Clinical significance of minimal residual disease in childhood acute lymphoblastic leukemia and its relationship to other prognostic factors

THESIS

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بسم الله الرحمن الرحيم

و فوق كل ذى علم عليم

صدق الله العظيم

سورة يوسف الآية رقم: (٧٦)

To my mother and father

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AIDS:	Acquired immunodeficiency syndrome
ALL:	Acute lymphoblastic leukemia
AML:	Acute myeloid leukemia
ANC:	Absolute Neutrophilic Count
B-ALL:	B cell Acute Lymphoblastic Leukemia
BCR:	Breakpoint Cluster Region
BFM:	Berlin-Frankfurt-Muenster
BM:	Bone Marrow
BMT:	Bone Marrow Transplantation
CD:	Cluster of Differentiation
CIBMTR	Center for International Blood and Marrow Transplant Research
CML:	Chronic Myeloid Leukemia
CNS:	Central Nervous System
COG:	Children oncology group
CR:	Complete Remission
CSF:	Cerebrospinal Fluid
CT:	Computed Tomography
DFCI:	Dana-Farber Cancer Institute
DFS:	Disease Free Survival
DNA:	Deoxyribonucleic Acid
EBV:	Epstein-Barr virus
EFS:	Event Free Survival
FAB:	French, American, British classification
FCM:	Flow cytometry
FISH:	fluorescence in situ hybridization
FSC:	Forward scatter
GST:	glutathione S-transferase

GVHD: Graft Versus Host Disease
GVL: Graft Versus Leukemia
HDAC: High Dose Ara-C
HDMTX: High Dose Methotrexate
HIV: human immunodeficiency virus
HR: High Risk
HLA: human lymphocyte antigen
HSCT: Hematopoietic Stem Cell Transplantation
HTLV: Human lymphotropic viruses
Ig: Immunoglobulin
IM: Intramuscular
IT: Intrathecal
I.V: Intravenous
LCR: Ligase Chain Reaction
MLL: Mixed lineage leukemia
MALT: mucosa-associated lymphoid tumors
MP: mercaptopurine
MRC: Medical Research Council
LDH: Lactate Dehydrogenase
MoAB: Monoclonal Antibodies
MRD: Minimal Residual Disease
MRI: Magnetic Resonance Imaging
MTX: Methotrexate
NK: natural killer
OS: Overall survival
PAS: Periodic Acid Schiff
PB: Peripheral Blood
PCR: Polymerase Chain Reaction
Ph: Philadelphia Chromosome

POG:	Pediatric oncology group
PR:	Partial Remission
RNA:	Ribonucleic acid
RQ-PCR:	Real time Quantitative polymerase Chain Reaction
SCT:	Stem Cell Transplantation
SKY:	spectral karyotyping
SR:	Standard Risk
SSC:	Sideward scatter
SJCRH	St Jude Children Research hospital
T-ALL:	T cell Acute Lymphoblastic Leukemia
TBI:	Total Body Irradiation
TCR:	T – Cell Receptors
TdT:	Terminal Deoxynucleotidyl Transferase
TG:	thioguanine
TLC:	Total Leucocytic Count
TPMT:	Thiopurine methyl transferase
WBC:	White Blood Cells
WHO:	Word Health Organization
WT1:	Wilms tumor 1

ABSTRACT

BACKGROUND :ALL is the most common cancer diagnosed in children and represents 23 %of cancer diagnoses among children younger than 15 years . Better understanding of the characteristics of leukemic cells have led to novel ways of detecting morphologically occult leukemic cells)i.e., minimal residual disease, MRD (by detecting abnormalities in protein expression detectable by flow cytometry .MRD is the presence of cells following chemotherapy below the level of morphologic detection .Flow cytometric techniques which assess MRD should achieve a sensitivity of at least 1/10000 leukemic cells in bone marrow) 0.01 .(% MRD has emerged as the most powerful tool for assessing response to chemotherapy, and MRD positivity has been demonstrated to be the strongest independent risk factor for relapse among other clinical and biological risk factors . Detection of leukemia associated phenotypes via flow cytometry is applicable in almost all cases, fast, and relatively inexpensive.

PURPOSE : The aim of this study was to determine if the presence of minimal residual disease by flow cytometry at bone marrow of cases with acute lymphoblastic leukemia in children during treatment had any correlation with various clinical and biological risk factors such as :age, gender, initial TLC, initial CSF, IPT, DNA index, cytogenetics and the effect of minimal residual disease on outcome of ALL patients regarding relapse free survival and overall survival.

PATIENTS AND METHODS : Between the periods from January 2009 to February 2010, seventy five children who presented to the pediatric oncology outpatient clinic, National Cancer Institute, Cairo University, were enrolled in the study .Patients were 46 males)61.3 (%and 29 females)38.7 (%with a male :female ratio 1.58:1 .Their ages ranged from 4 months to 18 years with mean age of 8.5

years .All patients received therapy according to the St .Jude Total XV protocol modified .They were evaluated for response to treatment by minimal residual disease at bone marrow at day 15 and at end of induction on day 42 .

RESULTS : Twenty four patients were MRD positive at bone marrow at day 15 and 51 patients were MRD negative .It was significant correlation that the 18 months relapse free survivals for these patients were 67.4 %for positive patients and 86.7 %for negative patients)P value =0.034.(quantification of the level of MRD positivity was also of significant value as patients whose level was between 10^{-4} to 10^{-3} had RFS of 73.2 %while patients whose level was above 10^{-3} had RFS of 53.6) %P value =0.043 .(

At day 42 MRD, 18 cases were positive and 57 cases were negative and it was highly significant correlation that the 18 months RFS of these two groups were 57.8 %and 88 %respectively)P value =0.002 .(MRD was studied in relation to the common clinical and biological risk factors of ALL and it was found that MRD post induction was frequently present with unfavorable risk criteria but all of these were of low significant value except for initial TLC, DNA index)P value =0.032, 0.006 respectively .(Post induction MRD was an independent prognostic factor for ALL patients regarding RFS and OS.

CONCLUSIONS : The presence of minimal residual disease at day 15 and post induction at day 42 can affect the outcome of patients with ALL regarding RFS and OS and its presence is frequently found in patients with adverse clinical and biological factors but it was revealed that MRD post induction was an independent prognostic factors among these factors to detect the outcome of patients.

Key words :ALL -minimal residual disease -flow cytometry

Introduction