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Second malignant neoplasms after treatment of childhood cancer

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Abstract

The aim of this study was to determine therapy-related risk factors for the development of second malignant neoplasm (SMN) after childhood cancer. The German Childhood Cancer Registry (GCCR) registers all childhood malignancies since 1980 including SMN. A nested case–control study with 238 SMN cases and 450 controls was conducted. A confirmatory, as well as an explorative, analysis was performed. Radiotherapy showed a small effect on the risk of SMN for doses $\geqslant$65 Gy. Regarding the chemotherapeutical agents, we saw increased Odds Ratios (OR) for high doses of cyclophosphamide (CP $>$8000 mg/m^2 OR = 6.3 (95% Confidence Interval (CI): 1.3–30.2)), cisplatinum (DDP $>$435 mg/m^2 OR = 2.8 (95% CI: 1.1–6.7)) and mercaptopurine (MP $>$5000 mg/m^2 OR = 4.5 (95% CI: 1.1–18.9)). Patients jointly receiving high doses of MP ($>$5000 mg/m^2) and dexamethasone (DEXA $\geqslant$1200 mg/m^2) had an OR = 6.9 (95% CI: 1.2–40.3). Our results could be added to those of other investigations to give indications for modifying future therapeutic strategies for childhood cancer.

Keywords: Second malignant neoplasms; Childhood cancer; Therapy-related; Case–control study; Epidemiology; Registry

1. Introduction

The prognosis of children with a malignancy has improved considerably in the past 30 years due to improvements in therapy. A major contribution to this has come from clinical trials such as those of the Society for Paediatric Oncology and Haematology (Gesellschaft für Pädiatrische Onkologie und Hämatologie, GPOH).

The effective, but aggressive, therapy protocols may have late effects, of which second malignant neoplasms (SMN) are among the most severe. Increasing evidence can be found in the literature that a certain fraction of SMN are therapy-induced. For example, it is well known that chemotherapy with alkylating agents or epipodophyllotoxins is a risk factor for the development of acute leukaemia in the first 7–10 years after diagnosis [1–5]. Furthermore, radiotherapy has been shown to be associated with an increased risk for solid tumours 10–15 years after treatment and later [6–11].

The paediatric oncologists thus find themselves challenged to develop minimally toxic therapy strategies, while retaining maximum effectiveness regarding survival and quality of life.

The German Childhood Cancer Registry (GCCR) registers all childhood malignancies for all of Germany since 1980, including East Germany since 1991 [12]. In a previous publication, we described the incidence rates of SMN in Germany registered by the GCCR up to 1995 [13]. The present case–control study was conducted to investigate the influence of cancer therapy on the risk of developing a SMN using data from this population-based registry. The aim of the study was to determine the potential risk of each single chemotherapeutical agent, as well as of radiotherapy.

2. Patients and methods

2.1. Patients with SMN

The GCCR systematically registers all malignancies, including SMN, diagnosed at age $<$15 years among German residents since 1980 (completeness approximately

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95%) [12]. There is no age restriction for SMN. Active follow-up is performed in cooperation with the clinical trials, the co-operating hospitals and the parents and patients themselves. Within the project reported here, we extended the inclusion criteria for primary malignancies of patients with SMN to patients diagnosed at age <25 years and residents of Switzerland, Austria, and the Netherlands. The SMN diagnosis and the diagnosis combinations were all validated by the principle investigators of the corresponding clinical trials. A total of 276 patients with SMN were known to the registry with a diagnosis of first and secondary neoplasm between 1 January 1980 and 28 February 1998. The cohort included 24 203 patients, with 16 293 patients known to be alive up to 28 February 1998. The median follow-up of the patients alive was 6 years (range 0–18 years).

2.2. Nested case–control study

Based on these 276 patients, a nested case–control study was performed. Retinoblastoma as a primary neoplasm was excluded ($n=13$), because of insufficient follow-up data. For some SMN cases, no therapy data was obtained ($n=13$) or no matching control could be found ($n=12$). Finally, 238 cases with SMN were included and matched 1:2 to controls from the registry by year of diagnosis of the primary malignancy, gender, year of birth and type of primary malignancy according to the International Classification of Childhood Cancer as presented in Parkin and colleagues in Ref. [14]. A control had to be alive and free of SMN by the age of occurrence of the SMN of its matched case. For some SMN cases, only one control was available, a total of 450 controls were finally included in the study. We obtained detailed data on diagnostic parameters, therapy, clinical history and vital status for cases and controls from the treating hospitals.

