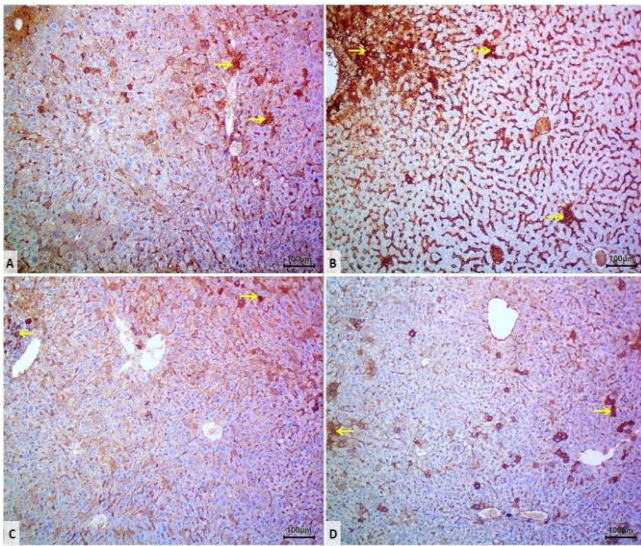


Figure 10: Immunohistochemical expression of CD2 in the rat's liver. [A& B]: 4 months and [C& D]: 5 months. [A] 1 hour radiation treatment group: showing moderate expression (score 2+) (arrows) as brown stain. [B]: 1.5 hour radiation treatment group showing intense expression (score 3+) (arrows). [C]: 1 hour radiation treatment group showing weak expression (score 1+) (arrows). [D]: 1.5 hour radiation treatment group showing moderate expression (score 2+) (arrows) (brown DAB staining; 100×; scale bar = 100 μm).

As detailed in Table 2, it indicates that the 1.5-hour irradiation groups exhibited the highest mean architecture preservation scores in both spleen and intestine (3.5 ± 0.2

and 3.6 ± 0.2 , respectively), indicating intact tissue integrity. Similarly, CD2 expression scores were significantly elevated in these groups (3.5 ± 0.2), suggesting robust and sustained T cell activation. In contrast, non-irradiated controls showed markedly lower scores across all parameters, confirming extensive tissue damage and diminished immune activity. These statistical findings collectively validate the histopathological observations and provide quantitative evidence of the protective and immunomodulatory advantages conferred by prolonged alpha particle irradiation.

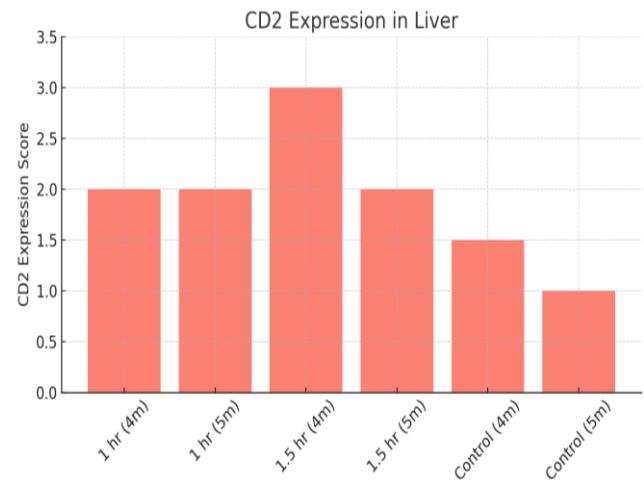


Figure 11: Immunohistochemical expression of CD2 in the rat liver across treatment groups (brown DAB staining; 100×; scale bar = 100 μm).

Table 2: Comparative quantitative analysis of tissue architecture preservation and CD2 expression scores among experimental groups (Mean $\pm$ SD)

Group	Treatment	Spleen Architecture Score (Mean $\pm$ SD)	Intestine Architecture Score (Mean $\pm$ SD)	Liver Architecture Score (Mean $\pm$ SD)	CD2 Expression Score (Mean $\pm$ SD)
1	1-hour irradiated, 4 months	2.5 $\pm$ 0.3	2.7 $\pm$ 0.2	2.4 $\pm$ 0.3	2.8 $\pm$ 0.2
2	1-hour irradiated, 5 months	2.0 $\pm$ 0.4	3.0 $\pm$ 0.1	2.5 $\pm$ 0.2	2.5 $\pm$ 0.3
3	1.5-hour irradiated, 4 months	3.5 $\pm$ 0.2	3.6 $\pm$ 0.2	3.3 $\pm$ 0.2	3.5 $\pm$ 0.2
4	1.5-hour irradiated, 5 months	2.8 $\pm$ 0.3	3.2 $\pm$ 0.3	3.0 $\pm$ 0.3	3.0 $\pm$ 0.3
5	Control, 4 months	1.5 $\pm$ 0.2	1.6 $\pm$ 0.2	2.0 $\pm$ 0.3	2.0 $\pm$ 0.3
6	Control, 5 months	1.2 $\pm$ 0.3	1.3 $\pm$ 0.2	1.5 $\pm$ 0.2	1.5 $\pm$ 0.2

