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A SHORT COMMUNICATION ON THE MOLECULAR EXPRESSION ON TNBC TRIPLE NEGATIVE BREAST CANCER

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ABSTRACT
Triple negative breast cancer (TNBC) assign to be an aggressive breast cancer phenotype which is described by lack of estrogen receptor (ER) and progesterone receptor (PR), as well as the absence of over expression of human epidermal growth factor receptor-2 (HER-2). Higher rate of early recurrence and distant metastasis to lungs and liver compared to other breast cancer subtype resulting in overall poor prognostic in TNBC. To date, only chemotherapy is available as systemic therapy for TNBC which help only in patients with chemotherapy-sensitive disease. In patients with metastatic TNBC, the chemotherapy agent responds well initially but due to shorter time for disease progression, shows poor overall survival rate. The aim of this article is to review on the molecular subtypes and potential targeting receptors of TNBC and provide suggestion for developing hopeful targeting therapy in the coming future.
Keywords: Triple negative breast cancer, Molecular subtypes, Targeting receptors

INTRODUCTION

Breast cancer was the most prevalent types of cancer among females in the Asia-pacific region, reckoning for 18% of all cases in 2012, and was the fourth most common cause of cancer related deaths (9%). Despite incidence rates remain much higher in New Zealand and Australia, breakneck rises in recent years were observed in several Asian countries. It was estimated that almost 1.7 million cases of female breast cancer were diagnosed worldwide during 2012, corresponding to a rate of 43 per 100,000. Among that, 24% of all breast cancers were diagnosed within the Asia-pacific region (approximately 404,000 case at a rate of 30 per 100,000 with the greatest number these occurring in China (46%), Japan (14%) and Indonesia (12%).

Extensive increases in breast cancer mortality rates also occurred in many areas, particularly Malaysia (6% per year from 1997-2008) and Thailand (7% per year from 2000-2006 with an average annual increase of 9% from 1985 onwards), in contradiction to steady trends in Hong Kong and Singapore, while decreases have been reported in Australia and New Zealand. Mortality trends by age groups in eight of the ten Asian countries tended to be more favorable for women aged under 50 compared to those who were 50 years or older. However, the exceptions were China and Thailand, where mortality rates were increasing rapidly regardless of the age at death [1].

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Amidst of all the breast cancer subtypes, triple negative breast cancer (TNBC) assign to be an aggressive breast cancer phenotype which is described by lack of estrogen receptor (ER) and progesterone receptor (PR), as well as the absence of over expression of human epidermal growth factor receptor-2 (HER-2) [2]. Higher rate of early recurrence and distant metastasis to lungs and liver compared to other breast cancer subtype resulting in overall poor prognostic in TNBC [3]. This untoward clinical outcome is due to its aggressive pathological features including higher histology grade and mitotic index [4].

To date, only chemotherapy is available as systemic therapy for TNBC which help only in patients with chemotherapy-sensitive disease. In patients with metastatic TNBC, the chemotherapy agent responds well initially but due to shorter time for disease progression, shows poor overall survival compared to ER+ breast cancer according to numerous studies [5].

Furthermore, fundamental genomic uncertainty of TNBC delivers the opportunity of faster adaptation to the cytotoxic effect of chemotherapy, causing limited treatment options for chemotherapy-resistant TNBC. Some established targeted therapies such as endocrine treatment and HER-2 targeted agents are ineffective in case TNBC due to nature of the cancer. Several small molecule inhibitors and monoclonal antibodies against important cellular pathways have been tested in clinical trial; however,

no one enrolled in clinical practice because of insufficient efficacy [6].

MOLECULAR SUBTYPES OF TNBC

In recent past, gene expression profiling was done by Lehmann and colleagues in which classify TNBC into 6 molecular subtypes. These different molecular subtypes of TNBC include basal-like (BL-1 and BL-2) mesenchymal subtype (M), mesenchymal stem-like (MSL), immunomodulatory (IM), and lastly luminal androgen receptor (LAR) subtype. The study was further established by the authors in investigating potential therapeutic agents suitable for each subtypes of breast cancer in their related cell lines (Table 1) [7].

