Trabectedin in Malignant Pleural Mesothelioma: Results From the Multicentre, Single Arm, Phase II ATREUS Study

Trabectedin in malignant pleural mesothelioma: results from the multicentre, single arm, phase II ATREUS Study

Cortinovis D [1*], Grosso F [2*], Zucali PA [3], Pasello G [4], Tiseo M[5], Ardizzoni A[6], Hollander L [7], Carlucci L [7], Galli F [7], Torri V[7], Rulli E[7], *3 nome Monza*, *2 nome Alessandria*, Bidoli P[1], Ceresoli GL [8#], D'Incalci M[7#], on behalf of ATREUS investigators§

1. UO Oncologia Medica, ASST Ospedale San Gerardo, Monza
2. Mesothelioma Unit, Azienda Ospedaliera SS Antonio e Biagio e Cesare Arrigo, Alessandria
3. Istituto Clinico Humanitas - IRCCS Milano
4. Istituto Oncologico Veneto IRCCS Parma
5. Azienda Ospedaliero-Universitaria di Parma
6. Medical Oncology Unit, Sant'Orsola-Malpighi Hospital, Bologna
7. Department of Oncology, Istituto di Ricerche Farmacologiche Mario Negri IRCCS, Milan
8. Department of Oncology, Cliniche Humanitas Gavazzeni, Bergamo

* dr. Cortinovis and dr. Grosso contributed equally as first authors.
# co-last authors
§ the list of co-authors is reported in appendix

Corresponding author:
Maurizio D'Incalci
Department of Oncology
Istituto di Ricerche Farmacologiche Mario Negri IRCCS, Milan
Mail: maurizio.dincalci@marionegri.it
Phone: +39 0239014571

ClinicalTrials.gov Registration Number: NCT02194231

ABSTRACT

Background: New therapeutic approaches in unresectable malignant pleural mesothelioma (MPM) are eagerly awaited. Trabectedin is an antitumor agent with a direct effect on cancer cell proliferation and a modulating action on tumour microenvironment. The ATREUS study explored the activity and safety of trabectedin in patients with unresectable epithelioid and non-epithelioid MPM.

Patients and methods: Two separate cohorts of patients received trabectedin as second-line treatment in epithelioid MPM and as first or second-line therapy in biphasic and sarcomatoid MPM. Treatment was given intravenously at an initially planned dose of 1.3 mg/m2 every 3 weeks, until progression or unacceptable toxicity. The primary endpoint in both cohorts was progression-free survival rate at 12 weeks (PFS_{12wks}).

Results: Overall, 145 patients were enrolled; 78 (54%) had epithelioid and 67 (46%) non-epithelioid MPM. PFS_{12wks} in 62 evaluable patients with epithelioid MPM was 43.5% (80%CI 34.9% to 52.5%); median PFS and OS in this patient subgroup were 2.4 months (IQR 1.2-5.4) and 9.0 months (IQR 3.6-15.1), respectively. PFS_{12wks} in 52 evaluable patients with non-epithelioid MPM was 30.8% (90%CI 20.3% to 42.9%); median PFS and OS in this patient subgroup were 1.7 months (IQR 1.2-4.0) and 5.4 months (IQR 2.3-10.7), respectively. Trabectedin starting dose was emended due to excess of liver toxicity. Eighty-four (64%) patients received 1.3 mg/m^2, 48 (36%) were treated at 1.1. mg/m^2. The most common grade 3-4 toxicities were hepatotoxicity, leukopenia/neutropenia and fatigue. Grade 3-4 hepatotoxicity was reported overall in 78 (59%) patients; it was observed in 59 (70%) patients treated at 1.3 mg/m2, and in 19 (40%) treated at 1.1 mg/m2.

Conclusions: In our trial trabectedin showed modest clinical activity, at the expense of relevant liver toxicity. Further development of this drug in MPM at full doses is not warranted. Lower doses in combination with immunological or micro-environmental modulators could be a promising strategy in this disease.

Keywords: trabectedin, mesothelioma, second line treatment, non-epithelioid, hepatotoxicity, tumour microenvironment

KEY MESSAGE

Trabectedin showed a signal of activity in this phase II trial in epithelioid and non-epithelioid malignant pleural mesothelioma (MPM), but treatment-related hepatotoxicity was relevant. The unique mechanism of action of this drug suggests that combination at lower doses with micro-environmental modulators could be a valuable strategy in MPM.

