

بسم الله الرحمن الرحيم



شبكة المعلومات الجامعية التوثيق الالكتروني والميكروفيلم



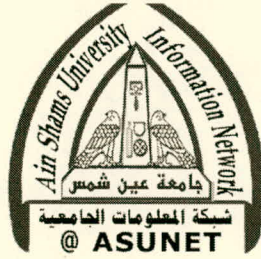
شبكة المعلومات الجامعية

جامعة عين شمس

التوثيق الالكتروني والميكرو فيلم

قسم

نقسم بالله العظيم أن المادة التي تم توثيقها وتسجيلها
علي هذه الأفلام قد أعدت دون أية تغيرات



يجب أن

تحفظ هذه الأفلام بعيدا عن الغبار

في درجة حرارة من ١٥-٢٥ مئوية ورطوبة نسبية من ٢٠-٤٠%

To be Kept away from Dust in Dry Cool place of
15-25- c and relative humidity 20-40%

بعض الوثائق الأصلية تالفة

بالرسالة صفحات لم ترد بالاصل

**THE USE OF FLUORESCENCE IN-SITU
HYBRIDIZATION (FISH) IN DETECTION OF
CHROMOSOME 4 ANEUPLOIDY IN PATIENTS WITH
ACUTE LYMPHOBLASTIC LEUKEMIA (AN
IMPORTANT PROGNOSTIC FACTOR)**

Thesis

Submitted for Partial Fulfillment of Master Degree
of Clinical and Chemical Pathology

By

Nevine Mohamed Bahaa El-Din Fouad

M.B., B.Ch.

Supervised By

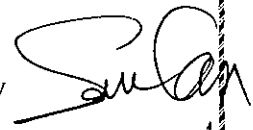
Prof. Dr. Azza Ahmed Mohamed

Professor of Clinical and Chemical Pathology
Cairo University



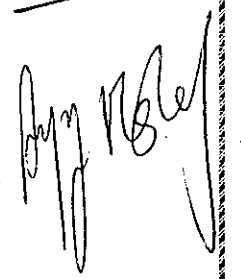
Prof. Dr. Mervat El-Ansary

Professor of Clinical and Chemical Pathology
Cairo University



Dr. Sherine Mahmoud Shawki

Lecturer of Clinical and Chemical Pathology
Cairo University



**Faculty of Medicine
Cairo University
2001**



الإدارة العامة للدراسات العليا والبحوث

قسم الامتحانات

السيد مدير إدارة كلية
معهد

الدراسات العليا

تحت طيبة وبعد . . .

٥ / ٢ / ١١٥

٢٢٦
١ / ٢٢

بالإشارة لكتابكم رقم ٢٦ في ١ / ٢٢ / ١١٥ بشأن موافقة مجلس كلية
معهد في ٢١ / ١٢ / ١٤٠٠ على تشكيل لجان الحكم للطلاب الآتية أسماؤهم :

مسل	الاسم	الدرجة	التخصص
١ -	نعمية سيد مزارا	ماجستير	بنا لربنا (العلم والكمبيوتر) أكتوبر ٩٨
٢ -	رانيا ناجي	~	~ ~ ~ ~ ~ ٩٧
٣ -	مصطفى النور	~	إدارة شؤون كليات ~ ٩٨
٤ -	كريم عبد الحليم	~	إدارة شؤون ~ ~ ~
٥ -			
٦ -			
٧ -			
٨ -			
٩ -			
١٠ -			

ننتهي اليكم انه بعرض الموضوع على د. د. نائب رئيس الجامعة لشئون الدراسات
العليا والبحوث اشر سيادته في ٢٥ / ١ / ٢٠١١م بالموافقة على اقتراح الكلية
المعهد وتفضلوا بقبول فائق الاحترام ،،

