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Paclitaxel Promotes Tumor-Infiltrating Macrophages in Breast Cancer

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Abstract

Objective: Breast cancer remains the most threatening triggers of cancer death in women. Drug resistance inevitably leads to the weakness of treatment for breast cancer. Macrophages, as one of the most abundant immune cells in tumor immune-infiltrating microenvironment, involves in cell survival, migration, and invasion of breast cancer. **Methods:** In this study, we compared the proportions of macrophages in patients with breast cancer with and without paclitaxel treatment, and investigated the targeted genes associated with macrophages for paclitaxel response. To explore the relationship between drug-related genes and breast cancer prognosis, survival analysis based on the drug-related genes were performed by website of Kaplan-Meier plotter with the threshold of significant *P* value < .05. **Results:** Compared to the normal samples, we revealed that paclitaxel significantly enhanced the ratio of macrophages in the tumor microenvironment. Furthermore, the expression of 3 drug-related genes (IFT46, PEX11A, and TMEM223) were significantly negatively associated with the proportions of macrophages. And it is worth to notice that PEX11A and TMEM223 were associated with better progression-free survival outcomes of patients with breast cancer. Moreover, PEX11A was associated with longer overall survival time of breast cancer. **Conclusion:** Taken all together, all the findings support to gain a better understanding to the development of more effective therapies targeted with paclitaxel.

Keywords

paclitaxel, breast cancer, macrophages, drug resistance, drug-related genes

Abbreviations

BC, breast cancer; EGF, epidermal growth factor; IFT46, intraflagellar transport 46; OS, overall survival; PCC, Pearson correlation coefficients; PEX11A, peroxisome biogenesis factor 11A; PFS, progression-free survival; ROS, reactive oxygen species; TMEM223, transmembrane protein 223

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Introduction

Breast cancer (BC) is the most common cancer among women worldwide, with a rising trend in aggressive neoplasm in young women.[1,2] In China, BC occupied 12.2% of all newly diagnosed and 9.6% of all cancer mortality of tumor patients, respectively.[3] Reproductive and lifestyle factors, such as age, exogenous hormone, diet, body fatness, and physical activities can influence the risk of BC.[4-6] Over the past decade, a set of novel therapeutic strategies are developed for diagnosis to treatment[3]; however, more accurate and effective treatments are still worthy of attention.

Paclitaxel, as an inhibitor of microtubule depolymerization, is an effective chemotherapeutic agent in the therapy for BC.[7] To strengthen the effect of the antibody therapy, combining paclitaxel with lumretuzumab (anti-human epidermal growth factor [EGF] receptor 3; anti-HER3) or pertuzumab (anti-human EGF receptor 2; anti-HER2) is used to treat patients with metastatic BC. Compared with paclitaxel alone treatment,

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combined solution (eg, paclitaxel plus bevacizumab) prolongs progression-free survival (PFS), but not overall survival (OS).[8] Although accelerating rates of the application of paclitaxel, the paclitaxel-evoked pain syndrome is inevitable, accompanying with paclitaxel-sensitivity and paclitaxel-resistance.[9-11] Some functional pathways and related genes have investigated to regulate the validation process in paclitaxel-sensitive or paclitaxel-resistant cells. For example, IκBα super-repressor enhanced NF-κB pathway by inhibiting NF-κB DNA binding activity and Mn-SOD expression in paclitaxel sensitivity of BC cells.[12] Upregulating of AKT2 contributes to the increase of resistance to paclitaxel in BC cells.[13]

Understanding tumor immune-infiltrating microenvironment is an important link between tumorigenesis mechanism and therapeutic solutions.[14] Many theoretical and experimental evidences have illustrated that the tumor-associated macrophages, as a major component of immune infiltrate, play a crucial role in tumor immune-infiltrating microenvironment.[15-17] For instance, macrophages improve tumor cell migration and invasion via regulating EGF and vascular endothelial growth factor.[14,18] However, there is little evidence as to how to link the paclitaxel response and paclitaxel-evoked pain syndrome with the macrophages. In this study, we acquire gene expression and clinical data from GDAC FIREHOSE to examine the proportion of macrophages in patients with BC with and without paclitaxel treatment and to capture more characteristic the targeted genes associating with macrophages on paclitaxel response in paclitaxel-treated and without paclitaxel-treated patients with BC.

Materials and Methods

Data Collection

Gene expression and clinical data were acquired from the GDAC FIREHOSE database version 2016_01_28 (http://gdac.broadinstitute.org, RRID: SCR_016213). The bioinformatics method CIBERSORT (RRID: SCR_016955)[19] was used to estimate the proportions of immune cells in the tumor microenvironment. Combining with the immunophenotypic expression profiles provided by CIBERSORT, we selected 6 immune cell types (B cells, T cell CD8, T cells CD4, macrophages, dendritic, neutrophils) as subjects to calculate the proportions in each BC sample by using the R package {"EpiDISH"}.https://labs.genetics.ucla.edu/horvath/CoexpressionNetwork/, RRID: SCR_003302).

