Original Article

Efficacy of platinum in advanced triple-negative breast cancer with germline *BRCA* mutation determined by next generation sequencing

Nan Wang[1*], Kun Li[1*], Wenfa Huang[1*], Weiyao Kong[1], Xiaoran Liu[1], Weijie Shi[2], Feng Xie[2], Hanfang Jiang[1], Guohong Song[1], Lijun Di[1], Quanren Wang[3], Jianjun Yu[2], Huiping Li[1]

[1]Key Laboratory of Carcinogenesis and Translational Research (Ministry of Education/Beijing), Department of Breast Oncology, Peking University Cancer Hospital and Institute, Beijing 100142, China; [2]Huidu Shanghai Medical Sciences, Shanghai 201499, China; [3]Jiangsu Hengrui Medicine, Shanghai 200120, China

*These authors contributed equally to this work.

Correspondence to: Huiping Li. Key Laboratory of Carcinogenesis and Translational Research (Ministry of Education/Beijing), Department of Breast Oncology, Peking University Cancer Hospital and Institute, Beijing 100142, China. Email: huipingli2012@hotmail.com.

Abstract

Objective: To compare the efficacy of platinum- and non-platinum-based regimens as first-line treatment for advanced triple-negative breast cancer (TNBC) and analyze the relationship between their efficacy and *BRCA* gene status.

Methods: Retrospectively analyze clinical data of 220 patients diagnosed pathologically with advanced TNBC and treated at the Department of Breast Oncology, Peking University Cancer Hospital from 2013 to 2018 and evaluate the efficacy of chemotherapy. A total of 114 patients had *BRCA1/2* gene tested by next generation sequencing (NGS) using peripheral blood, and we analyzed the correlation between their efficacy and *BRCA1/2* gene status.

Results: Non-platinum-based chemotherapy (NPCT) was administered to 129 and platinum-based chemotherapy (PBCT) to 91 study patients. The clinical benefit rate (CBR) and median progression-free survival (PFS) were not statistically different between NPCT and PBCT groups. The median overall survival (OS) was 30.0 and 22.5 months for PBCT and NPCT group, respectively [P=0.090, hazard ratios (HR)=0.703]. *BRCA* status was assessed in 114 patients, 14 of whom had deleterious germline *BRCA1/2* (*gBRCA*) mutations (seven in each group). In PBCT group, the CBR was 85.7% and 35.1% for patients with and without deleterious *gBRCA* mutations, respectively (P=0.039). The median PFS were 14.9 and 5.3 months and median OS were 26.5 and 15.5 months for patients with and without deleterious *gBRCA* mutations, respectively (P=0.001, P=0.161, respectively). Patients in PBCT group had significantly greater rates of grade 3–4 anemia (5.5% *vs*. 0%) and thrombocytopenia (8.8% *vs*. 0%), whereas palmar-plantar erythrodysesthesia (12.4% *vs*. 0%) and peripheral neuropathy (8.6% *vs*. 1.1%) occurred more frequently in NPCT group.

Conclusions: Platinum-based regimens are more effective in patients with deleterious *gBRCA* mutations, but no difference in patients without *BRCA* gene mutations, so non-platinum is an option in patients without *BRCA* gene mutations considering the toxicity and side effect. And we recommend that patients with advanced TNBC should have *BRCA* gene test.

Keywords: Advanced breast cancer; triple negative; *BRCA* mutation; next-generation sequencing; platinum; efficacy

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Introduction

Triple-negative breast cancer (TNBC) is negative for expression of estrogen receptor (ER), progesterone receptor (PR) and human epidermal growth factor receptor 2 (HER2), and accounts for an estimated 15% of breast cancers (1). It is associated with a poor clinical outcome and high relapse rate (2). Because of the absence of hormonal receptors and HER2, chemotherapy is the mainstay of treatment of TNBC (3,4). TNBC shows a higher prevalence of *BRCA*1/2 mutations than other subtypes of breast cancer. It has been reported that 11%–20% of patients with TNBC carry germline *BRCA1/2* (*gBRCA*) mutations (3,5,6). *BRCA1* and *BRCA2* are tumor suppressor genes (7). They play an important role in homologous recombination (HR) repair, which is responsible for repairing interstrand crosslinks (ICL) and double strand breaks (DSB) (8,9). Mutations in *BRCA1/2* genes result in genome instability and lead to development of malignancy (9,10). Individuals harboring *BRCA* mutations have an increased risk of developing breast cancer, which is often of triple negative phenotype. TNBC accounts for 70% of breast cancers with *BRCA1* mutations and 16%–23% of those with *BRCA2* mutations (11). While most patients with sporadic TNBC do not have *BRCA1* mutations, evidence exists of *BRCA1* pathway dysfunction in these tumors, a state defined as BRCAness (12,13). The mechanism of platinum-based therapy, which is an effective treatment for TNBC (14), is the generation of both intrastrand crosslinks and ICL, which inhibit DNA replication and transcription and induce DSB, eventually leading to cell death (8,14,15). Several studies have investigated the role of PBCT in metastatic TNBC; however, their results are conflicting. The CBCSG006 trial (16) showed that cisplatin plus gemcitabine is superior to paclitaxel plus gemcitabine as first-line therapy for patients with metastatic TNBC. However, according to the TNT trial (17), carboplatin is not more active than docetaxel in an unselected cohort, being more active than docetaxel only in patients with germline-mutated *BRCA1/2* breast cancer. Moreover, the use of platinum-based regimens is limited because of their adverse effects and drug resistance associated with DNA damage repair (DDR) (8,14,18). Adverse effects of cisplatin, such as nephrotoxicity, neurotoxicity, and ototoxicity and of carboplatin, namely myelosuppression, limit their therapeutic effect of prolonging longevity (18). Because of DDR, tumors show intrinsic or acquired drug resistance to platinum-based regimens (8). To improve efficacy and decrease unnecessary use of platinum drugs, we retrospectively analyzed the clinical data of 220 patients with advanced breast cancer who had pathologically confirmed TNBC and were treated at the Department of Breast Oncology, Peking University Cancer Hospital from January 2013 to October 2018. We compared the efficacy of platinum- and non-platinum-based first-line therapy for advanced TNBC and analyzed the factors affecting the efficacy of these regimens.

