



UNIVERSITI PUTRA MALAYSIA

***CLINICAL EVALUATION AND PROTEOMIC PROFILES OF ACUTE
MYELOID LEUKEMIA PATIENTS, SAUDI ARABIA***

ALMAIMAN AMER ABDULRAHMAN

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**CLINICAL EVALUATION AND PROTEOMIC PROFILES OF ACUTE
MYELOID LEUKEMIA PATIENTS, SAUDI ARABIA**

By

ALMAIMAN AMER ABDULRAHMAN

**Thesis Submitted to the School of Graduate Studies, Universiti Putra Malaysia, in
Fulfillment of the Requirements of the Degree of Doctor of Philosophy**

December 2015

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DEDICATIONS

Dedicate to my father, my mother and my wife



Abstract of thesis presented to the Senate of Universiti Putra Malaysia in fulfillment of the requirements for the degree of Doctor of Philosophy

CLINICAL EVALUATION AND PROTEOMIC PROFILES OF ACUTE MYELOID LEUKEMIA PATIENTS, SAUDI ARABIA

By

ALMAIMAN AMER ABDULRAHMAN

December 2015

Chairman: Prof. Rasedee Abdullah, PhD

Faculty: Institute of Biosciences

Acute myeloid leukemia (AML) represents a group of clonal hematopoietic stem cell disorders which fail to differentiate and proliferate result in the accumulation of non-functional myeloblasts. In AML, response of patients to therapy is variables and there is no reliable marker to predict treatment response or prognosis of the disease. In this study, the main objective was to determine proteomic profiles of AML in patients referred to the King Faisal Specialist Hospital and Research Center, Riyadh, Saudi Arabia. The AML patients were categorized according to risk degree based on the probability of survival. Clinical data were collected from 22 patients (16 males and 6 females) with consent. A clinical data form was developed to collect information on the diagnosis, prevention, and follow-up monitoring of AML patients.

The clinical data showed that most patients (14) were cytogenetically and genetically normal and classified under the intermediate-risk category. Six patients expressed complex CCAAT/enhancer binding protein (CEBPA), FMS-related tyrosine kinase (FLT), isocitrate dehydrogenase (IDH) and Wilms tumor 1 (WT1) gene abnormalities, 2 patients showed inversion at chromosome 16 (inv.16) and 1 had t(15:17) chromosomal translocation.

The high abundance protein in the samples must be depleted before subjecting to proteomic analyses. In the current study, the Pierce Albumin and IgG Removal, Albumin Depletion, and ProteoPrep[®] Immunoaffinity Albumin and IgG Depletion Kits were compared in the optimization of the samples. The Pierce Albumin and IgG Removal Kit was found to be the best depletion method for high abundance proteins.

The protein expressions were compared among patients with different risk categories, at diagnosis and after remission. The AML samples from peripheral blood and bone marrow were analyzed by liquid chromatography tandem mass spectrometry (LC-MS/MS). At diagnosis, in both the peripheral blood and bone marrow samples, 21 proteins were significantly ($P < 0.05$) different in differential expressions among groups. The total number of proteins detected in peripheral blood was similar to those detected in bone marrow. However, there were differences in proteomic profiles between samples collected at diagnosis and remission. Most differentially expressed proteins were down-regulated in peripheral blood at remission. Among the AML cases in this study, 144 differentially expressed proteins were identified, and differences in their expression levels correlated with AML risk categories. The study showed that amphiregulin precursor (AR) (colorectum cell-derived) was expressed in low-risk AML patients only, while haptoglobin α and β and cytoplasmic tyrosyl-tRNA synthetase (EC 6.1.1.1) were highly expressed in intermediate-risk AML patients only. Lysine-arginine-ornithine-binding periplasmic protein (LAO) was expressed in intermediate and high-risk and not in the low-risk AML patients.

In conclusion, the study showed that complete clinical data are essential for the determination of correlation between proteomic profile, risk categories and prediction of response to chemotherapy in AML patients. AR, haptoglobin α and β and LAO may serve as potential biomarkers for the determination of risk categories and responsiveness of AML patients to chemotherapy.

Keywords: Acute myeloid leukemia, risk categories, proteomics.

Abstrak tesis yang dikemukakan kepada Senat Universiti Putra Malaysia sebagai memenuhi keperluan untuk ijazah Doktor Falsafah

**PENILAIAN KLINIKAL DAN PROFIL PROTEOMIK LEUKEMIA MIELOID
AKUT PESAKIT, ARAB SAUDI**

Oleh

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Leukemia mieloid akut (AML) mewakili sekumpulan gangguan sel dasar hematopoiesis berklon di mana kegagalan untuk membeza dan memproliferasi mengakibatkan terkumpulnya mieloblas yang tidak berfungsi. Dalam AML, gerak balas pesakit terhadap terapi adalah berbeza dan tiada satu penanda yang betul boleh dipercayai untuk meramal gerak balas terhadap rawatan atau prognosis penyakit ini. Dalam kajian ini, objektif utama adalah menentukan profil proteomik AML untuk pesakit yang dirujuk kepada King Faisal Specialist Hospital and Research Center, Riyadh, Arab Saudi. Pesakit AML dikategorikan mengikut tahap risiko berasaskan kemungkinan mandiri. Data klinikal telah dikumpulkan daripada 22 pesakit (16 lelaki dan 6 perempuan) dengan izin. Borang data klinikal telah diwujudkan mengumpul keterangan mengenai diagnosis, pencegahan, dan pemantauan susulan serta pesakit AML.

Data klinikal menunjukkan bahawa kebanyakan pesakit (14) adalah normal sitogenetik dan genetiknya dan diklasifikasi di bawah kategori risiko sederhana. Enam pesakit menyatakan kompleks protein pengikat CCAAT/peningkat (CEBPA), tirosine kinase terkait-FMS (FLT), isositrat dehidrogenase (IDH) dan keabnormalan gen tumor Wilms 1 (WT1), 2 menunjukkan pembalikan pada kromosom 16 (inv.16) dan 1 dengan translokasi kromosom t(15:17).

Protein berlebihan tinggi dalam sampel mesti disingkirkan sebelum perlakuan analisis proteomik. Dalam kajian ini, tiga kaedah untuk pengoptimuman sampel, iaitu kit Pierce Albumin and IgG Removal, Albumin Depletion, and ProteoPrep[®] Immunoaffinity Albumin and IgG Depletion telah dibanding. Kit Pierce Albumin and IgG Removal didapati kaedah yang terbaik untuk penyingkiran protein berlebihan tinggi.

Penyataan protein telah dibandingkan di kalangan pesakit berlainan kategori risikonya, semasa diagnosis dan selepas kebah. Sampel AML daripada darah periferi dan sumsum tulang telah dianalisiskan melalui spektrometri jisim seiring kromatografi cecair (LC-MS/MS). Pada masa diagnosis, dalam kedua-dua sampel darah periferi dan sumsum tulang, 21 protein didapati berbeza secara tererti ($P < 0.05$) dengan nyata pembezaannya di kalangan kumpulan. Bilangan protein yang dikesan dalam darah periferi sama dengan sumsum tulang. Bagaimanapun, terdapat kelainan dalam profil proteomik antara sampel yang diambil semasa diagnosis dan kebah. Kebanyakan protein nyata pembezaan telah terkawal-turun dalam darah periferi pada masa kebah. Di kalangan kes dalam kajian ini, 144 protein nyata pembezaan dikenal pasti, dan kelainan dalam tahap penyataan berkorelasi dengan kategori risiko AML. Kajian itu menunjukkan bahawa pelopor amfiregulin (AR) (terbitan sel kolorektum) ternyata dalam pesakit AML berisiko rendah sahaja, sementara haptoglobin α dan β dan sintetase tirosil-tRNA sitoplasma (EC 6.1.1.1) ternyata tinggi dalam pesakit AML berisiko sederhana sahaja. Protein periplasma pengikat-lisina-arginina-ornitina (LAO) ternyata dalam pesakit AML yang berisiko sederhana dan tinggi dan bukan dalam yang berisiko rendah.

Kesimpulannya, kajian ini menunjukkan bahawa data klinikal lengkap adalah perlu untuk penentuan korelasi antara profil proteomik, kategori risiko dan ramalan gerak balas terhadap kemoterapi dalam pesakit AML. Pelopor amfiregulin, haptoglobin α dan β dan LAO mungkin boleh bertindak sebagai biopenanda berpotensi dalam penentuan kategori risiko dan tahap gerak balas pesakit AML terhadap kemoterapi.

Katakunci: leukemia mieloid akut, kategori risiko, proteomik.

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I certify that a Thesis Examination Committee has met on 8 December 2015 to conduct the final examination of Almaiman Amer Abdulrahman A on his thesis entitled "Clinical Evaluation and Proteomic Profiles of Acute Myeloid Leukemia Patients, Saudi Arabia" in accordance with the Universities and University Colleges Act 1971 and the Constitution of the Universiti Putra Malaysia [P.U.(A) 106] 15 March 1998. The Committee recommends that the student be awarded the Doctor of Philosophy.

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LIST OF ABBREVIATIONS

ADH	Alcohol dehydrogenase
ALL	Acute lymphoid leukemia
AmBic	Ammonium bicarbonate
AML	Acute myeloid leukemia
BM	Bone marrow
BMC	Bone marrow cell
BMP	Bone marrow plasma
BSA	Bovine serum albumin
CBC	Complete blood count
CDK	Cyclin-dependent kinase
CEBPA	CCAAT/enhancer-binding protein
CHAPS	3[(3-Cholamidopropyl) dimethylammonio] propanesulfonic acid
CLL	Chronic lymphoid leukemia
CLP	Common lymphoid progenitor
CML	Chronic myeloid leukemia
CMP	Common myeloid progenitor
CN-AML	Normal cytogenetic-acute myeloid leukemia
CPF	Core binding factor
CR	Complete remission
CSF-1	Colony-stimulating factor 1
CV	Coefficient of variation
Da	Dalton
DBP	D-binding protein precursor
EDTA	Ethylenediaminetetraacetic acid
DE	Dimensional electrophoresis
Del	Deletion
DNA	Deoxyribonucleic acid
DTT	Dithiothreitol
ETO	Eight twenty-one proteins
FAB	French-american-british
FLT	Fms-like tyrosine
Hb	Hemoglobin
HCl	Hydrogen chloride
Hct	Hematocrit
HDMS	High-definition mass spectrometry
HIC	Hydrophobic interaction chromatography
HIDAC	High dosage cytarabine
HAS	Human albumin serum
HSCs	Hematopoietic stem cells
HTLV-1	Human T-lymphotropic virus
IA	Immunoaffinity