2.3. Therapy data

For most individuals, we either obtained detailed therapy information (18% cases, 52% controls) or information as to which standardised clinical trial protocol the patient had been assigned (77% cases, 35% controls). For the rest, adherence to a clinical trial could be assumed based on the diagnosis and year of diagnosis (5% cases, 12% controls). Patients without detailed information were thus included as if they had been treated exactly according to protocol, an assumption which could be verified for most of those for whom detailed information was available.

The doses (per m^2 body surface) of each chemotherapeutical agent were accumulated over all of the therapy and all relapses up until the time of the diagnosis of the SMN for cases, or for controls up until the time of the diagnosis of the SMN for the matched case. The aim of the study was to investigate the risk of the single agents quantitatively. While it would be desirable to summarise all doses in a group of drug (e.g. alkylating agents), rules for computing such a cumulative dose do not exist. For three drugs (methotrexate, mercaptopurine, thioguanine), we present the results distinguished between initial and maintenance therapy, as for these drugs we found different results according to the phase of treatment. History of radiotherapy was reported with specification of the irradiation site and total cumulative dose in grays (Gy) for the most irradiated site.

2.4. Statistical analysis

The data was analysed using conditional logistic regression for matched data. In the first step, the dose distribution of each chemotherapeutical agent was classified into three groups by tertiles with 'medication not given' as the reference class. Radiotherapy was handled analogously. Additionally, a four level variable with categories 'no therapy', 'chemotherapy only', 'radiotherapy only', and 'both' was investigated. In a later explorative analysis, additional data-driven cut-points were defined (based on dose–response curves fitted by non-parametric smoothing models) and multiple models including more than one therapy were investigated. Tumour stage of the primary malignancy was included as a covariate or subsetting variable into some of the regression analyses. Results are presented as Odds Ratios (OR) with their respective 95% Confidence Intervals (95% CI). A significant trend test ($P<0.05$) implies that the risk increases with dose. Significance in the explorative analyses should not be interpreted as in a confirmatory test.

3. Results

3.1. Descriptive results

An overview of the diagnoses of the 238 SMN cases is presented in Table 1. The most frequent diagnosis combinations were acute lymphoid leukaemia (ALL) as a primary malignancy followed by AML; and ALL followed by astrocytoma.

The median time intervals between primary and second malignancy are shown in Table 2. The intervals are likely to be underestimated, especially for solid tumours, because of the bias by the shorter observation times of more recently registered primary cases.

3.2. Radiotherapy

Regarding radiotherapy, the OR for very high doses ($\geqslant 65$ Gy) was increased, although this was not

Table 1
Distribution of diagnoses for primary and second malignant neoplasms (SMN) for cases included in the case–control study ($n = 238$)

First malignancy	Second malignant neoplasms											
	Leukaemia	Lymphoma	CNS tumour	Sympathetic nervous system	Retinoblastoma	Kidney tumour	Hepatic tumour	Bone tumour	Soft-tissue tumour	Germ cell tumour	Carcinoma	Other	Total
Leukaemia	30	19	29	1	1	2	0	2	2	1	17	1	105
Lymphoma	11	6	2	0	0	1	0	1	2	1	7	0	31
CNS tumour	9	1	10	1	0	0	0	2	1	0	4	1	29
Sympathetic nervous system	8	2	1	0	0	0	0	0	0	0	0	0	11
Kidney tumour	0	0	6	0	0	0	0	0	0	0	1	1	8
Hepatic tumour	1	0	0	0	0	0	0	0	0	0	0	0	1
Bone tumour	10	2	2	0	0	0	0	2	1	1	1	1	20
Soft-tissue tumour	11	1	3	0	1	0	0	6	1	0	1	1	25
Germ cell tumour	3	0	0	0	0	0	0	0	0	1	1	0	5
Carcinoma	1	0	0	0	0	0	0	0	1	0	0	0	2
Other	0	0	0	0	0	0	0	0	1	0	0	0	1
Total	84	31	53	2	2	3	0	13	9	4	32	5	238

CNS, Central Nervous System.

statistically significant (Table 3). Such high doses are rare and usually only seen cumulatively in relapsed cases. Correcting this OR for chemotherapy, the radiotherapy shows a small contribution to the risk of SMN on its own (OR = 1.6 (95% CI: 0.3–8.4)). The results (confirmatory analyses as well as explorative analyses) did not change when the radiation dose was included as a continuous variable in the model.

