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Current Status of Immune Checkpoint Inhibitors and Treatment Responsive Biomarkers for Triple-Negative Breast Cancer

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ABSTRACT

Triple-negative breast cancer (TNBC), accounting for about 10%–20% of all breast cancer cases, is characterized by its aggressive nature, high recurrence rates, and poor prognosis. Unlike other breast cancer subtypes, TNBC lacks hormone receptors and specific molecular targets, limiting therapeutic options. In recent years, immune checkpoint inhibitors (ICIs) have shown promise in treating TNBC by targeting immune evasion mechanisms. Despite these advancements, several issues remain unresolved, including low response rates in programmed cell death ligand 1 (PD-L1) negative TNBC subtypes and the challenge of predicting which patients will benefit from ICIs. Consequently, there is growing interest in identifying reliable biomarkers beyond PD-L1 expression. This review synthesizes recent studies to provide a comprehensive perspective on ICI therapy in TNBC, clarifying the status of single-agent ICI therapies and combination strategies, emphasizing the need for further research into biomarkers. These insights provide clues for more personalized and effective treatment approaches, ultimately aiming to improve clinical outcomes for patients with TNBC.

1 | Introduction

Breast cancer is the most common malignancy among women worldwide. According to recent global data, 2,261,419 new cases and 684,996 deaths from breast cancer were reported in 2020 [1–3]. Triple-negative breast cancer (TNBC), which is estrogen receptor negative, progesterone receptor negative, and human epidermal growth factor receptor negative, accounts for about 10%–20% of all breast cancer cases [4]. TNBC is characterized by poor differentiation and is highly invasive with a high risk of recurrence and distant metastasis [5–7]. General progress has been achieved for breast cancer in surgical treatments, chemotherapies, endocrine therapies, and targeted therapies [8]. However, most patients with TNBC have not derived benefit from current endocrine therapies or targeted therapies, and their responses to chemotherapy are often unsatisfactory. Therefore, more efficacious treatment options for TNBC are needed [9].

Abbreviations: Akt, protein kinase B; APC, antigen-presenting cell; CPS, combined positive score; CR, complete response; CTLA-4, cytotoxic T lymphocyte antigen 4; DC, dendritic cell; DCR, disease control rate; DoR, duration of response; EFS, event-free survival; FDA, Food and Drug Administration; GEP, gene expression profile; ICI, immune checkpoint inhibitor; ICs, immune cells; iDFS, invasive disease-free survival; MMR, mismatch repair; mTNBC, metastatic triple-negative breast cancer; ORR, objective response rate; OS, overall survival; pCR, pathologic complete response; PD-1, programmed death protein 1; PD-L1, programmed cell death ligand-1; PFS, progression-free survival; PI3K, phosphatidylinositol-3-kinase; PR, partial response; PTEN, phosphatase and tensin homolog; SHP, Src-homology-2-domain-containing protein tyrosine phosphatase; TCR, T-cell receptor; TIL, tumor-infiltrating lymphocyte; TMB, tumor mutational burden; TNBC, metastatic triple-negative breast cancer; TNBC, triple-negative breast cancer; TRAE, treatment-related adverse event; Treg, regulatory T cell.

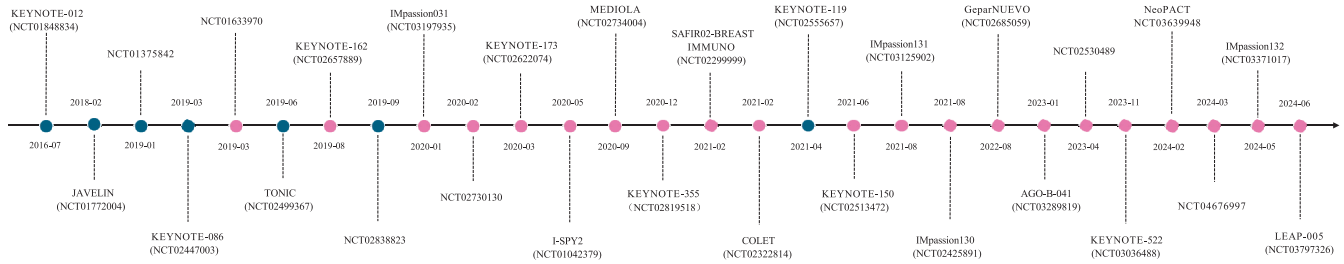

FIGURE 1 | The list of major clinical trials for ICIs in TNBC patients. Blue points: monotherapy; pink points: combination therapy.

Accumulated evidences indicate that immunotherapies and TROP2-directed antibody-drug conjugates have great potential as therapeutic strategies for breast cancer, including TNBC, as well as several other solid malignancies [6–11]. Compared with other types of breast cancer, TNBC has increased tumor-infiltrating lymphocytes (TILs), programmed cell death ligand-1 (PD-L1) expression, and genomic instability and mutational load [12–14], suggesting that immunotherapeutic approaches could be effective. Currently, these strategies mainly consist of cancer vaccines, oncolytic viruses, adoptive immune cell transfer and immune checkpoint inhibitors (ICIs).

In recent years, encouraging clinical results have ushered in a new era of using ICIs for the treatment of TNBC [15, 16]. Unfortunately, ICI treatment is not suitable for all TNBC patients, and thus, studies have been conducted to identify predictive biomarkers that can reliably predict a patient's response to ICI therapy [17]. In this review, we provide a broad, up-to-date summary of recent advances in the use of ICI therapy for TNBC, including molecular mechanisms, patient outcomes, and predictive biomarkers for identifying patients with TNBC who are most likely to benefit from ICI therapy.

2 | Molecular Mechanisms of ICIs in TNBC

The most studied ICI targets in breast cancers are the programmed death protein 1 (PD-1, also known as CD279), programmed death-ligand 1 (PD-L1, also known as CD274), and cytotoxic T lymphocyte antigen 4 (CTLA-4, also known as CD152) [18]. PD-1 is expressed on the outside surface of numerous immune cells, especially activated T cells, B cells, natural killer cells, activated monocytes, and dendritic cells, whereas PD-L1 exists on the surface of tumor cells [19, 20]. Binding of PD-L1 (on tumor cells) to PD-1 (on activated T cells) inhibits the migration and proliferation of T cells, decreases the release of anti-tumor cytokines, and thus prevents overactivation of immune responses, eventually leading to immunosuppression and escape of tumor cells from the anti-tumor immune response [21]. Binding between PD-1 and PD-L1 inhibits an immune response via several possible mechanisms: (1) recruitment of Src homology 2-containing protein tyrosine phosphatase-1 (SHP-1) and -2 (SHP-2), which dephosphorylates protein kinase B (Akt) and phosphatidylinositol-3-kinase (PI3K), and then inhibits T-cell receptor (TCR) signaling and T-cell activation [22–24]; (2) prevention of TCR signaling termination, which stops the coupling of antigen-carrying dendritic cells (DCs) with effector T cells and decreases the immune response [25], and (3) modulating the accumulation of regulatory T cells (Tregs) in the tumor immune microenvironment [26].