IAA	Iodoacetamide
ICE	Ifosfamide, carboplatin, and etoposide
IDH	Isocitrate dehydrogenase
IEF	Isoelectric focusing
IgG	Immunoglobulin
Inv	Inversion
IPA	Ingenuity pathway analysis
IPG	Immobilized ph gradient
LC/MS/MS	Liquid chromatography/ tandem mass spectrometry
LT-HSC	Long-term hematopoietic stem cell
M1	Acute myeloblastic leukemia with minimal maturation
M2	Acute myeloblastic leukemia with maturation
M3	Acute promyelocytic leukemia
M4	Acute myelomonocytic leukemia
M5	Acute monocytic leukemia
M6	Acute erythroid leukemia
M7	Acute megakaryocyte leukemia
MaPK	Mitogen-activated protein kinase
MALDI	Matrix-assisted laser desorption ionization
MDR	Multidrug resistant
MLL	Multi-lineage leukemia
MLP	Multiple lymphomatous polyposis
MPO	Myeloperoxidase
MRD	Minimal residual disease
MRN	Medical record number
MSE	Data-independent acquisition
Mg	Microgram
μl	Microliter
UPLC	Ultra performance liquid chromatography
NaCl	Sodium chloride
NK cells	Natural killer cells
NPM	Nucleophosmin
PAGE	Polyacrylamide gel electrophoresis
PB	Peripheral blood
PBC	Peripheral blood cell
PBS	Phosphate-buffered saline
PBP	Peripheral blood plasmas
PBMC	Peripheral blood mononuclear cell
PCA	Principle component analysis
PH	Partial pressure of (element)
Plt	Platelet
PML	Promyelocytic leukemia
PMSF	Phenylmethylsulfonyl fluoride
PTM	Post-translational modification

RAR	Retinoic acid receptor
RBC	Red blood cell
RBM	Round per minute
Rh	Rhesus
RNA	Ribonucleic acid
RT	Room temperature
RUNX1	Runt-related transcription factor 1
SDS	Sodium dodecyl sulfate
ST-HSC	Short-term hematopoietic stem cell
TCR	T-cell antigen receptor
TET2	Ten eleven translocation 2
TKD	Tyrosine kinase domain
TOF	Time-of-flight
Tris	Tris (hydroxymethyl) aminomethane
WBC	White blood cell
WHO	World health organization
WT1	Wilms tumor 1

CHAPTER 1

INTRODUCTION

1.0 Leukemia and proteomics

Acute and chronic hematological malignancies or “blood cancers” form a distinct subset of cancers that originate in the bone marrow or lymph nodes (Aquino, 2002; Wartenberg *et al.*, 2008). Leukemia is a group of disorders characterized by the accumulation of abnormal leukocytes in the bone marrow and blood circulation. The occupation of these abnormal cells may cause bone marrow failure (Lane and Gilliland, 2010).

Leukemogenesis is a multistep process with several factors that could influence normal hematopoiesis, including over-expression of certain oncogenes, aberrant intracellular pathways, behavior of oncoproteins and expression of chromosom (Reikvam *et al.*, 2011). Acute myeloid leukemia (AML) is a hematological malignancy characterized by clonal proliferation of immature myeloid precursors and arrest in the maturation of these cells (Rubnitz *et al.*, 2010). These abnormalities cause accumulation of leukemic blast cells in the bone marrow and eventually in peripheral blood. The bone marrow infiltration by the immature cells causes a decrease in blood cell production and thereby reducing the peripheral blood numbers of mature erythrocytes (RBCs), leucocytes (WBCs) and platelets (Plts), causing anemia, thrombocytopenia that eventually lead to hemorrhage, neutropenia and loss of resistance to infections. Thus, in AML, patients are initially presented with symptoms often related to pancytopenia, anemia, weakness, fatigue, infections and/or bleeding (Hamid, 2013).

The incidence of AML is about five cases per 100,000 individuals annually and it accounts for approximately 80 % of acute leukemias in adults and 15 to 20 % in children (Robison *et al.*, 2002; Deschler and Lübbert, 2006; Chen *et al.*, 2014). In infants, leukemia is unique in epidemiological, biological, and clinical characteristics. Most of the acute lymphoblastic leukemia (ALL) and AML cases in infants are characterized by high leukocyte count, extramedullary disease, propensity for expression of lymphoid and myeloid phenotypic markers, and translocation of the mixed-lineage leukemia (MLL) (ALL-1, HRX, Htrx-1) gene at chromosome band 11q23 (Reikvam *et al.*, 2011), rapid tumor cell proliferation and a predominance of blast cells. Acute lymphoblastic leukemia is the most common form of leukemia which accounts for 75 to 80 % of all acute leukemia cases in children and adolescents, while AML accounts for 15 to 20% (Yeates *et al.*, 2009). With modern intensive chemotherapy and supportive care, the prognosis of ALL and to a lesser extent AML in children has improved significantly over the past decades (Shipley and Butera, 2009).

The majority of leukemia studies focused on the correlation between leukemic cell phenotypes with cytogenetic and clinical response with the induction and consolidation therapies. Others have evaluated clinical outcomes of different AML subtypes. Few studies have demonstrated the potential of proteomics in the characterization, diagnosis and treatment of hematological malignancies (Luczak *et al.*, 2012). The weakness of leukemia studies lies in the relatively low sample size and more importantly, in the lack of validation of results and with no follow-up pilot clinical trial on the discoveries from their studies.

Currently, apart from the clinical evaluation coupled with hematological, cytogenetic and molecular tests to monitor patient's responses, there is no available objective protein marker for accurate prediction and monitoring of treatment response in AML. There is therefore a need to develop protein markers that could be complementary to currently diagnostic tools in the management of AML patients.

Recent advancements in proteomic technologies have generated vast interest in the pursuit for biomarkers for various hematological malignancies. Large-scale analysis of proteins from cells, tissues, fluids, or organs, using the classical proteomic platforms has yielded information on the disease heterogeneity at different stages of development and progression. Global differential protein profiling analyses or expression proteomics have resulted in identification of disease-related or tissue-specific proteins that could potentially be useful as disease biomarkers (Alaiya *et al.*, 2011).

This quest to translate basic discoveries into patient care regimes has resulted in many proteomic studies describing potential biomarkers. It is disappointing that despite the technological advances, very few studies have found way into clinical trials. The majority of the published proteomic studies have limitations, including inadequate sample representation, non-standardized sample handling and processing, and extreme dynamic range of serum proteins and above the modest sample size, which does not accurately represent the clinical cohorts (Eun *et al.*, 2004; Visanji *et al.*, 2012). It is therefore important that polypeptides identified as potential biomarkers are validated in appropriately processed, well-characterized clinical samples using established techniques, such as immunochemistry or PCR-based methods (Pitkanen and Lukasiuk, 2011).

The understanding of the molecular mechanisms in treatment responses and disease recurrence is limited in AML. Proteomics is the logical approach because proteins change with cellular responses to internal environment, external stimuli, and disease development.

The majority of available biomarkers are single biomolecules with low sensitivity and specificity. Therefore, the use of a combination of measurable protein panels for a more accurate diagnosis and prediction of disease prognosis in AML.

1.1 Hypothesis

Proteomic profiling in combination with clinical data would support the diagnosis, prediction of progression, and risk categorization of acute myeloid leukemia.

1.2 Problem statement

Cancer is among the main causes of death due to disease in developed countries and developing countries (Siegel *et al.*, 2014). Several studies have demonstrated the potential of proteomics and clinical data in the characterization, diagnosis, and treatment of AML (Luczak *et al.*, 2012). However, to date no protein marker has been identified that can accurately be used in the prediction and determination of treatment response in AML. The discovery of AML diagnostic protein markers will complement the currently available diagnostic tools in the management of AML patients.

1.3 General objective

The general goal of this study was to evaluate the AML risk categories and correlate their global protein expressions with the response to induction and consolidation treatments and overall outcome of the disease.

1.4 Specific objectives of the study

The specific objectives of the study are to:

1. Evaluate the clinical data of acute myeloid leukemia.
2. Correlate the proteins fingerprinting with clinical data of AML patients.
3. Optimize high abundance plasma protein removal for accurate and reproducible proteomic analysis in AML.
4. Determine the proteomic profiles of AML using Synapt G2 platform.

REFERENCES

- Abeloff, M. D., Armitage, J. O., Lichter, A. S. & Niederhuber, J. E. (Eds.). (2000). *Clinical Oncology* (2nd ed.) (pp. 2536-2563). New York: Churchill Livingstone.
- Ahmed, N., Barker, G., Oliva, K., Garfin, D., Talmadge, K., Georgiou, H., Quinn, M. & Rice, G. (2003). An approach to remove albumin for the proteomic analysis of low abundance biomarkers in human serum. *Proteomics*, 3(10): 1980-1987.
- Ahmed, N. & Rice, G. E. (2005). Strategies for revealing lower abundance proteins in two-dimensional protein maps. *Journal of Chromatography B*, 815(1-2): 39-50.
- Alaiya, A. A., Al-Mohanna, M., Aslam, M., Shinwari, Z., Al-Mansouri, L., Al-Rodayan, M., Al-Eid, M., Ahmad, I., Hanash, K., Tulbah, A., Bin Mahfooz, A. & Adra, C. (2011). Proteomics-based signature for human benign prostate hyperplasia and prostate adenocarcinoma. *International Journal of Oncology*, 38(4): 1047-1057.
- Alaiya, A., Fox, J., Bobis, S., Matic, G., Shinwari, Z., Barhoush, E., Marquez, M., Nilsson, S. & Holmberg, A. R. (2014). Proteomic analysis of soft tissue tumor implants treated with a novel polybisphosphonate. *Cancer Genomics Proteomics*, 11(1): 39-49.
- Al-Moghrabi N., Nofel A., Al-Yousef N., Madkhali S., Bin Amer S. M., Alaiya, A., Shinwari., Z., Al-Tweigeri, T., Karakas, B., Tulbah, A. & Aboussekhra, A. (2014). The molecular significance of methylated BRCA1 promoter in white blood cells of cancer-free females. *BioMedical Central Cancer*, 14: 830.
- American Cancer Society. (2014). *Cancer Facts & Figures 2014*. American Cancer Society: Atlanta, GA.
- Anderson, N. L. & Anderson, N. G. (2002). The human plasma proteome: history, character, and diagnostic prospects. *Molecular & Cellular Proteomics*, 1(11): 845-867.
- Aquino, V. M. (2002). Acute myelogenous leukemia. *Current Problems in Pediatric and Adolescent Health Care*, 32(2): 50-58.
- Arber, D. A., Stein, A. S., Carter, N. H., Ikle, D., Forman, S. J. & Slovak, M. L. (2003). Prognostic impact of acute myeloid leukemia classification. *American Journal of Clinical Pathology*, 119(5): 672-680.
- Ashraf, N. & Irshad, S. (2012). Molecular genetics of acute myeloid leukemia. *Advances in Life Sciences*, 2(4): 112-117.
- Bacher, U., Kern, W., Schnittger, S., Hiddemann, W., Haferlach, T. & Schoch, C. (2005). Population-based age-