Materials and methods

Patients

From January 2013 to October 2018, 3,367 patients diagnosed with breast cancer were treated at the Department of Breast Oncology, Peking University Cancer Hospital, 416 of whom (12.4%) were diagnosed with TNBC pathologically, 265 of whom having advanced stage disease. The inclusion criteria were as follows: 1) pathological diagnosis of advanced TNBC; 2) Eastern Cooperative Oncology Group (ECOG) score ≤2; 3) received at least two cycles of treatment or underwent at least one response evaluation; and 4) had measurable lesions. Patients with incomplete clinical data (n=45) were excluded, leaving 220 patients for analysis. The screening process is showed in *Figure 1*. Their clinical data were collected from medical records. Informed consent was obtained from all patients before commencement of treatment.

Evaluation

Efficacy was analyzed according to clinical benefit rate (CBR), progression-free survival (PFS) and overall survival (OS). Tumor response was evaluated according to the Response Evaluation Criteria in Solid Tumors (RECIST) version 1.1 by computed tomography scan or magnetic resonance imaging. Evaluations were performed every 6–12 weeks or whenever the patient's condition changed. CBR was defined as the proportion of patients who achieved CR, PR, or SD for at least 24 weeks. PFS was defined as the time from starting treatment to identification of disease progression or death from any cause. OS was defined as the

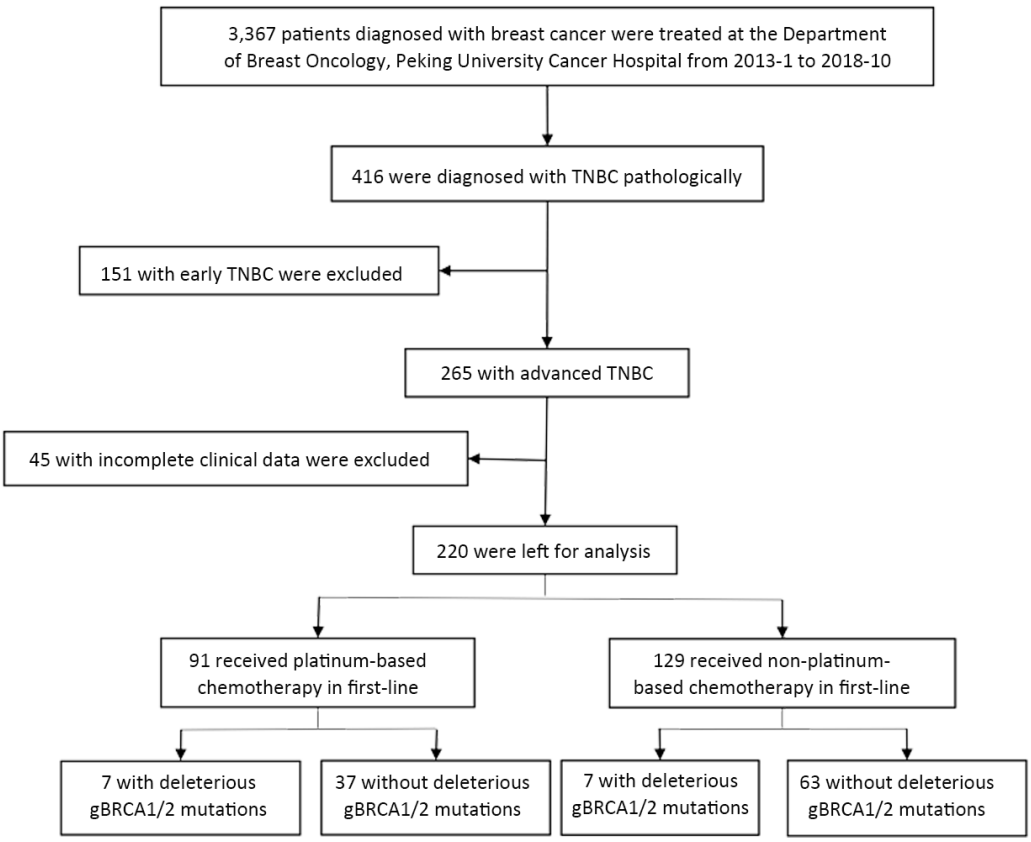

Figure 1 Screening process.

time from starting first-line treatment to death from any cause. Patients who survived without progression, died from any cause or were lost from follow-up were censored at the date of last follow-up (31 October 2018) or of their last contact. Adverse events were graded according to the National Cancer Institute Common Terminology Criteria for Adverse Events (CTCAE) version 4.0.

Follow-up

We followed up by regular inpatient, outpatient or telephone every 8–12 weeks. Every follow-up period, the clinicians would record the result of computed tomography scan or magnetic resonance imaging as well as the adverse events of treatment. The last follow-up date was 31 October 2018.

DNA extraction

Peripheral blood was collected in ethylene diamine tetraacetic acid (EDTA) Vacutainer tubes and processed within 3 h. Genomic DNA was extracted from peripheral blood lymphocytes (PBLs) by using a DNeasy Blood and Tissue Kit (Qiagen, Hilden, Germany) according to the manufacturer's instructions.

Next-generation sequencing (NGS)

Sequence of *BRCA1/2* gene has been enriched and sequenced by high throughput platform. All exons, 20 base pairs proximal to the 5' end and 10 base pairs distal to the 3' end of each exon were analyzed. Detected variations included single point mutations and small indels. Clinically important (pathogenic or likely pathogenic) mutations identified by the high throughput DNA sequencing method were verified by Sanger DNA sequencing analysis.

The variants were classified into the following five categories according to American College of Medical Genetics and Genomics (ACMG) Standards and Guidelines: pathogenic, likely pathogenic, benign, likely benign, and uncertain significance (19). In this study, pathogenic and likely pathogenic mutations were treated as deleterious *gBRCA* mutations.

Statistical analysis

Statistical analyses were carried out using IBM SPSS Statistics (Version 22.0; IBM Corp., New York, USA). Categorical data are presented as numbers and percentages and continuous data as medians and ranges. Pearson's χ^2 or Fisher's exact tests were used for comparison of categorical variables. PFS and OS were estimated using the Kaplan-Meier method and compared using the log-rank test. A multivariate Cox regression model was also performed to compute hazard ratios (HR) and 95% confidence interval (95% CI) and adjusting for prognostic variables. All P values were two sided and P<0.05 was considered to denote statistical significance.