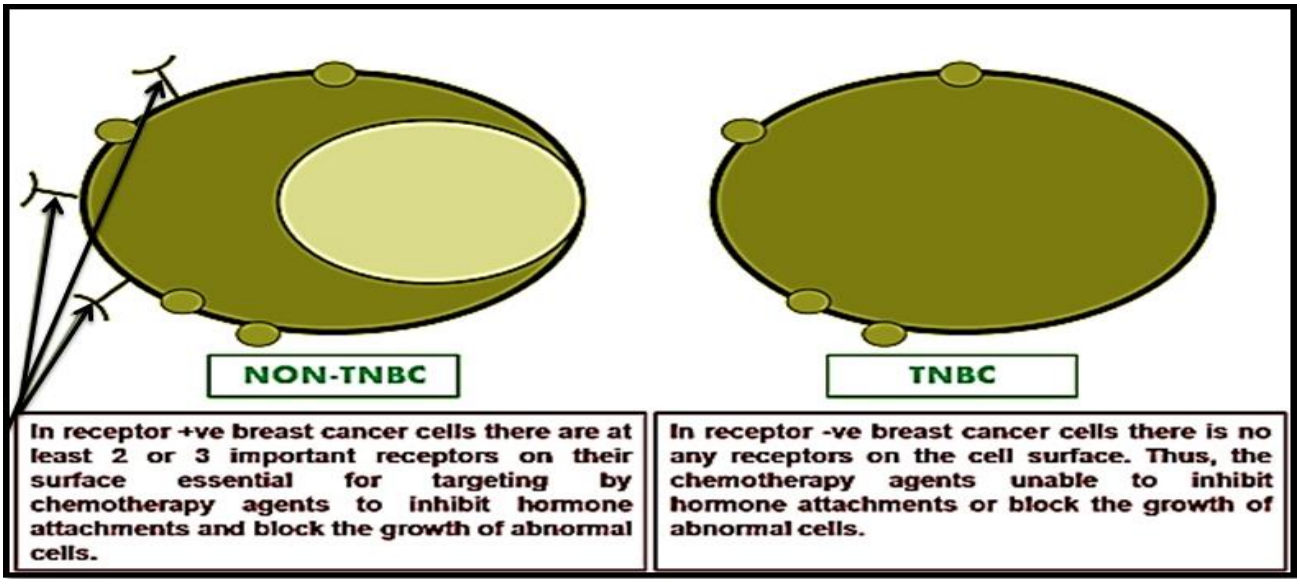

Figure 1: Graphical illustration of Non-TNBC and TNBC

Table 1 TNBC subtypes, their genetic characteristics, and potential therapeutic agents

Subtypes	Genetic characteristics	Therapeutic agent
BL-1 and BL-2	Higher expressions of cell cycle and DNA damage response genes	Cisplatin-Alkylating agent
M and MSL	Enhanced gene expression for epithelial-mesenchymal transition and growth factor pathways	Dactolisib (NVP-BEZ235)-PI3K/mTOR inhibitor Dasatinib- Abl-src inhibitor
LAR	AR signalling	Bicalutamide-AR antagonist

Source: *Lehmann et al*.

Abbreviations: BL-1, basal-like-1; BL-2, basal-like-2; M, mesenchymal; MSL, mesenchymal stem-like; LAR, luminal androgen receptor; AR, androgen receptor; PI3K, phosphatidylinositol 3-kinase; mTOR, mammalian target of rapamycin; abl/src, tyrosine kinase inhibitors).

In another study by the same authors, the distribution of TNBC subtypes separately was analyzed by using the intrinsic subtype tool. The results indicate that almost every TNBC subtypes constitute mainly basal-like (BL) intrinsic subtype (BL-1, BL-2, IM, and M) except for MSL and LAR subtypes. The LAR subtype was belonging to HER-2 and luminal B, whereas, the MSL subtype carrying BL, normal-like and luminal B [8].

