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Original Research

Clinical and genetic risk factors define two risk groups of extracranial malignant rhabdoid tumours (eMRT/RTK)☆

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☆ Dedicated to Professor Ivo Leuschner, member of the founding team of EU-RHAB, who died much too early.

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Abstract ***Introduction:*** Extracranial rhabdoid tumours are rare, highly aggressive malignancies primarily affecting young children. The EU-RHAB registry was initiated in 2009 to prospectively collect data of rhabdoid tumour patients treated according to the EU-RHAB therapeutic framework.
Methods: We evaluated 100 patients recruited within EU-RHAB (2009–2018). Tumours and matching blood samples were examined for *SMARCB1* mutations by sequencing and cytogenetics.
Results: A total of 70 patients presented with extracranial, extrarenal tumours (eMRT) and 30 with renal rhabdoid tumours (RTK). Nine patients demonstrated synchronous tumours. Distant metastases at diagnosis (M+) were present in 35% (35/100), localised disease (M0) with (LN+) and without (LN−) loco-regional lymph node involvement in 65% (65/100). *SMARCB1* germline mutations (GLM) were detected in 21% (17/81 evaluable) of patients. The 5-year overall survival (OS) and event-free survival (EFS) rates were 45.8 ± 5.4% and 35.2 ± 5.1%, respectively. On univariate analyses, age at diagnosis (≥12 months), M0-stage, absence of synchronous tumours, absence of a GLM, gross total resection (GTR), radiotherapy and achieving a CR were significantly associated with favourable outcomes. In an adjusted multivariate model presence of a GLM, M+ and lack of a GTR were the strongest significant negative predictors of outcome.
Conclusions: We suggest to stratify patients with localised disease (M0), GTR+ and without proof of a GLM (5-year OS 72.2 ± 9.9%) as 'standard risk'. Patients presenting with one of the features M+ and/or GTR− and/or GLM+ belong to a high risk group (5-year, OS 32.5 ± 6.2%). These patients need novel therapeutic strategies such as combinations of targeted agents with conventional chemotherapy or novel experimental approaches ideally within international phase I/II trials.

1. Introduction

Extracranial rhabdoid tumours (RT) are rare, aggressive malignancies arising predominantly in infants and young children <3 years. Most common locations are the kidneys (RTK − RT of the kidney) and other soft tissues (eMRT − extracranial, extrarenal malignant RT) (e.g. head and neck, liver, thorax, retroperitoneum) [1–3].

Within the UK and Germany the age standardized annual incidence rates of extracranial RT are 5–5.7 per million at age 1 and decrease to 0.1–0.2 at age 5 [1,4]. In the United States the annual incidence of extracranial RT among children less than 15 years is given as 0.19–0.32 per million [5].

Extracranial rhabdoid tumours are defined by distinct histologic appearance. Essentially all harbour

bi-allelic inactivation of the tumour suppressors *SMARCB1* [6,7] or rarely *SMARCA4* [8–10] in chromosomal regions 22q11.23 and 19p13.2, respectively. While recurrent genetic alterations explaining clinical heterogeneity have not been identified, DNA-methylation and expression profiling studies have uncovered distinct molecular subgroups of rhabdoid tumours. While the significance for their intracranial counterpart ATRT is well established [11] and while methylation profiling is an important asset in the diagnostics of childhood CNS tumours, its role is much less clear in sarcomas and associated neoplasias [12,13]. Approximately 25–30% of patients exhibit a germline mutation in *SMARCB1* (or *SMARCA4*) [14].

Survival rates for extracranial RT have not improved over recent years [3,15–20]. Tomlinson et al. reported a 23.2% 4-year OS for 142 patients with RTK (1969–2002) [15]. Analyses of data from the SEER database described a 33 ± 3.4% 5-year OS for 229 patients with malignant rhabdoid tumours of any anatomical region (1986–2005) [17]. In an EpSSG study (2005–2014) 3-year EFS and OS were 32.3% and 38.4%, respectively [20]. Recently, Cheng et al. demonstrated very poor 3- and 5-year OS of 23.7% and 18.4% for 53 patients with extracranial RT [21].

Due to the rarity of extracranial RT and a dearth of controlled clinical trials large data sets of uniformly treated patients are rare. As stratification criteria for soft tissue and kidney tumours have to be considered, the potential significance of clinical and genetic risk factors moves to the fore.

To develop strata for guiding future therapy, we evaluated whether clinical factors such as age, sex, metastases, synchronous tumours, and *SMARCB1* (germline) mutation status may help to identify patients at distinct risk.