Whereas PD-1–induced immunosuppression mainly occurs during the later stages of tumor progression and mostly regulates effector T cells in peripheral tissues, CTLA-4–mediated immunosuppression occurs during the early stage of antigen presentation [27]. CTLA-4 (CD152) and CD28 are expressed on the external surface of activated T cells and are responsible for T cell immune regulation [28]. They bind two distinct ligands, CD80 (B7.1) and CD86 (B7.2), which are found on the surface of professional antigen-presenting cells (APCs) or other immune mediators and regulate signal pathway activation of productive immunity at an early stage. When the immune system recognizes an antigen, CD28 first interacts with CD80 and CD86, and this interaction activates T cells by increasing T-cell antigen receptor signaling, leading to upregulation of CTLA-4 [9]. CTLA-4, which is structurally similar to CD28, binds to CD80 and CD86 on the surface of APCs and then blocks interaction of T cells with APCs, leading to down-regulation of T cells [29, 30]. Therefore, targeted inhibition of CTLA-4 activates and promotes immune cell proliferation, and this increases the anti-tumor immune response.

Currently, many drugs that target PD-1/PD-L1 or CTLA-4 have been or are being investigated for the treatment of various types of cancers. Among these ICIs, several PD-1/PD-L1 inhibitors, including pembrolizumab [31], nivolumab [32, 33], cemiplimab [34], atezolizumab [35], avelumab [36], and durvalumab, are approved for clinical use in various types of cancers. The U.S. Food and Drug Administration (FDA) recently approved pembrolizumab in combination with chemotherapy for the treatment of TNBC [37]. Moreover, several CTLA-4 inhibitors, including ipilimumab and tremelimumab, have also been approved or are being tested for the treatment of multiple cancer types [38–41], including TNBC [42].

3 | Clinical Updates With ICIs for TNBC

3.1 | Locally Advanced or Metastatic TNBC

Early clinical trials examining different applications of ICIs are considered "proof-of-concept" studies. These studies examined the efficacy and safety of ICIs alone (monotherapy) or in combination with other therapies (combination therapy), for locally advanced or metastatic TNBC (mTNBC) (Figure 1 and Table 1).

TABLE 1 | Clinical trials that examined the use of ICIs for TNBC treatment.

Clinical trial number	Author, year	Phase	Sample size (n)	Regimen	ORR	mPFS (months)	mOS (months)	HR (95% CI)	Response
Locally advanced or metastatic TNBC									
PD-1 inhibitors									
NCT01848834 KEYNOTE-012	Nanda et al. 2016 [43]	Ib	32	Pembrolizumab	18.5%	1.9	11.2		+
NCT02447003 KEYNOTE-086	Adams et al. 2019 [44, 45]	II	Cohort A: 170	Pembrolizumab	5.3%	2.0	9.0		+
			Cohort B: 84	Pembrolizumab	21.4%	2.1	18.0		+
NCT02555657 KEYNOTE-119	Winer et al. 2021 [46]	III	622	Pembrolizumab	9.6%		9.9	0.97 (0.82–1.15)	–
NCT02838823	Bian et al. 2019 [47]	I	20	Toripalimab	5%	1.8			+
NCT02499367 TONIC	Voorwerk et la., 2019 [48]	II	67	Nivolumab + chemotherapy	20%	1.9			+
NCT02657889 KEYNOTE-162	Vinayak et al. 2019 [49]	I/II	55	Pembrolizumab + niraparib	21%	2.3			+
NCT02819518 KEYNOTE-355	Cortes et al. 2020 [50]	III	847	Pembrolizumab + chemotherapy		7.5		0.82 (0.69–0.97)	+
NCT02730130	Ho et al. 2020 [51]	II	17	Pembrolizumab + radiotherapy	17.6%	2.6	7.6		+
NCT02513472 KEYNOTE-150	Tolaney et al. 2021 [52]	Ib/II	Stratum 1:66 Stratum 2:101	Pembrolizumab + eribulin Pembrolizumab + eribulin	25.8% 21.8%				
NCT03797326 LEAP-005	Chung et al. 2024 [53]	II	31	Pembrolizumab + lenvatinib	32%	5.1	11.4		+
PD-L1 inhibitors									
NCT01772004 JAVELIN	Dirix et al. 2018 [54]	Ib	58	Avelumab	5.2%	1.4	9.2		+
NCT01375842	Emens et al. 2019 [55]	I	116	Atezolizumab	10.0%	1.4	8.9		+
NCT02299999 SAFIR02-BREAST IMMUNO	Bachelot et al. 2021 [56]	II	82	Durvalumab		2.7	21.2		+
NCT02425891 IMpassion130	Emens et al. 2021 [57]	III	451	Atezolizumab + nab-paclitaxel		7.5	21.0	0.87 (0.75–1.02)	+
NCT03125902 IMpassion131	Miles et al. 2021 [58]	III	651	Atezolizumab + paclitaxel	63%	6.0	22.1	1.11 (0.76–1.64)	–
NCT01633970	Adams et al. 2019 [59]	Ib	33	Atezolizumab + nab-paclitaxel	39.4%	5.5	14.7		+

(Continues)

TABLE 1 | (Continued)

Clinical trial number	Author, year	Phase	Sample size (n)	Regimen	ORR	mPFS (months)	mOS (months)	HR (95% CI)	Response
NCT02322814 COLET	Brufsky et al. 2021 [60]	II	32	Atezolizumab + cobimetinib + nab-paclitaxel	33.4%	3.8	11.0		+
			31	Atezolizumab + cobimetinib + paclitaxel	29.0%	7.0	NE		+
NCT02734004 MEDIOLA	Domchek et al. 2020 [61]	I/II	34	Durvalumab + olaparib	63.3%	4.9	20.5		+
NCT03371017 IMpassion132	Dent et al. 2024 [62]	III	595	Atezolizumab + chemotherapy	40%	4.2	12.1	0.95 (0.74–1.22)	–
Early TNBC									
PD-1 inhibitors									
NCT01042379 I-SPY2	Nanda et al. 2020 [63]	II	69	Pembrolizumab + paclitaxel	pCR 60%				+
NCT03036488 KEYNOTE-522	Takahashi et al. 2023 [64] Schmid et al. 2024a [65] Schmid et al. 2024b [66]	III	1174	Pembrolizumab + chemotherapy	pCR 58.7%		5-year EFS%: 81.3% 5-year OS: 86.6%	OS: 0.65 (0.51–0.83)	+
NCT02622074 KEYNOTE-173	Schmid et al. 2020 [67]	Ib	60	Pembrolizumab + chemotherapy	pCR 60%				+
NCT03289819 NeoImmunoboost (AGO-B-041)	Fasching et al. 2023 [68]	II	50	Pembrolizumab + nab-paclitaxel	pCR 66%				+
NCT03639948 NeoPACT	Sharma et al. 2024 [69]	II	115	Pembrolizumab + carboplatin + docetaxel	pCR 58%		estimatedEFS% at 36 months: 86%		+
NCT04676997	Zheng et al. 2024 [70]	II	23	Camrelizumab + nab-paclitaxel: 4 cycles Camrelizumab + epirubicin + cyclophosphamide: 4 cycles	pCR 65.0%				+
PD-L1 inhibitors									
NCT03197935 IMpassion031	Mittendorf et al. 2020 [71]	III	333	Atezolizumab + chemotherapy	pCR: 58%				+
NCT02685059 GeparNUEVO	Loibl et al. 2022 [72]	II	174	Durvalumab + chemotherapy			OS% 95.2%		–
NCT02530489	Yam et al. 2023 [73]	II	37	Atezolizumab + nab-paclitaxel	pCR 46%				+