مدير إدارة
الدراسات العليا

٢٢٦
١ / ٢٢

محضر

اجتماع لجنة الحكم على الرسالة المقدمة من
الطبيب / د. محمد السيد فؤاد
توطئة للحصول على درجة الماجستير / الدكتورة
في الباثولوجيا الباطنية والكلى

تحت عنوان : باللغة الانجليزية : The use of fluorescence in-situ hybridization (FISH) in detection of chromosome & aneuploidy in patients with acute lymphoblastic leukemia
: باللغة العربية : استعمال طريقة التبريد بالاشعاع الذاتى لدراسة الكروموسومات في مرضى سرطان الدم الليمفوي الحاد

بناءً على موافقة الجامعة بتاريخ ٢٥ / ١ / ٢٠٠١ تم تشكيل لجنة الفحص والمناقشة للرسالة المذكورة أعلاه على النحو التالي :-

- (١) الأستاذ الدكتور / عزت أحمد أحمد عن المشرفين
 - (٢) الأستاذ الدكتور / عزت محمد محمد الراسم
 - (٣) الأستاذ الدكتور / محمود محمد عن المحصلين في الاسم
- بعد فحص الرسالة بواسطة كل عضو منفردا وكتابة تقارير منفردة لكل منهم لعائلات اللجنة مجتمعة فسي
يتم السبت بتاريخ ٢١ / ٢ / ٢٠٠١ - ١١ / ٢ / ٢٠٠١ بقرعة مدعى "ب"

بكلية الطب - جامعة القاهرة وذلك لمناقشة الطالب في جلسة علنية في موضوع الرسالة والنتائج التي توصل
إليها وكذلك الأعمال العملية التي قام عليها البحث .

قرار اللجنة : قبول الرسالة والسماح للأستاذ محمد السيد فؤاد

توقيعات أعضاء اللجنة :-

المشرف المتحن

المتحن الداخلي

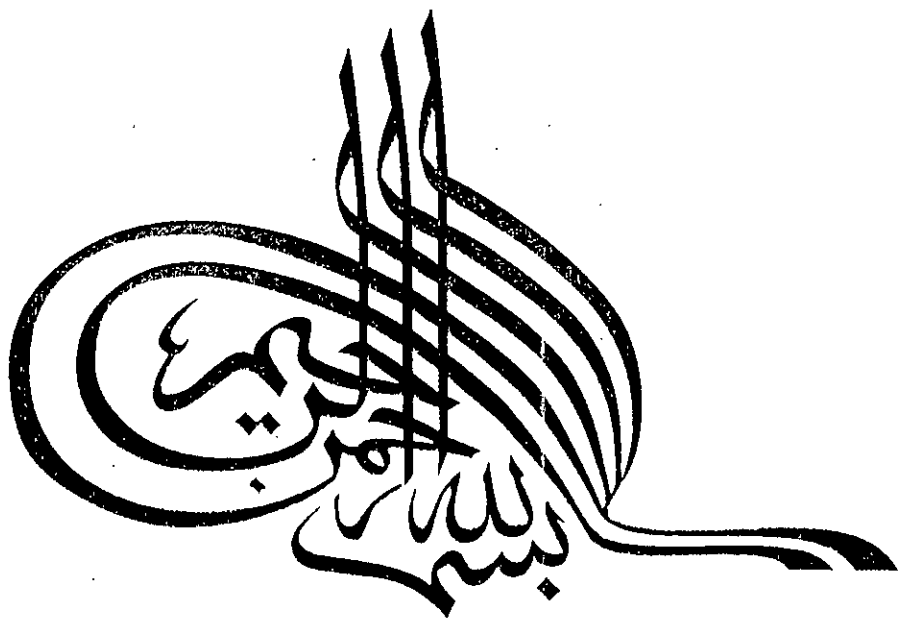
المتحن الخارجي

محمد السيد فؤاد

محمد السيد فؤاد

محمد السيد فؤاد

(صام) ٢١ / ٢ / ٢٠٠١



ABSTRACT

ALL is the commonest form of childhood cancer and represents clonal proliferation of transformed hemopoietic cells as a result of genetic changes.

