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ORIGINAL ARTICLE – BONE AND SOFT TISSUE SARCOMAS

Sirolimus in Advanced Epithelioid Hemangioendothelioma: A Retrospective Case-Series Analysis from the Italian Rare Cancer Network Database

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ABSTRACT

Background. The aim of this study was to report on sirolimus activity in a series of patients with hemangioendothelioma (HE) treated at the National Cancer Institute, Milan (Istituto Nazionale Tumori; INT) and within the Italian Rare Cancer Network ("Rete Tumori Rari"; RTR).

Methods. We retrospectively reviewed patients with advanced and progressing epithelioid hemangioendothelioma (EHE) treated with sirolimus at the INT and/or within the RTR. Pathologic review and molecular analysis for *WWTR1* rearrangement were performed. Sirolimus was administered until unacceptable toxicity or progression, with the dose being adjusted to reach target plasma levels of 15–20 ng/dL. Responses were assessed using the Response Evaluation Criteria In Solid Tumors (RECIST) criteria.

Results. Since 2005, 18 patients (17 EHE, 1 retiform HE; 1 locally advanced, 17 metastatic; *WWTR1* rearrangement: 16) have been identified, with 17/18 patients being evaluable for response. Mean sirolimus daily dose was 4.5 mg. According to RECIST, best responses in EHE were 1 partial response (PR), 12 stable disease (SD), and 3 progressive disease (PD); the patient with retiform HE also achieved a PR, lasting >2 years. Four patients with a reversed interval progression on interruption were observed. Median overall survival was 16 months, and median progression-free survival was 12 months (range 1–45), with four patients progression-free at 24 months. The clinical benefit (complete response [CR] + PR + SD $\geq$6 months) was 56 %. Seven patients receiving sirolimus experienced an increase in pleural/peritoneal effusion plus worsening of tumor-related symptoms; six of these patients died within 1–8 months from evidence of effusion progression, while a RECIST PD was assessed in two of seven patients.

Conclusions. A clinical benefit was achieved in 56 % of patients receiving sirolimus, which lasted >24 months in four patients. Most patients with pleural effusion did not benefit from sirolimus and had a poor outcome.

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INTRODUCTION

Epithelioid hemangioendothelioma (EHE) is an exceedingly rare sarcoma.[1,2] In the World Health Organization (WHO) classification, vascular sarcomas include EHE, along with four other subtypes of hemangioendothelioma (HE), i.e. kaposiform, retiform (RHE), composite, and pseudomyogenic.[2] EHE can be distinguished from other HE subtypes and epithelioid angiosarcoma by the presence of two specific translocations, *WWTR1-CAMTA1* and *YAP1/TFE3*, detected in approximately 90 and 10 % of cases, respectively.[3–5] These two fusions are responsible for the nuclear expression of CAMTA1[6] and TFE3.[5]

The natural history of EHE is usually indolent and unpredictable. Even if, in the literature, the proportion of metastatic patients is probably >20–30 %,[2,7] the metastatic phase of disease does not necessarily require treatment. Interestingly, the presence of pleural involvement was recently reported to correlate with poor prognosis in thoracic EHE.[8] In addition, progressive patients may require medical therapy.

We report on the activity of the mammalian target of rapamycin (mTOR) inhibitor sirolimus in a retrospective series of 17 patients with progressive EHE and one patient with RHE treated at the Istituto Nazionale Tumori, Milan, Italy (INT) and within the Italian Rare Cancer Network ("Rete Tumori Rari"; RTR).

MATERIALS AND METHODS

Patient Selection

We retrospectively identified 18 patients with advanced HE (17 EHE, 1 RHE) consecutively treated at the INT (15 patients) or included in the RTR database by three other institutions (three cases) between January 2005 and November 2015. All cases had signs of clinical and/or radiographic progression during the previous 6 months, as detailed in the Results section. Eastern Cooperative Oncology Group (ECOG) performance status $\leq$3 and adequate bone marrow and organ function were requested in all cases. Pathologic and radiologic review of all patients was performed.

All patients provided written informed consent to data collection and non-conventional treatment, and Institutional Review Board approval was requested.

Morphology and Immunophenotype

Diagnosis was reviewed and confirmed according to the WHO classification,[2] and diagnostic and functional immunoprofile assessment was performed using the antibodies and conditions reported in electronic supplementary Table 1. Expression and phosphorylation of 4E-BP1 was assessed in patients whose adequate material was available.