Results

Patient characteristics

Non-platinum-based chemotherapy (NPCT) was administered to 129 (58.6%) of the 220 patients and platinum-based chemotherapy (PBCT) to the remaining 91 (41.4%) as first-line therapy. Their baseline characteristics are presented in *Table 1*. The overall median age was 49 (range, 21–75) years old, being 51 (25–75) years old in the NPCT and 46 (21–66) years old in the PBCT group. The median disease-free survival (DFS) was 19.9 (95% CI: 16.4–23.4) months. Patients in the PBCT group were younger at onset and had shorter DFS than those in the NPCT group (P=0.002). The median DFS was 24.3 (0–192.8 months, 95% CI: 20.6–28.0) months in the NPCT group and 14.3 (0–144.6 months, 95% CI: 11.9–16.7) months in the PBCT group (P=0.002) (*Figure 2A*). However, there were no significant differences in other baseline characteristics between these two groups.

BRCA gene detection

BRCA1/2 gene testing was performed on 35 patients in Beijing Genomics Institution (BGI) clinical laboratories, on 33 patients in Peking University Cancer Hospital, on 61 patients in Huidu Shanghai clinical laboratory, and on 13 patients in other centers (some patients were tested more than once). The results are summarized in *Table 2*. *BRCA1/2* gene testing was performed at least twice in different centers in 25 of them and the concordance was 100% (*Table 3*). Thus, we analyzed CBR, PFS and OS using a combination of all these results.

Signature of BRCA mutation

In all, 114 patients underwent *BRCA1/2* gene testing, 14 (12.3%) of whom were found to have deleterious *gBRCA* mutations, seven in the NPCT and seven in the PBCT group. Nine had mutations of uncertain significance, the rest being (likely) benign mutations or wild type. These findings are presented in *Table 4* and the mutation sites in *Figure 3*. The PFS of patients with deleterious *gBRCA* mutations is compared with that (6.4 months) of all 220 patients in *Figure 4*.

In the NPCT group, patients with *BRCA2* mutation sites in c.4240delA and c.6628G>T had longer PFS. When these two patients were excluded, the PFS was 3.7 (95% CI: 1.4–6.0) months for patients with deleterious *gBRCA* mutations in the NPCT group; this does not differ significantly from the PFS (5.1 months) of patients without such mutations (P=0.220). The OS for the five patients with *gBRCA* mutations in the NPCT group could not be calculated because there were too few data.

Response and survival

The median follow-up time after recurrence or metastasis was 14.3 (range, 1.7–97.0) months. Overall, the CBR was 48.1% (62/129) for the NPCT and 51.6% (47/91) for the PBCT group; this difference is not significant (P=0.600). There was also no significant difference in median PFS (6.0 months, 95% CI: 4.6–7.4 for the NPCT and 6.6 months, 95% CI: 5.1–8.1 for the PBCT group) (P=0.907) (*Figure 2B*). CBR and median PFS did not differ significantly in these two groups within the subgroups of age of onset (≤50 years *vs.* >50 years), DFS (≤24 months *vs.* >24 months), and visceral metastases. CBR and median PFS were also analyzed according to site of metastases. The PBCT group tended to have better outcomes in patients with liver and chest wall metastases; however, neither difference was statistically significant (*Figure 2C*). Only eight patients developed brain metastases, two of whom had received PBCT. There were too few patients with brain metastases to compare CBR and PFS between the groups. CBR and median PFS are shown in *Table 5*. The median OS was around 7.5 months longer in the PBCT (30.0 months, 95% CI: 17.5–42.4) than NPCT group (22.5 months, 95% CI: 14.4–30.7); this difference was not statistically significant (P=0.090, HR=0.703, 95% CI: 0.466–1.059) (*Figure 2D*).

Additionally, subgroup analyses were performed. In the PBCT group, the CBR was 85.7% (6/7) for patients with deleterious *gBRCA* mutations and 35.1% (13/37) for those

Table 1 Baseline characteristics of patients according to type of first-line therapy

Characteristics	NPCT (n=129) [n (%)]	PBCT (n=91) [n (%)]	P
Age (year)			
Median (range)	51 (25–75)	46 (21–66)	0.001
≤50	64 (49.6)	62 (68.1)	0.006
>50	65 (50.4)	29 (31.9)	
Family history			
Breast/ovarian cancer	12 (9.3)	9 (9.9)	0.416
Other cancers*	16 (12.4)	17 (18.7)	
No	101 (78.3)	65 (71.4)	
Histology of primary tumor			
Invasive ductal carcinoma	113 (87.6)	79 (86.8)	0.063
Invasive lobular carcinoma	8 (6.2)	1 (1.1)	
Others	8 (6.2)	11 (12.1)	
Diagnosed triple negative			
Primary tumor	111 (86.0)	79 (86.8)	0.870
Metastatic sites	18 (14.0)	12 (13.2)	
Tumor grade			
I	1 (0.8)	0 (0)	0.624
II	49 (38.0)	30 (33.0)	
III	44 (34.1)	33 (36.3)	
Unknown	35 (27.1)	28 (30.8)	
Ki67 index			
≤14%	12 (9.3)	9 (9.9)	0.459
15%–50%	50 (38.8)	26 (28.6)	
>50%	45 (34.9)	39 (42.9)	
Unknown	22 (17.1)	17 (18.7)	
DFS (month)			
Median (95% CI)	24.3 (20.6–28.0)	14.3 (11.9–16.7)	0.002
≤24	66 (51.2)	69 (75.8)	0.000
>24	63 (48.8)	22 (24.2)	
Metastatic site			
Node	56 (43.4)	43 (47.3)	0.573
Bone	31 (24.0)	20 (22.0)	0.722
Chest wall	23 (17.8)	18 (19.8)	0.714
Lung	41 (31.8)	30 (33.0)	0.853
Liver	24 (18.6)	13 (14.3)	0.399
Brain	6 (4.7)	2 (2.2)	0.554
Other sites	13 (10.1)	20 (22.0)	0.015
Number of metastases			
1	88 (68.2)	52 (57.1)	0.153
2	22 (17.1)	25 (27.5)	
≥3	19 (14.7)	14 (15.4)	
Visceral metastasis**			
No	72 (55.8)	51 (56.0)	0.973
Yes	57 (44.2)	40 (44.0)	

DFS, disease-free survival; 95% CI, 95% confidence interval; NPCT, non-platinum-based chemotherapy; PBCT, platinum-based chemotherapy; *, Including digestive tract, pancreatic and prostate cancer and blood system tumors; **, Including lung, liver and brain metastases.

