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## NKG2D介导NK细胞对鼻咽癌细胞杀伤作用的体外研究

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### NKG2D Mediated Cytotoxicities of NK Cells Against Human Nasopharyngeal Carcinoma Cell Line (CNE2) in Vitro

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全文: PDF (258 KB) HTML (0 KB) 输出: BibTeX | EndNote (RIS) 背景资料

**摘要** 目的 探讨鼻咽癌CNE2细胞表面HLA- I 类分子表型和NKG2D配体的表达情况, 进一步了解其对同种异体NK细胞杀伤活性的影响。方法 流式细胞仪检测NKG2D的配体MICA、MICB、ULBP1、ULBP2、ULBP3在K562、CNE2细胞的表达情况。PCR-SSP法分析CNE2细胞HLA-A、B、Cw分型和NK细胞KIR分型。LDH释放法测定5例健康者NK细胞在不同效靶比时对K562、CNE2细胞的杀伤活性, 效靶比20: 1时观察抗NKG2D配体的单抗对NK细胞杀伤K562、CNE2细胞活性的影响。结果 CNE2细胞表达MICA、MICB、ULBP2, 不表达ULBP1、ULBP3。K562细胞表面表达MICA、MICB、ULBP1、ULBP2、ULBP3。5例健康者NK细胞抑制性KIR与CNE2细胞表面的HLA- I 类分子之间存在错配。效靶比5: 1、10: 1、20: 1、30: 1时NK细胞对K562、CNE2细胞的杀伤活性分别为(29.02±0.45)%、(10.50±2.17)%、(44.43±1.36)%、(27.68±1.47)%、(57.82±1.35)%、(36.99±3.13)%、(71.24±2.36)%、(55.00±2.20)%, 在各效靶比时NK细胞对K562细胞的杀伤活性较CNE2细胞明显增强(P=0.000); 在效靶比20: 1时anti-MICA、anti-MICB、anti-ULBP1、anti-ULBP2、anti-ULBP3可明显抑制NK细胞对K562细胞的杀伤活性, 与阻断前相比有显著性差异(P=0.000); anti-MICA、anti-MICB、anti-ULBP2可明显抑制NK细胞对CNE2细胞的杀伤活性, 与阻断前相比有显著性差异(P<0.01), 但anti-ULBP1、anti-ULBP3不能阻断NK细胞对CNE2细胞的杀伤活性。结论 NKG2D配体影响NK细胞对靶细胞的杀伤活性, 提高NKG2D配体的表达有可能提高NK细胞的抗肿瘤活性。

**关键词:** 自然杀伤细胞 NKG2D 杀伤细胞免疫球蛋白样受体

**Abstract:** Objective To analyze HLA-class I molecules and the expression of NKG2D ligands in human nasopharyngeal carcinoma cell line (CNE2) and their effects on cytotoxicity of natural killer (NK) cells. Methods The expression of NKG2D ligands on the surface of CNE2 and K562 cells were analyzed by flow cytometry. The HLA2 class I molecules in CNE2 cells and killer cell immunoglobulin-like receptors (KIR) expressed by NK cells (isolated from 5 healthy persons) were analyzed by PCR-SSP. Cytotoxicities of NK cells against CNE2 and K562 cells were detected by LDH releasing assay at different effect-to-target cell ratios (E:T). In blocking experiments, different anti-NKG2D ligands monoclonal antibodies (mAbs) were added to the target cells at 20:1 E:T ratio. Results It was found that MICA, MICB, ULBP2 were expressed by CNE2, ULBP1, ULBP3 were not detectable on CNE2; K562 expressed all the NKG2D ligands. There were mismatches between inhibitory KIRs expressed by NK cells and HLA-class I molecules expressed by the CNE2 cells. NK cells displayed highly in vitro cytotoxicity against K562 and CNE2 cells with analysis of (29.02±0.45)%, (10.50±2.17)%; (44.43±1.36)%, (27.68±1.47)%; (57.82±1.35)%, (36.99±3.13)%; (71.24±2.36)%, (55.00±2.20)% respectively at 5:1, 10:1, 20:1, 30:1 E:T ratios (P=0.000). Blocking experiments confirmed that killing of K562 by NK cells was efficiently inhibited by anti-MICA mAb, anti-MICB mAb, anti-ULBP1 mAb, anti-ULBP2 mAb and anti-ULBP3 mAb. anti-MICA mAb, anti-MICB mAb, anti-ULBP2 mAb could partially inhibit the cytotoxicity of NK cells against CNE2 cells, whereas anti-ULBP1 mAb and anti-ULBP3 mAb could not inhibit the cytotoxicity of NK cells. Conclusion Expression of NKG2D ligands is correlated with the cytotoxicity of NK cells. NK2 mediated cytolytic activity may be boosted by engineering cells expressing high levels of activating NKG2D ligands.

**Key words:** Natural killer cell NKG2D Killer cell immunoglobulin-like receptor

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