Scores were semi-quantitatively graded from 1 (poor) to 4 (excellent) based on histopathological and immunohistochemical evaluations. Data are presented as mean $\pm$ SD; significant differences were determined using one-way ANOVA followed by Tukey's test ($P < 0.05$).

Mechanism of alpha particle-irradiated hydatid protoscoleces

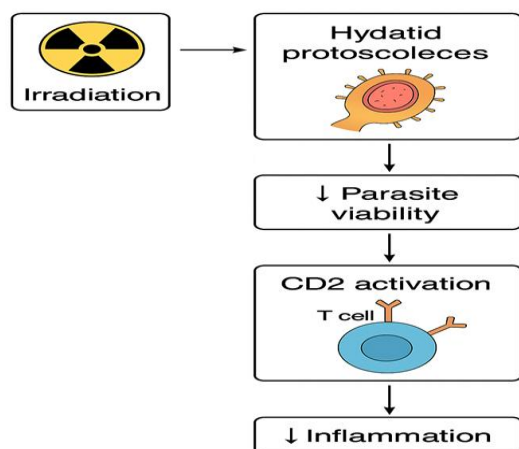


Figure 12: Schematic diagram summarizing the proposed immunomodulatory mechanism of alpha particle-irradiated hydatid protoscoleces: reduced parasite viability leads to CD2-mediated T cell activation and decreased tissue inflammation.

Discussion

The present study explored, with critical depth, the histopathological and immunohistochemical impacts of pre-irradiated hydatid protoscoleces using nuclear particles on experimentally infected rats. The results strongly corroborate the initial hypothesis that nuclear particles, alpha, irradiation effectively reduces protoscolex viability, preserves organ architecture, and modulates host immune responses through enhanced CD2 expression.

As clearly illustrated in Figure 1, the marked reduction in splenic histopathological damage in rats receiving protoscoleces irradiated for 1.5-hours can be attributed to diminished parasite viability, which consequently reduces local inflammatory-mediated tissue destruction. This finding aligns with previous studies showing that X-ray and gamma radiation significantly compromise the structural integrity of protoscoleces and decrease infectivity (8-10). Importantly, the near-intact architecture observed in spleen sections from the 1.5-hour group underscores a strong tissue-protective effect, potentially mediated by early and robust immune activation.

Similarly, as depicted in Figure 2, the intestinal sections revealed that prolonged irradiation markedly preserved villous integrity and minimized mucosal and glandular necrosis. Such outcomes further support the hypothesis that irradiation-mediated protoscolex inactivation prevents subsequent inflammatory cascades and secondary tissue degradation. The preservation of the muscular layer and reduced immune infiltration in the 1.5-hour group (Figure 2) highlight a potential mechanistic synergy between direct parasiticidal effects and modulated host immune responses.

Although liver tissues were not the primary investigative focus, the reduced periportal inflammation and better hepatocyte preservation seen in the 1.5-hour group (Figures 3-5) suggest systemic protective effects, potentially mediated by decreased parasite burden and modulated cytokine signaling. This systemic effect is of translational relevance, as it indicates a broader therapeutic potential beyond local tissue containment.

The immunohistochemical findings further deepen the mechanistic understanding of these protective effects. As shown in Figures (6-11) CD2 expression was markedly upregulated in irradiated groups, particularly after 1.5 hours of exposure, at four months post-infection. The strong CD2

expression in the spleen and intestine (Figures 6-9) suggests robust T-cell activation, which plays a crucial role in orchestrating effective anti-parasitic responses through enhanced cytotoxicity and cytokine production (15,16).

Moreover, the observed slight reduction in CD2 expression at five months, particularly in the 1.5-hour group, may reflect a transition toward immune homeostasis after an initial phase of hyperactivation. This dynamic modulation prevents excessive chronic inflammation and associated tissue damage, thereby preserving organ function. This nuanced temporal pattern parallels the concept of T-cell exhaustion described in chronic parasitic infections, where initial hyperactivation is followed by a regulatory decline to mitigate immunopathology (25).