2. Materials and methods

2.1. The EU-RHAB registry

The EUropean registry for RHABdoid tumours (EU-RHAB) provides a system of high quality reference diagnostics and expert counselling for diagnostic and therapeutic measures and a consensus treatment recommendation. EU-RHAB prospectively collects high quality data of patients with rhabdoid tumours of all anatomic locations treated according to EU-RHAB recommendation across participating European countries (Germany, Austria, Switzerland, Russia, Hungary, the Netherlands, Denmark, the UK, Sweden, Czech Republic, Poland, Slovakia, Portugal and Spain). Inclusion criteria cover (1) histopathological diagnosis of an (extracranial) rhabdoid tumour, according to WHO criteria confirmed by central pathology review [22], (2) age below 18 years and (3) informed consent by legal guardians to collect patient-related data. EU-RHAB has received continuous approval by the Ethics Committee of the University of Münster (ID 2009-532-f-S, last amendment 12/2016).

2.2. Consensus multimodal therapy

An overview of treatment recommendations, details on drug doses and protocol details are given elsewhere [11,23] (Supplemental Figure 1).

2.3. Diagnostic measures

The Pathology Reference Centre (Ivo Leuschner (+) and ChV, Kiel, since 2019 ChV, Bonn, Germany) reviewed all tumour samples according to WHO criteria and routinely included immunohistochemistry for SMARCB1/INI1 and SMARCA4/BRG1 [11,24]. Radiological response was evaluated according to criteria of the German National Reference Centre for Radiology (TK, Augsburg, Germany).

2.4. Toxicity

Toxicity was assessed according to version 3.0 of the Common Terminology Criteria for Adverse Events. Reporting of serious adverse events to the registry office was requested, but not monitored.

2.5. Tumour tissue collection and genetic analyses

All samples were collected at diagnosis. Cytogenetic studies including fluorescence-*in-situ*-hybridisation (FISH) as well as molecular studies including multiplex ligation-dependent probe amplification (MLPA) and sequencing of *SMARCB1* were performed at reference centres in Kiel (until 2016) and Ulm (since 2016), Germany (RSi), and Hamburg, Germany (RSch), as previously described [25,26] on blood samples and tumour tissues. DNA for MLPA and sequencing was isolated from FFPE tumour material. Reference sequences were: NM_003073.3/NP_003064.2 for *SMARCB1*.

2.6. Statistical analyses

Overall survival (OS) and event-free survival (EFS) rates were determined according to Kaplan–Meier estimates. OS was defined as the time from diagnosis until death of any cause. EFS was defined as time from diagnosis until first progression, relapse, death of any cause or last visit. Time-dependent factors such as radiotherapy (RTx), complete remission (CR) and maintenance therapy (MT) were evaluated using Cox regression for time-dependent covariates. p-values were regarded significant for $p \leq 0.05$ without adjustment for multiplicity.

3. Results

3.1. The majority of extracranial rhabdoid tumours present at an advanced stage

Clinical data of 100 patients, registered between 03/2009 and 01/2018 were analysed. Half of the patients were younger than 1 year at diagnosis. In 30 patients (30%) the tumour was located in the kidneys (RTK), and in 70% (70/100) extracranial, extrarenal (eMRT); most commonly in cervical and thoracic regions, and in the liver (Table 1a). Disease without loco-regional lymph node involvement (M0, LN−) was observed in 51% (51/100). A total of 14% (14/100) were diagnosed with loco-regional lymph node involvement (M0, LN+) and distant metastases (M+) at diagnosis were present in 35% (35/100) of patients. Nine patients (9%) demonstrated synchronous, multifocal tumours. Synchronous tumours were differentiated from metastatic spread by the concomitant occurrence of two major lesions in two separate compartments (e.g. brain and kidney) with or without signs of loco-regional M+ disease.

The IRS and SIOP stage distributions are shown in Supplemental Figures 2A and B. Clinical and treatment variables of these patients are summarised in Tables 1a, b and Supplemental Table 1.

Structural variants of SMARCB1 are most common in primary rhabdoid tumours and in germline.

Results on underlying genetic alterations of *SMARCB1* (tumour and/or blood) were available for 86% (86/100) of patients. For 51 of these, complete genetic data including germline and somatic mutational status comprising FISH, MLPA and sequencing were generated (Supplemental Figure 3). Among *SMARCB1* alterations 73.5% (n = 75/102 alleles) were structural variants. Single nucleotide variants were detectable in 26.5% (n = 27/102 alleles). All of the detected alterations were truncating including one splice site mutation predicted to be truncating. A total of 21% (17/81) demonstrated *SMARCB1* germline mutations. Only one of nine patients with synchronous tumours had no detectable GLM. Here heterozygous whole gene/partial deletions predominated (Supplemental Table 2). None of the tumours demonstrated loss of *SMARCA4*.

Table 1a
Clinical characteristic of 100 patients with extracranial rhabdoid tumours.