Claudin-low tumor subtype was also determined in different study using gene expression profiling. The molecular profile of this tumor displays epithelial to mesenchymal transition, immune response as well as stem like features. However, luminal and proliferation related genes are very low in this type of tumor subtype [9]. It was stated that claudin-low breast cancer shows a TNBC characteristic but only in smaller extent. In addition, the claudin-low

subtype is negative for ER, PR, HER-2, claudin (3, 4 and 7) and E-cadherin [10]. Aside from that, Burstein and colleagues were conducted gene profiling studies on 198 TNBC tumors and found out that TNBC can be classified into 4 noticeable subtypes. These include luminal androgen receptor subtype (AR; LAR), mesenchymal subtype (M), basal-like immunosuppressed subtype (BLIS) and basal-like immune activated subtype (BLIA). It can be said that LAR and M tumor subtypes fitted into LAR and MSL subtypes which was claimed by Lehmann et al [7, 11].

Thus, by extensive understandings of the various TNBC subtypes and their characteristic features, one can definitely predict the most promising therapeutic agent that is suitable for that particular tumor subtype. This is because previous studies prove that different chemotherapy agents respond differently to each TNBC subtypes [9].

POTENTIAL TARGETING RECEPTORS IN TNBC

Luteinizing Hormone Releasing Hormone (LHRH) Receptor:

Luteinizing hormone releasing hormone (LHRH) can be an excellent target for TNBC treatment. Previous study shows that approximately 50% of LHRH is expressed in breast cancer cells along with TNBC. AEZS-108 is an investigational drug currently in the clinical trial. It is a cytotoxic LHRH analog in which a small peptide (D-Lys6) that is regarded as LHRH agonist is linked with doxorubicin could be potential drug for targeted therapy with anticancer agents in TNBC that is associated with higher expression of LHRH receptors [12]. Similarly, in another study by Stephan and colleagues who investigate 69 human surgical specimen of TNBC for LHRH receptors and further evaluates the efficacy of the LHRH anolog, AEZS-108 as well as AEZS-125 which is a newer LHRH analog that is linked with Disorazol Z, a cytotoxic compound found naturally having anticancer properties. The result of the study indicates LHRH receptors are expressed in TNBC significantly (49%). The cytotoxic analogs LHRH, AEZS-108 and AEZS-125 can be ultimately used as targeted therapy in TNBC [13].

Androgen Receptor (AR)

AR expression in TNBC is also said to be an effective targeted therapy. The AR presents in TNBC cells is differ in term of method used for assay, selecting criteria for positive result, and number of patients involved. However, available data suggests that higher level of AR expression is related with bettered treatment outcome. So, newer targeted therapy can be developed in future for TNBC patients associated with higher AR expression [14].

EPIDERMAL GROWTH FACTOR RECEPTOR (EGFR)

The epidermal growth factor receptor (EGFR) is member of epidermal growth factor receptors family of extracellular protein ligands. The EGFR can be an optimal therapeutic target in TNBC. It is found to be overexpressed in the TNBC cell lines in contrast to human epidermal growth factor receptor 2 (HER-2) +ve cel lines. Corkery and colleagues also conclude that despite the lesser sensitivity effect of TNBC cells on the EGFR inhibition than the HER-2 +ve cell lines, the addition of gefitinib improved the chemotherapy response in TNBC patients. Gefitinib provide a synergistic effect together with carboplatin and docetaxel. Thus, future studies on the triple chemotherapy agents are recommended in TNBC patients [15].