INTRODUCTION

Malignant pleural mesothelioma (MPM) is an aggressive tumor commonly associated with asbestos exposure [1]. Its incidence is growing throughout most of the world; it has already peaked in the US, but is still increasing in most European and Asian countries [2]. Only a minority of cases is eligible for multimodal therapy including surgery, while most patients are diagnosed with a diffuse disease and are candidates for medical treatment only [3]. The combination of pemetrexed with a platinum agent has been established as the standard of care in systemic therapy for MPM [4-6], but the prognosis for these patients remains dismal. New therapeutic approaches are very much encouraged, especially for second line treatment of epithelioid patients, as well as for patients with sarcomatoid or biphasic histology in any line, whose outcome is particularly poor [7].

Trabectedin, an originally natural marine product now obtained by a semi-synthetic process, is an antitumor agent with a complex mechanism of action [8, 9]. It binds to the minor groove of DNA and interacts with DNA binding proteins such as some transcription factors and DNA repair proteins. It has a direct effect on cancer cell proliferation and survival, and an effect on tumor microenvironment [10]. In particular, trabectedin has been shown to reduce the number of tumor-associated macrophages (TAM) and the production of several inflammatory and angiogenic factors as IL6 and VEGF, and chemokines as CCL2 [11]. This is of potential therapeutic interest, due to the growing evidence that TAM and inflammatory cytokines play a major role not only in the proliferation, angiogenesis and malignant behavior of cancer cells but also causing an immunosuppressed tumor environment. This aspect is particular evident in the immunosuppressed MPM microenvironment as demonstrated in several murine models of asbestos induced pleural mesothelioma and in macrophage function tests co-cultured with human mesothelioma cell lines [12, 13]. The peculiar mechanism of action of trabectedin prompted us to speculate that it could be an effective drug against mesothelioma, a disease related to asbestos-induced chronic inflammation, with a high number of TAM commonly found in pathological specimens [14-17].

Based on this preclinical background, the ATREUS study aimed to identify a signal for the activity of trabectedin in patients with unresectable MPM, both in epithelioid and non-epithelioid tumors.

PATIENTS AND METHODS

Study design and participants

The ATREUS phase II, single arm, multicentre study aimed to explore the activity of trabectedin as second-line treatment in epithelioid MPM and as first or second-line treatment in biphasic and sarcomatoid MPM. Eligible patients were required to be at least 18 years, and to have a histologically confirmed diagnosis of unresectable MPM, an Eastern Cooperative Oncology Group (ECOG) performance status of 0-1, and at least one measurable lesion according to the modified Response Evaluation Criteria for Solid Tumors (mRECIST) for mesothelioma [18]. Adequate hepatic, renal and haematological functions were also needed. Patients with severe comorbidities (particularly diabetes or other conditions contraindicating the use of high-dose steroids) were excluded, as well as patients with brain metastases. Additional exclusion criteria included presence of a concurrent or previous malignancy (except for in-situ cervical cancer and basal cell carcinoma of the skin, adequately treated), unless there was no evidence of disease for at least 5 years. Full inclusion and exclusion criteria are reported in Supplementary Table 1.

The study complied with the Declaration of Helsinki and was done in accordance with Good Clinical Practice guidelines, and approved by the ethics committees of all study sites. All patients provided written informed consent before enrolment. The study protocol is registered with ClinicalTrials.gov, NCT02194231.

Procedures

Patients received intravenous trabectedin at the dose of 1.3 mg/m2 infused over three hours through a central venous catheter, on day 1 of a 21-day cycle. Treatment was administered until progressive disease, unacceptable toxicity, or patient or physician decision. Pre-medication with 20mg of intravenous dexamethasone was given one hour prior to infusion as prophylaxis for liver toxicity [19]. Dose reductions of trabectedin were predefined to allow management of adverse events. Dose of

subsequent cycles was reduced in case of grade 4 febrile neutropenia, or in case of grade 4 neutropenia lasting more than 5 days; or in case of a platelet count lower than 25.000/mm3. The dose of trabectedin was also reduced for any grade 3 or 4 non-hematological toxicity, except for grade 3-4 nausea/vomiting or isolated gammaglutamiltransferase increase. Once a dose reduction occurred for any reason, no dose escalation in subsequent cycles was allowed. Two drug dose level reductions were planned, firstly to 1.1 mg/m2 and subsequently to 0.9 mg/m2. Due to the occurrence of a high rate of grade 3-4 hepatotoxicity with the starting dose of 1.3 mg/m2 (see Results section), the study was amended, and the initial dose was reduced to 1.1 mg/m2.

