Plasma Cytokine Profiles at Diagnosis in Pediatric Patients With Non-Hodgkin Lymphoma

Karin Mellgren, MD, PhD,† Chris Juul Hedegaard, PhD,‡ Kjeld Schmiegelow, MD, PhD,§‖ and Klaus Müller, MD, PhD‡‖*

Summary: Non-Hodgkin lymphoma (NHL) has been associated with elevated levels of inflammatory and immune-regulating cytokines, and polymorphisms in the genes encoding interleukin (IL)-10 and tumor necrosis factor (TNF)-α have been associated with increased incidence of certain subtypes of NHL. The aim of the present study was to screen for a broader spectrum of growth factors and inflammatory mediators and to compare the profiles in different subtypes of NHL in pediatric patients. Serum samples were collected at diagnosis from 31 pediatric patients diagnosed with NHL admitted at Rigshospitalet, Copenhagen, between 1995 and 2008. Cytokines and growth factors were measured in serum using the Luminex platform by application of a 30-plex kit. Levels of IL-6, IL-2R, IL-10, TNF-RI, and macrophage inflammatory protein-1α were significantly higher in patients with anaplastic large-cell lymphoma compared with patients diagnosed with B-cell lymphomas and lymphoblastic lymphomas. High levels of IL-4, IL-13, TNF-RI, and epidermal growth factor were associated with a poorer general condition at diagnosis. The present study suggests that NHL subgrouping and the general condition of pediatric patients at diagnosis are associated with plasma levels of growth factors and inflammatory mediators at presentation.

Key Words: Non-Hodgkin lymphoma, cytokine profile, biological markers, pediatric patients

(*J Pediatr Hematol Oncol* 2012;34:271–275)

Immune mechanisms play a critical role in the pathogenesis of lymphomas.[1] Longitudinal studies in subjects infected by human immunodeficiency virus, as well as case-control studies in healthy populations that develop B-cell lymphomas, have provided important new insights into the role of chronic B-cell stimulation in the development of lymphomas.[2–4] Various cytokines have been found to be elevated in serum samples preceding the diagnosis of lymphoma by many years,[5–8] suggesting a genetic impact on cytokine production with a potential impact on the individual risk of developing lymphoma. Indeed, associations of polymorphisms of genes encoding tumor necrosis factor-α (TNF-α), lymphotoxin-α, and interleukin (IL)-10 with diffuse large B-cell lymphoma have been found in adults,[9] and they are thought to play an essential role in pathogenesis of this disease.[10] However, very little is known about the specific contribution of these immune mediators in the group of aggressive non-Hodgkin lymphomas (NHL) in children.

Various studies suggest that the biological and clinical characteristics of malignant lymphoma are influenced by the interaction between tumor cells and their nonmalignant microenvironment—including connective tissue cells, specialized organ cells, and cells belonging to the immune system.[11,12] Thus, mediators released by the surrounding healthy tissues may play a role by regulating the proliferation of the malignant cells, and growth factors and inflammatory mediators released from those may influence the function and activation of healthy tissue cells.

The composition of the tumor microenvironment most likely varies between different classes of malignant lymphomas, and a wide range of mediators may potentially play a role in these interactions. Despite this, cytokine profiling in NHL has previously been restricted to a rather narrow spectrum of mainly leukocyte-derived cytokines such as IL-1, IL-6, and TNF.

In the present single-center study we have screened plasma levels of a broad spectrum of both leukocyte-derived and tissue-derived cytokines and growth factors at diagnosis of pediatric NHL to relate their pattern to the immunophenotype and to the general condition and symptoms of the patient at diagnosis. The number of cases in the present study is too small to enable correlation of cytokine levels with the risk of relapse.

