P#ST37

Use of Intra-Gross Examination Touch Prep Slides to Facilitate Turnaround Time (TAT) Advantage Using the Biocartis Idylla

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Introduction

As the role of molecular biomarkers for clinical management of cancer patients expands, the demand for rapid molecular results is increasing. We sought to reduce TATs for specific actionable genetic alterations by utilizing touch preparation (TP) samples as substrate material for the Biocartis Idylla system, a fully automated molecular assay platform.

Methods

Diff-Quik (DQ) stained TPs were prospectively collected from non-small cell lung carcinoma (NSCLC) and colorectal adenocarcinoma (CRC) resection specimens at the time of gross examination. If tumor cells were present microscopically, they were tested using the Biocartis Idylla system for *EGFR*, *KRAS*, *BRAF*, *NRAS* and microsatellite instability (MSI) depending on tumor type. The accuracy of this approach was evaluated with subsequent corresponding formalin fixed paraffin embedded (FFPE) material.

Results

- 16 CRCs and 13 NSCLCs were tested.
- Among CRCs, 14, 13, 6 and 13 had valid *KRAS*, *NRAS*, *BRAF* and MSI results, respectively.
- 4 *KRAS* mutations and 1 *BRAF* mutation were identified in CRCs (Table 1).
- 2/5 MSI-High cases demonstrated discordant results (C5 and C12, Table 3), called MSI-H by Idylla but microsatellite stable by confirmatory testing.
- Among NSCLCs, 13, 12 and 13 had valid *EGFR*, *KRAS* and *BRAF* results, respectively.
- 2 *EGFR* mutations and 2 *KRAS* mutations were identified in NSCLCs (Table 2).
- All calls with valid mutational results were concordant with orthogonal analysis except one NSCLC case (L10) in which both a false positive (FP) *EGFR* and false negative (FN) *BRAF* mutation was evident on Idylla.
- Further evaluation of discordant cases was performed by testing unstained slides, which resulted in correct findings (Example case presented in Figures 1-3).
- Select cases with invalid results were re-run on the Idylla utilizing FFPE material or unstained TPs and subsequently yielded valid results.
- A subset of samples (n=22) tested with rapid intent were used for analysis of TATs. 17/22 had partial results, defined as at least one result, or complete results available on the same day as the pathology report release (Table 4).

Discussion

The aim of our study was to investigate the use of TPs on the Idylla system to reduce TATs such that results would be available with routine surgical pathology reports. Our approach of collecting TP samples directly from tissue and testing them via the Idylla system significantly reduced potential TATs. Samples processed with rapid intent often provided results the same day as or before the pathology report release. Our study suggests that DQ stain may cause interference, leading to invalid and FP results, thus use of a similar approach to accelerate TAT would require alternate methods to confirm tumor presence.

References

1. De Luca C, Conticelli F, Leone A et al. Is the Idylla EGFR Mutation Assay feasible on archival stained cytological smears? A pilot study. J Clin Pathol. 2019;72(9):609-614.
2. De Luca C, Gragnano G, Pisapia P et al. EGFR mutation detection on lung cancer cytological specimens by the novel fully automated PCR-based Idylla EGFR Mutation Assay. J Clin Pathol. 2017;70(4):295-300.

Table 1 Mutation Concordance for Colorectal Adenocarcinoma Cases

Case	Idylla *KRAS*	Idylla *NRAS/BRAF/EGFR*	NGS or direct sequencing	Concordance
C1	G12A	WT	*KRAS* G12A	Concordant
C2	G12V	WT	*KRAS* G12V	Concordant
C3	WT	*NRAS/EGFR* – WT *BRAF* - Invalid	No mutations detected	Concordant/partial invalid
C4	WT	WT	No mutations detected	Concordant
C5	WT	*NRAS/EGFR* – WT *BRAF* - Invalid	No mutations detected	Concordant/partial invalid
C6	WT	*NRAS/EGFR* – WT *BRAF* – V600E	*BRAF* V600E	Concordant
C7	G12A	*NRAS/BRAF*-Invalid *EGFR* - WT	*KRAS* G12A	Concordant/partial invalid
C8	WT	*NRAS/EGFR* – WT *BRAF* - Invalid	*KRAS* N116H	Concordant/partial invalid*
C9	WT	*NRAS/EGFR* – WT *BRAF* – Invalid	No mutations detected	Concordant/partial invalid
C10	G12D	WT	*KRAS* G12D	Concordant
C11	WT	*NRAS/EGFR* – WT *BRAF* - Invalid	No mutations detected	Concordant/partial invalid
C12	WT	WT	*KRAS* G10dup	Concordant*
C13	WT	*NRAS/EGFR* – WT *BRAF* - Invalid	*BRAF* V600E	Concordant/partial invalid
C14	WT	*NRAS/EGFR* – WT *BRAF* - Invalid	No mutations detected	Concordant/partial invalid
C15	Invalid	Invalid	*KRAS* G13D	Invalid
C16	Invalid	Invalid	No mutations detected	Invalid