Chromosome abnormalities are an important prognostic factor in childhood ALL. Recently, a subset of patients with hyperdiploid ALL and trisomy 4 has been reported to have a very favorable event-free survival and so, they can be treated with less toxic antimetabolite therapy.

In the current study chromosome 4 detection by FISH technique was done on 20 children suffering from ALL and matched with 10 normal children.

Trisomy 4 was detected in 11 patients (55%) at different frequencies ranging from 7 to 57%. Trisomy 4 was significantly higher in L1 cases when compared to L2 cases. It was also higher in CR patients when compared to relapsed patients.

Thus, FISH represents a simple, rapid, sensitive and specific technique that can be used for detection of chromosomal abnormalities in interphase nuclei as well as metaphase spreads with a high success rate. It can be used at diagnosis, during the course of the disease and is also useful to monitor patients after therapy.

Key words : ALL , FISH , Chromosome 4 .

ACKNOWLEDGMENT

I would like to express my deepest gratitude and respect to *Prof. Dr. Azza Ahmed Mohamed*, Professor of Clinical and Chemical pathology, Cairo University, for her generous help and vast experience she offered me unlimitedly. Her kindness, patience and support makes her so much loved by everyone.

My deepest appreciation and admiration to *Prof. Dr. Mervat El-Ansary*, Professor of Clinical and Chemical Pathology, Cairo University, under whose supervision I had the honor and pleasure to proceed with this work. Her constant guidance, meticulous supervision, fruitful remarks and technical experience she offered me, make me so indebted to her. She is by all means such a wonderful person whom I think so highly of.

I would like also to express my greatest thanks to *Dr. Shereen Mahmoud Shawki*, Lecturer of Clinical and Chemical pathology, Cairo University, for her great assistance, sincere guidance and precious time she offered me throughout this work.

Special thanks to all my professors and colleagues for their great help and support.

Also, I wish to thank my patients for their cooperation without which this work could not be possible.

And last, but by no means least, I would like to thank my friends, my family and especially my husband for all their patience, love, support and sacrifice which made this work possible.

LIST OF ABBREVIATIONS

ADA:	Adenine deaminase
ALL:	Acute lymphoblastic leukemia
AML:	Acute myeloid leukemia
ANLL:	Acute non-lymphoblastic leukemia
ATL:	Adult T-cell leukemia
BM:	Bone marrow
BMT:	Bone marrow transplantation
CALLA:	Common acute lymphoblastic leukemia antigen
CC:	Conventional cytogenetics
CCT:	Conventional cytogenetic technique
CD:	Cluster of differentiation
CGH:	Comparative genomic hybridization
CLL:	Chronic lymphatic leukemia
CML:	Chronic myeloid leukemia
CNS:	Central nervous system
CR:	Complete remission
CSF:	Cerebrospinal fluid
del:	Deletion
DNA:	Deoxy ribonucleic acid
EBV:	Epstein Barr virus
EDTA:	Ethylene diamine tetra-acetic acid
EFS:	Event-free survival
FAB:	French American British group
FCM:	Flow cytometry
FISH:	Fluorescence in-situ hybridization
HTLV-I:	Human T-cell leukemia virus type I
HTLV-II:	Human T-cell leukemia virus type II
idic:	Isodicentric chromosome
Ig:	Immunoglobulin
ISCN:	International System for Human Cytogenetic Nomenclature
LDH:	Lactic dehydrogenase
MDS:	Myelo-dysplastic syndrome
MRD:	Minimal residual disease
NHL:	Non-Hodgkin's lymphoma
PB:	Peripheral blood
PCR:	Polymerase chain reaction
Ph:	Philadelphia chromosome
PWS:	Prader-Willi syndrome
RT-PCR:	Reverse transcription polymerase chain reaction
SKY:	Spectral karyotyping
STS:	Sequence tagged sites
TCR:	T-cell receptor
TLC:	Total leucocytic count
WBCs:	White blood cells
WHO:	World Health Organization
YAC:	Yeast artificial chromosome

