A Whole-Genome-Informed Pipeline for Neoantigen Discovery in Solid Tumors: Integrating SNV, Splice Variant, and Exon–Transposon Junction Analysis to Enable Personalized Cancer Vaccines

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1 INTRODUCTION

- ✓ Personalized cancer vaccines (PCVs) are promising novel immunotherapy.
- ✓ In the phase II KEYNOTE-942 trial[1], the addition of a PCV to anti-PD-1 therapy improved recurrence-free survival in melanoma, and several phase III trials are now ongoing[2-4].
- ✓ Vaccine development has mainly focused on canonical antigens derived from SNVs and indels identified by whole-exome sequencing (WES).
- ✓ Recently, non-canonical antigens, including those arising from splicing variants and exon-transposon junctions, have been reported as potential targets[5-6].

1) The Lancet, **403** (10427), 632-644 (2024). 2) Journal of Clinical Oncology, **42**, TPS9616 (2024). 3) Journal of Clinical Oncology, **42**, TPS8116 (2024). 4) Cancer Research, **85**, CT251 (2025). 5) Nature, **639**, 463–473 (2025). 6) Nature Reviews Cancer, **24** (2), 123-140 (2024).

2 OBJECTIVES

To develop and apply a Whole-Genome Sequencing (WGS)- and RNA-seq-based AI-driven pipeline for the comprehensive prediction for canonical and non-canonical antigens to advance next-generation PCVs.

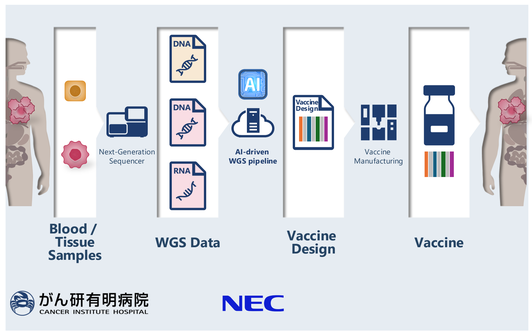
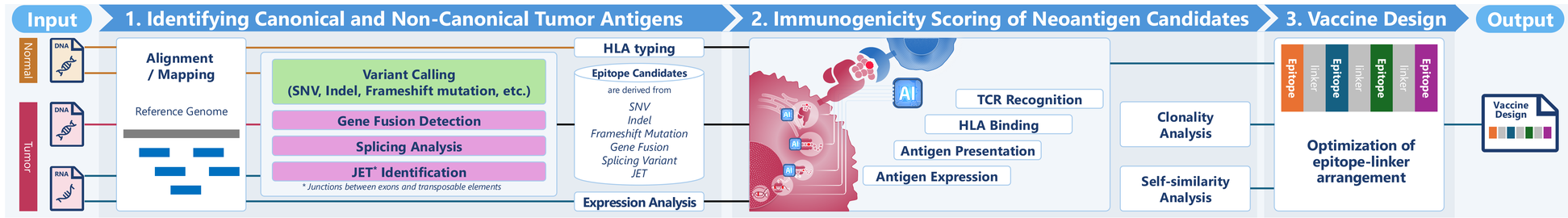


Figure: End-to-End Process for Personalized Cancer Vaccine Development and Administration

3 METHODS



Data from 43 breast and 158 sarcoma cases who participated in the national WGS project at our institution.

Canonical Antigen: Generated by SNVs, in-frame insertions/deletions, or frameshift mutations that lead to abnormal protein sequences with potential immunogenicity. These are typically detected by WES.

Non-canonical Antigen: Derived from gene fusions, splicing variants, or exon–transposon junctions (JETs). These are not necessarily associated with mutations but result in aberrant protein sequences.