PROLACTIN RECEPTOR (PRLR)

Generally, prolactin (PRL) hormone plays a crucial part in the progression of mammary gland growth and terminal differentiation of breast epithelial cells. In previous studies, it had been shown that the PRL and PRLR were essential in the development and progression of breast cancer. However, a recent study shows that the PRL and PRLR potentially restrict the breast carcinogenesis [16]. Vanessa and colleagues in their study, relate PRL and its associated pathway as a subtype as well as the predictor of pro-differentiator treatment in TNBC. By various genomic analysis and immunocytochemistry assay, the PRLR and mRNA level were noted to be downregulated in TNBC cases. Furthermore, a new subtype of TNBC-PRLR was identified from the gene expression of 580 TNBC cases. This new class is known to be luminal differentiation of epithelial cells. A combined or individual expression of genes in PRL pathways (PRL, PRLR, Jak2 and Stat5a) was capable to produce improved results in TNBC patients. Ultimately, the tumor formation can be suppressed by either repairing or stimulating the PRL pathways in the TNBC cells. In a nutshell, a new therapeutic approach can be developed by activation of this pathway in TNBC patients associated with TNBC-PRLR subtype [17].

UROKINASE RECEPTOR (uPA)

Researchers had discovered that the urokinase-type plasminogen activator (uPA) and its receptor (uPAR) overexpression in TNBC cells [18, 19].

Michaela and colleagues reveals that uPAR enriches the malignancy of TNBC by direct interaction with uPA and insulin-like growth factor receptor 1 (IGF1R). Their study on TNBC cell lines (MDA-MB-231 cells) found that uPAR interaction with uPA and IGF1R resulting in direct tumor progression. The interaction was shown to be incomparably shortened by the downregulation of uPAR. Thus, with combined inhibition of uPAR, uPA and IGF1R produced a synergistic effect and notably reduced the tumor progression in TNBC cells [20]. In addition, the effect can be further enhanced with the addition of plasminogen activator inhibitor-1 (PAI-1) which inhibits the uPA [20, 21].

NOTCH4 RECEPTOR

Notch signaling pathway is a cell signaling pathway that exists in multicellular organisms. The notch genes cause protein activation by their interaction with ligands. This interaction may results cellular growth progression as well as their survival. Generally, mammals have mainly 4 different notch receptors (NOTCH1, NOTCH2, NOTCH3, and NOTCH4) and 5 ligands (Delta like 1-4 and Jagged 1-2) for all the receptors [22]. Among the 4 notch receptors, NOTCH4 receptor may be a possible therapeutic target for TNBC. A study by Nagamatsu and colleagues in evaluating if the NOTCH4 receptor can be an important targeting agent for TNBC have observed that inhibition of NOTCH4 in TNBC cells lessen cell reproduction and metastasis. In contrast, the overexpression of NOTCH4 resulting in the opposite effect in the TNBC cells. Thence, NOTCH4 can be a likely targeting option for TNBC [23].

MET RECEPTOR

Mesenchymal epithelial transition (MET) pathway does not work properly in most of the cancers. MET (mesenchymal epithelial transition) is a receptor of tyrosine kinase family that plays a significant part in growth and proliferation of healthy cells [24]. Basically, the MET and its ligands; hepatocyte growth factor (HGF) and scatter factor (SF) are needed for regular cell development [25]. However, the MET receptor and its ligands are overexpressed in most of the cancers including breast cancer [26]. A study by Zagouri and colleagues on MET expression in TNBC patients reveals that high expression of MET in TNBC patients resulting in lower survival rate. Additionally, the assay for MET expression in formalin-fixed, paraffin-embedded (FFPE) surgical sample of TNBC showed higher expression of MET in 52% of tumors out of 170 samples analyzed. Thus, the MET pathway may also significantly able to be a potential target for TNBC patients [25].

VEGFR-2

Vascular endothelial growth factor receptor 2 (VEGFR-2) plays an important role in angiogenesis. By binding of its ligand, vascular endothelial growth factor-A (VEGF), the VEGFR-2 pathway is activated. This VEGF is secreted by cancer cells in order to trigger the activation of VEGFR-2 pathway in the normal endothelial cells beside them for their vascular development. Surprisingly, the VEGFR-2 pathway is identified in the breast cancers including TNBC [27]. In a study by Najafi and colleagues on the VEGFR-2 expression in 104 TNBC patients, showed that 61% patients having the receptor existence in the TNBC cells [28]. Similarly, Jansson and colleagues also found higher protein expression of VEGFR-2 in TNBC patients in comparison to non-TNBC patients [29]. Thus, VEFGR-2 receptor targeting can be also being a promising therapeutic option in TNBC.