Fluorescence in situ hybridization (FISH) analysis was performed on 2-μm-thick formalin-fixed, paraffin-embedded (FFPE) tissue samples with BAC probes (Children's Hospital Oakland Research Institute BACPAC Resource Center, Oakland, CA, USA) mapping at the 5′ (RP11-941L15) and 3′ (RP11-1151O19) end of WWTR1 and labeled in spectrum green and spectrum orange fluorochromes, respectively. Probe labeling and FISH treatment were carried out according to standard procedures. WWTR1 translocated cells displayed split green and orange signals (break-apart FISH pattern).

Treatment

Patients started sirolimus at a dosage of 5 mg/day (once daily) continuously, until progression or unacceptable toxicity. They were asked to always take the drug at the same time and in the same conditions (i.e. fasting or after a low-fat meal). The plasma level of sirolimus was checked after 10–15 days from the start of treatment, and then monthly, and the daily dose of sirolimus was consistently adjusted to reach target plasma levels of 15–20 ng/dL. Sirolimus was withheld for hematologic grade $\geq$3 and non-hematologic grade $\geq$2 adverse events (AEs), as defined according to the National Cancer Institute Common Toxicity Criteria 3.0, and restarted after recovery to grade <2 or <1 in case of hematologic or non-hematologic adverse events, respectively.

Clinical Assessment

Blood count and biochemistry were evaluated at baseline, at 2 weeks, and then monthly throughout the study period. Symptomatic changes and AEs were recorded. Disease status was assessed at baseline by whole-body computed tomography (CT) scan, including brain evaluation, CT of the sites of disease, and bone scan. An [18F]fluorodeoxyglucose–positron emission tomography (FDG–PET) scan was performed in a subgroup of patients. CTs were repeated after 4–6 weeks of treatment, at 3 months, and then every 3 months.

Efficacy Assessment

Response was assessed using Response Evaluation Criteria In Solid Tumors (RECIST) 1.1.[9] The clinical benefit rate (CBR) was defined as RECIST complete response (CR)+ partial response (PR)+ stable disease (SD) at 6 months.

We also recorded the presence/evolution of pleural/peritoneal effusion at baseline and all along the study period. Patients who stopped sirolimus due to worsening of tumor-related symptoms (pain, dyspnea, asthenia, anorexia, fever, weight loss) and worsening of pleural/peritoneal effusion without evidence of RECIST progressive disease (PD) were also recorded.

Overall survival (OS) and RECIST-based progression-free survival (PFS) were estimated using the Kaplan–Meier method. All patients who received at least one dose of sirolimus were included in the analysis. Patients without evidence of RECIST progression who interrupted sirolimus due to worsening of tumor-related symptoms and pleural/peritoneal effusion were considered treatment failure only at the time of evidence of RECIST progression or death. Patients who interrupted sirolimus for any reason without evidence of RECIST progression were scored at the last tumor assessment. Death was considered an event regardless of the cause. Patients alive or lost to follow-up were censored at the last contact.

In addition to conventional RECIST-based PFS, we assessed an 'extended PFS' as an exploratory endpoint, adding to RECIST progressions those cases where the pleural/peritoneal effusion, tumor-related symptoms, and general conditions worsened without evidence of RECIST PD.

RESULTS

Eighteen patients with progressive, advanced HE (17 EHE, 1 RHE) received sirolimus, with 17 patients being evaluable for response (one patient could not be assessed due to early interruption). Two patients are still receiving therapy and 16 patients stopped sirolimus (reasons for discontinuation: five had RECIST PD, of which three were primary and two were secondary progression; five had worsening of general condition, tumor-related symptoms, and pleural/peritoneal effusion without evidence of RECIST PD; and six for other reasons).

Patients

Table 1 summarizes the patient characteristics. All metastatic cases were metastatic at disease onset and none had evidence of a clear soft tissue or bone primary lesion. Pleural/peritoneal effusion at baseline was detectable in six cases.

Overall, 17/18 cases had a confirmed pathologic diagnosis of EHE, while one case had a final diagnosis of RHE. FISH analysis for *WWTR1* rearrangement was performed in 17/18 cases (in one case, tissue for the analysis was not available) and culminated in a positive result in 16 patients, and a negative result in 1 patient. The latter had a morphology consistent with an RHE, with extensive areas with solid growth and endothelial hobnail cells, in addition to the typical testis-like pattern.[2]

4E-BP1 was assessed in five cases and was found to be expressed and phosphorylated (moderate to strong phosphorylation) in all (Fig. 1).