In agreement with the summarized data presented in Table 1, the sustained moderate CD2 expression at later stages, especially in the liver (Figures 10 and 11), indicates that irradiation-induced immune priming continues to exert protective effects even in distant organs. This observation suggests potential systemic immunomodulatory benefits that could be strategically harnessed for future therapeutic applications.

When juxtaposed with prior research, the findings of this study not only confirm the parasitocidal efficacy of radiation modalities (26,27) but also introduce a novel dimension of immune activation, specifically via CD2-mediated T-cell responses. This dual effect positions alpha particle irradiation as a promising adjunctive strategy, potentially valuable in endemic regions where surgical and chemotherapeutic interventions remain limited.

Notably, our findings corroborate and extend the observations of Bao *et al.* (8,9) and Zhao *et al.* (10), who documented significant structural compromise of *Echinococcus* protoscoleces upon exposure to various forms of radiation. However, our study uniquely integrates a functional immunological dimension by quantifying CD2 expression and proposing mechanistic cytokine interactions which were not addressed in earlier studies. Moreover, the consistent upregulation of CD2 parallels results from Brudno *et al.* (16), emphasizing the centrality of T cell activation in parasite clearance. Conversely, the moderate decline in CD2 expression at later stages may reflect a homeostatic feedback mechanism akin to T cell exhaustion described by Taylor *et al.* (25), indicating a finely tuned balance between immune activation and regulation. This nuanced dynamic underscores the translational potential of irradiation as an immunomodulatory adjunct rather than a purely cytotoxic modality.

Future investigations should aim to incorporate comprehensive cytokine profiling (e.g., IL-1 β , IL-6, TNF- α , nitric oxide, PGE2) and advanced techniques such as flow cytometry and multiplex assays to elucidate the precise immune networks involved (11,28). Moreover, optimizing irradiation parameters to balance maximal parasitocidal

impact with minimal immunopathological risk remains a critical translational goal.

Future studies should incorporate detailed temporal cytokine profiling and flow cytometric analysis of splenic and hepatic lymphocyte subsets to delineate the exact cellular contributors to the observed immune modulation. Additionally, long-term follow-up experiments assessing recurrence rates and survival outcomes in larger animal cohorts would be invaluable. Finally, exploring combinatorial approaches integrating irradiation with low-dose chemotherapeutics or immunoadjuvants may offer synergistic benefits and pave the way toward translational application in endemic human populations.

Importantly, future studies should include cytokine profiling using ELISA or multiplex assays to provide a more comprehensive understanding of the systemic immune modulation induced by alpha particle-irradiated protoscoleces. Such data would complement the observed CD2 expression patterns and elucidate additional molecular pathways involved in parasite clearance and tissue preservation.

The comparative analysis of CD2 expression scores across the experimental groups, as shown in Table 2 and illustrated in the box plot, highlights a clear upregulation of CD2 in irradiated groups, particularly after 1.5-hour exposure at four months. This finding suggests that radiation treatment may modulate T-cell activity and enhance local immune responses within the spleen and intestinal tissues, potentially contributing to parasite clearance or immune reprogramming. These observations are in partial agreement with previous studies reporting radiation-induced immunomodulation in parasitic infections (29-32). As illustrated in Figure 12, the proposed mechanism integrates parasite inactivation with enhanced CD2-mediated immune responses. However, further mechanistic investigations are needed to elucidate the exact signaling pathways involved.

Conclusion

In conclusion, pre-irradiation of hydatid protoscoleces with nuclear particles effectively reduced parasite viability, preserved organ architecture, and enhanced CD2-mediated T cell responses in rats. These findings support the potential of nuclear particle irradiation as a dual parasitocidal and immunomodulatory strategy which could inform future therapies for hydatidosis. Further investigations are recommended to explore comprehensive cytokine profiles and long-term immune modulation effects.

Acknowledgments

The authors express their sincere gratitude to the College of Education for Pure Science, University of Mosul, for providing laboratory facilities and technical support throughout this study.

Conflict of Interest

The authors declare no conflict of interest.

