Ella Antikainen

IMMUNE RECONSTITUTION AFTER CHILDHOOD CANCER DEPENDS ON TREATMENT INTENSITY

Syventävien opintojen kirjallinen työ

Kevätlukukausi 2022

Ella Antikainen

IMMUNE RECONSTITUTION AFTER CHILDHOOD CANCER DEPENDS ON TREATMENT INTENSITY

Kliininen laitos

Kevätlukukausi 2022

Vastuuhenkilö: Schuez-Havupalo Linnea

TURUN YLIOPISTO

Lääketieteellinen tiedekunta

ANTIKAINEN, ELLA: Immune reconstitution after childhood cancer depends on treatment intensity

Syventävien opintojen kirjallinen työ

Hematologia

Toukokuu 2022

The aim of this study was to analyze immune reconstitution in the pediatric population after completion of cancer treatment. We retrospectively studied all children (N=79) who had been treated for cancer at our center in Turku University Hospital starting treatment during the years 2014-2019. In a separate dataset we merged all leukemia patients who started treatment during the years 2014-2019 including all high-risk leukemia patients treated between 2009-2019 (N=39). Patients were classified in high risk and low risk treatment intensity groups based on a previously validated tool ITR-3. We collected data on baseline parameters, treatment-related aspects and post-treatment immunological recovery, namely neutrophil and lymphocyte counts, immunoglobulin levels, CD19, CD4 and NK cell counts. Immunological parameters were followed until their normalization.

Our data showed a fast recovery of the above-named immunological parameters for the majority of current pediatric oncologic treatments. Treatment for high-risk leukemia, AML, medulloblastoma and mature B-cell lymphoma was associated with prolonged recovery times for a substantial proportion of cases. These patients with prolonged recovery time were mostly classified within the higher ITR-3 group, which means slower immune reconstitution can be anticipated for certain treatment groups.

According to our study, as childhood cancer treatment protocols have advanced to be less toxic, they also often cause less immunosuppression. This means that post-treatment transmission-based precautions would not be needed in most cases. Nevertheless, patients treated with high intensity treatment need special follow-up after completion of childhood cancer treatment.

Immune reconstitution after childhood cancer depends on treatment intensity

Ella Antikainen, Anu Huurre, MD, PhD, Marika Grönroos, MD, PhD, Laura Korhonen, MD, PhD, [a] , Päivi Lähteenmäki, MD, PhD, Schuez-Havupalo Linnea

[a] Department of Pediatrics and Adolescent Medicine, Turku University Hospital and University of Turku;

Corresponding author: Schuez-Havupalo Linnea

Keywords: Cellular immunity, malignant diseases, chemotherapy, children

Abbreviations:

Funding Source:

Word count of the abstract: 292

Word count of the text: 3880

Number of references: 6

The number of graphic presentations: 6 tables and 1 figure

Disclosure of potential conflict of interest: None

Abstract

Objective: The aim of this study was to analyze immune reconstitution after completion of chemotherapy in the pediatric population. Only a few previous studies examine immune system recovery after completed cancer treatment. **Procedure**: We analyzed all children (N=79) who had been treated for cancer starting treatment during the years 2014-2019 at our center at the Turku University Hospital, Finland. In a separate dataset we merged all leukemia patients who started treatment during the years 2014-2019 and all high risk leukemia patients starting treatment during the years 2009-2019 (N=39 patients). Stem cell transplant patients were excluded.
We retrospectively collected data on baseline parameters, treatment-related aspects and post-treatment immunological recovery, namely neutrophil and lymphocyte counts, immunoglobulin

