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Investigating the role of alpha particles in gene expression levels of cytokines in rats infected with cystic echinococcosis

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Abstract

Hydatidosis is a widespread parasitic disease caused by larval stage protoscoleces of Echinococcus granulosus. The present study investigated the effect of protoscoleces after exposure to alpha particles on the expression of immune response-related genes in rats. This study observed gene expression of TNF-α, IL-6, and IL-10 in rats infected with hydatid cysts. In this study, six groups of male albino rats were used; some groups were injected intraperitoneally, with protoscoleces exposed to alpha particles for either 1 or 1.5 hours, while the control group received protoscoleces. After four to five, the rats were euthanized. The levels of gene activity for markers like TNF-α, IL-6, and IL-10 were examined in different organs of rats at time intervals. The results showed that alpha particle exposure led to significant changes in the expression of these cytokines depending on both dose and duration of exposure. The expression of alpha particles after 1 and 1.5 hours resulted in notable changes in IL-6, IL-10, and TNF-α gene expression in rats, especially after four months, where the injection of irradiated protoscoleces led to an increase in TNF- α expression. The exposure to alpha particles influenced the inflammatory response at both the transcriptional and functional levels. Furthermore, prolonged exposure to alpha particles demonstrated a dose-dependent effect, which enhances gene expression changes.

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Introduction

Hydatid echinococcosis is a widespread parasitic disease caused by metacestode (1-3) characterized by the development of primary cysts in the liver, lung, and other organs (4,5). Hydatidosis is a result of incidental ingestion of *E. granulosus* eggs, often through contact with hosts such as lamb, camel, horses, goats, and pigs (6-8). The hydatid cyst of *E. granulosus* develops as fluid-filled structures in tissue organs and is composed of two main layers: an Internal nucleated germinal stratum and an external acellular plated layer. These layers are enclosed with a fiber capsule produced by the host (9). The location and the size of the cysts within the affected organ ruptures, which causes complications and triggers immune responses; all these are affected by the infectious change, which is very much changing (10-12). Mahmoud *et al.* (13) showed that the

immune response of the intermediate host is critical in determining the interaction between the host and parasite, and the larval stage secretion compound has an influence on intermediate host specialized cells in order to avert the immune responses of the hosts. Parasites have improved the diversity of mechanisms, which boost their existence in the organs and help prevent chronic infection. Environmental factors, including radiation, play an important role in how the immune system works since exposure to radiation can influence gene expression levels and significantly impact the functions of cells (14-16). When researching how radiation affects genes that are relevant to the system's functions, it becomes apparent that inflammatory cytokines like TNF-α and interleukins (IL-6 and IL-10) are key players in regulating immune responses and inflammation (17). TNF-α plays a role in activating cells and starting inflammatory signaling pathways, as well as promoting tissue repair through programmed cell death being involved in cell death mechanisms (18). On the other hand, Supino *et al.* (19) observed that IL-10 plays a role in acute immune response and is recognized for its inflammatory characteristics, which help to control exaggerated immune excessive and maintain immune balance (20,21). Previous Studies have found that exposure to radiation results in changes in the levels of cytokines like TNF-α, IL-10, and IL-6 over time, depending on both the dose and duration of exposure. Lower doses can boost the system, and higher doses may suppress it due to stress or damage to cells (22,23). Moreover, the impact of radiation varies depending on how much time passes after exposure, underscoring the need to examine how genes respond at different time points and doses of radiation (24-27).

The present study aims to investigate how radiation influences the alterations in gene expression of TNF- α , IL-6, and IL- 10 after exposing the body to irradiated protoscoleces, emphasizing the effect of radiation dosage and the time after exposure on immune system reactions. The findings from this research could help elucidate the molecular pathway involved in radiation-induced immune modulation and could support the development of strategies to mitigate radiation-related immune damage.

Materials and methods

Ethical approval

Ethical approvals reference number UM.VET.2024.125, dated January 2, 2024, for handling animals, was awarded by the Institutional Animal Care and Use Committee in the University of Mosul, College of Veterinary Medicine.

