B-07

Identification of Tumor Suppressor Genes in Nasopharyngeal Carcinoma

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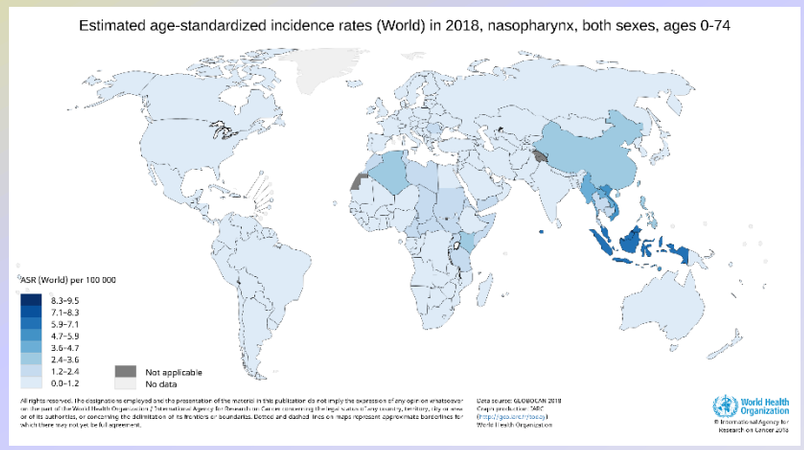
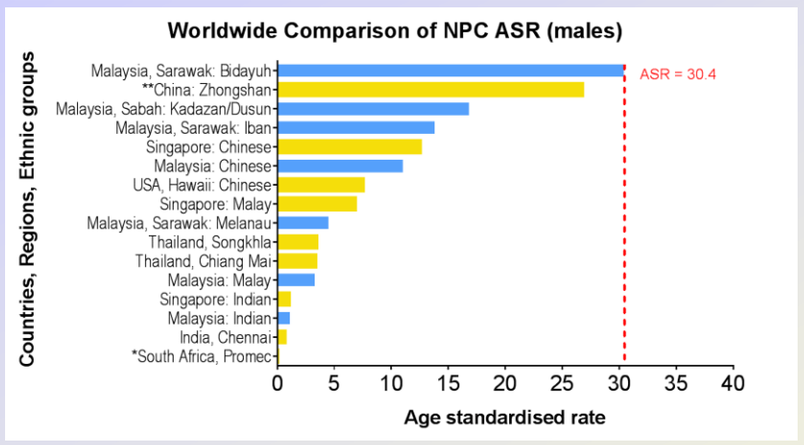
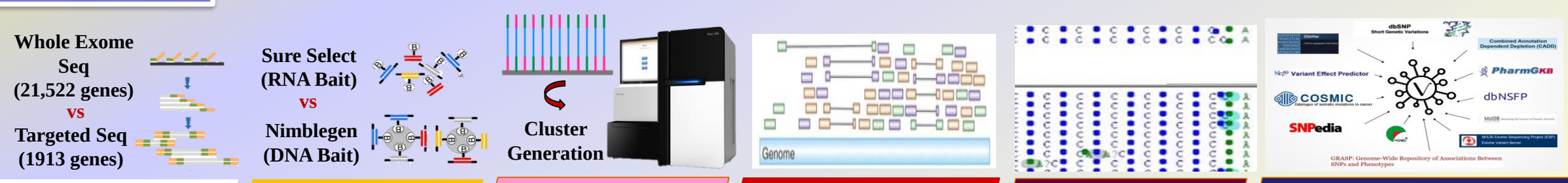
INTRODUCTION