Patients were followed up weekly for the first two treatment cycles, than every 3 weeks during the remaining study period. Physical examination, evaluation of performance status, and registration of adverse events and concomitant medication were performed at each study visit. Complete blood cell count and chemistry tests were collected. Chest and abdomen CT scans were done every 6 weeks from the date of first treatment until week 12, and subsequently every 9 weeks. A local radiologist at each study centre assessed response evaluation according to mRECIST criteria for MPM [18]. An independent central radiological review was planned. Progressing patients were followed up for survival only. All adverse events (AEs) were recorded prospectively according to the National Cancer Institute's Common Terminology Criteria for Adverse Events (NCI CTCAE), version 4.0. Serious Adverse Events (SAE) were also reported.

Statistical methods

Endpoints

The primary endpoint of the trial was progression-free survival at 12 weeks (PFS_{12wks}) defined as the proportion of patients who were alive and progression-free at the second CT scan assessment, performed 12 weeks after treatment start. Since clinical worsening could anticipate radiological findings, clinical progression, defined as significant increase in pain, cough or dyspnoea, or general deterioration of clinical conditions, was considered as event for PFS_{12wks}. PFS_{12wks} was assessed in

the per-protocol (PP) population that included all patients with no major violations of eligibility criteria who had received at least 12 weeks of treatment. Patients who interrupted treatment for disease progression or death before 12 weeks were included in the analysis as failure. Patients who did not progress or died within 12 weeks from treatment start and without a disease evaluation between the 11^{th} and the 13^{th} week were considered as not evaluable for the analysis, unless the absence of progressive disease was confirmed in the disease evaluations after the 13^{th} week.

Secondary endpoints were objective response rate (ORR), progression free survival (PFS), overall survival (OS) and toxicity. ORR was defined as the proportion of assessable patients achieving a complete (CR) or partial response (PR) based on mRECIST criteria for MPM [16]. PFS was defined as the time from the date of treatment start to the date of disease progression or death from any cause, whichever occurred first. Patients not progressed or died while on study or lost to follow-up were censored at their last disease evaluation date. OS was defined as the time from the date of treatment start to the date of death from any cause. Patients not reported as having died at the end of the study were censored at the last date they were known to be alive.

Sample size

The study design for each cohort is reported in Figure 1. According to Simon's optimal two-stage design, the study was sized to reveal, at a one-sided α level of 10%, whether in patients with epithelioid mesothelioma the PFS_{12wks} was $\leq$25%, and at the same time reveal, at a β level of 15%, whether the PFS_{12wks} was $\geq$40%. Overall, 24 and 62 patients had to be evaluable for the first and second stage, respectively. To allow for a 20% of not eligible patients, approximately 74 patients with epithelioid MPM were planned to be recruited. In the cohort of patients with non-epithelioid (sarcomatoid or biphasic) MPM, adopting the Fleming design with A'Hern's approach, the study was sized to reject, with a one-sided α level of 5%, the null hypothesis that the $PFS_{12}w$ was $\leq$15% and to have 95% power to reveal whether the $PFS_{12}w$ was $\geq$35%. To allow for a 20% of not eligible patients, the total number of patients to be registered in this cohort was approximately 67.

Statistical analyses

Continuous variables were expressed as medians with their interquartile range [IQR]. $PFS_{12}w$ rate was provided with its 95% confidence intervals (CIs) for epithelioid patients and with its 95%Cis for sarcomatoid/biphasic patients. Logistic regression models were used to explore the relationship between PFS_{12wks} and patients' demographical and clinical characteristics. The results were provided as odds ratio (OR) and 95%CI. Univariate and multivariable Cox proportional hazards models were used to analyze the impact of patients' demographical and clinical characteristics on survival outcomes. The results were provided as hazard ratios (HRs) and 95%CIs. Statistical models included as covariates age, sex, ECOG performance status, asbestos exposure, smoking history, disease staging prior to treatment start, trabectedin starting dose and the previous chemotherapy treatment for the group of patients with sarcomatoid/biphasic MPM. Survival curves were estimated by the Kaplan-Meier (KM) method.

The toxicity profile was evaluated in the safety population that included all patients without major violations of the eligibility criteria who had received at least one treatment dose. After the database lock, each toxicity was recorded according to MedDRA dictionary. For any single toxicity, the maximum grade experienced by each subject was reported overall, by initial dose of trabectedin and according to naïve or pre-treated status. All analyses were done with SAS software, versions 9.4 (SAS Institute).