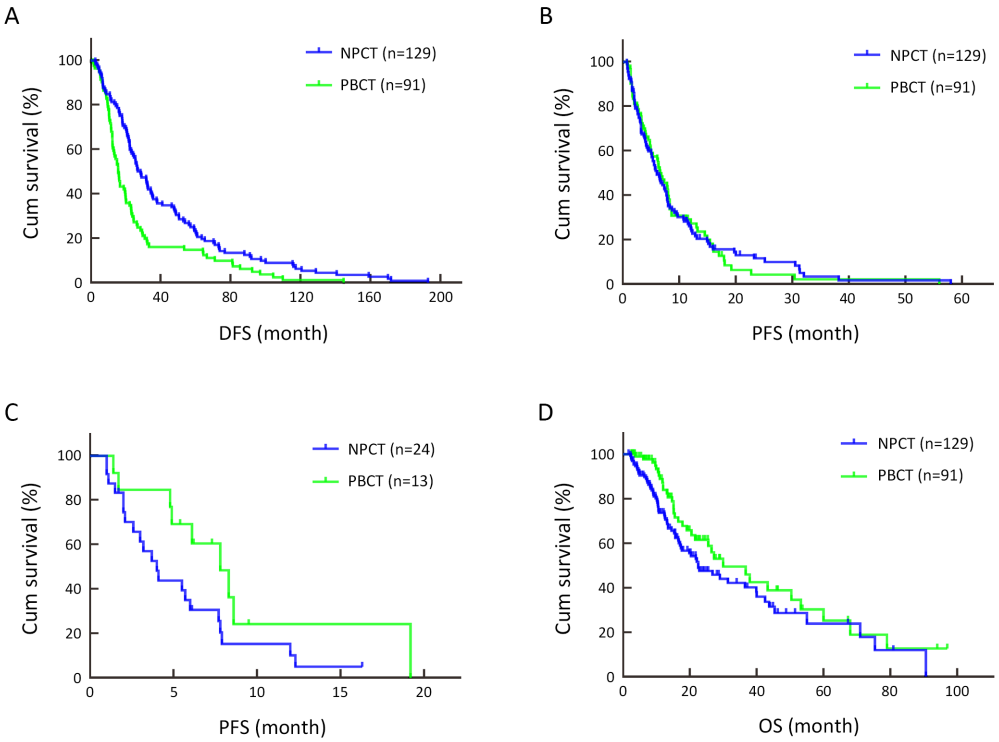

Figure 2 DFS, first-line PFS and OS according to treatment group. (A) DFS according to treatment group (P=0.002); (B) First-line PFS according to treatment group (P=0.907); (C) First-line PFS in patients with liver metastases according to treatment group (P=0.078); (D) OS from start of treatment of patients according to treatment group (P=0.090). DFS, disease-free survival; PFS, progression-free survival; OS, overall survival.

Table 2 Sources and results of *BRCA* gene testing

Results	BGI	Hospital*	Huidu**	Other centers
Total	35	33	61	13
Positive	4	5	8	2
Negative	31	28	53	11

BGI, Beijing Genomics Institution; *, Peking University Cancer Hospital; **, Huidu Shanghai clinical laboratory.

without; these response rates differ significantly (P=0.039). The median PFS was significantly longer in patients with *BRCA* mutations; namely, 14.9 months (95% CI: 6.9–22.9) for patients with deleterious *gBRCA* mutations *vs*. 5.3 months (95% CI: 4.0–6.6) for those without (P=0.001) (*Figure 5A*). By multivariate Cox regression model after adjusting for age of onset, DFS, tumor grade and visceral metastasis, the risk of progression was reduced by 88.7% for patients with deleterious *gBRCA* mutations compared to those without (P=0.008, HR=0.113, 95% CI: 0.023–0.566) (*Table 6*). The median OS was nearly 11 months longer in patients with deleterious *gBRCA* mutations (26.5 months *vs*. 15.5 months, 95% CI: 10.9–20.2); however, this difference was not statistically significant (P=0.161) (*Figure 5B*, *Table 7*).

In the NPCT group, CBR, PFS and OS were quite close between patients with and without deleterious *gBRCA* mutations. The CBR was 57.1% (4/7) and 39.7% (25/63) for patients with and without deleterious *gBRCA* mutations, respectively (P=0.627), the median PFS was 5.8 months (95% CI: 1.2–10.4) and 5.1 months (95% CI: 3.8–6.4) for patients with and without deleterious *gBRCA* mutations, respectively (P=0.677) (*Figure 5C*), and the median OS was 14.5 months (95% CI: 10.4–18.6) for patients without deleterious *gBRCA* mutations (*Figure 5D*). Only one of the seven patients with deleterious *gBRCA* mutations died; thus, there were too few data to calculate the OS (*Table 7*).

Adverse events

Adverse events were recorded in 180 patients, the most frequent is leucopenia, the incidence of which was similar in the two groups. Leucopenia occurred in 44.3% of the patients in the NPCT group and in 41.8% of the patients in the PBCT group. Other adverse events of any grade that

Table 3 Concordance of *BRCA* findings from different sources

No. of patients	BGI	Hospital*	Huidu**	Other centers
16	Negative	–	Negative	–
31	Negative	Negative	–	–
32	Negative	–	Negative	–
47	–	Negative	Negative	–
49	Negative	Negative	Negative	–
55	Negative	–	Negative	–
58	Negative	Negative	Negative	–
61	Negative	–	Negative	–
83	Positive	Positive	Positive	–
87	Negative	–	Negative	–
105	–	Positive	Positive	–
107	–	–	Positive	Positive
123	–	Negative	Negative	–
125	Negative	–	Negative	–
130	–	VUS	VUS	–
143	–	Negative	Negative	–
180	–	Negative	Negative	–
181	VUS	VUS	–	–
186	–	Negative	Negative	–
189	–	VUS	VUS	–
195	–	Negative	Negative	–
210	Positive	Positive	–	–
211	–	–	Negative	Negative
213	–	Negative	Negative	–
217	–	Negative	Negative	–

BGI, Beijing Genomics Institution; VUS, variant of uncertain significance in detected site; *, Peking University Cancer Hospital; **, Huidu Shanghai clinical laboratory.