All patients were evaluated with CT; PET was performed in 11 cases. Fourteen patients showed RECIST progression within 6 months before starting treatment, and in the remaining four cases there was evidence of clinical progression defined as worsening of tumor-related symptoms (pain, dyspnea, asthenia, fever) and pleural (three cases) or peritoneal (one case) effusion, without matching the criteria for RECIST PD.

Median treatment duration was 4 months (range 2 weeks–45 months). Patients started sirolimus 5 mg/day (once daily) in all but two cases, who started with 3 mg/day. Based on the plasma level, the dosage of 5 mg/day was confirmed in 11 patients along the study period. A dose adjustment was required in seven cases (Table 1). The mean daily dose of sirolimus was 4.5 mg (range 2–8).

Overall, sirolimus was fairly well tolerated. The main hematologic toxicities were neutropenia (three cases, grade 3: one) and thrombocytopenia (one case, grade 3), while the main non-hematologic toxicities included mucositis (four cases, grade 1–3), infection (six cases, grade 1–3), dysmenorrhea (four cases, grade 1–2), hypercholesterolemia (seven cases, grade 1–3), and hypertriglyceridemia (four cases, grade 1–2). Toxicity always resolved when sirolimus was discontinued.

Response

Table 2 summarizes the clinical findings. Overall, 1/16 EHE patients evaluable by RECIST had a PR (6 %). In addition, 12 SDs (75 %) and 3 PDs (19 %) were observed. One patient achieving a PR also showed symptomatic improvement 2 weeks after starting sirolimus, and remained on therapy for >36 months. Among patients with a RECIST SD, a minor tumor shrinkage (<30 % decrease in maximum diameter) was detected in four of them. Patients with an RHE also achieved a PR lasting >2 years. The CBR as defined by RECIST was 56 %.

A decrease in PET uptake was detected in 10/11 evaluable patients but, in four cases, PET evaluation was inconsistent with other clinical signs. In two patients with PET response, CT showed a RECIST PD, while in the other two cases with PET response, CT showed a worsening of the effusion and systemic symptoms.

