The effect of radiation on motility in *Caenorhabditis elegans*

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Abstract

Background

Organisms are exposed to a variety of stresses, including chemicals and radiation. The biological effects of radiation depend on the dose, radiation quality, and cell types. Linear Energy Transfer (LET) is the indicators of radiation effects. High-LET radiation, such as carbon ion beams induces more complicated DNA damage than low-LET radiation such as γ rays. On the other hand, the effects of high-LET radiation on the tissue and individual level are not yet fully understood.

In *Caenorhabditis elegans*, it is known that carbon ion beams irradiation and γ-rays irradiation decrease the motility of adult worms. Therefore, in the present study, I first aimed to establish a motility evaluation system suitable for the analysis of radiation effects in *C. elegans*. Next, using the established experimental system, the effects of radiation on motility were clarified using several types of radiation, dose, and genetic condition. In addition, region-specific microbeam irradiation was performed to clarify the important regions for radiation-induced motility reduction. Moreover, I observed changes in whole-body motility after irradiation, and clarified whether the reduced motility is restored or whether it leads to individual death. Then, I focused on autophagy, the degradation mechanism of damaged proteins and organelles, and investigated its involvement in radiation effects.

methods

In this study, *C. elegans* were cultured to adulthood and irradiated with carbon beams or γ rays for motility analysis and autophagy induction analysis. Wild Type worms and mutant strains were obtained from the Caenorhabditis Genetic Center. For the γ rays irradiation, a cobalt-60 source was used. For carbon ion beam irradiation, carbon ion ($^{12}C^{5+}$) particles were accelerated by an AVF cyclotron at Takasaki Ion Accelerators for Advanced Radiation Application (TIARA) facility at QST-Takasaki. For site-specific irradiation, *C. elegans* was encapsulated in a microfluidic chip and irradiated with accelerated carbon ion particles at each Φ60 μm range. For motility analysis, crawling motility on agar plate and swimming motility in buffer solution were measured and evaluated. For the detection of autophagy induction, GFP fluorescence was observed over time using a GFP reporter strain of autophagy marker gene, *lgg-1*.

Results and Discussion

First, to investigate the effects of radiation on motility, a motility analysis system was constructed to analyze crawling motility on an agar plate. Using this system, I evaluated motility immediately after carbon ion beams irradiation or γ rays irradiation, found a dose-dependent decrease in motility.

Next, I observed changes in whole body motility after irradiation to clarify whether the reduced motility is recovered or lead to death. As a result, I found that both carbon ion beams irradiation and γ rays irradiation caused partial recovery of motility within 24 hours. Similarly, I measured swimming motility in buffer solution after irradiation, and observed the radiation induced motility defect and partial recovery as well. The results suggest that radiation-induced motility effect could be classified into two types:

reversible and irreversible effects.

To clarify important regions for the irradiation induced decrease in motility, region-specific microbeam irradiation was performed and motility was analyzed. As a result, the motility after irradiation of the anterior half of the body was lower than that after irradiation of the posterior half. This suggests that the anterior half of the body is important in the motility of radiation.

In addition, several genes known to function in the radiation response were selected and analyzed to elucidate the pathways involved in the recovery of motility. The results suggest that *atm-1*, a master regulator gene of radiation response, which is the *C. elegans* ortholog of the mammalian Ataxia telangiectasia mutated gene (*ATM*), is involved in the recovery response to radiation.

Since autophagy, a degradation mechanism of damaged proteins and organelles, was considered a possible mechanism for the recovery of reduced motility, GFP fluorescence was observed using a GFP reporter strain of *lgg-1*, a marker gene for this mechanism. As a result, I found that autophagy was induced in the pharynx, neurons, and intestine.

Conclusion

I found that the effects of radiation on motility are more important in the anterior half of the body. Furthermore, it was found that radiation-induced motility could be classified into two types: reversible and irreversible effects. The reversible response was found to recover within 24 hours of irradiation in both high-LET and low-LET radiation. ATM-1 was found to be involved in this motility recovery response. Autophagy was also found to be induced during motility recovery.