	Total
Median age [months]	**11.5 (0–206)**
Age group [n = 100]	
<12	50
12–36	21
>36	29
Origin [n = 100]	
Germany, Austria, Switzerland	69
Other countries	31
Sex [n = 100]	
Female	52
Male	48
Localisation [n = 100]	
RTK	**30**
Right	18
Left	12
eMRT	**70**
Cervical	14
Thoracic	13
Liver	9
Lumbosacral	4
Retroperitoneum	4
Orbit	4
Cervico-thoracic	3
Pelvic soft tissue	3
Scrotal	2
Mandible	2
Nasopharynx	1
Peritoneum	1
Adrenal gland	1
Vagina	1
Thigh	2
Hand	1
Heart	1
Parotid	1
Pre-auricular	1
Bladder	1
Cheeks	1
Metastasis [n = 100]	
M0, LN−	51
M0, LN+	14
M+	35
Synchronous tumours (with ATRT)	9
eMRT	6
RTK	3
Post-surgical tumour staging	
eMRT – IRS stage [n = 70]	**70**
I	14
II	12
IIIa	12
IIIb	10
IV	22
RTK – local SIOP stage [n = 30]	**30**
I	3
II	7
III	20

M0, LN−; localised disease without loco-regional lymph node involvement, M0, LN+; localised disease with loco-regional lymph node involvement, M+; distant metastasis.

3.2. Clinical factors associated with outcome

OS and EFS estimates of the whole cohort at 5 years were 45.8 ± 5.4% and 35.2 ± 5.1%, respectively (Figure 1A). At the time of analyses 49 of 100 patients had died. The median follow-up of survivors was 37 months (range 2–101). Age at diagnosis was a significant determinant of overall survival with patients ≥12 months at diagnosis demonstrating superior outcome. However, EFS was not significantly different for patients above or below 12 months at diagnosis (Figure 1B). Distant metastases (M+) at diagnosis were associated with a significantly inferior survival

Table 1b
Therapy outline and outcome of 100 patients with extracranial rhabdoid tumours.

	Total [n]
Gross total resection [n = 100]	
Yes	53
No	47
Any radiotherapy [n = 100]	
Yes	56
No	44
High dose chemotherapy [n = 100]	
Yes	21
No	79
Maintenance therapy [n = 100]	
Yes	21
No	79
Complete remission (of all sites involved) [n = 100]	
Yes	59
*After surgery**	16
+ chemotherapy	32
+ radiotherapy	11
No	41
Progression [n = 100]	
No	43
PD on CT**	36
PD after CT***	21
SAE [n = 13]	**13**
VOD	10
Encephalopathy	1
Severe infection	1
AML	1
Present status [n = 100]	
Complete remission	45
Stable disease	4
Progressive disease	2
Death	49

* A total of 16 patients achieved complete remission after surgery (IRS I = 10, IRS II = 3, local SIOP I = 2, local SIOP II = 1).
** Progression on chemotherapy, analysed within 4 months from diagnosis.
*** Progression after chemotherapy, analysed at 12 months from diagnosis, SAE; serious adverse event, VOD; veno-occlusive disease, AML; acute myeloid leukaemia.

compared to localised disease (M0) (Figure 1C). Patients with localised disease, with (M0, LN+) and without loco-regional lymph node involvement (M0, LN−) demonstrated distinct, however not significantly different, 5-year OS and EFS rates. Furthermore, synchronous tumours were associated with a significantly inferior survival (5-year OS 0% vs. 51.2 ± 5.7% without synchronous tumours). Anatomic location of the tumour seems to have an impact on survival; the 5-year OS for RTK was inferior to eMRT; however, this did not reach significance (larger numbers are needed) (Supplemental Table 1). Postsurgical stage at diagnosis was also an important factor for outcome. Patients with IRS stages I/II/IIIb and local SIOP stage I had superior 5-year OS versus IRS IIIa/IV and local SIOP III (Table 2).

3.3. Treatment and survival

A total of 60% of patients completed chemotherapy. GTR was associated with significantly superior OS ($p < 0.05$; Figure 1D), however the difference in 5-year OS and EFS estimates of RTK patients with GTR compared to patients with incomplete resections was even higher [51.9% vs. 0% ($p = 0.0003$) and 50.7% vs. 0% ($p = 0.0016$)]. Radiotherapy was also of prognostic importance. Patients who were treated by RTx survived in 56.6 ± 6.9% ($p < 0.05$). Median age at RTx was 27 months (5–210 months).

Improvement in survival was neither seen for patients treated by HDCT nor with a maintenance regimen. Nevertheless, patients who had achieved a CR upon treatment had significantly superior outcome (Supplemental Table 1). A total of 16 patients achieved CR after surgery, n = 32 after additional chemotherapy, and n = 11 following RTx.