When investigating the location of the second tumour, 45% of the cases had their SMN within the radiation field (RT field) of the primary malignancy. For 13 out of 17 thyroid carcinomas, it is known that the tumour lay within the RT field; for the remaining four this can be assumed.

In more than 60% of all patients, primary tumours and relapses were treated with a combined chemo- and radiotherapy. The OR of 'combined radiation and chemotherapy' (67% cases, 63% controls) versus 'none of these' (3% cases, 5% controls) was 3.0 (95% CI: 1.1–8.4).

3.3. Results for specific agents of chemotherapy

The results of the confirmatory, as well as of the explorative, analyses for different chemotherapeutical agents are presented in Table 4. For comparison, we added the evaluation of the carcinogenic risk to humans of the International Agency for Research on Cancer (IARC) [15] for these agents.

3.3.1. Alkylating agents

Significant risk increases could be shown for cyclophosphamide (CP), cisplatinum (DDP), dacarbacine (DTIC) and, with restrictions, for procarbazine (PRO), mostly for high doses. A multiple model analysis for CP did not alter the results of the explorative analyses notably, i.e. the effect of CP is independent of other concurrent therapies (including radiotherapy).

The risk of SMN increased with increasing dose of DDP (trend test significant). A small group of those treated with high doses of DDP was also given high doses of dacarbacine (DTIC > 2700 mg/m^2); these were almost all cases with SMN. Correcting for DTIC slightly decreased the risk attributable to DDP alone (OR = 2.0 (95% CI: 0.5–7.5)). Including any other therapies simultaneously in a model with DDP did not change the risk estimates for DDP noticeably. DDP showed an effect all by itself.

Table 2
Median time interval between primary and SMN ($n = 238$)

	Time interval (years) (min–max)
All malignancies	5, 2 (10 days–16, 2)
Second leukaemias	3, 3 (6 months–11, 7)
Second solid tumours	6, 5 (10 days–16, 2)

DTIC did not show a clear dose–response relationship. However, doses >2688 mg/m^2, led to a significant 6.7-fold risk. A large fraction of this may be explained by correlated doses of DDP and radiotherapy. Correcting for those, only a non-significant 2.2-fold (95% CI: 0.3–18.9) increase of risk could be ascribed to DTIC. Only a few patients were given DTIC. High doses were given to 7 patients only, the 5 SMN cases among them were each also given >430 mg/m^2 DDP.

3.3.2. Antimetabolites

Up to a dose of 5000 mg/m^2, Mercaptopurine (MP) led to only a small risk increase, whereas 5000 mg/m^2 the risk was increased 4.5-fold. MP-treated patients are usually also given the cortisone dexamethasone (DEXA), especially those receiving high MP doses. The doses are highly positively correlated. Correcting for this, the risk of a high dose of MP alone was no longer significant (OR = 3.1 (95% CI: 0.7–14.5)). The database is not large enough to investigate interactions. Assuming an additive effect, receiving a high dose of MP (>5000 mg/m^2) and a high dose of DEXA ($\geqslant 1200$ mg/m^2) together would result in a 6.9-fold (95% CI: 1.2–40.3) increased risk.

For methotrexate (MTX) and cytarabine (ARA-C), a spurious, non-monotonous association could be seen. These medications were given in doses differing by orders of magnitude across the therapy protocols. For MTX, doses higher than 19 600 mg/m^2 showed an increased OR.

3.3.3. Epipodophyllotoxins

There were no clear dose–response relationships for etoposide (VP16) and teniposide (VM26). The explorative analyses yielded slightly increased risks for medium doses. Cases who were treated with medium doses of any of these cannot be identified as a special group, however, cases with medium doses of VM26 also received relatively high doses of CP, and cases who received medium doses of VP16 also received high doses of DDP and DTIC. It is possible that the seemingly non-monotonous relationships are artefacts caused by these correlations. High doses of VP16 (>5000 mg/m^2) were given to only 3 out of 131 cases and controls.

