A New Technology for Simultaneous Preservation of Biomolecules and Morphology in Tissues Facilitates Biomarker Development

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Impact of ischemia time and stabilization method analysed with Affymetix Human Genome U219 Array on the GeneTitan hybridization, wash and scanning station. Liver needle biopsies from 5 patients (P1-P5), exposed to different ischemia time points (T0-3), were either snap-frozen (Cryo) or fixed and stabilised with PAXgene Tissue (PAX). (a) RNA quality control (b) GDM analysis.

Results

Morphology and Antigenicity

PFPE samples show well-preserved morphology and antigenicity, comparable to FFPE samples. For some antibodies less harsh retrieval procedures can be used. (a) H&E staining of human stomach tissue. (b) CD 20 staining of human spleen tissue (c) Ki-67 staining of human liver.

Nucleic Acids

High correlation of gene signature in PFPE and cryo-preserved samples (R²=0.99), but decreased correlation (R²=0.99) and major gene-to-gene variations in FFPE samples.

DNA integrity and performance in multiplex PCR of PFPE samples. (a) High molecular mass bands indicating good DNA integrity were visible for FFPE and cryo-preserved samples but not for PFPE samples. Genomic DNA extracted from corresponding FFPE, PFPE, and snap-frozen (Cryo) samples of 5 human breast cancer cases was separated on 1% agarose gels and visualized with ethidium bromide. (b) Multiplex PCR of eight fragments of different human genes. PFRF (222 bp), C0706 (314 bp), X07 (144 bp), AGTR1 (833 bp), C014 (1652 bp), C930 (765 bp), C080 (943 bp) and C2019 (951 bp).

Conclusions

New Tissue Stabilisation Technology PAXgene Tissue provides

- Preservation of morphology and antigenicity resembling FFPE samples.
- Excellent RNA quality and strong correlation of multiple mRNA profiles with snap frozen samples and the phosphorylation levels are well-preserved.
- PFPE samples are comparable to cryo-preserved samples in reverse-phase protein microarray (RPMA) experiments.

Innovative Aspects

- Simultaneous high-quality preservation of biomolecules and morphology in clinical tissue samples.
- Direct correlation of morphological disease phenotypes with alterations of biomolecules.
- Multimodal biomarker studies in a routine clinical setting.
- Molecular analyses of lesions where a collection of snap-frozen material is impossible.

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