levels, CD19, CD4 and NK cell counts. Immunological parameters were followed until their normalization. Treatment intensity was stratified according to a previously validated tool (ITR-3). We analyzed the effects of treatment intensity on normalization of the above-mentioned immunological parameters within ALL patients and across the entire treatment range.
Results: Our data showed that treatment intensity has a major effect on immune system recovery after completion of treatment. At completion of treatment or at 4 months post-treatment most patients had normal immunological parameters both in high and low treatment intensity groups, but patients classified in the high intensity group had low parameters more often than patients in the low intensity group. **Conclusion:** Our data suggest a fast recovery of the above-named immunological parameters for the majority of current pediatric oncologic treatments. Treatment for high risk leukemia, AML, medulloblastoma, and mature B-cell lymphoma was associated with prolonged recovery times for a substantial proportion of cases. Patients with prolonged recovery times were mostly classified within the higher ITR-3 group, which means slower immune reconstitution can be anticipated for certain treatment groups.

Introduction

Cancer treatments have various effects on the patients' immune system. Cancer treated pediatric patients are usually immunosuppressed. Meanwhile cancer itself might already have affected their immune system. Transmission-based precautions often lead to a relatively isolated lifestyle of the pediatric patient and thus burden the entire family.
Currently there are limited prospective data on reconstitution of the immune system after pediatric oncological treatment (Torben Ek et al. 2005; Judit Gadó et al. 2006; Gabor T. Kovacs et al. 2008; Sofia Kosmidis et al. 2008)
Treatments for pediatric cancers have advanced so that the majority of patients can be cured (G.T. Kovacs et al. 2008). Still, during treatment patients are immunosuppressed and may have lethal infections as a treatment complication. The degree of immunosuppression depends on treatment intensity which varies considerably depending on disease and the treatment used. Isolation policies are often in place, but they differ internationally and between treatment centers. The majority of treatment centers set limitations in order to avoid viral infections that may cause postponement of cancer treatments and, on the other hand, extra hospitalizations because of fever and other symptoms. We aimed to study the effect of different treatment programs on the immune system and immune system recovery.

In this study, we analyzed post-treatment immunological recovery including neutrophil and lymphocyte counts, immunoglobulin levels, CD19, CD4 and NK cell counts. Secondly, we aimed to analyze the relationship between treatment intensity and normalization of the above-mentioned immunological parameters.

Based on the literature, our hypothesis was that treatment intensity would have a major effect on immune system recovery after treatment. It would be important for physicians to have more comprehensive information of the patient data for treatment decisions regarding isolation policies. Treatments are continuously developing, and modern treatments aim for minimal toxicity while optimizing survival outcomes. Therefore, it is important to examine the above research questions as a function of time considering changes in treatment protocols.

Patients and methods

Patients

We retrospectively studied 79 children who had been treated for malignant diseases at the Turku University Hospital between 2014-2019. In a separate dataset we merged all leukemia patients who underwent treatment during the years 2014-2019 including chemotherapy-treated high risk leukemia patients treated between 2009-2019 (N=39 patients). The children were aged 0 to 16 years at the time of diagnosis. In this study we included all children who had been treated with cytostatic treatment with or without radiotherapy but excluded children who underwent stem cell transplantation. We excluded patients with relapses during treatment but included patients who experienced a relapse after completed cancer treatment, if relapse treatments did not chronologically interfere with our follow-up of immunological parameters (Table 5).

Cancer treatments were split into two groups based on their intensity. Treatment intensity was stratified according to a previously validated tool (ITR-3) (Kazak A et al. 2012). ITR-3 categorizes patients into 4 groups. Given the inclusion criteria to our study we combined ITR-3 group 1+2 to form the low treatment intensity group and ITR-3 group 3+4 to form the high treatment intensity group.

