Immune checkpoint inhibitors a new player in the therapeutic game of mesothelioma: New reality with new challenges

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Abstract

Malignant pleural mesothelioma (MPM) is a rare and orphan thoracic malignancy, with a poor prognosis as majority of patients are diagnosed as unresectable MPM with no significant improvements in the therapeutic strategy for over a decade. However, the recent approval of immune checkpoint inhibitors (ICI) in treatment-naïve patients with unresectable MPM marks a significant step forward fand hope for the treatment of this disease. In this narrative review, we discuss the biological rationale to use ICI in the treatment of MPM. Indeed, we summarize the current evidence for the efficacy of ICI in MPM and discuss several unresolved challenges regarding the use of ICI in this disease such as the best upfront immune approach in MPM (ICI versus ICI plus chemotherapy), the optimal sequential treatment approach according to first-line treatment, and potential role of predictive biomarkers.

Keywords: mesothelioma, immune checkpoint inhibitors, nivolumab, pembrolizumab, ipilimumab

Introduction

Malignant pleural mesothelioma (MPM) is a rare and orphan thoracic malignancy, accounting for 2.1 cases per 100,000 people per year, and including epithelioid, sarcomatoid and biphasic histologic subtypes [1]. MPM is usually diagnosed at advanced stages due to the absence of early symptoms, which reduces the options for radical-curative approaches. While we are witnessing years of exciting progress for patients with advanced lung cancer patients, limited therapeutic improvements have been made in unresectable MPM since the approval of platinum-pemetrexed in 2004 as the backbone chemotherapy strategy [2,3]. In the phase 3 MAPS trial, the addition of bevacizumab to first-line platinum-pemetrexed chemotherapy improved the overall survival (OS) over chemotherapy alone. However, this strategy is not worldwide accepted by health authorities as bevacizumab was not compared with placebo [4], as well as in the phase 3 LUME-Meso trial, the addition of nintedanib (a multi-tyrosine kinase inhibitor with antiangiogenic properties) to chemotherapy did not increase survival as compared with placebo of patients with epithelioid unresectable MPM [5]. Finally, the U.S. Food and Drug Administration (FDA) approved in the first-line setting the tumor treating fields (TTFields) in combination with platinum-pemetrexed based on the OS results of the single arm phase II STELLAR trial, even if firm conclusions about the real efficacy of this strategy are hampered by the lack of a control arm [6].

With the aim to extend the survival benefit of induction platinum-pemetrexed, maintenance chemotherapy has also been explored in MPM. The phase II ALLIANCE study reported that pemetrexed as maintenance after four cycles of induction platinum-pemetrexed chemotherapy did not improve survival as compared to placebo. [7]. In contrast, in the phase II NVALT19 trial, switch maintenance with gemcitabine after first-line platinum-pemetrexed chemotherapy, improved progression free survival (PFS) compared with placebo, but the OS impact of this strategy remains to be proven as the trial was not powered for this [8]. Therefore, there is insufficient evidence to support maintenance treatment in patients with MPM [9]. For second-line, no standard treatment approach exists, available options are of limited efficacy and based on scant evidences [9]. Not surprisingly, based on this desertic journey in the therapeutic landscape, the life expectancy of patients

with MPM remains suboptimal with a median OS of less than 1 year and a 5-year OS of approximately 5% [10].

Recently, the introduction of immune checkpoint inhibitors (ICI) in the therapeutic strategy of MPM has shed a new light of hope for this orphan disease, especially in first-line setting. In this review, we provide a summary of the recent therapeutic advances in MPM in light of ICI, as well as current challenges of this strategy regarding the best immune approach in the first-line setting and the role of sequential treatment strategies at progression.