Table 4 Summary of *BRCA* mutations

Variables	NPCT (n=70)	PBCT (n=44)
Positive	7	7
Negative	63	37
(Likely) benign/Wild type	62	29
VUS	1	8

VUS, variant of uncertain significance in detected site; NPCT, non-platinum-based chemotherapy; PBCT, platinum-based chemotherapy.

occurred in at least 15% of patients in either group were neutropenia (20.2% and 26.4%), fatigue (22.5% and 15.4%) and nausea (13.2% and 15.4%). Significantly more patients in the PBCT group had grade 3–4 anemia (which occurred in 0% of the patients in the NPCT group and 5.5% of the patients in the PBCT group) and thrombocytopenia (0% and 8.8%), whereas significantly more patients had palmar-plantar erythrodysesthesia (PPE) (12.4% and 0%) and peripheral neuropathy (8.6% and 1.1%) in the NPCT group. There were no treatment-related deaths (*Table 8*).

Discussion

In this study, we found that PBCT and NPCT achieved similar overall CBR, PFS and OS in our cohort of patients with advanced TNBC. However, in the PBCT group, patients with deleterious *gBRCA* mutations had higher CBR and longer PFS, and a non-significant tendency to longer OS than those without such mutations. No such differences were observed in the NPCT group. Additionally, we calculated the prevalence of deleterious *gBRCA* mutations and summarized the types of these mutations.

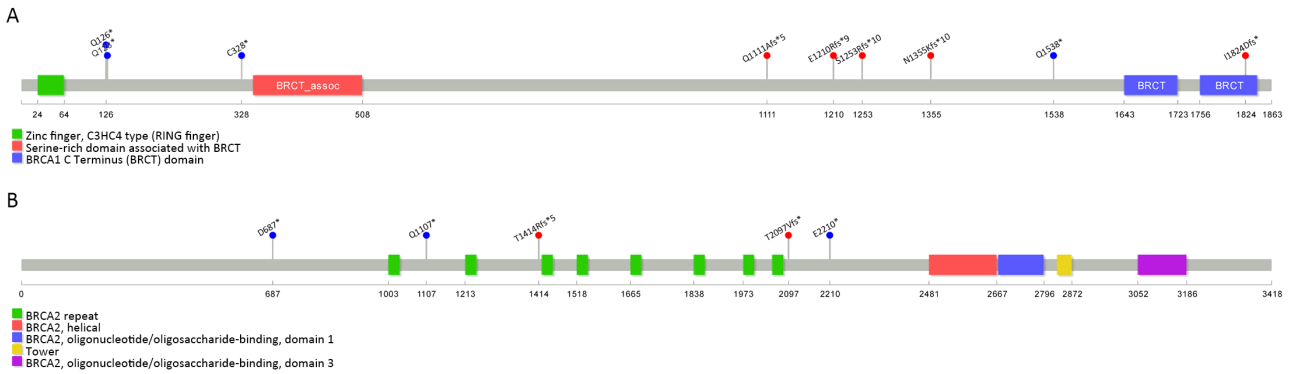

Figure 3 Locations of deleterious germline *BRCA1/2* (*gBRCA*) mutations. (A) *BRCA1*; (B) *BRCA2*.

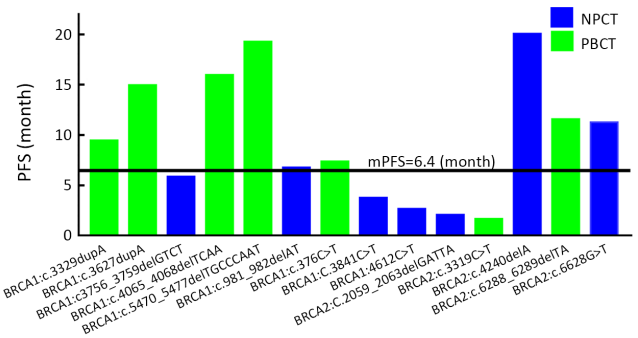

Figure 4 Progression-free survival (PFS) of patients with deleterious germline *BRCA1/2* (*gBRCA*) mutations according to treatment group. mPFS, median PFS.

The prevalence of *BRCA1/2* mutation is low level among patients with sporadic cancer and healthy individuals, but is higher in patients with risk factors such as TNBC and positive family history (20,21). In a Chinese cohort, Xie *et al.* found that the rate of *BRCA1/2* gene mutations in breast cancer was 5.3% overall, being the highest at 11.2% in those with triple negative disease (6). Patients with TNBC and a family history of breast and/or ovarian cancer have a higher rate of *BRCA* gene mutations, approximately 12.7% (20). In our study, the overall rate was 12.3% (14/114) in patients with TNBC and 23.5% (4/17) in patients with a family history of breast or ovarian cancer, these rates are comparable to those reported previously. However, the rate in patients with a family history of breast or ovarian cancer was higher than previously reported, and this discrepancy was possibly attributable to the small sample size.

Over 2,000 different mutations in *BRCA1/2* genes have been reported (22). Breast cancer risk varies by type and location of *BRCA1/2* mutations (23). Most deleterious mutations lead to truncated proteins that are nonfunctional (22,24). The BRCA1 protein contains a RING domain, a nuclear localization sequence (NLS), a CHK2 phosphorylation site on S988, a coiled-coil domain, and a BRCT domain (23,25). BRCA2 protein contains eight BRC repeats that bind RAD51, the DNA-binding domain that may facilitate BRCA2 binding to both single- and double-stranded DNA, an NLS and a cyclin-dependent kinase (CDK) phosphorylation site that also binds RAD51 (23,25). Mutations in the RING domain (c.72-192), the coiled coil domain (c.3759-3819, c.4191-4272), and the BRCT domain (c.4926-5169, c.5268-5526) of *BRCA1* are associated with high risk of breast cancer (23). Additionally, mutations in BRC repeats (c.3006-6255), the DNA-binding domain (c.7437-8001) and OB folds (c.8010-8400, c.9156-9570) have an impact on BRCA2 function (22,23). Mutations in these domains result in homologous recombination deficiency (HRD) and may result in increased sensitivity to platinum-based regimens. In this study, 114 patients had *BRCA* gene testing by using NGS; the identified deleterious mutations are summarized in *Table 9*, *Figure 3*. All deleterious mutations were located upstream or in the middle of the domain named above and were either frameshift or nonsense, which result in truncated proteins and loss of normal function.