Data collection and definitions

Routine blood tests including immunological parameters were taken from all children at around 1 and 4 months post-treatment and up until their normalization thereafter. Not all subjects had immunological parameters available for both time points (1 and 4 months post treatment), but all

had data available at either point depending on the normalization of parameters. Due to these missing data we merged time points such as to determine, whether counts had normalized by 4 months post-treatment. We furthermore collected data on clinical baseline parameters and treatment-related aspects. Immunological parameters investigated included neutrophil and lymphocyte counts, immunoglobulin levels, CD19, CD4 and NK cell counts.
Normalization of results was based on the age-appropriated reference values for immunoglobulin levels. CD19, CD4 and NK cell stratification was based on their normal levels (Table 2 and Table 3).
Analyses of CD19 cells and immunoglobulins excluded patients with rituximab treatment. The patients were classified to have neutropenia if their neutrophil level was less than 1 10 9/l at the time of first blood tests after completed chemotherapy.

Statistics

In our analyses p values less than 0.05 were regarded as statistically significant.
Background variables were compared between treatment intensity groups using the Chi-Square test (Fisher's exact test for small numbers) and Mann Whitney U test. The latter was applied since patients' ages did not follow a normal distribution.
For the leukemia cohort, recovery of immunological parameters was analyzed stratifying by treatment groups using the Fisher's exact test. For the main study cohort, the effect of treatment intensity on recovery of immunological outcome measures was first estimated using the Chi-square test (Fisher's exact test for small numbers). In a second step, adjusted binary logistic regression analysis was performed including all applicable background variables (age, sex, treatment intensity, pre-existing chronic disease, previous repeated viral respiratory infections). Apart from treatment intensity, none of the background variables were found to have any significant effect. Thus, the variables of pre-existing chronic disease and repeated viral infections were dropped in a stepwise fashion so that in the final model we only included the variables of age, sex, and treatment intensity. Adjusted odds ratios (aOR) with 95% confidence intervals (CI) were determined. All statistical analyses were carried out using IBM SPSS Statistics for Windows, version 27 (IBM Corp., Armonk, NY).

Ethics

All the data analyzed were collected as part of routine diagnostics and treatment. Data analysis was carried out in connection with a quality evaluation procedure and did therefore not require ethical approval. Data were handled in a strictly anonymous manner.

Results

Study population characteristics

The mean age of all patients at the time of diagnosis was 6.43 years (0 years to 16 years). Distribution of gender was comparable between groups, as 41 (51%) of the patients were girls and 39 (49%) boys. In our data 52 (65%) of the children were at daycare or at school at the time of diagnosis, 13 children (16%) were still in home care and the rest 15 (19%) we couldn't tell from the patient data. (Table 1).

In our high intensity group 42% of the patients underwent radiation therapy in addition to cytostatic treatment, while in the low intensity group radiation therapy was not delivered. The stratification of ITR-3 (Kazak A et al. 2006) explains this finding, as addition of treatment modalities increases the risk grouping by definition. There were no statistically significant differences relating to the occurrence of multiple respiratory infections or serious infections prior to the diagnosis of malignancy between the groups (Table 1).

Normalization of immunological parameters

In our dataset including all patients, lymphopenia was much more common in the higher treatment intensity group as 25/31 (80%) of patients were lymphopenic at completion of treatment. In the low intensity group 15/47 (32%) of patients had lymphopenia at completion of treatment. (Table 2). CD4 levels were low at completion of treatment or after 4 months in 5/48 (10%) of patients in the low intensity group and in the higher intensity group in 13/31 (42%) of patients (Table 2). There was a clear difference between the treatment intensity groups, but this finding – in combination with the analyses for lymphopenia – also showed a relatively fast normalization of results post treatment.

In the high intensity treatment group 6/29 (21%) of patients had low NK count and 6/27 (22%) had low IgG count. In the low intensity treatment group, 2/45 (4%) patients had a low NK count and 4/44 (9%) had low IgG, being less than in group 2. All in all, patients in the high intensity group had significantly lower counts in immunological parameters compared to the lower intensity group. (Table 2)

