Chinese Journal of Chemistry 2005 23 (9): 1247-1252 ISSN: 0567-7351 CN: 31-1320/06

FULL PAPERS

聚四氟乙烯片基上寡核苷酸原位的合成及金标银染检测

谭美军[1]，汤建新*，[a]，陈洪*[2]

[1]湖南工业大学湖南省绿色包装和生物纳米技术重点实验室, 株洲 412008
[2]中南林学院林业部经济林育种与栽培重点实验室, 株洲 412006

收稿日期 2005-2-24 修回日期 2005-6-27 网络版发布日期 接受日期

摘要 采用H_2/N_2混合气体等离子体处理了聚四氟乙烯片基，XPS光电子能谱证实在其表面接枝了大量氨基，并直接用于DNA原位合成和金标银染进行检测。研究结果表明：等离子体处理的聚四氟乙烯片基可直接用于DNA原位合成和金标银染进行检测，能明显识别正配和错配的碱基序列，其检测灵敏度可达10 pmol/L。正配与错配1、2、3个碱基的灰度之比为：72:44.4:22.5:11.4。论文还将聚四氟乙烯片基上先进行寡核苷酸原位合成和点样试验，然后再进行金标银染检测。结果发现前者的检测灵敏度比后者明显高一个数量级，且其信号强度是后者的1.8倍。上述结果表明：等离子体处理的聚四氟乙烯片基可发展为高密度基因芯片新片基。

关键词 寡核苷酸,原位合成,金标银染,等离子体,聚四乙烯

分类号

In situ Synthesis of Oligonucleotide and Detection of Gold-label-silver-stain on PTFE Slices

TAN Mei-Jun[1], TANG Jian-Xin*,[1], CHEN Hong*,[2]

[1] Key Laboratory of Green Packaging and Biological Nanotechnology of Hunan Province, Hunan Polytechnic University, Zhuzhou, Hunan 412008, China
[2] Key Laboratory of Non-timber Breeding and Cultivation of Ministry of Forestry of China, Central South Forestry University, Zhuzhou, Hunan 412006, China

Abstract Poly(tetrafluoroethylene) (PTFE) was treated with plasma in a mixture of nitrogen and hydrogen (1∶ 2 in volume). X-ray photoelectron spectroscopy (XPS) demonstrated the success of amino group grafting. The as-treated PTFE slices were successively applied to the *in situ* synthesis of oligonucleotides. With the detection of gold-label-silver-stain, the hybridization signals were recorded with a gel document and analysis system. A target DNA concentration as low as 10 pmol/L could be detected. The complementary and mismatched sequences were distinguished clearly, and the ratio of background-subtracted gray scale values for perfect match∶ 1 base mismatch∶ 2 base mismatch∶ 3 base mismatch was 72∶ 44.4∶ 22.5∶ 11.4. The sensitivity of *in situ* synthesis system was 1 order of magnitude higher than that of spotting system, and the signal of the former was about 1.5 times stronger than that of the latter under the same target DNA concentration. These plasma modified PTFE slices might open novel prospects for the *in situ* synthesis of DNA micro-arrays.

Key words oligonucleotide gold-label-silver-stain *in situ* synthesis plasma poly(tetrafluoroethylene)

DOI:

通讯作者 汤建新 jxtang0733@163.com; chenhongcs@126.com