Immunophenotype in MPM

The evolution of mesothelioma is strongly contingent on the inflammatory response to asbestos. The persistence of asbestos fibers in the pleural cavity results in long-term activation of macrophages, chronic inflammation and in the end transformation of mesothelial cells to MPM [11]. MPM cells induce an immunosuppressive tumor microenvironment (TME) by for example attracting cancer associated fibroblast and myeloid cells (the latter usually being immunosuppressive in MPM). Immune promoting myeloid cells switch to an immunosuppressive phenotype upon cytotoxic T-cell influx with the upregulation of PD-L1. These in turn inhibit cytotoxic T-cell influx [12]. PD-L1 expression occurs in 18% to 53% of MPM samples, being more frequently expressed in non-epithelioid histologies and the detection of PD-L1 is almost invariably associated with worst outcomes [13–18]. Of note, immunologic phenotypes in MPM differ based on PD-L1 status and histologic subtype. PD-L1 positive MPM tend to have higher infiltration of CD45+ CD3+ CD8+ T cells than PD-L1 negative tumors. Additionally, CD8+ T cells in PD-L1 positive tumors also tend to have a higher expression of the inhibitory markers PD-1 and TIM-3. Likewise, non-epithelioid MPM is also associated with higher CD8 positive T-cell infiltration in the tumor microenvironment than epithelioid histology [19]. Indeed, MPMs also contain abundant CD68+ and CD163+ macrophages with comparable levels across histology subtypes and association with shorter survival, suggesting a negative effect of these myeloid cells [20]. This data suggests the immune-complexity of tumor microenvironment in MPM.

Based on immune gene expression analysis, an attempt has been made to classify MPMs into 3 distinct sub-entities in 87 archival tumors from advanced-stage MPM. Forty percent of cases were classified in group 1 (immune desert), whereas the rest were classified in group 2 (higher B-cell and antigen presentation-related gene expression) and group 3 (higher T-cell related gene expression), suggesting that a significant number of MPMs are inflamed tumors [21]. In other cohort of 516 MPM samples three different groups could be made based on presence of T-helper 2 and cytotoxic T-cells. The group with low T-helper 2 cells and high cytotoxic T-cell levels (8.5% of the total group) had the best survival, and on a transcriptional level, upregulation of immune pathways was observed in this goup [22]. Therefore, it suggests a novel immune-based signature with potential clinical relevance.

Characterization of the immune microenvironment has also led to identification of new markers of immune suppression. V-domain Ig-containing suppressor of T-cell activation (VISTA) is one such novel immune checkpoint that is present in up to 85% of patients with mesothelioma, almost exclusively in epithelioid mesothelioma. VISTA is also expressed on T-cells and is involved in suppression of T-cell activation [23]. VISTA and PD-L1 were expressed on tissue microarray of immunotherapy-naive MPMs in 85% and 38% of samples, respectively, with significantly higher expression of VISTA in epithelioid subtype, whereas PD-L1 was significantly higher in sarcomatoid tumors compared with other subtypes [24]. Despite its functions as an immunosuppressive molecule, expression of VISTA has been associated with improved survival in mesothelioma independent of histology subtype [23,24].

Despite typically having low tumor mutational burden (TMB), as detected by next-generation sequencing (NGS) technologies [25], MPMs may obtain favorable outcome with ICI (see below). This could be related with potential neoantigen expression driven by structural chromosomal rearrangements. This fact, along with the possible contribution of germline mutations and the exposure to asbestos, may explain a high neoantigen burden in MPM resulting in ICI efficacy despite an otherwise low NGS-defined TMB and a variable PD-L1 expression [26,27]. Consequently, this evidence points to a subset of patients with MPM, who might benefit with immunotherapy-based regimens.

Single-agent ICI therapy in the salvage setting

In patients with advanced MPM who progressed on previous platinum-based chemotherapy, the guidelines recommend different strategies such as retreatment with pemetrexed (specially in patients with progression at least 6 months after the first-course of pemetrexed), vinorelbine or gemcitabine [9]. However, this resulted in limited outcomes with a response rate (RR) ranging from 9.8% to 19%, median PFS from 2.3 to 3.3 months, and median OS below to 10 months [28–30]. Similar results have been reported in a single phase II trial with lurbinectedin as second- or third-line (RR 5%, PFS 4.1 months, and median OS: 11.1 months), showing activity regardless of histology, or outcome on prior treatment [31]. Therefore, new therapeutic approaches are eagerly awaited.