3.3.4. Antibiotics

Bleomycin (BLEO) was given to only a few patients. High doses (given to 3% of the cases and 1% of the controls) led to a non-significant 6.7-fold increase in risk. Some of this effect is explained by these patients also being treated with CP, the risk ascribable to high doses of BLEO (>80 mg/m^2) only is OR = 4.6 (95% CI: 0.5–42.2). Almost all BLEO-treated patients also received CP with a positively correlated dose.

3.3.5. Corticosteroids

DEXA led to a noticeable and monotonous risk increase (trend test $P = 0.07$, confirmatory analysis). Some of this effect could be ascribed to other correlated therapies such as MP or Doxorubicin (ADR), but not all (see MP for discussion of joint effect).

3.4. Additional results

The study intended to detect a potential risk increase (one-sided question). However, the exploratory analysis suggested that some therapies also seem to have a small protective effect for higher doses. These are: Maintenance therapy with TG (TG_D), with MP (MP_D) and with MTX (MTX_D) as well as L-Asparaginase (ASP) and actinomycin D (AMD) (Table 4). Including stage as a confounder in the models either made no difference or, as the treatment regime often depends directly on stage, made the analysis impossible. Matching by age and requiring survival of the controls until the occurrence of SMN in the case, often led to concordance with regard to stage in a matched group.

Out of 238 patients with SMN, 9 (4%) were reported to have undergone bone marrow transplantation (BMT) in the treatment of the first malignancy compared with 6 (1%) controls. Despite the small numbers, this difference was significant ($P < 0.05$).

Genetic anomalies were observed in 13 cases (3 trisomy 21, 3 balanced translocations, 3 monosomy, 1 Turner Syndrome, 3 other chromosomal anomalies) and in 4 controls (3 trisomy 21, 1 Turner Syndrome), making them 5-fold more frequent in the case ($P < 0.001$). Analyses, excluding patients with known genetic anomalies, yielded the same or only marginally changed

Table 3
Results for radiotherapy as a potential risk factor for SMN ($n = 238$ cases, $n = 450$ controls)

Dose (Gy) classified by 33th and 66th percentile	% Exposed cases/controls per percentile	OR (95% CI)	Dose (Gy) data-driven cut points	% Exposed cases/controls per dose group	OR (95% CI)
$\leqslant 18$	27/28	1.1 (0.6–1.8)			
$>18 \leqslant 35$	19/19	1.1 (0.6–1.9)	<65	65/65	1.1 (0.7-1.7)
>35	21/19	1.2 (0.7–2.3)	$\geqslant 65$	2/1	2.1 (0.6-7.7)

OR, odds Ratio; 95% CI, 95% Confidence Intervals.

Table 4
Results for chemotherapeutical agents as potential risk factors for second malignant neoplasms ($n = 238$ cases, $n = 450$ controls)

