An-Najah National University

Faculty of Graduate Studies

Prevalence and Microbiology Profile of Febrile Neutropenia among Acute Lymphoblastic Leukemic Pediatric Patients

By

Rania Ghanem

Supervisor

Prof. Waleed Sweileh

This Thesis is submitted in Partial Fulfillment of the Requirements for the Degree of Masters of Clinical pharmacy, Faculty of Graduate Studies An-Najah National University, Nablus, Palestine.

2019

Prevalence and Microbiology Profile of Febrile Neutropenia among Acute Lymphoblasti Leukemic Pediatric Patients

By

Rania Ghanem

This thesis was defended successfully on 14 / 11 / 2019 and approved by:

Defense committee Members	**Signature**
– **Prof. Waleed Sweileh/ Supervisor**	.....
– **Dr. Hussien Halak / External Examiner**	.....
– **Dr. Adham Abu Taha / Internal Examiner**	.....

Dedication

Strength doesn't come from what you can do. It comes from overcoming the things you once thought you couldn't.

– Rikki Rogers -

I would like to dedicate this work to my family who support me a lot during the time I was doing this masters. I would really admit that without their love and supporting hands I wouldn't be able to be what I am now.

Especial gratefulness to my parents (my father and my mother) who grew me up to be that responsible person. I am very dedicated to my sister as well. I promise you all to let you be proud of me whatever the challenges are.

I am very thankful to my husband Fadi and my three little gorgeous daughter Meray, Maria and Yasmin for being there to shine my life and give me that much of reinforcement and love.

Acknowledgment

My thanks, respect and appreciation go to my supervisor Prof. Waleed Sweileh for his ongoing advice, support and patience. It was a great honour and pleasure to work with you.

Special gratitude to Huda Al-Masri Pediatric Cancer Department team at Beit Jala Governmental Hospital in Bethlehem for always being there to help and support especially Dr. Mohammad Najajrah (head of the department).

I am very grateful to my colleagues at Al-Quds University for their continuous support in the hard time especially Dr. Ahmad Amro.

I would like to acknowledge Dr. Hussien Halak and Dr. Adham Abu Taha for their subjective criticism and advice.

Finally, I would like to thank the professors of the clinical pharmacy department at An-Najah National University.

الإقرار

أنا الموقع أدناه مقدم الرسالة التي تحمل العنوان:

Prevalence and Microbiology Profile of Febrile Neutropenia among Acute Lymphoblastic Leukemic Pediatric Patients

أقر بأن ما اشتملت عليه هذه الرسالة إنما هي نتاج جهدي الخاص، باستثناء ما تمت الإشارة إليه حيثما ورد، وأن هذه الرسالة ككل، أو أي جزء منها لم يقدم لنيل أية درجة أو لقب علمي أو بحثي لدى أية مؤسسة تعليمية أو بحثية أخرى.

Declaration

The work provided in this thesis, unless otherwise referenced, is the researcher's own work, and has not been submitted elsewhere for any other degree or qualification.

Student's Name: **الطالب اسم :**

Signature: **التوقيع :**

Date: **التاريخ :**

List of Contents

List of Tables

List of Figures

List of Abbreviations

Abbreviation	**Full Name**
ALL	Acute Lymphoblastic leukemia
AML	Acute myeloid leukemia
AK	Amikacin
AMP	Ampicillin
ANC	Absolute neutrophil count
BSI	Blood stream infection
CBC	Complete blood count
C. albicans	*Candida albicans*
CFZ	Ceftazidime
C. luteola	*Chryseonomas luteola*
CIP	Ciprofloxacin
CFM	Cefixime
CXM	Cefuroxime
CNS	Central nervous system
CONS	*Coagulase negative staphylococcus aureus*
CSF	Cerebro spinal fluid
CTR	Ceftriaxone
CTX	Cefotaxime
CVC	Central venous catheter
E. cloacae	*Enterobacter cloacae*
E. coli	*Escherichia coli*
E. coli ESBL	*Escherichia coli extended spectrum beta-lactamase*
ERT	Etrapenem
FN	Febrile neutropenia
FUO	Fever of unknown origin
GIT	Gastro-intestinal tract
IMP	Imipenem
IT	Intra-thecal
IV	Intra-venous
K. pneumonia	*Klebsiella pneunomia*
K. pneumonia ESBL	*Klebsiella pneunomia extended spectrum beta- lactamase*
MER	Meropenem
MOH	Ministry of health
MRI	Magnetic resonance imaging
MRSA	*Methicillin-resistant Staphylococcus aureus*
NCI	National cancer institute
OX	Oxacillin
P. aerogenosa	*Pseudomonas aerogenosa*
PIT	Piperacilline/tazobactam
PCR	Poly chain reaction

PEN	Penicillin
PMNs	Polymorphonuclear cells
ROS	Reactive oxygen species
S. aureus	*Staphylococcus aureus*
S. epidermidis	*Streptococcus epidermidis*
S. pyrogens	*Streptococcus pyrogens*
S. saccharolyticus	*Streptococcus saccharolyticus*
STD	Standard deviation
TEP	Teicoplanin
UTI	Urinary tract infection
VAN	Vancomycin

Prevalence and Microbiology Profile of Febrile Neutropenia among Acute Lymphoblastic Leukemic Pediatric Patients

By
Rania Ghanem
Supervisor
Prof. Waleed Sweileh

Abstract

Background: neutropenia is a life-threatening complication among patients suffering from acute lymphoblastic leukemia (ALL). It is a consequence of the disease pathophysiology and as a side effect of cancer chemotherapeutic agents. Febrile neutropenia (FN) is an indication of infections among these immune compromised patients with high rates of morbidity and mortality. The immediate intervention in the treatment of FN infections with the appropriate empiric antimicrobial agent/s, is the corner stone in saving the life of the patient.

Objectives: to determine the prevalence of febrile neutropenia among pediatric ALL patients. To assess the microbiology profile for each febrile neutropenia episodes in order to identify the microbes that are responsible for FN. To evaluate their sensitivity patterns profiles of the obtained positive cultures.

Methodology: a descriptive retrospective study, that had been carried out at Huda Al-Masri Pediatric Cancer Department at Beit Jala Governmental Hospital in Bethlehem during the period from 1/1/2014 – 1/6/2018. Medical records of 83 pediatric ALL patients were reviewed by means of symptoms,

vital signs, different blood tests, FN, positive cultures, microbiology profile and antibiotic sensitivity patterns were also analyzed.

Results: Sixty three patients had a total of 161 FN episodes. The majority were due to non-bacterial FN episode 54%. Bacterial origin of FN attacks were about 45%. Gram-positive bacteria accounted for 62.5% of the bacterial infections, while Gram-negative bacteria were about 37.5% of them.

Among Gram-positive, *CONS* were the most abundant pathogen 58.7%, followed by *S.aureus* 15.2%. Both were isolated mostly from blood culture samples. *CONS* was found to be 100% sensitive to tiecoplanin, vancomycin, ceftriaxone and cefotaxime. While it was least sensitive to ampicillin and penicillin 18.5% and 7.4% respectively.

Regarding Gram-negative bacteria, *E.coli ESBL* was the most frequent pathogen 42.9% that is isolated mostly from blood samples, followed by *E.coli* 17.9% which is isolated commonly from urine samples. *E.coli ESBL* showed 83.3% sensitivity towards amikacin, meronem, imipenem and ertapenem while it was 25% sensitive to cefotaxime.

Conclusion: This study showed the high prevalence 94% of FN among ALL pediatrics. Gram-positive bacteria were more frequent in occurrence with *CONS* are the most abundant pathogen with high sensitivity to tiecoplanin, vancomycin, ceftriaxone and cefotaxime. *E.coli ESBL* was the predominant Gram-negative microbe with lower sensitivity behavior. It is recommended that the empiric antibiotic therapy for the management of FN infection

should be revised, re-evaluated and updated according to the bacterial behavior towards antibiotics.

Keywords: Pediatric, acute lymphoblastic leukemia (ALL), febrile neutropenia.

Chapter (1)

Introduction

1.1- Background

Childhood malignancies are rare. They are of two types: solid and hematological malignancies. Leukemia is one of the hematological diseases and it is divided into acute and chronic leukemia. Acute leukemia includes: acute lymphoblastic leukemia (ALL) and acute myeloid leukemia (AML), in which there is malignant proliferation of precursors either to lymphoid blood cells (ALL), or to myeloid blood cells (AML) [*Pui*, 1997].

Leukemia is the most common malignancy in childhood. ALL accounts about 75% of the cases, while AML accounts for 20% of the cases [*Pui*, 1997]. The peak incidence for ALL is between the age of two and six years old [*Carroll and Bhatla*, 2016].

In the USA, there are more than 3000 new cases of ALL among children each year [*Siegel et al.*, 2017].

In February 2017, the Palestinian ministry of health (MOH) announced that cancers among pediatric patients represent 7.8% of all cancer patients. About 31.7% of those pediatrics are less than five years old. The majority of them 30.2% were diagnosed with leukemia, brain and central nervous system (CNS) cancer were by 18.5%, non-Hodgkin lymphoma by about 7.4%, multiple myeloma accounted for 5.8%, and lymph node

carcinoma was about 4.2%. The most leading cause of death among these Palestinian children are: brain and CNS cancer by about 43.9 % while for leukemia by 26.8% [*Ministry of Health*, 2018].

1.2- Pathophysiology and risk factors of ALL

Acute lymphoblastic leukemia (ALL) is abnormal cancerous transformation and proliferation of lymphoid progenitor cells in the bone marrow, blood and extramedullary sites [*Terwilliger and Abdul-Hay*, 2017].

This clonal disease of lymphoid cell may originate from B-cells or T-cells as a result of genetic mutations in blood progenitor cells, resulting in un-programmed ability to self-renewal thus preventing the developmental process of the progenitor cells to proliferate and differentiate [*Greaves*, 1997]. The accumulation of these immature cells and blasts in bone marrow will inhibit the normal hematopoiesis process leading to anemia and thrombocytopenia. Moreover, these cells will infiltrate from the bone marrow, reticulo-endothelial system and extra-medullary sites to blood stream in huge amount [*Alvarnas et al.*, 2015].

There are many predisposing factors that may increase the risk for ALL. Chromosomal abnormalities; like chromosomal translocations: t(12;21) [ETV6-RUNX1], t(1;19) [TCF3-PBX1], t(9;22) [BCR-ABL1], as well as to many chromosomal rearrangements of mixed-linked leukemia (MLL). Genetic syndromes had been also identified to have higher risk of ALL such as; Down syndrome, Fanconi anemia and many others [*Chessells*

et al., 2001; *Terwilliger and Abdul-Hay*, 2017]. Other environmental factors which may precipitate ALL include; exposure to ionized radiations, certain chemicals and pesticides, viruses (for example; Epstein-Barr virus and Human immune-deficiency virus) [*Terwilliger and Abdul-Hay*, 2017]. During childhood males are at higher risk to develop ALL than females [*Hunger and Mullighan*, 2015]. Lim and his co-worker had identified the incidence of pediatric ALL in respect to race and ethnic groups; hispanics were found to have the higher incidence, then after came the whites and lastly the blacks [*Lim et al.*, 2014].