Several phase Ib/II clinical trials either with pembrolizumab [13,32], nivolumab [33–35] or avelumab [36], as well as real word data cohorts [37–39] have assessed the role of ICI in previously treated MPM. In this subset, ICI reported a RR of ~20% and median PFS and OS of 4 and 12 months, respectively, with half of the patients alive at 1 year (Table 1). Although these results have to be interpreted cautiously as patients enrolled in phase Ib trials may be overselected (and only PD-L1 positive tumors allowed) [13], globally these results in terms of RR and OS were slightly better than historical data, especially in terms of long-term benefit.

Based on these promising results two phase 3 clinical trials were launched, assessing the role of ICI in patients with previously treated unresectable MPM: the PROMISE [40] and the CONFIRM trials [41], which have reported opposite results in terms of survival benefit (Table 1). The PROMISE trial randomized 144 patients with PD-L1 unselected MPM with progression on previous platinum-based chemotherapy and unselected for PD-L1 status to pembrolizumab (200 mg every 3 weeks, Q3W) versus institutional choice of single-agent chemotherapy (gemcitabine or vinorelbine). Although pembrolizumab significantly improved the RR over chemotherapy (22% *vs*. 6%, p=0.002), pembrolizumab neither improved the primary PFS endpoint by independent review (2.5 *vs*. 3.4 months, HR: 1.06; 95% CI: 0.73-1.53; p = 0.76) nor the OS (10.7 *vs*. 12.4 months; HR = 1.12, 95% CI: 0.74-1.69; p = 0.59), even after adjusting for crossover. Grade 3-5 treatment-related adverse events (TRAEs) occurred in 19.4% of patients in the pembrolizumab arm and in

25.7% of patients in the chemotherapy arm, leading to treatment discontinuation in 6.9% and 7.1% of cases, respectively [40]. The CONFIRM trial randomized (2:1) 332 patients with previously treated MPM to nivolumab or placebo. Nivolumab compared with placebo achieved the two co-primary investigator-assessed endpoints with longer PFS (3.0 *vs* 1.8 months, HR 0.62; 95% CI, 0.49-0.78; p<0.001) and OS (9.2 *vs.* 6.6 months; HR, 0.72; 95% CI: 0.55-0.94; p < 0.02, still immature). Grade 3-4 TRAEs were reported in 19% in the nivolumab arm and in 6.3% on the placebo arm. Treatment discontinuation due to toxicity occurred in 13.1% and 2.7%, respectively [41].

Although indirect-trial comparisons should be undertaken with caution, baseline characteristics in both trials were similar in terms of epithelioid histology (90% *vs*. 88%), patients with ECOG 0 (29% *vs*. 20%) and median age (69 years *vs*. 70 years) [40,41]. However, a higher proportion of PD-L1 positive tumors was included in PROMISE compared with the CONFIRM (63% *vs*. 37%) and the PROMISE only allowed one previous line before pembrolizumab, whereas 56% of patients in CONFIRM trial received nivolumab as third-line treatment strategy [40,41]. Finally, in the PROMISE trial the comparator arm was chemotherapy and 63% of patients received pembrolizumab at progression, with no survival improvement after adjusting for crossover [40]. In contrast, in the CONFIRM trial nivolumab was compared with placebo and only 13% of patients in the placebo arm received nivolumab at progression [41]. Despite these differences and based on the results coming from the CONFIRM trial, ICI is now a potential rational and medically useful option for patients with unresectable, platinum-relapsed MPM, in the absence of any contraindication.