Therapy refractory disease and relapse were very poor prognostic factors. In total, 57 patients suffered from relapses or progressions. In 35 patients PD occurred locally only, 9 patients demonstrated local relapse and distant metastases as well. Thirteen patients suffered from relapse of distant metastasis (CNS = 6, lungs = 6, multiple = 1). Survival upon relapse or early progression was in general poor; only 11.1 ± 5.8% of the patients with a lack of response or progression on chemotherapy (analysed within 4 months from diagnosis) and 26.6 ± 24.9% with early relapse (analysed at 12 months from diagnosis) were alive 5 years following diagnosis. Patients without progression demonstrated 5-year OS rates of 91 ± 3.9% (Supplemental Table 1).

3.4. Germline mutation of SMARCB1 is an important risk factor

Presence of a GLM was observed in 17/81 patients and had significant predictive power on univariate testing as well as in a multivariate model (Figure 1E). No further significant correlations were detected between genetic and clinical factors.

3.5. Toxicity of treatment

Toxicity was noteworthy, but manageable. All patients demonstrated grade 3 or 4 haematologic toxicity at any time during therapy. Thirteen patients had SAEs (severe adverse events). Ten experienced VOD (veno-occlusive disease), one CNS toxicity (encephalopathy), one a severe infection and one a secondary AML. Four SAEs were associated with a lethal outcome (Supplemental Table 3).