Interactome Analysis of Targeted Gene on Paclitaxel Response

To investigate the targeted genes on paclitaxel response, the drug-related response data were obtained from Genomics of Drug Sensitivity in Cancer (GDSC) database (http://www.cancerrxgene.org/, RRID: MGI: 5909258). There are only 12 cell lines (HCC1187, HCC1599, HCC2157, HCC2218, COLO-824, DU-4475, EVSA-T, MRK-nu-1, OCUB-M, MFM-223, CAL-148, BT-474) involved in paclitaxel drug response from GDSC. Furthermore, these cell lines were classified into drug-resistance and drug-sensitivity using SL Forward-Backward methods. The brief description of procedures was as follow: (1) To randomly classify one cell line into drug-resistant cells. If fold change (FC) value of the average 50% inhibitory concentration (IC50) between the drug-resistant and drug-sensitive cell line was of > 2, it proceeded to the next step, otherwise randomly restarted. (2) To randomly capture a drug-sensitive cell line into the drug-resistant cell line, and then calculate the FC value of the IC50 between the 2 cell lines. If the FC value was greater than that in step (1), we moved to the next; otherwise, returned to the drug-sensitive cell line. (3) To repeat step (2) for screening all the drug-sensitive cell lines. (4) To randomly selected a drug-resistant cell line into sensitive cell lines, and then calculate the FC value of the IC50 between 2 cell lines. If the FC value was greater than that in step (1), it moved to the next; otherwise, returned to the drug-resistant cell line. (5) To repeat step (4) for screening all the drug-sensitive cell lines. After clarifying the drug-resistant and drug-sensitive cell lines, we screened all drug-related genes in expression profiles of these cell lines from GDSC and calculated the FC values and P values of t test. Finally, the genes with P value $<$ .05 and (FC) > 2 or FC $< 1/2$ were considering as the potential-targeted gene on paclitaxel response.

The Effect of Macrophages on Drug-Related Genes

To further evaluate the effect of macrophages on drug-related genes, Pearson correlation coefficients (PCC) between drug-related gene expression and macrophage ratio were calculated by the {cor} and {cor.test} functions in the R environment. And the genes with PCC < -0.5 or PCC > 0.5 were considered as associating with macrophages.

Prognostic Analysis of Drug-Related Genes

To explore the relationship between drug-related genes and BC prognosis, survival analysis based on the drug-related genes were performed by website of Kaplan-Meier plotter (https://kmplot.com/analysis/index.php?p=service&cancer=breast) with the statistically significant P value$<$ .05. The Kaplan-Meier plotter is an online database based on expression data and clinical data from BC, gastric cancer, lung cancer, and ovarian cancer. The patient samples were divided into 2 groups according to the median expression of the gene. Then, Kaplan-Meier survival curve was plotted to obtain the prognostic value of drug-related genes. Briefly, the drug-related genes were uploaded into the database respectively to get the Kaplan-Meier curve.

Results

Paclitaxel Upregulated Tumor-Infiltrating Macrophages

To investigate the effects of paclitaxel on the immune-infiltrating microenvironment of patients with BC, we obtained