PATIENTS AND METHODS

Patients

Serum samples were collected at diagnosis of NHL in 31 patients (25 boys and 6 girls) treated and followed at Copenhagen University Hospital Rigshospitalet during the period 1987 to 2008, after approval by the local ethics committee. Median (range) age at diagnosis was 9.3 years (range, 1.3 to 15.9 y). Sixteen patients were diagnosed with B-NHL and treated according to Berlin-Frankfurt-Muenster protocols.[13,14] Nine patients were diagnosed with lymphoblastic lymphoma, 8 patients with T-cell phenotype, and 1 patient with pre–B-cell phenotype were treated according to a Nordic Society of Pediatric Hematology and Oncology protocol [modified acute lymphoblastic leukemia (ALL) protocol].[15] Six patients with anaplastic large-cell lymphoma (ALCL) were treated according to the Berlin-Frankfurt-Muenster ALCL protocol[16] or the international ALCL-99 study protocol.[17] One of the patients suffered from a preexisting immune deficiency (X-linked lympho-

Received for publication July 27, 2011; accepted November 21, 2011.
From the *Department of Paediatrics, The Queen Silvia's Hospital for Children and Adolescents; †The Sahlgrenska Academy, University of Gothenburg, Göteborg, Sweden; ‡Institute for Inflammation Research, Department of Rheumatology; §Faculty of Health Sciences, University of Copenhagen; and ‖Paediatric Clinic II, Rigshospitalet, Copenhagen, Denmark.
Supported by grants from the Swedish Childhood Cancer Foundation. The authors declare no conflict of interest.
Reprints: Karin Mellgren, MD, Department of Paediatrics, The Queen Silvia's Hospital for Children and Adolescents, S-41685 Göteborg, Sweden (e-mail: karin.mellgren@vgregion.se).

TABLE 1. Clinical Data of the 31 Patients Included in This Study

Study No.	Age (y)	Sex	Stage	Diagnosis	Event	EFS	Current Status	Condition at Diagnosis
51997607	10.9	Female	Stage IV	ALCL	Alive in CR-1	152.7	Alive in CR-1	Normal
51998606	8.9	Male	Stage III	ALCL	Alive in CR-1	138.0	Alive in CR-1	Bedridden
51998609	12.8	Male	Stage III	ALCL	Alive in CR-1	122.7	Alive in $\geq$CR-2	Bedridden
51998610	12.0	Female	Stage IV	ALCL	Alive in CR-1	146.0	Alive in CR-1	Reduced activity
52005611	4.5	Male	Stage III	ALCL	Relapse	9.4	Dead—after relapse(s)	Reduced activity
52007606	1.3	Male	Stage III	ALCL	Relapse	9.4	Dead—after relapse(s)	Normal
51995607	13.9	Female	Stage III	B-NHL	Relapse	6.0	Dead—after relapse(s)	Reduced activity
51995610	5.3	Male	Stage III	B-NHL	Relapse/SMN	181.0	Alive in $\geq$CR-2	Reduced activity
51996605	5.9	Female	Stage I	B-NHL	Alive in CR-1	165.8	Alive in CR-1	Bedridden
51996606	7.1	Male	Stage I	B-NHL	Alive in CR-1	163.5	Alive in CR-1	Few symptoms
51996608	9.2	Male	Stage IV	B-NHL	Alive in CR-1	169.1	Alive in CR-1	Normal
51999601	4.3	Male	Stage III	B-NHL	Alive in CR-1	135.4	Alive in CR-1	Few symptoms
51999604	10.1	Male	Stage I	B-NHL	Alive in CR-1	126.8	Alive in CR-1	Normal
52002601	6.7	Male	Stage III	B-NHL	Alive in CR-1	95.7	Alive in CR-1	Reduced activity
52002606	4.0	Male	Stage I	B-NHL	Alive in CR-1	87.8	Alive in CR-1	Normal
52004613	8.5	Male	Stage III	B-NHL	Alive in CR-1	74.1	Alive in CR-1	Few symptoms
52004617	13.5	Female	Stage III	B-NHL	Alive in CR-1	65.2	Alive in CR-1	Reduced activity
52005613	15.9	Male	Stage III	B-NHL	Induction failure	0.3	Early death	Reduced activity
52005614	5.9	Male	Stage III	B-NHL	Alive in CR-1	62.3	Alive in CR-1	Reduced activity
52006601	7.5	Male	Stage III	B-NHL	Alive in CR-1	41.9	Alive in CR-1	Normal
52007610	15.5	Male	Stage II	B-NHL	Alive in CR-1	35.7	Alive in CR-1	Few symptoms
52008608	4.0	Male	Stage III	B-NHL	Alive in CR-1	29.5	Alive in CR-1	Few symptoms
51998608	9.3	Male	Stage III	Pre-B LBL	Alive in CR-1	141.0	Alive in CR-1	Reduced activity
51996603	8.8	Male	Stage III	T-cell LBL	Alive in CR-1	168.4	Alive in CR-1	Bedridden
51998611	9.5	Male	Stage IV	T-cell LBL	Alive in CR-1	143.7	Alive in CR-1	Few symptoms
52000601	10.7	Male	Stage III	T-cell LBL	Alive in CR-1	121.8	Alive in CR-1	Normal
52004611	11.1	Female	Stage III	T-cell LBL	Alive in CR-1	75.2	Alive in CR-1	Reduced activity
52004616	13.6	Male	Stage III	T-cell LBL	Alive in CR-1	66.8	Alive in CR-1	Reduced activity
52005610	14.0	Male	Stage III	T-cell LBL	Relapse	6.0	Dead—after relapse(s)	Few symptoms
52005612	13.0	Male	Stage III	T-cell LBL	Alive in CR-1	59.4	Alive in CR-1	Reduced activity
52006602	5.4	Male	Stage III	T-cell LBL	Alive in CR-1	43.0	Alive in CR-1	Normal