1.3- Diagnosis of ALL

The presence of more than 20% of lymphoblasts within the bone marrow or peripheral blood is diagnosed as ALL. Other laboratory tests are needed to confirm the diagnosis and to identify the risk stratification; blood morphology, flow cytometry, immunophenotyping and other cytogenic and molecular genetic tests. For evaluating CNS involvement lumbar puncture with cerebrospinal fluid (CSF) analysis is required, in case of CNS is affected brain magnetic resonance imaging (MRI) should be done. Complete blood count CBC, blood smear, full serum chemistry and coagulation profile are also needed for full evaluation of the disease [*Alvarnas et al.*, 2015; *Terwilliger and Abdul-Hay*, 2017].

1.4- Symptoms of ALL

ALL patients will present with many symptoms as a result of; excessive infiltration by blast cells in the bone marrow, infiltration of these

blasts in the tissues and other effects due to the release of cytokines by these malignant cells.

Acute leukemia patients will suffer from; anemia, weakness, pallor, thrombocytopenia, bleeding, neutropenia and susceptibility to infections and fever due to low immunity, hepatosplenomegaly and lymphadenopathy. Other organs may be affected by the infiltrated cells such as; CNS and testes [*Alvarnas et al.*, 2015; *Terwilliger and Abdul-Hay*, 2017].

1.5- Treatment of ALL

Treating children will ALL includes: multi-drug combination of chemotherapeutic agents in addition to intrathecal (IT) chemotherapy as CNS protective measures.

There are many protocols for the treatment of ALL, all include three main phases of aggressive chemotherapy:

1- **Remission induction phase:** it is the first treatment step. It intends to kill the leukemic cells in the bone marrow and peripheral blood in order to reduce the cancer cells burden.

2- **Consolidation/intensification phase:** it is the second step of treatment. This step kills any leukemia malignant cells that are still found in the body and may cause retrogression later.

3- **Maintenance phase:** it is the third step in the treatment protocols. The purpose of this phase is to kill leukemia cancer cells that might grow again and also it prevents retrogression [*Kato and Manabe*, 2018].

The pediatric ALL patients at Huda Al-Masri Pediatric Cancer Department at Beit Jala Governmental Hospital in Bethlehem; where this study was conducted, were treated according to AIEOP-ALL protocol, see table (1.1) for detailed protocol.

Table (1.1): AIEOP-BFM ALL 2000 treatment phases, adopted from [*Conter et al.*, 2014].

Drug/administration route	Daily dose (mg/m^2)	Day
Prephase		
Prednisolone/P.O-IV	60	1-7
MTX/IT	By age	1
Induction: protocol IA		
Vincristine/IV	1.5 (max. 2 mg)	8, 15, 22, 29
Prednisolone/P.O-IV	60	8-28 then tapered
Or Dexamethasone/P.O-IV	10	8-28 then tapered
Daunorubicin/IV	30	8, 15, 22, 29
L-Asparaginase/IM	5000 IU/m^2	12, 15. 18, 21, 24, 27, 30, 33
MTX/IT	By age	1, 15, 29
Consolidation: protocol IB		
Cyclophosphamide/IV	1000	36, 64
Mercaptopurine/ P.O	60	36 – 63
Cytarabine/IV-SC	75	38-41, 45-48, 52-55, 59-62
MTX/IT	By age	38, 52
HR block 1		
Dexamethasone/P.O-IV	20	1-5
Vincristine/IV	1.5 (max. 2 mg)	1, 6
Cytarabine/IV	2000 x 2	5
MTX/IV	5000	1
Leucovorin rescue	15 mg/ m^2/dose	42, 48, 54 h after start MTX
Cyclophosphamide/IV	200 (q 12h x 5)	2-4
L-Asparaginase/IM	10000 IU/m^2	6
MTX/IT	By age	1
HR block 2		
Dexamethasone/P.O-IV	20	1-5

Vindosine/IV	3	1, 6
Daunorubicin/IV	30	5
MTX/IV	5000	1
Leucovorin rescue	15 mg/ m^2/dose	42, 48, 54 h after start MTX
Ifosphomide/IV	800 (q 12h x 5)	2- 4
L-Asparaginase/IM	10000 IU/m^2	6
MTX/IT	By age	1
HR block 3		
Dexamethasone/P.O-IV	20	1-5
Cytarabine/IV	2000 (q 12h x 4)	1, 2
Etoposide/IV	100 (q 12h x 5)	3-5
L-Asparaginase/IM	10000 IU/m^2	6
MTX/IT	By age	5
Reinduction (protocol II)		
Dexamethasone/P.O-IV	10	1-21 then tapered
Vincrisine/IV	1.5 (max. 2 mg)	8, 15, 22, 29
Doxorubicin/IV	25	8, 15, 22, 29
L-Asparaginase/IM	10000 IU/m^2	8, 11, 15, 18
6- Thioguanine/P.O	60	36- 49
Cyclophosphamide/IV	1000	36
Cytarabine/IV-SC	75	38-41, 45-47
MTX/IT	By age	38, 45
Cranial irradiation	By age	
Reinduction (protocol III)		
Dexamethasone/P.O-IV	10	1-14 then tapered
Vincristine/IV	1.5 (max. 2 mg)	1, 8
Doxorubicin/IV	30	1, 8
L-Asparaginase/IM	10000 IU/m^2	1, 4, 8, 11
6- Thioguanine/P.O	60	15- 28
Cyclophosphamide/IV	500	15
Cytarabine/IV-SC	75	17-20, 24-27
MTX/IT	By age	17, 24
Cranial irradiation	By age	
Interim maintenance		
Meractopurine/P.O	50	Daily
MTX/P.O	20	Weekly
Continuation phase		
Meractopurine/P.O	50 *	Daily
MTX/P.O	20 *	Weekly

*Doses should be adjusted according to WBCs count (target range: 2000- 3000 cell/µl).

The duration of each chemotherapy cycle in AIEOP-ALL protocol is shown in figure (1.1).

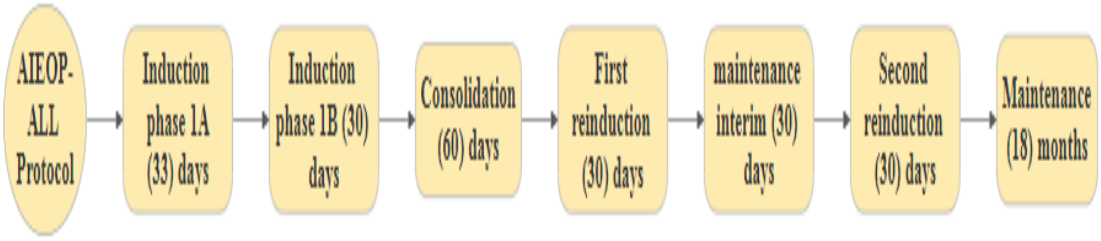


Figure (1.1): the duration of each chemotherapy cycle in AIEOP-ALL protocol.

1.6- The role of neutrophils

Neutrophils are also called polymorphonuclear cells (PMNs), they are produced in the bone marrow from stem cells within the hematopoiesis process then after, and they proliferate and differentiate into mature neutrophils that are characterized by the presence of granules. The mature neutrophils then migrate to the circulation where they live for short period of time ranging from hours to days unless they have to act as the body's defensive mechanism at an infection site where they remain active for (2-6) days.

They kill the microbes (bacteria, fungal, microbes in the blood) by antimicrobial and bactericidal proteins which they secrete from the granules or by activating certain metabolic pathways, which at the end will destroy the pathogen's DNA [*Borregaard*, 2010; *Dale*, 2005; *Nathan*, 2006; *Pillay et al.*, 2010; *Tak et al.*, 2013].

1.7- Chemotherapy induced neutropenia

Most chemotherapeutic agents will cause myelosuppression in a dose-dependent manner. The highly cytotoxic agents for bone marrow are: alkylating agents, pyramidine analogs, anthracyclines, anthraquinones, nitrosoureas, methotrexate, mitomycin C and hydroxyurea. As a result the host protective mechanisms will be affected negatively as neutropenia is induced. Moreover anticancer therapy produces reactive oxygen species (ROS), causing induced-somatic oxidative DNA damage leading to cellular aging and the senescence process, resulting in apoptosis or permanent cell-cycle arrest. The end point event will be bone marrow cytotoxicity, myelosuppression, and immunosuppression, this will reduce the number of neutrophils production causing neutropenia [*Xiao et al.*, 2017].

Hence the patient will be at high risk for infectious diseases due to suppressing the production of neutrophils and by damaging the cells of the gastrointestinal tract [*Crawford et al.*, 2004].

1.8- Consequences of neutropenia

Pediatric ALL patients will suffer from neutropenia due to:

1- The cytotoxic effects of the leukemic cells [*Crawford et al.*, 2004].

2- The aggressive chemotherapy treatment which suppresses the hematopoietic system, and destroy the body's defensive mechanism due to neutropenia [*Crawford et al.*, 2004; *Xiao et al.*, 2017].

Thus the patient will be at serious high risk for infections and life-threatening infections caused by bacteria, viruses, fungus and many other pathogens.

1.9- Neutropenia and febrile neutropenia (FN)

Neutropenia is defined as the reduction in the absolute neutrophil count (ANC) in the blood to less than 1500 cell/µl (normal level). Neutropenia is classified into five grades in which the (ANC)is as following: grade (I) neutropenia The ANC is (1500-1000 cell/µl), grade (II) neutropenia the ANC is (1000- 500 cell/µl) without fever, grade (III) in which the ANC is (1000- 500 cell/µl) and fever is present as well, grade (IV) neutropenia the ANC is (< 500 cell/µl) with the presence of life threatening consequence and grade (V) is the death of the patient.

Neutropenia can be also determined as mild for grade (I), moderate for grade (II and), sever for grade (III) and life threatening condition is for grade (IV). These conditions will expose the patient to serious opportunistic infections [*Dale*, 2005; *Nirenberg et al.*, 2006].

Febrile neutropenia (FN) is a life threatening condition that is described in patients who are receiving chemotherapy and are neutropenic. It is defined as having; neutropenia: ANC ≤ 500 cell/µl **or** ANC is expected to fall to ≤ 500 cell/µl within the next 48 hour, **and** at the same time are having fever ≥ 38.3 ºC on one occasion or, a temperature of ≥ 38.0 ºC that is

maintained for one hour [*Dale*, 2005; 2009; *Freifeld et al.*, 2011; *Swati et al.*, 2010; *Vossen et al.*, 2018].

It had been shown in previous pediatric studies that the risk factors for chemotherapy induced febrile neutropenia (FN) are: early stage/first cycle of therapy, chemotherapy dose intensity, bone marrow involvement, presences of central venous lines and any previous attacks of febrile neutropenia [*Nirenberg et al.*, 2006; *Stabell et al.*, 2008; *Wicki et al.*, 2008], table (1.2) demonstrates FN risk factors.

Table (1.2): FN risk factors, adopted from: [*Nirenberg et al.*, 2006].