- specific incidences of cytogenetic subgroups of acute myeloid leukemia. *Hematologica*, 90(11): 1502-1510.
- Asif, M., Ofelia, B., Joycelyn, C. & Asmara, K. (2012). Tumor Registry Annual Report (2011). King Faisal Specialist Hospital and Research Centre, Riyadh, Saudi Arabia.
- Bacher, U., Kohlmann, A., Haferlach, C. & Haferlach, T. (2009). Gene expression profiling in acute myeloid leukaemia (AML). *Best Practice & Research Clinical Hematology*, 22(2): 169-180.
- Bai, J., He, A., Zhang, W., Huang, C., Yang, J., Yang, Y. & Zhang, Y. (2013). Potential biomarkers for adult acute myeloid leukemia minimal residual disease assessment searched by serum peptidome profiling. *Proteome Science*, 11: 39.
- Balkhi, M. Y., Trivedi, A. K., Geletu, M., Christopheit, M., Bohlander, S. K., Behre, H. M. & Behre, G. (2006). Proteomics of acute myeloid leukaemia: cytogenetic risk groups differ specifically in their proteome, interactome and post-translational protein modifications. *Oncogene*, 25(53): 7041-7058.
- Bellei, E., Bergamini, S., Monari, E., Fantoni, L. I., Cuoghi, A., Ozben, T. & Tomasi, A. (2011). High-abundance proteins depletion for serum proteomic analysis: concomitant removal of non-targeted proteins. *Amino Acids*, 40(1): 145-156.
- Berger, U., Maywald, O., Pfirrmann, M., Lahaye, T., Hochhaus, A., Reiter, A., Hasford, J., Heimpel, H., Hossfeld, D. K., Kolb, H. J., Loffler, H., Pralle, H., Queisser, W. & Hehlmann, R. (2005). Gender aspects in chronic myeloid leukemia: long-term results from randomized studies. *Leukemia*, 19(6): 984-989.
- Bienz, M., Ludwig, M., Mueller, B. U., Oppliger Leibundgut, E., Ratschiller, D., Solenthaler, M., Fey, M. F. & Pabst, T. (2005). Risk assessment in patients with acute myeloid leukemia and a normal karyotype. *Clinical Cancer Research*, 11(15): 1416-1424.
- Björhall, K., Miliotis, T. & Davidsson, P. (2005). Comparison of different depletion strategies for improved resolution in proteomic analysis of human serum samples. *Proteomics*, 5(1): 307-317.
- Boyd, R. S., Dyer, M. J. & Cain, K. (2010). Proteomic analysis of b-cell malignancies. *Proteomics*, 73(10): 1804-1822.
- Bradford, M. M. (1976). A rapid and sensitive method for the quantitation of microgram quantities of protein utilizing the principle of protein-dye binding. *Analytical Biochemistry*, 72: 248-254.
- Brand, J., Haslberger, T., Zolg, W., Pestlin, G. & Palme, S. (2006). Depletion efficiency and recovery of trace markers from an ultiparameter immunodepletion column. *Proteomics*, 6(11): 3236-3242.

- Breems, D. A., Van Putten, W. L., Huijgens, P. C., Ossenkoppele, G. J., Verhoef, G. E., Verdonck, L. F. & Löwenberg, B. (2005). Prognostic index for adult patients with acute myeloid leukemia in first relapse. *Journal of Clinical Oncology*, 23(9): 1969-1978.
- Bruserud, O., Hovland, R., Wergeland, L., Huang, T. & Gjertsen, B. (2003). Flt3-mediated signaling in human acute myelogenous leukemia (AML) blasts: a functional characterization of Flt3-ligand effects in AML cell populations with and without genetic Flt3 abnormalities. *Haematologica*, 88(4): 416-428.
- Bryder, D., Rossi, D. J. & Weissman, I. L. (2006). Hematopoietic stem cells: the paradigmatic tissue-specific stem cell. *The American Journal of Pathology*, 169(2): 338-346.
- Burnett, A. K. (2002). Acute myeloid leukemia: Treatment of adults under 60 years. *Reviews in Clinical and Experimental Hematology*, 6(1): 26-45.
- Cagnetta, A., Adamia, S., Acharya, C., Patrone, F., Miglino, M., Nencioni, A., Gobbi, M. & Cea, M. (2014). Role of genotype-based approach in the clinical management of adult acute myeloid leukemia with normal cytogenetics. *Leukemia Research*, 38(6): 649-659.
- Chauhan, P. S., Ihsan, R., Singh, L. C., Gupta, D. K., Mittal, V. & Kapur, S. (2013). Mutation of NPM1 and FLT3 genes in acute myeloid leukemia and their association with clinical and immunophenotypic features. *Disease Markers*, 35(5): 581-588.
- Chen, K. H., Liu, H. C., Liang, D. C., Hou, J. Y., Huang, T. H., Chang, C. Y. & Yeh, T. C. (2014). Minimally early morbidity in children with acute myeloid leukemia and hyperleukocytosis treated with prompt chemotherapy without leukapheresis. *Journal of the Formosan Medical Association*, 113(11): 833-838.
- Chen, Y. Y., Lin, S. Y., Yeh, Y. Y., Hsaio, H. H., Wu, C. Y., Chen, S. T. & Wang, A. H. J. (2005). A modified protein precipitation procedure for efficient removal of albumin from serum. *Electrophoresis*, 26(11): 2117-2127.
- Cheson, B. D., Bennett, J. M., Kopecky, K. J., Büchner, T., Willman, C. L., Estey, E. H., Schiffer, C. A., Doehner, H., Tallman, M. S. & Lister, T. A. (2003). Revised recommendations of the international working group for diagnosis, standardization of response criteria, treatment outcomes, and reporting standards for therapeutic trials in acute myeloid leukemia. *Journal of Clinical Oncology*, 21(24): 4642-4649.
- Chuang, S. S. & Li, C. Y. (1997). Useful pane of antibodies for the classification of acute leukemias by immunohistochemical methods in bone marrow trephine biopsy sections. *American Journal of Clinical Pathology*, 107(4): 410-418.

- Cohen, A., Rovelli, A., Bakker, B., Uderzo, C., Van Lint, M. T., Esperou, H., Gaiero, A., Leiper, A. D., Dopfer, R., Cahn, J. Y., Merlo, F., Kolb, H. J. & Socie, G. (1999). Final height of patients who underwent bone marrow transplantation for hematological disorders during childhood: a study by the Working Party for Late Effects-EBMT. *Blood*, 93(12): 4109-4115.
- Colantonio, D. A., Dunkinson, C., Bovenkamp, D. E. & Van Eyk, J. E. (2005). Effective removal of albumin from serum. *Proteomics*, 5(15): 3831-3835.
- Craig, F. E. & Foon, K. A. (2008). Flow cytometric immunophenotyping for hematologic neoplasms. *Blood*, 111(8): 3941-3967.
- Cristea, I. M., Gaskell, S. J. & Whetton, A. D. (2004). Proteomics techniques and their application to hematology. *Blood*, 103(10): 3624-3634.
- Cui, J. W., Wang, J., He, K., Jin, B. F., Wang, H. X., Li, W., Kang, L. H., Hu, M. R., Li, H. Y. & Yu, M. (2004). Proteomic analysis of human acute leukemia cells: insight into their classification. *Clinical Cancer Research*, 10(20): 6887-6896.
- Cui, J. W., Wang, J., He, K., Jin, B. F., Wang, H. X., Li, W. & Zhang, X. M. (2005). Two dimensional electrophoresis protein profiling as an analytical tool for human acute leukemia classification. *Electrophoresis*, 26(1): 268-279.
- Daraki, A., Zachaki, S., Koromila, T., Diamantopoulou, P., Pantelias, G. E., Sambani, C. & Manola, K. N. (2014). The G516T CYP2B6 germline polymorphism affects the risk of acute myeloid leukemia and is associated with specific chromosomal abnormalities. *PLoS One*, 9(2): e88879.
- Dardé, V. M., Barderas, M. G. & Vivanco, F. (2007). Depletion of high-abundance proteins in plasma by immunoaffinity subtraction for two-dimensional difference gel electrophoresis analysis. *Methods in Molecular Biology*, 357: 351-364.
- Dayarathna, M., Hancock, W. S. & Hincapie, M. (2008). A two-step fractionation approach for plasma proteomics using immunodepletion of abundant proteins and multi-lectin affinity chromatography: Application to the analysis of obesity, diabetes, and hypertension diseases. *Journal of Separation Science*, 31(6-7): 1156-1166.
- Dekker, L. J., Bosman, J., Burgers, P. C., van Rijswijk, A., Freije, R., Luider, T. & Bischoff, R. (2007). Depletion of high-abundance proteins from serum by immunoaffinity chromatography: a MALDI-FT-MS study. *Journal of Chromatography B*, 847(1): 65-69.
- Deng, W., Lee, J., Wang, H., Miller, J., Reik, A., Gregory, P. D., Dean, A. & Blobel, G. A. (2012). Controlling long-range genomic interactions at a native locus by targeted tethering of a looping factor. *Cell*, 149(6): 1233-1244.
- Deschler, B. & Lübbert, M. (2006). Acute myeloid leukemia: epidemiology and etiology. *Cancer*, 107(9): 2099-2107.

- Dillman, R., Davis, R., Green, M., Weiss, R., Gottlieb, A., Caplan, S., Kopel, S., Preisler, H., McIntyre, O. & Schiffer, C. (1991). A comparative study of two different doses of cytarabine for acute myeloid leukemia: a phase III trial of Cancer and Leukemia Group B. *Blood*, 78(10): 2520-2526.
- Döhner, H., Estey, E. H., Amadori, S., Appelbaum, F. R., Büchner, T., Burnett, A. K., Dombret, H., Fenaux, P., Grimwade, D., Larson, R. A., Lo-Coco, F., Naoe, T., Niederwieser, D., Ossenkoppele, G. J., Sanz, M. A., Sierra, J., Tallman, M. S., Löwenberg, B. & Bloomfield, C. D.; European LeukemiaNet. (2010). Diagnosis and management of acute myeloid leukemia in adults: recommendations from an international expert panel, on behalf of the European LeukemiaNet. *Blood*, 115(3): 453-474.
- Doulatov, S., Notta, F., Laurenti, E. & Dick, J. E. (2012). Hematopoiesis: a human perspective. *Cell Stem Cell*, 10(2): 120-136.
- Downing, J. R. (1999). The AML1-ETO chimaeric transcription factor in acute myeloid leukaemia: biology and clinical significance. *British Journal of Haematology*, 106(2): 296-308.
- Echan, L. A., Tang, H. Y., Ali-Khan, N., Lee, K. & Speicher, D. W. (2005). Depletion of multiple high-abundance proteins improves protein profiling capacities of human serum and plasma. *Proteomics*, 5(13): 3292-3303.
- Enany, S., Yoshida, Y., Magdeldin, S., Bo, X., Zhang, Y., Enany, M. & Yamamoto, T. (2013). Two dimensional electrophoresis of the exo-proteome produced from community acquired methicillin resistant Staphylococcus aureus belonging to clonal complex 80. *Microbiological Research*, 168(8): 504-511.
- Erickson, P., Gao, J., Chang, K., Look, T., Whisenant, E., Raimondi, S., Lasher, R., Trujillo, J., Rowley, J. & Drabkin, H. (1992). Identification of breakpoints in t(8;21) acute myelogenous leukemia and isolation of a fusion transcript, AML1/ETO, with similarity to Drosophila segmentation gene, runt. *Blood*, 80(7): 1825-1831.
- Estey, E. H. (2000). How I treat older patients with AML. *Blood*, 96(5): 1670-1673.
- Estey, E. & Döhner, H. (2006). Acute myeloid leukaemia. *The Lancet*, 368(9550): 1894-1907.
- Eun, J. P., Choi, H. Y. & Kwak, Y. G. (2004). Proteomic analysis of human cerebral cortex in epileptic patients. *Experimental Molecular Medicine*, 36(2): 185-191.
- Evans, T. & Felsenfeld, G. (1989). The erythroid-specific transcription factor eryf1: A new finger protein. *Cell*, 58(5): 877-885.
- Faca, V., Coram, M., Phanstiel, D., Glukhova, V., Zhang, Q., Fitzgibbon, M., McIntosh, M. & Hanash, S. (2006). Quantitative analysis of acrylamide