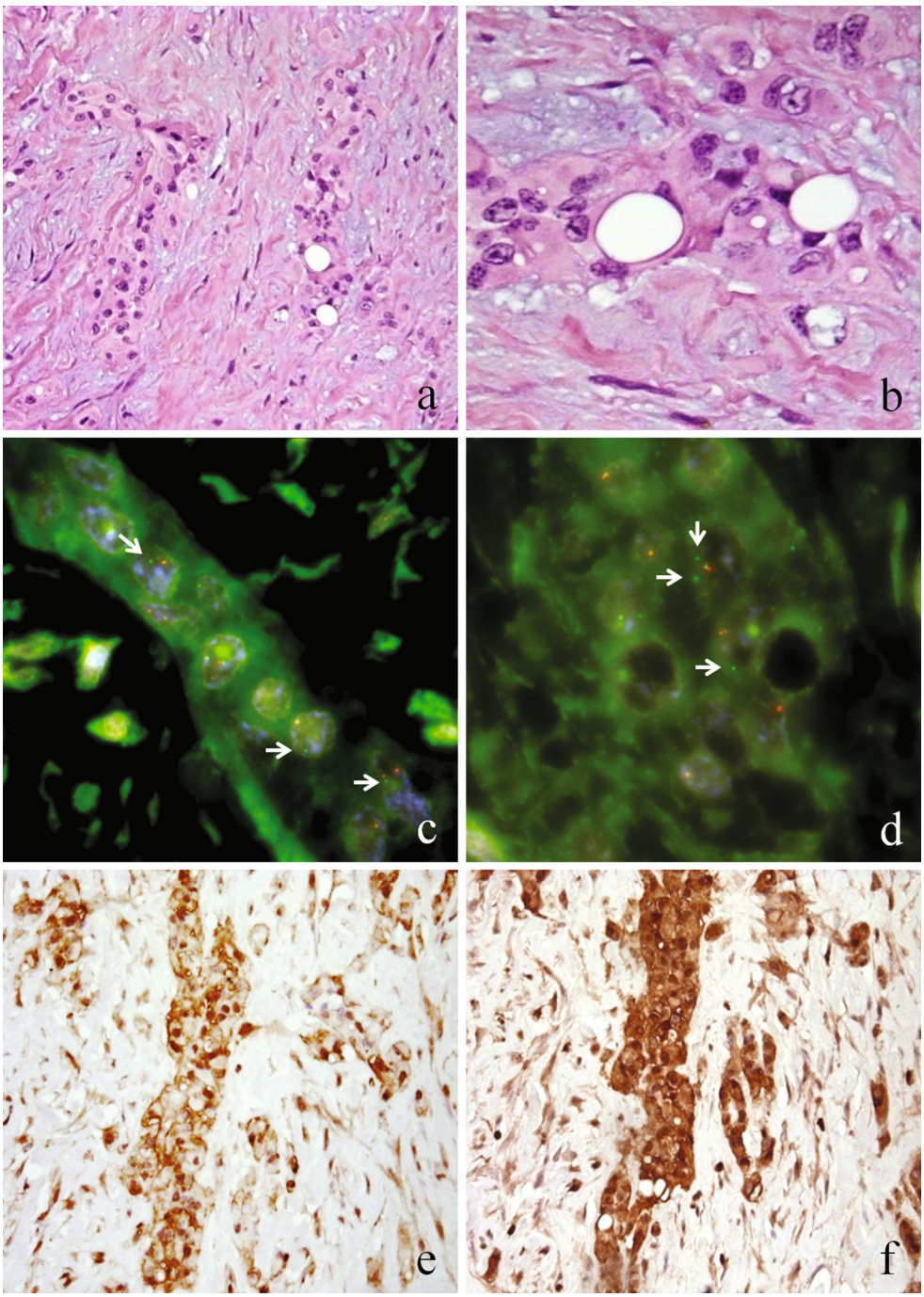

FIG. 1 Pathology and 4E-BP1 expression/phosphorylation. The micrographs highlight the morphologic, cytogenetic and immunophenotypic-based functional characteristics of case number 4 (Tables 1, 2). **a**, **b** Hematoxylin and eosin slides with the cords of tumor cells containing intracytoplasmic lumina embedded into hyaline stroma. **c**, **d** Results of the FISH analysis for *WWTR1* gene rearrangement. *White arrows* indicate the translocated portion of the gene (*green* or *red signals*). The cord-like tumor strands express 4E-BP1 protein (**e**), which looked to be moderately–strongly phosphorylated in the majority of tumor cells when challenged with P-4E-BP1 antibody (**f**). *FISH* fluorescence in situ hybridization

At a median follow-up of 41 months, median OS was 16 months (range 2–50). Median RECIST-based PFS was 12 months (range 1–45), with four patients progression-free at 24 months (electronic supplementary Fig. 1). Median 'extended PFS' was 10 months (range 1–45).

Four patients discontinued sirolimus while responsive/stable (patient choice: three patients; surgery: one patient). All stabilized again after sirolimus rechallenge (Fig. 2). One patient received a complete surgical resection of the residual disease while stable after 6 months of sirolimus; however,

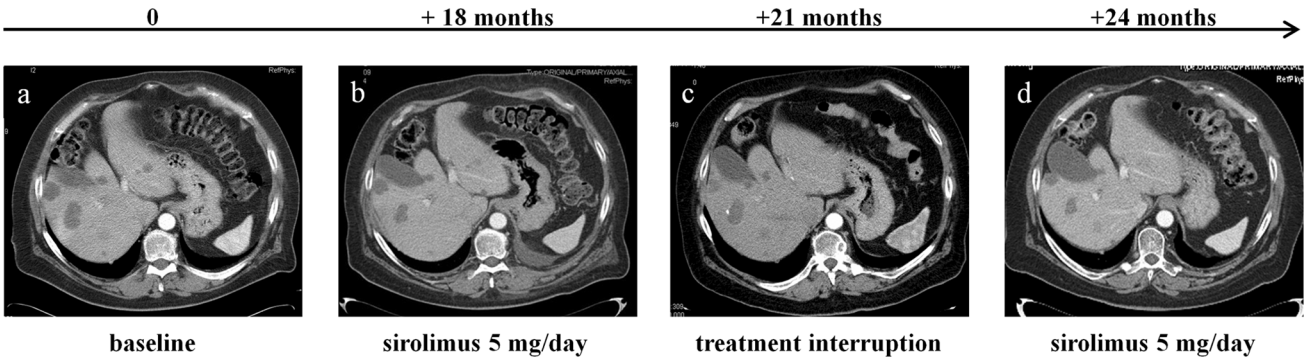

FIG. 2 Response to sirolimus and interval progression. CT scan (arterial phase after contrast medium) of patient number 1 (Tables 1, 2). Multiple liver metastases from EHE at baseline (**a**) and after 18 months of treatment with sirolimus 5 mg/day (**b**), with response. Treatment with sirolimus was discontinued at 18 months, with the evidence of a disease progression 3 months later (**c**). At this point, sirolimus was rechallenged, with a new response detected 3 months later (**d**). *CT* computed tomography, *EHE* epithelioid hemangioendothelioma

the patient relapsed 19 months after surgery and restarted sirolimus, with new disease stabilization. All these patients were scored at the first evidence of progression.