Targeted capture of genomic regions of interest for massively parallel sequencing allows simultaneous screening of mutations in hundreds of loci in genetically heterogeneous human diseases. It has become a widely used method in genetic testing because it leads to increased cost-effectiveness, shorter turn-around-time, higher accuracy and accessibility[1,2]. Nasopharyngeal Carcinoma (NPC) is uncommon in many parts of the world with the age-standardize rates of about 2.2 per 100,000[3] but is one of the most prevalent cancers in Southeast Asia especially among the Bidayuhs and Kadazans, with its aetiology understudied[4]. Familial clustering shows that individuals with first degree family history of NPC have elevated risk of NPC with odd ratios ranged from 2 to 20 as compared to those with no family history of NPC[5]. As genetic factors such as tumor suppressor genes that confers susceptibility to NPC remain unclear and understudied in different ethnic groups in Malaysia, we conduct Whole Exome Sequencing and Targeted Sequencing in 24 NPC cases with family history of NPC to evaluate the feasibility of targeted sequencing as well as to identify genetic factors that may lead to development of NPC.





METHODOLOGY



Genomic DNA Library Preparation → Targeted DNA Enrichment → Paired-End Sequencing → Alignment to Human Reference Genome → Variant Detection (SNVs and Indels) → Variant Annotation (DBSNP, Annovar etc)

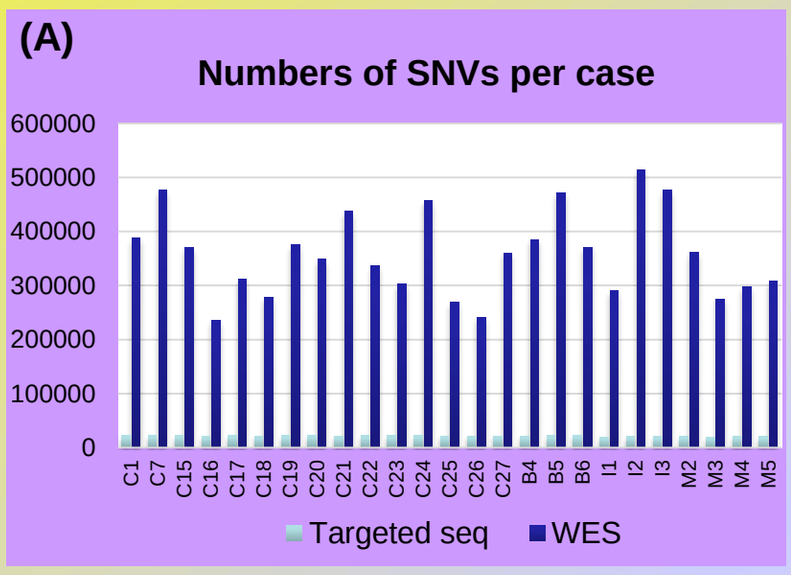
RESULT

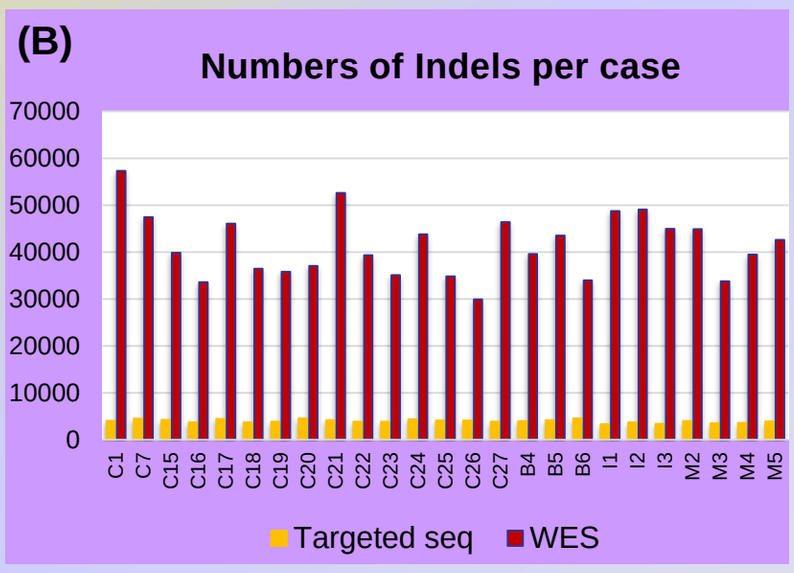
Table 1. Alignment and coverage statistics of whole exome sequencing and targeted sequencing for 24 NPC cases with family history of NPC. SNVs, Single Nucleotide Variants. Indels, small insertions or deletions.