References

- Hogea MO, Ciomaga BF, Muntean MM, Muntean AA, Popa MI, Popa GL. Cystic echinococcosis in the early 2020s: a review. *Trop Med Infect Dis.* 2024;9(2):36. DOI: [10.3390/tropicalmed9020036](https://doi.org/10.3390/tropicalmed9020036)
- Hajjafari A, Sadr S, Santucci C, Masala G, Bayat M, Lotfalizadeh N, Borji H, Partovi Moghaddam S, Hajjafari K. Advances in detecting cystic echinococcosis in intermediate hosts and new diagnostic tools: a literature review. *Vet Sci.* 2024;11(6):227. DOI: [10.3390/vetsci11060227](https://doi.org/10.3390/vetsci11060227)
- Pal M, Alemu HH, Marami LM, Garedo DR, Bodena EB. Cystic echinococcosis: a comprehensive review on life cycle, epidemiology, pathogenesis, clinical spectrum, diagnosis, public health and economic implications, treatment, and control. *Int J Clin Exp Med Res.* 2022;6(2):131-141. DOI: [10.26855/ijcemr.2022.04.005](https://doi.org/10.26855/ijcemr.2022.04.005)
- Zhai S, Yang Y, Zhou Y, Lai Q, Li K, Liu S, Li W, Gao F, Guan J. *Echinococcus granulosus*-induced liver damage through ferroptosis in rat model. *Cells.* 2025;14:328. DOI: [10.3390/cells14050328](https://doi.org/10.3390/cells14050328)
- Alpaca Rodriguez LR, Kirschbaum-Chrem JP, Romero G, Villanueva E, Ugas Charcape CF. The many faces of pediatric hydatid disease: a pictorial review. *Pediatr Radiol.* 2024. DOI: [10.1007/s00247-024-06080-7](https://doi.org/10.1007/s00247-024-06080-7)
- Aydin Y, Ulaş AB, Dostbil A. Current management of pulmonary hydatid cyst. *Eurasian J Med.* 2025;57(1):1-7. DOI: [10.5152/eurasianjmed.2025.24761](https://doi.org/10.5152/eurasianjmed.2025.24761)
- Yönder H, Elkan H. Management of a liver hydatid cyst patient who fistulized the diaphragm and subsequently presented with thorax rupture: a case report. *J Cardiothorac Surg.* 2024;19(1):596. DOI: [10.1186/s13019-024-02585-3](https://doi.org/10.1186/s13019-024-02585-3)
- Bao YX, Mao R, Qi HZ, Zhang YF, Ni YQ, Xie ZR, Aziguli T, Wen H. X-ray irradiation against *Echinococcus multilocularis* protoscoleces in vitro. *Chin J Parasitol Parasit Dis.* 2011;29:208-211. [\[available at\]](#)
- Bao YX, Zhang YF, Ni YQ, Xie ZR, Qi HZ, Mao R, Yang YG, Wen H. Effect of 6 MeV radiotherapy on secondary *Echinococcus multilocularis* infection in rats. *Chin J Parasitol Parasit Dis.* 2011;29:127-129. [\[available at\]](#)
- Zhao Y, Gui W, Zhang Y, Mo G, Li D, Chong S. Inhibitory effect of ionizing radiation on *Echinococcus granulosus* hydatid cyst. *Diseases.* 2019;7(1):23. DOI: [10.3390/diseases7010023](https://doi.org/10.3390/diseases7010023)
- Alfathi M, Alabdaly Y, Al-Hayyali F. Ameliorative effect of spirulina against gentamicin toxicity in liver and kidney tissues of male rat. *Egypt J Histol.* 2023;46(4):1666-1675. DOI: [10.21608/ejh.2022.155247.1750](https://doi.org/10.21608/ejh.2022.155247.1750)
- Khalaf AT, Abdalla AN, Ren K, Liu X. Cold atmospheric plasma (CAP): a revolutionary approach in dermatology and skincare. *Eur J Med Res.* 2024;29(1):487. DOI: [10.1186/s40001-024-02088-9](https://doi.org/10.1186/s40001-024-02088-9)
- Ruprecht NA, Singhal S, Schaefer K, Charkoftaki G, Cekanaviciute E, Kennedy AR, Goukassian DA, Taylor AM, Nikitaki Z, Georgakilas AG. A review: multi-omics approach to studying the association between ionizing radiation effects on biological aging. *Biology.* 2024;13(2):98. DOI: [10.3390/biology13020098](https://doi.org/10.3390/biology13020098)
- Wadhwa A, Moreno-Villanueva M, Crucian B, Wu H. Synergistic interplay between radiation and microgravity in spaceflight-related immunological health risks. *Immun Ageing.* 2024;21(1):5. DOI: [10.1186/s12979-024-00449-w](https://doi.org/10.1186/s12979-024-00449-w)
- Jo Y, Sim HI, Yun B, Park Y, Jin HS. Revisiting T-cell adhesion molecules as potential targets for cancer immunotherapy: CD226 and CD2. *Exp Mol Med.* 2024. DOI: [10.1038/s12276-024-01317-9](https://doi.org/10.1038/s12276-024-01317-9)
- Brudno JN, Maus MV, Hinrichs CS. CAR T cells and T-cell therapies for cancer: a translational science review. *JAMA.* 2024. DOI: [10.1001/jama.2024.19462](https://doi.org/10.1001/jama.2024.19462)
