

SUPPLEMENTAL MATERIAL

Next Gene Sequencing (NGS)

Patients were screened using next-generation sequencing (NGS) with TruSight Oncology (TSO) 500 (Illumina, San Diego, CA, USA), a panel targeting 500+ cancer genes as described previously.^{1 2} To examine *BRAF* mutation in all samples, DNA library was developed by a hybrid capture-based TSO 500 DNA/RNA NextSeq Kit, following the manufacturer's protocol. Sequence data of all samples were measured to identify clinically relevant class of genomic alterations. Annotation of filtered data acquired from the TSO 500 pipeline were done through the Ensembl Variant Effect Predictor Annotation Engine¹ with information from databases, such as gnomAD genome and exome, 1000 genomes, COSMIC, dbSNP, ClinVar, RefSeq, and Ensembl.

REFERENCES

1. Pestinger V, Smith M, Sillo T, *et al.* Use of an Integrated Pan-Cancer Oncology Enrichment Next-Generation Sequencing Assay to Measure Tumour Mutational Burden and Detect Clinically Actionable Variants. *Mol Diagn Ther* 2020;24:339-49.
2. Karasaki T, Nakajima J, Kakimi K. Neoantigens and Whole-Exome Sequencing. *Gan To Kagaku Ryoho* 2016;43:791-7.