Correlation between Pleural/Peritoneal Effusion Evolution and Outcome

Overall, 9/18 patients suffered from pleural and/or peritoneal effusion (six at baseline, three developed afterwards). In seven of these patients (five pleural, two peritoneal effusion) the effusion worsened while receiving sirolimus, which was coupled with a worsening of tumor-related systemic symptoms, pain, dyspnea, and general condition, without evidence of RECIST PD in 5/7 patients. Six of seven cases had a rapid fatal outcome, dying in 1–8 months (Fig. 3).

DISCUSSION

Since January 2005, we have been treating 18 consecutive patients with progressive, advanced HE (17 EHE, 1 RHE) with continuous-dosing sirolimus. Among the 16 EHE patients evaluable for response according to RECIST, we observed 1 PR (6 %) lasting >3 years, 12 SDs (75 %), and 3 PDs (19 %). A PR was also achieved in the patient affected by RHE. A minor tumor shrinkage was detected in four cases. An interval progression was observed in 4/4 patients who stopped sirolimus while they were stable, with further disease stabilization after rechallenge. Median PFS was 12 months, with four patients being progression-free at 24 months; median OS was 16 months. Seven patients experienced an increase in their pleural and/or peritoneal effusion while receiving sirolimus, along with worsening of tumor-related symptoms and general conditions; 6/7 died in 1–8 months. Among these patients, only 2/7 had a PD according to RECIST.

This was a small, retrospective case-series analysis. Nonetheless, to the best of our knowledge, this is the largest reported series of EHE treated with medical therapy. Patients in this series had a cytogenetically confirmed diagnosis and a progressive tumor. EHE is a rare disease in which differential diagnosis with angiosarcoma can be challenging. In addition, EHE is often marked by indolent behavior and requires medical therapy only in selected cases, although it is not considered sensitive to the conventional agents used in sarcomas. Thus, patients in need of medical therapy have no standard options available.

Data on the activity of medical therapy in EHE are scanty. Two responses and four SDs were reported to bevacizumab in seven EHE patients treated within a phase II study that also included angiosarcoma.[10] Sorafenib achieved two PRs in 15 EHE patients treated within another phase II study enrolling different vascular sarcoma subtypes.[11] Responses lasted 2 and 9 months, respectively, for a 9-month PFS of 30 % in the EHE subgroup. In these studies, patients were treated irrespective of any evidence of progression before starting the experimental therapy. Case reports are available on responses to sunitinib[12,13] and pazopanib.[14,15] Among drugs with an expected antiangiogenic and/or immunomodulatory effect, interferon,[16,17] celecoxib,[17] and thalidomide [18] showed some activity in EHE. Finally, two case reports describe the activity of sirolimus. The first describes metastatic EHE in a child with Maffucci–Ollier syndrome, who achieved a PR lasting >16 months,[19] while the second report refers to an EHE patient treated with sirolimus within a phase I study, with a response lasting >3 years.[20] Of note, besides pazopanib, none of these agents are approved for EHE.

Our analysis suggests that sirolimus can be effective in progressing EHE, achieving a tumor growth arrest in most; however, dimensional responses were uncommon. A major criticism could be that a stable disease cannot be taken as a

