Performance of Exome Sequencing Panels: From Targeted Screening to Comprehensive Coverage

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Exome sequencing panels are critical tools in genomics, enabling the detection of genetic variations associated with various diseases, especially cancer. The Twist Exome 2.0 Panel provides a comprehensive approach to whole exome sequencing, covering 36.5 Mb of the human genome and offering a broader perspective for the diagnosis of inherited diseases. In contrast, the KAPA HyperPETE Newborn Screening Panel (Roche) is a targeted panel designed for the screening of genetic disorders in newborns, offering high sensitivity for small, well-defined regions.

Presented comparative study highlights the advantage of Twist Exome 2.0 solution Twist Exome 2.0 is engineered to identify rare and inherited diseases, as well as germline cancers.

Its exceptional uniformity and minimal off-target rate ensure top-tier sequencing efficiency, allowing high-quality data collection with reduced sequencing efforts.

Offering comprehensive coverage of key genetic databases—including RefSeq, CCDS, GenCode, ClinVar, ACMG73, and more—this panel also incorporates clinically significant non-coding pathogenic and likely pathogenic variants.

Twist Exome 2.0 delivers the benefits of multiple clinical panels in one customizable solution, streamlining your diagnostic and research needs.

Newborn Screening Panel

- KAPA HyperPETE Newborn Screening Panel (size 294 kb)

Table 1. The selected QC metrics of WES NGS plex

Sample ID	Qubit Results		Bioanalyzer 2100 Results
	c [ng/µl]	c [nM]	Average Fragment Length [bp]
Plex 1	2,2	10,2	321

Newborn Screening Panel

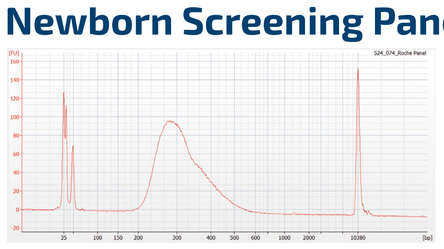
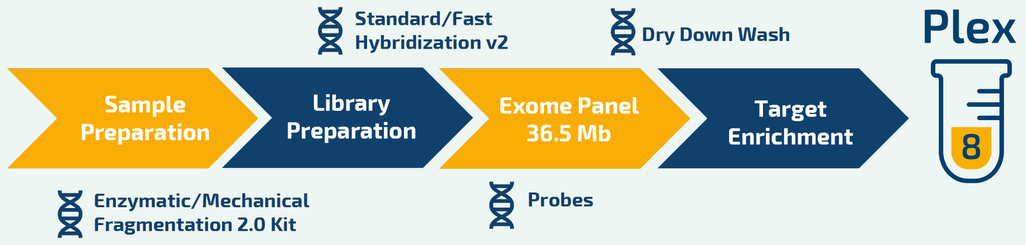


Figure 1. Electrophoretogram of the plex containing NGS libraries for WES with the KAPA HyperPETE Newborn Screening Panel.

Twist Exome Panel



Panel Size:

- Twist Exome 2.0 panel (size 36,5 Mb)

Plex QC:

Qubit
- Plex with 8 samples
- Concentration of the plex was 19,8 ng/µl; 93,2nM

Bioanalyzer 2100:
- Bioanalyzer 2100: Average fragment length was 322 bp (Table 2)
- No adapter dimers were detected (Figure 2)

Table 2. The selected QC metrics of WES NGS plex

Sample ID	Qubit Results		Bioanalyzer 2100 Results	
	c [ng/µl]	c [nM]	Average Fragment Length [bp]	Dimers [%]
Plex 2	19,8	93,2	322	0

Twist panel

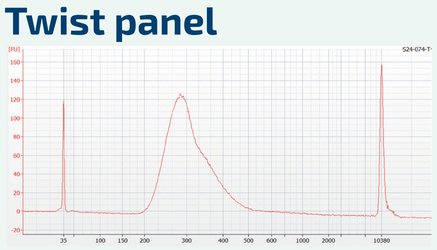


Figure 2. Electrophoretogram of the plex containing NGS libraries for WES with the Twist Exome 2.0 panel.

Sequencing and Bioinformatic:

Sequencing
- All plexes were successfully sequenced on NovaSeq X Plus
- The sequencing chemistry used: 10 B chemistry

- Plex 1 with the KAPA HyperPETE Newborn Screening Panel achieved the required 4 million reads per plex

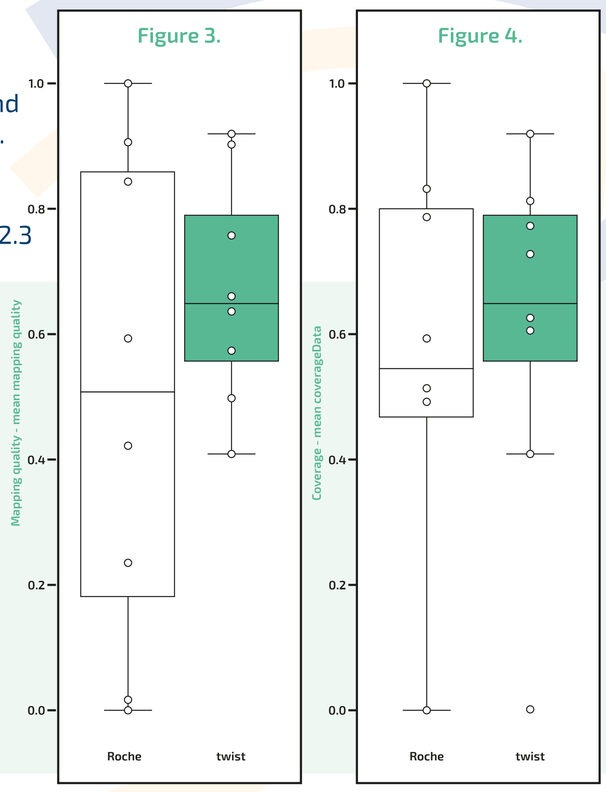
- Plex 2 with the Twist panel achieved the required 480 million reads per plex

Bioinformatic analysis and reference mapping
- Sequence quality control – FastQC v0.11.9
- Reads aligning to the TWIST panel regions were extracted and downsampled to standardize the read count across samples.
- Aligning to the hg38 reference genome, variant calling – DRAGEN v4.2.7
- BAM file metrics, mapping quality and coverage – Qualimap v2.3

The evaluated bioinformatics parameters indicate higher stability and reliability in mapping to the reference genome and complete coverage of target regions for the Twist EF 2.0 panel (Figures 3 and 4).

Figure 3. The Twist Exome 2.0 Panel (black) demonstrates higher consistency in mapping quality with lower variability in values compared to the KAPA HyperPETE Panel (red), indicating more stable read alignment to the reference genome.

Figure 4. The Twist Exome 2.0 panel demonstrates coverage comparable to the KAPA HyperPETE Panel but with significantly lower deviation. The lower variability in coverage indicates higher stability and reliability of the Twist Panel in ensuring uniform coverage of target regions.



Conclusion

In this study, we compared the technical performance of the Twist Exome 2.0 Panel and the KAPA HyperPETE Newborn Screening Panel. Despite the expectation that a small, targeted panel would exhibit greater uniformity and stability, our results show that the Twist Exome 2.0 Panel outperforms the smaller panel in mapping quality and coverage consistency. This suggests that the advanced probe design and optimization of the Twist panel provide a significant advantage, making it a reliable choice for high-quality exome sequencing and comprehensive variant detection.