Exposure[a] (IARC-Class[b])	Median dose (mg/m^2) (min–max)	Dose (mg/m^2) classified by 33rd and 66th percentile	% Exposed cases/controls per percentile	OR (95% CI)	Dose (mg/m^2) data-driven cut-points	% Exposed cases/controls per dose group	OR (95% CI)
Alkylating agents							
CP (group 1)	3000 (387–18 863)	⩽2022	18/21	1.0 (0.5–1.8)	⩽4000	45/49	0.9 (0.5–1.6)
		>2022⩽3333	20/20	1.2 (0.6–2.1)	>4000⩽8000	9/6	1.7 (0.8–3.6)
		>3333	24/18	1.6 (0.9–2.8)[c]	>8000⩽12 000	3/1	6.3 (1.3–30.2)
					>12 000	5/2	8.7 (1.9–40.0)[c]
DDP (group 2A)	320 (14–720)	⩽240	3/6	0.5 (0.2-1.4)	⩽150	2/2	1.1 (0.3–4.1)
		>240⩽435	4/4	1.9 (0.7-4.9)	>150⩽320	2/6	0.2 (0.1–1.1)
		>435	7/3	2.8 (1.1-6.7)[c]	>320⩽500	6/4	3.4 (1.1–10.4)
					>500	3/1	2.8 (0.9–8.6)
DTIC (group 2B)	2000 (618-4000)	⩽1589	1/1	3.3 (0.5–23.4)			
		>1589⩽2688	0.4/2	0.7 (0.1–6.5)	⩽2500	2/2	1.7 (0.4–7.0)
		>2688	2/0.5	6.7 (1.0–44.3)[c]	>2500	2/1	5.4 (0.9–32.4)
IFO (group 3)	18 000 (2000–98 909)	⩽10 873	9/9	1.0 (0.5–1.8)	⩽10 000	9/9	1.0 (0.5–1.8)
		>10 873⩽30 000	11/9	1.3 (0.7–2.5)	>10 000⩽40 000	13/12	1.1 (0.6–2.0)
		>30 000	7/10	0.3 (0.1–0.8)	>40 000	5/8	0.4 (0.1–1.0)
PRO (group 2A)	1400 (238–8400)	⩽1148	3/3	1.8 (0.5–6.0)	⩽1000	3/2	2.3 (0.7–7.6)
		>1148⩽2800	7/2	8.3 (1.7–39.9)	>1000⩽2200	4/2	4.6 (1.2–17.4)
		>2800	0.4/3	0.2 (0.0–2.0)	>2200⩽5000	2/2	1.9 (0.4–10.1)
					>5000	0.4/2	0.3 (0.03–2.7)
Antimetabolites							
ARA-C	1813 (280–46 615)	⩽1800	28/25	1.7 (0.5–5.8)	⩽1000	6/3	2.9 (0.7–12.0)
		>1800⩽2376	9/10	1.3 (0.4–5.0)	>1000⩽4800	39/40	1.2 (0.4–4.3)
		>2376	17/18	1.4 (0.4–4.8)	>4800⩽11 000	2/1	2.9 (0.4–19.8)
					>11 000⩽21 000	3/5	0.8 (0.2–3.5)
					>21 000	5/6	0.9 (0.2–4.1)
MP (group 3)	3057 (500–6480)	⩽2550	14/14	1.6 (0.6–4.1)	⩽3000	20/19	1.7 (0.7–4.3)
		>2550⩽3125	15/12	2.0 (0.8–5.4)	>3000⩽5000	18/20	1.5 (0.6–3.8)
		>3125	12/14	1.3 (0.5–3.4)	>5000	3/1	4.5 (1.1–18.9)
MTX (group 3)	4000 (20–168 000)	⩽2067	17/18	1.3 (0.7–2.7)	⩽15 000	36/33	1.7 (0.9–3.2)
		>2067⩽19 646	18/18	1.7 (0.8–3.5)	>15 000⩽40 000	16/14	2.2 (0.98–4.8)
		>19 646	20/16	2.4 (1.1–4.9)[c]	>40 000⩽100 000	1/2	0.6 (0.1–3.4)
					>100 000	2/2	1.2 (0.2–7.3)
TG	840 (280–5760)	⩽840	18/17	1.6 (0.8–3.0)			
		>840⩽1008	10/8	1.6 (0.8–3.4)	⩽2800	37/35	1.5 (0.8–2.8)
		>1008	12/13	1.2 (0.5–2.7)	>2800	4/3	1.3 (0.3–5.0)

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Table 4 (*continued*)