Six patients were diagnosed with ALCL, 16 with B-NHL, and 9 with lymphoblastic lymphoma. Two of the 6 patients with ALCL had a stage IV disease.
ALCL indicates anaplastic large-cell lymphoma; CR, complete remission; EFS, event-free survival; LBL, lymphoblastic lymphoma; NHL, non-Hodgkin lymphoma; SMN, second malignant neoplasm.

proliferative syndrome). Clinical data are given in Table 1. General condition at diagnosis was evaluated according to a 5-scale, standardized evaluation by the responsible physician, and the presence or absence of B-symptoms was recorded according to international criteria.

Methods

Sera were collected from patients at diagnosis and stored at -80°C until analysis. Collected sera were assessed for the following mediators: IL-1β, IL-1 receptor antagonist (IL-1ra), IL-2, soluble IL-2 receptor (sIL-2R), IL-4, IL-5, IL-6, IL-7, IL-8, IL-10, IL-12 (p40/p70), IL-13, IL-15, IL-17, TNF-α, TNF-RI, TNF-RII, interferon (IFN)-α, IFN-γ, macrophage inflammatory protein (MIP)-1α/chemokine ligand 3 (CCL3), MIP-1β/CCL4, IFN-γ-induced protein 10 kDa [IP-10/C-X-C motif chemokine 10 (CXCL10)], monokine induced by IFN-γ (MIG/CXCL9), Eotaxin/CCL11, Regulated on Activation, Normal T Expressed and Secreted (RANTES/CCL5), monocyte chemotactic protein-1/CCL2, vascular endothelial growth factor, granulocyte-macrophage colony-stimulating factor (GM-CSF), granulocyte colony-stimulating factor (G-CSF), epidermal growth factor, fibroblast growth factors basic, and hepatocyte growth factor (HGF), using a 30-plex kit from Invitrogen (Taastrup, Denmark). Samples were applied to a Luminex 100 IS (Luminex Corp., Austin, TX) and analyzed using StarStation version 2.0 software (Applied Cytometry Systems, Sheffield, UK).

Statistical Methods

Patient characteristics and serum levels of immune mediators are reported as median values and range. Event-free survival (EFS) is defined as the time from diagnosis to relapse of malignant disease, secondary malignancy, or death from any cause. Overall survival and EFS are estimated using the Kaplan-Meier method and given as percent ± SE.