RESULTS

From July 2013 to February 2018, 145 patients were enrolled in seven Italian sites; 78 patients (54%) had epithelioid MPM and 67 (46%) non-epithelioid MPM. A biphasic and sarcomatoid subtype was diagnosed in 38 (58.5%) and 27 (41.5%) cases, respectively. For two patients the information about the subtype was not available. Figure 2 summarize the study flowchart. Three patients in the epithelioid cohort and four in the non-epithelioid cohort were excluded from the analysis due to major protocol violations (due to signature of an informed consent non-compliant with the amended version); additionally, two patients in the epithelioid and 4 in the non-epithelioid group did not start the study treatment. Trabectedin was therefore administered in 73 patients with epithelioid histology as second-line treatment; among 59 patients with non-epithelioid tumours who actually received the planned treatment, 22 (37%) were treated in the first-line setting and 47 (63%) as second-line therapy. The main baseline characteristics of enrolled patients (including those who were enrolled in the study but did not receive treatment) are summarised in Table 1 and Supplementary Table 2. Results are reported according to histological cohorts, except for the safety results, which are reported according to the initial dose of trabectedin.

Epithelioid MPM cohort

Baseline characteristics of the 75 patients with epithelioid MPM and no major violations of eligibility criteria are listed in Table 1 and Supplementary Table 2. Two patients (2.7%) did not start treatment due to withdrawal of consent and symptomatic disease progression. The main reasons for treatment interruption were radiological disease progression (51 patients, 69.9%) after a median of 4 cycles and clinical disease progression (7 patients, 9.6%) after a median of 3 cycles. No CR was observed among the 70 patients with at least one disease evaluation. Overall, 5 patients achieved a PR (ORR 7.1%, 95%CI 2.4% to 15.9%), whereas 40 (57.1%) had stable disease (SD). Out of 66 patients included in the PP population, 62 were evaluable for PFS_{12wks}. The proportion of patients alive and without progression at 12 weeks was 43.5% (27 patients, 80%CI 34.9% to 52.5%, 95%CI 31.0 to 56.7%).

Out of 73 treated patients, 25 patients were enrolled by the centre of the radiologist who performed the review, no CT scan was done in 2 patients and only the baseline CT scan was done for 4 patients. Therefore, 84 CT scans from 42 patients were independently revised. Concordance was observed in 64/84 assessments (76.2%). Considering the radiological central review, PFS_{12wks} was 36.5% (23 patients out 63 evaluable patients, 80%CI 28.4% to 45.3%). Overall, 65 patients (98.5%) progressed and 64 (97.0%) died and all had progression or died during the study. Median PFS was 2.4 months (IQR 1.2 to 5.4 months; Figure 3A). At multivariable analysis, older age was associated with a better PFS (HR [1 year increase] 0.92, 95%CI 0.89 to 0.95, p<0.001), whereas stages III/IV (HR 3.04, 95%CI 1.30 to 7.11, p=0.010) compared to stages I/II had a significantly shorter PFS. The median OS was 9.0 months (IQR 3.6 to 15.1 months; figure 3B). No significant correlation of OS with patient demographical and clinical characteristics was detected.

Biphasic/sarcomatoid MPM cohort

Table 1 shows the baseline characteristics of the 63 enrolled patients with non-epithelioid (biphasic/sarcomatoid) MPM and no major violations of eligibility criteria. Four patients (6.3%) did not start treatment due to withdrawal of consent (2 patients), death (1 patient) and deterioration of clinical conditions (1 patient). Main reasons for treatment interruption were radiological disease progression (36 patients, 61%) after a median of 3.5 cycles, death (9 patients, 15%) after a median of 2 cycles, and clinical disease progression in 8 patients (14%) after a median of 4 cycles. No CR was observed among the 47 patients with at least one disease evaluation. Only one patient (2.1%, 95%CI 0.1% to 11.3%) had a PR, 30 patients (63.8%) achieved a SD. Out of 54 patients included in the PP population, 52 were evaluable for PFS_{12wks}. The proportion of patients alive and without progression at 12 weeks was 30.8% (16 patients, 90%CI 20.3% to 42.9%, 95%CI 18.7 to 45.1%). Out of 59 treated patients, 16 patients were enrolled by the centre of the radiologist who performed the review and no CT scan were done in 9 patients. Therefore, 54 CT scans from 34 patients were revised. Concordance

was observed in 41/54 assessments (75.9%). Considering the radiological central review, PFS_{12wks} was 25.0% (13 patients out 52 evaluable patients, 90%CI 15.5% to 36.8%).

Overall, 48 patients (88.9%) progressed, 52 (96.3%) died and all had progression or died during the study. Median PFS was 1.7 months (IQR 1.2 to 4.0 months; Figure 3A). At multivariable analysis, a significantly shorter PFS was detected for patients with a smoking habit (HR 1.99, 95%CI 1.02 to 3.88, p=0.042). Median OS was 5.4 months (IQR 2.3 to 10.7 months; figure 3B). At multivariable analysis, ECOG performance status correlated with a worse OS (HR [1 vs 0] 2.79, 95%CI 1.36 to 5.73, p=0.005).