The prevalence and spectrum of *BRCA1* and *BRCA2* mutations are heterogeneous in diverse groups of individuals. For example, Ashkenazi Jews are prone to well-described founder mutations in *BRCA1* (187delAG and 5385insC) and *BRCA2* (6174delT) (22). These founder mutations constitute more than 90% of mutations in Ashkenazi Jews but occur less frequently in other populations (26). One of our 14 patients with deleterious mutations was found to have *BRCA1* c.5470_5477 delTGCCCAAT and another patient was found to carry *BRCA1* c.981_982delAT. These two mutations occur frequently in Chinese individuals, suggesting that they are also potential founder mutations in Chinese population

Table 5 Efficacy according to clinically important factors, including type of chemotherapy regimen

Variables	CBR [% (n/N)]			PFS (95% CI)		
	NPCT (n=129)	PBCT (n=91)	P	NPCT (n=129)	PBCT (n=91)	P
Total	48.1 (62/129)	51.6 (47/91)	0.600	6.0 (4.6–7.4)	6.6 (5.1–8.1)	0.907
Age of onset (year)						
≤50	53.1 (34/64)	56.5 (35/62)	0.708	6.4 (4.9–7.9)	7.3 (5.9–8.7)	0.950
>50	43.1 (28/65)	41.4 (12/29)	0.878	5.7 (2.3–9.1)	4.5 (1.7–7.3)	0.713
DFS (month)						
≤24	31.8 (21/66)	44.9 (31/69)	0.118	4.5 (3.4–5.6)	6.2 (4.4–8.0)	0.333
>24	65.1 (41/63)	72.7 (16/22)	0.511	7.9 (7.0–8.8)	8.6 (0.8–16.4)	0.965
Visceral metastasis	47.4 (27/57)	50.0 (20/40)	0.798	6.0 (4.2–7.8)	6.7 (4.0–9.4)	0.939
Non-visceral metastasis	48.6 (35/72)	52.9 (27/51)	0.636	6.5 (4.7–8.3)	6.6 (3.6–9.6)	0.866
Lung metastasis	48.8 (20/41)	46.7 (14/30)	0.860	6.0 (3.5–8.5)	6.2 (3.0–9.4)	0.687
Liver metastasis	37.5 (9/24)	61.5 (8/13)	0.188	4.0 (2.6–5.4)	7.8 (5.0–10.6)	0.078
Chest wall metastasis	34.8 (8/23)	55.6 (10/18)	0.183	5.0 (2.1–7.9)	6.6 (2.1–11.1)	0.096
Bone metastasis	51.6 (16/31)	55.0 (11/20)	0.813	5.7 (1.3–10.1)	8.0 (6.6–9.4)	0.980
Lymph node metastasis	48.2 (27/56)	48.8 (21/43)	0.951	7.3 (4.9–9.7)	6.2 (4.4–8.0)	0.765

DFS, disease-free survival; NPCT, non-platinum-based chemotherapy; PBCT, platinum-based chemotherapy; CBR, clinical benefit rate; PFS, progression-free survival; 95% CI, 95% confidence interval.

(20). We also detected three deleterious mutations on *BRCA2* (c.4240delA, c.6288_6289delTA, and c.6628G>T), which resulted in nonfunctional truncated proteins but were rarely reported. *BRCA2* c.4240delA is a deletion of "A" at the 4240th nucleotide of *BRCA2* gene which results in a frameshift mutation and premature truncation of the BRCA2 protein. The gene with this mutation can only encode 1,417 amino acids while the wild type one can encode 3,418 amino acids. Similarly, *BRCA2* c.6288_6289delTA is a frameshift mutation, resulting in the change of the amino acid 2097 from Thr to Val and consequently a premature truncated protein. This variant is not reported in Clinvar (http://www.ncbi.nlm.nih.gov/clinvar), 1000 genomes (http://www.1000genomes.org), NHLBI-ESP 6500 exome project (http://evs.gs.washington.edu/EVS), and the Exome Aggregation Consortium databases (http://exac.broadinstitute.org/). We classified it as likely pathogenic according to ACMG Guidelines. With a further review of the patient's family history of cancer, the patient's mother had ovarian cancer supporting that this mutation is deleterious. *BRCA2* c.6628G>T is a nonsense mutation, leading to a truncated BRCA2 protein at amino acid 2210 (Glu). While this variant is not reported in Clinvar, BIC (Breast Cancer Information Core; http://research.nhgri.nih.gov/bic/), and UMD-BRCA2 (Universal Mutation Database; http://www.umd.be/BRCA2/) databases, it has been reported pathogenic with one case in LOVD database (Leiden Open Variation Database; http://www.lovd.nl) and one case in Japan (27). In our study, the patient also reported a family history of breast and liver cancer. These three mutations may be warranted for further screening to determine if they are specific to Chinese or Asian population.

Multiple genes, including *BRCA1/2*, *ATM*, *RAD51* and *BRIP1*, are involved in HR (28); in this study, we mainly studied the *BRCA1/2* gene. *BRCA* gene mutations can lead to HRD, resulting in failure to repair DNA double strand breaks and thus increasing sensitivity to agents aimed at DNA (5,10,29). Although most patients with sporadic TNBC do not have *BRCA1* mutations, there is evidence of BRCA1 pathway dysfunction in these tumors (13). Platinum-based regimens are effective treatments for advanced breast cancer and damage DNA by cross-linking with DNA, thereby killing tumor cells (30). Therefore, platinum-based regimens should be more effective in TNBC, especially TNBC with *BRCA* gene mutations (30,31).