Note: +: positive results, –: negative results.

Abbreviations: EFS: event-free survival; mOS: median overall survival; mPFS: median progression-free survival; ORR: objective response rate; pCR: pathologic complete response; PD-1: programmed death protein 1; PD-L1: programmed cell death ligand-1; PFS: progression-free survival; TNBC: triple-negative breast cancer.

3.1.1 | Monotherapy With PD-1 Inhibitors

Recent studies, including KEYNOTE-012, KEYNOTE-086, and KEYNOTE-119, have explored pembrolizumab monotherapy as a treatment for advanced TNBC, with mixed efficacy results. The KEYNOTE-012 phase Ib trial examined the efficacy and safety of pembrolizumab monotherapy for the treatment of PD-L1+ (PD-L1 expression ≥1% of tumor cells) advanced TNBC (cohort A, $n=32$), many of whom (46.9%) had undergone multiple prior therapies [43]. The objective response rate (ORR) was 18.5%, with one patient achieving a complete response (CR). Although 15.6% of patients experienced grade 3 or higher treatment-related adverse events (TRAEs), these findings highlighted some anti-tumor activity in a heavily pretreated population [43]. In cohort A ($n=170$) of the KEYNOTE-086 phase II trial, which evaluated the efficacy and safety of pembrolizumab monotherapy in previously treated TNBC, the ORRs were low, at 5.3% in the overall population and 5.7% in the PD-L1 positive (combined positive score [CPS] ≥1) population, with a median progression-free survival (PFS) of 2.0 months and a median overall survival (OS) of 9.0 months [44]. However, in cohort B ($n=84$) of KEYNOTE-086, which focused on untreated, PD-L1 positive (CPS ≥1) TNBC patients, the ORR was 21.4%, with a median PFS of 2.1 months and a median OS of 18.0 months, and a low rate of adverse effects [45]. These results suggest better efficacy in treatment-naive, PD-L1-positive patients and underscored the importance of PD-L1 status in response to pembrolizumab.

The KEYNOTE-119 phase III multi-center trial compared pembrolizumab to chemotherapy in previously treated TNBC patients [46]. While the primary analysis did not show a statistically significant OS benefit in patients with low CPS (either CPS ≥10 or CPS ≥1), an exploratory post hoc analysis revealed longer OS in patients with a higher CPS (≥20) in the pembrolizumab group (14.9 vs. 12.5 months), suggesting pembrolizumab may offer survival benefits in this subgroup [46]. Another preliminary phase I open-label dose escalation study (NCT02838823) evaluated JS001 (a novel PD-1 antibody) in advanced TNBC patients who experienced failure after multi-line standard systemic therapy [47]. Results showed limited efficacy (ORR of 11.1%) and a median PFS of 1.8 months, suggesting that while PD-1 inhibitors can be safe, but their efficacy in heavily pretreated TNBC remains limited.

These trials showed that PD-1 inhibitor monotherapy has demonstrated modest anti-tumor activity and an acceptable safety profile in advanced TNBC, especially in PD-L1-positive, treatment-naive patients. However, the effectiveness of ICIs in broader TNBC populations remains limited, underscoring the need for potential combination approaches to improve outcomes.

3.1.2 | Combination Therapy With PD-1 Inhibitors

Based on the encouraging results with monotherapies of ICIs, clinical trials of combination regimens with ICIs, specifically PD-1 inhibitors like pembrolizumab and nivolumab, with chemotherapy for advanced or metastatic TNBC were conducted.

The TONIC trial explored a short induction period with agents like cyclophosphamide, doxorubicin, or cisplatin before nivolumab in 67 patients with mTNBC who had received previous chemotherapy. The results showed an improved ORR in patients with doxorubicin (35%) or cisplatin (23%) induction compared to no induction (17%) or with irradiation or cyclophosphamide induction (8%) [48]. This suggests that selective induction strategies can potentially prime the tumor microenvironment, enhancing the efficacy of PD-1 inhibitors. In another phase II clinical trial (NCT02657889), pembrolizumab in combination with niraparib (a poly ADP-ribose polymerase inhibitor) achieved an ORR of 21% and a DCR of 49% in advanced TNBC patients [49]. *BRCA* mutated tumors showed higher response rates, with an ORR of 47% versus 11%, a DCR of 80% versus 33%, and a median PFS of 8.3 versus 2.1 months, respectively [49]. This highlighted the importance of genetic profiling in TNBC to optimize combination strategies for maximum efficacy.

Based on the PFS results from the interim analysis of the phase III KEYNOTE-355 trial, the U.S. FDA provided accelerated approval of pembrolizumab plus chemotherapy as first-line treatment for unresectable, locally recurrent or metastatic, PD-L1 positive (CPS ≥10) TNBC [hazard ratio (HR): 0.82 (0.69–0.97)] [50]. At the protocol-specified final analysis of this trial, pembrolizumab plus chemotherapy significantly improved mOS in patients with higher CPS scores (≥10: 23.0 vs. 16.1 months) compared to chemotherapy alone. However, in patients with a CPS ≥1, the two treatment groups had a similar median OS (17.6 vs. 16.0 months) [74], underscoring the need to refine patient selection based on PD-L1 levels.