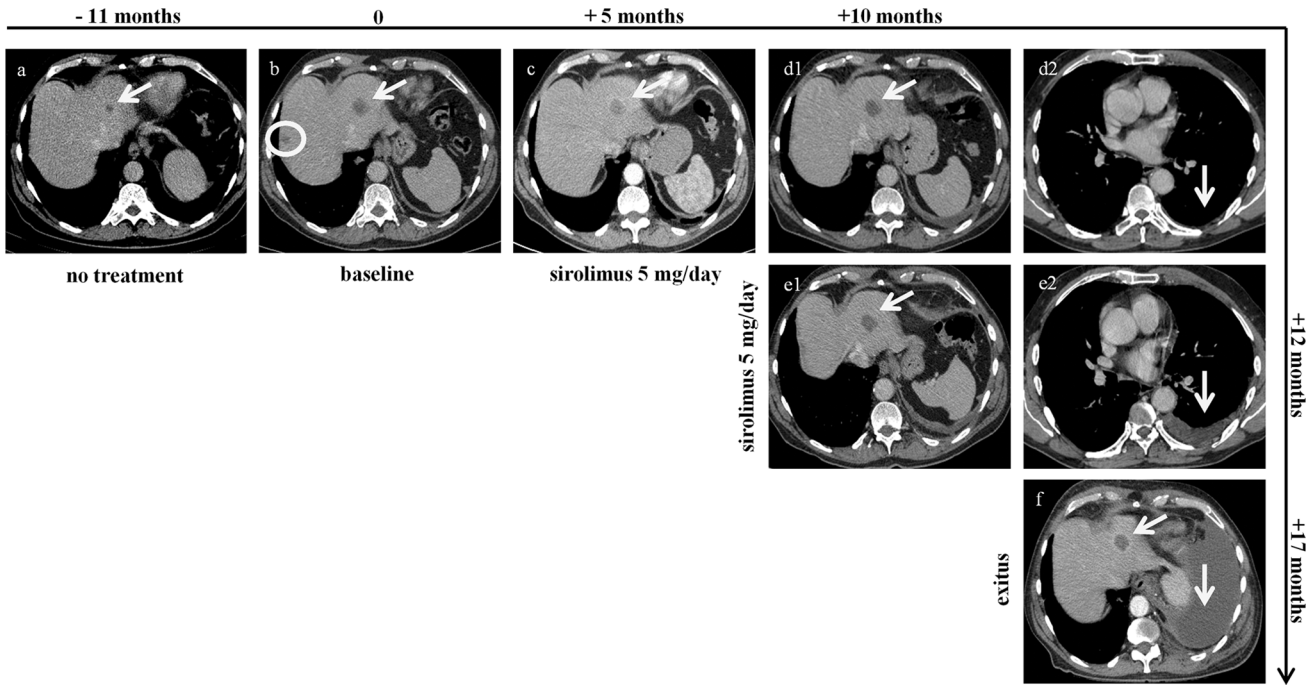

FIG. 3 Progression of pleural effusion and tumor-related systemic symptoms after response to sirolimus. CT scan of patient number 12 (Tables 1, 2). Multiple liver metastases from EHE were stable after 5, 10, and 12 months (**c**, **d**, **e**) of therapy with sirolimus compared with baseline (**b**), in a previously progressing patient (**a**, **b**). At 10 months, pleural effusion was not present (**d2**). The presence of pleural effusion was detected for the first time at 12 months (**e2**), together with worsening of tumor-related systemic symptoms while under sirolimus and in the lack of other radiologic signs of progression [i.e. liver metastases were stable (**e**), such as those located in the bone and lung (not shown), and no new lesions appeared]. The pleural effusion worsened quickly (**f**), as did the general condition of the patient, who died 5 months later without any other radiologic signs of progression. *CT* computed tomography, *EHE* epithelioid hemangioendothelioma

sign of activity in an indolent tumor such as EHE. Thus, a prospective controlled study would be needed in principle, but, given the rarity of this disease, there are major obstacles, at least under conventional methodological approaches. On the other side, we only treated progressive patients and, in four cases, progression was evident at sirolimus discontinuation, and a new response was achieved after rechallenge. Finally, tumor response was long-lasting in four patients receiving treatment for > 2 years.

Furthermore, RECIST criteria were found to be inadequate to catch disease progression since variations in serosal effusion is not considered by itself to be a sign of progression. In our analysis we reviewed all radiological assessments from baseline and found that the appearance/increase of effusion, particularly pleural effusion, was usually associated with other systemic signs, such as asthenia/prostration, weight loss, fever, and, more so, pain and dyspnea, even in those patients in which no other radiologic signs of progression [i.e. new lesion(s) and/or an increase in size of known lesion(s)] were detectable. This was followed in most cases by rapid deterioration of general conditions and death, with six patients dying within 1–8 months after the evidence of the effusion worsening, compared with a 16-month median OS in the whole series. Interestingly, the 10-month median 'enlarged PFS', obtained by considering as PD the worsening of pleural/peritoneal effusion and systemic tumor-related symptoms, is inferior to the 12-month median PFS according to RECIST. This observation is in line with data recently reported by Anderson et al., who found a correlation between the presence of pleural involvement and poor prognosis in lung and/or pleural vascular sarcomas, among which EHE.[8] Unfortunately, sirolimus was ineffective in the majority of these patients, with 7/9 cases with pleural/peritoneal effusions worsening in spite of therapy. This underlies the strong need for new active drugs to treat EHE.