- Abaza S. Extracellular vesicles and host-parasite interactions. *Parasitol United J.* 2023;16(1):12-31. DOI: [10.21608/PUJ.2023.202887.1208](https://doi.org/10.21608/PUJ.2023.202887.1208)
- Smyth JD, Barrett NJ. Procedures for testing the viability of human hydatid cysts following surgical removal especially after chemotherapy. *Trans R Soc Trop Med Hyg.* 1980;74(5):649-652. DOI: [10.1016/0035-9203\(80\)90157-1](https://doi.org/10.1016/0035-9203(80)90157-1)
- Smyth J. In vitro culture of *Echinococcus* spp. *Proceedings of the 13th Int. Congr. Hydatidology.* Madrid, 1985; 85: 84-89. Doi:10.1016/0020-7519.
- Cember H. Introduction to health physics. 3rd ed. Oxford: Pergamon Press; 1996. 129-131 p.
- Kassim YY. Validity of the diameter-length method for determining the bulk etch rate of a CR-39 detector. *Nucl Instrum Methods Phys Res B.* 2021;487:8-11. DOI: [10.1016/j.nimb.2020.11.008](https://doi.org/10.1016/j.nimb.2020.11.008)
- Shahab B, Shnawa BH. Immunohistochemical and histopathological characterization of immune changes in the host-tissue reaction site of murine cystic echinococcosis. *Adv Anim Vet Sci.* 2022;10(11):2367-2375. DOI: [10.17582/journal.aavs/2022/10.11.2367.2375](https://doi.org/10.17582/journal.aavs/2022/10.11.2367.2375)
- Alturkistani HA, Tashkandi FM, Mohammedsalem ZM. Histological stains: a literature review and case study. *Glob J Health Sci.* 2016;8(3):72. DOI: [10.5539/gjhs.v8n3p72](https://doi.org/10.5539/gjhs.v8n3p72)
- Rüfer A, Nilius H, Hermine O, Niedoszytko M, Oude Elberink JN, Bonadonna P, Shoumariyeh K, Gulen T, Hartmann K, Sabato V, Angelova-Fischer I. Prognostic impact of expression of CD2, CD25, and/or CD30 in/on mast cells in systemic mastocytosis: a registry study of the European Competence Network on Mastocytosis. *Leukemia.* 2025;39(3):675-683. DOI: [10.1038/s41375-024-02504-3](https://doi.org/10.1038/s41375-024-02504-3)
- Taylor CH, Friberg IM, Jackson JA, Arriero E, Begon M, Wanelik KM, Paterson S, Bradley JE. Living with chronic infection: persistent immunomodulation during avirulent haemoparasitic infection in a wild rodent. *Mol Ecol.* 2023;32(5):1197-1210. DOI: [10.1111/mec.16819](https://doi.org/10.1111/mec.16819)
- Pohle S, Ernst R, MacKenzie C, Spicher M, Romig T, Hemphill A, Gripp S. *Echinococcus multilocularis*: the impact of ionizing radiation on metacestodes. *Exp Parasitol.* 2011;127(1):127-134. DOI: [10.1016/j.exppara.2010.06.007](https://doi.org/10.1016/j.exppara.2010.06.007)
- Ulger S, Barut H, Tunc M, Aydin E, Aydinkarahaliloglu E, Gokcek A. Radiation therapy for resistant sternal hydatid disease. *Strahlenther Onkol.* 2013;189(6):508-509. DOI: [10.1007/s00066-013-0322-5](https://doi.org/10.1007/s00066-013-0322-5)
- Carroll SL, Pasare C, Barton GM. Control of adaptive immunity by pattern recognition receptors. *Immunity.* 2024;57(4):632-648. DOI: [10.1016/j.immuni.2024.03.014](https://doi.org/10.1016/j.immuni.2024.03.014)
- Constanzo J, Faget J, Ursino C, Badie C, Pouget JP. Radiation-induced immunity and toxicities: the versatility of the cGAS-STING pathway. *Front Immunol.* 2021;12:680503. DOI: [10.3389/fimmu.2021.680503](https://doi.org/10.3389/fimmu.2021.680503)
- Berzaghi R, Tornaas S, Lode K, Hellevik T, Martinez-Zubiaurre I. Ionizing radiation curtails immunosuppressive effects from cancer-associated fibroblasts on dendritic cells. *Front Immunol.* 2021;12:662594. DOI: [10.3389/fimmu.2021.662594](https://doi.org/10.3389/fimmu.2021.662594)
- Kaur AP, Alice A, Crittenden MR, Gough MJ. The role of dendritic cells in radiation-induced immune responses. *Int Rev Cell Mol Biol.* 2023;378:61-104. DOI: [10.1016/bs.ircmb.2022.12.001](https://doi.org/10.1016/bs.ircmb.2022.12.001)
- Mao RB, Zhang WB, Qi HZ, Jiang T, Wu G, Lu PF, Shang G, Xu L, Hao J, Shou X, Li H, Li J, Zhang S, Bao Y, Wen H. Efficacy of radiotherapy for the treatment of cystic echinococcosis in naturally infected sheep. *Infect Dis Poverty.* 2017;6:88. DOI: [10.1186/s40249-017-0301-7](https://doi.org/10.1186/s40249-017-0301-7)

