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STAT3 and the resistance ability of HSC to ionizing radiation

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Abstract:

HSC is a very important part of the blood system and has a functional transcription factor, STAT3. When STAT3 is damaged, it is very likely to lead to death of HSC and even the whole organism. The ionizing radiation, on the other hand, is ubiquitous in the universe and is sure to cause damage to HSC. There is no doubt that the ionizing radiation damage to STAT3 is a big obstacle we need to overcome in our journey to explore space. Thus, in this work we will use the bone marrow hematopoietic stem cells of mice to further study the regulation pathway between STAT3 and the resistance ability of HSC to ionizing radiation.

Keywords: HSC, STAT3, IR

1. Introduction

HSC is a kind of multipotent stem cell whose niches are perivascular in the bone marrow [1]. As shown in Figure 1, HSC has the potential to self-renew and differentiate into many different kinds of blood cells. It plays a very important role in maintaining the normal function of the blood system. However, scientists believe that after exposure to a certain range of ionizing radiation (IR) doses, IR will injure HSC by damaging the DNA sequence, which is the primary cause of death [2]. They already confirmed that IR can damage the HSC by impairing HSC's ability to self-renew by induction of HSC differentiation, and finally, it leads to organism death. But more details about the pathway of how specific damage of DNA to the HSC differentiation are still unknown. Since we do know that STAT3 is one of the most important factors which can regulate the transcription of DNA, the main topic of this passage is to explore the relationship between HSC differentiation and the extent of STAT3 expression. With the knowledge of the induction pathway, we can improve the IR resistance ability of HSC by controlling the expression of STAT3. Thus, it may assist in the further development of cell culture in the field of aerospace exploration.

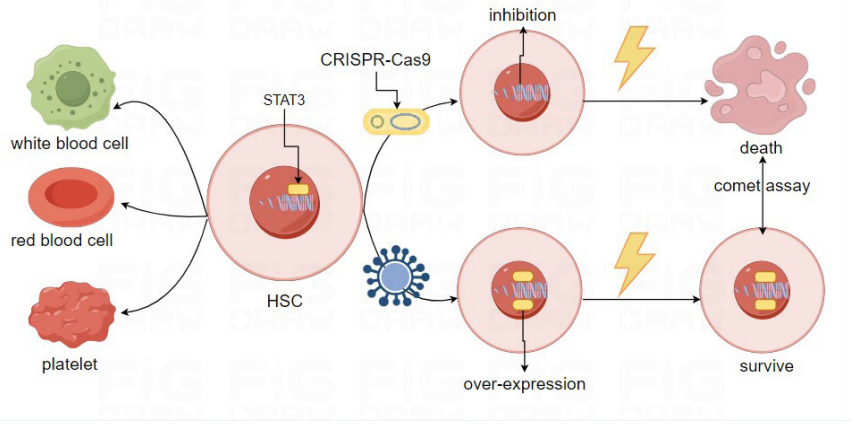

Figure 1. The main function of HSC in the blood system and the brief schematic of the experimental principle

2 Method

2.1 Signal transducer and activator of transcription 3 (STAT3)

STAT3 proteins are potential cytoplasmic transcription factors. Cytokines and growth factors can motivate STAT3 [3]. So far, scientists have discovered 7 members in the STAT family, and these members share an overall general structure [4]. STAT3 takes part in many biological processes, such as cell differentiation and angiogenesis [5]. STAT3 directly interacts with DNA, acting as a signal transducer and transcription factor in cells. Its cytoplasmic activation results in dimerization and nuclear translocation, and it is involved in the transcription of a large number of target genes [6]. However, since so many other proteins and factors are included in the transcription of DNA, we need first to check if the STAT3 is related to the IR-resistance ability of HSC.

2.2 The clustered regularly interspaced short palindromic sequence repeats-Cas (CRISPR-Cas)

CRISPR-Cas is one type of gene editing technology. By using a natural defense mechanism of bacteria, it can modify DNA. By recording the DNA sequence related to STAT3 expression, the bacteria could recognize the specific sequence and knock the chosen genes out after being inserted into the cells [7]. We will use CRISPR-Cas to control the variable of STAT3 in the first experiment.

2.3 Comet assay

The comet assay is a functional method to detect nuclear DNA damage in individual eukaryotic cells [8]. By determining the intensity of the comet tail relative to the head, it shows the extent of broken DNA [9]. In the following experiments, we will use this method to measure the extent of DNA damage, which inversely stands for IR-resistance ability of HSC.

2.3.1 Question 1: if STAT3 can decrease DNA damage?

We choose to use the bone marrow hematopoietic stem cells of mice as the model for the following experiments. In order to verify the mentioned question, we need to culture two groups of HSC: group A, as the control group, uses normal HSC; group B, as the experimental group, uses CRISPR-Cas system to knock out STAT3 genes in order to achieve zero expression level. Then apply different concentrations of IR to the HSC and control other irrelevant variables during the culture. Use the comet assay to measure the damage extent. Draw the graph of DNA damage extent against different groups. To show that the STAT3 can increase the IR-resistance ability of HSC, we expect to see that group A has a much lower extent of DNA damage than group B.

2.3.2 Question 2: if over-expressing STAT3 genes increase the IR resistance ability of HSC?

Next, we hope to improve the resistance ability of HSC by over-expressing STAT3 genes. According to recent research, scientists have already shown that the activation of an ectopic expression of STAT3 could promote DNA replication. Here, we want to test if this ability could help the damaged DNA repair itself under exposure to IR and increase the resistance ability. Thus, we can clone the genes into a plasmid and then transfect or transduce the plasmid to our cultured HSC to over-express STAT3 genes. Then expose the cells IR and use the comet assay to measure the extent of DNA damage. Repeat these experiments under different concentrations of IR to get a conclusive result. If the over-expression of the STAT3 can increase

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the resistance ability, we expect to see the groups of HSC with plasmid transfection have lower DNA damage extent than the control groups.

3 Conclusion

According to the results of these experiments, we consider that the resistance ability of HSC to ionizing radiation is positively correlated with the extent of expression of STAT3 in HSC. However, even in this pathway, many other correlating factors are still not scientifically studied, so the maximum IR resistance ability of HSC is still unknown. We hope that this research will be helpful for the research of genetic modification of space organisms in the future.

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