Table 2 Mutation Concordance for Lung Adenocarcinoma Cases

Case	Idylla *EGFR*	Idylla *KRAS*	Idylla *BRAF*	NGS or direct sequencing	Concordance
L1	Exon 19 deletion	WT	WT	*EGFR* DEL746_750	Concordant
L2	WT	G12C	WT	*KRAS* G12C	Concordant
L3	WT	WT	WT	*KRAS* G13C *BRAF* G466L	Concordant*
L4	WT	Invalid	WT	No mutations	Concordant/invalid
L5	WT	WT	WT	No mutations	Concordant
L6	WT	WT	WT	No mutations	Concordant
L7	WT	WT	WT	*EGFR* A767_V769dup	Concordant*
L8	WT	WT	WT	No mutations	Concordant
L9	WT	WT	WT	No mutations	Concordant
L10	L861Q	WT	WT	*BRAF* V600E	Discordant
L11	WT	G12V	WT	*KRAS* G12V	Concordant
L12	WT	WT	WT	No mutations	Concordant
L13	WT	WT	WT	No mutations	Concordant

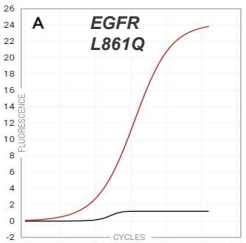

Figure 1: Case L10, Diff-Quik TP slide, chamber A, *EGFR* total CQ = 21.82, dCQ = 3.26

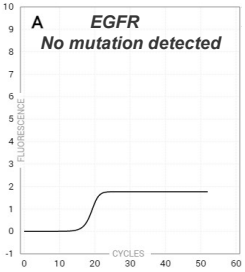

Figure 2: Case L10, Unstained TP slide, chamber A, CQ = 17.83

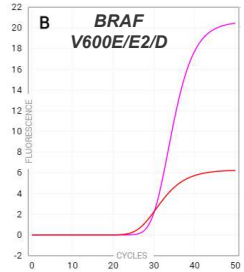

Figure 3: Case L10, Unstained TP slide, chamber B, SPC CQ = 28.70, dCQ = 2.14

Table 3 MSI Concordance for Colorectal Adenocarcinoma Cases

Case	MSI Idylla	MSI	Concordance
C1	MSS	MSS	Concordant
C2	MSS	MSS	Concordant
C3	MSI-H	MSI-H	Concordant
C4	MSS	MSS	Concordant
C5	MSI-H	MSS	Discordant
C6	MSI-H	MSI-H	Concordant
C7	MSS	MSS	Concordant
C8	MSS	MSS	Concordant
C9	MSS	MSS	Concordant
C10	MSI-H	MSI-H	Concordant
C11	Invalid	MSS	Invalid
C12	MSI-H	MSS	Discordant
C13	Invalid	MSI-H	Invalid
C14	MSS	MSS	Concordant
C15	Invalid	MSS	Invalid
C16	MSS	MSS	Concordant

Table 4 Rapid Idylla Result Turnaround Availability

Case	Idylla Results Available	Days compared to final pathology sign out date
L1	Partial results	7 days before
L2	Partial results	37 days after
L3	Complete results	2 days before
L4	Complete results	Same day
L5	Complete results	1 day after
L6	Complete results	11 days after
L7	Complete results	1 day before
C1	Partial results	7 days before
C2	Partial results	8 days before
C3	Partial results	5 days before
C4	Complete results	6 days before
C5	Complete results	4 days before
C6	Complete results	Same day
C7	Complete results	1 day before
C8	Complete results	4 days before
C9	Complete results	4 days after
C10	Complete results	4 days before
C11	Complete results	11 days after
C12	Complete results	Same day
C13	Complete results	Same day
C14	Partial results	10 days before

Tables 1-4 Legend: WT = wild-type, MSS= microsatellite stable, MSI-H = microsatellite instability high, N/A = none applicable, * = mutation detected by orthogonal assay is not evaluated by Idylla