Safety Evaluation

Overall, 132 patients were included in the safety population. Eighty-four (64%) patients received trabectedin at the starting dose of 1.3 mg/m^2, and the remaining 48 (36%) patients were treated at the starting dose of 1.1. mg/m^2. Table 2 summarizes the frequency of adverse events occurred in at least 10% of patients. The most common grade 3-4 toxicities were hepatotoxicity, leukopenia/neutropenia and fatigue. Grade 3-4 hepatotoxicity was reported overall in 78 (59.1%) patients; it was observed in 59 (70.2%) patients treated at the starting dose of 1.3 mg/m2, and in 19 (39.6%) of patients treated at 1.1 mg/m2. Grade 3-4 neutropenia was observed overall in 27 (20.5%) patients, decreasing from 23.8% to 14.6% according to the dose level. Overall grade 3-4 fatigue was 8.3%, and was similarly observed at the two dose levels. Fifty-one serious adverse events occurred in 44 (30.3%) patients; of these, 18 (35.3%) were judged as at least possibly treatment-related. One toxic death occurred due to multi-organ failure.

DISCUSSION

Our phase II, single arm, multicentre study, conducted in two separate cohorts of patients with epithelioid and non-epithelioid MPM, showed a signal of activity of trabectedin in both subsets of the study population. Trabectedin was administered in the second-line setting in epithelioid tumors, and in either first or second-line setting in non-epithelioid cases. Unfortunately, a high rate of severe hepatotoxicity was observed at the initially planned dose of 1.3 mg/m2, leading to a protocol amendment to a lower dose of 1.1 mg/m2.

MPM is a disease with a generally poor prognosis, and few effective therapeutic options. Upfront treatment for unresectable patients has not changed in the past 15 years [4, 5]. There is no standard second-line treatment for those who progress during or after first-line pemetrexed/platinum chemotherapy [20]. Single agent vinorelbine or gemcitabine, or re-challenge with a pemetrexed-based regimen are commonly used, but their activity is limited [21, 22]. Several targeted therapies and immunotherapy with immune checkpoint inhibitors have failed to improve patient outcome in this setting [23-26]. Clearly, new treatment strategies are needed. Although current guidelines do not differentiate therapy recommendations of unresectable MPM according to histological subtypes, biphasic and sarcomatoid tumours appear to benefit less from current interventions [7, 27], and optimal treatment of these patients is still an unmet need.

Trabectedin is an antineoplastic agent that has been approved for the treatment of advanced soft tissue sarcoma and ovarian cancer [28, 29]. DNA damage and cell cycle arrest in tumour cells accounts for only part of its complex mechanism of action [8, 11]. In preclinical models, trabectedin was shown to selectively induce a reduction of TAM, with a marked inhibition of the production of inflammatory cytokines, chemokines and angiogenic factors [30]. Furthermore, reduced TAM infiltration and decreased angiogenesis were observed in tumour samples from patients with sarcoma treated with trabectedin, compared with pre-treatment biopsy samples [11]. These observations raise the question

as to whether the combination of trabectedin with antiangiogenic agents and/or immune-checkpoint inhibitors might increase its effectiveness [31].

In early Phase I trials, anecdotal activity of trabectedin in a few mesothelioma patients was reported [32, 33]. More recently, in a preclinical study, trabectedin showed a dose-dependent cytotoxic effect on several MPM cells in vitro and on an intraperitoneal MPM xenograft model in vivo; the drug activity was synergistically enhanced by co-administration of cisplatin and experimental bcl-2 inhibitors in vitro [34]. In another study, activity of trabectedin against patient-derived mesothelioma xenografts was reported [35, 36]. To the best of our knowledge, ATREUS is the first clinical study of this compound in MPM patients. In our trial, trabectedin was moderately active in both epithelioid and non-epithelioid MPM, with a PFS_{12wks} of 43.5% (95%CI 31.0-56.7%) and 30.8% (95%CI 18.7-45.1%) in the two cohorts, respectively. Disease control rate (PR+SD) was 64.2% and 65.9% in the two groups. These results, based on investigator assessment, were confirmed by a blinded radiological central review, that is strongly recommended in clinical trials on MPM [3].