A phase II trial (NCT02730130) reported an ORR of 17.6% with pembrolizumab and radiotherapy in heavily pretreated patients with mTNBC [51]. Three of these patients achieved CR (one with DoR up to 108 weeks), and one patient achieved stable disease for 22 weeks, suggesting radiotherapy as a potential priming mechanism to boost ICI efficacy in selected patients. Also, the KEYNOTE 150 phase Ib/II trial (NCT02513472) evaluated the efficacy and safety in 167 patients with mTNBC (≤2 prior systemic anticancer therapies) treated with pembrolizumab in combination with eribulin [52]. ORRs were 25.8% (95% CI: 15.8–38.0) for stratum 1 with 66 untreated patients and 21.8% (95% CI: 14.2–31.1) for stratum 2 with 101 patients after 1–2 line treatment. PD-L1+ patients (CPS ≥1) had numerically higher ORR than PD-L1- patients, particularly in stratum 1 (34.5% vs. 16.1%; $p<0.001$) and stratum 2 (24.4% vs. 18.2%; $p<0.05$) [52]. In the TNBC cohort of the phase 2 LEAP-005 Study (NCT03797326) who received lenvatinib (tyrosine kinase inhibitor) plus pembrolizumab after one or two lines of therapy, ORR was 32% (95% CI, 17%–51%), mPFS was 5.1 (95% CI, 1.9–11.8) months, and the mOS was 11.4 (95% CI, 4.1–21.7) [53] months, suggesting that combining PD-1 inhibitors with tyrosine kinase inhibitors could be a viable option for enhancing treatment response.

These trials emphasize that while PD-1 inhibitors show some efficacy in locally advanced or metastatic TNBC, combination therapies and patient selection based on biomarkers like PD-L1 status are essential for optimizing outcomes. Numerous ongoing phase III trials continue to explore different combination approaches, aiming to extend the benefits of PD-1 inhibitors to a broader TNBC population. For example, TROPION-Breast05 (NCT06103864) is investigating pembrolizumab + Dato-DXd with or without durvalumab for PD-L1+ locally recurrent

inoperable or metastatic TNBC, ASCENT-04 (NCT05382286) is assessing pembrolizumab + Sacituzumab Govitecan-hziy [an antibody–drug conjugate (ADC) targeting anti-trophoblast cell-surface antigen 2] for previously untreated locally advanced inoperable or metastatic TNBC, and MK-2870-011/TroFuse-011 (NCT06841354) examined the effectiveness of pembrolizumab + Sacituzumab Tirumotecan in patients with previously untreated locally recurrent unresectable advanced or metastatic TNBC expressing PD-L1 at CPS less than 10.

3.1.3 | Monotherapy With PD-L1 Inhibitors

Several early-phase studies have investigated PD-L1 inhibitors as monotherapy for metastatic or locally advanced previously treated TNBC (≤3 prior lines of cytotoxic therapy), showing varying degrees of efficacy. In a phase 1 JAVELIN study (NCT01772004), monotherapy with avelumab was investigated in 168 patients with heavily pretreated metastatic breast cancer, including 58 with TNBC [54]. The ORR was 5.2% in patients with TNBC, and a higher ORR (22.2%) was observed in PD-L1+ patients than in PD-L1− patients (2.6%) [54]. Although this study was a relatively small phase I trial, the results provided a therapeutic rationale for further clinical evaluation of avelumab for patients with advanced TNBC.

The first-in-human phase 1 dose-escalation trial (NCT01375842) evaluated the safety and tolerability of atezolizumab in patients with solid tumors, including TNBC. For patients with TNBC after any number of prior systemic treatments, the mPFS was 1.4 months and the mOS was 17.6 months. Patients with PD-L1+ expression (>1% PD-L1 expression on immune cells) had longer OS than those with PD-L1- expression (<1%) (10.1 months vs. 6.0 months; log rank $p = 0.002$). The ORR was numerically higher in patients who received atezolizumab as first-line therapy than in those who received it as second-line or later therapy (24% vs. 6%) [55].

The exploratory subgroup analysis of 82 mTNBC patients (≤1 prior lines of chemotherapy) from the phase II SAFIR02-BREAST IMMUNO trial (NCT02299999) of durvalumab (vs. maintenance chemotherapy) indicated that durvalumab prolonged OS (21.2 vs. 14.0 months; $p = 0.037$) [56].

3.1.4 | Combination Therapy With PD-L1 Inhibitors

The Impassion130 phase III study (NCT02425891) evaluated atezolizumab (vs. placebo) plus chemotherapy (nab-paclitaxel) in 902 patients with advanced TNBC, who had no previous targeted therapy or chemotherapy [57]. The addition of atezolizumab improved OS (21.0 months) compared to chemotherapy alone (18.7 months) [$p = 0.077$, HR: 0.87 (0.75–1.02)], particularly in PD-L1-positive patients (25.4 vs. 17.9 months) [57]. Although the overall survival benefit was not statistically significant for the entire cohort, the combination showed potential for PD-L1-positive patients. The phase III Impassion131 clinical trial (NCT03125902) evaluated atezolizumab + paclitaxel (vs. placebo + paclitaxel) as first-line treatment in 651 patients with unresectable locally advanced or metastatic TNBC, who had no previous targeted therapy, endocrine therapy, or chemotherapy [58]. In PD-L1-positive patients, PFS (6.0 vs. 5.7 months) and final OS (22.1 vs. 28.3 months) were comparable between the two groups. However, the atezolizumab arm had a higher unconfirmed ORR (63% vs. 55%) and longer median DoR (7.2 vs. 5.5 months), suggesting that combining atezolizumab with chemotherapy may offer durable responses.

Other trials of combination therapies with PD-L1 inhibitors also provided promising results. The phase Ib trial (NCT01633970) of 33 patients with advanced or locally recurrent TNBC (≥2 prior systemic cytotoxic regimens) showed that atezolizumab + nab-paclitaxel was effective, with an ORR of 39.4%, a DCR of 51.5%, a mPFS of 5.5 months, and a mOS of 14.7 months [59]. The phase II randomized COLET trial (NCT02322814) evaluating cobimetinib (a MEK inhibitor) plus taxane chemotherapy (paclitaxel/nab-paclitaxel) with or without atezolizumab as first-line treatment for patients with locally advanced or metastatic TNBC reported a modest clinical response. Respectively, the confirmed ORR was 38.3% versus 20.9% and mPFS was 5.5 months versus 3.8 months in the cobimetinib plus paclitaxel arm versus the placebo plus paclitaxel arm. Moreover, the confirmed ORR was 34.4% versus 29.0% and mPFS was 3.8 months versus 7.0 months, respectively, in the cobimetinib + atezolizumab + paclitaxel versus the cobimetinib + atezolizumab + nab-paclitaxel arms [60]. Of interest, further exploratory analysis for the COLET trial showed a trend toward improved ORR and PFS in PD-L1 positive patients treated with cobimetinib + atezolizumab + paclitaxel/nab-paclitaxel [60]. In addition, in the multicenter, open-label, phase 1/2, MEDIOLA basket trial (NCT02734004), the combination of durvalumab and olaparib (PPAR inhibitor) in the cohort with germline *BRCA*-mutated TNBC (≥2 prior systemic cytotoxic regimens) achieved an ORR of 63.3%, with a mPFS of 4.9 months and a mOS of 20.5 months [61], suggesting combining immunotherapy with DNA repair inhibitors could be particularly effective for this subgroup of TNBC patients. However, a recent phase III trial IMpassion132 in rapidly relapsing PD-L1-positive aTNBC found that atezolizumab plus chemotherapy resulted in an ORR of 40% versus 28% in chemotherapy placebo [HR: 0.95 (0.74–1.22)] [62]. The mPFS was 4.2 (95% CI 3.7–5.6) months in the atezolizumab arm compared to 3.6 (95% CI 3.4–4.2) months in the placebo arm, and the mOS was 12.1 (95% CI 10.1–15.1) in the atezolizumab arm [62] compared to 11.2 (95% CI 9.0–13.3) months in the placebo arm [62], indicating a modest benefit in this challenging patient population.