To achieve an unbiased overview of the data, including immune mediators, disease characteristics, and outcome parameters, a principal component analysis (PCA) including all variables was carried out using the SIMCA-*P*+ software, version 18.0 (Umetrics, Umeå, Sweden). Variables exhibiting significant influence on the PCA model were then subjected to univariate and correlation analyses. Correlations between cytokine levels and immunophenotype, events, general condition, hemoglobin (Hb) level, and lactate dehydrogenase (LDH) at diagnosis were compared using Mann-Whitney *U* test or Kruskal-Wallis test as appropriate. Differences in proportions were assessed with the χ^2 test. Correlations between Hb and LDH levels at diagnosis and concentration of the different immune markers were calculated using the Spearman ρ. Calculations were performed using the SPSS/PC+18.0 statistical program (SPSS Inc., Chicago, IL).

TABLE 2. Median Value and Range of Plasma Levels of Immune Mediators in the Different Subgroups of NHL

Subtype	IL-6 (pg/mL)	IL-2R (pg/mL)	MIP-1α (pg/mL)	IL-10 (pg/mL)	TNF-RI (pg/mL)
B-NHL	0 (0-64)	661 (279-2558)	37 (0-59)	8 (0-27)	1392 (180-1894)
ALCL	49 (11-135)	7509 (778-21,636)	113 (47-145)	27 (13-219)	2464 (441-6307)
LBL	0 (0-4300)	578 (330-4704)	29 (0-2998)	4 (0-8)	1031 (308-1317)
P	< 0.001	< 0.001	< 0.001	< 0.001	0.011

Indeed, median values of serum levels of IL-6, IL-2R, MIP-1α, IL-10, and TNF-RI were significantly higher in the 6 patients diagnosed with ALCL as compared with patients with lymphoblastic lymphoma or B-NHL.

ALCL indicates anaplastic large-cell lymphoma; B-NHL, B-cell non-Hodgkin lymphoma; IL, interleukin; LBL, lymphoblastic lymphoma; MIP, macrophage inflammatory protein; TNF-RI, tumor necrosis factor receptor I.

RESULTS

Patients

Median follow-up time for the patients was 87.8 months (range, 0.3 to 268.7 mo). Twenty-five patients were alive in CR-1 at the end of follow-up, and 1 patient was alive in CR-2. Four patients relapsed within 10 months from diagnosis, and 1 patient suffered a very late relapse/second cancer (second malignant neoplasm) 181 months from diagnosis. This patient, a boy from a family with X-linked lymphoproliferative disease, suffered various relapses.[18] All 4 patients with relapse and 1 patient with induction failure died. The patient who suffered a second malignant neoplasm was successfully treated and is still alive.At 10 years, overall survival was at 84% ± 7% and EFS was 84% ± 7%. Four patients (13%) were severely sick and bedridden at diagnosis, 12 patients (39.5%) had reduced activity, 7 patients (22.5%) had few symptoms, and 8 patients (26%) had normal activity at diagnosis.

PCA analysis

According to the PCA analysis, the tested variables differed according to the immunophenotype of the lymphoma, and ALCL cytokine profile was significantly different from that of the other NHL subgroups. The levels of some immune mediators were significantly associated with lymphoma subtype, namely IL-2R, CCL11, IL-10, TNF-RI, and IL-6. In addition, fibroblast growth factor basic, CXCL9, and CCL3 appeared to be of importance for this difference. These observations were further tested by univariate and multivariate statistical methods as described below.

Levels of Immune Mediators

At diagnosis, several mediators were detectable above the sensitivity of the assay. These mediators represented T-cell activating cytokines and soluble receptors, including both T helper 1 (Th1)-associated mediators {IFN-γ [84 pg/mL (range, 29 to 187 pg/mL)], sIL-2R [744 pg/mL (range, 279 to 21,636 pg/mL)]} and Th2-associated mediators {IL-4 [83 pg/mL (range, 0 to 132 pg/mL)], IL-5 [21 pg/mL (range, 0 to 693 pg/mL)], IL-10 [7 pg/mL (range, 0 to 219 pg/mL)], CCL11 [92 pg/mL (range, 26 to 569 pg/mL)]}. Moreover, a number of growth factors were found to be elevated at diagnosis. These included GM-CSF [93 pg/mL (range, 0 to 2589 pg/mL)], EGF [241 pg/mL (range, 0 to 576 pg/mL)], and HGF [285 pg/mL (range, 99 to 669 pg/mL)].

