Evaluation of TNBC-PDX Models for Personalized Medicine

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Abstract

Background:
Triple-negative breast cancer (TNBC) is known to be highly heterogeneous compared to other subtypes. Establishing reliable preclinical models is essential for personalized treatment strategies, such as selecting effective drugs for each patient. Patient-derived xenograft (PDX) models, created by transplanting patient tumors into immunodeficient nude mice, are considered to more accurately replicate tumor diversity and drug responses. This study aimed to evaluate the retention of clinically relevant biomarkers in TNBC-PDX models.
Methods:
With approval from the Kobe University Institutional Review Board, tumor tissues were collected from TNBC patients who met the inclusion criteria and provided informed consent. The tissues were implanted into nude mice to establish TNBC-PDX models. Expression of key biomarkers—EGFR, AR, CK5/6, HER2, and Trop-2—was assessed in both the original patient tumors and corresponding PDX tumors using immunohistochemistry.
Results:
EGFR and Trop-2 expression was retained in all models. However, AR expression was not preserved. CK5/6 and HER2 showed variable expression across cases.
Conclusion:
TNBC-PDX models are promising tools for drug evaluation in personalized medicine. However, since some biomarkers, such as AR and CK5/6, may not be consistently retained, prior validation of biomarker expression is essential for accurate therapeutic evaluation.

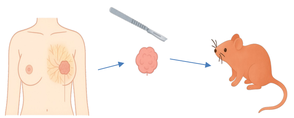
Introduction

TNBC remains one of the most aggressive breast cancer subtypes, lacking established molecular targets such as ER, PgR, and HER2. PDX models have emerged as indispensable preclinical tools that faithfully reproduce the histological and molecular features of original tumors, thus bridging laboratory research with clinical application. However, it remains unclear whether these models consistently retain therapeutic targets essential for developing personalized treatment strategies. We analyzed the immunohistochemical concordance of key biomarkers— Trop-2, EGFR, AR, CK5/6, and HER2—between patient tumors and corresponding TNBC-PDX models, revealing both preserved and lost expressions. Building on these findings, the present study evaluates TNBC-PDX models as precision-medicine platforms, emphasizing how molecular validation across generations can enhance their translational reliability. This approach aims to optimize individualized drug testing, particularly for antibody–drug conjugates and novel targeted therapies in TNBC.

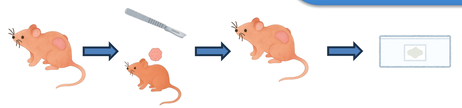
Material and Methods

PDX establishment:
Fresh tumor tissues obtained from TNBC patients undergoing surgery at Kobe University Hospital were orthotopically implanted into the mammary fat pads of immunodeficient mice. Each specimen was divided into small fragments and transplanted into two to three BALB/c-nu mice under sterile conditions. Tumor growth was monitored weekly, and engrafted tumors were serially passaged to establish stable PDX lines. All procedures were performed under approved institutional animal welfare protocols.
Ethics Board of Kobe University (Ethics Committee No.B200065 and B200065-1)
Animal Care and Use Committee of Oriental BioService Inc.(approval no. ma-K1804_2024)

Immunohistochemical analysis:
Tumor sections from both patient specimens and corresponding PDX tissues were subjected to immunohistochemical staining for key biomarkers: Trop-2, EGFR, AR, CK5/6, and HER2. Standardized protocols were followed using HRP-labeled secondary antibodies and DAB visualization. Two independent investigators evaluated staining intensity and distribution according to ASCO/CAP guidelines. Expression concordance between patient and PDX tissues was assessed to determine marker preservation.



Tumor samples were obtained from patients and transplanted into nude mice. (Murata et al 2020)



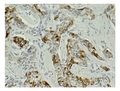
Once the tumors reached an adequate size, they were passaged, and tumor tissues were collected and preserved for analysis at each passage. (Inoue et al 2025)

Results

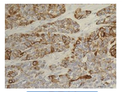
PDX establishment: Successful engraftment was achieved in 8 of 10 cases (80%).

Immunohistochemical analysis : Representative results are shown below.

CK5/6



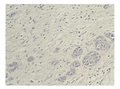
Patient



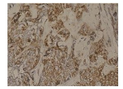
PDX

All patient tumors were positive; however, one PDX model (K006) showed a complete loss of expression. In case K012 in the figure, the expression is preserved in the PDX.

HER2



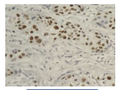
Patient



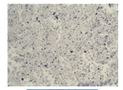
PDX

Expression patterns were concordant in five of six cases, while one case(K007) showed increased IHC score(from 0 to 2) in the figure.

AR



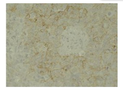
Patient



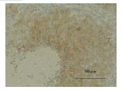
PDX

Two patient tumors were positive, but all corresponding PDX tumors became negative. In K006 shown in the figure, the original tumor is positive, whereas the PDX is negative.

Trop-2

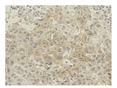


Patient

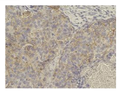


PDX

EGFR



Patient



PDX

For EGFR and Trop-2, expression was preserved in all cases.

Immunohistochemical Profile of Patient and PDX Tumors

Number		Histology	CK5/6	HER2	EGFR	AR	Trop-2
K006	patient	invasive breast cancer of no special type	+	0	+	+	+
	PDX	-	-	0	+	-	+
K007	patient	invasive breast cancer of no special type	+	0	-	-	+
	PDX	-	+	2	-	-	+
K008	patient	invasive breast cancer of no special type	+	0	+	-	+
	PDX	-	+	0	+	-	+
K010	patient	metaplastic carcinoma	+	2	-	+	+
	PDX	-	+	2	-	-	+
K012	patient	invasive breast cancer of no special type	+	1	-	-	+
	PDX	-	+	1	-	-	+
K014	patient	invasive breast cancer of no special type	+	0	-	-	+
	PDX	-	+	0	-	-	+

Discussion

In this study, several molecular markers showed discordant expression between patient tumors and their corresponding TNBC-PDX models. While EGFR and HER2 tended to remain stable, AR and CK5/6 frequently demonstrated loss or alteration after engraftment. These changes likely arise from the intrinsic intratumoral heterogeneity of TNBC, where diverse subclones coexist. During transplantation into immunodeficient mice, selective pressures may favor clones better suited to the murine microenvironment. The loss of AR expression may reflect the androgen-poor conditions of nude mice, which disadvantage AR-positive cells. Additionally, the absence of human immune components may further alter tumor phenotypes.

Conclusion

At the Department of Breast Surgery, Kobe University, we successfully established TNBC PDX models with an 80% success rate.
We compared the expression of molecular markers between the original patient tumors and their corresponding PDX tumors, confirming that the PDX system functions as a highly practical and reliable preclinical model.
In breast cancer treatment, selecting therapeutic agents that match the molecular characteristics of each patient's tumor is essential. Likewise, in preclinical drug development, it is necessary to verify whether the target molecular markers are expressed in the models being used.

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