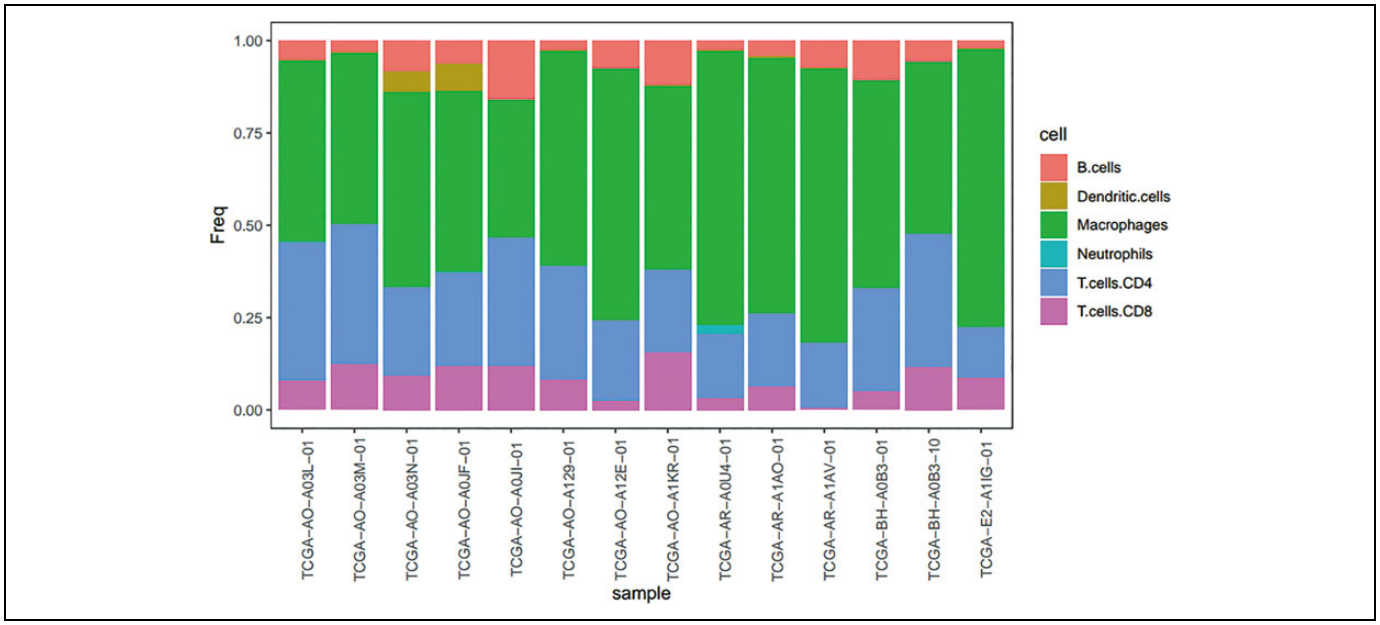

Figure 1. The frequency of 6 immune cells in BC-related tissues. All 13 BC-related tissues and control (TCGA-BH-A0B3-10) are obtained from the GDAC FIREHOSE. BC indicates breast cancer.

the frequency of 6 immune cells (Figure 1) in 13 BC-related patients who received paclitaxel in the GDAC FIREHOSE database (Table 1) and one normal control nontumor sample (TCGA-BH-A0B3-10).

To further clarify the ratio variations of immune-related cells, we randomly selected 12 adjacent nontumor samples in patients with BC without taking paclitaxel treatments to match the cohort size of 13 tumor samples. The proportion of macrophages was upregulated in paclitaxel-treated patients with BC. The detailed results showed that mean proportion in patients was 0.6168 but in normal samples was 0.4957 (Table 2, Figure 2A). On the other hand, the mean proportion of other immune cell types was not changed so much, such as B cells in patients was 0.0609 and in normal samples was 0.0670; T cells CD4 even reduced, in patients it was 0.2318 but in normal samples it was 0.2555. Besides, we randomly selected 50 BC tissues and 50 adjacent nontumor samples in patients with BC without paclitaxel-treatment. No obvious difference was detected (Figure 2B). As a consequence, we may deduce that paclitaxel-induced macrophage infiltration in the tumor microenvironment of BC. The results showed that paclitaxel upregulated the proportion of macrophages, compared to the control.

Potential Drug-Related Genes

According to gene expression profiles between cell lines of drug-resistant (HCC2157, HCC2218, COLO-824, DU-4475, EVSA-T, MFM-223, CAL-148, BT-474) and drug-sensitive (HCC1599, HCC1187, MRK-nu-1, OCUB-M) (Table 3), we screened 62 potential drug-related genes. Further, the correlation between gene expression level and the ratio of macrophages within each sample allow us to obtain drug macrophages–related genes. Three targeted genes were found, including intraflagellar transport 46 (IFT46), peroxisome biogenesis factor 11A (PEX11A), and transmembrane protein 223 (TMEM223). The expression of these genes were significantly negatively associated with the proportions of macrophages ($\gamma_{IFT46} = -0.604$; $\gamma_{PEX11A} = -0.698$; $\gamma_{TMEM223} = -0.616$; Figure 3).

Prognostic Analysis of Drug-Related Genes

Kaplan-Meier plotter was performed to explore the relationship between drug-related genes and BC prognosis. The results revealed that PEX11A and TMEM223 were associated with longer PFS of patients with BC, and the log-rank test *P* value were 3.3e-08 and 1.8e-07. Peroxisome biogenesis factor 11A was associated with longer OS of BC and the log-rank test *P* value was .002. However, the gene IFT46 had not been found in Kaplan-Meier plotter online database (Figure 4).

Discussion

Paclitaxel is a frontline antineoplastic agent widely applied in the treatment of BC, ovarian cancer, gastric cancer, and lung cancer.[20-23] In this study, compared with nonpaclitaxel-treated patients with BC, paclitaxel can upregulate the proportion of macrophages in the tumor-infiltrating cells of tumor microenvironment in paclitaxel-treated patients with BC. Similar results have shown that paclitaxel may restore tumor-induced macrophages production of proimmune factors (eg, immunostimulatory cytokine IL-12) in tumor-bearing host.[24] Low-dose paclitaxel suppresses the M2 macrophages by acting on Toll-like receptor 4, but induces the M1 macrophages in gastric cancer.[22]

Table 1. Details of Patients With BC With Paclitaxel.