Exposure[a] (IARC-Class[b])	Median dose (mg/m^2) (min–max)	Dose (mg/m^2) classified by 33rd and 66th percentile	% Exposed cases/controls per percentile	OR (95% CI)	Dose (mg/m^2) data-driven cut-points	% Exposed cases/controls per dose group	OR (95% CI)
Epipodophyllotoxins							
VM26	331 (142–1320)	⩽325	3/5	0.6 (0.3–1.5)	⩽400	7/8	0.9 (0.5–1.7)
		>325⩽594	5/4	1.3 (0.6–2.8)	>400⩽800	7/4	1.8 (0.8–3.7)
		>594	6/4	1.7 (0.8–3.6)	>800	0.4/1	0.5 (0.1–4.4)
VP16	1000 (155–9682)	⩽900	7/8	0.8 (0.4-1.6)	⩽500	2/4	0.4 (0.1–1.4)
		>900⩽1350	5/6	0.8 (0.4–2.0)	>500⩽1800	12/12	1.0 (0.6–1.9)
		>1350	8/6	1.6 (0.8–3.3)	>1800⩽5000	5/3	2.1 (0.8–5.2)
					>5000	0.4/0.5	1.1 (0.1–12.0)
Antibiotics							
ADR (group 2A)	123 (15–679)	⩽120	35/31	1.2 (0.7–2.2)	⩽90	14/18	0.8 (0.4–1.5)
		>120⩽160	15/18	0.8 (0.4–1.5)	>90⩽210	42/37	1.2 (0.7–2.0)
		>160	22/24	0.8 (0.4–1.5)	>210⩽350	5/8	0.6 (0.2–1.3)
					>350	11/10	1.2 (0.5–2.8)
AMD (group 3)	6 (0.9–62)	⩽5.4	7/6	0.8 (0.3–2.3)	⩽3	2/2	0.6 (0.2–2.4)
		>5.4⩽8.4	7/6	0.7 (0.2–2.2)	>3⩽10	13/11	0.7 (0.3–2.1)
		>8.4	4/7	0.3 (0.1–1.1)	>10	3/5	0.3 (0.1–1.3)
BLEO (group 2B)	80 (17–138)	⩽40	1/1	0.9 (0.2–4.5)	⩽40	1/1	0.8 (0.1–4.0)
		>40⩽90	1/1	1.6 (0.3–8.4)	>40⩽80	0.4/1	0.8 (0.1–9.2)
		>90	2/1	–[d]	>80	3/1	6.7 (0.8–59.4)
DNR (group 2B)	120 (42–360)	⩽120	18/20	1.1 (0.5–2.6)	⩽110	6/7	1.0 (0.4–2.9)
		>120⩽144	12/11	1.4 (0.6–3.4)	>110⩽230	33/33	1.3 (0.6–2.9)
		>144	13/12	1.3 (0.5–3.2)	>230	4/4	1.4 (0.5–4.3)
Vinca-alkaloids							
VIN (group 3)	18 (6–48)	⩽12	1/0.2	7.9 (0.7–93.2)			
		>12⩽24	1/1	1.9 (0.3–14.4)	⩽10	0.4/0.2	2.3 (0.1–41.2)
		>24	0.4/1	0.7 (0.1–7.2)	>10	2/2	1.3 (0.3–6.0)
VCR (group 3)	12 (1.5–90)	⩽9	25/29	0.9 (0.5–1.8)			
		>9⩽13	28/23	1.4 (0.7–2.8)	⩽8	12/18	0.6 (0.3–1.3)
		>13	24/27	0.8 (0.4–1.6)	>8	65/60	1.1 (0.6–2.0)
VDS	9 (2–20)	⩽6.2	1/4	0.1 (0.02–1.1)	⩽4	0.4/1	0.3 (0.03–2.8)
		>6.2⩽12	6/4	1.6 (0.7–3.6)	>4⩽8	1/3	0.5 (0.1–1.8)
		>12	2/1	1.7 (0.5–5.9)	>8⩽12	5/3	1.5 (0.6–3.4)
					>12	2/1	1.7 (0.5–6.1)

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Table 4 (*continued*)

Exposure[a] (IARC-Class[b])	Median dose (mg/m^2) (min–max)	Dose (mg/m^2) classified by 33rd and 66th percentile	% Exposed cases/controls per percentile	OR (95% CI)	Dose (mg/m^2) data-driven cut-points	% Exposed cases/controls per dose group	OR (95% CI)
Corticosteroids							
DEXA	245 (13–1432)	⩽210	12/13	1.2 (0.6–2.1)	⩽200	12/12	1.2 (0.6–2.1)
		>210⩽300	14/14	1.4 (0.7–2.8)	>200⩽800	25/23	1.4 (0.8–2.6)
		>300	13/10	1.6 (0.8–3.0)	>800	2/1	2.6 (0.7–9.7)
PRED (group 3)	1834 (9–13 818)	⩽1680	15/18	0.8 (0.4–1.8)	⩽2000	40/43	0.9 (0.4–1.9)
		>1680⩽1839	20/19	1.1 (0.5–2.4)	>2000⩽5500	13/11	1.2 (0.5–2.7)
		>1839	18/18	1.1 (0.5–2.4)	>5500	1/1	0.9 (0.2–5.7)
Other							
ASP (U/m^2)	145 000 (30 000–840 769)	⩽120 000	16/14	1.5 (0.5–4.3)	⩽80 000	4/4	1.2 (0.4–4.1)
		>120 000⩽177 231	13/14	1.3 (0.4–3.8)	>80 000⩽220 000	28/28	1.4 (0.5–4.1)
		>177 231	14/14	1.4 (0.5–4.0)	>220 000⩽400 000	10/7	2.4 (0.7–7.8)
					>400 000	1/4	0.5 (0.1–2.6)
Drugs in maintenance therapy							
MTX_D	1479 (200–7925)	⩽1141	17/13	0.9 (0.3–2.8)	⩽500	1/1	1.1 (0.1–8.6)
		>1141⩽1595	13/14	0.7 (0.2–2.1)	>500⩽2200	36/35	0.8 (0.3–2.5)
		>1595	11/16	0.6 (0.2–1.8)	>2200⩽4500	3/5	0.4 (0.1–1.7)
					>4500	1/2	0.9 (0.2–5.6)
MP_D	25 200 (3600–76 494)	⩽19 444	16/13	1.0 (0.4–2.9)	⩽20 000	17/14	1.0 (0.4–2.8)
		>19 444⩽27 678	14/14	0.8 (0.3–2.4)	>20 000⩽45 000	23/26	0.8 (0.3–2.1)
		>27678	11/15	0.6 (0.2–1.9)	>45 000	0.4/2	0.2 (0.02–1.8)
TG_D	25 146 (837–36 500)	⩽14 519	2/2	1.0 (0.2–4.3)			
		>14 519⩽25760	2/2	0.4 (0.1–2.4)	⩽20 000	2/2	0.6 (0.1–2.6)
		>25 760	0.4/2	0.2 (0.0–1.4)	>20 000	2/4	0.4 (0.1–1.3)

