论著

噬菌体肽库与NCI-H1299细胞筛选肺癌特异性结合多肽ZS-9的初步研究

王军业^{1,2},石磊³,戴淑琴^{1,2,4},潘琴³,巫玮³,臧林泉^{3 \triangle},戎铁华^{1,2 \triangle} 1华南肿瘤学国家重点实验室,2中山大学肿瘤防治中心胸科,4检验科,广东 广州 510060; 3 广东药学院药科学院药理教研室,广东 广州 510006

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摘要 目的:利用噬菌体肽库与肺癌NCI-H1299细胞筛选出肺癌特异性结合的多肽,研究其亲和力和特异性。方法:以肺癌细胞NCI-H1299为靶细胞,肺二倍体成纤维细胞MRC-5为吸附细胞,与噬菌体随机十二肽库进行三轮筛选,挑取单克隆扩增并测序,进行生物信息学分析、比对;利用ELISA、细胞免疫化学、组织免疫化学方法测定多肽的亲和力和特异性。结果:经过三轮减性筛选发现,随机挑选的9个单克隆中,其中1个对〖JP〗NCI-H1299和A549均具有较高亲和力,将其命名为ZS-9,测序结果为CAT AAT AAG CAT CTT CCG TCT ACG CAG CCT CTT GCG,根据测序结果推导出ZS-9的氨基酸序列HNKHLPSTQPLA,生物信息学分析表明ZS-9的氨基酸序列与美国国立生物技术信息中心(NCBI) GenBank DNA序列数据库和Swiss-Prot蛋白数据库中的已知基因和蛋白无同源性,表明我们筛选到一种新的肺癌相关抗原的配体。结论:利用噬菌体随机十二肽成功筛选出与肺癌细胞NCI-H1299和A549具有较高亲和力的多肽ZS-9,为肺癌的早期诊断和靶向治疗奠定基础。

关键词 <u>噬菌体肽库</u>; <u>肺肿瘤</u>; <u>NCI-H1299细胞</u>; <u>多肽ZS-9</u> 分类号 Q7, R73, R9

Screening of lung cancer specific peptide ZS-9 from a phage display peptide library and NCI-H1299 cells

WANG Jun-ye^{1, 2}, SHI Lei³, DAI Shu-qin^{1,4}, PAN Qin³, WU Wei³, ZANG Lin-quan³,RONG Tie-hua^{1, 2}

1Skate Key Laboratory of Oncology in South China, Guangzhou 510060, China; 2Department of Thoracic Surgery, Cancer Center, SUN Yat-sen University, Guangzhou 510060, China; 3Department of Pharmacology, Guangdong Pharmaceutical University, Guangzhou 510006, China; 4Department of Laboratory Medicine, Cancer Center, SUN Yat-sen University, Guangzhou 510060, China. E-mail: zanglq@yahoo.com; rongth@sysucc.com

Abstract

=AIM: To obtain the polypeptides specifically bound to NCI-H1299 cell line from peptide libraries and to identify these polypeptides affinity and specific to lung cancer.
METHODS: The lung cancer NCI-H1299 cell line was used as the antigen and MRC-5 was used as control for subtraction biopanning from a phage display peptide library. The positive and specific binding clones were identified by cell enzyme-linked immunosorbent assay (ELISA) and immunochemistry staining. Those DNA sequences of identified clones were sequenced, and the amino acid sequence was deduced and analyzed with bioinformatics.
RESULTS: After 3 rounds of panning, 9 phage clones were identified by ELISA, one of them specially bound to the NCI-H1299 and A549. The DNA sequence result of ZS-9 was CAT AAT AAG CAT CTT CCG TCT ACG CAG CCT CTT GCG. Hence, the polypeptide sequence was HNKHLPSTQPLA deduced from its DNA sequence. The amino acid sequence was analyzed in NCBI and Swiss-Prot, the results showed that it has no similarity with the known proteins sequence in the database. All these results showed that we have discovered a novel lung cancer surface associated antigen ligand.
CONCLUSION: A peptide which is specific binding to lung cancer cell line NCI-H1299 and A549 has been selected from phage display peptide libraries. Therefore, it provides a potential tool for early diagnosis of lung cancer or targeted drug delivery in chemotherapy.

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肺肿瘤; NCI-H1299细胞; 多肽Z

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Key words Phage library Lung neoplasms NCI-H1299 cells Peptides ZS-9

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通讯作者 臧林泉;戎铁华 rongth@sysucc.com