Patients	Stage	T	M	N	Age	Gender	Used drug
TCGA-BH-A0B3	Stage IIb	T 2	M 0	N 1a	53	Female	paclitaxel; cyclophosphamide; doxorubicin
TCGA-AO-A03L	Stage IIIa	T 3	M 0	N 2	34	Female	paclitaxel; tamoxifen; doxorubicin; lupron; cyclophosphamide
TCGA-AO-A03M	Stage I	T 1c	M 0	N 0 (i-)	29	Female	paclitaxel; tamoxifen; cyclophosphamide; doxorubicin
TCGA-AO-A03N	Stage Ib	T 2	M 0	N 1a	59	Female	tesetaxel; gemcitabine; vinorelbine; xgeva; paclitaxel; cyclophosphamide; femara; aromasin; arimidex; tamoxifen; fulvestrant; doxorubicin
TCGA-AO-A0JF	Stage IIa	T 1c	M 0	N 1a	68	Female	paclitaxel; doxorubicin; cyclophosphamide; letrozole (femara); anastrozole (arimidex)
TCGA-AO-A0JI	Stage IIa	T 1c	M 0	N 1	56	Female	paclitaxel; arimidex (anastrozole); cyclophosphamide; doxorubicin
TCGA-AO-A129	Stage IIb	T 2	M 0	N 1a	29	Female	paclitaxel; doxorubicin; cyclophosphamide
TCGA-AO-A12E	Stage IIb	T 3	M 0	N 0 (i+)	51	Female	paclitaxel; cyclophosphamide; doxorubicin; tamoxifen
TCGA-AO-A1KR	Stage IIa	T 2	M 0	N 0	51	Female	paclitaxel; cyclophosphamide; doxorubicin
TCGA-AR-A0U4	Stage IIa	T 2	M 0	N 0	54	Female	paclitaxel; cytoxan; doxorubicin
TCGA-AR-A1AO	Stage IIa	T 1	M 0	N 1	47	Female	paclitaxel; cytoxan; doxorubicin; tamoxifen
TCGA-AR-A1AV	Stage IIb	T 2	M 0	N 1	68	Male	paclitaxel; doxorubicin; cytoxan; tamoxifen
TCGA-E2-A1IG	Stage IIb	T 2	M 0	N 1mi	45	Female	paclitaxel; tamoxifen; cyclophosphamide; letrozole; doxorubicin

Abbreviation: BC, breast cancer.

Table 2. Details of Microenvironment Between BC Patients' Tissues With Paclitaxel and Normal Samples Without Paclitaxel.

Immune cell types	Mean proportions in patients	Mean proportions in normal samples
B cells	0.0609	0.0620
T cells CD8	0.0894	0.0803
T cells CD4	0.2318	0.2555
Macrophages	0.6169	0.4957
Dendritic cells	0.0012	0.0105

Abbreviation: BC, breast cancer.

However, paclitaxel-induced peripheral sensory neuropathy is well established and is a serious problem that requires chemotherapy in combination with paclitaxel.[9,25] Previous study have shown that the paclitaxel-induced mechanical allodynia is due to the increase of macrophages induced by high-dose paclitaxel, and macrophages infiltrate the dorsal root ganglion.[25] The paclitaxel-induced mechanical allodynia by minocycline is alleviated via inhibiting the loss of intraepidermal nerve fiber.[25] Recent studies have shown that the Toll-like receptor signaling (eg, TLR4) and Nod-like receptor signaling (eg, NOD2) may involve in the development of neuropathic pain.[26,27] Moreover, pathological pain may also be attributed to the release of pro-inflammatory cytokines (eg, some sensitizing primary afferent neurons).[28,29] On the other hand, increased tumor-infiltrating macrophages can enhances the response to chemotherapy and inhibit tumor-initiating cells.[30] As a neoadjuvant paclitaxel chemotherapy, paclitaxel can increase the proportion of macrophages which may lead to either enhance the antitumor immune response or induce paclitaxel-evoked pain syndrome. Furthermore, better understanding of more trait-related genes will help improve the quality of life and survival of patients with BC with paclitaxel chemotherapy.

The 3 drug-related genes (IFT46, PEX11A, TMEM223) screened were significantly negatively correlated with macrophages in BC. Besides, PEX11A and TMEM223 were associated with longer PFS of patients with BC. Peroxisome biogenesis factor 11A is associated with prolonged OS in BC. The intraflagellar transport (IFT) system has been well established to connect kinesin motor protein (linked by IFTB) and dynein motor protein (linked by IFTA).[31] The transmembrane transport protein IFT46 is a member of IFTB complex and is involved in the biosynthesis in cell organelles, and the maintenance and transport of IFT proteins.[31,32] However, few studies have investigated the relationship between IFTB complex and tumor-infiltrating macrophages. This is the first study that IFT46 has been linked to paclitaxel-responsive macrophages of BC. Peroxisomes are highly involved in cellular functions (eg, reactive oxygen species [ROS] metabolism).[33] Peroxisome biogenesis factor 11A is a factor of peroxisomes, which plays a key role in the regulation of peroxisome proliferator-activated receptor alpha in ROS metabolism and lipid metabolism.[33,34] Reactive oxygen species is involved in regulating the occurrence of tumor-associated macrophages and alternatively activating macrophages (M2) differentiation.[35] Therefore, PEX11A may regulate M2 macrophages via ROS metabolism under the combination of chemotherapy and paclitaxel. Transmembrane protein 223 as a transmembrane protein, we assumed that it may be involved in the phagocytosis of macrophages.[36,37] However, there is not enough experimental evidence or literature to evaluate the response of drug-related genes to macrophages in the tumor microenvironment.