Two single-arm phase 2 trials with tremelimumab (an anti-CTLA4) in second-line in patients with MPM showed good activity (disease control rate, DCR 31-52%, median PFS and OS of 6.2 and 11 months, respectively) [42,43]. By contrast, in the randomized phase 2b DETERMINE study, second- or third-line treatment with tremelimumab did not improve the OS compared with placebo in patients with unresectable mesothelioma (7,7 months *vs.* 7,3 months, HR 0·92, 95% CI: 0.76-1·12, p=0.41) [44].

Combination ICI strategies in the salvage setting

With the aim to boost the immune response in MPM [40], and given the results observed in other solid tumors [41-43], three phase II trials have explored the combination of PD(L)-1 and CTLA4 inhibitors in MPM . The efficacy of nivolumab plus ipilimumab has been assessed in the INITIATE [45] and MAPS2 trials [35], whereas the NIBIT-MESO-1 trial [46] evaluated durvalumab plus tremelimumab (Table 2). Although a combination strategy may lead to interesting DCRs compared with monotherapy, adding anti-CTLA4 to anti-PD-1 is associated with an increased risk of adverse events (AEs), especially of grades 3–4 (26% *vs.* 14%, for the combination and the single agent PD-1 inhibitor, respectively, in the MAPS2 non-comparative randomized trial) [35]. The safety profile, along with the lack of randomized comparisons with other treatments and the new potential strategies in the first-line setting that include ICI limit the applicability of this combination in daily clinical practice in the second-line setting.

ICI in first-line setting

Based on the encouraging activity of the ICI combination in thesalvage setting, this strategy was tested in the first line setting. The phase 3 CheckMate 743 trial, randomized 605 patients with unresectable MPM to receive either nivolumab and ipilimumab (n=303) or platinum plus pemetrexed chemotherapy (n=302). Notably, patients were not selected by histology nor PD-L1 status. ICI significantly improved OS by 4 months compared to chemotherapy, with median OS of 18.1 versus 14.1 months (HR 0.74, 96.6% CI 0.60–0.91; p=0,0020. Figure 1). The 2-year OS was 41% and 27%, respectively, despite the fact that 20% of patients in the control arm received ICI at the time of progression. The difference in median OS was more pronounced in patients with non-epithelioid histology (18.1 *vs*. 8.8 months, HR 0.46, 95% CI 0.31-0.68), and in PD-L1 positive (cut-off ≥ 1% by 28-8 assay test) tumors (18.0 *vs*. 13.3 months; HR 0.69, 95% CI 0.55-0.87). Of note, the proportion of grade 3-4 TRAEs were similar in both arms (˜30%) [47]. Based on these results, the

combination of nivolumab and ipilimumab was approved by the FDA in October 2020 for previously untreated patients with unresectable or advanced MPM.

Chemotherapy can elicit immune stimuli in mesothelioma models [48,49]. The combination of chemotherapy plus ICI has been explored in two single arm phase 2 trials in patients with untreated and unresectable MPM, the DREAM [14] and the Pre505 trials [50]. Both trials have assessed the combination up to 6 cycles of durvalumab plus platinum and pemetrexed, followed by maintenance durvalumab if no progression (Figure 1). Both trials have reported very promising results, with a RR of ~50%, median PFS of ~7montts, and median OS of 18.4 to 20.4 months, with 70% and 40% of patients alive at 1-year and 2-years, respectively. The combination was well tolerated in both studies [14,50]. These chemotherapy plus ICI results provide a new therapeutic approach to be explored in MPM.

Currently, three ongoing phase 3 trials are exploring different immune-strategies in first line setting in patients with unresectable MPM. The DREAM3R trial (NCT04334759), explores durvalumab plus platinum-pemetrexed versus chemotherapy alone; the IND227 trial (NCT02784171) evaluates pembrolizumab-chemotherapy –versus chemotherapy alone; and finally, the BEAT-Meso trial (NCT03762018) compares atezolizumab plus bevacizumab and platinum-pemetrexed to platinum-pemetrexed-bevacizumab. The primary endpoint of all these trials is OS.