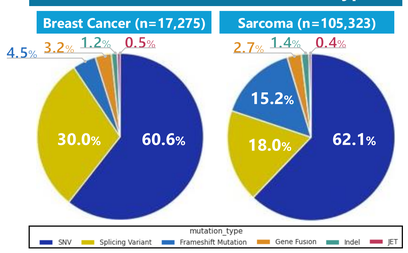
4 RESULTS

Patient Characteristics

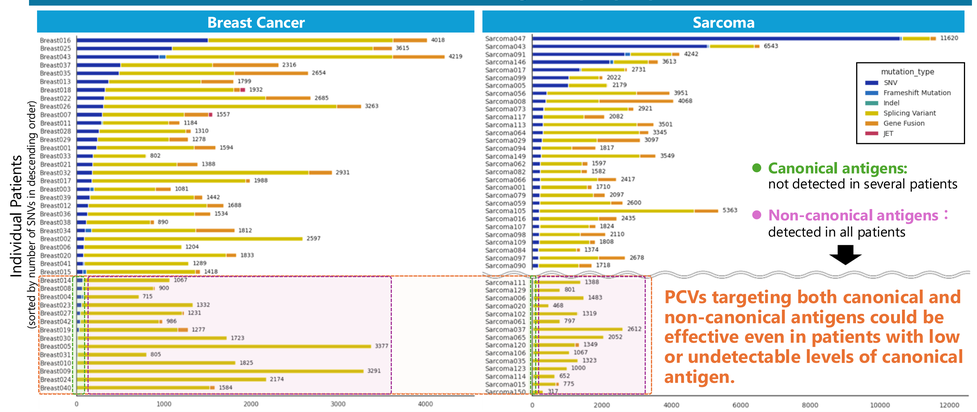
Breast Cancer (n=43)		
Median Age; years (range)	53	(20-80)
Sex, female; n (%)	43	(100)
Settings; n (%)		
Stage I-III	32	(74.4)
Metastatic/Recurrent	11	(25.6)
Subtype; n (%)		
Luminal	23	(53.5)
TNBC	14	(32.6)
HER2	1	(2.3)
Unknown	5	(11.6)
Race; n (%)		
Japanese	42	(97.7)
Other	1	(2.3)

Sarcoma (n=158)		
Median Age; years (range)	61	(15-90)
Sex, Male:Female; n (%)	92:66	(58.2:41.8)
Histologic Subtype; n (%)		
Liposarcoma	38	(24.1)
UPS	21	(13.3)
Leiomyosarcoma	19	(12.0)
Myxofibrosarcoma	14	(8.9)
Osteosarcoma	12	(7.6)
Ewing sarcoma	6	(3.8)
Others	48	(30.4)
Race; n (%)		
Japanese	155	(98.1)
Other	3	(1.9)

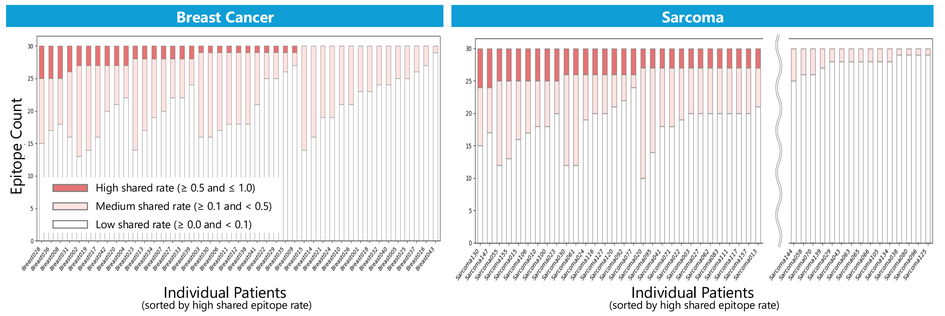
Distribution of Detected Variant Types



Number of Predicted Immunogenic Epitopes per Patient



Distribution of Shared Rates among the Top 30 Predicted Immunogenic Epitopes per Patient



The low ratio of shared epitopes among patients suggests a high degree of individual variability, highlighting the need for personalized cancer vaccines.

5 CONCLUSION

- ✓ We developed an AI-driven prediction pipeline based on WGS and RNA-seq data.
- ✓ This pipeline successfully identified numerous non-canonical antigens beyond the scope of WES.
- ✓ These findings may contribute to the development of next-generation personalized cancer vaccines targeting both canonical and non-canonical antigens.
- ✓ Future Work: Validate the immunogenicity of non-canonical antigens through in vivo and ex vivo experiments.

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