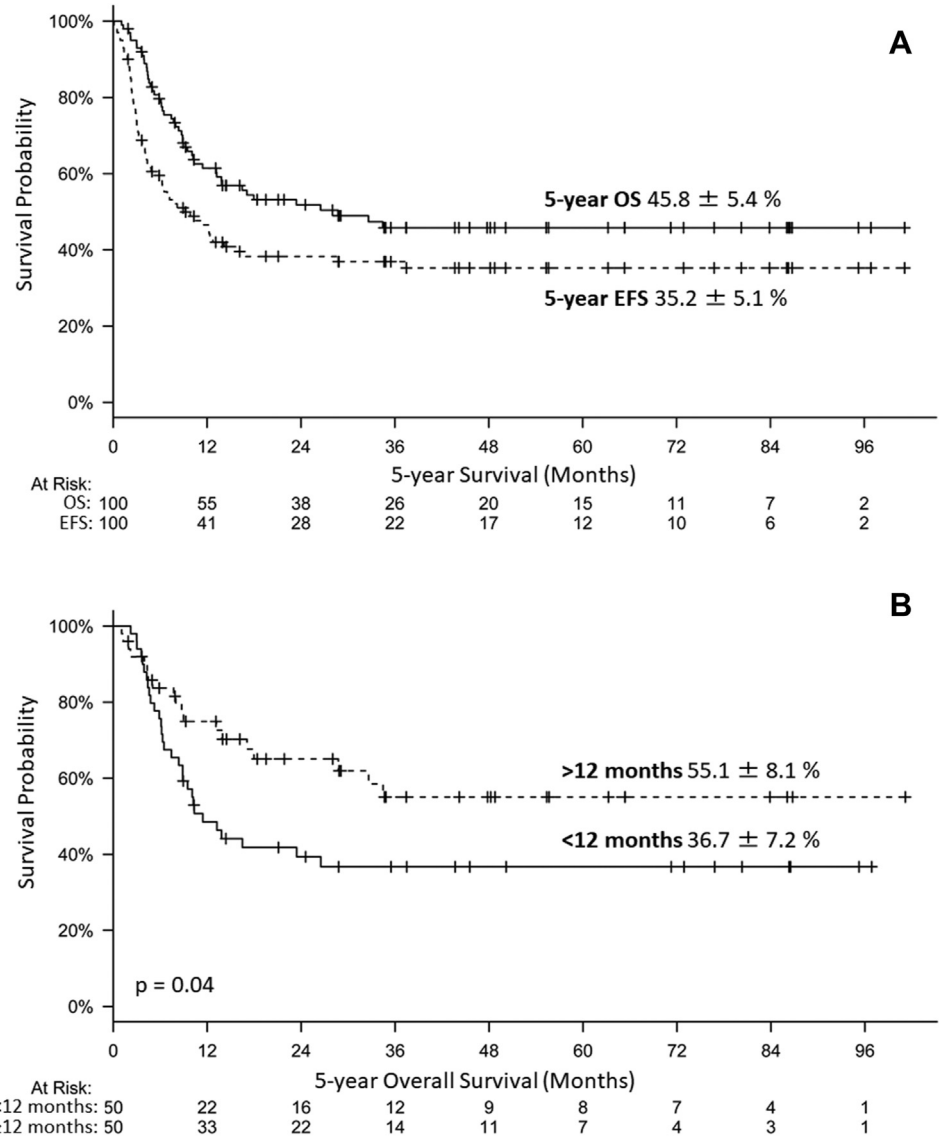

Fig. 1. **Five-year overall survival (OS) of 100 consecutive patients treated according to the EU-RHAB consensus therapy.** (A) The 5-year OS of 100 patients with extracranial RT was 45.8 ± 5.4%, while the 5-year event-free survival (EFS) of the same cohort was 35.2 ± 5.1%. (B) The 5-year OS was 55.1 ± 8.1% for patients diagnosed at the age of 1 year and older and 36.7 ± 7.2% for those <1 year at diagnosis. (C) The 5-year OS was 16.5 ± 6.8% for patients with distant metastasis (M+) at diagnosis and 62.1 ± 6.7% for patients with localised disease (M0) with and without loco-regional lymph node involvement. (D) The 5-year OS was 59.2 ± 7.4% for patients who achieved gross total resection (GTR+) and 31.4 ± 7.4% for those with incomplete resection (GTR−). (E) The 5-year OS was 6.3 ± 6.1% for patients diagnosed with germline mutation (GLM+) and 55.6 ± 7% for those without germline mutation (GLM−).

3.6. A combined clinical and genetic risk model for the stratification of extracranial RT

All risk factors were included in a multivariate, stepwise Cox-regression model to create a risk model. Only distant metastases (M+), GTR+ and GLM + remained independent prognostic factors (Table 2). Employing this model, we differentiated two risk groups (Figure 2A and B):

1) a standard risk group (SR = 27) [localised disease with (M0, LN+) and without loco-regional lymph node involvement (M0, LN−), and GTR+ and GLM−] demonstrated significantly superior 5-year OS and EFS values 72.2 ± 9.9% and 58.3 ± 10.4%, compared to.

2) a high risk group (HR = 66) presenting with one of the features M+ and/or GTR− and/or GLM+ (5-year OS 32.5 ± 6.2%, EFS 22.1 ± 5.4%).

4. Discussion

EU-RHAB provides a large and clinically well-annotated cohort of patients with extracranial RT.

Our analyses comprise currently one of the largest cohorts of extracranial rhabdoid tumours treated within

