Секция «Тезисы на английском языке»

Applying CRISPR / Cas9 genome editing technology to knockout the urokinase receptor gene for treatment of neuroblastoma

Научный руководитель – Semina Ekaterina Vladimirovna

Shmakova Anna Andreevna

Студент (специалист)

Московский государственный университет имени М.В.Ломоносова, Факультет фундаментальной медицины, Кафедра биологической и медицинской химии, Москва, Россия

E-mail: pr.me@mail.ru

Neuroblastoma is one of the most common tumors in children (under 15 years old), which accounts for about 10% of all children oncology. One of the specific features of the tumor is its ability to rapid progression and early metastasis; the presence of metastases is poor prognostic factor of neuroblastomas [1]. Several studies have shown that in non-differentiated tumor types, which are characterized by unfavorable histology, of these neuroblastoma, overexpression of urokinase (uPA) and its receptor (uPAR) is observed. At the same time a high level of uPA and uPAR correlated with a high risk of invasion and metastasis of neuroblastoma and is considered as a prognostic marker of poor prognosis [2]. Thus, the use of effective approaches to reduce gene expression of *uPA* and *uPAR* is regarded as one of the most promising approaches to the so-called a gene-specific targeted neuroblastoma treatment.

In our work we applied CRISPR / Cas9 genome editing technology for knocking out *uPAR* gene in the linear culture of mouse neuroblastoma cells Neuro2a. We created nonviral plasmid construction containing nickase Cas9 sequence and two guide sequences directed to the first exon of *uPAR* gene. This system makes a double-stranded breaks in the *uPAR* gene repair of which leads to impairment of gene function. To analyze the effectiveness of created plasmid transfection of neuroblastoma was performed, with subsequent evaluation of the level of membrane expression of *uPAR*. After three transfections of Neuro2a more than half of the cells (52%) lacked *uPAR* expression on the cell membrane. Cell sorting after third transfection enabled to select clones containing no uPAR to study their proliferation. It has been found that the degree of *uPAR* silening influences the rate of cell proliferation, and the lowest rate of proliferation were observed in clones Neuro2a, where uPAR was absent.

Thus, our findings enable the use of CRISPR / Cas9 technology to obtain cells with low *uPAR* expression which can become a model for further study of the role of the urokinase system in the development, progression and metastasis of neuroblastoma. Knocking out *uPAR* gene in tumor cells by using this technique can be considered as a possible therapeutic approach to reduce the proliferation of tumor cells in the treatment of neuroblastoma.

Источники и литература

1) 1. Stroganova A.M., Karseladze A.I. Neuroblastoma: morphological structure, molecular and genetic characteristics and prognostic factors // The success of molecular oncology. 2016. №1. P. 32-43.
2) 2. Li, Peng et al. Role of urokinase plasminogen activator and its receptor in metastasis and invasion of neuroblastoma // Journal of Pediatric Surgery. 2004. № 39(10). P.1512 - 151