Animal subjects

Rats 6 to 8 weeks old and weighed between 200 and 250 grams were used for this study. These rats were kept in a controlled laboratory setting with a temperature of around 22°C and a normal dark cycle of 12 hours. They had access to food and water throughout the study to maintain their well-being and ensure they were comfortable during the research period. All animal-related procedures followed guidelines.

Collection and preparation of hydatid cysts

Hydatid cysts were collected from the livers of sheep acquired from slaughterhouses in Mosul city, Iraq. The fertility of these cysts was confirmed by observing the presence of viable protoscoleces upon their opening. In order to determine the viability of protoscolices. A solution of 0.01 % Eosin was used. Samples showing a viability rate of 99% were chosen for research and experimentation. The cyst's outer layer was sterilized using a 1 percent iodine alcohol solution administered using cotton, and the hydatid fluid was extracted using a 20 mL syringe equipped with an 18-gauge needle. The content of the cyst was aspirated using a plastic

pipette before thoroughly rinsing the interior with phosphate-buffered saline (PBS) at a pH of 7.2. The liquid containing protoscoleces was transferred to sterile test tubes and subjected to three cycles of centrifugation at 3000 rpm for 5 minutes. To minimize contamination, penicillin 2000 IU/L and streptomycin 1 g/L were added to the PBS during the second and third washes. After each centrifugation step, the supernatant was discarded, and the remaining pellet containing protoscoleces was resuspended in fresh, sterile PBS (28).

Viability assessment

Protoscolex viability was determined by mixing $20 \,\mu\text{L}$ of the protoscolex suspension with an equal volume of 0.1% eosin on a glass slide. The mixture was examined under a light microscope. Viable protoscoleces were identified by their intact greenish and reflective membranes, which indicated their ability to exclude the eosin dye. Non-viable protoscoleces that absorbed the dye and appeared red were counted. Additionally, the motility of the protoscoleces was observed as a primary indicator of viability. The viability of protoscoleces was determined using the following formula; viability (%) = (total number of protoscoleces/number of viable protoscoleces) *100. This assessment was conducted in triplicate, and the average viability rate was recorded. Only protoscoleces with a viability rate of 99% were selected for use in the experiments (29).

Irradiation of protoscoleces with alpha particles

A 100 μ L suspension of protoscoleces was exposed to alpha particles (3.4 MeV energy) emitted from a radioactive source ²⁴¹Amershium ²⁴¹(Am). The distance between the source and the samples was maintained at 1.77 cm (30). The exposure durations were set at 5, 20, 45, 60, and 90 minutes. Following irradiation, the viability of the protoscoleces was evaluated using the eosin staining method under a light microscope. The survival rate was calculated to analyze the impact of nuclear particle exposure on protoscolex viability. Each rat was given intraperitoneally 2000 protoscoleces mixed in PBS solution to ensure the supply of protoscoleces for additional research purposes.

Experimental design and animal groups

Thirty-six male albino rats were divided into six groups randomly, with three rats in each group. Group 1 rats were injected intraperitoneally with protoscoleces and exposed to alpha particles for 1 hour; then, after four months, rats were euthanized. Group 2 rats were injected intraperitoneally with protoscoleces and exposed to alpha particles for 1 hour. The rats were then euthanized after five months. Group 3 rats were injected intraperitoneally with protoscoleces and exposed to alpha particles for 1.5 hours. Then, after four months, the rats were euthanized. Group 4 rats were injected intraperitoneally with protoscoleces exposed to alpha particles for 1.5 hours and were euthanized after five months.

Group 5 (Positive control) rats were injected with non-irradiated protoscoleces and euthanized after four months. Group 6 (Positive control) rats received non-irradiated protoscoleces and were euthanized after five months. During the research period, all the rats were kept in standard laboratory conditions.