In summary, our preliminary evidence suggested that paclitaxel can increase the proportion of tumor-infiltrating macrophages in BC, which may mediate the regulation of drug-related genes associated with macrophages. This study provides a potential basis of alleviating paclitaxel-induced peripheral sensory neuropathy for chemotherapy.

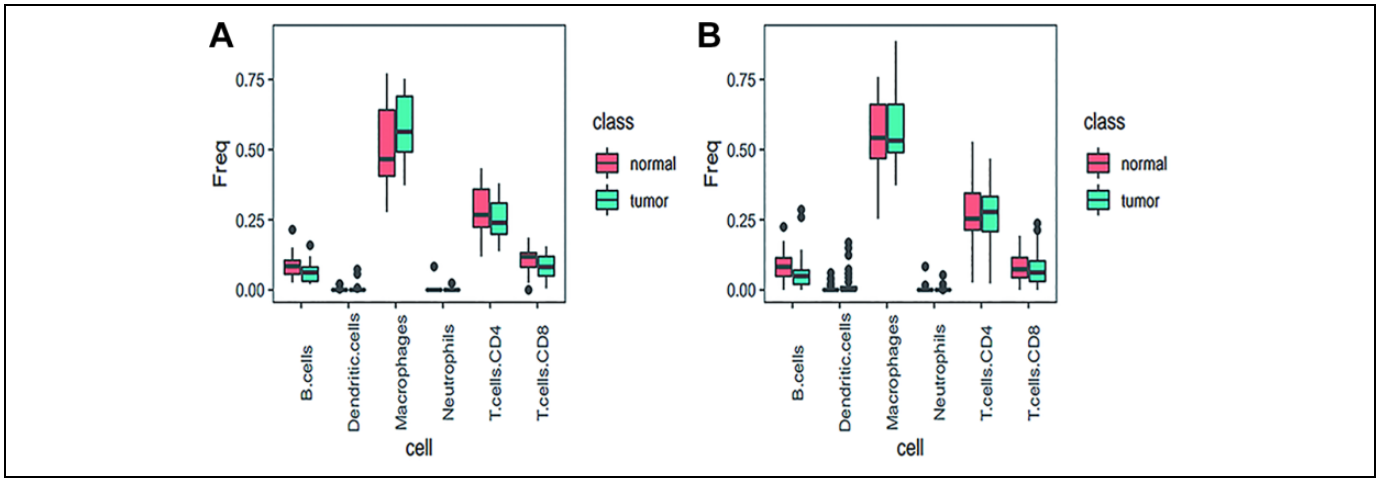

Figure 2. The effect of paclitaxel treatment on immune cells. A, Comparison of breast cancer tissue and adjacent nontumor samples in patients with BC with paclitaxel treatments. B, Comparison of breast cancer tissue and adjacent nontumor samples in patients with BC without paclitaxel treatments.

Table 3. Details of GDAC Cell Lines.

Class	Cell lines
Resistant	HCC2157, HCC2218, COLO-824, DU-4475, EVSA-T, MFM-223, CAL-148, BT-474
Sensitive	HCC1599, HCC1187, MRK-nu-1, OCUB-M

