SmartChip™ TE

FLEXIBLE DESIGNS, SUPERIOR COVERAGE

Fast NGS Target Enrichment workflow for unparalleled sample uniformity and coverage

COVERAGE - Massively parallel single-plex reactions for greater coverage

EFFICIENT - Library prep and target enrichment in a single step with a less than 4 hour workflow

DYNAMIC - Add or remove assays without affecting the entire panel

Targeted sequencing maximizes clinical researchers' time and minimizes sequencing costs, while avoiding the complication of whole genome sequencing. Current multiplex and hybrid capture methods allow the targeting of specific regions, but these methods can be cumbersome, expensive, and can suffer from low coverage, and off-target reads.

The SmartChip TE, based on massive parallel single-plex PCR, resolves these issues and provides panels with high coverage, no off-target reads, and no gaps that require costly Sanger backfilling. Combined with a simple one step sequencing ready workflow (Figure 1), SmartChip TE gives you the easiest and most complete enrichment method available (Figure 2).

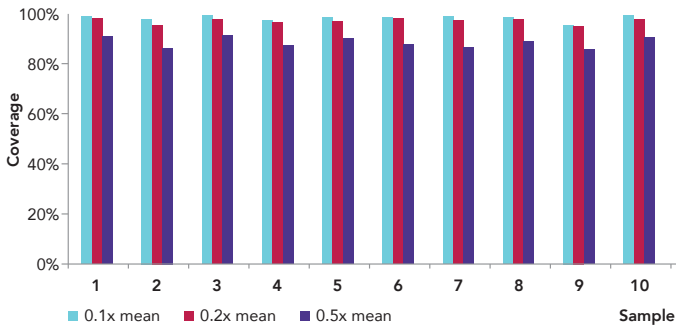

FIGURE 2. Uniformity at 0.1x, 0.2x and 0.5x of the mean coverage is shown for a 140kb target region. Data is plotted for 10 samples on a single MiSeq run. For all samples, >95% of bases are covered at 0.1x and 0.2x of the mean coverage (blue and red bars, respectively) and >85% of bases are covered at 0.5x of the mean coverage (purple bars).

TABLE 1. SMARTCHIP MULTISAMPLE NANODISPENSER

CUSTOM DNA PANELS CONFIGURATIONS

Max. Assay #	Samples	Max. Assay #	Samples
48	96	**432**	12
72	72	**648**	8
96	54	**864**	6
108	48	**1296**	4
216	24	**2592**	2
288	18	**5184**	1
360	14	—	—

MultiSample NanoDispenser permits a highly flexible assay configuration based on experimental requirements.

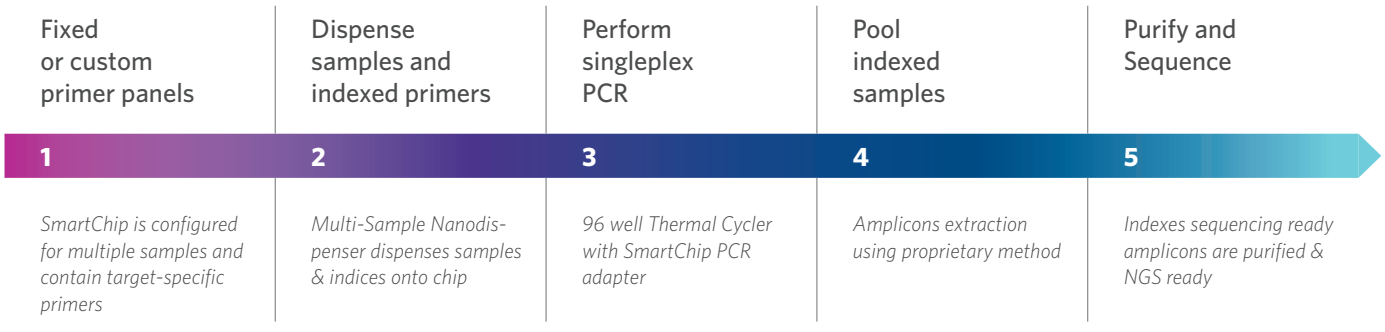

FIGURE 1. Seq-Ready TE MultiSample Workflow - One-step target enrichment and library prep

Dynamic panels with consistent results

Customizing multiplex PCR based designs can be frustrating. Adjusting amplicon pools to prevent unwanted interactions creates many panel iterations, and still leads to a design with gaps and low sample-to-sample reproducibility. With Seq-Ready TE MultiSample Custom DNA panels using SmartChip single-plex technology, you can quickly design custom targeted panels with high coverage and uniformity. SmartChip's 5,184 individual wells provide flexible assay configurations and samples optimized for any target you choose (Table 1). Unlike multiplex methods, design iteration is quick and simple; targets can be added and removed without disrupting the gap-free panel you already have.