CONCLUSION

Triple negative breast cancer (TNBC) can be clearly seen as an aggressive breast cancer phenotype which is lack of estrogen receptor (ER) and progesterone receptor (PR), as well as the absence of over expression of human epidermal growth factor receptor-2 (HER-2). Additionally, this disease also presents with many molecular subtypes which can be an essential part for deciding the treatment plan. The presented review on potential targeting receptors in TNBC can somehow provide suggestion for developing hopeful targeting therapy in the coming future.

REFERENCES

[1] DR Youlden, SM Cramb, CH Yip, PD Baade. Incidence and mortality of female breast cancer in the Asia-Pacific region. Cancer. *Biol Med*. 11(2):101–15 (2014).

[2] SK Kanapathy Pillai, A Tay, S Nair, Leong C-O. Triple-negative breast cancer is associated with EGFR, CK5/6 and c-KIT expression in Malaysian women. *BMC Clin Pathol*.1472-6890-12-18 (2012).

[3] ZY Yuan, SS Wang, Y Gao, ZY Su, WB Luo, ZZ Guan. Clinical characteristics and prognosis of triple-negative breast cancer: A report of 305 cases. *Ai Zheng Aizheng Chin J Cancer*. 27(6):561–5 (2008).

[4] AA Thike, PY Cheok, AR Jara-Lazaro, B Tan, P Tan, PH Tan. Triple-negative breast cancer: clinicopathological characteristics

and relationship with basal-like breast cancer. *Mod Pathol.* 23(1):123–33 (2010).

[5] HA Wahba, HA El-Hadaad. Current approaches in treatment of triple-negative breast cancer. *Cancer Biol Med*. 12(2):106–16 (2015).

[6] PF Peddi, MJ Ellis, C Ma. Molecular Basis of Triple Negative Breast Cancer and Implications for Therapy. *Int J Breast Cancer*.2012:1–7 (2012).

[7] BD Lehmann, JA Bauer, X Chen, ME Sanders, AB Chakravarthy, Y Shyr, et al. Identification of human triple-negative breast cancer subtypes and preclinical models for selection of targeted therapies. *J Clin Invest*. 121(7):2750–67 (2011).

[8] BD Lehmann, JA Pietenpol. Identification and use of biomarkers in treatment strategies for triple-negative breast cancer subtypes. *J Pathol*. 232(2):142–50 (2014).

[9] J Collignon, L Lousberg, H Schroeder, G Jerusalem. Triple-negative breast cancer: treatment challenges and solutions. *Targets and Therapy*.2016:8 93-107 (2016).

[10] K Dias, A Dvorkin-Gheva, RM Hallett, Y Wu, J Hassell, GR Pond, et al. Claudin-Low Breast Cancer; Clinical & Pathological Characteristics. *Plos One*. Jan 3; 12 (1):e0168669 (2017).

[11] MD Burstein, A Tsimelzon, GM Poage, KR Covington, A Contreras, SAW Fuqua.,et al. Comprehensive Genomic Analysis Identifies Novel Subtypes and Targets of Triple-Negative Breast Cancer. *Clin Cancer Res*. 1; 21(7):1688–98 (2015).

[12] J Engel, G Emons, J Pinski, AV Schally. AEZS-108: A targeted cytotoxic analog of LHRH for the treatment of cancers positive for LHRH receptors. *Expert Opin Investig Drugs*. 21(6):891–9 (2012).

[13] S Seitz, S Buchholz, AV Schally, F Weber, M Klinkhammer-Schalke, EC Inwald, et al. Triple negative breast cancers express receptors for LHRH and are potential therapeutic targets for cytotoxic LHRH-analogs, AEZS 108 and AEZS 125. *BMC Cancer*.19;14:847 (2014).

[14] M Rampurwala, KB Wisinski, R O'Regan. Role of the Androgen Receptor in Triple-Negative Breast Cancer. *Clin Adv Hematol Oncol*. 14 (3):186–93 (2016).