In order to investigate how alpha particles impact the gene activity of response-related genes, like TNF-α, IL-6, and IL-10, a specific procedure was implemented involving the retrieval of ribonucleic acid (RNA). This RNA was extracted from the tissue of treated rats. Then, it was transformed into DNA (cDNA) utilizing reverse transcription technology techniques. Following this process, the expression levels of the target genes were evaluated using real-time polymerase chain reaction (RT qPCR). RNA was obtained by following the instructions provided in the TransZol UP kit. Checked for quality using a spectrophotometer with absorbance ratios falling between 1. This RNA was then transformed into cDNA through the use of reverse transcriptase enzyme along with primers.

This study created primers for each gene of interest, such as TNF- α IL-6 and IL-10, and also designed a primer for a reference gene like GAPDH to ensure result accuracy. The qRT PCR reaction was carried out using an instrument with the reaction conditions programmed for 40 amplification cycles. The data analysis was done using the method to calculate the change in gene expression levels between the treated and control groups. This approach enabled the assessment of how alpha particle exposure impacts the expression of genes related to inflammation and immune responses, offering insights into the processes that drive the biological impact of radiation.

Gene expression analysis of TNF- α , IL-6, and IL-10 using RT-PCR

The levels of gene expression for TNF-α (tumor necrosis factor-α), IL-6 (interleukin- six), and IL-10 (interleukin- ten) were assessed through a technique called reverse transcription polymerase chain reaction (RT PCR). The procedure includes stages; the present study extracted RNA using an RNA extraction kit called Transzol UP, following the instructions provided by the manufacturer in the protocol guidebook. 200 microliters of chlorofluorocarbon were introduced to the Trizol-containing sample. Thoroughly mix it before allowing it to rest at room temperature for 3 minutes. After spinning the tubes 10,000 times the force of gravity for 15 minutes, the sample was split into two layers in a centrifuge machine. A layer with RNA in water and a bottom layer with compounds. The top portion was delicately moved to a tube before gently blending in an amount of pure ethanol. The blend was moved to a spin column and spun at 12,000 times gravity for half a minute; the liquid that passed through was thrown away. This process was repeated two times. 500 µL of washing solution (CB9) was added to the column, followed by centrifugation at

 $12,\!000 \times\! g$ for 30 seconds. Another 500 μL of washing solution (WB9) was added to the column, and the sample was centrifuged at $12,\!000 \times\! g$ for 30 seconds. An additional centrifugation step was performed at $12,\!000 \times\! g$ for 2 minutes at room temperature to remove any ethanol residues. The column was transferred to a new tube, and $25{-}200~\mu L$ of RNase-free water was added to dissolve the RNA. The extracted RNA was stored at -80°C for subsequent steps. The quality and quantity of the RNA were confirmed using a NanoDrop spectrophotometer before proceeding to the reverse transcription step for cDNA synthesis. The quality and strength of the RNA collected were checked with a Biodrop tool by putting a 1 μl RNA sample in the device following a spin to evenly spread it out before noting down the measurements.

The mRNA that was taken out was converted into cDNA through a process involving reverse transcriptase enzyme and a kit based on the instructions provided by the manufacturer. To prepare the primer for use, the experiment was left at 25°C for 10 minutes. Then, combine it with the EasyScript RT/RI Enzyme Mix before being incubated at 42°C for 15 minutes to facilitate transcription of the RNA sample. The enzyme is then deactivated by heating the reaction mixture to 85°C for 5 minutes in preparation for RT PCR analysis. Primers were utilized in conjunction with housekeeping genes for calibration to measure the levels of gene expression accurately (31).

Determining gene expression for 2FPGS

The level of gene expression was determined by comparing the CT values of the target gene and the housekeeping gene in test and control samples. The formula for calculating Δ CT is CT (target, test) - CT (ref, test) = Δ CT (control). The $\Delta\Delta$ CT (Δ CT test- Δ CT control) is calculated using the same formula (32).