The signal for activity achieved in the ATREUS study is further validated by the results of the recent SAKK 17/16 phase II trial of lurbinectedin, a novel compound closely related to trabectedin [37], in the second- and third-line MPM setting [38]. In this trial, 42 patients with pretreated MPM were recruited; all had progressed during or after pemetrexed/platinum therapy; 10 were also pre-treated with immunotherapy. Histology was epithelioid in 33, and biphasic/sarcomatoid in 9. PFS_{12wks}, the trial primary endpoint, was met by 22/42 patients (52.4%; 90% CI: 38.7-63.5%) with a disease control rate of 52%. No significant difference in PFS_{12wks} was observed in epithelioid versus non-epithelioid cases and in patients with prior immunotherapy versus those without. The lurbinectedin data seem to compare favorably with the results of the ATREUS trial with trabectedin; however, the SAKK trial enrolled a smaller number of patients, with only few biphasic/sarcomatoid cases, and no central radiological review, precluding any definite conclusion, particularly in the non-epithelioid setting. It should be noted that trabectedin and lurbinectedin have a similar mode of action, but different

pharmacokinetic properties [37]. In particular, lurbinectedin has a much lower volume of distribution than trabectedin, possibly due to its higher water solubility and to its binding to alfa-1 acidic glycoprotein [39]. The different pharmacokinetic features of lurbinectedin could explain the different pattern of toxicity of the two drugs. In our trial we observed a very high rate of hepatotoxicity of trabectedin given at 1.3 mg/m2, which was not expected based on the available data on patients with other diseases like soft tissue sarcomas [28, 40, 41]. Pre-treatment with steroids, which was found to markedly reduce the hepatotoxicity of the drug in sarcoma patients [19], was also applied to the ATREUS patients, with less success. A possible partial explanation for this discrepancy may be related to the 3-hour schedule of trabectedin infusion in our trial. It has been in fact reported that the hepatotoxicity of trabectedin was generally lower by administering the drug as a prolonged infusion rather than as shorter schedules [42-44]. However, by decreasing the dose to 1.1 mg/m2, grade 3-4 hepatotoxicity in ATREUS patients dropped from 70% to 40%, with a rapid recovery that allowed treatment continuation in most patients.

In conclusion, the ATREUS trial showed some signals of activity of trabectedin in MPM patients, confirmed by a central radiological review, at the expense of relevant liver toxicity. This toxicity profile, together with the small survival benefit observed in the study, precludes the additional development of trabectedin in MPM as single agent with the investigated schedule. However, its unique mechanism of action suggests that its combination at lower doses with immunological or microenvironmental modulators could be a successful strategy to be explored in mesothelioma patients, including those with non-epithelioid histology.

REFERENCES

1. Carbone M, Adusumilli PS, Alexander HR, Jr. et al. Mesothelioma: Scientific clues for prevention, diagnosis, and therapy. CA Cancer J Clin 2019; 69: 402-429.
2. Delgermaa V, Takahashi K, Park EK et al. Global mesothelioma deaths reported to the World Health Organization between 1994 and 2008. Bull World Health Organ 2011; 89: 716-724, 724A-724C.
3. Tsao AS, Lindwasser OW, Adjei AA et al. Current and Future Management of Malignant Mesothelioma: A Consensus Report from the National Cancer Institute Thoracic Malignancy Steering Committee, International Association for the Study of Lung Cancer, and Mesothelioma Applied Research Foundation. J Thorac Oncol 2018; 13: 1655-1667.
4. Vogelzang NJ, Rusthoven JJ, Symanowski J et al. Phase III study of pemetrexed in combination with cisplatin versus cisplatin alone in patients with malignant pleural mesothelioma. J Clin Oncol 2003; 21: 2636-2644.
5. Ceresoli GL, Zucali PA, Favaretto AG et al. Phase II study of pemetrexed plus carboplatin in malignant pleural mesothelioma. J Clin Oncol 2006; 24: 1443-1448.
6. Santoro A, O'Brien ME, Stahel RA et al. Pemetrexed plus cisplatin or pemetrexed plus carboplatin for chemonaive patients with malignant pleural mesothelioma: results of the International Expanded Access Program. J Thorac Oncol 2008; 3: 756-763.
7. Mansfield AS, Symanowski JT, Peikert T. Systematic review of response rates of sarcomatoid malignant pleural mesotheliomas in clinical trials. Lung Cancer 2014; 86: 133-136.
8. D'Incalci M, Galmarini CM. A review of trabectedin (ET-743): a unique mechanism of action. Mol Cancer Ther 2010; 9: 2157-2163.
9. Larsen AK, Galmarini CM, D'Incalci M. Unique features of trabectedin mechanism of action. Cancer Chemother Pharmacol 2016; 77: 663-671.
10. D'Incalci M, Badri N, Galmarini CM, Allavena P. Trabectedin, a drug acting on both cancer cells and the tumour microenvironment. Br J Cancer 2014; 111: 646-650.
11. Germano G, Frapolli R, Belgiovine C et al. Role of macrophage targeting in the antitumor activity of trabectedin. Cancer Cell 2013; 23: 249-262.
12. Rehrauer H, Wu L, Blum W et al. How asbestos drives the tissue towards tumors: YAP activation, macrophage and mesothelial precursor recruitment, RNA editing, and somatic mutations. Oncogene 2018; 37: 2645-2659.
13. Izzi V, Chiurchiu V, D'Aquilio F et al. Differential effects of malignant mesothelioma cells on THP-1 monocytes and macrophages. Int J Oncol 2009; 34: 543-550.
14. Thapa B, Salcedo A, Lin X et al. The Immune Microenvironment, Genome-wide Copy Number Aberrations, and Survival in Mesothelioma. J Thorac Oncol 2017; 12: 850-859.
15. Yap TA, Aerts JG, Popat S, Fennell DA. Novel insights into mesothelioma biology and implications for therapy. Nat Rev Cancer 2017; 17: 475-488.