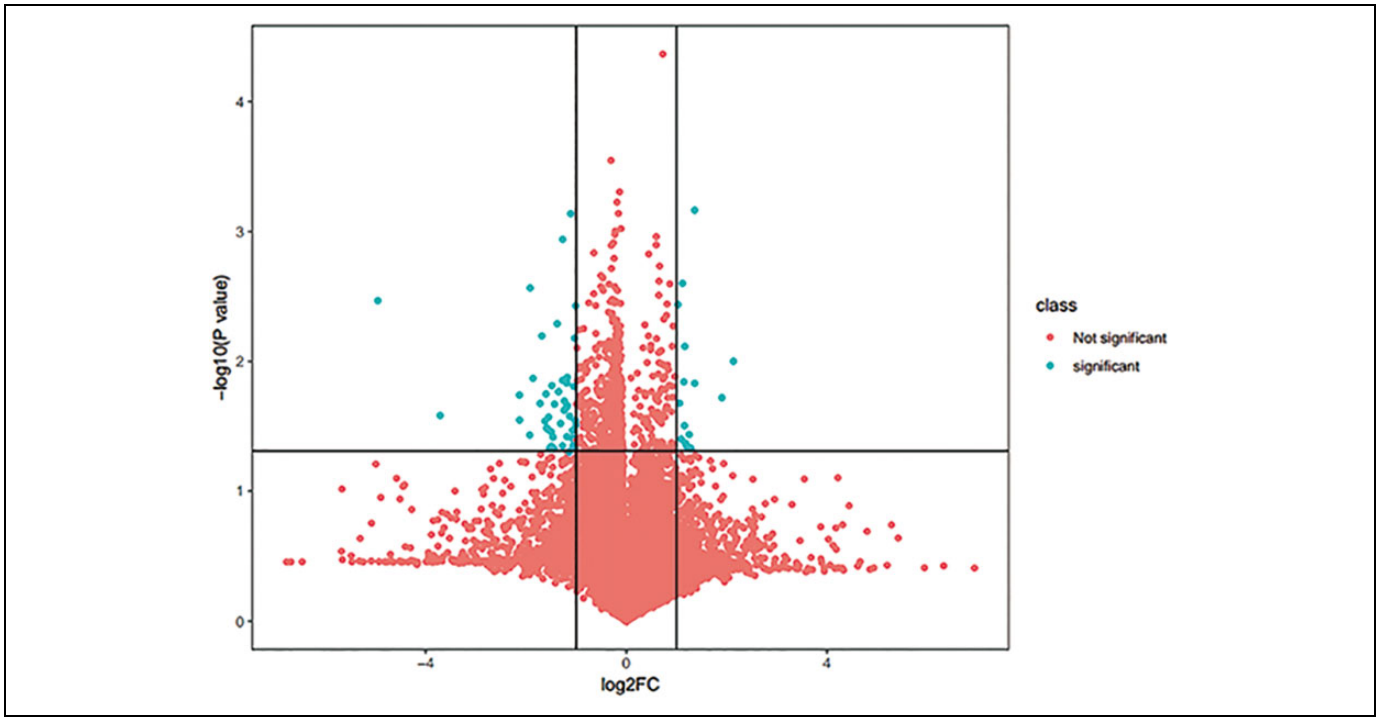

Figure 3. Potential targeting genes on paclitaxel response. Three targeted genes were found, including intraflagellar transport 46 (IFT46), peroxisome biogenesis factor 11A (PEX11A), and transmembrane protein 223 (TMEM223). The expression of these genes were significantly negatively associated with the proportions of macrophages ($\gamma_{IFT46} = -0.604$; $\gamma_{PEX11A} = -0.698$; $\gamma_{TMEM223} = -0.616$).