Clinical trials have shown that the use of platinum-based

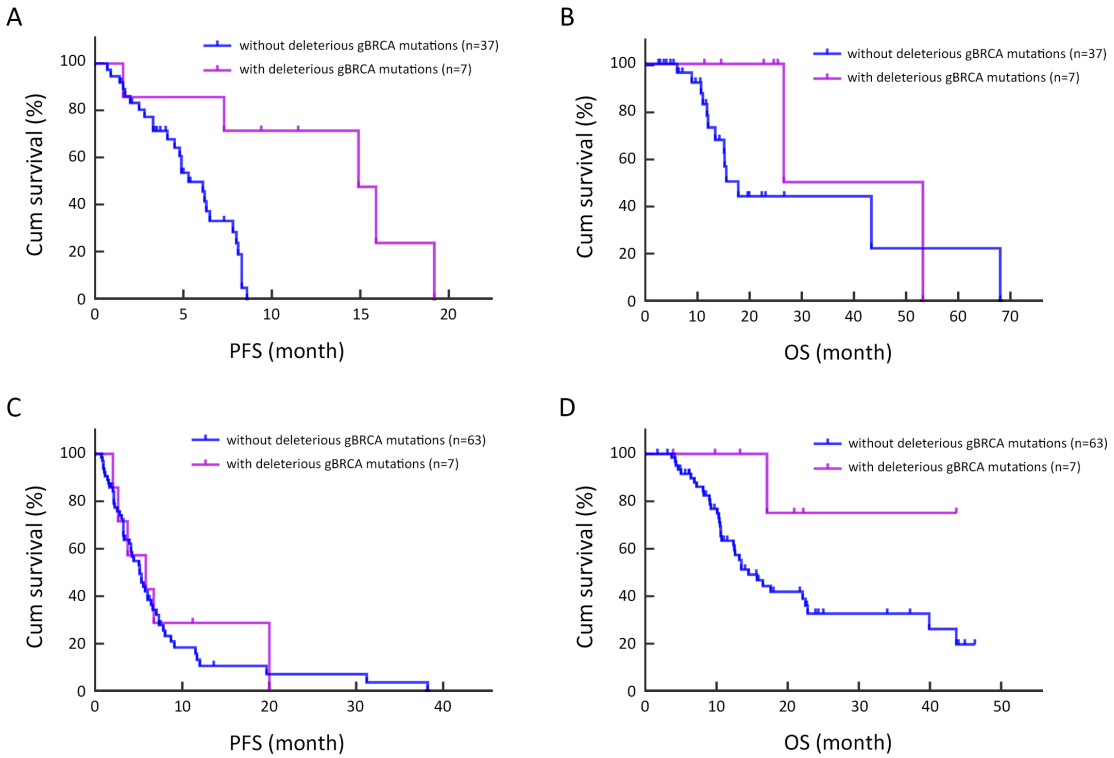

Figure 5 First-line progression-free survival (PFS) of patients with or without deleterious germline *BRCA1/2* (*gBRCA*) mutations in subgroup analysis. (A) First-line PFS of patients with or without deleterious *gBRCA* mutations in platinum-based chemotherapy (PBCT) group (P=0.001); (B) Overall survival (OS) for platinum-based treatment of patients with or without deleterious *gBRCA* mutations in PBCT group (P=0.161); (C) First-line PFS of patients with or without deleterious *gBRCA* mutations in non-platinum-based chemotherapy (NPCT) group (P=0.677); (D) OS from non-platinum-based treatment of patients with or without deleterious *gBRCA* mutations in NPCT group (P=0.075).

Table 6 Multivariate Cox analyses for DFS in PBCT group

Variables	HR (95% CI)	P
Age of onset (≤50 *vs*. >50) (year)	0.576 (0.233–1.420)	0.230
DFS (≤24 *vs*. >24) (month)	0.391 (0.096–1.596)	0.191
Tumor grade (III *vs*. II)	1.740 (0.545–5.557)	0.350
Visceral metastasis (visceral *vs*. non-visceral)	0.823 (0.368–1.839)	0.635
gBRCA mutation (with *vs*. without)	0.113 (0.023–0.566)	0.008

DFS, disease-free survival; PBCT, platinum-based chemotherapy; *gBRCA*, germline *BRCA1/2*; HR, hazard ratio; 95% CI, 95% confidence interval.

regimens as neoadjuvant chemotherapy can improve the pathologic complete response (pCR) rates of patients with *BRCA* gene mutations and thus improve the OS (32,33). The CBCSG006 trial in patients with advanced TNBC (16) found that cisplatin plus gemcitabine was superior to paclitaxel plus gemcitabine as first-line therapy (PFS, 7.73 months *vs.* 6.47 months, P=0.009). However, in our study, we identified no overall difference in efficacy between platinum- and non-platinum-based therapy (CBR, 51.6% *vs.* 48.1%, P=0.600, median PFS, 6.6 months *vs.* 6.0 months, P=0.907). However, among patients receiving platinum-based regimens, the CBR and PFS were statistically superior in patients with deleterious *gBRCA* mutations, namely, 85.7% *vs.* 35.1%, respectively, P=0.039, and 14.9 months *vs.* 5.3 months, respectively, P=0.001. Additionally, the TNT trial (17) found that platinum-based regimens achieve a better objective response rate (68% *vs.* 33.3%, P=0.03) and PFS (6.8 months *vs.* 4.4 months,

Table 7 CBR, PFS and OS with or without deleterious *gBRCA* mutations according to type of chemotherapy

Results	PBCT (n=44)			NPCT (n=70)		
	With (n=7)	Without (n=37)	P	With (n=7)	Without (n=63)	P
CBR	85.7% (6/7)	35.1% (13/37)	0.039	57.1% (4/7)	39.7% (25/63)	0.627
PFS [median (95% CI)] (month)	14.9 (6.9–22.9)	5.3 (4.0–6.6)	0.001	5.8 (1.2–10.4)	5.1 (3.8–6.4)	0.677
OS [median (95% CI)] (month)	26.5 (–)	15.5 (10.9–20.2)	0.161	–	14.5 (10.4–18.6)	0.075

CBR, clinical benefit rate; PFS, progression-free survival; OS, overall survival; *gBRCA*, germline *BRCA1/2*; PBCT, platinum-based chemotherapy; NPCT, non-platinum-based chemotherapy; 95% CI, 95% confidence interval.