Statistical analysis

A statistical analysis using ANOVA can provide insights into the significance of the observed differences between groups (n=3; P \leq 0.05). Gene Expression Folding (2^- $\Delta\Delta$ Ct) values were used to evaluate the changes in IL-6, IL-10, and TNF- α gene expression levels across the experimental groups (33).

Results

Genes expression in the liver of rats after 4 months of injection with protoscoleces

Table 1 shows an increase in IL-6 expression 1.3 and 1.04 compared to the control group after 1 and 1.5 hours of exposed protoscolecs to alpha particles, respectively. An increase in IL-10 gene expression 6.82 and 1.14 showed after 1 and 1.5 hours of exposure to protoscoleces to alpha particles, respectively, compared to the control group. The gene expression TNF- α reported significantly 4.5 more than

the control group after 1 hour of exposure to protoscoleces to alpha particles; there was no increase of the gene expression fold TNF- α associated with the control group

after 1.5 hours of exposure to protoscoleces to alpha particles.

Table 1: Gene expression levels gene in liver after 4 months

| Gene | Groups | Target gene | Housekeeping gene | Target gene Δ | Control Δ | ΔΔ | Expression/folding |
|-------|---------|-------------|-------------------|---------------|-----------|-------|--------------------|
| | Control | 34.22 | 24.73 | 8.71 | 8.71 | 0 | 1 |
| IL-6 | 1hour | 34.52 | 26.21 | 8.31 | 8.71 | -0.4 | 1.3 |
| | 1.5hour | 32.75 | 23.97 | 8.78 | 8.71 | 0.07 | 1.04 |
| | Control | 35.81 | 24.73 | 11.08 | 11.08 | 0 | 1 |
| IL-10 | 1hour | 34.52 | 26.21 | 8.31 | 11.08 | -2.77 | 6.82 |
| | 1.5hour | 35.24 | 23.97 | 11.27 | 11.08 | 0.19 | 1.14 |
| TNF-α | Control | 28.9 | 24.73 | 4.17 | 4.17 | 0 | 1 |
| | 1hour | 28.21 | 26.21 | 2.0 | 4.17 | -2.17 | 4.5 |
| | 1.5hour | 28.14 | 23.97 | 4.17 | 4.17 | 0 | 1 |

Genes expression in the liver of rats after 5 months of injection with protoscoleces

The results, as shown in table 2, reported that IL-6 gene expression increased after 1 and 1.5 hours of exposed protoscoleces to alpha particles 6.06 and 3.58, respectively, compared to the control group. As can be seen from the gene expression, IL-10 reported significantly more increase of 7.51 and 2.71 after 1 and 1.5 hours of exposed protoscoleces to alpha particles, respectively, compared to the control group. An increase can be seen from the data in the gene expression level of TNF- α 1.68 after 1 hour of exposure protoscoleces to alpha particles. On the other hand, the decrease was inhibited by 0.44 after 1.5 hours compared to the control group.

Genes expression kidney of rats after 4 months of injection with protoscoleces

Table 3 shows an increase in IL-6 expression 1.09 compared to the control group after 1 hour, while a reduction in IL-6 expression 0.84 occurred compared to the control group. An increase can be seen from the data in the gene

Table 2: Gene expression levels gene in liver after 5 months

expression level of IL-10 2.98 and 1.41 after 1 and 1.5 hours of exposure protoscoleces to alpha particles compared to the control group (Table 3). A moderate can be seen from the data in the gene expression level of TNF- α 1 after 1 hour of exposure protoscoleces to alpha particles. On the other hand, the decrease was inhibited by 0.11 after 1.5 hours (Table 3).

Genes expression kidney of rats after 5 months of injection with protoscoleces.