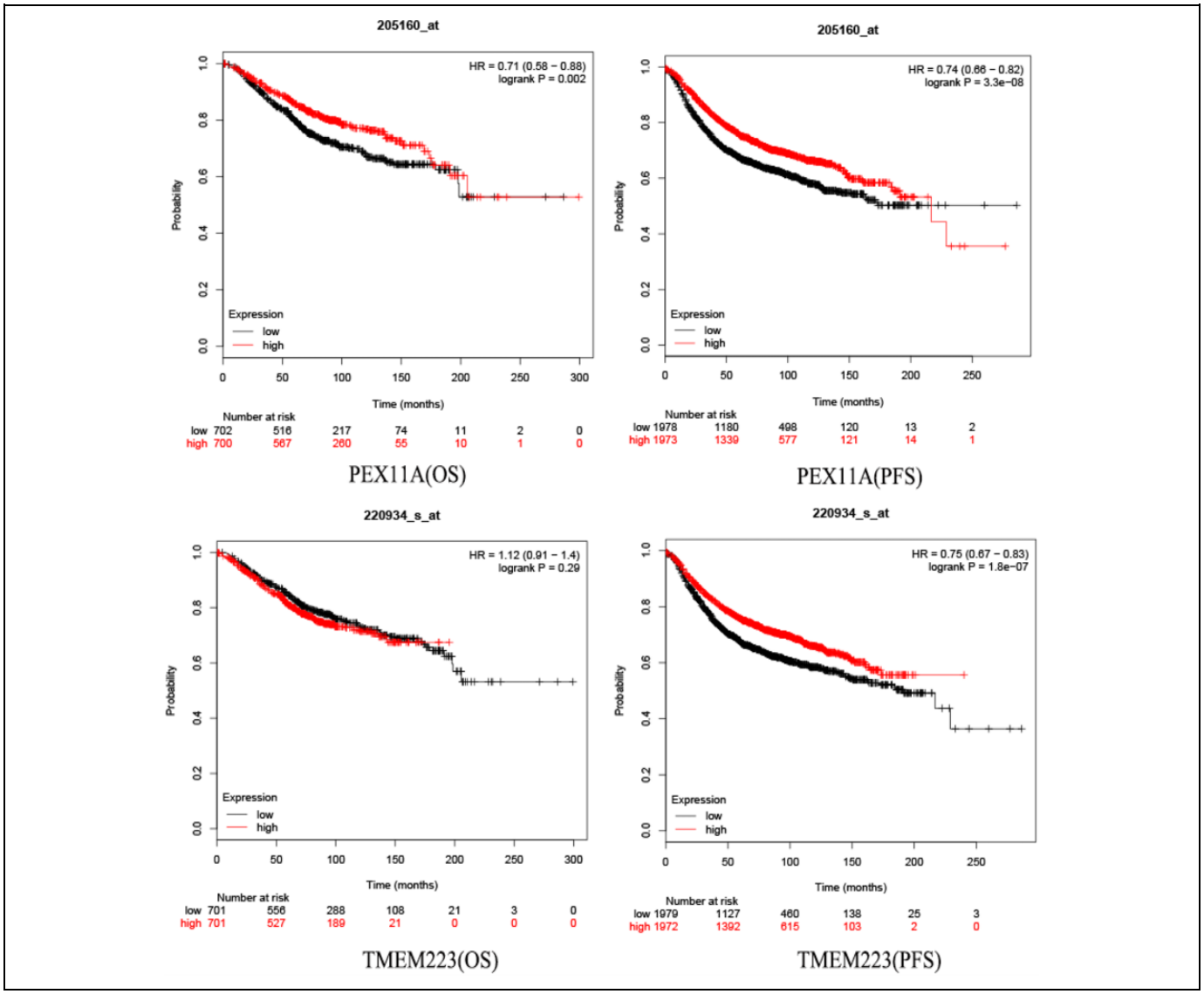

Figure 4. Prognostic analysis of drug-related genes. Kaplan-Meier plotter database prognostic analysis of PEX11A, TMEM223 (overall survival [OS]; progression-free survival [PFS]). PEX11A indicates peroxisome biogenesis factor 11A; TMEM223, transmembrane protein 223.

Authors' Note

Our study did not need approval because gene expression and clinical data were acquired from the GDAC FIREHOSE v2016_01_28 database (http://gdac.broadinstitute.org, RRID: SCR_016213).

Declaration of Conflicting Interests

The author(s) declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

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References

1. Oh H, Eliassen AH, Beck AH, et al. Breast cancer risk factors in relation to estrogen receptor, progesterone receptor, insulin-like growth factor-1 receptor, and Ki67 expression in normal breast tissue. *NPJ Breast Cancer*. 2017;3:39.
2. Coughlin SS. Epidemiology of Breast Cancer in Women. *Adv Exp Med Biol*. 2019;1152:9-29.
3. Wu C, Du S, Zhang J, Liang AL, Liu YJ. Exosomes and breast cancer: a comprehensive review of novel therapeutic strategies from diagnosis to treatment. *Cancer Gene Ther*. 2017;24(1):6-12.
4. Collaborative Group on Hormonal Factors in Breast Cancer. Breast cancer and hormone replacement therapy: collaborative