Several ongoing clinical trials are examining the safety and efficacy of various combination therapies of different PD-L1 inhibitors for locally advanced TNBC or mTNBC, including NCT02685059, which is evaluating the therapeutic efficacy of durvalumab + taxane-anthracycline chemotherapy.

3.1.5 | CTLA-4 Inhibitors

Preliminary clinical evidence suggested that CTLA-4 inhibitors have potential for the treatment of TNBC, but such evidence was based only on several small studies. A pilot study (NCT02536794) showed that second-line durvalumab plus tremelimumab resulted in an ORR of 42.9% (3/7) in patients with mTNBC, and that the responders had a higher CD8 expression and tumor

mutational burden (TMB) than the non-responders [75]. Although many clinical trials were investigating the efficacy and toxicity of ipilimumab for treating solid tumors [76–78], only one pilot study evaluated the safety and tolerability of first-line ipilimumab therapy in patients with early-stage breast cancer [79], and studies on ipilimumab for advanced TNBC are lacking.

3.2 | Early TNBC

3.2.1 | PD-1 Inhibitors

Based on the encouraging results of ICIs for treating locally advanced TNBC or mTNBC, numerous recent studies have been initiated to investigate their efficacy for early TNBC, especially used in neoadjuvant combination therapy.

The phase II I-SPY2 trial showed a significantly higher pathological complete response (pCR) rate in patients with TNBC receiving neoadjuvant pembrolizumab + chemotherapy compared to chemotherapy alone (60% vs. 22%), underscoring the potential benefit of incorporating pembrolizumab early in treatment [63].

The phase Ib KEYNOTE-173 (NCT02622074) trial showed that neoadjuvant pembrolizumab + chemotherapy was tolerable and efficacious for the treatment of early-stage TNBC, with an overall pCR of 60% (range 30%–80%) and both the 12-month EFS and OS rates ranging from 80% to100% among the six arms [67]. Further exploratory analysis also demonstrated that the pCR rate was positively correlated with both PD-L1 expression (defined as CPS ≥ 1) and TILs [67], suggesting a potential predictive role for these markers in response to the treatment combination.

The phase III KEYNOTE-522 study (NCT03036488) of neoadjuvant and adjuvant chemo-immunotherapy in 1174 patients with previously untreated stage II or III TNBC showed that pembrolizumab + chemotherapy significantly improved pCR rate (58.7% vs. 40.0%; $p < 0.001$) [64], 5-year event-free survival (EFS) [81.3% (78.4–83.9) vs. 72.3% (67.5–76.5)] [65] and 5-year OS [86.6% vs. 81.7%; $p = 0.002$, HR: 0.65 (0.51–0.83)] [66] compared to chemotherapy alone. Adverse events were predominantly observed during the neoadjuvant phase of treatment and were consistent with the previously established safety profiles of pembrolizumab and chemotherapy. Based on primary outcomes (pCR and EFS), FDA approved its application for high-risk early-stage TNBC in combination with chemotherapy as neoadjuvant treatment or continued as a single agent as adjuvant treatment after surgery [80].

The phase II AGO-B-041 trial (NCT03289819) showed pembrolizumab in combination with nab-paclitaxel for early-stage TNBC achieved an overall pCR of 66.0% (95% CI: 51.2%–78.8%) [68]. The phase II NeoPACT trial (NCT03639948) reported neoadjuvant pembrolizumab and carboplatin plus docetaxel for TNBC resulted in a pCR of 58% (95% CI, 48%–67%), with an estimated 3-year EFS of 86% in all patients [69]. Another phase II trial (NCT04676997) neoadjuvant camrelizumab plus chemotherapy for early-stage TNBC led to a total pCR rate of 65% [70].

Taken together, these results indicate that neoadjuvant pembrolizumab combined with chemotherapy has significant clinical potential for the treatment of early-stage TNBC. Accordingly, TROPION-Breast04 (NCT06112379) is an ongoing phase III trial assessing neoadjuvant pembrolizumab plus chemotherapy followed by adjuvant pembrolizumab with or without chemotherapy in participants with previously untreated TNBC; ASCENT-05 (NCT05633654) is assessing Sacituzumab Govitecan-hziy and pembrolizumab for TNBC with residual invasive disease after surgery and neoadjuvant therapy; another NCT06393374 is investigating adjuvant sacituzumab tirumotecan + pembrolizumab for TNBC who received neoadjuvant therapy and did not achieve a pCR at surgery; while another NCT05675579 trial is investigating neoadjuvant pembrolizumab + Sacituzumab Govitecan therapy for immunochemotherapy-resistant early-stage TNBC.

3.2.2 | PD-L1 Inhibitors

The phase III IMpassion031 trial (NCT03197935) showed that neoadjuvant atezolizumab + chemotherapy versus chemotherapy alone led to a significantly improved pCR (58% vs. 41%, $p = 0.0044$) in patients with early-stage TNBC [71]. The benefit of atezolizumab was found to be unrelated to PD-L1 status. Atezolizumab-related adverse events were manageable and largely attributed to chemotherapy, marking atezolizumab as a feasible neoadjuvant option for early-stage TNBC.

In the GeparNUEVO phase II window trial (NCT02685059), 174 patients with early-stage primary TNBC were randomized to receive neoadjuvant durvalumab or placebo for 2 weeks, followed by durvalumab or placebo plus nab-paclitaxel for 12 weeks and then by durvalumab or placebo plus epirubicin/cyclophosphamide for 8 weeks. Significantly higher 3-year invasive disease-free survival (iDFS) and OS rates were observed in the durvalumab arm than in the placebo arm (85.6% vs. 77.2%, log-rank, $p = 0.036$ and 95.2% vs. 83.5%, $p = 0.006$, respectively) [72]. Among patients who achieved a pCR, the 3-year iDFS rate was also significantly higher in the durvalumab arm than in the placebo arm (95.5% vs. 86.1%). Multivariate regression analysis confirmed the long-term iDFS and OS efficacy for durvalumab, which was independent of pCR.