Reference group are the cases and controls not exposed to the respective agent.

[a] For full name of the abbreviations see the Appendix.

[b] Where available, the evaluation of the International Agency for Research on Cancer (IARC) is added [15]: group 1 = agent is carcinogenic to humans; group 2A = agent is probably carcinogenic to humans; group 2B = agent is possibly carcinogenic to humans; group 3 = agent is not classifiable as to its carcinogenicity to humans; group 4 = agent is probably not carcinogenic to humans.

[c] Test of trend $P < 0.05$.

[d] Not analysable.

results for radiotherapy and chemotherapeutical agents (explorative analysis). Other congenital malformations were only moderately more frequent (4.2% versus 3.3%, respectively).

An overview of the results showing increased risks for SMN is given in Table 5.

4. Discussion

The GCCR has been recording malignancies in children for 20 years. The GCCR is capable of conducting population-based analyses across all diagnoses. To ensure a complete registration of SMN by the GCCR an active long-term follow-up is carried out.

The distribution of the primary diagnoses in the registry population is comparable to that of other registries [14], with the exception of brain tumours, for which the registration rate is relatively low in Germany [16].

Data regarding the therapy of the primary malignancies were analysed with the help of a nested case–control study. Chemotherapy and radiation were investigated as potential risk factors for SMN.

The Children's Cancer Group of the US pointed out that if expected doses of treatment are substituted for the actual doses in studies of late effects of cancer survivors, great care should be used in conclusions about the effects of chemotherapy dosage [17]. As we did not have actual doses for all of the patients at our disposal, it was necessary to estimate the bias of the results. To do this, all chemotherapy doses were recalculated as specified in the clinical trial protocol and were compared with the cumulative doses actually administered (where available). Both data sets compared very well with one another ($>90\%$ agreement when classified into tertiles, data not shown).

A further source of bias could be the exclusion of the patients with retinoblastoma and the loss of cases without therapy data or matching partners. Concerning year of birth, year of diagnosis, gender or first diagnosis (besides retinoblastoma), the excluded cases did not form a special group. Thus, the results are unlikely to be affected to a relevant degree.

The matching criteria we used for the case–control study, especially diagnosis and year of diagnosis, may have introduced some 'overmatching', as diagnosis and year of diagnosis are major determinants for the assignment to a specific therapy protocol. This may result in a certain degree of underestimation of the effect of a medication and of the radiotherapy. A case and its control may differ in therapy only for individual reasons, when a new protocol was initiated just the previous or next year, when a case and its control fall into different risk or stage groups, and in cases where 1 patient had a relapse with according treatment and the other had not. The power of a matched study is determined not so much by the total number of cases available, but by the number of match groups, where at least one control falls into a different dose category as the respective case (discordant match triplets). Especially for therapies given rarely, the likelihood of having a sufficient number of discordant triplets is very low.