The results, as shown in table 4, reported that IL-6 gene expression increased after 1 hour of exposed protoscoleces to alpha particles 2.46 compared to the control group. There was no increase of gene expression IL-6 after 1.5 hours 0.51 compared to the control group. A decrease can be seen from the data in the gene expression level of IL-10 0.59 after 1 hour of exposure protoscoleces to alpha particles. On the other hand, a slight increase inhibited 1.07 after 1.5 hours compared to the control group (Table 4). An increase can be seen from the data in the gene expression level of TNF- α 7.5 and 8.87 after 1 and 1.5 hours of exposure protoscoleces to alpha particles compared to a control group (Table 4).

| Gene | Groups | Target gene | Housekeeping gene | Target gene Δ | Control Δ | ΔΔ | Expression/folding |
|-------|---------|-------------|-------------------|---------------|-----------|-------|--------------------|
| IL-6 | Control | 35.35 | 24.73 | 10.62 | 10.62 | 0 | 1 |
| | 1hour | 34.23 | 26.21 | 8.02 | 10.62 | -2.6 | 6.06 |
| | 1.5hour | 32.75 | 23.97 | 8.78 | 10.62 | -1.84 | 3.58 |
| | Control | 34.46 | 24.73 | 9.73 | 9.73 | 0 | 1 |
| IL-10 | 1hour | 33.03 | 26.21 | 6.82 | 9.73 | -2.91 | 7.51 |
| | 1.5hour | 32.26 | 23.97 | 8.29 | 9.73 | -1.44 | 2.71 |
| TNF-α | Control | 27.07 | 24.73 | 2.34 | 2.34 | 0 | 1 |
| | 1hour | 27.8 | 26.21 | 1.59 | 2.34 | -0.75 | 1.68 |
| | 1.5hour | 27.48 | 23.97 | 3.51 | 2.34 | 1.17 | 0.44 |

Table 3: Gene expression levels gene in kidney after 4 months

| Gene | Groups | Target gene | Housekeeping gene | Target gene Δ | Control Δ | ΔΔ | Expression/folding |
|-------|---------|-------------|-------------------|---------------|-----------|-------|--------------------|
| IL-6 | Control | 32.36 | 23.57 | 8.79 | 8.72 | 0.25 | 0.84 |
| | 1hour | 35.98 | 25.51 | 10.47 | 10.47 | 0 | 1 |
| | 1.5hour | 33.8 | 24.91 | 8.89 | 10.47 | -1.58 | 2.98 |
| IL-10 | Control | 34.56 | 23.57 | 10.99 | 10.47 | 0.5 | 1.41 |
| | 1hour | 27.5 | 25.51 | 1.99 | 1.99 | 0 | 1 |
| | 1.5hour | 26.91 | 24.91 | 2.0 | 1.99 | 0.01 | 1 |
| TNF-α | Control | 28.69 | 23.57 | 5.12 | 1.99 | 3.13 | 0.11 |
| | 1hour | 32.36 | 23.57 | 8.79 | 8.72 | 0.25 | 0.84 |
| | 1.5hour | 35.98 | 25.51 | 10.47 | 10.47 | 0 | 1 |

Table 4: Gene expression levels gene in kidney after 5 months

| Gene | Groups | Target gene | Housekeeping gene | Target gene Δ | Control Δ | ΔΔ | Expression/folding |
|-------|---------|-------------|-------------------|---------------|-----------|-------|--------------------|
| IL-6 | Control | 35.22 | 25.51 | 9.71 | 9.71 | 0 | 1 |
| | 1hour | 33.32 | 24.91 | 8.41 | 9.71 | -1.3 | 2.46 |
| | 1.5hour | 34.23 | 23.57 | 10.66 | 9.71 | 0.95 | 0.51 |
| | Control | 34.33 | 25.51 | 8.82 | 8.82 | 0 | 1 |
| IL-10 | 1hour | 34.51 | 24.91 | 9.6 | 8.82 | 0.75 | 0.59 |
| | 1.5hour | 32.36 | 23.57 | 8.79 | 8.82 | -0.1 | 1.07 |
| TNF-α | Control | 31.8 | 25.51 | 6.29 | 6.29 | 0 | 1 |
| | 1hour | 32.28 | 24.91 | 3.37 | 6.29 | -2.92 | 7.5 |
| | 1.5hour | 26.81 | 23.57 | 3.24 | 6.29 | -3.05 | 8.87 |