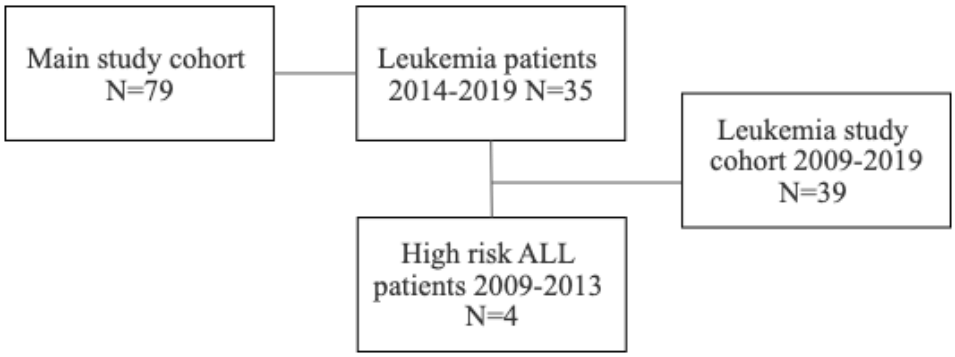
In our separate dataset including only leukemia patients, all of the AML patients (N=8) showed lymphopenia at completion of treatment. In the high risk ALL group 4/7 (57%) of patients were lymphopenic and in the standard/intermediate risk ALL only 2/24 (8%) had lymphopenia at

completion of treatment ($p<0.001$). (Table 3). Neutrophil counts had normalized by 1 month post treatment in all but 1 leukemia patient. This patient was treated according to the high risk protocol.

Adjusted binary logistic regression analysis with background variables (age, sex, treatment intensity, pre-existing chronic disease, previous repeated viral respiratory infections) did not show any significant effect for any of the variables, except for the variable of treatment intensity. When analyzing cases stratified by diagnoses, treatment for high-risk leukemia, AML, medulloblastoma, and mature B-cell lymphoma was associated with prolonged recovery times for a substantial proportion of cases. High-risk leukemia patients (N=6) had low IgG in 5/6 (83%) and lymphopenia in 4/6 (67%) of cases. AML patients (N=8) had lymphopenia in all cases and low CD4 in 4/8 (50%) cases. All medulloblastoma patients (N=5) had low CD4 and lymphopenia. Mature B-cell lymphoma patients (N=9) had lymphopenia in 7/9 (88%) cases and low IgG in 4/9 (44%) cases. (Table 7).

Figure 1

Study cohort



Note. Main study cohort: all pediatric patients starting cytostatic treatment for malignant diseases at Turku University Hospital 2014-2019.
The leukemia study cohort consisted of all pediatric leukemia patients (standard risk, intermediate risk, high risk patients) starting treatment 2014-2019 and high risk patients who started treatment 2009-2013. Stem cell transplant patients were excluded.
Please refer to the methods section for details on other exclusion criteria.

Table 1

Patient characteristics across groups.

	n/N (%)	Lower intensity treatment (group 1) n/N (%)	Higher intensity treatment (group 2) n/N (%)	P-values
Age at diagnosis in years, median (1st Quartile)	79	4.0 (2.0)	7.0 (4.0)	0.032*)
Sex, male n/N (%)	39/ 79 (49%)	26/ 48 (54%)	13/ 31 (42%)	0.28**)
Solid tumor	44/ 79 (56%)	24/48 (50%)	20/ 31 (65%)	0.21**)
Radiation therapy	13/ 79 (16%)	0/ 48	13/ 31 (42%)	NA
Pre-existing chronic disease	10/ 79 (13%)	4/ 48 (8%)	6/ 31 (19%)	0.18***)
Multiple respiratory infections previous to diagnosis	7/ 79 (9%)	2/ 48 (4%)	5/ 31 (16%)	0.11***)
Serious infections previous to diagnosis	1/ 79 (1%)	0/ 48	1/ 31 (3%)	NA

Note. Main study cohort. Does not include leukemia patients from 2009-2013.

Chronic disease does not include skin atopy or allergies.

