GENE EXPRESSION PROFILING OF TRANSPLANT FFPE BIOPSIES: COMPARISON OF A CUSTOM TAQMAN® LOW DENSITY ARRAY AND QUANTITATIVE NUCLEASE-PROTECTION ASSAY

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Introduction

We previously developed a custom Taqman® Low Density Array (TLDA) containing 47 markers to monitor changes of gene expression in FFPE renal and small bowel allografts. In this work we carried out a parallel comparison of expression profiles of marker genes obtained with TaqMan® technology and with the automated quantitative nuclease-protection assay (qNPA) system developed by HTG Molecular, Inc.

Materials and Methods

Kidney, small bowel (SB) and heart transplant FFPE biopsies with different levels of rejection were selected for the study. 5 to 6 curls of 10µm were cut from each FFPE biopsy for analysis in a Custom TLDA developed in University of Miami Transplant Laboratories. This custom array contains 47 markers to monitor kidney and small bowel cellular rejection, and includes immune, inflammatory and apoptosis genes. We used the HTG Edge System with the Immuno-Oncology Expression assay (HTG-IOE) containing 26 markers, or the HTG EdgeSeq Oncology Biomarker assay (HTG-ESOB) containing 2558 genes formatted for NGS. In all cases expression data was presented as differential expression relative to Normal Donor (ND) or No Rejection (NR).

Results

Similar Expression Profiles in Kidney FFPE Biopsies of Markers Analyzed by TLDA and HTG-IOE Arrays

These are examples of a total of 10 markers

Markers in HTG-IOE Array with Potential Value for Expression Profiles of Kidney FFPE Biopsies

These are examples of a total of 15 markers

Expression Profile in Small Bowel FFPE Biopsies analyzed by HTG-ESOB

Conclusions

• This study showed that expression analysis carried out with TLDA and qNPA are comparable.
• The qNPA technology developed by HTG Molecular Diagnostics, Inc. is a good alternative to evaluate expression of solid organ transplant markers in FFPE biopsies.
• We found that the HTG-IOE (96-well format) could be an excellent automated tool to evaluate custom panels with reduced number of markers in a clinical setting.
• The HTG-ESOB (HTG EdgeSeq) allows evaluation of a large number of genes, but it includes an NGS step that makes it expensive and time consuming.
• The HTG-ESOB (HTG EdgeSeq) is a good exploratory tool for searching relevant markers.

References