Treatment related
- Previous history of severe neutropenia with similar chemotherapy
- Type of chemotherapy (anthracyclines and platinum-based regimens)
- Planned relative dose intensity greater than 80%
- Preexisting neutropenia or lymphocytopenia
- Extensive prior chemotherapy
- Concurrent or prior radiation therapy to marrow-containing bone

Patient related
- Older age
- Female gender
- Poor performance status
- Poor nutritional status (e.g., low albumin)
- Decreased immune function
- Open wounds or active tissue infection
- Comorbidities
  - Chronic obstructive pulmonary disease
  - Cardiovascular disease
  - Liver disease (elevated bilirubin, alkaline phosphatase)
  - Diabetes mellitus
  - Low baseline hemoglobin

Cancer related
- Bone marrow involvement with tumor
- Advanced cancer
- Elevated lactate dehydrogenase (lymphoma)

1.10- Consequences of febrile neutropenia on chemotherapy

The occurrence of febrile neutropenia will affect the treatment protocol negatively as it will cause:

- Treatment discontinuation.

- Dose delay.

- Dose reduction [*Badr et al.*, 2016].

1.11- Diagnosis of febrile neutropenia

Full patient's history information should be obtained including: details of the given chemotherapy, presence of prophylactic antibiotic, current steroid use, recent surgeries and drug allergies.

Figure (1.2) illustrates the systemic initial assessment and investigation of FN that should be followed.

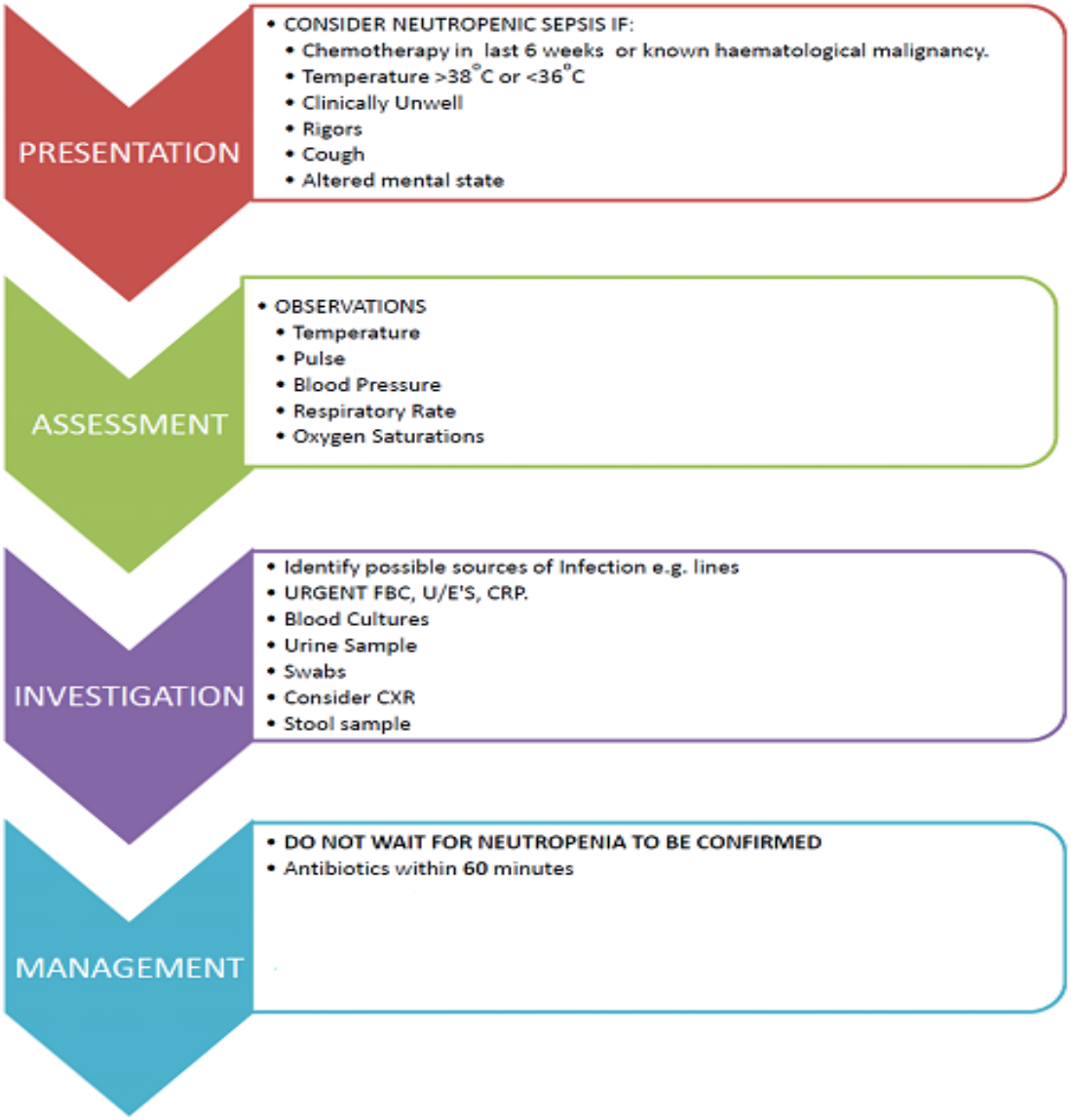


Figure (1.2): Initial assessment of febrile neutropenia, extracted from [*NHS*, 2017].

1.12- Treatment of Febrile Neutropenia

The occurrence of FN in oncology patients is a sign of infection which may progress very rapidly to life-threatening condition in such immune compromised patients. The immediate intervention with the appropriate empiric antimicrobial therapy according to the suspected microbe in addition to wide coverage against bacteria is a crucial point in the management of FN

to save the life of the patient, figure (1.3) shows therapeutic escalation approach for the treatment of septic FN [*Vossen et al.*, 2018].

Moreover, prophylactic antifungal agent should be administered twice weekly. Fungal infection in FN should be also treated immediately [*Vossen et al.*, 2018].

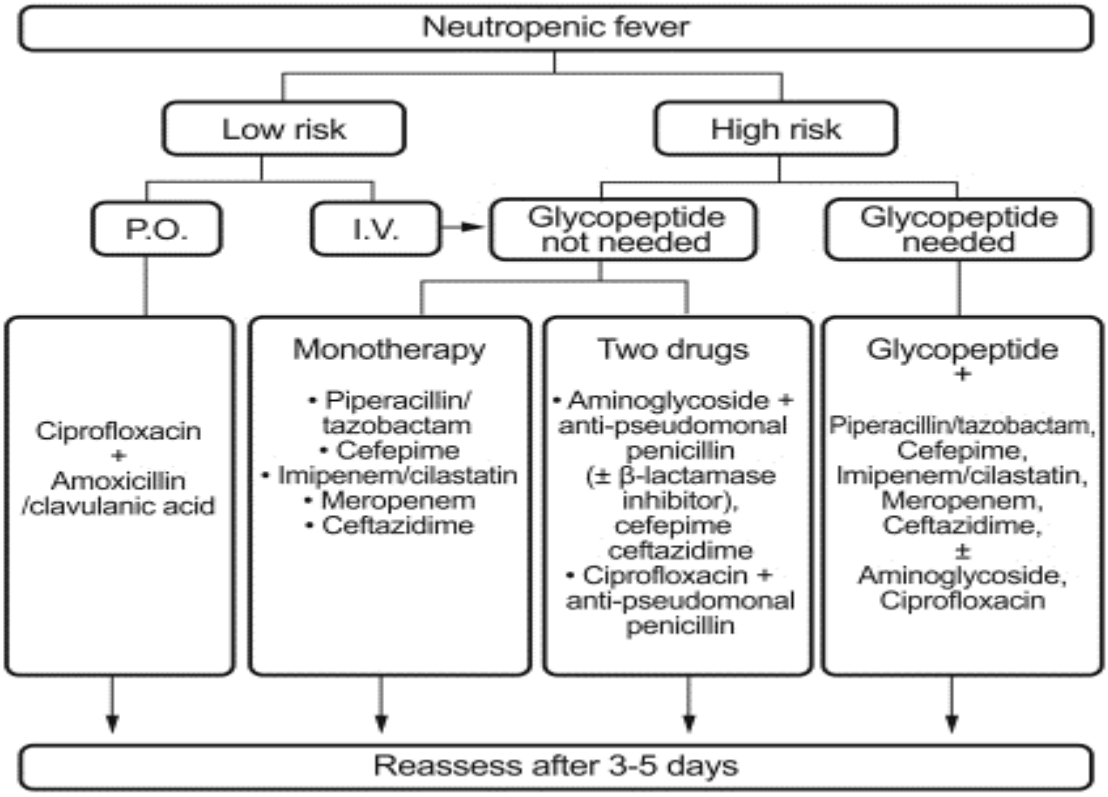


Figure (1.3): FN treatment escalation chart, extracted from [*LCCH*, 2018; *D G Lee et al.*, 2011].

The guidelines had recommended the use of single dose of 5µg/kg/day subcutaneously of granulocyte colony-stimulating factor (G-CSF) within 24-72 hours following the last chemotherapy as a prophylactic measure to chemotherapy induced neutropenia. However, some studies had suggested

that the initiation of G-CSF within less than 24 hours from the last chemotherapy may benefit certain patients [*Klastersky et al.*, 2016; *Limvorapitak and Khawcharoenporn*, 2015; *Ludwig et al.*, 2019].

1.13- Empiric Antimicrobial Treatment of FN at Huda Al-Masri Pediatric Cancer Department at Beit Jala Governmental Hospital

The followed empiric antimicrobial treatment of FN at Huda Al-Masri Pediatric Cancer department is adopted from;

1- NCCN Practice Guidelines in Oncology – v.1.2011.

2- Guidelines for Preventing Infectious Complications among Hematopoietic Cell Transplantation Recipients: A Global Perspective 2009

3- IDSA Guidelines for Febrile Neutropenia CID2011

Appendix (1) represents the followed guideline at the previously mentioned hospital.

The empiric antibiotic therapy that had been used in practice at Huda Al-Masri Pediatric Cancer department was ceftazidime or piperacillin/tazobactam combination. Metronidazole had been used for anaerobic bacteria. Carbapenems and vancomycin were used in case of penicillin allergy.

1.14- Statement of the Problem

Febrile neutropenia is a life threatening complication of both acute lymphoblastic leukemia and chemotherapy treatment; that if not treated immediately and properly can be lethal.

This study had identified the prevalence of FN and the type of the causative pathogens that are responsible of infections and their resistance pattern at Huda Al-Masri Pediatric Cancer Department at Beit Jala Governmental Hospital in Bethlehem.

1.15- Significance of the study

According to our knowledge, this was the first study to be conducted in Palestine among pediatric patients who are diagnosed with acute lymphoblastic leukemia (ALL). The study aimed to re-evaluate and to establish the policy of the empirical therapy that is currently followed at Huda Al-Masri Pediatric Cancer Department at Beit Jala Governmental Hospital in Bethlehem.

1.16- Study objectives

The objectives of this study are:

a- Determine the prevalence of febrile neutropenia among pediatric ALL patients.

b- Determine the microbiology profile for febrile neutropenia episodes.

c- Determine the resistance profile for positive cultures obtained from febrile neutropenic pediatric patients.