Genes expressions in the intestine of rats after 4 months of injection with protoscoleces

Table 5 shows a decrease in IL-6 expression of 0.65 compared to the control group after 1 hour of exposed protoscoleces to alpha particles. On the other hand, a slight increase was recorded after 1.5 hours compared to the control group. An increase in IL-10 gene expression 7.11 was shown after 1 hour of exposure to protoscoleces to alpha particles compared to the control group. No increase in gene expression of IL-10 after 1.5 hours was detected compared to the control group (Table 5). It can be seen from the data in table 5 that the gene expression TNF- α reported a significantly greater increase of 13.7 than the control group after 1 hour of exposure protoscoleces to alpha particles; there was no increase of the gene expression fold TNF- α associated with the control group after 1.5 hour of exposure protoscoleces to alpha particles.

Genes expression intestine of rats after 5 months of injection with protoscoleces The results, as shown in table 6, reported that IL-6 gene

The results, as shown in table 6, reported that IL-6 gene expression increased by 5.46 after 1 hour of exposed protoscoleces to alpha particles. A slight reduction of 0.92 of gene expression IL-6 was found after 1.5 hours compared to the control group. As can be seen from the gene expression IL-10 in table 6, the gene expression IL-10 reported a significantly greater decrease of 0.35 after 1 hour of exposed protoscoleces to alpha particles. An increase in gene expression IL-10 1.16 occurred after 1.5 hours compared to the control group (Table 6). An increase can be seen from the data in the gene expression level of TNF- α 3.11 after 1 hour of exposure protoscoleces to alpha particles. On the other hand, the decrease was inhibited by 0.19 after 1.5 hours compared to the control group (Table 6).

Table 5: Gene expression levels gene in intestine after 4 months

| Gene | Groups | Target gene | Housekeeping gene | Target gene Δ | Control Δ | ΔΔ | Expression/folding |
|-------|---------|-------------|-------------------|---------------|-----------|-------|--------------------|
| IL-6 | Control | 34.36 | 25.98 | 8.38 | 8.38 | 0 | 1 |
| | 1hour | 36.75 | 27.77 | 8.98 | 8.38 | 0.6 | 0.65 |
| | 1.5hour | 32.57 | 24.32 | 8.25 | 8.38 | -0.13 | 1.09 |
| | Control | 34.36 | 25.98 | 8.38 | 8.38 | 0 | 1 |
| IL-10 | 1hour | 33.32 | 27.77 | 5.55 | 8.38 | -2.83 | 7.11 |
| | 1.5hour | 37.27 | 24.32 | 12.95 | 8.38 | 4.57 | 0.1 |
| TNF-α | Control | 28.18 | 25.98 | 2.2 | 2.2 | 0 | 1 |
| | 1hour | 26.19 | 27.77 | -1.58 | 2.2 | -3.78 | 13.7 |
| | 1.5hour | 27.89 | 24.32 | 3.57 | 2.2 | 1.37 | 0.38 |

Table 6: Gene expression levels gene in intestine after 5 months

| Gene | Groups | Target gene | Housekeeping gene | Target gene Δ | Control Δ | ΔΔ | Expression/folding |
|-------|---------|-------------|-------------------|---------------|-----------|-------|--------------------|
| IL-6 | Control | 35.5 | 25.98 | 9.52 | 9.52 | 0 | 1 |
| | 1hour | 34.84 | 27.77 | 7.07 | 9.52 | -2.45 | 5.46 |
| | 1.5hour | 33.96 | 24.32 | 9.64 | 9.52 | 0.12 | 0.92 |
| IL-10 | Control | 34.45 | 25.98 | 8.47 | 8.47 | 0 | 1 |
| | 1hour | 37.75 | 27.77 | 9.98 | 8.47 | 1.51 | 0.35 |
| | 1.5hour | 32.57 | 24.32 | 8.25 | 8.47 | -0.22 | 1.16 |
| TNF-α | Control | 27.1 | 25.98 | 1.12 | 1.12 | 0 | 1 |
| | 1hour | 27.25 | 27.77 | -0.52 | 1.12 | -1.64 | 3.11 |
| | 1.5hour | 27.81 | 24.32 | 3.49 | 1.12 | 2.37 | 0.19 |