16. Minnema-Luiting J, Vroman H, Aerts J, Cornelissen R. Heterogeneity in Immune Cell Content in Malignant Pleural Mesothelioma. Int J Mol Sci 2018; 19.
17. Salaroglio IC, Kopecka J, Napoli F et al. Potential Diagnostic and Prognostic Role of Microenvironment in Malignant Pleural Mesothelioma. J Thorac Oncol 2019; 14: 1458-1471.
18. Byrne MJ, Nowak AK. Modified RECIST criteria for assessment of response in malignant pleural mesothelioma. Ann Oncol 2004; 15: 257-260.
19. Grosso F, Dileo P, Sanfilippo R et al. Steroid premedication markedly reduces liver and bone marrow toxicity of trabectedin in advanced sarcoma. Eur J Cancer 2006; 42: 1484-1490.
20. Kindler HL, Ismaila N, Armato SG, 3rd et al. Treatment of Malignant Pleural Mesothelioma: American Society of Clinical Oncology Clinical Practice Guideline. J Clin Oncol 2018; 36: 1343-1373.
21. Zauderer MG, Kass SL, Woo K et al. Vinorelbine and gemcitabine as second- or third-line therapy for malignant pleural mesothelioma. Lung Cancer 2014; 84: 271-274.
22. Ceresoli GL, Zucali PA, De Vincenzo F et al. Retreatment with pemetrexed-based chemotherapy in patients with malignant pleural mesothelioma. Lung Cancer 2011; 72: 73-77.
23. Scherpereel A, Wallyn F, Albelda SM, Munck C. Novel therapies for malignant pleural mesothelioma. Lancet Oncol 2018; 19: e161-e172.
24. Maio M, Scherpereel A, Calabro L et al. Tremelimumab as second-line or third-line treatment in relapsed malignant mesothelioma (DETERMINE): a multicentre, international, randomised, double-blind, placebo-controlled phase 2b trial. Lancet Oncol 2017; 18: 1261-1273.
25. Scherpereel A, Mazieres J, Greillier L et al. Nivolumab or nivolumab plus ipilimumab in patients with relapsed malignant pleural mesothelioma (IFCT-1501 MAPS2): a multicentre, open-label, randomised, non-comparative, phase 2 trial. Lancet Oncol 2019; 20: 239-253.
26. Popat S, Curioni-Fontecedro A, Polydoropoulou V et al. LBA91_PRA multicentre randomized phase III trial comparing pembrolizumab (P) vs single agent chemotherapy (CT) for advanced pre-treated malignant pleural mesothelioma (MPM): Results from the European Thoracic Oncology Platform (ETOP 9-15) PROMISE-meso trial. Annals of Oncology 2019; 30.
27. Meyerhoff RR, Yang CF, Speicher PJ et al. Impact of mesothelioma histologic subtype on outcomes in the Surveillance, Epidemiology, and End Results database. J Surg Res 2015; 196: 23-32.
28. Demetri GD, von Mehren M, Jones RL et al. Efficacy and Safety of Trabectedin or Dacarbazine for Metastatic Liposarcoma or Leiomyosarcoma After Failure of Conventional Chemotherapy: Results of a Phase III Randomized Multicenter Clinical Trial. J Clin Oncol 2016; 34: 786-793.
29. Monk BJ, Herzog TJ, Kaye SB et al. Trabectedin plus pegylated liposomal Doxorubicin in recurrent ovarian cancer. J Clin Oncol 2010; 28: 3107-3114.
30. Dossi R, Frapolli R, Di Giandomenico S et al. Antiangiogenic activity of trabectedin in myxoid liposarcoma: involvement of host TIMP-1 and TIMP-2 and tumor thrombospondin-1. Int J Cancer 2015; 136: 721-729.