1.17- Research questions

No.	Research question	Hypothesis	Statistics
1)	What is the prevalence of FN among children suffering from ALL?	No hypothesis	Descriptive analysis
2)	What are the most bacterial pathogen that are responsible for infection in FN patient?	No hypothesis	Descriptive analysis
3)	What is the susceptibility pattern of the bacteria that are causing FN in pediatric ALL patients?	No hypothesis	Descriptive analysis

Chapter (2)

Literature review

2.1- Prevalence of FN among pediatric ALL patients

It had been found in a research done at St. Jude Children's Research Hospital, Memphis in the USA, that occurrence of infections in pediatric ALL patients is (10) times more in low- and middle-income countries compared to high-income countries [*Caniza et al.*, 2015]. Therefore**,** assessing the type, frequency and severity of infections in oncology patients (adults or pediatrics) is a must for every single cancer center especially those treating hematological malignancies in order to prevent and control any life-threatening conditions in such patients who are immunocompromised as mentioned above.

Many studies from Finland, Greece, Germany, Morocco and Taiwan had recorded that the prevalence of FN episodes in ALL pediatric patients is ranging between (1- 4.2 episode/patient) [*Depasse et al.*, 2013; *Katsimpardi et al.*, 2006; *Lex et al.*, 2001; *Li et al.*, 2017; *N Ozdemir et al.*, 2016; *Rahiala et al.*, 1998].

2.2- Infections etiology and microbiological profile of FN among ALL patients

Febrile neutropenia is an emergent complication in hematological malignant patients as the patient may face a life-threatening infections due

to neutropenia and leukopenia. This condition is serious in terms of morbidity and mortality. Thus, detecting and identifying the causative microorganism and its microbiological profile is the key in saving the patients' life.

In a retrospective study done at King Abdullah University Hospital in Jordan, microbial profile of positive blood cultures in pediatrics was as following; Gram-positive bacteria was the most common pathogen 60% of the cases, 29% had Gram-negative bacteria and 11% with fungal infections. The most predominant Gram-positive bacteria was *Coagulase Negative Staphylococcus* (*CONS*) 59%, while *Staphylococcus aureus (S. aureus)* accounted for 28%. For Gram-negative bacteria 25% of the cases were infected with *Klebsiella pneumonia (K. pneumonia)*, while 19% were for *E.coli*. Candida species were responsible for all fungal infections. Regarding susceptibility pattern in this study, *Methicillin-resistant Staphylococcus aureus (MRSA)* was seen in 10 blood cultures of *S. aureus*, while one sample was intermediate to vancomycin. The isolated *Enterococcus* bacteria were sensitive to vancomycin and tiecoplanin except one which was intermediate to vancomycin. For Gram-negative bacteria, 10% of them were extended spectrum beta-lactamase producing (ESBL). All the *Pseudomonas aerugenosa (P.aerugenosa)* samples were susceptible to piperacilline-tazobactam and carbapenems antibiotics. The *Enterobacter species* were found to be sensitive to fourth generation cephalosporines, carbapenems and aminoglycosides [*Al-Sweedan et al.*, 2012].

Al Omar and colleagues had worked out on a prospective observational study among pediatrics at King Hussein Cancer Center in Jordan. 18% of the episodes were positive for pathogens. About 50% of them were Gram-positive bacteria, however, 20% were Gram-negative bacteria, 25% were viral infection and 5% were fungal infections. Bacteremia was found in 60% of the episodes. In which, 75% were gram-positive bacteria represented as: *CONS* (n=2), *Enterococcus faecalis* (n=1),

Streptococcus saccharolyticus (S. saccharolyticus) (n=1), Peptostreptococcus spp. (n=1), *Neisseria cinerea* (n=1), *Gram-positive Bacillus (n=1),* and *Viridans streptococci (V. streptococci)* (n=1). One episode of bacteremia was with mixed Gram-positive bacteria: (*CONS* and *V. streptococci*). Gram-negative was about 25% of bacteremia episodes, with *E. coli* (n=2) and *K. pneumonia* (n=1). It has been found that patients with central lines were having significant positive blood cultures compared to those without central line catheters. The non-bacteremia clinically documented infections accounted 10% of the episodes (2 episodes): 1 episode of *Proteus vulgaris* obtained from urine culture and 1 episode of *MRSA* obtained from a nasal swab culture. The sites of the clinically documented infections were as following: respiratory 62.1%, urinary 17.2%, gastrointestinal 17.2%, and one episode of mixed urinary and respiratory infections 3.5% [*Al Omar et al.*, 2013].

In another retrospective study conducted at department of pediatrics, King Hussein Cancer Center in Jordan, had assessed the causative pathogen

from positive urine cultures in children with cancer. 41% of the cases were diagnosed with solid tumors while 26% of them had hematological malignancies. The most isolated bacteria were gram-negative 84%, with *E.coli* was the predominant one 51%. Followed by *K.pneumonia* 9% and *P.aeruginosa* 8%. Nearly 37% of the isolated gram-negative bacteria were extended spectrum beta-lactamase (ESBL)-producers, and 3% showed multi-drug resistant manner. For gram-positive bacteria, *CONS* were the most causative pathogen 6.6% followed by *Enterococci* 5%. In this study gram-positive bacteria showed susceptible behavior to standard antibiotics [*Hirmas et al.*, 2017].

However, In Lebanon Gram-negative organisms were the predominant pathogens that are responsible for FN in malignant patients [*Ghosn et al.*, 1994; *Hamzeh et al.*, 2000; *Kanafani et al.*, 2007; *Kanj et al.*, 2001]. According to a prospective study at the American University of Beirut Medical Center. 104 episodes were studied in 64 patients over 1.5 years. Bacteremia was documented in 29% of the episodes. Around 60% of them were gram-negative bacilli, with *E.coli* as the predominant Gram-negative organism. While 40% were Gram-positive cocci., with *CONS* as the most prevalent Gram-positive agent [*Hamzeh et al.*, 2000].

Another prospective observational cohort study done at the same center, showed that 78.8% of the bloodstream infections were due to Gram-negative bacteria, were *E. coli* has was the most isolated pathogen followed by *P. aeruginosa*. However, (33.3%) of the blood stream infections were as

a result Gram-positive pathogens, with the majority were *CONS* bacteria [*Kanafani et al.*, 2007].

The goal of a cross-sectional study performed at King Abdulaziz Medical City-Western Region (KAMC-WR), in Saudi Arabia, was to evaluate the FN treatment guideline. One hundred FN episodes were studied and it was surprising that 55% of patients were treated with a guideline that is not compliant treatment. This is due to incorrect dosing of the used antibiotics. About 19% of the FN episodes were due to bacteria, in which gram-negative pathogens were predominate in 16 episodes 74%, whereas gram-positive pathogens were seen in 3 episodes 16% of episodes. Mixed pathogen infections (Gram-negative and Gram-positive were detected in 10% of the FN episodes. Regarding Gram-negative bacteria, *E. coli* was the most common microbe obtained from six isolates, in which four isolated were *E. coli ESBL* and the remained two isolates were susceptible to penicillin. *K. pneumonia* was isolated from three episodes, one was *K. pneumonia ESBL*, while the remaining isolated were to penicillin. *P. aeruginosa* was identified from three isolates, in which all were sensitive to ceftazidime and piperacillin/tazobactam. *Acinetobacter baumannii* were detected in two episodes, one of them was resistant to imipenem, and however both were sensitive to colistin. For Gram-positive bacteria, there were two isolates of *Enterococcus species* which were susceptible to ampicillin, one *Micrococcus luteus* isolate that was found to be sensitive to piperacillin/tazobactam, *CONS* which was sensitive to vancomycin, and *methicillin-sensitive Staphylococcus aureus* [*Naeem et al.*, 2018].

An observational Iraqi study, had evaluated the bacterial and its antimicrobial resistance profile in FN children with hematological malignancies over a period of two years (2012 and 2013). Positive bacteria cultures were 40.4% and 48%, while multi-microbial isolates were seen in 18.4% and 11.1% of the sample during 2012 and 2013 respectively. Gram-negative bacteria had been the most common pathogen during the study duration (2012 and 2013) as follow: 63.6% and 68.1% respectively. The distribution of Gram-positive bacteria was 36.3% in 2012 while 31.8% in 2013. *E. coli* and *P. aeruginosa* were the most abundant Gram-negative microbes. Whereas, *Streptococcus epidermidis (S. epidermidis)* and *S. aureus* were the predominant Gram-positive isolates. No significant differences in the bacterial distribution over the tow study period. Regarding susceptibility patterns in this study, many of the isolated Gram-negative and Gram-positive bacteria showed high resistance to many antibiotics with significant differences in the susceptibility behavior of the bacteria isolates over the two study periods [*Almaziny*, 2014].

A study held in Egypt to assess the characteristics and antimicrobial susceptibility of Gram-negative bacteria showed that, 30% of the samples were for *E. coli* followed by 24.5% for *P. aeruginosa* then after 18.7% for *Acinetobacter baumannii/haemolyticus. . E.coli* was abundantly isolated from stool 66.7%. Urine 49.2% and skin 38.3% samples. While the most common isolated Gram-negative bacteria isolated from sputum 35.1%, throat 34.6% and blood 21.6% was for *Acinetobacter baumannii/haemolyticus*. Testing the susceptibly pattern demonstrated that

were resistant to most tested antibiotics such as non-β-lactam agents for example aminoglycosides (gentamycin) and quinolones (cipsofloxacin, gatifloxacin and levofloxacin). Carbapenems resistance had also been reported, as the previously mentioned pathogen showed resistance to imipenem: (*Acinetobacter species* 65.2%, *Pseudomonas* 42.7%, *Enterobacter species* 25%, *Klebsiella* 10.5% and (*E. coli* 3.9%) [*Eldomany and Abdelaziz*, 2011].

A retrospective study performed at the pediatric ward of the National Cancer Institute (NCI) at Cairo University in Egypt, had evaluated the blood stream infections (BSI) in FN pediatric cancer. In this study Gram-positive bacteria were the most common pathogens, followed by Gram-negative microbes and mixed infections as; 51.2%, 29.6% and 13.7% respectively. Among Gram-positive bacteria *CONS*, *S. aureus* and *Streptococcus species* were detected in BSI by 16.2%, 13.4% and 12.1% respectively. The most common BSI caused by Gram-negative bacteria were *Acinetobacter species* 6.7% and *Pseudomonas species* 5.5%. Fungemia accounted for 2.7% of the episodes, with *aspergillus fumigatus* is the predominant fungi.

Mixed BSI were observed as a mix of (fungi + Gram-positive cocci, fungi + Gram-negative bacteria, mixed Gram-positive cocci, Gram-positive cocci + Gram-negative bacteria, Gram-positive cocci + Gram-positive bacilli and finally Gram-negative and Gram-positive bacilli. Tests of the sensitivity

pattern found that 41.7% of the isolated *S. aureus bacteria* were *MRSA.* [*H El-Mahallawy et al.*, 2005].

Tunisian Observational study conducted at Aziza Othmana Univerity Hospital, identified that out of 94 FN episodes, 65% were fever of unknown origin FUO, 28.7% were microbiologically documented infections and 6.3% were clinically documented. Of the microbially documented infections, 48.1% were Gram-negative bacteria with *Klebsiella* is the dominant pathogen, 40.7% were Gram-positive organisms with *Staphylococcus* as the most common bacteria and 11.1% were mixed of Gram-positive and Gram-negative bacteria. 83% of the *Staphylococcus* bacteria were resistant to oxacillin [*Jeddi et al.*, 2010].