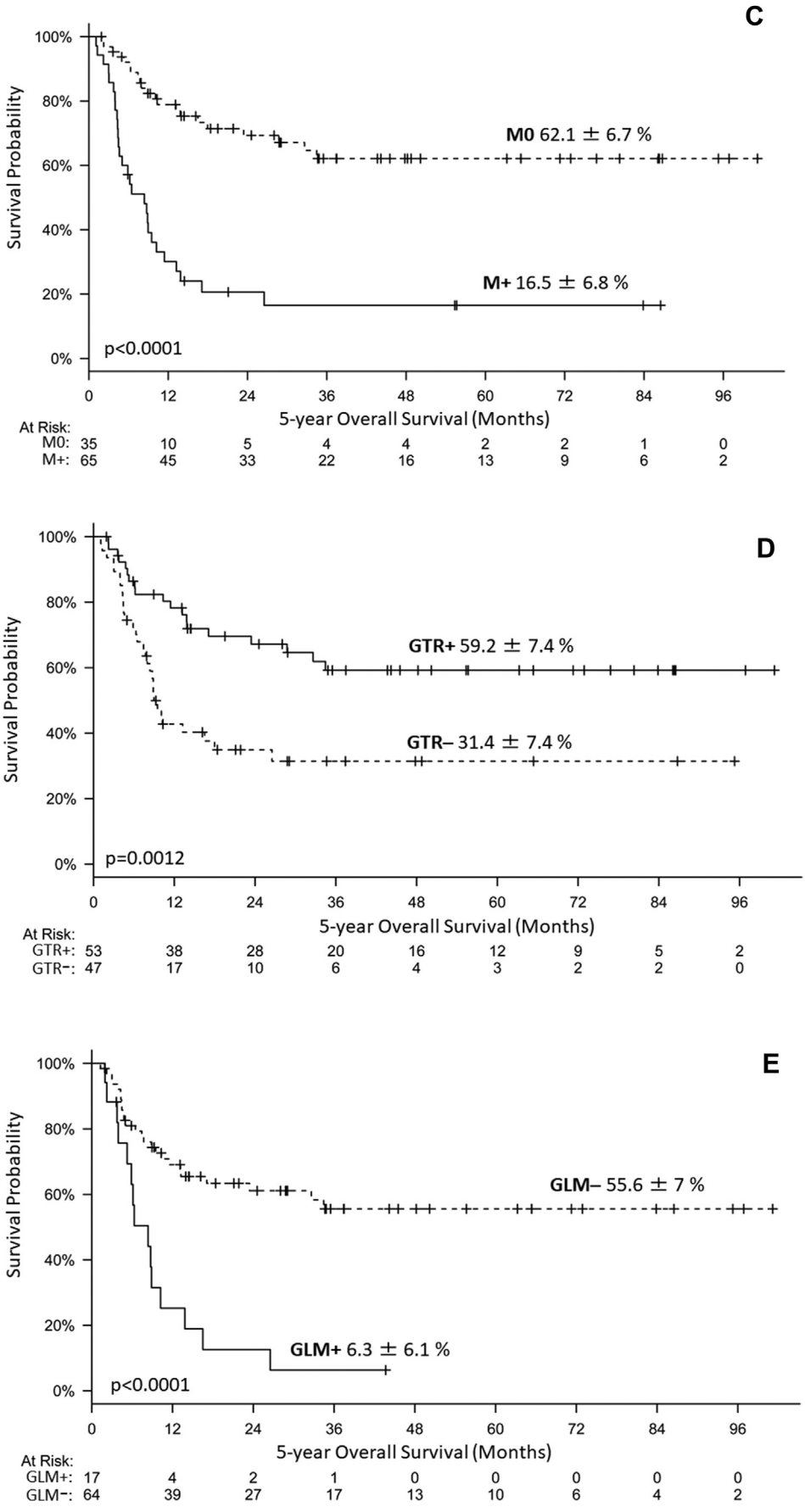

Fig. 1. (*continued*).

Table 2
Risk factors of overall survival according to univariate and multivariate analyses.

Prognostic Factors	Univariate analysis	Multivariate analysis	
	p-value	RR (95% CI)	*p*-value
GLM yes versus **no**	**<0.0001**	**8.5** (3.1–22.9)	**<0.0001**
M+ versus **M0**	**<0.0001**	**7.3** (3.1–17.3)	**<0.0001**
GTR yes versus **no**	**0.0012**	**0.46** (0.22–0.93)	**0.03**
RTx **yes** versus **no**	**0.0003**	0.42 (0.16–1.04)	0.06
Age <1 year versus **≥1 year**	**0.04**		n.s.
SYN **yes** versus **no**	**0.0023**		n.s.
IRS I/II/IIIb versus **IIIa/IV**	**<0.0001**		n.s.
SIOP local stage **I** versus **III**	**0.048**		n.s.

Age, location, germline mutation (GLM), metastatic stage (M-stage), distant metastasis (M+), localised disease (M0) with and without loco-regional lymph node involvement, synchronous tumours (SYN), gross total resection (GTR), IRS stage, SIOP stage, conventional chemotherapy, radiotherapy (RTx), high-dose chemotherapy, maintenance therapy, complete remission, progression or early relapse, and genetic subgroups were examined. Factors with significance on a univariate and multivariate level are listed. RR; relative risk, CI; confidence interval, n.s.; not significant.

the same therapeutic framework. We demonstrated stable, improved long-term (96 months) overall survival of 45.8 ± 5.4% in line with or better than other series [15–20]. Moreover, a substantial part of patients (SR) demonstrated a remarkable long-term survival.

4.1. Clinical and genetic markers for stratification of extracranial RT

Similar to recent studies [NWTS (<5 months 8.8%) [15], SEER series (<12 months 17%) [17], EpSSG (<12 months 20.1%) [20], and according to our own data (<12 months 36.7 ± 7.2%), age remains a significant determinant of survival. In contrast to the EpSSG study we did not demonstrate its significance on multivariate analysis. An age cut-off was determined at 12 months, since children under 12 months are in general not irradiated.

One of our independent prognostic factors was distant metastasis. Only 16.5 ± 6.8% of our patients with distant metastases survived for 5 years or longer (2-year OS in EpSSG study 13%) [20]. Another independent prognostic factor was achievement of a GTR, which was associated with a significant superior overall survival (59.2 ± 7.4%). Our results revealed a significant survival advantage for patients with lower IRS and local SIOP stage. Patients diagnosed with stages IRS I/II (n = 26) and local SIOP I (n = 3) presented significantly superior 5-year overall survival rates when compared to results from other groups (5-year OS IRS stage I 74 ± 13.2%, IRS II 66.7 ± 13.6%, local SIOP I 100%), e.g. the NWTS study (OS for local SIOP stage I/II was 41.8%) [15] or the SEER series [17]. We conclude that gathering data across nations in a collaborative effort will facilitate a clearer distinction of risk factors.

Based on currently available data, the role of radiotherapy in the treatment of extracranial RT cannot be defined conclusively. The SEER series indicated improved survival for patients treated by RTx [17]. However, in the NWTS series after adjusting for age and stage, the significant effect of RTx disappeared [15]. Brennan et al. did not confirm a significant benefit of radiotherapy [20]. In our series patients treated with RTx survived significantly longer (5-year OS, 56.6 ± 6.9%) compared to those not treated by RTx. Still, the benefits of RTx in patients with SIOP stage I or IRS I await further definition. None of 5/16 patients with IRS I/local SIOP I stage treated without RTx relapsed. More data pooling of international series is urgently needed. The role of HDCT in extracranial RT remains ill-defined. Similar to the SIOP studies [19] we could not confirm any survival advantage of HDCT ($p > 0.05$). We corroborated the important role of achieving a CR as identified by the NWTS [17] and the EpSSG [20]. Patients with CR demonstrated 5-year OS rates of 53.9 ± 6.8%. Progressive disease on therapy and relapse remained important poor prognostic factors. Thirty-six percent of patients demonstrated progress on chemotherapy and 21% exhibited early relapse after chemotherapy (in EpSSG progression 49.5%) [20].