- labeled serum proteins by LC-MS/MS. *Journal of Proteome Research*, 5: 2009–2018.
- Falini, B., Mecucci, C., Tiacci, E., Alcalay, M., Rosati, R., Pasqualucci, L., La Starza, R., Diverio, D., Colombo, E., Santucci, A., Bigerna, B., Pacini, R., Pucciarini, A., Liso, A., Vignetti, M., Fazi, P., Meani, N., Pettrossi, V., Saglio, G., Mandelli, F., Lo-Coco, F., Pelicci, P. G. & Martelli, M. F. (2005). Cytoplasmic nucleophosmin in acute myelogenous leukemia with a normal karyotype. *New England Journal of Medicine*, 352(3): 254-266.
- Fang, X. & Zhang, W. W. (2008). Affinity separation and enrichment methods in proteomic analysis. *Proteomics*, 71(3): 284-303.
- Feng, J. T., Liu, Y. K., Song, H. Y., Dai, Z., Qin, L. X., Almofti, M. R., Fang, C. Y., Lu, H. J., Yang, P. Y. & Tang, Z. Y. (2005). Heat-shock protein 27: A potential biomarker for hepatocellular carcinoma identified by serum proteome analysis. *Proteomics*, 5(17): 4581-4588.
- Forman, D., Stockton, D., Møller, H., Quinn, M., Babb, P., De Angelis, R. & Micheli, A. (2003). Cancer prevalence in the UK: results from the EUROPREVAL study. *Annals of Oncology*, 14(4): 648-654.
- Frantzi, M., Bhat, A. & Latosinska, A. (2014). Clinical proteomic biomarkers: relevant issues on study design & technical considerations in biomarker development. *Clinical and Translational Medicine*, 3(1), 7.
- Galvani, M., Hamdan, M., Herbert, B. & Righetti, P. G. (2001a). Alkylation kinetics of proteins in preparation for two-dimensional maps: a matrix assisted laser desorption/ionization- mass spectrometry investigation. *Electrophoresis*, 22(10): 2058-2065.
- Galvani, M., Rovatti, L., Hamdan, M., Herbert, B. & Righetti, P. G. (2001b). Protein alkylation in the presence/absence of thiourea in proteome analysis: A matrix assisted laser desorption/ionization-time of flight-mass spectrometry investigation. *Electrophoresis*, 22(10): 2066-2074.
- Gast, M. C. W., Schellens, J. H. & Beijnen, J. H. (2009). Clinical proteomics in breast cancer: a review. *Breast Cancer Research and Treatment*, 116(1): 17-29.
- Goldstone, A. H., Burnett, A. K., Wheatley, K., Smith, A. G., Hutchinson, R. M. & Clark, R. E. (2001). Attempts to improve treatment outcomes in acute myeloid leukemia (AML) in older patients: the results of the United Kingdom Medical Research Council AML11 trial. *Blood*, 98(5), 1302-1311.
- Gelfand, C. A. & Omenn, G. S. (2011). Preanalytical variables for plasma and serum proteome analyses. In A. R. Ivanov, & A. V. Lazarev (Eds.), *Sample Preparation in Biological Mass Spectrometry* (pp. 269-289). Netherlands: Springer.

- Golub, T., Weinstein, H. & Grier, H. (1997). Acute Myelogenous Leukemia. In P. Pizzo, & D. Poplack (Eds.), *Principles and Practice of Pediatric Oncology* (pp.463-482). Philadelphia: Lippincott-Raven.
- Golub, T. R., Barker, G. F., Lovett, M. & Gilliland, D. G. (1994). Fusion of PDGF receptor β to a novel ets-like gene, tel, in chronic myelomonocytic leukemia with t(5;12) chromosomal translocation. *Cell*, 77(2): 307-316.
- Golub, T. R., Slonim, D. K., Tamayo, P., Huard, C., Gaasenbeek, M., Mesirov, J. P., Coller, H., Loh, M. L., Downing, J. R., Caligiuri, M. A., Bloomfield, C. D. & Lander, E. S. (1999). Molecular classification of cancer: class discovery and class prediction by gene expression monitoring. *Science*, 286(5439): 531-537.
- Grandjean, M., Dieu, M., Raes, M. & Feron, O. (2013). A new method combining sequential immunoaffinity depletion and differential in gel electrophoresis to identify autoantibodies as cancer biomarkers. *Journal of Immunological Methods*, 396(1-2): 23-32.
- Greaves, M. (1997). Aetiology of acute leukaemia. *The Lancet*, 349(9048): 344-349.
- Greenlee, R. T., Hill-Harmon, M. B., Murray, T. & Thun, M. (2001). Cancer Statistics, 2001. *CA: A Cancer Journal for Clinicians*, 51(1): 15-36.
- Grimwade, D., Hills, R. K., Moorman, A. V., Walker, H., Chatters, S., Goldstone, A. H., Wheatley, K., Harrison, C. J. & Burnett, A. K. (2010). Refinement of cytogenetic classification in acute myeloid leukemia: determination of prognostic significance of rare recurring chromosomal abnormalities among 5876 younger adult patients treated in the United Kingdom Medical Research Council trials. *Blood*, 116(3): 354-365.
- Grimwade, D., Walker, H., Oliver, F., Wheatley, K., Harrison, C., Harrison, G., Rees, J., Hann, I., Stevens, R., Burnett, A. & Goldstone, A. (1998). The importance of diagnostic cytogenetics on outcome in AML: analysis of 1,612 patients entered into the MRC AML 10 trial. *Blood*, 92(7): 2322-2333.
- Guo, H., Ma, O., Speck, N. A. & Friedman, A. D. (2012). Runx1 deletion or dominant inhibition reduces Cebpa transcription via conserved promoter and distal enhancer sites to favor monopoiesis over granulopoiesis. *Blood*, 119(19): 4408-4418.
- Gurney, J. G., Severson, R. K., Davis, S. & Robison, L. L. (1995). Incidence of cancer in children in the United States. Sex-, race-, and 1-year age-specific rates by histologic type. *Cancer*, 75(8): 2186-2195.
- Guyton, A. C. & Hall, J. E. (1996). Red Blood Cells, Anemia and Polycythemia. In A. C. Guyton, & J. E. Hall (Eds.), *Textbook of Medical Physiology* (pp. 353-71). Philadelphia: W.B. Saunders.

- Gygi, S. P., Corthals, G. L., Zhang, Y., Rochon, Y. & Aebersold, R. (2000). Evaluation of two-dimensional gel electrophoresis-based proteome analysis technology. *Proceedings of the National Academy of Sciences*, 97(17): 9390-9395.
- Hahn, C. K., Berchuck, J. E., Ross, K. N., Kakoza, R. M., Clauser, K., Schinzel, A. C. & Stegmaier, K. (2009). Proteomic and genetic approaches identify Syk as an AML target. *Cancer Cell*, 16(4): 281-294.
- Hamid, G. A. (2013). Acute Leukemia Clinical Presentation. In M. Guenova, & G. Balatzenko (Eds.), *Leukemia*. DOI: 10.5772/53531
- Hannan, N. J., Stoikos, C. J., Stephens, A. N. & Salamonsen, L. A. (2008). Depletion of high-abundance serum proteins from human uterine lavages enhances detection of lower-abundance proteins. *Journal of Proteome Research*, 8(2): 1099-1103.
- Hawkrige, A. M., Muddiman, D. C., Hebblein, D. M., Cataliotti, A. & Burnett, J. C. (2008). Effect of plasma protein depletion on BNP-32 recovery. *Clinical Chemistry*, 54(5): 933-934.
- Heerema, N. A. & Raimondi, S. C. (2013). Cytogenetics of Acute Leukemia. In P. H. Wiernik, J. M. Goldman, J. P. Dutcher, & R. A. Kyle (Eds.), *Neoplastic Diseases of the Blood*. New York: Springer.
- Hegedus, C. M., Gunn, L., Skibola, C. F., Zhang, L., Shiao, R., Fu, S., Dalmasso, E. A., Metayer, C., Dahl, G. V., Buffler, P. A. & Smith, M. T. (2005). Proteomic analysis of childhood leukemia. *Leukemia*, 19(10): 1713-1718.
- Hehlmann, R., Berger, U. & Hochhaus, A. (2005). Chronic myeloid leukemia: a model for oncology. *Annals of Hematology*, 84(8): 487-497.
- Hehlmann, R., Berger, U., Pffirmann, M., Hochhaus, A., Metzgeroth, G., Maywald, O., Hasford, J., Reiter, A., Hossfeld, D. K., Kolb, H. J., Löffler, H., Pralle, H., Queiszer, W., Griesshammer, M., Nerl, C., Kuse, R., Tobler, A., Eimermacher, H., Tichelli, A., Aul, C., Wilhelm, M., Fischer, J. T., Perker, M., Scheid, C., Schenk, M., Weisz, J., Meier, C. R., Kremers, S., Labedzki, L., Schmeiser, T., Lohrmann, H. P. & Heimpel, H. (2003). Randomized comparison of interferon [alpha] and hydroxyurea with hydroxyurea monotherapy in chronic myeloid leukemia (CML-study II): prolongation of survival by the combination of interferon alpha and hydroxyurea. *Leukemia*, 17(8): 1529-1537.
- Hehlmann, R., Hochhaus, A. & Baccarani, M. (2007). Chronic myeloid leukaemia. *The Lancet*, 370(9584): 342-350.
- Herbert, B., Galvani, M., Hamdan, M., Olivieri, E., MacCarthy, J., Pedersen, S. & Righetti, P. G. (2001). Reduction and alkylation of proteins in preparation of two-dimensional map analysis: Why, when, and how? *Electrophoresis*, 22(10): 2046-2057.