A retrospective cohort study performed in Morocco, to assess FN among ALL and AML pediatrics in two hematological oncology centers in Morocco. It has been found that the prevalence of FN among AML and ALL patients was (3 episodes / patient) and (1 episode / patient) respectively. Pneumonitis and mucositis were the most common clinically identified infections. Among Gram-positive bacteria, *CONS* was most abundant agent. While for Gram-negative bacteria, *E. coli* was the prevalent pathogen. 35% of FN episodes were fever of unknown origin (FUO). The mortality rate due to FN in this study was 11.3% and 9.2% in patients with AML and ALL respectively [*Depasse et al.*, 2013].

A study performed among children with cancer in the Children's hospital of Tabriz in Iran, in which 60.4% of cases were diagnosed with

hematological malignancy, with ALL accounts for 35.4% of them. It had been shown that 67.7% and 32.3% of bacteria isolated from blood cultures were Gram-positive and Gram-negative pathogens, respectively. The most common isolated microbes are *CONS* 28.1%, *S. aureus* 24%, and *K. pneumoniae* 9.4%.

The resistance profile for Gram-positive pathogens was as following: Penicillin, ceftizoxime, cefotaxime, erythromycin, ceftriaxone, oxacillin, co-trimoxazole, gentamicin, ampicillin, cephalexin, amikacin, ciprofloxacin, chloramphenicol, rifampin and vancomycin; 95%, 83.6%, 81.7%, 79.9%, 77.4%, 72.9%, 71.2%, 65.1%, 63.6%, 60.3%, 55.7%, 46.5%, 41%, 32.7% and 25.5% respectively. While for Gram-negative bacteria, the resistance profile was as follow: imipenem, cefixime, ceftazidime, cephalexin, ceftriaxone, cefotaxime, ceftizoxime, rifampin, gentamicin, amikacin, chloramphenicol, co-trimoxazole, and ciprofloxacin; 14.8%, 75%, 84%, 90.5%, 62.1%, 66.6%, 67.8%, 33.3%, 40%, 38.4%, 51.8%, 57.1%, and 9.7% respectively. Moreover, 88% of *CONS* and 77.3% of *S. aureus* bacteria were found to be resistant to oxacillin. On the over hand, *S. pneumoniae* and *V. streptococcus*, and 50% of non-group A, B or D streptococci were resistant to penicillin [*Rezaee et al.*, 2017].

A Turkish retrospective study, evaluated FN episodes in pediatric hematologic malignant patients. 40.5% of FN episodes were clinically documented, while 36.5% were microbiologically proven infections and 23% of the episodes were FUO. The site of the infections were as follow;

33.4% for mucositis and 24.7% for pneumonia. 75.3% of microbiologically documented infections were of positive blood cultures, in which Gram-negative bacteria were the highest prevalent 47.2%. Gram-positive microbes accounted for 38.2%, and 9.1% were fungal infections. Polymicrobial infections were seen in 5.5% of the episodes. The most abundant Gram-negative agent was *Klebsiella species* 30.8%, while the most commonly Gram-positive isolated bacteria was *Staphylococcus species* 85.7%. The major fungal agent isolated was *Candida species* 80%. It has been estimated that 13.2% of the patients (9 patients) died in this study. Seven of those patients died because of Gram-negative bacteremia, while two cases died due to fungemia [*Kar et al.*, 2017].

Another retrospective study done at Istanbul University in Turkey in 2016, found that the FN in ALL pediatric patients was 3.6 episode/ per patient. 50% of those FN episodes were due to FUO. The episodes were distributed as follow: 22% bacteremia, 38% pulmonary infections, 29% gastrointestinal infections, 18% urinary tract infections, 9% otolaryngological or dental infections, 4.5% dermatological and soft tissue infections and 1.5% meningitis infection. Out of twenty five pulmonary infections; five were due to fungal pneumonia, one case was atypical pneumonia and one because of viral bronchiolitis. 86% of the microbially documented infections were due to bacteria. In this study the prevalence of Gram-positive and Gram-negative bacteria were equal 50%. Eight percent of the infections were caused by viruses, and 6% were fungal infections. Among Gram-positive bacteria, *coagulase negative staphylococcus* was the

most common agent, *E. Coli* was the predominant Gram-negative isolate [*N Ozdemir et al.*, 2016].

Many Indian studies showed that Gram-negative bacteria are the most prevalent pathogens among FN patients with hematological malignancies [*Malabagi et al.*, 2015; *Mangaraj et al.*, 2015; *Siddaiahgari et al.*, 2014; *Swati et al.*, 2010]. Malabagi and co-workers found that 63.64% of the positive cutures were of Gram-negative bacilli organisms, in which E.coli was the most common. These Gram-negative bacilli were susceptible to imipenem, however their sensitivity profile to other antibiotics was as follows: 85.71%, 78.26%, 69.52%, 63.64%, 41.66% and 47.05% for pipercillin-tazoactum, meropenem, cefoperazone-sulbactum, amikacin, ceftazidime, ciprofloxacin respectively. The mortality rate due to septicemia with Gram-negative bacilli was 13.5% [*Malabagi et al.*, 2015]. Mangaraj and his collegues showed that the sensitivity pattern of Gram-negative bacilli was 100% sensitive to colistin and 93.8% to teigecycline. However, the resistance behavior towards cephalosporins and carbepenems was shocking. For example 68.8% of *E.coli* were sensitive to carbapenems, while 37.5% of *E.coli* were sensitive to ceftriaxone. About 64.3% of *K. pneumoniae* and 37.5% of *E. coli* were ESBL producers. The *Acinetobacter species* were sensitive to most of the tested antibiotics. [*Mangaraj et al.*, 2015].

Reviewing the positive cultures obtained from FN patients with hematological cancers in Pakistan in a prospective study, had demonstrated that 35.39% of FN were microbiologically proven. Gram-negative bacteria

was found in 85% of the isolates and *E. coli* was the most common pathogen. Gram-positive microbes accounted 15% of the isolates with *S. aureus* as the major agent. The susceptibility pattern of *E.coli* was as follow: cefoperazone/sulbactam 96%, amikacin 88%, carbapenem agents 66.6% and piperacillin/tazobactam 48%. Regarding the sensitivity profile of Gram-positive bacteria *MRSA* was 100% susceptible to vancomycin; While there were two isolates that were *vancomycin resistant enterococci (VRE)* [*Maqsood et al.*, 2015].

In an observational study by [*Latiff et al.*, 2002] in Malaysia, revealed that 50.6% of FN episodes were microbiologically proven infection. Bacteremia and fungal infections accounted 32.2% and 13.4% respectively. FOU was about 39.3% of the FN episodes.

Among bacteremia, Gram-negative pathogens accounted 64%, while Gram-positive microbes were 16%. Regarding Gram-negative bacteria, the *Klebsiella* group was about 28% with 16% of them were *extended spectrum beta lactamase (ESBL) producing Klebsiella* and 12% were multi-resistant *Klebsiella*. Regarding the Gram-positive microbes, *S. aureus* were the predominant pathogen 8%. Nearly 20% of FN cases were due to fungemia.

2.3- Summary

Febrile neutropenia is an urgent and serious complication in patients with hematological malignancy especially during chemotherapy treatment, as the patient will be at high risk for life-threatening infections. The

immediate intervention with the appropriate antibiotic empiric therapy is the corner stone in reducing mortality and morbidity. There are many factors that should be taken into account while determining the antibiotic of choice such as; infection focus, patient's infection risk factors, causative microorganism and its sensitivity behavior towards different antibiotics. The selection of the empiric antibiotic regimen in such cases should be individualized to each institute regardless to the international guidelines. Therefore, each center should identify the most common pathogens that are responsible for FN among malignant patients. Moreover, susceptibility pattern should be assessed from time to time, followed by re-evaluation for the used empiric therapy, as the bacteria may change its sensitivity for antimicrobial agents. In addition, to changes in the prevalence of the causative pathogen with time.

Chapter (3)

Methodology

3.1- Study Design

- This study is a descriptive retrospective study.

3.2- Study population

- Pediatric patients diagnosed with ALL and who had attend Huda Al-Masri Pediatric Cancer Department at Beit Jala Governmental Hospital in Bethlehem from 1/1/2014 – 1/6/2018.

3.2.1- Inclusion criteria

1. Pediatric patients who are (1-18) years old.
2. Diagnosed with acute lymphoblastic leukemia (ALL).
3. Receiving chemotherapy treatment at Huda Al-Masri Pediatric Cancer Department at Beit Jala Governmental Hospital in Bethlehem.
4. Patients with ANC ≤ 500 cell/µl **or** ANC is expected to fall to ≤ 500 cell/µl within the next 48 hour, **and** at the same time are having fever ≥ 38.3 ºC on one occasion or, a temperature of ≥ 38.0 ºC that is maintained for one hour.

3.2.2- Exclusion criteria

1- Patients who are less than one year old or those who are older than 18 years old.

2- Patients who had incomplete medical files.

3.3- Sampling and sample size

All the 83 pediatric patients that fit the inclusion criteria between the period (1/1/2014 -1/6/2018).

3.4- Data Collection

1. Patients' medical records were reviewed.
2. The data were recorded on a pre-designed data collection form.
3. The collection form contents (see appendix 2):

*) The patient's demographic characteristics (age, gender, address,…).

*) Type of malignancy.

*) Type and date of last chemotherapy given.

*) Patient's medical history and other co-morbid diseases if present.

*) Presence of the central line.

*) Vital signs.

*) Clinical manifestations.

*) Fever and the duration of the fever.

*) Laboratory results and duration of neutropenia will be determined.

*) Site of infection.

*) Culture results (the causative microbe and the pathogen susceptibility results)

*) Prophylactic antibiotic and anti-fungal agents which were used before FN episode.

*) The empiric therapy used and any add on antibiotics, antifungal or antiviral agents.

3.5 Clinical analysis

3.5.1- Microbiology

1. Microbiology data were collected from culture reports to determine the causative organism that is responsible for the infection (bacteria or fungus).
2. The culture results were done by the microbiology lab at Beit Jala Governmental Hospital in Bethlehem:

a) Samples were cultured according to (CLSI 2017) guidelines using different culture medias (macconkey agar, blood agar, bile escolin agar, thioglycolate broth and chocolate agar). If cultures were positive, bacteria was identified by colony morphology, Gram stain and other biochemical tests according to (CLSI 2017) guideline.

b) The antimicrobial susceptibility pattern of the isolated bacteria was determined by Kirby-Bauer disc diffusion method. The bacteria was tested against different antibiotics, and the inhibition zone was recorded as sensitive or resistant according to the Clinical and Laboratory Standards Institute (CLSI 2017) guideline.

3. The sensitivity pattern of those pathogens at Huda Al-Masri Pediatric Cancer Department at Beit Jala Governmental Hospital in Bethlehem was assessed.

3.6 Data analysis

The data were analyzed using Statistical Package for the Social Sciences (SPSS) version 23. The frequency was calculated to determine the prevalence of FN, while the mean ± standard deviation (STD) was computed for the purpose to find out other descriptive statistical analysis and to draw graphics for microbiology and culture sensitivity profiles.

3.7 Ethical Approvals

Approvals from the college of graduate studies at An-Najah National University and Institutional Review Board (IRB), and the Palestinian Ministry of Health (MOH) were obtained before starting the study.

A permission was acquired from the MOH in order to access of the electronic patients' medical record.

