IGT® UMI Adapter & UDI Primer (for Illumina) —

Molecular tags for Illumina platform, facilitating low-frequency mutation detection in ctDNA

Circulating tumor DNA (ctDNA) refers to extracellular DNA present in body fluids such as plasma, urine, and cerebrospinal fluid, measuring approximately 70~200 bp in length, it primarily originates from necrotic or apoptotic tumor cells, exosomes, and circulating tumor cells. While ctDNA analysis effectively addresses challenges in frequent tissue sampling and tumor heterogeneity, most ctDNA samples exhibit low detection rates, with approximately 10%~50% showing matching endogenous tags.

To address these technical challenges, based on self-developed TargetSeq One® hybridization capture system, by integrating paired-end Universal Molecular Identifiers(UMI), iGeneTech can realize PCR and random sequencing error correction, significantly enhancing detection sensitivity and specificity in liquid biopsy applications. This innovative solution demonstrates broad applicability across multiple clinical fields, including early cancer diagnosis and staging, therapeutic efficacy evaluation, recurrence monitoring, and prognosis prediction in oncology.

Product Overview

The IGT® UMI Adapter & UDI Primer (for Illumina) comprises a paired-end UMI adapter mix and UDI primers, specifically designed for TruSeq library prep on Illumina. The paired-end UMI adapter mix contains 64 fixed UMI adapters of 6 bp length, generating 4,096 unique combinations that ensure sufficient diversity to distinguish individual DNA molecules. With an edit distance of ≥ 3 between UMI sequences, this design enhances accuracy in correcting errors during library prep, target region capture and sequencing processes of DNA molecules. Additionally, the product line offers 384 UDI sequences for customer selection, effectively meeting the polling requirements for ultra-high data yields per lane in high-throughput sequencing platforms.

