摘　要

胶质瘤是脑神经细胞癌变形成的恶性肿瘤，约占颅内肿瘤的 50%，具有发病率高，治愈率低的特点。而恶性肿瘤能够无限增殖和转移的关键在于血管的新生。VEGF 是主要的血管生成促进因子，且 VEGF 对促进缺血性心脏病、 缺血缺氧性脑病、肾脏疾病、骨骼疾病的恢复具有重要意义；因为其对内皮细胞、淋巴血管增殖的作用，VEGF 成为研究热点，通过对 VEGF 及其信号通路的研究与探讨，极有可能研发出抗肿瘤的重要方法与手段。

本课题采用 Trizol 的方法，从 293T 细胞中提取总基因组，再经过反转录 PCR，并使用我们设计的引物， 达到 VEGF 基因的扩增；并通过酶切连接的方式构建重组质粒 pcDNA3.1-VEGF，我们利用限制性核酸内切酶进行酶切鉴定重组质粒，经过电泳显示出大小为 1926 bp 的 DNA 条带，表明重组质粒 pcDNA3.1-VEGF 构建成功；转染 C6 细胞，SDS-PAGE 结果显示，出现一条大小约为 45KD 的蛋白条带，说明重组质粒转入 C6 细胞并成功表达。采用 MTT 法鉴定 VEGF 基因表达对 C6 细胞产生的影响。MTT 结果显示，转入重组质粒 pcDNA3.1-VEGF 的 C6 细胞，培养 48　h 后，细胞生长加快。本课题的研究内容，证明了 VEGF 基因可促进 C6 细胞的增殖和转移，为 VEGF 抗体药物研发提供基础。

关键词　VEGF　C6 细胞　pcDNA3.1　表达　基因

Effect of VEGF gene expression on C6 cells

Abstract

Glioma is a malignant tumor formed by canceration of brain nerve cells, accounting for about 50% of intracranial tumors.The key to the unlimited proliferation and metastasis of malignant tumors lies in the regeneration of blood vessels.VEGF is the main factor promoting angiogenesis, and it is of great significance to promote the recovery of ischemic heart disease, hypoxic-ischemic encephalopathy, kidney disease and bone disease.Because of its effect on endothelial cells and lymphatic vascular proliferation, VEGF has become a research hotspot. Through the study and discussion of VEGF and its signaling pathway, it is highly possible to develop important anti-tumor methods and means.In this study, Trizol method was used to extract the total genome from 293T cells, which was then subjected to reverse transcription PCR, and the primer designed by us was used to achieve the amplification of VEGF gene.The recombinant plasmid pcdna3.1-vegf was constructed by enzyme digestion and ligation. The recombinant plasmid was identified by enzyme digestion with restriction endonucleoidase. The DNA band of 1926 bp was shown by electrophoresis, indicating that the recombinant plasmid pcdna3.1-vegf was successfully constructed.The sds-page results of transfected C6 cells showed a protein band of about 45KD, indicating that the recombinant plasmid was successfully expressed in C6 cells.The effect of VEGF gene expression on C6 cells was determined by MTT assay.MTT results showed that C6 cells transferred into recombinant plasmid pcdna3.1-vegf were cultured for 48 h, and the cell growth was accelerated.The research content of this project proves that VEGF gene can promote the proliferation and transfer of C6 cells, providing a basis for the development of anti-vegf drugs.

Key words:VEGF Glioma cells pcDNA3.1 express gene