LIST OF TABLES

	<i>Page</i>
Table (1): Major immunologic subtypes of acute lymphoblastic leukemia and associated prognosis.....	8
Table (2): Selected karyotypic changes in acute lymphoblastic leukemia and associated features.....	9
Table (3): Prognostic factors in acute lymphoblastic leukemia ...	16
Table (4): Total therapy regimen for ALL.....	22
Table (5): Clinical data of ALL patients.....	80
Table (6): Haematological data in studied groups	80
Table (7): Frequency of ALL cases according to FAB classification	83
Table (8): Immunophenotypic types of ALL cases	83
Table (9): Results of FISH for chromosome 4 in the control group	86
Table (10): Chromosome 4 aberrations by FISH in ALL cases	88
Table (11): Comparison of FISH results between patients and controls	88
Table (12): Comparison between haematological data and FISH results in L1 and L2 cases.....	89
Table (13): Comparison between CR and relapsed ALL patients...	91

LIST OF FIGURES

	<i>Page</i>
Figure (1): Stages of mitosis.	25
Figure (2): Stages of cell cycle.	25
Figure (3): Structure of the chromosome.	27
Figure (4): A normal G-banded male karyotype.	29
Figure (5): Types of translocation.	34
Figure (6): Partial karyotype showing a 14q21q Robertsonian translocation.	35
Figure (7): A pericentric (A) and a paracentric (B) inversion	36
Figure (8): Partial karyotype showing a ring chromosome 18.	37
Figure (9): Clinical data of ALL patients.	81
Figure (10): Haematological data of ALL patients.	82
Figure (11): Morphological types of ALL cases.	84
Figure (12): Immunophenotypic types of ALL cases.	85
Figure (13): Trisomy percent in controls and ALL patients.	90
Figure (14): Comparison between trisomy 4% in L1, L2 and L3 cases	92
Figure (15): Comparison between trisomy 4% in CR and relapsed patients.	93
Figure (16): Regression analysis showing correlation between 3 spots and TLC among CR patients.	94
Figure (17): Regression analysis showing correlation between 3 spots and TLC among relapsed patients	95
Figure (18): Interphase FISH for chromosome 4 in controls	96
Figure (19): Interphase FISH in a patient of trisomy chromosome 4	96
Figure (20): Chromosome 4 showing trisomy and disomy	97
Figure (21): Chromosome 4 showing trisomy and disomy	97
Figure (22): Chromosome 4 Showing monosomy	98
Figure (23): An example of diffuse signal	98

CONTENTS

	<i>Page</i>
List of Abbreviations	i
List of Tables	ii
List of Figures.....	iii
INTRODUCTION AND AIM OF THE WORK.....	1
REVIEW OF LITERATURE	
I-ACUTE LYMPHOBLASTIC LEUKEMIA.....	3
•Definition.....	3
•Epidemiology.....	3
•Etiology	4
•Classification	6
•Clinical Picture.....	11
•Laboratory Features	14
•Prognosis	15
•Treatment.....	21
II-CHROMOSOME IDENTIFICATION AND ANALYSIS	23
•Human Chromosomes.....	23
•The Cell Cycle	24
•Structure of Chromosomes.....	26
•Techniques of Chromosome Analysis	27
•Nomenclature For Normal and Abnormal Karyotypes	31
•Types of Chromosomal Abnormalities.....	32
<i>A-Numerical chromosomal abnormalitie.....</i>	<i>32</i>
<i>B-Structural chromosomal abnormalities.....</i>	<i>33</i>
III-CYTOGENETIC ABNORMALITIES in CHILDHOOD ALL	38
•Numerical Chromosomal Abnormalities	38
•Structural Chromosomal Abnormalities	42