プラズマ遺伝子導入法を用いた細胞医療の有用性検討

Examination of usefulness of cell medicine using plasma gene transfer method

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We investigated the possibility of cell medicine by plasma gene transfection method using a mouse type I diabetes model. Insulin gene coded plasmid is transferred into cell by micro plasma treatment and is expressed in mouse hepatocytes and blood cells. Moreover, the random integration of the transgene into the genome was very low compared to existing methods such as electroporation. These results are suggested that the micro plasma gene transfection method enables practical application such as cell medicine.

1. Introduction

The Gene transfer is a technique for creating functional cells by incorporating genes artificially. The conventional gene transfer method has problems such as damage to cell, low transfer efficiency, and side effects caused by genome rearrangement and random integration after transfer. Therefore, gene therapy has not been put into practical use except for fatal diseases such as cancer and serious genetic diseases. Our laboratory aims to establish medical application technology for the cure of intractable diseases by plasma gene transfer method which is less damaging than the conventional gene transfer method and has succeeded in gene transfer in many types of cells.

As one of the many intractable diseases, we focused on type I diabetes, in which the β cells in the pancreas are destroyed and the patient cannot produce insulin by himself. We have started to investigate that possibility of practical application of cell medicine by plasma gene transfection method

2. Method and Result

This paper investigated genomic integration into target cells using plasma gene transfer. In preparation for the experiment, a plasmid for mouse insulin expression and red fluorescence was constructed.

At the same time, gene transfer by using various gene transfer methods was performed to CHO cells, which are frequently used for gene expression, and the expressed cells were cultured for 21 days. As a result, a large number of colonies could be confirmed by the electroporation method and the lipofection method, but almost no colonies were formed by the plasma method. From this was sugested that the plasma method causes almost no random integration

Fig.1. Map of mouse insulin expression Plasmid

3.Conclusion

By using the microplasma method, it was revealed that the plasma method has very low random integration into chromosomes.

In the future, we will develop introduction conditions to improve the introduction efficiency, confirm the chromosomal abnormalities of the obtained insulin-expressing cells, and verify that safe gene therapy is possible by plasma gene transfection method.

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