*) Mann Whitney U test

**) Chi-square

***) Fisher's exact test

Table 2

Treatment Intensity and Lymphocyte Subset at end of the treatment and after 4 months.

	n/N (%)	Lower intensity treatment (group 1) n/N (%)	Higher intensity treatment (group 2) n/N (%)	P-values
Lymphopenia at completion of treatment	40/ 79 (51%)	15/ 47 (32%)	25/ 31 (80%)	<0.001**)
Low CD4	18/ 79 (23%)	5/ 48 (10%)	13/ 31 (42%)	0.002***)
Low NK	8/ 74 (11%)	2/ 45 (4%)	6/ 29 (21%)	0.050***)

Low CD19 *)	3/ 69 (4%)	2/ 42 (5%)	1/ 27 (4%)	1.0***)
Low IgG *)	10/ 71 (14%)	4/ 44 (9%)	6/ 27 (22%)	0.16***)

Note. Main study cohort. Does not include leukemia patients from 2009-2013.

Low CD4: CD4 count below 400 E6/l by 4months post-treatment

Low NK: NK count below 90 E6/l by 4 months post-treatment

Low CD19: CD19 count below 90 E6/l by 4 months post-treatment

Low IgG: IgG below reference range 4 months post-treatment

Lymphopenia: Lymphopenia at completion of treatment

*) patients having received Rituximab excluded

**) Chi-square

***) Fisher's exact test

Table 3

Recovery of immunological parameters for leukemia patients.

	Diagnosis			
	ALL SR/IR	ALL HR	AML	P-values
	n/N (%)	n/N (%)	n/N (%)	
Low CD4	3/ 24 (13%)	2/ 7 (29%)	4/ 8 (50%)	0.080
Low NK	0/ 23	2/ 5 (40%)	0/ 7	NA
Low CD19	2/ 23 (9%)	1/ 5 (20%)	0/ 8	0.46*)
Low IgG	2/ 24 (8%)	6/ 7 (86%)	1/ 8 (13%)	<0.001
Lymphopenia	2/ 24 (8%)	4/ 7 (57%)	8/ 8 (100%)	<0.001

Note. Only leukemia patients analyzed in this data.

Low CD4: CD4 count below 400 E6/l by 4months post-treatment

Low NK: NK count below 90 E6/l by 4 months post-treatment

Low CD19: CD19 count below 90 E6/l by 4 months post-treatment

Low IgG: IgG below reference range 4 months post-treatment

Lymphopenia: Lymphopenia at completion of treatment

P: Fisher's exact test

NA: not applicable

*) P-value without AML patients

Table 4

Adjusted logistic regression: Risk of prolonged time to CD4 count recovery

Variable	aOR	95% CI	P-values
Sex	2.83	0.83-9.70	0.10
Age group (pre-school vs school)	2.04	0.62-6.72	0.24
Treatment intensity group	6.64	1.88-23.45	0.003

Note. Main study cohort.

Discussion

Our data showed that most children have normal immunological parameters already by 4 months post cancer treatment. Immunological parameters were normal in almost all patients within the lower intensity group at completion of treatment or after 4 months, as only 10% of patients had low CD4 counts, 4% of patients had low NK counts and 9% of patients had low IgG. Within the high treatment intensity group there were more patients who showed prolongation of abnormal counts after 4 months of completion of treatment than in the low intensity group. Within the high treatment intensity group 42% of patients had low CD4 levels, 21% of patients had low NK count and 22% had low IgG count by 4 months post-treatment.