Patients with RTK have a very dismal prognosis following relapse or progression, which may be attributable to a higher rate of GLM in RTKs. In fact, 33% of all completely characterized patients with RTKs presented a GLM contrasting with 16% in eMRT. Our data are unique in that we were able to prospectively analyse a large proportion of our patients for constitutive and somatic mutations of *SMARCB1*. Presence of a GLM proved to be a robust, significant, independent prognostic marker as verified in 81 patients; however, the number of GLM may be underestimated due to cryptic GLM cases with e.g. mosaicism. Patients with a GLM did very poorly with 5-year survival rates in the range of only 6.3 ± 6.1%. When adjusting for age (≥1 and <1 year of age) significance was maintained highlighting this, an important HR-marker.

4.2. Proposal of a clinical and genetic risk model for stratification

Using our multivariate model we recognise two risk groups. Patients at standard risk (SR) demonstrated significantly superior 5-year OS rates, compared to those of a high risk group (HR).

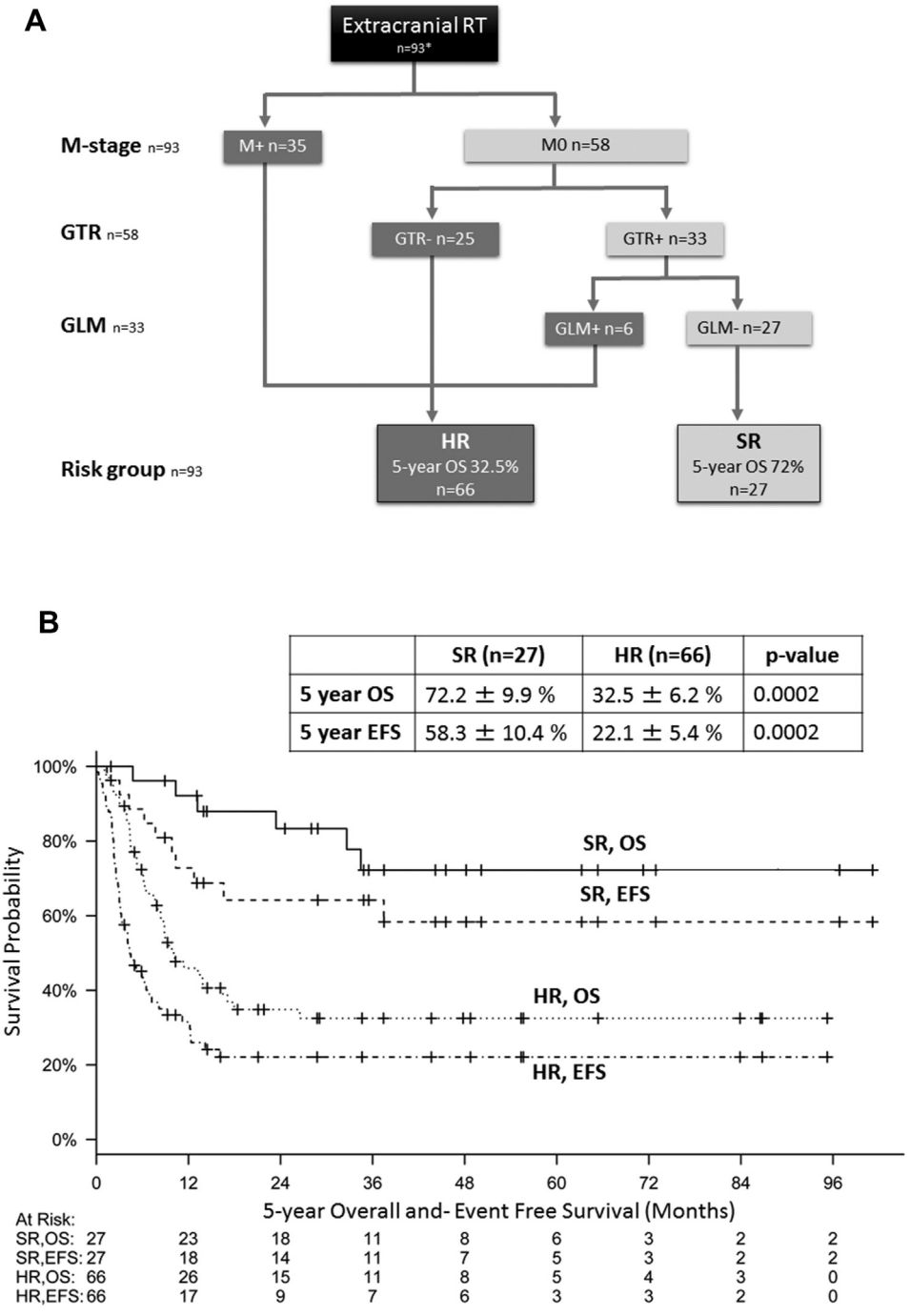

Fig. 2. **A combined clinical and genetic risk model for stratification in extracranial RT.** (A) Potential Risk Model for the stratification of extracranial rhabdoid tumours. M-stage [M+; distant metastasis, M0; localised disease with and without loco-regional lymph node involvement], gross total resection (GTR) and germline mutation (GLM) may predict the potential risk of patients affected by extracranial RT independently of any other clinical or known genetic factor. Using this model we differentiated two risk strata: the HR group was characterized by patients presenting with one of the three high risk factors (M+ and/or GTR− and/or GLM+). If one of the three negative prognostic factors was present, the patient was added to HR group. Among a total of 19 patients with unavailable GLM status (n = 12/19 patients had M+ and/or GTR−) inclusion into the HR stratum was possible. The standard risk group (SR) containing patients with absence of all of three negative prognostic factors (M0 and GTR+ and GLM−). Only patients with absence of all three negative prognostic factors were taken to multivariate analysis. ∗In contrast to the HR group for n = 7/19 patients despite the availability of M0 and GTR+, stratification was not possible due to missing GLM data. (B) Kaplan-Meier estimates of patients with risk factors, such as metastatic stage (M-stage), gross total resection (GTR) and germline mutation (GLM), were analysed for their 5-year overall (OS) and event-free survival (EFS). Two risk strata were delineated: standard risk group (SR) with 5-year OS and EFS rates of 72.2 ± 9.9% and 58.3 ± 10.4% and high risk (HR) with 5-year OS and EFS rates of 32.5 ± 6.2% and 22.1 ± 5.4%.

Should SR patients be treated further with standard conventional chemotherapy approaches such as the EU-RHAB or EpSSG approach, and do HR patients benefit from novel treatment strategies? Furthermore, are there patients apt for reduced treatment intensity?