Controversies

Several challenges regarding ICI in MPM remain to be answered in the coming future (Figure 2)

Best treatment option in the first-line setting

Nivolumab plus ipilimumab has been adopted as the new potential standard of care in the first-line setting, however, chemo-immunotherapy combinations resulted in promising results in phase 2 clinical trials (Figure 1) potentially mirroring the data

reported in the Checkmate 743 trial, questioning about the best upfront treatment approach.

In the CheckMate 743 trial, analysis of OS data clearly shows that the effect of nivolumab plus ipilimumab across the ITT population is heterogeneous: in the overall population, PFS curves and OS curves cross approximately seven and four months, respectively, after treatment initiation, with chemotherapy performing better than the ICI doublet during this time period. This pattern is even more relevant in PD-L1 negative MPM. In this subset, the PFS curves with ICI combination underperforms compared with chemotherapy during the first 16 months after randomization. Altogether this suggests that a substantial number of patients progress rapidly and die within the first months under treatment without obtaining any meaningful benefit from immunotherapy [47]. These data also highlight the potential risk of hyper-progressive disease with ICIs in a largely unselected patient population. Although this pattern of progression has not been reported in the CheckMate 743 trial, in the salvage setting, an exploratory analysis from the MAPS2 trial reported ~7% of patients with hyper-progressive disease (50% increase of the sum of the diameters of largest lesions) during treatment with nivolumab plus ipilimumab [51]. In patients with advanced non-small cell lung cancer (NSCLC) unselected for PD-L1 status, the addition of chemotherapy to immunotherapy in the first-line setting has avoided the crossover of survival curves [52] thus reducing the risk of hyper-progressive disease [53]. Following the same principle, an upfront chemo-immunotherapy combination would possibly give similar results in MPM, especially in PD-L1 negative patients, although it must be confirmed in phase 3 clinical trials.

The non-epithelioid MPM subtypes (sarcomatoid and biphasic) are characterized by worst outcomes due to their aggressiveness and chemo-resistance. However, these seems benefit the most from ICI. Albeit Checkmate 743 is formally positive in the intention-to-treat population, the subgroup analysis, powered by the stratification based on histology, clearly suggests a superiority of nivolumab and ipilimumab in non-epithelioid MPM over chemotherapy (HR 0.46, 95% CI 0.31-0.68) but not in the epithelioid subtype (HR 0.86, 95% CI: 0.69-1.08)[47]. Although two single arm phase 2 trials with durvalumab and chemotherapy have also included patients with

non-epithelioid MPM, the number is too limited to perform sub-analyses according to the hsistology [14,50].

The ongoing phase 3 trials in the first-line setting with chemo-immunotherapy strategy (NCT04334759, NCT02784171, NCT03762018) may help to elucidate the role of this therapeutic approach, with special focus in PD-L1 negative tumors and non-epithelioid subtype, as all histologies are allowed. However, the control arm in these trials is chemotherapy with or without bevacizumab, making difficult to obtain firm conclusions about the best approach in first-line setting when considering immunotherapy combinations.

Best treatment strategy at progression

The treatment paradigm is rapidly evolving in MPM, introducing the challenge of treatment sequences. As the FDA recently approved the combination of nivolumab plus ipilimumab in first-line setting, patients progressing to this treatment become candidates to platinum and pemetrexed combination. However, in most countries, chemotherapy still remains the standard first-line treatment. Although nivolumab is a potential second-line treatment in MPM, ICIs are not always the most feasible treatment strategy in platinum-refractory tumors. As an example, in advanced NSCLC, rapid progression on chemotherapy, high tumor burden and poor performance status were negative prognostic factors and correlated with early risk of death on second-line with nivolumab [54]. In contrast, the combination of chemotherapy plus antiangiogenic therapy resulted in reasonable outcomes even in platinum-refractory tumors [55,56]. The phase II RAMES study has reported that gemcitabine plus ramucirumab (an antiVEGFR2) significantly improved the OS compared with gemcitabine (13.8 months *vs*. 7.5 months, HR 0.71; 70% CI: 0.59-0.85, p = 0.057) as second-line treatment in patients with MPM regardless of age, histological subtype and time to progression after first-line treatment [30]. Even if short time to progression after first line treatment in MPM should be proved to be a negative prognostic factor for second-line treatment with ICI, considering the lack of survival benefit observed in the PROMISE trial [40], gemcitabine plus ramucirumab may become a potential strategy in this subgroup of patients,