- Hiddemann, W., Kern, W., Schoch, C., Fonatsch, C., Heinecke, A., Wörmann, B. & Büchner, T. (1999). Management of acute myeloid leukemia in elderly patients. *Journal of Clinical Oncology*, 17(11): 3569-3576.
- Ho, A. D., Schetelig, J., Bochtler, T., Schaich, M., Schäfer-Eckart, K., Hänel, M. & Berdel, W. E. (2015). Allogeneic Stem Cell Transplantation Improves Survival in Patients with Acute Myeloid Leukemia Characterized by a High Allelic Ratio of Mutant FLT3-ITD. *Biology of Blood and Marrow Transplantation*, 1-8.
- Hoffbrand, V. & Moss, P. (2011). *Essential Haematology*. Hoboken, NJ: John Wiley & Sons.
- Holčápek, M., Jirásko, R. & Lída, M. (2012). Recent developments in liquid chromatography–mass spectrometry and related techniques. *Journal of Chromatography A*, 1259: 3-15.
- Holmes, T., Yan, F., Ko, K. H., Nordon, R., Song, E., O'Brien, T. A. & Dolnikov, A. (2012). Ex vivo expansion of cord blood progenitors impairs their short-term and long-term repopulating activity associated with transcriptional dysregulation of signalling networks. *Cell Proliferation*, 45(3): 266-278.
- Hourigan, C. S. & Karp, J. E. (2010). Development of therapeutic agents for older patients with acute myelogenous leukemia. *Current Opinions in Investigational Drugs*, 11(6): 669-677.
- Hourigan, C. S. & Karp, J. E. (2013). Minimal residual disease in acute myeloid leukaemia. *Nature Reviews Clinical Oncology*, 10(8): 460-471.
- Huang, L., Harvie, G., Feitelson, J. S., Gramatikoff, K., Herold, D. A., Allen, D. L., Amunngama, R., Hagler, R. A., Pisano, M. R. & Zhang, W. W. (2005). Immunoaffinity separation of plasma proteins by IgY microbeads: meeting the needs of proteomic sample preparation and analysis. *Proteomics*, 5(13): 3314-3328.
- Hughes, T., Deininger, M., Hochhaus, A., Branford, S., Radich, J., Kaeda, J., Baccarani, M., Cortes, J., Cross, N. C. P., Druker, B. J., Gabert, J., Grimwade, D., Hehlmann, R., Kamel-Reid, S., Lipton, J. H., Longtine, J., Martinelli, G., Saglio, G., Soverini, S., Stock, W. & Goldman, J. M. (2006). Monitoring CML patients responding to treatment with tyrosine kinase inhibitors: review and recommendations for harmonizing current methodology for detecting BCR-ABL transcripts and kinase domain mutations and for expressing results. *Blood*, 108(1): 28-37.
- Hütter, G., Letsch, A., Nowak, D., Poland, J., Sinha, P., Thiel, E. & Hofmann, W. K. (2009). High correlation of the proteome patterns in bone marrow and peripheral blood blast cells in patients with acute myeloid leukemia. *Journal of Translational Medicine*, 7: 7.

- Infante-Rivard, C., Fortier, I. & Olson, E. (2000). Markers of infection, breast-feeding and childhood acute lymphoblastic leukaemia. *British Journal of Cancer*, 83(11): 1559-1564.
- Inutan, E. & Trimpin, S. (2010). Laserspray ionization (LSI) ion mobility spectrometry (IMS) mass spectrometry. *Journal of the American Society for Mass Spectrometry*, 21(7): 1260-1264.
- Jemal, A., Murray, T., Ward, E., Samuels, A., Tiwari, R. C., Ghafoor, A., Feuer, E. J. & Thun, M. J. (2005). Cancer statistics, 2005. *CA: A Cancer Journal for Clinicians*, 55(1): 10-30.
- Jemal, A., Siegel, R., Ward, E., Murray, T., Xu, J., Smigal, C. & Thun, M. J. (2006). Cancer Statistics, 2006. *CA: A Cancer Journal for Clinicians*, 56(1): 106-130.
- Jemal, A., Siegel, R., Xu, J. & Ward, E. (2010). Cancer statistics,(2010). *CA: A Cancer Journal for Clinicians*, 60(5): 277-300.
- Jemal, A., Thomas, A., Murray, T. & Thun, M. (2002). Cancer Statistics, 2002. *CA: A Cancer Journal for Clinicians*, 52(1): 23-47.
- Jourdan, E., Boissel, N., Chevret, S., Delabesse, E., Renneville, A., Cornillet, P. & Dombret, H. (2013). Prospective evaluation of gene mutations and minimal residual disease in patients with core binding factor acute myeloid leukemia. *Blood*, 121(12): 2213-2223.
- Kalil, N. & Cheson, B. D. (1999). Chronic lymphocytic leukemia. *The Oncologist*, 4(5): 352-369.
- Kanaujiya, J. K., Lochab, S., Pal, P., Christopheit, M., Singh, S. M., Sanyal, S., Behre, G. & Trivedi, A. K. (2011). Proteomic approaches in myeloid leukemia. *Electrophoresis*, 32(3-4): 357-367.
- Kaushansky, K. (2006). Lineage-specific hematopoietic growth factors. *New England Journal of Medicine*, 354(19): 2034-2045.
- Karp, J. E., Garrett-Mayer, E., Estey, E. H., Rudek, M. A., Smith, B. D., Greer, J. M. & Pagel, J. M. (2012). Randomized phase II study of two schedules of flavopiridol given as timed sequential therapy with cytosine arabinoside and mitoxantrone for adults with newly diagnosed, high-risk acute myelogenous leukemia. *Haematologica*, 97(11): 1736-1742.
- Keating, M. J. (2002). Chronic Lymphocytic Leukemia. In E. S. Henderson, T. A. Lister. & M. F. Greaves (Eds.), *Leukemia*, 7th Edition (pp. 656-691). Philadelphia: Saunders.
- Khanna-Gupta, A., Abayasekara, N., Levine, M., Sun, H., Virgilio, M., Nia, N., Halene, S., Sportoletti, P., Jeong, J. Y., Pandolfi, P. P. & Berliner, N. (2012). Upregulation of translation initiation factor eIF4E in nucleophosmin 1 haploinsufficient cells results in changes in CCAAT enhancer binding protein

- alpha activity: implications in myelodysplastic syndrome and acute myeloid leukemia. *Journal of Biological Chemistry*, 287(39): 32728-32737.
- Kipps, T. J. (2001). Chronic Lymphocytic Leukemia and Related Diseases. In E. Beutler, B. S. Collier, M. A. Lichtman, T. J. Kipps, U. & Seligsohn (Eds.), *Williams Hematology* (pp. 1163-1194). New York: Oxford University Press.
- Klaus, M., Haferlach, T., Schnittger, S., Kern, W., Hiddemann, W. & Schoch, C. (2004). Cytogenetic profile in de novo acute myeloid leukemia with FAB subtypes M0, M1, and M2: a study based on 652 cases analyzed with morphology, cytogenetics, and fluorescence in situ hybridization. *Cancer Genetics and Cytogenetics*, 155(1): 47-56.
- Klepin, H. D. (2016). Myelodysplastic Syndromes and Acute Myeloid Leukemia in the Elderly. *Clinics in geriatric medicine*, 32(1), 155-173.
- Koleva, R. I., Ficarro, S. B., Radomska, H. S., Carrasco-Alfonso, M. J., Alberta, J. A., Webber, J. T., Luckey, C. J., Marcucci, G., Tenen, D. G. & Marto, J. A. (2012). C/EBP α and DEK coordinately regulate myeloid differentiation. *Blood*, 119(21): 4878-4888.
- Konuma, T., Takahashi, S., Uchida, N., Kuwatsuka, Y., Yamasaki, S., Aoki, J. & Masuko, M. (2015). Effect of granulocyte colony-stimulating factor combined conditioning in cord blood transplantation for myelodysplastic syndrome and secondary acute myeloid leukemia: a retrospective study in Japan. *Biology of Blood and Marrow Transplantation*, 21(9):1632–1640.
- Krishnankutty, B., Bellary, S., Kumar, N. B. R. & Moodahudu L. S. (2012). Data management in clinical research: An overview. *Indian Journal of Pharmacology*, 44(2): 168-172.
- Kuick, R., Misek, D.E., Monsma, D.J., Webb, C.P., Wang, H., Peterson, K.J., Pisano, M., Omenn, G.S. & Hanash, S.M. (2007). Discovery of cancer biomarkers through the use of mouse models. *Cancer Letter* 249, 40–48.
- Lafiura, K. M., Bielawski, D. M., Posecion, N. C., Ostrea, E. M., Matherly, L. H., Taub, J. W. & Ge, Y. (2007). Association between prenatal pesticide exposures and the generation of leukemia-associated T(8;21). *Pediatric Blood and Cancer*, 49(5): 624-628.
- Lane, S. W. & Gilliland, D. G. (2010). Leukemia stem cells. *Seminars in Cancer Biology*, 20(2): 71-76.
- Laurier, D., Valenty, M. & Tiramarche, M. (2001). Radon exposure and the risk of leukemia: a review of epidemiological studies. *Health Physics*, 81(3): 272-288.
- Lee, W. C. & Lee, K. H. (2004). Applications of affinity chromatography in proteomics. *Analytical Biochemistry*, 324(1): 1-10.

- Lekstrom-Himes, J. & Xanthopoulos, K. G. (1998). Biological role of the CCAAT/enhancer-binding protein family of transcription factors. *Journal of Biological Chemistry*, 273(44): 28545-28548.
- Leung, W., Hudson, M. M., Strickland, D. K., Phipps, S., Srivastava, D. K., Ribeiro, R. C., Rubnitz, J. E., Sandlund, J. T., Kun, L. E., Bowman, L. C., Razzouk, B. I., Mathew, P., Shearer, P., Evans, W. E. & Pui, C. H. (2000). Late effects of treatment in survivors of childhood acute myeloid leukemia. *Journal of Clinical Oncology*, 18(18): 3273-3279.
- Lin, B., White, J. T., Wu, J., Lele, S., Old, L. J., Hood, L. & Odunsi, K. (2009). Deep depletion of abundant serum proteins reveals low abundant proteins as potential biomarkers for human ovarian cancer. *Proteomics-Clinical Applications*, 3(7): 853-861.
- Liu, Y., Aiello, A. & Zinkel, S. S. (2012). Bid protects the mouse hematopoietic system following hydroxyurea-induced replicative stress. *Cell Death Differentiation*, 19(10): 1602-1612.
- Liu, T., Qian, W. J., Mottaz, H. M., Gritsenko, M. A., Norbeck, A. D., Moore, R. J. & Smith, R. D. (2006). Evaluation of multiprotein immunoaffinity subtraction for plasma proteomics and candidate biomarker discovery using mass spectrometry. *Molecular & Cellular Proteomics*, 5(11): 2167-2174.
- Lockhart, D. J. & Winzler, E. A. (2000). Genomics, gene expression and DNA arrays. *Nature*, 405(6788): 827-36.
- López-Pedrerá, C., Villalba, J. M., Siendones, E., Barbarroja, N., Gómez-Díaz, C., Rodríguez-Ariza, A., Buendía, P., Torres, A. & Velasco, F. (2006). Proteomic analysis of acute myeloid leukemia: Identification of potential early biomarkers and therapeutic targets. *Proteomics*, 6 (Suppl 1): S293-S299.
- Lowenberg, B., Downing, J. R. & Burnett, A. (1999). Acute myeloid leukemia. *New England Journal of Medicine*, 341(14): 1051-1062.
- Löwenberg, B., Griffin, J. D. & Tallman, M. S. (2003). Acute myeloid leukemia and acute promyelocytic leukemia. *Hematology 2003 American Society of Hematology Education Program*, 2003: 82-101.
- Luczak, M., Kazmierczak, M., Handschuh, L., Lewandowski, K., Komarnicki, M. & Figlerowicz, M. (2012). Comparative proteome analysis of acute myeloid leukemia with and without maturation. *Proteomics*, 12(18): 5734-5748.
- Magdeldin, S., Enany, S., Yoshida, Y., Xu, B., Zhang, Y., Zureena, Z., Lokamani, I., Yaoita, E. & Yamamoto, T. (2014). Basics and recent advances of two dimensional-polyacrylamide gel electrophoresis. *Clinical Proteomics*, 11(1): 16.
- Mahn, A., Reyes, A., Zamorano, M., Cifuentes, W. & Ismail, M. (2010). Depletion of highly abundant proteins in blood plasma by hydrophobic interaction