Chapter (4)

Results

4.1) Characteristics of pediatric ALL patients enrolled in the study

In this study, 83 pediatric ALL patients' medical records were reviewed and 63 pediatric ALL patients were found to be potentially eligible based on the inclusion criteria set during the period from 1/1/2014 to 1/6/2018. Twenty patients were excluded as they have incomplete medical files at Huda Al-Masri Pediatric Cancer Department at Beit Jala Governmental Hospital in Bethlehem.

Table (4.1) describes the pediatric ALL patients' characteristics who were included in our study. Males were about 32 (50.8%) and females were about 31 (49.2%). Patients who had been admitted with FN were about 59 (93.7%) of the pediatric ALL patients, while those who had never developed FN were about 4 (6.3%) of the pediatric ALL patients. The age range of the patients in this study was (1- 14) years old with mean average age was (5.7 ± 3.6). Fifty five 87.3% patients were diagnosed as having pre-B ALL type, while eight 12.7% were having T-cell ALL type. Three 4.8% patients were suffering from Down syndrome in addition to ALL.

Table (4.1): Patients' characteristics who are diagnosed with FN in the hospital.

Variable	N (%)
No. of Patient included in the study	63 (100)
No. of Patients diagnosed with FN	59 (93.7)
No. of patients who did not develop FN	4 (6.3)
Gender	
Males	32 (50.8)
Females	31 (49.2)
Average age (years) ± STD	5.7 ± 3.6 Range (1-14)
ALL- Type	
Pre-B ALL	55 (87.3)
T-cell ALL	8 (12.7)
Down syndrome	
Yes	3 (4.8)*
No	60 (95.2)

*All Down Syndrome All cases were of Pre-B All-Type.

4.2) Characteristics of FN episodes

4.2.1) Characteristics of FN episodes among Pediatric ALL patients

The total number of FN episodes were 161 episodes. The range of FN episodes were; (1- 6) episodes with the mean average number of FN episodes per patient was (2.5 ± 1.5). The prevalence of FN among ALL pediatric patients was found to be 94 %.

For all patients with FN the mean ± (STD) for each episode was calculated in order to describe the characteristics of the FN episodes. The mean initial temperature upon diagnosis with FN episodes was (39 C^0 ± 0.1; range: 39- 41 C^0), the average duration of fever was found to be (2.6 days ±

0.9; range: 1- 6). The mean initial ANC was (280 cell/µl ± 39; range: 0-500), while the mean hemoglobin level (Hgb) was (9.8 g/dL ± 0.4). The average duration of neutropenia lasted for (16.2 days ± 3.1; range: 4 -30). Moreover, the average onset of FN after chemotherapy administration was (4.1 days ± 1.2; range: 0- 20), the data are presented in table (4.2).

Table (4.2): The characteristics of FN episodes (mean of the mean ± STD).

Variable	Result Mean of the mean ± STD	Range
Initial temperature (C^0)	39 ± 0.1	(39-41)
Duration of fever (days)	2.6 ± 0.9	(1-6)
Initial ANC (cell/µl)	280 ± 39	(0-500)
Hgb level (g/dL)	9.8 ± 0.4	(6-13)
Duration of neutropenia (days)	16.2 ± 3.1	(4-30)
Onset of FN after chemotherapy treatment (days)	4.1 ± 1.2	(0-20)

4.3) Treatment settings and frequency of FN episodes (n= 161)

Forty episodes (25% of the episodes) occurred in pediatric ALL patients while they are in the induction phase 1A chemotherapy cycle, 24 (15%) of the attacks developed during induction phase 1B period, 16 (10%) developed in the consolidation cycle, 25 (22%) developed during the first reinduction period, 18 (11%) developed in the second reinduction cycle, 4 (3%) of the attacked occurred during the maintenance interim period and 19 (12%) of the FN episodes developed in the maintenance cycle. Three patients who were poor responders to the AIEOP-ALL protocol were on Flag

protocol when they had developed 5 (3%) FN attacks, data are shown in figure (4.1).

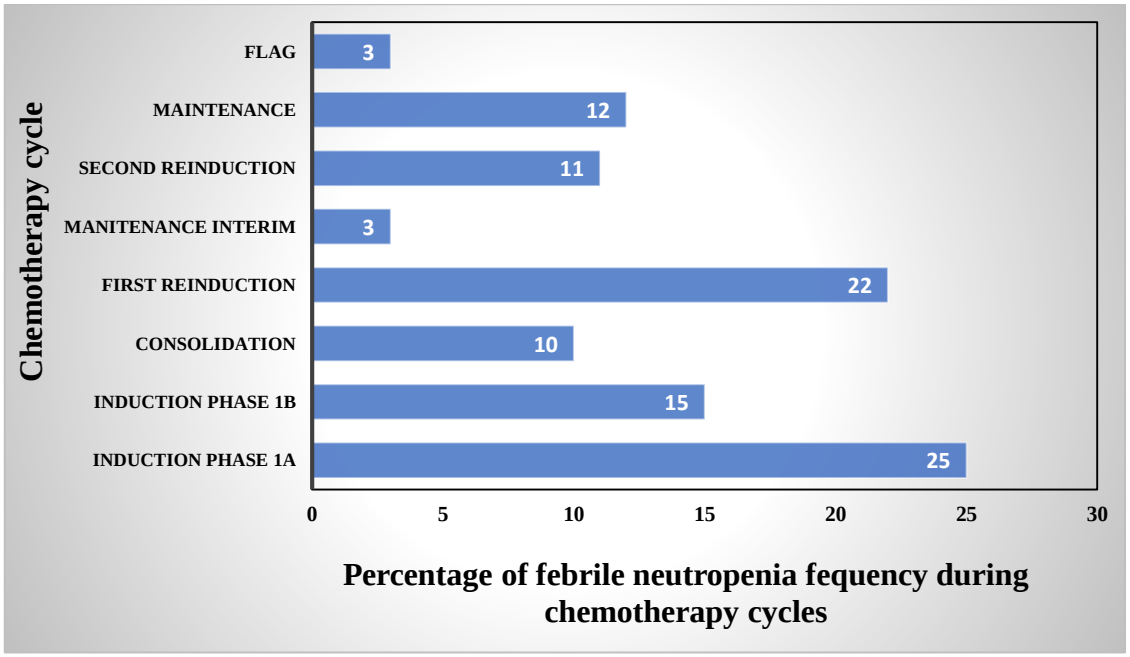


Figure (4.1): The frequency of FN episodes by chemotherapy cycle.

4.4) Etiology of FN episodes

Among 161 FN episodes, the majority 87 (54%) of the FN episodes were due to fever of non-bacterial origin. Among which, fever of unknown origin (FUO) accounted for 71 (44%) of them, while confirmed viral infections accounted for 8 (5%), among those viral infections 4 (2.5%) were reported to be *H1N1* infection, 3 (1.2%) of *Varicella zoster virus* (chicken pox infections) and 1(0.6%) was due to *Mumps virus* infection.

FN attacks due to fungal infections accounted for 6 (3.7%), one of which was confirmed to be *Candida albicans* (*C. albicans*). There were 2

(1.3%) FN attacks due to parasitic infections, one FN case was due to *Entemebia histolitica* while the other case was due to *Giardiasis* infection.

The total bacterial origin of FN episodes were found to be 72 (45%); in which Gram-positive bacteria and Gram-negative bacteria were about 45 (28%) and 27 (17%) of the total attacks respectively.

Gram-positive bacteria and Gram-negative bacteria reported to be 62.5% and 37.5% of the bacterial FN episodes respectively, the data are represented in table (4.3).

There were 2 (2.7%) FN episodes as a result of polymicrobial infections (mixed bacterial and non-bacterial infections) in which one case was due to (fungal infection + Gram-positive bacterial infection) while the other case was due to (fungal infection + Gram-negative bacterial infection), the data are illustrated in a figures (4.2) and (4.3).

Table (4.3): The etiology of FN episodes.

Etiology of FN episodes	N (%)
Non-bacterial origin of FN	**87 (54)**
- FUO	71 (44)
- Confirmed viral infections	8 (5)
- Fungal infections	6 (3.7)
Bacterial origin of FN	**72 (45)**
- Gram-positive bacteria	45 (28)
- Gram-negative bacteria	27 (17)
Polymicrobial infections	**2 (2.7)**
- Gram-positive bacteria+ fungal	1 (1.35)
- Gram-negative bacteria+ fungal	1 (1.35)
Total	**161 (100)**

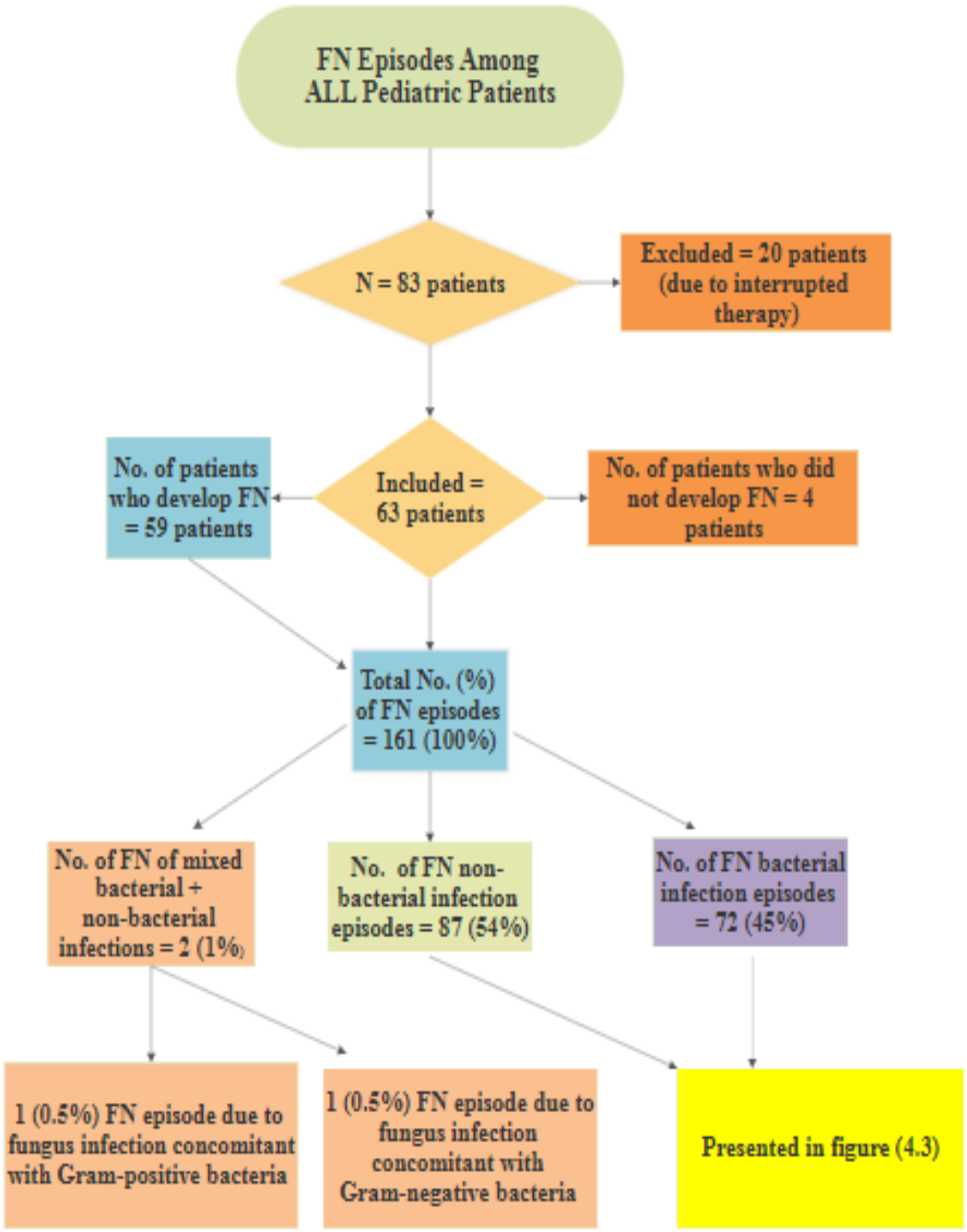


Figure (4.2): Etiology of FN episodes among ALL pediatric patients.