Designed panels not only offer high coverage and uniformity with no gaps, but also provide high sample-to-sample reproducibility. Unlike multiplex panels, which can suffer from primer competition causing variable gaps to occur, Seq-Ready panels achieve coverage uniformity every run (Figure 2). This is all accomplished without expensive and time consuming post-enrichment and normalization.

Efficient workflow with precision delivery

Target enrichment workflows based on multiplex PCR or hybrid capture are long, difficult, and labor intensive, making them a challenge to integrate into clinical research labs. The Seq-Ready TE ligation-free workflow simultaneously enriches target regions and incorporates sequencing adapters and barcodes to produce sequencing ready libraries in one simple step (Figure 3). Utilizing the MultiSample NanoDispenser an operator can complete the streamlined workflow in under 4 hours, with under 45 minutes of hands-on time.

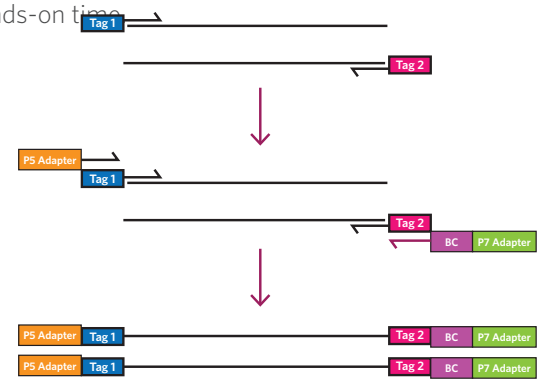

FIGURE 3: Nested primer sets simultaneously enrich a large number of target regions and incorporate sequencing adapters and indices to produce sequencing ready libraries in one step.

TABLE 2. SEQ-READY BRCA1/2 PANEL

COVERAGE ANALYSIS SUMMARY & SPECIFICATIONS	METRIC
Bases in target regions	19,092
Percent aligned base reads	>97%
Mean coverage depth [N = 24]	~ 5,000
Uniformity of base coverage [> 0.05 of mean]	99.3%
Uniformity of amplicon coverage [>0.05 of mean]	99.8%
Target base coverage at 1x	100%
Target base coverage at 30x	100%
Target base coverage at 100x	99.7%

The Seq-Ready BRCA1/2 panel provides SmartChip TE's high level of uniformity and coverage in an off the shelf design

Off the shelf BRCA1/2 Panel

The Seq-Ready TE MultiSample BRCA1/2 Panel contains 139 unique PCR primer pairs that target 100% of the coding regions of BRCA1 and BRCA2 tumor suppressor genes. By leveraging massively parallel single-plex chemistry, we are able to provide high coverage and uniformity across a 19Kb region of interest with inputs as low as 60ng for up to 24 samples (Table 2). As with our custom panels, the BRCA1/2 panel is a one step enrichment and library preparation ligation-free workflow that requires no post-enrichment, or sample-to-sample normalization.

The Seq-Ready TE MultiSample BRCA1/2 panel is designed with primers that have been optimized for 100% coverage of all targeted coding exons and exon-intron boundaries of BRCA1 and BRCA2 genes. Amplicons span at least 45 bp beyond the targeted coding exon and exon-intron boundaries to ensure that the regions of interest are sequenced. Amplicons overlap to cut down on false negatives due to rare variants and polymorphisms. To reduce the possibility of dropouts, no SNPs with frequency under 0.5% are included in the last 10 nucleotides of the 3' end of primers. Primer redundancy allows for maximum coverage in SNP dense regions.

For Research Use Only. Not for use in diagnostic procedures.

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