	Whole Exome Sequencing	Targeted Sequencing
Total no. of Genes Analysed	21,522	1,913
Average Total Reads Passing Filter	47,281,522	7,209,895
Average Number of Aligned Reads	46,844,872	6,450,093
Average % of Aligned Reads	96.8	93.2
Average Total SNVs identified	387,342	22,671
Average Total Indels identified	57,298	4,037

Table 2. Potentially pathogenic variants identified among the 24 NPC cases with family history of NPC.

Gene	Sample ID	Ref	Alt	AAChange.refGene	Mutation	Affected Pathway
TTLL3	C12, M1	TC	T	c.2450delC:p.S817X	nonsense	Cytosol
MSH2	C15	C	T	c.C28T:p.Q10X	nonsense	DNA Repair
DNAH6	B3	G	T	c.G4210T:p.E1404X	nonsense	Axonemal Dynein Complex
TLR6	M5	G	A	c.C2269T:p.Q757X	nonsense	Innate Immune System





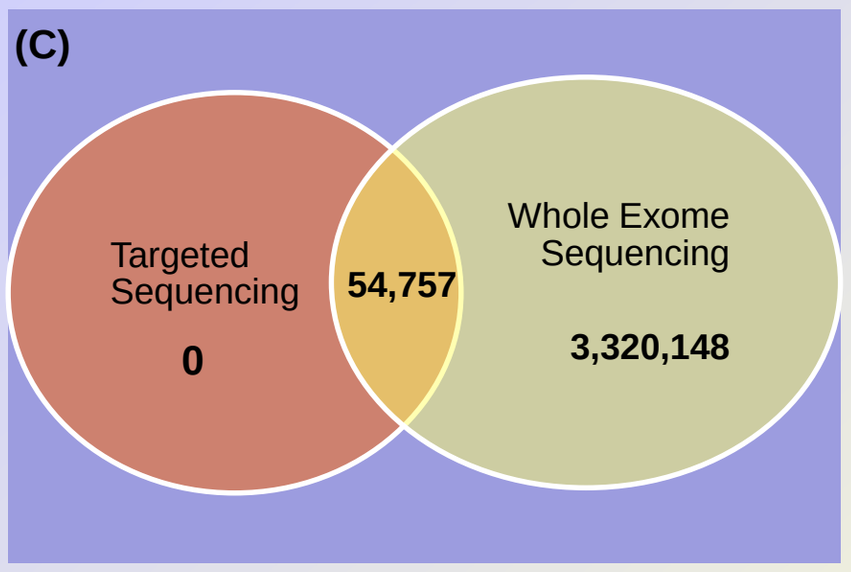


Figure 1. Total numbers of **A)** SNVs or **B)** Indels found in 24 NPC cases with family history of NPC by Targeted Sequencing and WES. **C)** The concordance of SNVs and Indels between these two sequencing platforms. SNVs, Single Nucleotide Variants. Indels, small insertions or deletions. WES, Whole Exome Sequencing.

DISCUSSION & CONCLUSION

Genetic susceptibility to cancer is usually caused by dominantly inherited heterozygous mutations of tumour suppressor genes. However, tumour suppressor genes which confer such increased risk of NPC are understudied among various high risk groups in Malaysia. Comprehensive analysis were performed to identify potential susceptibility genes and in-depth characterization and validation of the target genes should be done to evaluate the association of such genes in pathogenesis of NPC. In our study, the targeted sequencing data demonstrates very high correlation with the WES data and our results show that both platforms are useful for the identification of potential tumour suppressor genes in NPC. It is found that in NPC, pathways related to innate immune system and DNA repair are affected. This will further enhance our understanding in the molecular basis of the disease and shed light for future prevention, diagnosis, gene therapy and/or management of NPC.

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