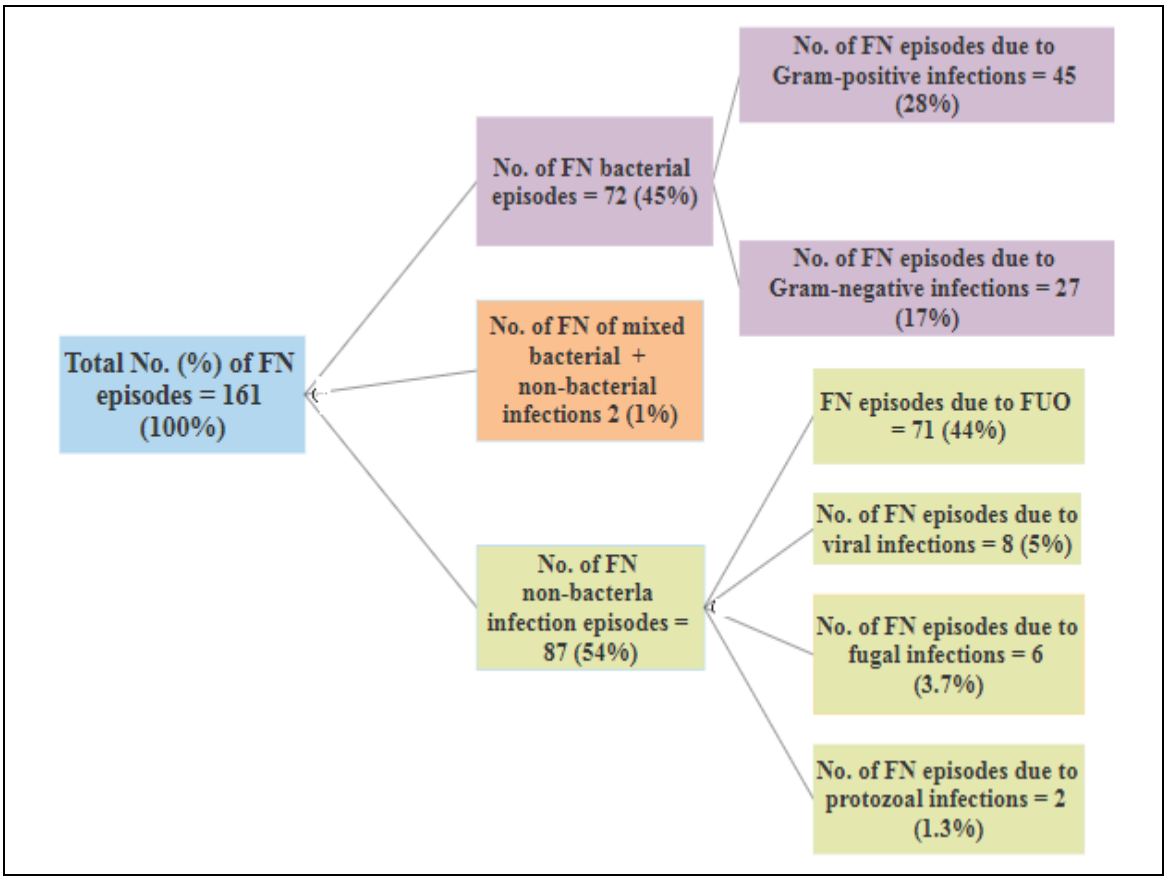
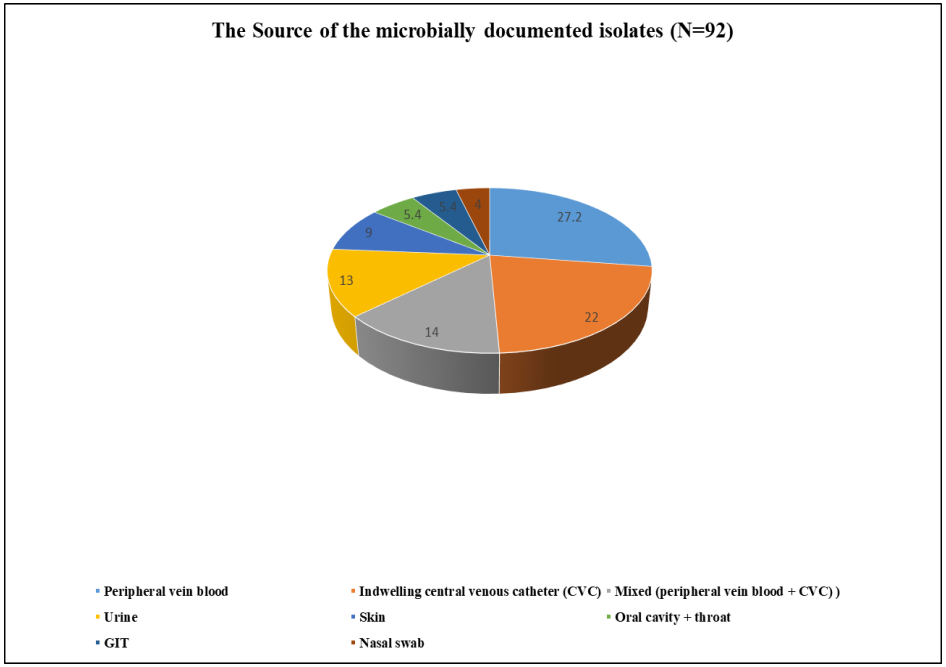


Figure (4.3): Continuation of the etiology of FN episodes among ALL pediatric patients.

4.5) Source of microbiologically documented isolates (N= 92)

The documentation of the foci of the FN infections in the population of the study was proven in 92 (57.1%) of the episodes. Among these 92 episodes; blood infection obtained from peripheral vein blood cultures was the most abundant and accounted for 25 (27.2%), followed by blood infection isolated from indwelling central venous catheter (CVC) cultures by about 20 (22%), mixed blood infections (isolated from both peripheral vein blood and CVC were about 13 (14%). Documentation from Urine isolates accounted for 12 (13%), skin infections were documented to be 8 (9%), infections of the throat and the oral cavity were as 5 (5.4%), infections related

to GIT were about 5 (5.4%), while infections obtained by nasal swab culture were about 4 (4%), the data are presented in figure (4.



Source of the microbially documented isolates (n = 92)	**%**
Peripheral vein blood	27.2
Indwelling central venous catheter (CVC)	22
Mixed (peripheral vein blood + CVC))	14
Urine	13
Skin	9
Oral cavity + throat	5.4
GIT	5.4
Nasal swab	4
Total	**100**

Figure (4.4): Source of microbiologically documented isolates (N=92).

4.6) The isolated organisms in (92) FN infections with respect to the infectious site in ALL pediatrics

1. **Peripheral vein blood isolates:** The majority of the microbially documented FN infections 25 cases, were reported from peripheral vein blood cultures, in which Gram-positive bacteria accounted for 16 (64%) while Gram-negative bacteria were about 8 (32%). In addition, one isolate 4% FN episode was due to fungal isolate from peripheral vein blood. Among Gram-positive bacteria *CONS* were the most abundant pathogen 36% followed by *S. aureus, Micrococcus spp.* and *MRSA* 16%, 8% and 4% respectively. Regarding Gram-negative bacteria *E. coli ESBL* was most causative agent for FN episode 12%, followed by *E. coli*, *Enterobacter spp*. and *K.pneumonia ESBL* 8%, 8% and 4% respectively.

2. **Isolates from indwelling central venous catheter (CVC):** positive cultures obtained from CVC were about 20 isolates. Gram-positive bacteria were the most responsible agent 16 (80%), Gram-negative bacteria accounted for 3 (15%) and fungal infection occurred in one episode 5%. For Gram-positive bacteria *CONS* was the most common microbe 65% while *Micrococcus spp*. was reported in 15% of the FN attacks. Among Gram-negative bacteria there were equal distribution 5% for *E. coli ESBL*, *P.aerogenosa* and *Chryseonomas luteola* (*C. luteola*).

3. **Mixed isolates of peripheral vein blood and CVC:** thirteen FN episodes were isolated from mixed infections from both peripheral vein blood and CVC, in which it has been documented that Gram-positive bacteria to be most abundant pathogen 46% followed be Gram-negative bacteria and fungus 31% and 23% respectively. Regarding Gram-positive bacteria *CONS* bacteria was the most frequent causative agent 38% then after came *Micrococcus spp.* by about 8%. The majority of Gram-negative bacterial infection due to were *E. coli ESBL* 23% while 8% were for *Acenobacter spp*. Among fungal infections, one FN attack was proven to be *C. albicans.*

4. **Urinary tract infections (UTI):** UTIs occurred in 12 FN episodes. Gram-negative bacteria accounted for 84% of the cases, while Gram-positive bacteria isolated from 16% of the UTI. For Gram-negative bacterial infections *E. coli ESBL* was found to be the most responsible pathogen for UTIs 43%, *E. coli* was reported in 25% of those infections, while *K.pneumonia ESBL* and *K.pneumonia* were occurring in the same frequency of 8%. UTIs due to Gram-positive bacteria were equally distributed 8% between *S. aureus* and *Enterococcus spp.*

5. **Skin infection:** skin infections were documented in 8 FN episodes, and it was divided between bacterial skin infections 5 (62.5%) and viral skin infection 3 (37.5%). Regarding bacterial skin infections; Gram-positive bacteria accounted for 60% and Gram-negative

bacteria were about 40%. Among Gram-positive bacteria skin infections *S. aureus* was found in 40% of the cases while *Streptococcus pyrogens (S. pyrogens)* was in 20% of the cases. Describing the pathogens that are responsible for Gram-negative bacterial infection; *Enterobacter cloacae (E. cloacae)* and *P. aerogenosa* both were occurring in 20% of those infections. However, all viral skin infections were due to *Varicella zoster virus* (*chicken pox infection).*

6. **Throat and oral cavity infections:** bacterial throat infections were all due to Gram-positive bacteria 60% all were due to *S. pyrogen* bacteria. Regarding other oral cavity infections; *Mumps* virus was responsible for 50% of the attacks while the rest of the cases were due to oral candidiasis.

7. **Gastro-intestinal tract infections (GITs):** there were five FN attacks related to GITs, in which 3 (60%) were due to fungus. The rest 2 (40%) were caused by parasites; one case was a result of *Entemebia histolitica* while the other was caused by *Giardia lambia*.

8. **Nasal culture isolates:** all the isolated cultures obtained from nasal swab 4 (100%) confirmed *H1N1* infection, the data were described in table (4.4).