mirroring data reported in NSCLC. Finally, ICI-naïve patients in good performance status experiencing progression to first- or second-line chemotherapy, should be candidate to nivolumab monotherapy, based on CONFIRM results [41], especially in the absence of clinical trials.

This sequential treatment approach may be modified in the coming future when results of ongoing phase 3 clinical trials with chemo-immunotherapy with or without bevacizumab will become available (NCT04334759, NCT02784171, NCT03762018), shifting again the therapeutic strategy in MPM and challenging again potential sequential approaches.

Predictive biomarkers

Another unmet need is the identification of predictive biomarkers of ICI effects. As in other malignancies, the predictive role of PD-L1 expression has also been assessed in MPM. Except for SP263, all other immunohistochemistry assays (SP142, 28-8 and 22C3) have reported an accurate PD-L1 immunostaining in this disease [57]. Of note, in up to one-third of MPM, PD-L1 expression appears discordant between paired lesions limiting, along with its dynamicity, the use as a predictive biomarker for therapy [58].

In the first-line setting the predictive role of PD-L1 has been explored. In the Checkmate 743 trial, PD-L1 was not a stratification factor, and the trial was enriched with PD-L1 positive tumors (77%). Although PD-L1 expression did not correlate with outcome (OS with nivolumab and ipilimumab in PD-L1 < 1% vs ≥ 1%, HR 0.87, 95% CI: 0.61-1.24), the magnitude of survival benefit was higher in PD-L1 positive tumors (18.0 *vs*. 13.3 months, HR 0.69, 95% CI: 0.55–0.87) than PD-L1 negative tumors (17.3 *vs*. 16.5 months, HR 0.94, 95% CI 0.62–1.40) [47]. In the two single arm phase 2 trials with durvalumab plus chemotherapy in first line setting, exploratory analysis reported that PD-L1 expression was not significantly associated with survival [14,50].

In previously treated patients who receive ICI in second-line or beyond, PD-L1 positive tumors (*i.e.* PD-L1 detected in ≥ 1% of tumor cells) derived better outcomes compared to PD-L1 negative tumors. The benefit increased with higher of PD-L1

expression [18,32,33,35–37,39,45], but without an optimal cut-off of PD-L1 positivity defined [32]. However, both phase III trials in this setting (PROMISE and CONFIRM), did not support PD-L1 expression as predictive biomarker [40,41]. Whether the ICI efficacy is truly dependent on the PD-L1 expression level is still controversial, but PD-L1 positive MPM may have a trend toward benefit with ICI.

Although patients with MPM have low TMB and intermediate T-cell inflamed tumor microenvironment [23,25,59], the predictive role of TMB assessed by whole exome sequencing has been explored in the Pre505 trial. Although longer OS with durvalumab plus chemotherapy was reported in tumors with high TMB versus low (27.9 *vs*. 14.2 months), the difference did not reach significance (p=0.21) [50].

BRCA-associated protein 1 (BAP1) is a tumor suppressor gene that is responsible for DNA damage repair, cellular differentiation and cell cycle progression. In MPM, *BAP1* inactivation either by mutation or copy number loss is common and reported in up to 60% of the cases [60–62]. Of interest, BAP1 haploinsufficiency strongly correlated with cytokine signaling and an inflammatory tumor microenvironment [63]. It would be relevant in the coming future to elucidate whether *BAP1*-mutant MPM derive greater benefit of ICI.