- chromatography for proteomic analysis. *Journal of Chromatography B*, 878(5-16): 1038-1044.
- Manola, K. N. (2009). Cytogenetics of pediatric acute myeloid leukemia. *European Journal of Haematology*, 83(5): 391-405.
- Margolin, J. & Poplack, D. (1997). Acute Lymphoblastic Leukemia. In P. Pizzo, & D. Poplack (Eds.), *Principles and Practice of Pediatric Oncology* (pp. 409-462). Philadelphia: Lippincott-Raven.
- Martorella A. & Robbins R. (2007). Serum peptide profiling: identifying novel cancer biomarkers for early disease detection. *Acta Biomedica*, 78 (Suppl 1): 123-128.
- Matondo, M., Bousquet-Dubouch, M. P., Gally, N., Uttenweiler-Joseph, S., Recher, C., Payrastra, B., Manenti, S., Monsarrat, B. & Burlet-Schiltz, O. (2010). Proteasome inhibitor-induced apoptosis in acute myeloid leukemia: a correlation with the proteasome status. *Leukemia Research*, 34(4): 498-506.
- Matrangolo, F. S., Liarte, D. B., Andrade, L. C., De Melo, M. F., Andrade, J. M., Ferreira, R. F., Santiago, A. S., Pirovani, C. P., Silva-Pereira, R. A. & Murta, S. M. (2013). Comparative proteomic analysis of antimony-resistant and-susceptible *Leishmania braziliensis* and *Leishmania infantum chagasi* lines. *Molecular and Biochemical Parasitology*, 190(2): 63-75.
- McCarthy D. J. & Smyth G. K. (2009). Testing significance relative to a fold-change threshold is a TREAT. *Bioinformatics*, 25(6):765-771.
- Mehta, P. (2012). ARUP Consult: The physician's guide to laboratory test selection and interpretation. *Journal of the American Medical Association*, 307(22): 2436-2437.
- Mengis, C., Aebi, S., Tobler, A., Dähler, W. & Fey, M. F. (2003). Assessment of differences in patient populations selected for or excluded from participation in clinical phase III acute myelogenous leukemia trials. *Journal of Clinical Oncology*, 21(21): 3933-3939.
- Meshinchi, S. & Arceci, R. J. (2007). Prognostic factors and risk-based therapy in pediatric acute myeloid leukemia. *The Oncologist*, 12(13): 341-355.
- Meyers, S., Downing, J. R. & Hiebert, S. W. (1993). Identification of AML-1 and the (8;21) translocation protein (AML-1/ETO) as sequence-specific DNA-binding proteins: the runt homology domain is required for DNA binding and protein-protein interactions. *Molecular and Cellular Biology*, 13(10): 6336-6345.
- Mills, K., Walsh, V., Gilkes, A., Sweeney, M., Mirza, T., Woodgate, L., Brown, G. & Burnett, A. (2000). High FUS/TLS expression in acute myeloid leukaemia samples. *British Journal of Haematology*, 108(2): 316-321.

- Milne, T. A., Briggs, S. D., Brock, H. W., Martin, M. E., Gibbs, D., Allis, C. D. & Hess, J. L. (2002). MLL targets SET domain methyltransferase activity to Hox gene promoters. *Molecular Cell*, 10(5): 1107-1117.
- Monteoliva, L. & Albar, J. P. (2004). Differential proteomics: an overview of gel and non-gel based approaches. *Briefings in Functional Genomics & Proteomics*, 3(3): 220-239.
- Moorman, A. V., Ensor, H. M., Richards, S. M., Chilton, L., Schwab, C., Kinsey, S. E., Vora, A., Mitchell, C. D. & Harrison, C. J. (2010). Prognostic effect of chromosomal abnormalities in childhood B-cell precursor acute lymphoblastic leukaemia: results from the UK Medical Research Council ALL97/99 randomised trial. *The Lancet Oncology*, 11(5): 429-438.
- Morton, L. M., Wang, S. S., Devesa, S. S., Hartge, P., Weisenburger, D. D. & Linet, M. S. (2006). Lymphoma incidence patterns by WHO subtype in the United States, 1992-2001. *Blood*, 107(1): 265-276.
- Mrózek, K., Döhner, H. & Bloomfield, C. D. (2007). Influence of new molecular prognostic markers in patients with karyotypically normal acute myeloid leukemia: recent advances. *Current Opinions in Hematology*, 14(2): 106-114.
- Mrózek, K., Marcucci, G., Paschka, P. & Bloomfield, C. D. (2008). Advances in molecular genetics and treatment of core-binding factor acute myeloid leukemia. *Current Opinion in Oncology*, 20(6): 711-718.
- Nerlov, C., Querfurth, E., Kulesa, H. & Graf, T. (2000). GATA-1 interacts with the myeloid PU.1 transcription factor and represses PU.1-dependent transcription. *Blood*, 95(8): 2543-2551.
- Ogawa, M. (1993). Differentiation and proliferation of hematopoietic stem cells. *Blood*, 81(11): 2844-2853.
- Ohshiro, K., Rosenthal, D. I., Koomen, J. M., Streckfus, C. F., Chambers, M., Kobayashi, R. & El-Naggar, A. K. (2007). Pre-analytic saliva processing affect proteomic results and biomarker screening of head and neck squamous carcinoma. *International Journal of Oncology*, 30(3): 743-749.
- Okuda, T., Van Deursen, J., Hiebert, S. W., Grosveld, G. & Downing, J. R. (1996). Aml1, the target of multiple chromosomal translocations in human leukemia, is essential for normal fetal liver hematopoiesis. *Cell*, 84(2): 321-330.
- Olson, J. S. 1989. *The History of Cancer: An Annotated Bibliography*. Connecticut: Greenwood Press, Inc.
- Owaidah, T. M., Al Beihany, A., Iqbal, M. A., Elkum, N. & Roberts, G. T. (2006). Cytogenetics, molecular and ultrastructural characteristics of biphenotypic acute leukemia identified by the EGIL scoring system. *Leukemia*, 20(4), 620-626.

- Paietta, E. (2002). Assessing minimal residual disease (MRD) in leukemia: a changing definition and concept? *Bone Marrow Transplantation*, 2(6): 459-465.
- Palmlad M., Tiss A. & Cramer R. (2009) Mass spectrometry in clinical proteomics— from the present to the future. *Proteomics-Clinical Applications*, 3(1): 6-17.
- Pantic, M. Novak, A., Marisavljevic, D., Djordjevic, V., Elezovic, I., Vidovic, A. & Colovic, M. (2000). Additional chromosome aberrations in acute promyelocytic leukemia: characteristics and prognostic influence. *Medical Oncology*, 17(4): 307-313.
- Parkin, D. M., Stiller, C. A., Draper, G. J. & Bieber, C. A. 1988. The international incidence of childhood cancer. *International Journal of Cancer*, 42(4): 511-520.
- Patel, J. P., Gönen, M., Figueroa, M. E., Fernandez, H., Sun, Z., Racevskis, J., Van Vlierberghe, P., Dolgalev, I., Thomas, S., Aminova, O., Huberman, K., Cheng, J., Viale, A., Socci, N. D., Heguy, A., Cherry, A., Vance, G., Higgins, R. R., Ketterling, R. P., Gallagher, R. E., Litzow, M., Van Den Brink, M. R. M., Lazarus, H. M., Rowe, J. M., Luger, S., Ferrando, A., Paietta, E., Tallman, M. S., Melnick, A., Abdel-Wahab, O. & Levine, R. L. (2012). Prognostic relevance of integrated genetic profiling in acute myeloid leukemia. *New England Journal of Medicine*, 366, 1079-1089.
- Peixoto, D., Dingli, D. & Pacheco, J. M. (2011). Modelling hematopoiesis in health and disease. *Mathematical and Computer Modelling*, 53(7-8): 1546-1557.
- Penque D (2009). Two-dimensional gel electrophoresis and mass spectrometry for biomarker discovery. *Proteomics-Clinical Applications* 3: 155-172.
- Perel, Y., Auvrignon, A., Leblanc, T., Michel, G., Reguerre, Y., Vannier, J. P., Dalle, J. H., Gandemer, V., Schmitt, C., Mechinaud, F., Lejars, O., Piguet, C., Couillaud, G., Pautard, B., Landman-Parker, J., Thuret, I., Aladjidi, N., Baruchel, A. & Leverger, G. (2005). Treatment of childhood acute myeloblastic leukemia: dose intensification improves outcome and maintenance therapy is of no benefit - multicenter studies of the French LAME (Leucemie Aigue Myeloblastique Enfant) Cooperative Group. *Leukemia*, 19(12): 2082-2089.
- Pernemalm, M., Lewensohn, R. & Lehtiö, J. (2009). Affinity prefractionation for MS-based plasma proteomics. *Proteomics*, 9(6): 1420-1427.
- Pession, A., Rondelli, R., Basso, G., Rizzari, C., Testi, A. M., Fagioli, F., De Stefano, P. & Locatelli, F. (2005). Treatment and long-term results in children with acute myeloid leukaemia treated according to the AIEOP AML protocols. *Leukemia*, 19(12): 2043-2053.
- Pitkanen, A. & Lukasiuk, K. (2011). Molecular biomarkers of epileptogenesis. *Biomarkers in Medicine*, 5(5), 629-33.