reanalysis of data from 51 epidemiological studies of 52 705 women with breast cancer and 108 411 women without breast cancer. *Lancet*. 1997;350(9084):1047-1059.
5. Kelsey JL, Gammon MD, John EM. Reproductive factors and breast cancer. *Epidemiol Rev*. 1993;15(1):36.
6. Romieu II, Amadou A, Chajes V. The role of diet, physical activity, body fatness, and breastfeeding in breast cancer in young women: epidemiological evidence. *Rev Invest Clín*. 2017;69(4): 193-203.
7. Galons JP, Altbach MI, Paine-Murrieta GD, Taylor CW, Gillies RJ. Early increases in breast tumor xenograft water mobility in response to paclitaxel therapy detected by non-invasive diffusion magnetic resonance imaging. *Neoplasia*. 1999;1(2):113-117.
8. Miller K, Wang M, Gralow J, et al. Paclitaxel plus bevacizumab versus paclitaxel alone for metastatic breast cancer. *N Engl J Med*. 2007;357(26):2666-2676.
9. Peters CM, Jimenez-Andrade JM, Jonas BM, et al. Intravenous paclitaxel administration in the rat induces a peripheral sensory neuropathy characterized by macrophage infiltration and injury to sensory neurons and their supporting cells. *Exp Neurol*. 2007; 203(1):42-54.
10. Rouzier R, Rajan R, Wagner P, et al. Microtubule-associated protein tau: a marker of paclitaxel sensitivity in breast cancer. *Proc Natl Acad Sci U S A*. 2005;102(23):8315-8320.
11. Gonzalez-Angulo AM, Morales-Vasquez F, Hortobagyi GN. Overview of resistance to systemic therapy in patients with breast cancer. *Adv Exp Med Biol*. 2007;608:1-22.
12. Patel NM, Nozaki S, Shortle NH, et al. Paclitaxel sensitivity of breast cancer cells with constitutively active NF-κB is enhanced by IκBα super-repressor and parthenolide. *Oncogene*. 2000; 19(36):4159-4169.
13. Cheng GZ, Chan J, Wang Q, Zhang W, Sun CD, Wang LH. Twist transcriptionally up-regulates AKT2 in breast cancer cells leading to increased migration, invasion, and resistance to paclitaxel. *Cancer Res*. 2007;67(5):1979-1987.
14. Pollard JW. Macrophages define the invasive microenvironment in breast cancer. *J Leukoc Biol*. 2008;84(3):623-630.
15. Cassetta L, Pollard JW. Repolarizing macrophages improves breast cancer therapy. *Cell Res*. 2017;27(8):963-964.
16. Lin EY, Pollard JW. Tumor-associated macrophages press the angiogenic switch in breast cancer. *Cancer Res*. 2007;67(11): 5064-5066.
17. Leek RD, Harris AL. Tumor-associated macrophages in breast cancer. *J Mammary Gland Biol Neoplasia*. 2002;7(2):177-189.
18. Lin EY, Li JF, Gnatovskiy L, et al. Macrophages regulate the angiogenic switch in a mouse model of breast cancer. *Cancer Res*. 2006;66(23):11238-11246.
19. Gentles AJ, Newman AM, Liu CL, et al. The prognostic landscape of genes and infiltrating immune cells across human cancers. *Nature Med*. 2015;21(8):938-945.
20. Ganz PA, Romond EH, Cecchini RS, et al. Long-term follow-up of cardiac function and quality of life for patients in NSABP protocol B-31/NRG oncology: a randomized trial comparing the safety and efficacy of doxorubicin and cyclophosphamide (AC) followed by paclitaxel with AC followed by paclitaxel and trastuzumab in patients with node-positive breast cancer with tumors overexpressing human epidermal growth factor receptor 2. *J Clin Oncol*. 2017;35(35):3942-3948.
21. Chan JK, Brady MF, Penson RT, et al. Weekly vs. every-3-week paclitaxel and carboplatin for ovarian cancer. *N Engl J Med*. 2016;374(8):738-748.
22. Yamaguchi T, Fushida S, Yamamoto Y, et al. Low-dose paclitaxel suppresses the induction of M2 macrophages in gastric cancer. *Oncol Rep*. 2017;37(6):3341-3350.
23. Bradley JD, Paulus R, Komaki R, et al. Standard-dose versus high-dose conformal radiotherapy with concurrent and consolidation carboplatin plus paclitaxel with or without cetuximab for patients with stage IIIA or IIIB non-small-cell lung cancer (RTOG 0617): a randomised, two-by-two factorial phase 3 study. *Lancet Oncol*. 2015;16(2):187-199.
24. Mullins DW, Burger CJ, Elgert KD. Paclitaxel enhances macrophage IL-12 production in tumor-bearing hosts through nitric oxide. *J Immunol*. 1999;162(11):6811-6818.
25. Liu CC, Lu N, Cui Y, et al. Prevention of paclitaxel-induced allodynia by minocycline: effect on loss of peripheral nerve fibers and infiltration of macrophages in rats. *Mol Pain*. 2010; 6:76.
26. Zhang H, Li Y, de Carvalho-Barbosa M, et al. Dorsal root ganglion infiltration by macrophages contributes to paclitaxel chemotherapy-induced peripheral neuropathy. *J Pain*. 2016; 17(7):775-786.
27. Santa-Cecília FV, Ferreira DW, Guimaraes RM, et al. The NOD2 signaling in peripheral macrophages contributes to neuropathic pain development. *Pain*. 2019;160(1):102-116.
28. Ma W, Eisenach J. Cyclooxygenase 2 in infiltrating inflammatory cells in injured nerve is universally up-regulated following various types of peripheral nerve injury. *Neuroscience*. 2003;121(3): 691-704.
29. Cui JG, Holmin S, Mathiesen T, Meyerson BA, Linderoth B. Possible role of inflammatory mediators in tactile hypersensitivity in rat models of mononeuropathy. *Pain*. 2000;88(3):239-248.
30. Mitchem JB, Brennan DJ, Knolhoff BL, et al. Targeting tumor-infiltrating macrophages decreases tumor-initiating cells, relieves immunosuppression, and improves chemotherapeutic responses. *Cancer Res*. 2013;73(3):1128-1141.
31. Shi L, Shi X, Shen Y. Intraflagellar transport 46 (IFT46) is essential for trafficking IFT proteins between cilia and cytoplasm in paramecium. *Sci Rep*. 2018;8(1):9259.
32. Ren H, Dong B, Fan Z, Meng D. Prokaryotic expression and purification of *Chlamydomonas reinhardtii* intraflagellar transport protein 46 (IFT46) and preparation of polyclonal antibody. *Sheng Wu Gong Cheng Xue Bao*. 2016;32(8):1124-1132.
33. Rodriguez-Serrano M, Romero-Puertas MC, Sanz-Fernandez M, Hu J, Sandalio LM. Peroxisomes extend peroxules in a fast response to stress via a reactive oxygen species-mediated induction of peroxin PEX11a. *Plant Physiol*. 2016; 171(3):1665-1674.
34. Anderson SP, Howroyd P, Liu J, et al. The transcriptional response to a peroxisome proliferator-activated receptor alpha (PPAR alpha) agonist includes increased expression of

proteome maintenance genes. *J Biol Chem*. 2004;279(50): 52390-52398.

35. Zhang Y, Choksi S, Chen K, Pobezinskaya Y, Linnoila I, Liu ZG. ROS play a critical role in the differentiation of alternatively activated macrophages and the occurrence of tumor-associated macrophages. *Cell Res*. 2013;23(7):898-914.
36. Liu D, Wang R, Grant AR, et al. Immune adaptation to chronic intense exercise training: new microarray evidence. *BMC Genomics*. 2017;18(1):29.
37. Zou F, Wang X, Han X, et al. Expression and function of tetraspanins and their interacting partners in B cells. *Front Immunol*. 2018;9:1606.