PET was not useful in response evaluation as 4/10 patients had a decrease in FDG uptake which was not consistent with other clinical signs showing a disease progression. By contrast, the improvement in symptoms of the first patient of this series prompted us to use sirolimus in the other EHE patients. Unfortunately, in our study PS was not routinely assessed for all patients, but the evaluation of changes in PS and/or other quality-of-life metrics can add to radiologic response assessment, especially when responses are not dimensional, and should be considered in future studies on new agents in EHE.

Translational studies have suggested an important role for the PI3 K-Akt-mTOR pathway in sarcomagenesis,[21–23]

TABLE 1 Patient characteristics

ID	Age, years	Sex	Pathologic diagnosis	FISH for *WWTR1*	Tumor extent at disease onset	Past surgery (yes/no)	Past medical therapy	Past RT	Tumor extent at baseline	Effusion at baseline (pleural–peritoneal)	PS (ECOG)	Evidence of RECIST PD before starting sirolimus
1	53	M	EHE	NA	Lung, liver, bone	only biopsy	Yes (adriamycin, interferon, thalidomide)	Yes	Lung, liver, bone	No	3	Yes
2	46	M	EHE	Pos	Pleura	Only biopsy	Yes (interferon)	No	Pleura	Yes (pleural)	2	N[a]
3	61	M	EHE	Pos	Bone (multiple sites), bone marrow,	Only biopsy	Yes (interferon)	Yes	Bone (multiple sites), bone marrow,	No	2	Yes
4	22	F	EHE	Pos	lung, liver	Only biopsy	Yes (paclitaxel)	No	Lung, liver	No	1	Yes
5	51	F	Retiform HE	Neg	Lung, liver, bone	Yes (liver)	No	Yes	Lung, liver, bone	No	0	Yes
6	48	F	EHE	Pos	Lung, liver	Only biopsy	Yes (interferon)	Yes	Lung, liver, bone	No	1	Yes
7	50	F	EHE	Pos	Liver (multiple lesions)	Only biopsy	Yes (interferon)	No	Liver (multiple lesions)	No	1	Yes
8	40	F	EHE	Pos	Liver (multiple lesions), lung	Yes (primary tumor)	No	No	Liver (multiple lesions), soft tissue	No	0	Yes
9	50	M	EHE	Pos	Lung, liver	Only biopsy	Yes (interferon + bevacizumab)	Yes	Lung, liver	No	0	Yes
10	41	F	EHE	Pos	LN (mediastinum), bone	Only biopsy	No	Yes	LN, bone	Yes (pleural)	2	N[a]
11	58	F	EHE	Pos	Lung, bone	Only biopsy	No	Yes	Lung, liver, bone	Yes (pleural)	2	Yes
12	68	M	EHE	Pos	Lung, liver	Only biopsy	No	No	Lung, liver, bone	No	0	Yes
13	28	F	EHE	Pos	Lung, liver	Yes (liver, peritoneum)	Yes (gemcitabine)	Yes	Lung, liver, peritoneum	Yes (peritoneal)	2	N[a]
14	53	F	EHE	Pos	Pleura	Only biopsy	Yes (adriamycin + ifosfamide; gemcitabine + docetaxel)	Yes	Bone, pleura	Yes (pleural)	2	N[a]
15	48	M	EHE	Pos	Lung, bone. LN	Only biopsy	Yes (interferon)	Yes	Lung, bone, LN	No	0	Yes
16	38	F	EHE	Pos	Liver (multiple lesions)	Only biopsy	No	Yes	Brain, liver	Yes (peritoneal)	2	Yes
17	44	M	EHE	Pos	Lung, liver	Only biopsy	No	No	Lung, liver	No	1	Yes
18	55	M	EHE	Pos	Lung, liver	Yes (liver)	Yes (adriamycin; interferon)	No	Lung, liver	No	2	Yes

M male, *F* female, *EHE* epithelioid hemangioendothelioma, *HE* hemangioendothelioma, *NA* not assessable, *Pos* positive, *Neg* negative, *LN* lymphonodes, *RT* radiation therapy, *PS* performance status, *ECOG* Eastern Cooperative Oncology Group, *FISH* fluorescence in situ hybridization, *RECIST* Response Evaluation Criteria In Solid Tumors, *PD* progressive disease