- Polaskova, V., Kapur, A., Khan, A., Molloy, M. P. & Baker, M. S. (2010). High-abundance protein depletion: Comparison of methods for human plasma biomarker discovery. *Electrophoresis*, 31(3), 471-482.
- Pulsoni, A., Pagano, L., Latagliata, R., Casini, M., Cerri, R., Crugnola, M., De Paoli, L., Di Bona, E., Invernizzi, R., Marmont, F., Petti, M., Rigolin, G., Ronco, F., Spadano, A., Tosti, M., Visani, G., Mele, A. & Mandelli, F. (2004). Survival of elderly patients with acute myeloid leukemia. *Haematologica*, 89(3), 296-302.
- Quero, C., Colome, N., Prieto, M. R., Carrascal, M., Posada, M., Gelpí, E. & Abian, J. (2004). Determination of protein markers in human serum: Analysis of protein expression in toxic oil syndrome studies. *Proteomics*, 4(2), 303-315.
- Quintás-Cardama, A., Zhang, N., Qiu, Y. H., Post, S., Creighton, C. J., Cortes, J., Coombes, K. R. & Kornblau, S. M. (2014). Loss of TRIM62 expression is an independent adverse prognostic factor in acute myeloid leukemia. *Clinical Lymphoma Myeloma and Leukemia*, 14(5): 401-410.
- Rabilloud, T. & Lelong, C. (2011). Two-dimensional gel electrophoresis in proteomics: a tutorial. *Proteomics*, 74(10): 1829-1841.
- Radtke, I., Mullighan, C. G., Ishii, M., Su, X., Cheng, J., Ma, J. & Downing, J. R. (2009). Genomic analysis reveals few genetic alterations in pediatric acute myeloid leukemia. *Proceedings of the National Academy of Sciences*, 106(31), 12944-12949.
- Raimondi, S. C. (2006). Cytogenetics of acute leukemias. *Childhood Leukemias*, 2: 235-271.
- Raimondi, S. C., Chang, M. N., Ravindranath, Y., Behm, F. G., Gresik, M. V., Steuber, C. P., Weinstein, H. J. & Carroll, A. J. (1999). Chromosomal abnormalities in 478 children with acute myeloid leukemia: clinical characteristics and treatment outcome in a cooperative pediatric oncology group study—POG 8821. *Blood*, 94(11): 3707-3716.
- Ranganathan, P., Yu, X., Na, C., Santhanam, R., Shacham, S., Kauffman, M., Walker, A., Klisovic, R., Blum, W., Caligiuri, M., Croce, C. M., Marcucci, G. & Garzon, R. (2012). Preclinical activity of a novel CRM1 inhibitor in acute myeloid leukemia. *Blood*, 120(9): 1765-1773.
- Rau, R. & Brown, P. (2009). Nucleophosmin (NPM1) mutations in adult and childhood acute myeloid leukaemia: towards definition of a new leukemia entity. *Hematological Oncology*, 27(4): 171-181.
- Rawstron, A. C., Green, M. J., Kuzmicki, A., Kennedy, B., Fenton, J. A. L., Evans, P. A. S., O'Connor, S. J. M., Richards, S. J., Morgan, G. J., Jack, A. S. & Hillmen, P. (2002). Monoclonal B lymphocytes with the characteristics of “indolent” chronic lymphocytic leukemia are present in 3.5% of adults with normal blood counts. *Blood*, 100(2): 635-639.

- Ravandi, F., Jilani, I., Estey, E., Kantarjian, H., Dey, A., Aguilar, C. & Albitar, M. (2007). Soluble phosphorylated fms-like tyrosine kinase: III. FLT3 protein in patients with acute myeloid leukemia (AML). *Leukemia Research*, 31(6): 791-797.
- Redaelli, A., Laskin, B. L., Stephens, J. M., Botteman, M. F. & Pashos, C. L. (2004). The clinical and epidemiological burden of chronic lymphocytic leukaemia. *European Journal of Cancer Care*, 13(3): 279-287.
- Rees, J. K. H., Gray, R. G. & Wheatley, K. (1996). Dose intensification in acute myeloid leukaemia: greater effectiveness at lower cost. Principal report of the Medical Research Council's ATML9 study. *British journal of haematology*, 94(1), 89-98.
- Reikvam, H., Hatfield, K. J., Kittang, A. O., Hovland, R. & Bruserud, Y. (2011). Acute myeloid leukemia with the t(8;21) translocation: clinical consequences and biological implications. *Journal of Biomedicine and Biotechnology*. 2011: 104631. doi:10.1155/2011/104631.
- Reya, T., Morrison, S. J., Clarke, M. F. & Weissman, I. L. (2001). Stem cells, cancer, and cancer stem cells. *Nature*, 414: 105-111.
- Ries, L. A. G., Eisner, M. P., Kosary, C. L., Hankey, B. F., Miller, B. A., Clegg, L., Mariotto, A., Feuer, E. J. & Edwards, B. K. (2005). SEER Cancer Statistics Review, 1975-2002, based on November 2004 SEER data submission, posted to the SEER web site 2005. Retrieved from: http://seer.cancer.gov/csr/1975_2002/.
- Robison, L. L. (2011). Late effects of acute lymphoblastic leukemia therapy in patients diagnosed at 0-20 years of age. *American Society of Hematology, Education Program*. 2011: 238-242.
- Robison, L. L., Mertens, A. C., Boice, J. D., Breslow, N. E., Donaldson, S. S., Green, D. M., Li, F. P., Meadows, A. T., Mulvihill, J. J. & Neglia, J. P. (2002). Study design and cohort characteristics of the childhood cancer survivor study: A multi-institutional collaborative project. *Medical and Pediatric Oncology*, 38(4): 229-239.
- Roche, S., Tiers, L., Provansal, M., Seveno, M., Piva, M. T., Jouin, P. & Lehmann, S. (2009). Depletion of one, six, twelve or twenty major blood proteins before proteomic analysis: the more the better? *Proteomics*, 72(6): 945-951.
- Rohrbacher, M. & Hasford, J. (2009). Epidemiology of chronic myeloid leukaemia (CML). *Best Practice & Research Clinical Haematology*, 22(3): 295-302.
- Rossi, L., Serafini, S., Pierigé, F., Antonelli, A., Cerasi, A., Fraternali, A., Chiarantini, L. & Magnani, M. (2005). Erythrocyte-based drug delivery. *Expert Opinion on Drug Delivery*, 2(2): 311-322.

- Rowe, J. M. (2009). Optimal induction and post-remission therapy for AML in first remission. *American Society of Hematology Education Program Book, 2009*, 2009(1), 396-405.
- Rowley, J. D. 1973. Identificaton of a translocation with quinacrine fluorescence in a patient with acute leukemia. *Annales de Genetique*, 16: 109-112.
- Rubnitz, J. E., Gibson, B. & Smith, F. O. (2010). Acute myeloid leukemia. *Hematology/oncology clinics of North America*, 24(1), 35-63.
- Sanderson, R. N., Johnson, P. R. E., Moorman, A. V., Roman, E., Willett, E., Taylor, P. R., Proctor, S. J., Bown, N., Ogston, S. & Bowen, D. T. (2006). Population-based demographic study of karyotypes in 1709 patients with adult acute myeloid leukemia. *Leukemia*, 20(3): 444-450.
- Saha, S., Banerjee, S., Banerjee, D., Chandra, S. & Chakrabarti, A. (2014). 2DGE and DIGE based proteomic study of malignant B-cells in B-cell acute lymphoblastic leukemia. *European Proteomic Association Open Proteomics*, 3: 13-26.
- Sandler, D. P. & Ross, J. A. (1997). Epidemiology of acute leukemia in children and adults. *Seminars in Oncology* 24(1): 3-16.
- Saslow, D., Solomon, D., Lawson, H. W., Killackey, M., Kulasingam, S. L., Cain, J., Garcia, F. A., Moriarty, A. T., Waxman, A. G. & Wilbur, D. C. (2012). American Cancer Society, American Society for Colposcopy and Cervical Pathology, and American Society for Clinical Pathology screening guidelines for the prevention and early detection of cervical cancer. *CA: A Cancer Journal for Clinicians*, 62(3): 147-172.
- Sassone-Corsi, P. (2012). Minireview: NAD⁺, a circadian metabolite with an epigenetic twist. *Endocrinology*, 153(1): 1-5.
- Schlenk, R. F., Döhner, K., Krauter, J., Fröhling, S., Corbacioglu, A., Bullinger, L., Habdank, M., Späth, D., Morgan, M. & Benner, A. (2008). Mutations and treatment outcome in cytogenetically normal acute myeloid leukemia. *New England Journal of Medicine*, 358(18): 1909-1918.
- Scott, A. S. & Fong, E. (2004). *Body Structures and Functions*, New York: Delmar Learning.
- Seam, N., Gonzales, D. A., Kern, S. J., Hortin, G. L., Hoehn, G. T. & Suffredini, A. F. (2007). Quality control of serum albumin depletion for proteomic analysis. *Clinical Chemistry*, 53(11): 1915-1920.
- Sekeres, M. A., Stone, R. M., Zahrieh, D., Neuberg, D., Morrison, V., De Angelo, D. J., Galinsky, I. & Lee, S. J. (2004). Decision-making and quality of life in older adults with acute myeloid leukemia or advanced myelodysplastic syndrome. *Leukemia*, 18(4): 809-816.

- Seul, K. J., Cho, H. S. & Ghim, S. Y. (2011). Characterization of a PyrR-deficient mutant of *Bacillus subtilis* by a proteomic approach. *Korean Journal of Microbiology and Biotechnology*, 39: 9-19.
- Sgambati, M. T., Linet, M. S. & Devesa, S. S. (2001). Chronic Lymphocytic Leukemia. Epidemiological, Familial, and Genetic Aspects. In B. D. Cheson (Ed.), *Chronic Lymphoid Leukemias* (pp. 33-62). New York: Marcel Dekker.
- Shaffer L. G., Slovak, M. L. & Campbell, L. G. (2013) *ISCN 2009: An International System for Human Cytogenetic Nomenclature (2013)*. Basel, Switzerland: S. Karger.
- Sherwood, L. (2010). *Human Physiology: From Cells to Systems*. Belmont: Cengage Learning.
- Shipley, J. L. & Butera, J. N. (2009). Acute myelogenous leukemia. *Experimental Hematology*, 37(6): 649-658.
- Siegel, R., Ma, J., Zou, Z. & Jemal, A. (2014). Cancer statistics, (2014). *CA: A Cancer Journal for Clinicians*, 64(1): 9-29.
- Slovak, M. L., Kopecky, K. J., Cassileth, P. A., Harrington, D. H., Theil, K. S., Mohamed, A., Paietta, E., Willman, C. L., Head, D. R., Rowe, J. M., Forman, S. J. & Appelbaum, F. R. (2000). Karyotypic analysis predicts outcome of preremission and postremission therapy in adult acute myeloid leukemia: a Southwest Oncology Group/Eastern Cooperative Oncology Group study. *Blood*, 96(13): 4075-4083.
- Smith M., Barnett M., Bassan R., Gatta G., Tondini C. & Kern W. (2004). Adult acute myeloid leukaemia. *Critical Reviews in Oncology/Hematology*, 50(3): 197-222.
- Smith, M. A., Ries, L. A. G., Gurney, J. G. & Ross, J. A. (1999). Leukemia. In L. A. G. Ries, M. A. Smith, J. G. Gurney, M. Linet, T. Tamra, J. L. Young, & G. R. Bunin (Eds.), *Cancer Incidence and Survival among Children and Adolescents: United States SEER Program 1975-1995* (pp. 17-31). Bethesda: National Cancer Institute, SEER Program.
- Swerdlow, S. H., Campo, E. & Harris, N. L. (2008). *WHO classification of tumours of haematopoietic and lymphoid tissues*. France: IARC Press.
- Tamahkar, E., Babaç, C., Kutsal, T., Pişkin, E. & Denizli, A. (2010). Bacterial cellulose nanofibers for albumin depletion from human serum. *Process Biochemistry*, 45(10): 1713-1719.
- Tarlock, K. & Meshinchi, S. (2014). Pediatric acute myeloid leukemia: Biology and therapeutic implications of genomic variants. *Pediatric Clinics of North America*. 62(1): 75-93.
- Tenen, D. G. (2003). Disruption of differentiation in human cancer: AML shows the way. *Nature Reviews Cancer*, 3(2): 89-101.

