CAPRI 细胞与 CIK 细胞分泌 IFN-γ 的研究

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目的：对比研究链式激活的免疫 (CAPRI) 细胞和细胞因子诱导的杀伤 (CIK) 细胞分泌 IFN-γ 的情况，以探讨两种免疫细胞体外抗肿瘤作用的差别。

方法：分别抽取 3 名健康志愿者外周血 50ml，用淋巴细胞分层液分离出 3 份单个核细胞 (PBMC) 悬液，均调整细胞浓度为 1×106/ml，每份细胞悬液平均分为两部分，一部分经过多种细胞因子得到 CAPRI 细胞，另一部分经细胞因子共同诱导收获 CIK 细胞，应用 ELISPOT 技术检测 IFN-γ 的分泌情况。

结果：CAPRI 细胞与 CIK 细胞在 1×106/ml、5×105/ml 两个不同细胞浓度下应用 ELISPOT 技术检测，CAPRI 细胞组 ELISPOT 检测的 IFN-γ 的斑点数分别为 (126.2±10.31)、(48.8±10.99)，均低于 CIK 细胞组 (409.3±7.76)、(159.3±15.45) (P ＜ 0.001/P ＜ 0.001)。

结论：CAPRI 细胞分泌 IFN-γ 的水平低于 CIK 细胞，对自体免疫细胞细胞因子检测提供了实验模型。

关键词：CAPRI 细胞；CIK 细胞；IFN-γ

Transcriptional regulation of TGF-β1 combined with miR200c mimics increase therapeutic efficacy of tumor vaccine B16F10/GPI-IL-21 in melanoma bearing mice

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The transforming growth factor β (TGF-β) in tumor environment can promote the upregulation of Zinc-finger E-box binding homeobox and repress miR-200c, and result in tumor growth, invasion, and metastasis by triggering epithelial to mesenchymal transition (EMT). To find a potential anti-cancer regimen for treatment of aggressive malignant melanoma, an investigation of the therapeutic effect and molecular mechanism is needed. To this end, we designed a therapeutic paradigm that using the transcriptional regulation of TGF-β1 combined with injection of miR200c mimics to enhance therapeutic effect of B16F10 cell vaccine expressing interleukin 21(IL-21) in the glycosylphosphatidylinositol (GPI)-anchored form (B16F10/GPI- IL-21) on melanoma bearing mice. An expression vector-based small hairpin RNA targeting TGF-β1 (shTGF-β1) and tumor vaccine B16F10/GPI-IL-21 were first developed, and the B16F10 cells stably transfected with shTGF-β1 were then selected. After C57BL/6 mice were vaccinated subcutaneous with inactivated B16F10/GPI-IL-21 vaccine three times at two week interval, the vaccinated mice were randomly divided into four therapeutic groups: the B16F10/ PBS group; B16F10/shTGF-β1 group; miR200 mimics group and B16F10/GPI-IL-21+ mB16F10/shTGF-β1+miRNA200c group. The immune responses, EMT associated molecular expression, tumorigenicity and metastasis of lungs, livers, kidneys, and inguen lymph nodes in mice were investigated, respectively. The results showed that all immunized and treated mice significantly indicated increases in the NK and CTL activities and IFN-γ level, decreases in TGF-β1, TNF-α and $CD4^+CD25^+$Treg compared with the mice treated with B16F10/PBS. In particular, the treatment with B16F10/GPI-IL-21+ B16F10/shTGF-β1+miRNA200c induced a strong immune responses and accompanied with