A phase II trial (NCT02530489) evaluated the efficacy of atezolizumab and nab-paclitaxel as the second phase of neoadjuvant therapy in patients with anthracycline-resistant early-stage TNBC [73], achieving a 46% pCR rate. Other clinical trials of neoadjuvant therapy with PD-L1 inhibitors are ongoing, including the GeparDouze study (NCT03281954) of neoadjuvant chemotherapy with atezolizumab followed by adjuvant atezolizumab in patients with TNBC, the A-BRAVE study (NCT02926196) of adjuvant avelumab therapy in patients with high-risk TNBC, and the Impassion030 study (NCT03498716) of atezolizumab combined with adjuvant anthracycline/taxane-based chemotherapy in patients with operable TNBC.

3.2.3 | CTLA-4 Inhibitors

Although some CTLA-4 inhibitors, such as ipilimumab, have been approved for use as monotherapy or in combination with

a PD-1/PD-L1 inhibitor for treatment of various malignancies [81–83], very few studies have investigated their efficacy for treating breast cancer. One pilot study of 19 patients with early-stage breast cancer showed that single-dose ipilimumab monotherapy or combination therapy with ipilimumab and cryoablation were safe and tolerable, and combination therapy was associated with increased levels of T helper type 1 cytokines as well as CD4+ and CD8+ T cells [79]. Additional clinical trials of CTLA-4 inhibitors for the treatment of early-stage TNBC are needed.

These trials collectively emphasize that ICIs, particularly pembrolizumab and atezolizumab, can improve outcomes in early-stage TNBC, with a growing body of evidence supporting their integration into neoadjuvant and possibly adjuvant settings. Future research is essential to validate biomarkers like PD-L1 and TILs for selecting patients who would benefit most from ICI-based therapies.

4 | Predictive Biomarkers for Response to ICIs

4.1 | PD-L1 Expression

PD-L1 expression has shown potential as a biomarker across solid tumors, including TNBC. Patel et al. [84] tested 654 tumor specimens from different sites and showed that the PD-L1 expression level was higher in TNBC than in other hormone-driven tumors of breast cancers (18% vs. 0%). Gatalica et al. [85] revealed that PD-L1 was significantly over-expressed in TNBC compared to luminal-type tumors (odd ratio = 1.70, $p < 0.001$). Zhang et al. [86] reported that PD-L1 expression was common in TNBC. In addition, multiple studies have provided evidence that PD-L1 is a useful biomarker for predicting the efficacy of ICIs in other solid tumors [87–89]. Despite these studies, the predictive value of PD-L1 as a reliable biomarker of treatment response to ICIs remains controversial, as many patients with PD-L1-negative tumors also respond to ICIs [90].

As previously described, clinical trials have demonstrated that ICIs improve therapeutic outcomes in patients with PD-L1 positive breast tumors, including TNBC [43, 54]. In some of these trials, PD-L1 expression levels were evaluated as biomarkers to be associated with or predict the therapeutic efficacy. In the randomized phase III KEYNOTE-119 trial, patients with mTNBC who had received previous systemic treatments for metastasis were randomized to receive second-line or third-line pembrolizumab monotherapy or chemotherapy. The ORR was similar between the two arms (30% vs. 33%). However, the ORR was correlated with PD-L1 levels in the pembrolizumab arm; the rates were 12%, 18%, and 26%, respectively, in patients with CPS ≥ 1, CPS ≥ 10, and CPS ≥ 20, whereas the rates were 9%, 9%, and 12%, respectively, in the chemotherapy arm [46]. In the phase 3 KEYNOTE-355 trial, patients with mTNBC were randomized to receive first-line pembrolizumab + chemotherapy or placebo + chemotherapy. The median PFS was longer in the pembrolizumab + chemotherapy arm than in the placebo + chemotherapy arm (7.5 vs. 5.6 months). Moreover, the median PFS in the pembrolizumab + chemotherapy arm, with CPS ≥ 1 and CPS ≥ 10, were 7.6 and 9.7 months, as compared to 5.6 months in both CPS populations in the placebo + chemotherapy arm [50].

In the phase III IMpassion130 study, first-line atezolizumab + nab-paclitaxel demonstrated longer PFS and numerically improved OS in patients with TNBC who had ≥ 1% PD-L1–expressing tumor-infiltrating immune cells, compared with nab-paclitaxel alone [57]. In the biomarker analysis of KEYNOTE-086 (NCT02447003), baseline tumor PD-L1 positive status was associated with improved clinical efficacy of pembrolizumab monotherapy [91]. Similarly, the JAVELIN study showed that a higher PD-L1 expression was associated with improved efficacy of atezolizumab in patients with mTNBC [52]. These studies have demonstrated that PD-L1 levels can identify those patients who can benefit from ICIs in advanced TNBC. In contrast, trials in early-stage TNBC, such as KEYNOTE-522 and IMpassion 031, demonstrated that the efficacy of neoadjuvant ICIs appears to be independent of PD-L1 expression [71, 92].

Potential biomarkers based on PD-L1 expression for the identification of patients with TNBC who respond to ICI treatment are shown in Table 2. It is important to note that although PD-L1 expression is regularly used to guide the selection of single-agent immunotherapy or combination therapy, inconsistencies may result due to variations in the methods to detect PD-L1 expression, the criteria to define PD-L1 positivity, and the heterogeneous nature of PD-L1 expression. Therefore, for current clinical applications, other predictive biomarkers for the response to ICI therapy in addition to PD-L1 are needed.

4.2 | TILs

Considering the complex mechanisms underlying tumor immunity, parameters related to TILs in the tumor microenvironment have been suggested as important biomarkers for the promotion of tumor immunogenicity [93]. Patients with TNBC have a higher proportion of tumor-infiltrating immune cells and higher TIL activity than those with other types of breast cancers, potentially making them respond better to ICI therapy [37, 94–98].

Dieci et al. [99] retrospectively analyzed the use of TIL levels after chemotherapy for predicting the prognosis of 278 patients with TNBC and reported a 5-year OS rate of 91% for patients with high levels of TILs (intratumoral and stromal TILs > 60%) compared with only 55% for patients with low levels of TILs, indicating that patients with high TILs obtain greater benefit from chemotherapy. In addition, Denkert et al. [96] showed that a high level of TILs (stromal TILs > 60%) was associated with longer survival in patients with TNBC, and a high residual level of TILs after neoadjuvant chemotherapy corresponded to a more favorable prognosis. A real-world study of 108 patients with TNBC who had at least 5 years of follow-up after surgical resection showed that those with high-level TILs (TILs > 60%) had significantly longer survival and better prognosis ($p < 0.05$) [98]. Moreover, in the biomarker analysis of KEYNOTE-086 (NCT02447003), stromal TILs were associated with improved clinical efficacy of pembrolizumab monotherapy [91]. Lotfinejad et al. conducted a meta-analysis of seven studies with 1152 patients and showed that TILs levels were a significant indicator for long-term OS and DFS [100]. Nevertheless, none of these studies evaluated the association between TIL levels and the

response to ICI combination therapy. An initial data analysis for the phase Ib KEYNOTE-173 trial [67] in patients with early-stage TNBC showed that those with higher pre- and on-treatment TIL levels had a higher pCR rate with neoadjuvant pembrolizumab + chemotherapy. However, whether the level of TILs is an independent predictor or prognostic factor for patients with early-stage TNBC in response to ICI therapy requires further investigation.