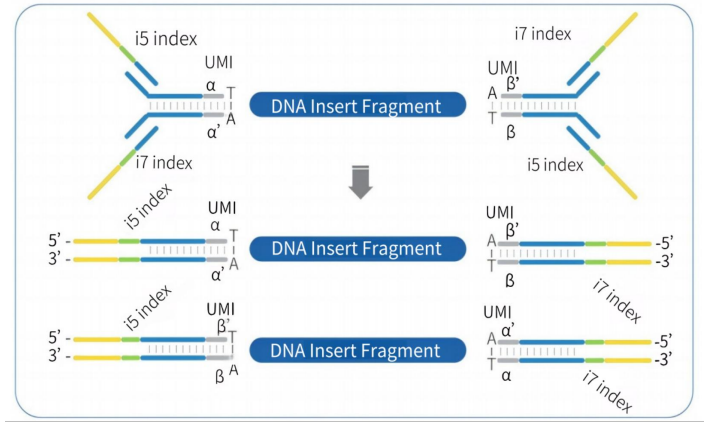

Figure1. IGT® UMI Adapter & UDI Primer (for Illumina) Library Prep Process

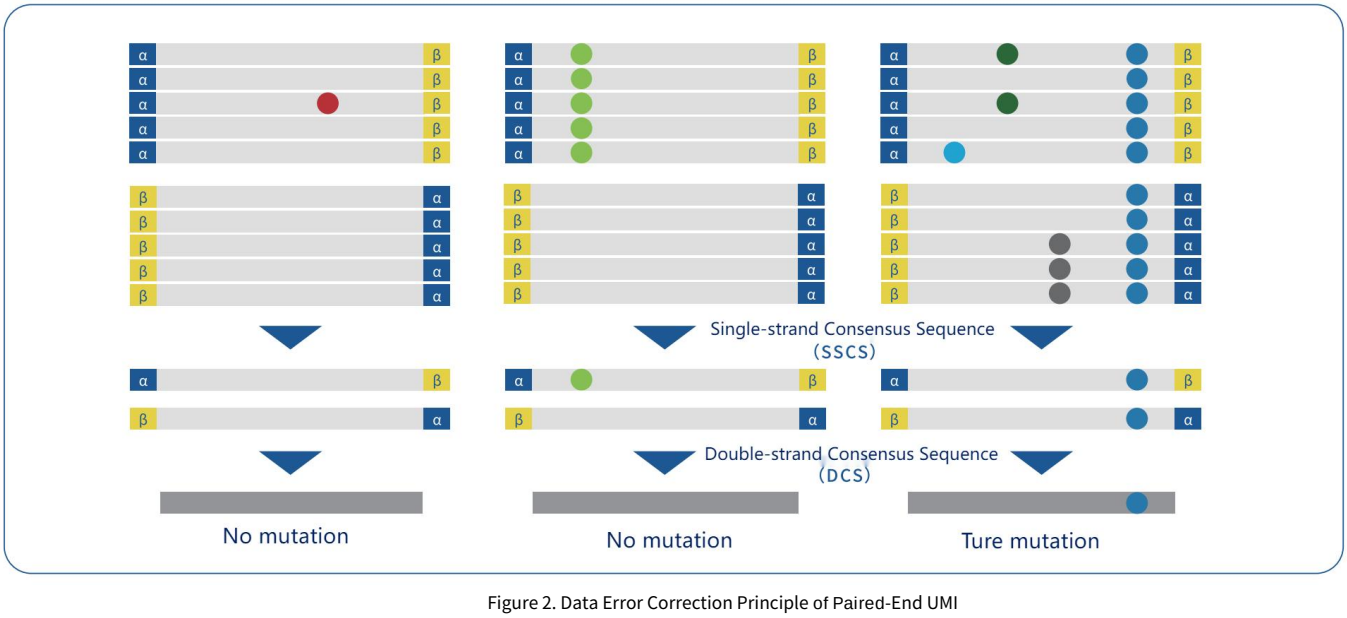

Figure 2. Data Error Correction Principle of Paired-End UMI

Product advantages

1 High adapter connection efficiency
Optimize the adapter design and synthesis to ensure high library conversion efficiency and high sensitivity detection of low frequency variation.

2 Assist to achieve accurate detection of low frequency variation
Stable detection of abundance as low as 0.02% of ctDNA. 99.9% of background noise can be filtered out by paired-end UMI correction.

3 Unique UMI sequence design
The design of UMI sequences was not completely random (edit distance ≥ 3) to avoid false positive variation caused by UMI sequence error.

4 Contains UDI amplification primers
It contains a paired-end Unique Dual Index (UDI) to avoid cross-contamination to the greatest extent.

Detection Cases

● Input 25 ng of 0.1% Horizon cfDNA standard (Horizon Multiplex I cfDNA, catalog # HD780) for library prep, capture with a 52 kb panel, followed by sequencing on the NovaSeq 6000 platform using PE150. Under conditions of 20,000× average sequencing depth in the target region before redundancy removal, all 8 known mutation sites were detected with 100% sensitivity (Table1).

Table 1. Sequencing Data of Horizon 0.1% cfDNA Reference Samples

EGFR L858R	*EGFR* T790M	*EGFR* delE746-A750	*EGFR V769* -D770insASV	*KRAS* G12D	*NRAS* Q61K	*NRAS* Q61K	*NRAS* A59T	*PIK3CA* E545K	Sensitivity
0.08%	0.10%	0.09%	0.06%	0.10%	0.10%	0.12%	0.16%	0.21%	100%

●The implementation of molecular labeling technology in low-frequency ctDNA detection significantly reduces false positive site identification number. After standard deduplication, 526 mutations with VAF below 3% were detected in the Horizon standard panel. When combining UMI with genome coordinate alignment for single-stranded consensus sequence (SSCS deduplication & correction) , 163 mutations with VAF < 3% were identified. In contrast, using UMI for cfDNA molecular positive/negative strand error correction(DCS)combination, only 15 low-frequency mutations with VAF < 3 % were detected, including 8 known mutation sites. The paired-end UMI technology demonstrated a 97% reduction in low-frequency allele detection rates compared to standard deduplication methods (Figure 3).

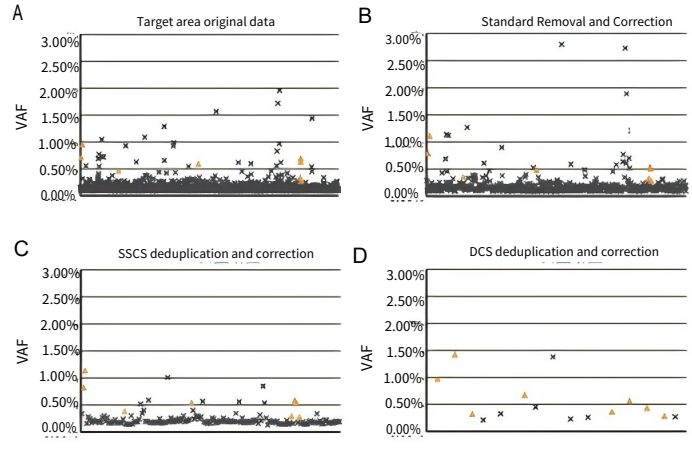

Figure3. IGT® UMI Adapter & UDI Primer (for Illumina) can significantly reduce background noise

Product Information

Product name	Specifications	Catalog #
IGT® UMI Adapter & UDI Primer 1-16 (for Illumina, tube)	16*1 rxn	C10131
IGT® UMI Adapter & UDI Primer 1-96 (forIllumina, plate)	96*1rxn	C10092
IGT® UMI Adapter & UDI Primer 97-192 (for llumina, plate)	96*1rxn	C10102
IGT® UMI Adapter & UDI Primer 193-288 (for Illumina, plate)	96*1rxn	C10112
IGT® UMI Adapter & UDI Primer 289-384 (for llumina, plate)	96*1rxn	C10122

*For more adapter reagents, please consult iGeneTech customer manager.

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