- Tenen, D. G., Hromas, R., Licht, J. D. & Zhang, D. E. (1997). Transcription factors, normal myeloid development, and leukemia. *Blood*, 90(2): 489-519.
- Thadikaran, L., Siegenthaler, M. A., Crettaz, D., Queloz, P. A., Schneider, P. & Tissot, J. D. (2005). Recent advances in blood-related proteomics. *Proteomics*, 5(12): 3019-3034.
- Thiel, A. T., Huang, J., Lei, M. & Hua, X. (2012). Menin as a hub controlling mixed lineage leukemia. *BioEssays*, 34(9): 771-780.
- Tomlins, S. A., Rhodes, D. R., Perner, S., Dhanasekaran, S. M., Mehra, R., Sun, X. W., Varambally, S., Cao, X., Tchinda, J., Kuefer, R., Lee, C., Montie, J. E., Shah, R. B., Pienta, K. J., Rubin, M. A. & Chinnaiyan, A. M. (2005). Recurrent fusion of TMPRSS2 and ETS transcription factor genes in prostate cancer. *Science*, 310(5748): 644-648.
- Torotra, G. J. & Grabowski, S. R. (1996). The Cardiovascular System: The Blood. In: G. J. Torotra, & S. R. Grabowski (Eds.), *Principles of Anatomy and Physiology* (598-605). New York: Harper Collins College.
- Twyman, R. M. (2004). SNP discovery and typing technologies for pharmacogenomics. *Current Topics in Medicinal Chemistry*, 4(13): 1423-31.
- Valk, P. J., Delwel, R. & Lowenberg, B. (2005). Gene expression profiling in acute myeloid leukemia. *Current Opinion in Hematology*, 12(1): 76-81.
- Valk, P. J. M., Verhaak, R. G. W., Beijten, M. A., Erpelinck, C. A. J., Van Doorn-Khosrovani, S. B. V. W., Boer, J. M., Beverloo, H. B., Moorhouse, M. J., Van Der Spek, P. J., Löwenberg, B. & Delwel, R. (2004). Prognostically useful gene-expression profiles in acute myeloid leukemia. *New England Journal of Medicine*, 350(16): 1617-1628.
- Vardiman, J. W., Harris, N. L. & Brunning, R. D. (2002). The World Health Organization (WHO) classification of the myeloid neoplasms. *Blood*, 100(7): 2292-2302.
- Vardiman, J. W., Thiele, J., Arber, D. A., Brunning, R. D., Borowitz, M. J., Porwit, A., Harris, N. L., Le Beau, M. M., Hellström-Lindberg, E., Tefferi, A. & Bloomfield, C. D. (2009). The 2008 revision of the World Health Organization (WHO) classification of myeloid neoplasms and acute leukemia: rationale and important changes. *Blood*, 114(5): 937-951.
- Varnum, S. M., Covington, C. C., Woodbury, R. L., Petritis, K., Kangas, L. J., Abdullah, M. S., Pounds, J. G., Smith, R. D. & Zangar, R. C. (2003). Proteomic characterization of nipple aspirate fluid: identification of potential biomarkers of breast cancer. *Breast Cancer Research and Treatment*, 80(1): 87-97.
- Verrills, N. M. (2006). Clinical proteomics: present and future prospects. *Clinical Biochemist Reviews*, 27(2), 99.

- Visanji, N. P., Wong, J. C., Wang, S. X., Cappel, B., Kleinschmidt-Demasters, B. K., Handler, M. H., Ochi, A., Otsubo, H., Rutka, J. T., Go, C., Weiss, S., Vinters, H. V., Hawkins, C. E., Desouza, L. V., Siu, K. W. & Hazrati, L. N. (2012). A proteomic analysis of pediatric seizure cases associated with astrocytic inclusions. *Epilepsia*, 53(3): e50-4.
- Wahlin, A., Hörnsten, P. & Jonsson, H. (1991). Remission rate and survival in acute myeloid leukemia: Impact of selection and chemotherapy. *European Journal of Haematology*, 46(4): 240-247.
- Walter, R. B., Kantarjian, H. M., Huang, X., Pierce, S. A., Sun, Z., Gundacker, H. M., Ravandi, F., Faderl, S. H., Tallman, M. S., Appelbaum, F. R. & Estey, E. H. (2010). Effect of complete remission and responses less than complete remission on survival in acute myeloid leukemia: a combined Eastern Cooperative Oncology Group, Southwest Oncology Group, and M. D. Anderson Cancer Center Study. *Journal of Clinical Oncology*, 28(10): 1766-1771.
- Walter, M. J., Payton, J. E., Ries, R. E., Shannon, W. D., Deshmukh, H., Zhao, Y. & Ley, T. J. (2009). Acquired copy number alterations in adult acute myeloid leukemia genomes. *Proceedings of the National Academy of Sciences*, 106(31): 12950-12955.
- Wang, J., Iwasaki, H., Krivtsov, A., Febbo, P. G., Thorner, A. R., Ernst, P., Anastasiadou, E., Kutok, J. L., Kogan, S. C., Zinkel, S. S., Fisher, J. K., Hess, J. L., Golub, T. R., Armstrong, S. A., Akashi, K. & Korsmeyer, S. J. (2005). Conditional MLL-CBP targets GMP and models therapy-related myeloproliferative disease. *The EMBO Journal*, 24(2): 368-381.
- Wang, Q. F., Cleaves, R., Kummalue, T., Nerlov, C. & Friedman, A. D. (2003). Cell cycle inhibition mediated by the outer surface of the C/EBP[alpha] basic region is required but not sufficient for granulopoiesis. *Oncogene*, 22(17): 2548-2557.
- Wang, S., Wang, Q., Crute, B. E., Melnikova, I. N., Keller, S. R. & Speck, N. A. (1993). Cloning and characterization of subunits of the T-cell receptor and murine leukemia virus enhancer core-binding factor. *Molecular and Cellular Biology*, 13(6): 3324-3339.
- Wartenberg, D., Groves, F. D. & Adelman, A. S. (2008). Acute lymphoblastic leukemia: Epidemiology and etiology. *Acute Leukemias* (pp.77-93). Springer Berlin Heidelberg.
- Wheatley, K., Burnett, A. K., Goldstone, A. H., Gray, R., Hann, I., Harrison, C., Rees, J., Stevens, R. & Walker, H. (1999). A simple, robust, validated and highly predictive index for the determination of risk-directed therapy in acute myeloid leukaemia derived from the MRC AML 10 trial. United Kingdom

- Medical Research Council's Adult and Childhood Leukaemia Working Parties. *British Journal of Haematology* 107(1): 69-79.
- White, C. N., Chan, D. W. & Zhang, Z. (2004). Bioinformatics strategies for proteomic profiling. *Clinical Biochemistry*, 37(7), 636-641.
- Whitman, S. P., Archer, K. J., Feng, L., Baldus, C., Becknell, B., Carlson, B. D., Carroll, A. J., Mrózek, K., Vardiman, J. W. & George, S. L. (2001). Absence of the wild-type allele predicts high prognosis in adult de novo acute myeloid leukemia with normal cytogenetics and the internal tandem duplication of FLT3 A cancer and leukemia group B study. *Cancer Research*, 61(19): 7233-7239.
- Widjaja, I., Naumann, K., Roth, U., Wolf, N., Mackey, D., Dangl, J. L., Scheel, D. & Lee, J. (2009). Combining subproteome enrichment and Rubisco depletion enables identification of low abundance proteins differentially regulated during plant defense. *Proteomics*, 9(1): 138-147.
- Wiemels, J. L., Xiao, Z., Buffler, P. A., Maia, A. T., Ma, X., Dicks, B. M., Smith, M. T., Zhang, L., Feusner, J., Wiencke, J., Pritchard-Jones, K., Kempfski, H. & Greaves, M. (2002). In utero origin of t(8;21) AML1-ETO translocations in childhood acute myeloid leukemia. *Blood*, 99(10): 3801-3805.
- Wilson, C. S., Davidson, G. S., Martin, S. B., Andries, E., Potter, J., Harvey, R., Ar, K., Xu, Y., Kopecky, K. J., Ankerst, D. P., Gundacker, H., Slovak, M. L., Mosquera-Caro, M., Chen, I. M., Stirewalt, D. L., Murphy, M., Schultz, F. A., Kang, H., Wang, X., Radich, J. P., Appelbaum, F. R., Atlas, S. R., Godwin, J. & Willman, C. L. (2006). Gene expression profiling of adult acute myeloid leukemia identifies novel biologic clusters for risk classification and outcome prediction. *Blood*, 108(2): 685-96.
- Wingo, P. A., Cardinez, C. J., Landis, S. H., Greenlee, R. T., Ries, L. A. G., Anderson, R. N. & Thun, M. J. (2003). Long-term trends in cancer mortality in the United States, 1930–1998. *Cancer*, 97(12): 3133-3275.
- Xie, Y., Davies, S. M., Xiang, Y., Robison, L. L. & Ross, J. A. (2003). Trends in leukemia incidence and survival in the United States (1973–1998). *Cancer*, 97(9): 2229-2235.
- Xuan, J., Yu, Y., Qing, T., Guo, L. & Shi, L. (2013). Next-generation sequencing in the clinic: promises and challenges. *Cancer Letters*, 340(2): 284-295.
- Yeates, K. O., Ris, M. D., Taylor, H. G. & Pennington, B. F. (2009). *Pediatric Neuropsychology: Research, Theory, and Practice*. New York: Guilford Press.
- Yergeau, D. A., Hetherington, C. J., Wang, Q., Zhang, P., Sharpe, A. H., Binder, M., Marin-Padilla, M., Tenen, D. G., Speck, N. A. & Zhang, D. E. (1997). Embryonic lethality and impairment of haematopoiesis in mice heterozygous for an AML1-ETO fusion gene. *Nature Genetics* 15(3): 303-306.

- Yunis, J. (1983). The chromosomal basis of human neoplasia. *Science*, 221(4607): 227-236.
- Zainal Ariffin, O. & Nor Saleha, I. (2011). *National Cancer Registry Report: Malaysia Cancer Statistics-Data and Figure (2007)*. Putrajaya: Ministry of Health Malaysia.
- Zhang, D. E., Hetherington, C. J., Meyers, S., Rhoades, K. L., Larson, C. J., Chen, H. M., Hiebert, S. W. & Tenen, D. G. (1996). CCAAT enhancer-binding protein (C/EBP) and AML1 (CBF alpha2) synergistically activate the macrophage colony-stimulating factor receptor promoter. *Molecular and Cellular Biology*, 16(3): 1231-40.
- Zhang, L., Wen, Q., Mao, H. P., Luo, N., Rong, R., Fan, J. J. & Yu, X. Q. (2013). Developing a reproducible method for the high-resolution separation of peritoneal dialysate proteins on 2-D gels. *Protein Expression and Purification*, 89(2): 196-202.
- Zhou M., Conrads T. P. & Veenstra T. D. (2005) Proteomics approaches to biomarker detection. *Briefings in Functional Genomics and Proteomics*, 4(1): 69-75.
- Zhu, K., Yan, F., O'neil, K. A., Hamler, R., Lubman, D. M., Lin, L. & Barder, T. J. (2004). Proteomic analysis using 2-d liquid separations of intact proteins from whole-cell lysates. *Current Protocols in Protein Science*, 23: 23.3. 1-23.3. 28.
- Zolg, J. W. & Langen, H. (2004). How industry is approaching the search for new diagnostic markers and biomarkers. *Molecular and Cellular Proteomics*, 3(4): 345-354.