Patients with high risk factors may benefit from inclusion into phase I trials using target specific (e.g. CDK4/6 inhibitor, ribociclib), but also mechanism specific, epigenetic approaches (e.g. HDAC-, DNMT- or EZH2-inhibitors) as frontline therapy [27,28]. Checkpoint inhibitors could be attractive tools in high risk extracranial RT [29–31]. A comprehensive review on experimental approaches for MRT is given elsewhere [32]. The evidence of the presence and importance of molecular subgroups of extracranial RT was first demonstrated by Chun et al. [12,33]. As additive information on molecular specifics and potential drug targets evolve, the number of subgroups may increase and subsequently the absolute number of patients belonging to a distinct subgroup may become smaller and smaller. For further analyses international collaborations are thus gaining even more importance.

Criteria for inclusion into the study were identical in participating countries. A certain inclusion bias can still not be excluded. In German speaking countries all patients were included according to identical inclusion criteria, as the investigators were contacted for each individual patient to provide expert counselling and diagnostic support. Nevertheless, we cannot exclude that some patients may be precluded access to the EU-RHAB treatment because they are deemed palliative at diagnosis or too young to receive intensive treatment. We estimate the number to be rather small however as in previous studies we have been able to demonstrate feasibility of the treatment approach in the youngest of children [16,34].

Recruiting sufficient numbers of patients along with biomaterial from many different institutions has tremendously helped in building a network of dedicated clinician scientists, a solid database for retrospective analyses, as well as in defining standards for genetic testing and therapy. Nevertheless, as individual risk groups are still rather small and analyses barely reach statistical significance we suggest to validate the data in an independent cohort, ideally in the setting of a multinational, controlled clinical trial.

CRediT author statement

MCF: Conceptualization, Methodology, Validation, Resources, Funding acquisition, Writing - Original Draft, Visualization, Supervision, Data Curation; RF: Conceptualization, Methodology, Writing - Original Draft, Supervision; NG: Conceptualization, Methodology; JG: Software, Formal analysis; KN: Validation, Writing - Original Draft, Project administration, Visualization, Data Curation; DK, MF, PH, TS, ST, WW, OB, CHF, LG, PHD, PGS, CK, KH, RK, AL, FA, MJG, CHM: Investigation; MB, KK, SL, TK, CV, PM, PHD, RS, RSi, SB, UK, MCF, RF, NG, KNY: Writing - Review.

Conflict of Interest statement

We hereby confirm, that there are no conflict of interest disclosures from any authors.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.ejca.2020.10.004.

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