31. Mantovani A, Marchesi F, Malesci A et al. Tumour-associated macrophages as treatment targets in oncology. Nat Rev Clin Oncol 2017; 14: 399-416.
32. Ryan DP, Supko JG, Eder JP et al. Phase I and pharmacokinetic study of ecteinascidin 743 administered as a 72-hour continuous intravenous infusion in patients with solid malignancies. Clin Cancer Res 2001; 7: 231-242.
33. Twelves C, Hoekman K, Bowman A et al. Phase I and pharmacokinetic study of Yondelis (Ecteinascidin-743; ET-743) administered as an infusion over 1 h or 3 h every 21 days in patients with solid tumours. Eur J Cancer 2003; 39: 1842-1851.
34. Hoda MA, Pirker C, Dong Y et al. Trabectedin Is Active against Malignant Pleural Mesothelioma Cell and Xenograft Models and Synergizes with Chemotherapy and Bcl-2 Inhibition In Vitro. Mol Cancer Ther 2016; 15: 2357-2369.
35. Licandro SA, Frapolli R, Bello E et al. Trabectedin activity in patient-derived mesothelioma xenografts. Cancer Research 2016; 76.
36. Ceriani L, Ferrari M, Zangarini M et al. HPLC-MS/MS method to measure trabectedin in tumors: preliminary PK study in a mesothelioma xenograft model. Bioanalysis 2015; 7: 1831-1842.
37. Romano M, Frapolli R, Zangarini M et al. Comparison of in vitro and in vivo biological effects of trabectedin, lurbinectedin (PM01183) and Zalypsis(R) (PM00104). Int J Cancer 2013; 133: 2024-2033.
38. Metaxas Y, Frueh M, Eboulet EI et al. 1843OSAKK 17/16 - Lurbinectedin as second or third line palliative chemotherapy in malignant pleural mesothelioma (MPM): A multi-center, single-arm phase II trial. Annals of Oncology 2019; 30.
39. Erba E, Romano M, Gobbi M et al. Ascites interferes with the activity of lurbinectedin and trabectedin: Potential role of their binding to alpha 1-acid glycoprotein. Biochem Pharmacol 2017; 144: 52-62.
40. Garcia-Carbonero R, Supko JG, Maki RG et al. Ecteinascidin-743 (ET-743) for chemotherapy-naive patients with advanced soft tissue sarcomas: multicenter phase II and pharmacokinetic study. J Clin Oncol 2005; 23: 5484-5492.
41. Kobayashi H, Iwata S, Wakamatsu T et al. Efficacy and safety of trabectedin for patients with unresectable and relapsed soft-tissue sarcoma in Japan: A Japanese Musculoskeletal Oncology Group study. Cancer 2019.
42. Ueda T, Kakunaga S, Ando M et al. Phase I and pharmacokinetic study of trabectedin, a DNA minor groove binder, administered as a 24-h continuous infusion in Japanese patients with soft tissue sarcoma. Invest New Drugs 2014; 32: 691-699.
43. Forouzesh B, Hidalgo M, Chu Q et al. Phase I and pharmacokinetic study of trabectedin as a 1- or 3-hour infusion weekly in patients with advanced solid malignancies. Clin Cancer Res 2009; 15: 3591-3599.
44. Lau L, Supko JG, Blaney S et al. A phase I and pharmacokinetic study of ecteinascidin-743 (Yondelis) in children with refractory solid tumors. A Children's Oncology Group study. Clin Cancer Res 2005; 11: 672-677.

ACKNOWLEDGMENTS

xxx

APPENDIX

List of participating institutions and coauthors

ASST Ospedale San Gerardo, Monza
Cortinovis D, Bidoli P, xxx
AO SS Antonio e Biagio e Cesare Arrigo, Alessandria
Grosso F, xx
Istituto Clinico Humanitas - IRCCS ,Milano
Zucali PA, xxx
Istituto Oncologico Veneto IRCCS, Parma
Pasello G, xxx
Azienda Ospedaliero-Universitaria di Parma
Tiseo M, xxx
Sant'Orsola-Malpighi Hospital, Bologna
Ardizzoni A, xxx
Cliniche Humanitas Gavazzeni, Bergamo
Ceresoli GL, xxx
Istituto Ricerche Farmacologiche Mario Negri , Milano
Hollander L, Carlucci L, Galli F, Torri V, Rulli E, D'Incalci M, Fabio Galli, Davide Poli.

FIGURE LEGEND

Figure 1: Study design. MPM: Malignant pleural mesothelioma; PFS_{12wks}: Progression Free Survival at 12 weeks.

Figure 2: Study flowchart. MPM: Malignant pleural mesothelioma; PP: Per Protocol

Figure 3: Kaplan Meier curves for Progression Free Survival (A) and Overall Survival (B) according to histological cohorts.