

The collaboration research for the Dual Graduate School between VNU and JAIST

[Title of collaboration research]: Development of devices for genetic analysis by using porous glass.

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[Reference home-page address]: <http://www.jaist.ac.jp/nmcenter/labs/tsukahara-www/>

[Other references]: T. Tsukahara and H. Nagasawa: Probe-on-carriers for Oligonucleotide microarrays (DNA Chips). *Sci Techno Adv Materials* (2004) **5**, 359-362.

S. Noguchi, T. Tsukahara, M. Fujita, R. Kurokawa, M. Tachikawa, T. Toda, A. Tsujimoto, K. Arahata, I. Nishino: cDNA microarray analysis of individual Duchenne muscular dystrophy patients. *Hum.Mol.Genet.* (2003) **12**, 595-600.

[Contents] To realize tailor-made medicine, cheap and accurate analytical methods for SNPs are indispensable. In accuracy, oligo-DNA microarrays are considerably behind the invader method, because most of in situ synthetic probes on arrays do not have enough fidelity. Moreover, arrays are too expensive for personal use although the comprehensive SNPs analysis can be accomplished. Therefore, a cost-effective method to manufacture arrays with precise oligonucleotides is necessary.

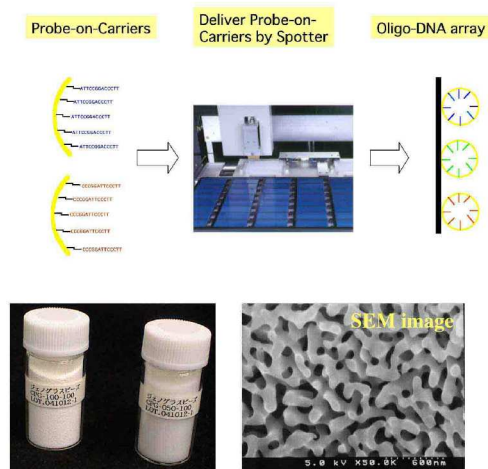
In this research, we are proposing "probe-on-carriers", immobilized oligonucleotide probes by using porous glasses on the solid phase, to make DNA analytical devices. In this method, each oligonucleotide is synthesized on porous glass as a solid phase-carrier and can be used as a probe for the complementary target sequence. Because probes are synthesized in the same manner as that used for the routine oligonucleotide synthesis, they show enough fidelity.

Probe-on-carriers carrying wild-type or mutated sequences of *p53* gene were synthesized on the porous glass with 200 nm in pore diameter. Then a suspended solution of probe-on-carriers was spotted on a slide glass with a pre-spin-coated polybutadiene adhesive to make a DNA microarray. Each probe-on-carrier was hybridized with a fluorescent-labeled perfect-match DNA fragment but not with a mismatch one. This result clearly demonstrated that

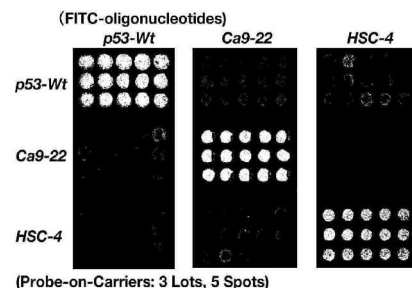
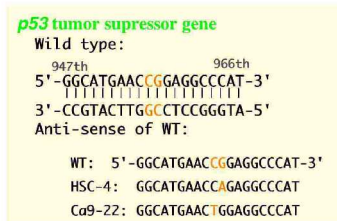
fixed probe-on-carriers acted as probes to detect one nucleotide substitution and may be used to manufacture DNA microarrays. This method can be used to construct accurate oligo-DNA arrays with 1,000 SNPs at a cost of less than 10 US dollar per an array.

Development of New Devices for SNPs Analysis

Schematic Representation for New DNA Chip



The highly controlled porous glass, "genoglass", was developed as a solid phase-carrier to synthesize probes for DNA chip. (Collaboration with Genoglass and Ebara Corp.)



Each probe-on-carrier was hybridized with the fluorescent-labeled perfect-match probes but not with mismatch probe. This result clearly demonstrated that fixed probe-on-carriers acted as probes to detect SNPs.