Too few discordant triplets may also be the reason why radiotherapy had a visible, but not statistically significant effect in the study. However, causality seems likely, as a large proportion of secondary solid tumours were located within the radiation field of the primaries. This has been published previously for SMN such as brain tumours, thyroid carcinoma, breast carcinoma, or osteosarcoma [6,8,10,11,18,19]. An elevated risk for radiotherapy, especially in children diagnosed before the age of 5 years, was found by a Scandinavian group [9]. We could not confirm this finding with our data (data not shown).

We saw significantly elevated risks of SMN for high doses of CP, DDP and DTIC (also when accounting for DDP and radiotherapy). Alkylating agents are regularly mentioned in the context of SMN [1,8,20,21]. We can thus confirm the increased risk of SMN.

The non-significant elevated risk caused by DEXA was not seen for other corticosteroids (prednisone (PRED), prednisolone (PREDO)). Thus, this observation

Table 5
Overview of increased risks for the development of a SMN found in the nested case–control study

Significantly increased risks	Non-significantly increased risks
High doses of: Cyclophosphamide (CP) Cisplatinum (DDP)	High doses of: Dexamethasone (DEXA) (corrected for MP) Bleomycin (BLEO) (corrected for CP) Dacarbacine (DTIC) (corrected for DDP and radiotherapy)
Mercaptopurine and dexamethasone together	Mercaptopurine (MP) (corrected for DEXA)
Bone marrow transplantation	High doses of radiotherapy
Genetic disorders (trisomy 21, balanced translocations, monosomy, Turner Syndrome, other chromosomal anomalies)	

is difficult to interpret. It may merely reflect the effects of other toxic substances, which were given to the same patients. MP and DEXA were given in highly correlated doses. Including MP or various combinations of the other therapies with a risk increasing effect in the regression model did not reduce the effect estimate of DEXA to zero. Apart from the potential artefacts in the data collection and the possibility of a chance finding, it may also be hypothesised that the immunosuppressive effect of DEXA may increase the risk of SMN.

Topoisomerase-II-inhibitors are frequently mentioned in the context of secondary leukaemias [5,22,23]. We could not show a distinct effect for VP16 and VM26 within the dose ranges observed in our study. The median dose of 1000 mg/m^2 VP16 given to our patients was much lower than in the studies which reported an increased risk of SMN; this difference in dose may explain the different outcomes.

The evaluation of the IARC is more or less in accordance with our results.

We saw a possibly protective effect especially for maintenance therapies. Certainly, a chance finding cannot be excluded. However, we also observed this for at least three different drugs administered as maintenance therapy, suggesting this is not a chance finding. Maintenance therapy may suppress the development of a SMN for some time and thus, at least, increase the time interval between the first and second malignancy.

A strong immunosuppressive effect is observed in children who underwent BMT during treatment of their first malignancy. Therefore, it was not surprising to see BMT more often in the patients with SMN compared with the control group. This observation represents the confirmation of a well known risk factor [24], as do the effects of the reported genetic disorders, which were 5 times more frequent in children with SMN.

5. Conclusions

Studies concerning the risk of SMN after childhood cancer, which are not restricted to defined malignancies or a special clinical study group, are rare. Our results add to the evidence that cancer therapy is included among the risk factors for the development of SMN. The consequences for future therapy have to be discussed very carefully. The SMN project at the GCCR continues. We will update the investigations presented here and address further questions regarding SMN after childhood cancer.

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Appendix

Drug therapies and their abbreviations in alphabetical order:

ADR, Doxorubicin (=Adriablastine, Adriamycin); AMD, Actinomycin D (=Dactinomycin); ARA-C, Cytarabine; ASP: L-Asparaginase; BLEO, Bleomycin; CP, Cyclophosphamide; DDP, Cisplatinum; DEXA, Dexamethasone; DNR, Daunorubicin; DTIC, Dacarbacine; IFO, Ifosfamide; MP, Mercaptopurine; MP_D, Mercaptopurine maintenance therapy; MTX, Methotrexate; MTX_D, Methotrexate maintenance therapy; PRED, Prednisone; PREDO, Prednisolone; PRO: Procarbazine; TG, Thioguanine; TG_D, Thioguanine maintenance therapy; VCR, Vincristine; VDS, Vindesine; VIN, Vinblastine; VM26, Teniposide; VP16, Etoposide

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