4.3 | TMB

In addition to PD-L1 expression, TMB, defined as the number of somatic mutations per megabase in the genomic sequence of targeted genes, has consistently been shown to be an important biomarker for predicting the treatment efficacy with ICI therapy across multiple cancer types [101]. In CHECKMATE-032, high TMB was associated with greater clinical improvement following ICI treatment in patients with small cell lung cancer [102]. Samstein et al. [103] demonstrated an association between high TMB and OS in ICI-treated patients ($n = 1662$), but not in non-ICI-treated patients ($n = 5371$). However, the heterogeneity of different cancer types and a lack of consensus regarding the cut-off value for defining high TMB remain challenges to the use of TMB as a biomarker for predicting treatment efficacy [104, 105]. Barroso-Sousa et al. [106] performed whole exome sequencing for patients with breast cancer and reported that the incidence of hypermutation was only 5%, with the rate being higher in metastatic tumors than in primary tumors, and the median TMB was 2.63 mut/Mb, which was different among tumor types and between metastatic and primary tumors. In addition, they also observed that patients with TMB ≥ 10 mut/Mb appeared to benefit from pembrolizumab-based therapy [106]. In a retrospective study of patients with TNBC who received ICI therapy in multiple clinical trials, patients with high TMB (≥ 10 mut/Mb) benefited more than those with low TMB (< 10 mut/Mb) from pembrolizumab-based therapy (PFS: 12.5 vs. 3.7 months, $p < 0.05$) [107]. These findings were confirmed by the KEYNOTE 158 (NCT02628067) study, in which high TMB (≥ 10 mut/Mb) was significantly associated with tumor response to pembrolizumab monotherapy in patients with previously treated, advanced solid tumors, including TNBC [105], and the biomarker analysis of the KEYNOTE-086 study, in which TMB levels were associated with improved clinical efficacy of pembrolizumab monotherapy in patients with previously treated mTNBC [91]. Additional studies, like the NIMBUS (NCT03789110) phase II prospective clinical trial, are currently in progress and expected to determine whether high TMB can be an adequate biomarker for predicting the survival time of mTNBC patients treating with ICIs.

4.4 | Emerging Biomarkers

In addition to PD-L1 CPS, sTIL, and TMB, other genomic indices have emerged as potential biomarkers for TNBC. For example, defective mismatch repair (MMR) genes have been established for predicting patient prognosis in many solid tumors, especially colorectal cancer [108, 109]. Özcan et al. [110] observed complete/partial loss of MMR, with or without high TIL levels, in a substantial proportion of patients with PD-L1 negative TNBC, suggesting that these biomarkers in addition to PD-L1 may help select patients who would derive the greatest benefit from ICI immunotherapy. However, the incidence of defective MMR and microsatellite instability-high (MSI-H) in patients with TNBC is extremely low [111], which may limit their predictive value as biomarkers.

Phosphatase and tensin homolog (PTEN), a tumor suppressor that downregulates Akt/PKB signaling, is involved in the complex molecular mechanism regulating PD-L1 expression [112]. PTEN knockdown leads to upregulation of PD-L1 and inhibition of T-cell proliferation, and agents targeting the PI3K pathway might increase adaptive immune responses [113]. Approximately half of TNBC tumors with PD-L1 expression have PTEN deletions [107]. A study in Chinese patients showed that PTEN deletion was present in 35% of basal-like breast tumors (mostly TNBC) [114]. Sasak et al. found that the PTEN loss rate was higher in tumors with high TIL levels than in tumors with low TIL levels (60% vs. 25%) [115]. Tavares et al. evaluated the association between PTEN expression and clinical outcomes and reported that lack of PTEN expression was associated with improved OS in non-metastatic TNBC [116]. Iqbal et al. showed that a high percentage of PTEN loss and increased insulin growth factor receptor-1 expression can predict early recurrence in TNBC [117].

Beyond genomic biomarkers, several studies evaluated the association between a series of potential biomarkers in the tumor microenvironment and clinical outcomes in patients with TNBC who were treated with pembrolizumab monotherapy. An exploratory analysis of the FUTURE trial suggests that a CD8 immunohistochemical score may be a potential biomarker for predicting immunotherapy outcomes in patients with the immunomodulatory subtype of TNBC [118]. Recently, the KEYNOTE-119 (NCT02555657) trial showed that the T-cell inflamed gene expression profile (GEP) was significantly associated with better clinical outcomes in patients with TNBC who were treated with pembrolizumab [91, 119]. In addition, the KEYNOTE-086 (NCT02447003) trial revealed that the T-cell-inflamed GEP was significantly associated with improved ORR, PFS, and OS with pembrolizumab monotherapy in TNBC [91, 119]. Notably, the sample sizes of these biomarker studies were small, which may limit the predictive value.

In conclusion, while PD-L1 remains a key biomarker for selecting patients with TNBC for ICI therapy, it has limitations in accurately predicting outcomes. Emerging biomarkers like TMB, TILs, and GEP offer additional predictive value, highlighting the need for a multi-biomarker approach. Additional research is essential to establish these markers' predictive value, optimize patient selection, and improve treatment outcomes in TNBC.

5 | Future Perspectives

Besides currently established immune checkpoints, future research may extend to explore emerging targets such as lymphocyte activation gene-3 (LAG-3), T cell immunoreceptor with Ig and ITIM domains (TIGIT), and T cell immunoglobulin and

TABLE 2 | Application of different biomarkers for predicting the response to ICI treatment in TNBC.

Biomarkers	Trials	Biomarker grouping	ORR	mPFS (months)	mOS (months)
Locally advanced or metastatic TNBC					
PD-L1	NCT02447003 KEYNOTE-086 Cohort A [44]	CPS ≥ 1	5.7%		
		CPS < 1	4.7%		
	NCT02555657 KEYNOTE-119 [46]	CPS ≥ 1	12.3%		10.7
		CPS ≥ 10	17.7%		12.7
		CPS ≥ 20	26.3%		14.9
	NCT02657889 KEYNOTE-162 [49]	PD-L1+	32%		
		PD-L1−	8%		
	NCT02819518 KEYNOTE-355 [74]	CPS ≥ 1	44.9%	7.6	17.6
		CPS ≥ 10	52.7%	9.7	23.0
	NCT01772004 JAVELIN [54]	≥ 1% tumor cells	2.4%	5.9 weeks	6.5
		≥ 5% tumor cells	4.3%	6.0 weeks	6.5
		≥ 25% tumor cells	0	6.0 weeks	9.2
		≥ 10% tumor-associated ICs	16.7%	6.1 weeks	11.3
	NCT03125902 IMpassion131 [58]	≥ 1% tumor cells		6.0	22.1
	NCT02322814 COLET [60]	≥ 1% tumor-infiltrating ICs	39.0%	7.0	
	NCT02513472 KEYNOTE-150 Stratum 1	CPS ≥ 1	34.5% 24.4%		
	KEYNOTE-150 Stratum 2				
	NCT03797326 LEAP-005 [53]	CPS ≥ 10	50%		
		CPS < 10	27%		
		< 1% tumor-infiltrating ICs	19%	3.7	
PD-L1 with others	NCT02425891 IMpassion130 [16]	≥ 1%, ICs		7.6	25.0
		< 1%, ICs		5.6	19.7
TMB	NCT02555657 KEYNOTE-119 [46]	≥ 10	14.3%		
		< 10	12.7%		