Surgery in MPM

ICIs have reported long term survival benefit in first-line setting in advanced unresectable MPM. The 1-year of 6 and 2-year OS data with immunotherapy (either nivolumab plus ipilimumab [47] or durvalumab plus chemotherapy [14,50]) mirrors the 1- and 2-year OS reported either with extrapleural pneumonectomy (EPP) or pleurectomy decortication (P/D) in resected MPM [64]. Therefore, questioning the real role of surgery, especially EPP, in this disease. Today it remains unknown whether surgery in resectable MPM improves survival or whether it is just a spurious effect related to the over-selection of patients with good prognostic factors enrolled in surgical studies. The ongoing MARS 2 trial aims to test the hypothesis that (extended) P/D plus chemotherapy improve the survival compared with chemotherapy alone (up to 6 cycles of platinum and pemetrexed) in surgically resectable MPM. The trial has completed the accrual and results are eagerly awaited

[65]. While the role of surgery is being defined, several ongoing clinical trials are testing (neo)adjuvant ICI in the early-stage MPM (NCT04177953, NCT04162015, NCT03918252) with the aim to improve the outcome of resectable MPM.

New potential immune strategies in the therapeutic landscape

Beside ICI, other immunotherapy strategies are actively investigated in MPM. Different phase I trials explored mesothelin (MSLN)-directed chimeric antigen receptor T cells (CAR-Ts) in MPM patients, showing acceptable toxicity profiles and moderate activity (recently reviewed by Castelletti et. [66]). Notably, these studies are highly heterogenous as they employed different generation of CAR-Ts delivered either intravenously or locally. In a phase I/II trial, locally delivered anti-MSLN CAR-Ts showed to be safe and active in patients with pre-treated advanced MPM or pleural solid tumor metastases [67,67]. In this trial CARTs were administered after lymphodepletion and with or without pembrolizumab, with the aim to overcome exhaustion. Similarly, a phase I trial with new generation CAR-T with improved persistence and resistant to PD-L1 mediated exhaustion is ongoing (NCT04577326). Similarly, T-cell receptor fusion construct (TRuC) are in development. The MSLN-directed TRuC TC-210 has shown manageable toxicity profile and some responses in MSLN-positive advanced tumors, including MPM [68]. Recently, local delivery of fibroblast activation protein (FAP)-directed CARTs has also been reported to be safe and feasible in patients with MPM [69]. Beside CARTs, immunotherapy using peripherally collected dendritic cells (DCs) and *in vitro* loaded with autologous or allogenic tumor lysate showed promising results in three phase I trials in MPM [70–72]. The phase III DENIM randomized trial (NCT03610360) is currently ongoing to evaluate this approach, while another phase I study (NCT03546426) is evaluating autologous DCs along with pembrolizumab.

Among vaccines, Galinpepimut-S, a Wilms Tumor-1 (WT-1) peptide-based vaccine, and CRS-207, a live, attenuated, double-deleted *Listeria monoctyogenes*, engineered to express MLSN have been studied. Despite the phase II study of Galinpepimut-S versus placebo in WT-1 positive MPM was closed due to futility [73], a trial combining this molecule with nivolumab is ongoing (NCT04040231). CRS-207

development has been halted, despite a phase I showed tolerability and moderate activity when combined with chemotherapy in unresectable chemo-naïve MPM [74] Other approaches under investigation include oncolytic vaccinia virus such as GL-ONC1 [75] (NCT01766739] and gene-mediated cytotoxic immunotherapy (GMCI) exploiting adenoviral vectors [76]. Finally, in cancer cells, integrating TTFields with anti-PD-1 therapy may further enhance antitumor immunity, hence achieve better tumor control, suggesting a potential future strategy in MPM [77]

Conclusions

In conclusion, similar to ~~toher~~ other thoracic malignancies, ICI shifted the treatment paradigm in unresectable MPM, making prolonged OS possible for a subgroup of these patients. To improve survival for more patients, research should focus on selecting the best ICI combination for every patient, as well incorporating ICI into the treatment strategy of resectable MPM. More emphasis needs to be placed on identifying tumor and immune related biomarkers to assist in patient selection for novel ICI-based strategies.