Discussion

Hydatid echinococcosis is a careless and inveterate parasitic illness caused by the protoscoleces (34). The immune responses against hydatid echinococcosis are singular for some characteristic to stimulate all of the cytokine's inhibitor and activator for immunity host, Thelper 1 (Th1) cells stimulate cytokines against infections hydatid cyst to allow host conservative immunity while the work of Thelper 2 (Th2) cells is inducing inhibitor cytokines which activation the chronic infection of hydatid cyst (35-38). The majority of parasite diseases are associated with IL-10 and IL-6, which are induced by Th2 cell response. These cytokines are responsible for the physiopathology of the contagion (39,40).

Tumor necrosis factor TNF- α is a pro-inflammatory interleukin related to the homicide of tumor cells; it has been observed that the interleukin supplies a rapid form of host defense versus infection (41,42). The present study indicates that exposure of protoscoleces to alpha particles before their injection into the peritoneal cavity of rats has a dose- and time-dependent effect on the expression of key immunerelated cytokines, including IL-6, IL-10, and TNF- α . The duration of radiation exposure, 1 and 1.5 hours, significantly influenced the magnitude and direction of gene expression changes, highlighting the complex biological responses to radiation-modified pathogens and their subsequent impact on host immune regulation.

IL-6 symbolizes an inflammatory substance associated with immune responses and tissue healing processes (43,44). This study indicates that rats injected with protoscoleces exposed to alpha particles for 1 hour displayed heightened interleukin expression at both 4- and 5-month intervals with a noticeable impact observed after the longer duration (for instance-6.82-fold increase in Liver). This implies a period of inflammation in response likely initiated by the tissue harm caused by the irradiated protoscoleces; these results agree with the findings of other studies (45). On the other hand, when exposed for 1.5 hours, IL-6 expression showed varying effects. Certain groups exhibited decreased levels of expression, for example, 0.84 times in the kidney organ after 4 months and 0.51 times after five months, indicating a

cellular response that adapts to the presence of radiationaltered protoscoleces (46). The present study shows a decrease in expression levels, which might suggest that there could be some unique immune system adjustments specific to the tissue itself; these findings are in agreement with those obtained by Chapli (47). The intestine tissue showed a reaction after 5 months in the 1-hour group with a significant increase in IL-6. In contrast, the group with prolonged exposure of 1.5 hours had nearly normalized expression, indicating that the body might have triggered healing processes or built up a tolerance to extended exposure, reducing the impact.

IL-10 functions as an inflammatory cytokine that is critical for regulating immune response and reducing excessive inflammation levels in the body, according to the research findings present in the study. In the liver, exposure of protoscoleces to low-dose radiation for 1.0 hours led to heightened IL-10 expression five months later, signaling an adaptive anti-inflammatory response within the host organism. This implies that being subject to moderate radiation exposure from the injected pathogen could trigger compensatory mechanisms aimed at mitigating inflammatory harm (48,49). Excessive exposure to radiation for 90 minutes resulted in a decreased IL-10 expression in groups like the kidneys and intestines for 90 minutes, suggesting immune system suppression or cell damage caused by the high levels of radiation in the protoscoleces introduced. Zhu et al. (50) show that reduction could be a result of cell death triggered by radiation within the injected pathogens, causing changes in the host response.