Correlation Between Cytokine Levels and Immunophenotype

The cytokine profiles of the subgroups of lymphoma patients showed significant differences for a number of immunoregulatory mediators, including IL-6, IL-2R, CCL3, IL-10, and sTNF-RI. Median levels of these mediators were significantly higher in ALCL as compared with the other subgroups (Table 2). All the 16 patients who were bedridden or had reduced activity at diagnosis had significantly higher levels of IL-4, IL-13, EGF, and TNF-RI as compared with those who had few or no symptoms (Table 3).

Cytokine Levels With Respect to LDH Levels and Hb Levels at Diagnosis

Median Hb level at diagnosis was at 117 g/L (range, 56 to 149 g/L). Eleven patients had Hb levels > 125 g/L at diagnosis, 13 patients had Hb levels of 100 to 125 g/L, and 6 patients had Hb < 100 g/L.

LDH was elevated to above 2 times the upper normal limit at diagnosis in 7 patients and was within normal limits in the remaining 24 patients. The LDH level at diagnosis correlated with levels of CXCL9 ($r_S = 0.42$; $P = 0.02$), TNF-RII ($r_S = 0.38$; $P = 0.034$), and CCL11 ($r_S = 0.38$; $P = 0.04$). Hb level at diagnosis was associated with TNF-RII ($r_S = -0.49$; $P = 0.005$), HGF ($r_S = -0.41$; $P = 0.025$), and G-CSF ($r_S = -0.37$; $P = 0.04$).

The incidence of events was not significantly associated with Hb levels. Likewise, none of the measured mediators was significantly related to the occurrence of events.

TABLE 3. Median Value and Range of Plasma Levels of Immune Mediators in Patients With Few or No Symptoms of Their Lymphoma at Diagnosis as Compared With the Group of Patients Who Presented With Reduced Activity or Who Were Severely Sick at Diagnosis

Cytokine	Few or No Symptoms	Reduced Activity or Bedridden	*P*
IL-13	52 pg/mL (44-1736)	47 pg/mL (14-57)	0.024
IL-4	98 pg/mL (0-121)	59 pg/mL (0-132)	0.024
EGF	159 pg/mL (34-470)	159 pg/mL (34-470)	0.013
sTNF-RI	1572 pg/mL (705-6307)	1089 pg/mL (180-2182)	0.008

Four patients were severely sick and bedridden at diagnosis, 12 patients had reduced activity, 7 patients had few symptoms, and 8 patients had normal activity at diagnosis. Levels of sTNF-RI were significantly higher in patients with few or no symptoms at diagnosis as compared with those who were severely sick.

EGF indicates epidermal growth factor; IL, interleukins; sTNF-RI, soluble tumor necrosis factor receptor I.

DISCUSSION

The data of the present study do not suggest that enhanced inflammation is a general phenomenon in children with NHL. Thus, median levels of the key inflammatory markers IL-6, TNF-α, sTNF-RI, and sTNF-RII were similar to those previously reported in healthy children.[19] Rather, the present study suggests that NHL patients may be differentiated on the basis of cytokine profiling, into subgroups reflecting NHL subtypes and clinical presentation. These profiles include cytokines, indicative of a combined Th1 and Th2 pattern of T-cell activation. It is not clear from the present study, however, whether these findings are correlated to a genetic predisposition of the individual or to other factors such as infection.