(Continues)

TABLE 2 | (Continued)

Biomarkers	Trials	Biomarker grouping	ORR	mPFS (months)	mOS (months)
TILs	NCT02425891 IMpassion130 [16]	sTILs < 10%		5.6	19.2
		sTILs ≥ 10%		8.3	25
Other Biomarkers	NCT02425891 IMpassion130 [16]	CD8 < 0.5%		5.6	19.2
		CD8 ≥ 0.5%		7.4	22.6
		BCRA mutant		7.4	28.9
		BCRA wild-type		7.2	20.8
Early TNBC					
	NCT02685059 GeparNUEVO [72]	PD-L1 ≥ 1% tumor cells	pCR 54.3%		
		PD-L1 < 1% tumor cells	pCR 30.0%		
		sTILs < 10%	pCR 36.4%		
		sTILs ≥ 10%	pCR 56.5%		
	NCT03639948 NeoPACT [69]	PD-L1+	pCR 75%		
		PD-L1−	pCR 40%		
		sTILs ≥ 30%	pCR 74%		
		sTILs < 30%	pCR 42%		

Abbreviations: CPS: combined positive score; ICs: immune cells; mOS: median overall survival; mPFS: median progression-free survival; ORR: objective response rate; pCR: pathologic complete response; PD-1: programmed death protein 1; PD-L1: programmed cell death ligand-1; sTILs: tumor-infiltrating immune cells; TNBC: triple-negative breast cancer.

mucin domain-containing protein 3 (TIM-3). Recent studies have demonstrated the potential clinical relevance of these molecules in TNBC. High levels of LAG-3 expression have been detected in TNBC patients, highlighting its role in tumor immunology [120, 121]. A Phase I/II study (NCT02460224) investigated the safety and efficacy of LAG525 alone or in combination with spartalizumab in patients with advanced malignancies, including TNBC, showing promising antitumor activity [122]. Moreover, the ongoing NCT06259162 trial is evaluating the relationship between LAG-3 expression and treatment outcomes in neoadjuvant chemotherapy combined with ICI. The NCT03499899 trial is also assessing the efficacy and safety of LAG525 (an anti-LAG-3 antibody) in combination with spartalizumab, with or without carboplatin, in advanced TNBC. Similarly, the TIGIT pathway is emerging as a promising therapeutic target [123]. Additionally, TIM3 expression on TILs has been associated with poor responses to neoadjuvant chemotherapy in patients with locally advanced TNBC [124]. These findings underscore the need for further investigation of LAG3, TIGIT, and TIM3 as potential therapeutic targets, which may ultimately lead to more personalized and effective treatment strategies for TNBC patients.

Despite the encouraging preclinical and clinical data that have introduced ICIs into clinical practice and helped shape the strategies for the diagnosis and treatment of TNBC, several unresolved issues remain. The first issue is identifying the patients most likely to benefit from ICIs. TNBC is highly heterogeneous, leading to considerable variability in responses to ICIs. Although PD-L1 is the most commonly used biomarker, its predictive reliability is limited due to inconsistencies in detection methods and cutoff thresholds across assays, as evidenced in trials like KEYNOTE-355 and IMpassion130. These differences highlight the need for standardized PD-L1 testing to improve patient screening. Other potential biomarkers, including TMB, MMR status, and TIL levels, also show promise but currently lack universally accepted cut-off values. For instance, while TMB has been associated with better responses to ICIs, variations in testing and its low incidence in TNBC limit its broad applicability as a clinical marker. Furthermore, distinct immune microenvironment profiles in early- versus advanced-stage TNBC may require stage-specific biomarker analyses. Addressing these gaps will be essential for refining patient stratification and optimizing ICI-based therapeutic strategies.

Optimizing treatment regimens is another area of focus, as ICI monotherapy has shown limited efficacy in TNBC. Combination therapies, such as ICIs with chemotherapy, targeted agents, or ADCs, are now under active investigation, with early studies demonstrating enhanced responses. Several ADCs such as Sacituzumab govitecan, Ladiratuzumab vedotin, Trastuzumab deruxtecan (DS-8201a) and Glembatumumab vedotin have

shown promising efficacy for TNBC, and Sacituzumab govitecan has been approved for advanced TNBC in the US and China [125, 126]. ICIs with these ADCs may synergize by leveraging ADCs' tumor-targeted cytotoxicity to induce immunogenic cell death, thereby enhancing antigen release and T-cell activation. These combinations aim to boost the immune response to tumors, and ongoing trials are assessing their efficacy and safety in more diverse patient populations.

Lastly, a deeper understanding of TNBC immunogenicity is crucial for better ICI response prediction. TNBC frequently has high TIL counts, which correlate with improved prognosis and ICI response, but the exact mechanisms driving immunogenicity and immune resistance are not yet fully understood. Further research is needed to identify new biomarkers or therapeutic targets, allowing for a more personalized approach to immunotherapy.

6 | Conclusions

Many clinical trials have been completed or are underway to determine the efficacy of ICIs in patients with TNBC. In general, improved responses to treatment with different combinations of ICIs with radiotherapy, chemotherapy, and targeted therapy have been observed. Therefore, such combination therapies are a main direction for the treatment of TNBC. Besides, the identification of the most appropriate predictive biomarkers (e.g., whether individual biomarkers such as TMB, TIL level, and PD-L1 expression or a combination of several of these biomarkers) to predict responses to ICI immunotherapy is a key focus of current research related to TNBC treatment. Standardizing these markers and refining treatment strategies will be essential for optimizing outcomes and advancing precision medicine in TNBC.

Author Contributions

All authors contributed to the conception, design, or planning of the study. Lingxia Wang contributed to drafting the manuscript. Xinran Wang and Yueping Liu contributed to reviewing or revising the manuscript for important intellectual content. All authors read and approved the final version of the manuscript.

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The authors have nothing to report.

Consent

The authors have nothing to report.

Conflicts of Interest

Lingxia Wang is an employee of MSD China. The other authors declare no conflicts of interest.

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The authors have nothing to report.

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