[a] No evidence of progression by RECIST, but clinical signs of progression in terms of worsening of pleural/peritoneal effusion and tumor-related symptoms

TABLE 2 Clinical findings

ID	Effusion at baseline (pleural–peritoneal)	Daily dose of sirolimus, start/final (mg)	Best response, RECIST	Best response, PET	Worsening of pleural–peritoneal effusion	Interval progression	Reason for treatment interruption	Treatment duration (months)	PFS RECIST	Extended PFS[a]	Status at last FU	OS (months)
1	No	5/4	PR	PR	No	Yes	Patient choice	40	40	40	DOAC	48
2	Yes (pleural)	5/5	SD	PR	Yes (pleural)	NA	Worsening of pleural effusion + systemic symptoms without RECIST PD	12	13	12	DOD	13
3	No	5/3	PD	PR	No	NA	RECIST PD	4	4	4	DOD	14
4	No	5/5	PD	PR	Yes (peritoneal)	NA	RECIST PD	3	3	3	DOD	42
5	No	5/5	SD	PR	No	Yes	Patient choice	30	30	30	AWD	46
6	No	5/5	SD	NE	No	NA	RECIST PD	2	2	2	LOST	13
7	No	5/8	SD	PR	No	Yes	Patient choice	45	45	45	AWD	50
8	No	5/5	SD	PR	No	Yes	Surgery	25	25	25	AWD	34
9	No	5/8	SD	NA	No	NA	Ongoing	3	3	3	AWD	3
10	Yes (pleural)	5/5	SD	PR	Yes (pleural)	NA	Worsening of pleural effusion + systemic symptoms without RECIST PD	3	4	3	DOD	4
11	Yes (pleural)	5/5	SD	NE	Yes (pleural)	NA	Worsening of pleural effusion + systemic symptoms without RECIST PD	2	8	2	DOD	8
12	No	5/5	SD	PR	Yes (pleural)	NA	Worsening of pleural effusion + systemic symptoms without RECIST PD	12	16	12	DOD	16
13	Yes (peritoneal)	3/5	NE	NE	No	NA	Patient choice	0.5	24	11	AWD	24
14	Yes (pleural)	3/2	PD	NE	Yes (pleural)	NA	RECIST PD	1	1	1	DOD	6
15	No	5/5	SD	PR	No	NA	RECIST PD	5	3	3	AWD	3
16	Yes (peritoneal)	5/3	SD	NE	No	NA	Patient choice	10	12	10	DOD	12
17	No	5/5	SD	CR	No	NA	Ongoing	4	4	4	AWD	4
18	No	5/4	SD	NE	Yes (peritoneal)	NA	Worsening of pleural effusion + systemic symptoms without RECIST PD	2	2	2	DOD	5

CR complete response, *PR* partial response, *SD* stable disease, *PD* progressive disease, *NA* not assessed, *NE* not evaluable, *PFS* progression-free survival, *OS* overall survival, *FU* follow-up, *DOAC* dead of other causes, *DOD* dead of disease, *AWD* alive with disease, *LOST* lost to follow-up, *RECIST* Response Evaluation Criteria In Solid Tumors, *PET* positron emission tomography

[a] PFS as defined in the Materials and methods Section of the main text

providing the rationale for testing the clinical utility of mTOR inhibitors in all STS. Unexpectedly, overall results were disappointing,[24,25] with some exceptions, such as perivascular epithelioid cell tumor (PEComa)[26]. The mechanisms sustaining the activity of sirolimus in EHE are still to be elucidated. Unfortunately, we had no fresh/frozen tissues to assess mTOR status by biochemical analysis. We investigated the status of 4E-BP1, an mTOR effector, in five cases, and in all cases we detected phosphorylation of 4E-BP1, which silences 4E-BP1, thus allowing 4E-mediated protein synthesis. These very preliminary results would suggest mTOR signaling activation but the results need to be confirmed by biochemical analysis in a larger series. Sirolimus is also known for its ability to modulate T cell differentiation,[27] therefore an immunomodulatory effect cannot be ruled out.

Remarkably, 17/18 patients were metastatic at presentation. Of course, there may be a selection bias in this medical series. However, the proportion of EHE patients developing metastases is conventionally reported to be relatively low, i.e. 20–30 %.[2,7] Interestingly, in most of them the site of origin of the tumor could not be identified.

CONCLUSION

Our analysis shows that sirolimus may stabilize the tumor in advanced and progressive HE but was not effective in more aggressive cases, particularly in those patients showing worsening of their pleural effusion.

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