This study provides preliminary evidence of an association between cytokine/growth factor profiles at diagnosis and NHL immunophenotypes in pediatric patients. In particular, patients with ALCL appeared to present with a distinct mediator profile characterized by high levels of IL-6, IL-2R, MIP-1α, IL-10, and sTNF-RI at diagnosis as compared with children with other types of lymphoma. The present study further extends our knowledge by the finding of a mediator profile that indicates the activation of non-hematopoietically derived tissue cells at diagnosis. Growth factors such as GM-CSF, EGF, and HGF were elevated at diagnosis in most of the patients with a pattern suggestive of differences between the subgroups of NHL. Tumor environment is believed to play a key role in the growth and survival of hematologic malignancies. In lymphomas, cancer cells are surrounded by numerous cell types, including stromal fibroblasts, endothelial cells, and cells of the immune system. Interactions between tumor cells and these surrounding cells are thought to be critical in the initiation and progression of the oncologic process, and individual variability in these interactions may play important roles in the clinical presentation and disease progression. In particular, Hodgkin lymphoma is known to be a malignant disease with a disturbed cytokine production[20] in which cytokines that process proliferative (IL-13 and IL-17), immunosuppressive (IL-10 and TGF-β), and regulatory functions (IL-5, IL-10, CCL5, CXCL9) may be involved in the cross-talk between the tumor cells and their microenvironment.[21]

Less is known about the potential impact of tissue reactions on tumor cell growth in NHL. In the present study, we found evidence of high levels of EGF and HGF at diagnosis. EGF has been shown to be expressed by lymphoma cells and mediate upregulation of early growth response protein 1 in stroma cells surrounding the lymphoma.[22] HGF is produced by stroma cells and stimulates epithelial cell proliferation, motility, morphogenesis, and angiogenesis in various organs through tyrosine phosphorylation of its receptor c-Met. It is involved in tissue regeneration and may be important for tissue protection during inflammatory disease through direct actions on macrophages, dendritic cells, and lymphocytes.[23,24] Emerging studies indicate a key role of HGF during tumor metastasis, and overproduction of HGF may accelerate tumor malignancy.[25] The present finding of elevated HGF in patients with B-NHL and ALCL may be explained by tissue responses to the tumor. Indeed, ALCL is known to be a highly immunogenic tumor, expressing the oncoprotein nucleophosmin-anaplastic lymphoma kinase (NPM-ALK), a product from the chromosomal translocation (t2;5 p23q35) that fuses NPM with ALK. NPM-ALK can associate with the cytoplasmic tail of CD30,[26] and may thereby be involved in tumor genesis possibly abrogating CD30-mediated nuclear factor-κ B activation.[27] The interaction between stroma cells and malignant cells in ALCL might be similar to what has been observed in Hodgkin lymphoma.

A poor general condition at diagnosis appears to be associated with worse outcomes in lymphoma patients (unpublished data from Nordic Society of Pediatric Hematology and Oncology register). In our study, the 16 patients who were bedridden or had reduced activity at diagnosis had significantly elevated levels of IL-13, IL-4, and EGF as compared with patients with few or no symptoms at diagnosis. Inflammatory cytokines, including TNF, are known to induce symptoms of general malaise including fever, anorexia, and sleepiness through systemically mediated effects on the central nervous system. In line with this, genetic polymorphisms associated with high TNF and lymphotoxin-α production have been found to influence prognosis in B-NHL and B-ALL of pediatric patients and to be associated with more events (relapse, progression, tumor-related mortality).[28] In a prospective study in adult patients, a possible correlation between plasma levels of IL-2, intercellular adhesion molecule, IFN-γ, and TNF-α and the risk for developing NHL was observed suggesting that a down regulation of Th1 cytokines might be associated with the risk of developing NHL.[8] In addition, Hb levels at diagnosis have been proposed as a prognostic marker in childhood ALL.[29] In previous studies, high levels of TNF-α and IFN-γ have been associated with low Hb levels in children with solid tumors,[30] and in vitro experiments have shown that both TNF-α and IFN-γ inhibit proliferation of erythroid progenitor cells.[31] In the present study, we did not find any significant association between Hb and TNF-α, which was generally found at levels close to the detection limit of the assay. Indeed, TNF-α is a cytokine difficult to measure reliably because of multimer formation and rapid degeneration, with the risk of overestimation or underestimation of TNF levels in biological fluids. The present study did, however, indicate an association between Hb levels and TNF-RII, which is considered an indicator for increased TNF activity.

