NCU – Summative report for 2014

Report submission date: 24 February 2015

Principal investigator: Kjeld Schmiegelow

Project title: EXPLORING AND IMPROVING THIOPURINE/METHOTREXATE MAINTENANCE THERAPY OF ACUTE LYMPHOBLASTIC LEUKEMIA

NCU grant received (€): 50.000

Project commencement and completion dates:
1 January 2013 – 31 December 2017

1. **Briefly describe the project in a language understandable to non-scientists (max. 100 words)**
   Metabolism of anticancer drugs is highly variable among patients. Therefore, standardized dosing will cause insufficient treatment in rapid metabolizers whereas patients with slow inactivation may suffer excessive side effects. In this project we optimize dosing of anticancer drugs based on the genetics of the patients and drug measurements throughout the treatment period and use this to test if a novel combination of anticancer drugs can improve the prognosis of patients with an adverse drug metabolism profiles.

2. **Summarize the major findings of the project (max. 400 words)**
   Sample collection from Danish, Nordic and Baltic depertments progresses as planned; we receive the weekly scheduled 80-100 patient samples for thiopurine/methotrexate metabolites analyses, thiopurine methyltransferase activity (TPMT) assay and genotyping. Laboratory analyses of these samples are ongoing (so far 150,000 individual metabolite analyses). In parallel, our assays for TPMT phenotyping and MTXpg have been improved to allow higher throughput, precision and robustness. Additional clinical data from Danish patients, as well as all patients on study with event (relapse or second cancer; hematological data, and drug doses) have been collected by two medical students (Maria Ebbesen and Tina Gerbek Pedersen) to be merged with toxicity and survival data.

   For the 6MP/thioguanine (6TG) combination therapy part of the study, M.D. Stine Nygaard Nielsen has been affiliated full-time with the study since September 2013. For accurate dosing of thioguanine (TG) she has obtained approval for and coordinated production with the British company Novalab for an oral suspension formulation of TG. Moreover, Stine has contributed to a recently published review

on ALL thiopurine/MTX maintenance therapy (2) and published a case report on 6MP 6TG combination therapy (3). High resolution exome-enriched single nucleotide popymorphism analyses (Illumina Omni2.5 Human) has been performed December 2014 for the first 800 Nordic/Baltic patients and awaits bioinformatic analysis with linkage to drug metabolism and cure rates. A recent Nature Genetics publication on the role of acquired NT5C2 mutations in ALL relapse and thiopurine resistance has led to a cooperation with the American authors, where the emergence of NT5C2 mutated ALL clones is backtracked and correlated with thiopurine metabolic profiles. A postdoc (Kathrine Ask) now working full time on this project has been employed at the laboratory since April 2014.

Preliminary analysis of pharmacological data has resulted in one published paper (1) on the influence of 6MP dose and TPMT status on risk of second malignancy and 4 manuscripts in preparation a) on the role of inosine triphosphate pyrophosphatase (ITPA) polymorphisms in thiopurine disposition, b) on prediction of post high-dose methotrexate hematological toxicity by thiopurine and methotrexate metabolites (Sophia Detner), c) on role of TMPT variants in early (ALL induction/consolidation) phase residual disease levels Emilie Brunner), and d) effect of TPMT variants on post HDM toxicity. A full-time biostatistician is affiliated with the study for the complex metabolite profiling. Finally, a pregraduate medicine student (Maria Pærregaard) is affiliated with the project working scientifically with MTXpg pharmacology.

3. Describe how the project has increased our knowledge of the prevention, cause and/or cure for cancer (max. 150 words)

See above 2.

4. Outline how Nordic cooperation has added value to this project (max. 100 words)

Since acute lymphoblastic leukemia is rare, research in this heterogenous disease requires access to large cohorts of patient treated according to identical guidelines in order to get sufficient statistical power for hypothesis testing. The Nordic/Baltic cooperation with common treatment protocols has not only added to, but is essential for the cohort-based research strategy in this study (and future studies). The present study is the so far largest clinical Nordic study on acute lymphoblastic leukemia involving both children and adults.