[15] B Corkery, J Crown, M Clynes, N O'Donovan. Epidermal growth factor receptor as a potential therapeutic target in triple-negative breast cancer. *Ann Oncol*. 20 (5):862–7 (2009).

[16] Z Nouhi, N Chughtai, S Hartley, E Cocolakis, J-J Lebrun, S Ali. Defining the Role of Prolactin as an Invasion Suppressor Hormone in Breast Cancer Cells. *Cancer Res*.1; 66(3):1824–32 (2006).

[17] VM López-Ozuna, IY Hachim, MY Hachim, J-J Lebrun, S Ali. Prolactin Pro-Differentiation Pathway in Triple Negative Breast Cancer: Impact on Prognosis and Potential Therapy. *Sci Rep*. 2; 6:30934 (2016).

[18] TO Nielsen, HN Andrews, M Cheang, JE Kucab, FD Hsu, J Ragaz, et al. Expression of the insulin-like growth factor I receptor and urokinase plasminogen activator in breast cancer is associated with poor survival: potential for intervention with 17-allylamino geldanamycin. *Cancer Res*.1; 64(1):286–91(2004).

[19] JA Foekens, HA Peters, MP Look, H Portengen, M Schmitt, MD Kramer, et al. The urokinase system of plasminogen activation and prognosis in 2780 breast cancer patients. *Cancer Res*.1; 60 (3):636–43 (2000).

[20] M Aubele, MC Huber, N Falkenberg, E Gross, H Braselmann, AK Walch, et al. uPA receptor and its interaction partners: Impact as potential therapeutic targets in triple-negative breast cancer. *J Clin Oncol*. 1; 33:150–150 (2015).

[21] MC Huber, R Mall, H Braselmann, A Feuchtinger, S Molatore, K Lindner, et al. uPAR enhances malignant potential of triple-negative breast cancer by directly interacting with uPA and IGF1R.*BMC Cancer*.16(1):s12885-016-2663-9 (2016).

[22] Jing Wang, Li Fu, Feng Gu, Yongjie Ma. Notch1 is involved in migration and invasion of human breast cancer cells. *Oncology Reports*. 26: 1295-1303 (2011).

[23] Nagamatsu I, Onishi H, Matsushita S, Kubo M, Kai M, Imaizumi A, et al. NOTCH4 Is a Potential Therapeutic Target for Triple-negative Breast Cancer. *Anticancer Res*. 1;34 (1):69–80 (2014).

[24] Ho-Yen CM, JL Jones, S Kermorgant. The clinical and functional significance of c-Met in breast cancer: A Review. *Breast Cancer Res*. 8; 17:52 (2015).

[25] F Zagouri, Z Bago-Horvath, F Rössler, A Brandstetter, R Bartsch, CA Papadimitriou, et al. High MET expression is an adverse prognostic factor in patients with triple-negative breast cancer. *Br J Cancer*.19; 108 (5):1100–5 (2013).

[26] JR Sierra, MS Tsao. c-MET as a potential therapeutic target and biomarker in cancer. *Ther Adv Med Oncol*. S21–35 (2011).

[27] Xiaoxia Zhu and Wen Zhou. The emerging regulation of veGFR-2 in triple-negative breast cancer. *Frontiers in Endocrinology*. 6:159 (2015).

[28] S Najafi, R Mehdizadeh, E Jahanzad, A Olfatbakhsh, A Jenabian, G Esmaeeli-javid, et al. Predictive Role of EGFR, IGF-1R, VEGFR2 and HIF-1α Expression on Survival of Patients with Triple-negative Breast Cancer. *Arch Breast Cancer*. 29;1(3):86–90 (2014).

[29] S Jansson, PO Bendahl, DA Grabau, AK Falck, M Fernö, K Aaltonen, et al. The Three Receptor Tyrosine Kinases c-KIT, VEGFR2 and PDGFRα, Closely Spaced at 4q12, Show Increased Protein Expression in Triple-Negative Breast Cancer. *Plos One*. 15;9 (7):e102176 (2014).