In conclusion, this small pilot study shows evidence that plasma cytokine and growth factor profiles differ according to the immunophenotype in patients at diagnosis of NHL. Interindividual differences in inflammatory response genes may play an important role in lymphoma-genesis, and larger population-based studies are necessary to investigate their role in pediatric NHL.

REFERENCES

1. Verdame E, Martinez-Maza O. Assessment of prediagnosis biomarkers of immune activation and inflammation: insight on the etiology of lymphoma. *J Proteome Res*. 2011;10:113–119.
2. Manovani A, Allavena P, Sica A, et al. Cancer-related inflammation. *Nature*. 2008;454:436–444.
3. Pasqualucci L, Neumeister P, Goossens T, et al. Hypermutation of multiple proto-oncogenes in B-cell diffuse large-cell lymphomas. *Nature*. 2001;412:341–346.
4. Epeldegui M, Widney DP, Martinez-Maza O. Pathogenesis of AIDS lymphoma: role of oncogenic viruses and B-cell activation-associated molecular lesions. *Curr Opin Oncol*. 2006;18:444–448.
5. Purdue MP, Lan Q, Martinez-Maza O, et al. A prospective study of serum soluble CD30 concentration and risk of non-Hodgkin lymphoma. *Blood*. 2009;114:2730–2732.