5. Publications resulting from the NCU research grant

1: Vang SI, Schmiegelow K, Frandsen T.L, Rosthøj, S, Nersting, J. Mercaptopurine metabolite levels are predictors of bone marrow toxicity following high-dose methotrexate therapy of childhood acute lymphoblastic leukaemia. Accepted for publication in 2015 in Cancer Chemotherapy and Pharmacology.

2: Levinsen M, Rosthøj S, Nygaard U, Heldrup J, Harila-Saari A, Jonsson OG, Bechensteen AG, Abrahamsson J, Lausen B, Frandsen TL, Weinshilboum RM, Schmiegelow K. Myelotoxicity after high-dose methotrexate in childhood acute leukemia is influenced by 6-mercaptopurine dosing but not by intermediate thiopurine methyltransferase activity. Cancer Chemother Pharmacol. 2015 Jan;75(1):59-66.

3: Nielsen SN, Frandsen TL, Nersting J, Hjalgrim LL, Schmiegelow K. Pharmacokinetics of 6-Thioguanine and 6-Mercaptopurine Combination Maintenance Therapy of Childhood ALL: Hypothesis and Case Report. J Pediatr Hematol Oncol. 2014 Aug 28. [Epub ahead of print] PMID: 25171455.

4: Schmiegelow K, Nielsen SN, Frandsen TL, Nersting J. Mercaptopurine/Methotrexate maintenance therapy of childhood acute lymphoblastic leukemia: clinical facts and fiction. J Pediatr Hematol Oncol. 2014 Oct;36(7):503-17.

5: Levinsen M, Rotevatn EØ, Rosthøj S, Nersting J, Abrahamsson J, Appell ML, Bergan S, Bechensteen AG, Harila-Saari A, Heyman M, Jonsson OG, Maxild JB, Niemi M, Söderhäll S, Schmiegelow K; Nordic Society of Paediatric Haematology, Oncology. Pharmacogenetically based dosing of thiopurines in childhood acute lymphoblastic leukemia: influence on cure rates and risk of second cancer. Pediatr Blood Cancer. 2014 May;61(5):797-802.

6: Lund B, Wesolowska-Andersen A, Lausen B, Borst L, Rasmussen KK, Müller K, Klungland H, Gupta R, Schmiegelow K. Host genome variations and risk of infections during induction treatment for childhood acute lymphoblastic leukaemia. Eur J Haematol. 2014 Apr;92(4):321-30.

7: Frandsen TL, Heyman M, Abrahamsson J, Vettenranta K, Åsberg A, Vaitkeviciene G, Pruunsild K, Toft N, Birgens H, Hallböök H, Quist-Paulsen P, Griškevičius L, Helt L, Hansen BV, Schmiegelow K. Complying with the European Clinical Trials directive while surviving the administrative pressure - an alternative approach to toxicity registration in a cancer trial. Eur J Cancer. 2014 Jan;50(2):251-9.

8: Schmiegelow K, Levinsen MF, Attarbaschi A, Baruchel A, Devidas M, Escherich G, Gibson B, Heydrich C, Horibe K, Ishida Y, Liang DC, Locatelli F, Michel G, Pieters R, Piette C, Pui CH, Raimondi S, Silverman L, Stanulla M, Stark B, Winick N, Valsecchi MG. Second malignant neoplasms after treatment of childhood acute lymphoblastic leukemia. J Clin Oncol. 2013 Jul 1;31(19):2469-76.

9: Rasmussen MM, Christensen RH, Gregers J, Heldrup J, Nersting J, Schmiegelow K. Can SLC19A1 80G>A polymorphisms predict risk of extremely delayed MTX excretion after high dose of methotrexate? J Pediatr Hematol Oncol. 2013 Jul;35(5):417-8.

10: Ebbesen MS, Nersting J, Jacobsen JH, Frandsen TL, Vettenranta K, Abramsson J, Wesenberg F, Schmiegelow K. Incorporation of 6-thioguanine nucleotides into DNA during maintenance therapy of childhood acute lymphoblastic leukemia-the influence of thiopurine methyltransferase genotypes. J Clin Pharmacol. 2013 Jun;53(6):670-4.