Childhood cancer is a rare disease and nowadays at least 70% of cases can be cured (G.T. Kovacs et al. 2008). Most common cancers in children are leukemias, lymphomas and brain tumors. Cancer treatments may last several years, involve a long period of intensive cytostatic treatment and in some cases radiotherapy. The immune system of children is not as developed as in adults, which means that some children might not have protection towards some of the most common viruses. (Toivonen et al. 2016)

Gabor T. Kovacs et al. could not detect any severe lymphopenia in chemotherapy treated children at 1 year after treatment. In our study 40 out of 79 (51%) patients showed lymphopenia at completion of treatment, but it seems that lymphocyte levels normalize quite soon after completing chemotherapy. Gabor T. Kovacs et al. found a significant ($p<0.001$) difference between leukemia and solid tumor patients in normalization of IgG levels. We did not categorize our patients into leukemia and solid tumor groups, but into high intensity and lower intensity treatment groups and found significant differences in normalization of immunological parameters between these two groups. In Gabor T. Kovacs et al. study, leukemic patients were one group and as we can see in our

data there is a difference in immunological reconstitution after treatment between AML, high risk ALL and standard/intermediate risk ALL patients. Low IgG levels were detected in 6/7 (86%) of high risk ALL patients but only in 2/24 (8%) of standard/intermediate risk ALL and 1/8 (13%) AML patients.

Our results show a rapid recovery of immunological parameters in the majority of patients. Childhood cancer treatment protocols have advanced to be less toxic and, according to our study, also cause less immunosuppression. This means that post-treatment transmission-based precautions are not needed in most cases. Nevertheless, within the high treatment intensity group prolonged recovery of immunological parameters was seen especially for high-risk leukemia, AML, medulloblastoma, and mature B-cell lymphoma. These patients need special attention and follow-up after completion of childhood cancer treatment.

References:

Ek, T., Mellander, L., Andersson, B., Abrahamsson, J., Immune reconstitution after childhood acute lymphoblastic leukemia is most severely affected in the high risk group. Pediatr Blood Cancer 2005;44(5):461-468.

Gadó, J., Schlick, B., Bárány, O., Németh, J., Múller, J., Csoka, M., Constantin, T., Vonnák, E., Böckskei, R., Hauser, P., Kovácks, G. T. The function of the immune system after the treatment of pediatric malignant diseases. Orv Hetil. 2006;147(36):1731-1738.

Kazak, A.E., Hocking, M.C., Ittenbach, R.F., Meadows, A.T., Hobbie, W., DeRosa B.W., Leahey A., Kersun, L., Reilly, A. A revision of the intensity of treatment rating scale: classifying the intensity of pediatric cancer treatment. Pediatrc Blood Cancer 2012;59(1):96-99.

Kovacs, G.T., Barany, O., Schlick, B., Csoka, M., Gado, J., Ponyi, A., Müller, J., Nemeth, J., Hauser, P., Erdelyi, D.J. Late Immune Recovery in Children Treated for Malignant Diseases. Pathol. Oncol. Res. 2008;14(4):391–397.

Kosmidis, S., Baka, M., Bouhoutsou, D., Doganis, D., Kallergi, C., Douladiris, N., Pourtsidis, A., Varvoutsi, M., Saxoni-Papageorgiou, F., Vasilatou-Kodmidis, H. Longitudinal assessment of immunological status and rate of immune recovery following treatment in children with ALL. Pediatrc Blood Cancer 2008;50(3):528-532.

Toivonen, L., Karppinen, S., Schuez-Havupalo, L., Teros-Jaakkola, T., Vuononvirta, J., Mertsola, J., … Peltola, V. Burden of Recurrent Respiratory Tract Infections in Children. The Pediatric Infectious Disease Journal 2016;35(12):e362–e369.

Table 5
Diagnoses, applied protocols, features of patients with prolonged recovery of CD4 lymphocyte subsets.