6. Gu Y, Shore RE, Arslan AA, et al. Circulating cytokines and risk of B-cell non-Hodgkin lymphoma: a prospective study. *Cancer Causes Control*. 2010;21:1323–1333.
7. Skibola CF, Bracci PM, Nieters A, et al. Tumor necrosis factor (TNF) and lymphotoxin-α (LTA) polymorphisms and risk of non-Hodgkin lymphoma in the InterLymph Consortium. *Am J Epidemiol*. 2010;171:267–276.
8. Saberi Hosnijeh F, Krop E, Scoccianti C, et al. Plasma cytokines and future risk of non-Hodgkin lymphoma (NHL): a case-control study nested in the Italian European prospective investigation into cancer and nutrition. *Cancer Epidemiol Biomarkers Prev*. 2010;19:1577–1584.
9. Stasi R, Zinzani L, Galieni P, et al. Clinical implications of cytokine and soluble receptor measurements in patients with newly-diagnosed aggressive non-Hodgkin's lymphoma. *Eur J Haematol*. 1995;54:9–17.
10. Rothman N, Skibola CF, Wang SS, et al. Genetic variation in TNF and IL10 and risk of non-Hodgkin lymphoma: a report from the InterLymph Consortium. *Lancet Oncol*. 2006;7:27–38.
11. De Jong D, Enblad G. Inflammatory cells and immune microenvironment in malignant lymphoma. *J Intern Med*. 2008;264:528–536.
12. Herreros B, Sanchez-Aguilera A, Piris MA. Lymphoma microenvironment: culprit or innocent? *Leukemia*. 2008;22:49–58.
13. Reiter A, Schrappe M, Yakisan E, et al. NHL-BFM 90 therapy study in treatment of malignant non-Hodgkin's lymphoma in children and adolescents. Part 3: an intermediate term analysis of the B-NHL/B-ALL. *Klin Padiatr*. 1994;206:242–252. (In German).
14. Woessmann W, Seidemann K, Mann G, et al. BFM Group. The impact of the methotrexate administration schedule and dose in the treatment of children and adolescents with B-cell neoplasms: a report of the BFM Group Study NHL-BFM95. *Blood*. 2005;105:948–958.
15. Márky I, Björk O, Forestier E, et al. Nordic Society of Pediatric Hematology and Oncology. Intensive chemotherapy without radiotherapy gives more than 85% event-free survival for non-Hodgkin lymphoma without central nervous involvement: a 6-year population-based study from the Nordic Society of Pediatric Hematology and Oncology. *J Pediatr Hematol Oncol*. 2004;26:555–560.
16. Seidemann K, Tiemann M, Schrappe M, et al. Short-pulse B-non-Hodgkin lymphoma-type chemotherapy is efficacious treatment for pediatric anaplastic large cell lymphoma: a report of the Berlin-Frankfurt-Münster Group Trial NHL-BFM 90. *Blood*. 2001;97:3699–3706.
17. Brugières L, Le Deley MC, Rosolen A, et al. Impact of the methotrexate administration dose on the need for intrathecal treatment in children and adolescents with anaplastic large-cell lymphoma: results of a randomized trial of the EICNHL Group. *J Clin Oncol*. 2009;27:897–903.
18. Gross TG, Filipovich AH, Conley ME, et al. Cure of X-linked lymphoproliferative disease (XLP) with allogenic hematopoietic stem cell transplantation. Report from the XLP registry. *Bone Marrow Transplant*. 1996;17:741–744.
19. Bjørnhart B, Svenningsen P, Gudbrandsdottir S, et al. Plasma TNF binding capacity in patients with juvenile idiopathic arthritis. *Int Immunopharmacol*. 2005;5:67–71.
20. Gross HJ, Hermann F, Drezler HG. Hodgkin's disease: a cytokine producing tumour: a review. *Crit Rev Oncol*. 1994;5:473–538.
21. Maggio E, van den BA, Diepstra A, et al. cytokines and their receptors in Hodgkin's lymphoma cell lines and tissues. *Ann Oncol*. 2002;13(suppl 1):52–56.
22. Bouchard F, Bélanger SD, Biron-Pain K, et al. EGR-1 activation by EGF inhibits MMP-9 expression and lymphoma growth. *Blood*. 2010;116:759–766.
23. Nakamura T, Mizuno S. Review: the discovery of hepatocyte growth factor (HGF) and its significance for cell biology, life sciences and clinical medicine. *Proc Jpn Acad Ser B*. 2010;86:588–610.
24. Trusolono L, Bertotti A, Comoglio PM. Met signaling: principles and functions in development, organ regeneration and cancer. *Nat Rev*. 2010;11:834–848.
25. Matsumoto K, Nakamura T. Hepatocyte growth factor and the Met system as a mediator of tumor-stromal interactions. *Int J Cancer*. 2006;119:477–483.
26. Hübinger G, Scheffrahn I, Müller E, et al. The tyrosine kinase NPM-ALK, associated with anaplastic large cell lymphoma, binds the intracellular domain of the surface receptor CD30 but is not activated by CD30 stimulation. *Exp Hematol*. 1999;27:1796–1805.
27. Horie R, Watanabe M, Ishida T, et al. The NPM-ALK oncoprotein abrogates CD30 signaling and constitutive NF-kappa B activation in anaplastic large cell lymphoma. *Cancer Cell*. 2004;5:353–364.
28. Seidemann K, Zimmermann M, Book M, et al. Tumor necrosis factor and lymphotoxin alfa genetic polymorphisms and outcome in pediatric patients with non-Hodgkin's lymphoma: results from Berlin-Frankfurt-Münster trial NHL-BFM 95. *J Clin Oncol*. 2005;23:8414–8421.
29. Rautonen J, Simes MA. Glycosylated and total hemoglobin concentrations as prognostic factors in childhood acute lymphoblastic leukemia. *Pediatr Hematol Oncol*. 1989;6:45–50.
30. Ek T, Mellander L, Abrahamsson J. Interferon γ and tumour necrosis factor α in relation to anemia and prognosis in childhood cancer. *Acta Paediatr*. 2005;94:435–437.
31. Broxmeyer HE, Williams DE, Lu L, et al. The suppressive influences of human tumor necrosis factors on bone marrow hematopoietic progenitor cells from normal donors and patients with leukemia: synergism of tumor necrosis factor and interferon-gamma. *J Immunol*. 1986;136:4487–4495.