Table 8 Drug-related adverse events

Adverse events	NPCT (n=129) [n (%)]			PBCT (n=91) [n (%)]		
	Grade 1–2	Grade 3	Grade 4	Grade 1–2	Grade 3	Grade 4
Hematological						
Leucopenia	38 (29.5)	14 (10.9)	5 (3.9)	24 (26.4)	11 (12.1)	3 (3.3)
Neutropenia	11 (8.5)	13 (10.1)	2 (1.6)	15 (16.5)	6 (6.6)	3 (3.3)
Febrile neutropenia	NA	2 (1.6)	0 (0)	NA	2 (2.2)	0 (0)
Anemia	4 (3.1)	0 (0)	0 (0)	3 (3.3)	4 (4.4)	1 (1.1)
Thrombocytopenia	2 (1.6)	0 (0)	0 (0)	3 (3.3)	5 (5.5)	3 (3.3)
Laboratory-assessed items						
Increased ALT/AST	6 (4.7)	0 (0)	0 (0)	9 (9.9)	0 (0)	0 (0)
Increased bilirubin	3 (2.3)	0 (0)	0 (0)	3 (3.3)	0 (0)	0 (0)
Non-hematological						
Nausea	16 (12.4)	1 (0.8)	NA	14 (15.4)	0 (0)	NA
Vomiting	9 (7.0)	1 (0.8)	0 (0)	4 (4.4)	1 (1.1)	0 (0)
Anorexic	19 (14.7)	0 (0)	0 (0)	6 (6.6)	0 (0)	0 (0)
Diarrhea	5 (3.9)	0 (0)	0 (0)	4 (4.4)	0 (0)	0 (0)
Abdominal distension	3 (2.3)	0 (0)	NA	5 (5.5)	0 (0)	NA
Constipation	5 (3.9)	0 (0)	0 (0)	3 (3.3)	0 (0)	0 (0)
Fatigue	27 (20.9)	2 (1.6)	NA	13 (14.3)	1 (1.1)	NA
Hyperhidrosis	15 (11.6)	0 (0)	NA	9 (9.9)	0 (0)	NA
Weight loss	3 (2.3)	0 (0)	NA	1 (1.1)	0 (0)	NA
Insomnia	13 (10.1)	1 (0.8)	NA	10 (11.0)	0 (0)	NA
Pain	13 (10.1)	0 (0)	NA	5 (5.5)	0 (0)	NA
Alopecia	17 (13.2)	NA	NA	10 (11.0)	NA	NA
PPE	16 (12.4)	0 (0)	NA	0 (0)	0 (0)	NA
Pruritus	8 (6.2)	0 (0)	NA	4 (4.4)	0 (0)	NA
Hyperpigmentation	7 (5.4)	NA	NA	5 (5.5)	NA	NA
Dyspnea	1 (0.8)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)
Palpitations	3 (2.3)	NA	NA	0 (0)	NA	NA
Stomatitis	8 (6.2)	0 (0)	0 (0)	2 (2.2)	0 (0)	0 (0)
Peripheral neuropathy	10 (7.8)	1 (0.8)	0 (0)	1 (1.1)	0 (0)	0 (0)
Headache	1 (0.8)	0 (0)	NA	0 (0)	0 (0)	NA
Dizziness	2 (1.6)	0 (0)	NA	1 (1.1)	0 (0)	NA

ALT, alanine aminotransferase; AST, aspartate aminotransferase; PPE, palmar-plantar erythrodysesthesia; NPCT, non-platinum-based chemotherapy; PBCT, platinum-based chemotherapy; NA, not applicable. No grade 5 adverse events were observed.

Table 9 Summary of deleterious *gBRCA* mutations

No. of patients	Gene	Location	Mutation type	AA change
193	*BRCA1*	c.3329dupA	Frameshift	Q1111Afs*5
126	*BRCA1*	c.3627dupA	Frameshift	E1210Rfs*9
105	*BRCA1*	c.3756_3759delGTCT	Frameshift	S1253Rfs*10
198	*BRCA1*	c.4065_4068delTCAA	Frameshift	N1355Kfs*10
210	*BRCA1*	c.5470_5477delTGCCCAAT	Frameshift	I1824Dfs*
107	*BRCA1*	c.981_982delAT	Nonsense	C328*
157	*BRCA1*	c.376C>T	Nonsense	Q126*
216	*BRCA1*	c.3841C>T	Nonsense	Q128*
98	*BRCA1*	c.4612C>T	Nonsense	Q1538*
10	*BRCA2*	c.2059_2063delGATTA	Frameshift	D687*
215	*BRCA2*	c.3319C>T	Nonsense	Q1107*
83	*BRCA2*	c.4240delA	Frameshift	T1414Rfs*5
136	*BRCA2*	c.6288_6289delTA	Frameshift	T2097Vfs*
113	*BRCA2*	c.6628G>T	Nonsense	E2210*

gBRCA, germline *BRCA1/2*.

P=0.002) in patients with *BRCA* gene mutations. We found no such difference in the NPCT group, suggesting that patients with advanced TNBC and deleterious *gBRCA* mutations gain more benefit from platinum-based regimens.

The median PFS of platinum-based regimens tended to be better in patients with liver and chest wall metastases; however, the difference between the PBCT and NPCT groups was not significant (P=0.078) (*Figure 2C*). Whether the efficacy of platinum-based regimens is related to metastatic sites and how to screen the patients who will benefit needs further study.

Limited information is available about OS. In the TNT trial (17), OS did not differ significantly between carboplatin and docetaxel either overall or in the *BRCA* subgroup. The CBCSG006 trial updated their survival data in 2018 (34) and reported identifying no statistical difference in overall OS between the cisplatin plus gemcitabine *vs.* paclitaxel plus gemcitabine arms, and no significant correlation between *gBRCA1/2* status and OS. The median follow-up time of our study was 14.3 (range, 1.7–97.0) months. As the median OS of TNBC after recurrence is about 9 months (35), the follow-up time of our study had covered the median OS in most TNBC patients. In our study, there was no statistically significant difference in OS, although OS tended to be longer in patients who received PBCT and harbored deleterious *gBRCA* mutations. Given that OS is influenced by many factors, including adverse effects of treatment and subsequent treatment, further investigation in larger cohorts is needed.

In our study, the most frequent adverse event was leucopenia and the PBCT group had significantly more grade 3–4 anemia and thrombocytopenia, which is in concordance with the results of the CBCSG006 trial (16). PPE and peripheral neuropathy occurred more frequently in the non-platinum-based group; these are associated with use of capecitabine.

The main limitation of this study is inherent to its retrospective design. Additionally, some clinical information, such as adverse effects, were missing because they were not documented in the medical records; these data may have influenced the identified associations to some extent.

Conclusions

Platinum-based regimens are more effective in patients with deleterious *gBRCA* mutations, but no difference in patients without *BRCA* gene mutations, so non-platinum is an option in patients without *BRCA* gene mutations considering the toxicity and side effect. And we recommend that patients with advanced TNBC should have *BRCA* gene test.

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Footnote

Conflicts of Interest: The authors have no conflicts of interest to declare.

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