التغيرات الكيميائية النسجية المناعية في تعبير CD2 لأعضاء مختلفة من الجرذان المعاملة بالجسيمات النووية ضد داء الاكياس العدوية

اميمة عادل نجم^١، سهيلة يعقوب يوسف^١ و ياسر يحيى قاسم^٢

^١ قسم علوم الحياة، ^٢ قسم الفيزياء، كلية التربية للعلوم الصرفة، جامعة الموصل، الموصل، العراق

الخلاصة

يعد داء الاكياس العدوية من الامراض الحيوانية المشتركة المتسببة عن الطور البرقي للمشوكة الحبيبية والذي يمثل مشكلة صحية خطيرة تعود الى موقع الكيس ومقاومته للأدوية. ثمة اساليب جديدة كالجسيمات النووية، جسيمات ألفا، قد تقلل من حيوية الطفيل وتعزز من مناعة المضيف. قُيِّمت هذه الدراسة تأثير الجسيمات النووية على الرؤيسات الاولى المعزولة من الاكياس العدوية وعلى التغيرات النسجية المرضية وعلى تعبير CD2 في كبد، طحال وأمعاء الجرذان المصابة لغرض

استخدامها كعلاج ضد داء الاكياس العدوية. تم عزل الرؤيسات الاولى من اكباد الاغنام وتعريضها للجسيمات النووية عند طاقة (٣,٣ ميكالكترون فولت) لمدة ساعة او ساعة ونصف. استخدم ٣٦ ذكراً من الجرذان وتم تقسيمها الى ٦ مجاميع حقنت داخل البريتون بالرؤيسات الاولى (المجموعتان الاولى والثانية؛ الرؤيسات الاولى مشععة لمدة ساعة، المجموعتان الثالثة والرابعة؛ الرؤيسات الاولى مشععة لمدة ساعة ونصف؛ المجموعتان الخامسة والسادسة؛ الرؤيسات الاولى غير مشععة). شرحت المجاميع بعد اربعة او خمسة أشهر من اصابتها. اجريت فحوصات نسجية مرضية وكيمياء نسجية مناعية لتعبير CD2 لغرض تقييم سلامة الانسجة والاستجابات المناعية، اذ لوحظ حدوث تغيرات مرضية نسجية قليلة وتعبير لـ CD2 قوي (+٣) في طحال وامعاء وكبد الجرذان بعد خمسة أشهر من حقنها بالرؤيسات الاولى المشععة بالجسيمات النووية لمدة ساعة ونصف مما يؤكد تنشيط الخلايا القاتلة الطبيعية (NK) مقارنة ببقية المجاميع الاخرى التي ظهر فيها تلف شديد في الانسجة. يمكن الاستنتاج من خلال هذه الدراسة بأن الرؤيسات الاولى المشععة تعمل كمضادات للطفيليات ومعدلات مناعية والتي يمكن تطويرها الى علاج ضد داء الاكياس العدوية. توصي الدراسة الحالية بإجراء المزيد من الدراسات اللاحقة لقياس السايوتوكينات ومؤشرات مناعية اخرى لتوضيح ميكانيكية السيطرة على داء الاكياس العدوية.