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MICE TESTES AND DUCT DEFERENCE DURING SPACE FLIGHT (RR-4 EXPERIMENT): CYTOSKELETON STRUCTURE AND ITS REGULATION

Abstract

To date, the influence of gravity to male reproductive system has not been well studied. The purpose of our work was to evaluate the levels of the cytoskeletal proteins and the mRNA levels of the genes that encode them in the testes and duct deferens of mice whose tissues were fixed in space flight conditions (experiment Rodent Research-4, SpaceX-10 mission, February 2017, USA). In addition, we sought to evaluate epigenetic events, in particular, the levels of 5-hydroxymethylcytosine and of the enzymes that regulate DNA methylation/demethylation. The samples, in accordance with the NASA-Roskosmos protocol "Utilization Sharing Plan on-board ISS" (signed on July 18, 2013), were delivered to Russia on dry ice without defrosting. The results suggested that there was no change in the levels of the studied cytoskeletal proteins (beta- and gamma-actin, alpha-actinin 1 and 4, beta-tubulin and desmin) in the flight group, although there was a decrease in Actn1 mRNA in the duct deferens and in beta-tubulin in the testes. The reasons for the changes in expression could be associated with a wide range of factors, such as histone modifications, but for higher mammals, it is most likely because of a change in the DNA methylation levels of CpG-islands in the promoter regions of genes. In this study, we attempted to analyze the total DNA methylation level by restriction analysis and, accordingly, CG-islands in the promoter regions of the cytoskeletal genes were studied; however, the isolated genomic DNA was not of a high enough quality to conduct such an analysis. Nevertheless, we were able to evaluate the level of 5-hydroxymethylcytosine (5hmC). There were no changes in the 5-hydroxymethylcytosine levels in the testes or duct deferens. The protein content of the S-phase methylase DNMT1, de novo methylase DNMT3a, and demethylases of the TET family (TET1, TET2, and TET3) of the flight group both in the testes and duct deferens remained at the same level as the control group. But, the expression of the Hdac1 deacetylase gene was reduced in the duct deferens, while the mRNA levels of the acetylase Hat1 remained constant; and in the testes, while the expression of the deacetylase was reduced, the expression of the acetylase was increased. This work was financially supported by the program for fundamental research SSC RF – IBMP RAS; program "Cell and Molecular Biology" of the RAS Presidium; Russian Academic Excellence Project 5-100.