Figure legends:

Figure 1. Summary of the efficacy of immune checkpoint inhibitors in first-line setting in advanced unresectable malignant pleural mesothelioma. RR: response rate. R: randomization. PFS: Progression Free Survival. OS: Overall Survival. Figure created based on data reported in references [14,47,50].

Figure 2. Current challenges with immune-strategy in first-line setting in advanced unresectable malignant pleural mesothelioma (MPM)

Table 1. Clinical trials and cohorts assessing the role of immune checkpoint inhibitors in previously treated malignant-pleural mesothelioma patients.

Trial	**Drug**	**N**	**RR (%)**	**DCR (%)**	**PFS (mo.)**	**OS (mo.)**	**1-year OS (%)**	**Grade ≥ 3 AEs (%)**	**Grade ≥3 ir-AE (%)**
KEYNOTE 028 [13]	Pembrolizumab (10 mg/Kg Q2W) *	25	20	72	5.4	18	63	20	8
CHICAGO [32]	Pembrolizumab (200 mg Q3W)	65	19	65	4.5	11.5	~50	18	5
MERIT [33]	Nivolumab (240 mg Q2W)	34	29	68	6.1	17.3	59	47	32
NIVO-MESS [34]	Nivolumab (3 mg/Kg Q2W)	34	24	47	2.6	11.8	50	26	8
MAPS2 [35]	Nivolumab arm (3 mg/Kg Q2W)	63	19	40	4.0	11.9	49	14	2
JAVELIN [36]	Avelumab (10 mg/Kg Q2W)	53	9.4	58	4.1	10.7	46	23	9
PROMISE ph 3 [40]	Pembrolizumab (200 mg Q3W)	73	22	45	2.5	10.7	45	NR	19
CONFIRM ph 3 [41]	Nivolumab (240 mg Q2W)	221	10.4	NR	3.0	9.2	40	45	NR
RWD [37]	Pembrolizumab (several schedules) #	93	18	48	3.1	7.2	25	8	NR
RWD [38]	Pembrolizumab (several schedules)	98	18	56	4.8	9.5	41	NR	8
RWD [39]	Nivolumab (3 mg/Kg Q2W)	107	10	37	2.3	6.7	31	NR	NR

RR: Response Rate. DCR: Disease Control Rate. PFS: Progression Free Survival. OS: Overall Survival. AEs: Adverse Events. ir-AES: immune-related adverse events. Q2W: every 2 weeks. RWD: Real World Data. NR: not reported. Ph: Phase. * This trial only includes PD-L1 positive tumors. #200 mg every 21 days (used mostly) to 10 mg/kg body weight every 14 days, 2 mg/kg body weight every 21 days, 2 mg/kg every 14 days, or200 mg every 14 days.

Table 2. Clinical trials assessing the role of the combination of immune checkpoint inhibitors in previously treated malignant-pleural mesothelioma patients.

	Nivolumab (N) + Ipilimumab (I)		Durvalumab (D) + Tremelimumab (T)
	INITIATE [45] N :240 mg Q2W + I: 1 mg/kg Q6W up to four times→ N until PD or 2 years	**MAPS2 (Arm B) [35] *** N: 3 mg/kg Q2W + I: 1mg/kg Q6W→ until PD or up to 2 years	**NIBIT-MESO [46]** T: 1 mg/kg + D: 20 mg/kg Q4W x 4 doses → D x 9 doses
N	34	57	40
End-Point	12 week DCR	12 week DCR	iRR
RR	29%	28%	25%
DCR	68%	52%	63%
PFS (mo)	6.2	5.6	5.7
OS (mo)	NR	15.9	16.6
Grade 3/4 AEs	34%	26%	18%

PD: Progression. iRR: immune Response Rate. DCR: Disease Control Rate. PFS: Progression Free Survival. OS: Overall survival. AEs: Adverse Events. Y: years. * Only reports outcome with nivolumab plus ipilimumab arm.