Disease	Applied protocols	Patients with prolonged CD4 recovery (n/N)	Additional features of patients with prolonged CD4 recovery
ALL	NOPHO ALL 2008	5/ 20	3 IR patients age 13, 15 and 16 (of which 1 Down sdr patient); 2 HR patients age 14 and 15.
	Interfant 06	0/ 1	
Philadelpia ALL	EsPhALL2010/ NOPHO version	0/ 1	
Acute myeloid leukemia	NOPHO-DBH-AML2012	4/ 7	
	Down AML 2007	0/ 1	
Hodgkin's lymphoma	EuroNet-PHL-C1 2006	1/ 5	TG2 and radiotherapy 31gy
Soft tissue sarcomas: Rhabdomyosarcoma	CWS guidance 2014	0/ 2	
	CWS 2006	0/ 1	
Soft tissue sarcomas: Synovial sarcoma	CWS guidance 2014	1/ 1	IRS2A; radiotherapy 31 gy
Malignant peripheral nerve sheath tumor	CWS guidance 2014	0/ 1	
Retinoblastoma	VEC-treatment a)	0/ 2	
Neuroblastoma	LINES protocol SIOPEN 2010	0/ 2	
Wilms tumor	Umbrella protocol SIOP-RTSG 2016	0/ 5	
Lymphoblastic lymphoma	Euro-LBL2014	2/0	
Mature B-cell lymphoma	B-NHL 2013 (NHL-BFM and NOPHO study groups)	8/2	R2
	BFM-NHL 2004	1/1	R4
	FAB/LMB96	1/1	group B

Anaplastic large cell lymphoma	ALCL-99	2/0	
Germ cell tumor	GCT-III 2005	1/0	
Medulloblastoma	SIOP-PNET5MB	5/5	CSI
Ependymoma	SIOP ependymoma	1/1	CSI
Low-grade glioma	LGG 2004	2/0	
High grade glioma	Radiotherapy; Temozolamide/ CCNU b) subsequently PCV-treatment. c)	1/0	
Hepatoblastoma	SIOPEL-6 2008	1/0	

Note.

a) VEC treatment: Vincristine, Etoposide, Carboplatin

b) Jakacki R et al.: Phase 2 study of concurrent radiotherapy and temozolomide followed by temozolomide and lomustine in the treatment of children with high-grade glioma: a report of the Children's Oncology Group ACNS0423 study. Neuro-Oncology. 2016.

c) procarbazine, CCNU, vincristine

Table 6

Reconstitution of immunological parameters by diagnoses.

Diagnosis	N (%)	Low CD4	Low NK	Low IgG	Low CD19	Lymphopenia
ALL SR/ IR	25	3 (12%)	0	3 (12%)	3 (12%)	6 (24%)
ALL HR	6	2 (33%)	2 (33%)	5 (83%)	0	4 (67%)
AML	8	4 (50%)	0	1 (13%)	0	8 (100%)
Lymphoblastic lymphoma	2	0	0	0	0	1 (50%)
ALCL	3		1 (33%)	0	0	2 (67%)
Mature B-cell lymphoma	9	3 (33%)	3 (33%)	4 (44%)	2 (22%)	7 (88%)
Hodgkin lymphoma	5	1 (20%)	0	2 (40%)	0	4 (80%)
Wilms tumor	5	0	0	1 (20%)	0	1 (20%)
Neuroblastoma	2	0	1 (50%)	0	0	1 (50%)
Soft-tissue sarcoma: Rhabdomyosarcoma	3	0	0	0	0	1 (33%)
Soft-tissue sarcoma: Non-rhabdomyosarcoma	1	1 (100%)	0	0	0	1 (100%)
Malignant peripheral nerve sheath tumor	1	0	0	0	0	1 (100%)
Germ cell tumor	1	0	0	0	0	0
Hepatoblastoma	1	0	0	0	0	0
Retinoblastoma	2	0	0	0	0	0
Medulloblastoma	5	5 (100%)	2 (40%)	2 (40%)	0	5 (100%)
Ependymoma	1	1 (100%)	0	0	0	1 (100%)
Low-grade glioma	2	0	0	0	0	0
High-grade glioma	1	0	0	0	0	1 (100%)

Note. Main study cohort.

The originality of this thesis has been checked in accordance with the University of Turku quality assurance system using the Turnitin OriginalityCheck service.