After 5 months of delay in the rise of IL-10 levels, post-treatment was observed primarily in the liver 1.0-hour group, suggesting a prolonged reaction against changes induced by exposure to alpha particles in protoscoleces was seen within that group. On the other hand, suppressed levels of IL-10 were noted in the kidney and intestine 1.5-hour group, which could indicate the effects of high doses of radiation on the immune system equilibrium over time. These results highlight the benefits of radiation exposure on immune responses, while excessive exposure to radiation may disrupt the balance between inflammatory and anti-inflammatory processes (51).

TNF-α plays an interleukin that triggers tissue inflammation. The current study found that TNF-α levels varied among different groups and time points due to the impact of radiation on immune regulation after exposing protoscoleces to alpha particles exposure varied, depending on the level of exposure 60 minutes. Another important finding was an increase in TNF-α expression in groups of liver and intestine after one hour. A possible explanation for this might be that a pro-inflammatory reaction is linked to changes induced by radiation in protoscoleces and their interactions with the hosts' immune system, which agrees with the findings of other studies (52,53). On the contrary, exposure for 90 minutes initially reduced the expression of TNF- α in the kidney after four months. These results seem to be consistent with other research, which found that the depletion or harm to immune cells is due to increased radiation levels (54,55).

Nonetheless, a subsequent rise in TNF- α expression showed after five months indicates a healing process following the earlier suppression. The difference between being exposed to radiation for 1 and 1.5 hours shows that moderate radiation exposure boosts activity by raising TNF- α levels, while prolonged exposure may initially suppress immune reactions but later trigger a secondary inflammatory response surge (56). The fluctuating levels of TNF- α expression suggest that radiation-induced immune adjustment is a developing process with lasting impacts on immune balance in reaction to irradiated protoscoleces (57).

Conclusion

The present study detected that alpha particles affect the levels of gene expression IL-6, IL-10, and TNF- α in the liver, kidney, and intestine of rats and could be used as therapeutic methods in the future against hydatidosis.

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Conflicted interest

None

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التحري عن دور جسيمات الفا في مستويات التعبير الجيني للسايتوكينات في الجرذان المصابة بداء المشوكات الكيسية

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الخلاصة

يعد داء الأكياس العدرية من الأمراض الطفيلية الواسعة الانتشار في جميع أنحاء العالم والذي يسببه الطور اليرقي لدودة المشوكات الحبيبية. تحرت الدراسة الحالية عن تأثير الرؤيسات الأولية بعد تعريضها لجسيمات الفا على تعبير الجينات المتعلقة بالاستجابة المناعية في

الجرذان. في هذه الدراسة تم ملاحظة التعبير الجيني لكل من عامل نخر الأورام والانترلوكين-٦ والانترلوكين-١٠ في اعضاء مختلفة من الجرذان المصابة بالأكياس العدرية. تم استخدام ست مجاميع من الجرذان بعضها حقنت، داخل البريتون، بالرؤيسات الأولية المعرضة لجسيمات الفا بفترات زمنية مختلفة (ساعة وساعة ونصف) بينما حقنت مجموعة السيطرة بالرؤيسات الأولية فقط بشرحت الجرذان بعد اصابتها بأربعة وخمسة أشهر وتم فحص مستويات مختلفة من الجينات المعلمة مثل عامل نخر الأورام والانترلوكين-٦ والانترلوكين-١٠ في كبد وامعاء وكلى الجرذان أظهرت النتائج أن التعرض لجسيمات الفا أدى إلى تغييرات ملحوظة في تعبير هذه السايتوكينات اعتمادا على الجرعة ومدة التعرض، إذ أدى التعرض بعد ساعة وساعة ونصف إلى زيادة ملحوظة في تعبير جين عامل نخر الأورام في الجرذان خاصة بعد ٤ أشهر. إن التعرض لجسيمات الفا إثر على الاستجابة الالتهابية على المستويين النسجي والوظيفي، إذ ان الفترات الزمنية للتعرض لجسيمات الفا أثرت على الاستجابات البيولوجية المعقدة للرؤيسات الأولية المعدلة اشعاعيا والتي ادت بدورها الى تنظيم الجهاز المناعي للمضيف.