Table (4.4): The infectious etiology to FN in pediatric ALL patients.

	Peripheral vein blood infections **(N= 25) (% =100)**	**Indwelling central venous catheter (CVC) infections** **(N= 20) (% =100)**	**Mixed peripheral vein blood + (CVC) infections** **(N = 13) (%=100)**	**Urine infections** **(N= 12) (100%)**	**Skin infections** **(N=8) (%= 100)**	**Throat and oral cavity infections** **(N=5) (%= 100)**	**GIT infections** **(N= 5) (%= 100)**	**Nasal swab cultures** **(N= 4) (% = 100)**
Gram-positive bacteria	*(N = 16) (% = 64)* *CONS* = 9 (36) *S. aureus* = 4 (16) *Micrococcus spp*. = 2 (8) *MRSA* = 1 (4)	*(N = 16) (% = 80)* *CONS* = 13 (65) *Micrococcus spp*. = 3 (15)	*(N = 6) (% = 46)* *CONS* = 5 (38) *Micrococcus spp*. = 1 (8)	*(N= 2) (% =16)* *S. aureus* = 1 (8) *Enterococcus spp*.= 1 (8)	*(N = 3) (% = 60)* *S. aureus* = 2 (40) *S. Pyrogens* = 1 (20)	*(N = 3) (% = 100)* *S. Pyrogen* = 3 (100)		

Gram-negative bacteria	*(N= 8) (%= 32)* *E.coli ESBL*= 3 (12) *E.coli* = 2 (8) *Enterobacter spp.* = 2 (8) *K. pneumonia ESBL*= 1 (4)	*(N= 3)(%= 15)* *E.coli ESBL*= 1 (5) *P.aerogenosa* = 1 (5) *Chryseonomas luteola* = 1 (5)	*(N = 4) (% = 31)* *E.coli ESBL* = 3 (23) *Acenobacter spp.* = 1 (8)	*(N = 10) (% = 84)* *E.coli ESBL*= 5 (43) *E.coli* = 3 (25) *K. pneumonia ESBL*= 1 (8) *K. pneumonia* = 1 (8)	*(N= 2) (% = 40)* *Enterobacter cloacae* = 1 (20) *P. aerogenosa* = 1 (20)			

Table (4.4): Continuation; The infectious etiology to FN in pediatric ALL patients.

	Peripheral vein blood infections **(N= 25) (% =100)**	**Indwelling central venous catheter (CVC) infections** **(N= 20) (% =100)**	**Mixed peripheral vein blood + (CVC) infections** **(N = 13) (%=100)**	**Urine infections** **(N= 12) (100%)**	**Skin infections** **(N=8) (%= 100)**	**Throat and oral cavity infections** **(N=5) (%= 100)**	**GIT infections** **(N= 5) (%= 100)**	**Nasal swab cultures** **(N= 4) (% = 100)**
Fungus	*(N=1) (%=4)* Fungus = 1 (4)	*(N = 1)(% =5)* Fungus = 1 (5)	*(N= 3) (%= 23)* Fungus = 3 (23) (One is *C.albicans)*					
Others					**Viral skin infection (N = 3) (%=37.5)**	**Other infections (N= 2) (40%)**	***Stool culture (N= 3) (% = 60)***	***Viral infections (N =4) (% = 100)***

					Varicella zoster (chicken pox) = (3) (100%)	**Fungus** = 1 (50) **Virus (*Mumps*)** = 1 (50)	Fungus = 3 (60) ***Parasites*** *(N= 2) (% = 40)* *Entemebia histolitica* (1) (20) *Giardia lambia* (1) (20)	*H1N1* = 4 (100)

4.7 Spectrum of Gram-positive bacterial isolates with respect to the most common source of infection

Among 72 FN episodes due to bacterial infections, Gram-positive bacteria were found to be the most common cause for bacterial FN episodes 45 (62.5%). *Coagulase negative staphylococcous CONS* bacteria was the most abundant 27 (58.7%) of the Gram-positive culture samples, followed by *S. aureus* and *Micrococcous spp*. 7 (15.2%) and 6 (13%) respectively. All those three pathogens were mostly isolated from blood samples. *S. pyrogen* were the most common cause for throat infections 3 (6.5%). *MRSA*, *Enterococcous spp*. *Group A streprococcous* occurred in the frequency of one infection (2.2%) for each and were obtained from sample of blood, urine and throat respectively, data are shown in table (4.4). During one FN attack, in which the sample was isolated from blood there was a mixed infection of Gram-positive bacteria *(Microccoucus spp.)* and fungal infection, this episode was not represented in table (4.5) due to complexity in calculation.

Table (4.5): Spectrum of Gram-positive bacteria in respect to the most source of infection.

Gram-positive bacteria (N= 45) (62.5)%		Most common source
Coagualse negative staphylococcous	27 (58.7)	Blood
Staphylococcous aureus	7 (15.2)	Blood
Micrococcous spp.	6 (13)	Blood
Streptococcous pyrogens	3 (6.5)	Throat
MRSA	1 (2.2)	Blood
Enterococcous spp.	1 (2.2)	Urine
Group A streptococcpous	1 (2.2)	Throat

4.8) Spectrum of Gram-negative bacterial isolates in respect to the most common source of infection

Gram-negative bacterial infection accounted about 27 (37.5%) of bacterial FN attacks. In which *E.coli ESBL* was the most abundant pathogen 12 (42.9%) and was mostly isolated from blood samples. *E.coli* and *Enterobacter spp.*, occurred in the frequency of 5 (17.9%) and 4 (14%) respectively, and were predominately isolated from urine and blood respectively. *K.peumonia ESBL* was equally found in urine and blood samples, in addition *P. aeroginosa* was equally isolated from blood and skin samples both pathogens occurred in the frequency of 2 (7.1%). *K. pneumonia*, *C. luteola* and *Acinobacter spp*. all were found in one infection (3.6%) of FN episodes for each bacteria, *K. pneumonia* was isolated mostly from urine while *C. luteola* and *Acinobacter spp* were both isolated from blood samples, data are present in table (4.5). During one FN episode of septicemia there was a mixed infection of Gram-negative bacteria *(K.peumonia ESBL)* and fungal infection, during this episode the patient died, this attack was not included in table (4.6) due to complexity in calculations.

Table (4.6): Spectrum of Gram-negative bacteria in respect to the most source of infection.

Gram-negative bacteria = (27) (37.8)%		**Most common source**
E.coli ESBL	12 (42.9)	Blood
E.coli	5 (17.9)	Urine
Enterobacter spp.	4 (14)	Blood
Klebsiella pneumonia ESBL	2 (7.1)	Blood/ Urine
Pseudomonas aeroginosa	2 (7.1)	Blood/Skin
Klebsiella pneumonia	1 (3.6)	Urine
Chryseonoma luteola	1 (3.6)	Blood
Acinobacter spp.	1 (3.6)	Blood

4.9) Antibiotic sensitivity pattern of Gram-positive bacteria

The sensitivity profile for Gram-positive bacteria revealed that; *CONS* isolates were found to be 100% sensitive to teicoplanin (TEP), vancomycin (VAN), ceftriaxone (CTR) and cefotaxime (CTX). However CONS bacteria was found to be 48.1% sensitive for oxacillin (OX), while its sensitivity pattern towards cefuroxime (CXM), meropenem (MER), ampicillin (AMP) and penicillin (PEN) was as follow; 37%, 29.6%, 18.5% and 7.4% respectively.

Regarding *S. aureus*, it was found to be 100% sensitivity for vancomycin (VAN) and oxacillin (OX). While it showed 71.4 % sensitivity towards ceftriaxone (CTR) and cefotaxime (CTX). Lower sensitivity pattern was present with teicoplanin (TEP) 57.1%. On the over hand, for peniciilin

(PEN) and meropenem (MER) it was 28.6%, while its sensitivity for cefuroxime (CXM) and ampicillin (AMP) was 14.3%.

The sensitivity profile for *Micrococcus spp*. was lower than the previous mentioned bacteria. It had been 83.3% sensitive towards teicoplanin (TEP), vancomycin (VAN), ceftriaxone (CTR) and cefotaxime (CTX). It was 66.6% sensitive to meropenem (MER). However, it was 33.3% sensitive towards cefuroxime (CXM) while it showed 16.6% towards penicillin (PEN) and ampicillin (AMP). There was no available data regarding *Micrococcus spp*. towards oxacillin (OX), the data are presented in table (4.7).

Table (4.7): Antibiotic sensitivity pattern for Gram-positive bacteria.

Gram-positive Bacteria	TEP N%	CXM N%	PEN N%	VAN N%	MER N%	CTR N%	CTX N%	OX N%	AMP N%
CONS **(n= 27)**	27 (100%)	10 (37%)	2 (7.4%)	27 (100%)	8 (29.6%)	27 (100%)	27 (100%)	13 (48.1%)	5 (18.5%)
S. aureus **(n = 7)**	4 (57.1%)	1 (14.3%)	2 (28.6%)	7 (100%)	2 (28.6%)	5 (71.4%)	5 (71.4%)	7 (100%)	1 (14.3%)
Micrococcus Spp**. (n= 6)**	5 (83.3%)	2 (33.3%)	1 (16.6%)	5 (83.3%)	4 (66.6%)	5 (83.3%)	5 (83.3%)	NA	1 (16.6%)

4.10) Antibiotic sensitivity pattern of Gram-negative bacteria

Gram-negative bacteria sensitivity pattern showed that *E.coli* isolates were 100% sensitive to amikacin (AK), cefotaxime (CTX) and ceftriaxone (CTR). The *E.coli* isolates were found to be 80% sensitive to gentamycin (GEN), combination of piperacilline/tazobactam (PIT), ceftazidine (CFZ) and cefuroxime (CXM). The sensitivity pattern towards meropenem (MER) was revealed to be 60%, while the sensitivity behavior against ciprofloxacin (CIP) and cefixime (CFM) was found to be 20%. There were no available data for *E.coli* isolates sensitivity pattern against imipenem (IMP) and ertapimen (ERT).

On the other hand, *E.coli ESBL* isolates showed 83.3% sensitivity pattern to amikacin (AK), meropenem (MER), imipenem (IMP) and etrapemen (ERT). 66.6% sensitivity behavior was shown against the combination of piperacillin/tazobactam (PIT), while for gentamycin (GEN) it was 50% and for ciprofloxacin (CIP) it was 41.6%. *E.coli ESBL* revealed 33.3% sensitivity pattern against cefixime (CFM), ceftazidime (CFZ), cefuroxime (CXM), ceftriaxone (CTR). However, for cefotaxime (CTX) it was 25%.

For *K.pneumonia* isolate it was 100% sensitive for amikacin (AK), ciprofloxacin (CIP), gentamycin (GEN), combination of piperacillin/tazobactam (PIT), meropenem (MER), cefixime (CFM), cefotaxime (CTX), ceftazidime (CFZ), cefuroxime (CXM) and ceftriaxone

(CTR). There were no available data for *K.pneumonia* isolate sensitivity pattern against imipenem (IMP) and ertapimen (ERT).

Regarding *K.pneumonia ESBL* isolates, 100% sensitivity pattern against amikacin (AK), gentamycin (GEN), combination of piperacillin/tazobactam (PIT) and meropenem (MER). 50% sensitivity behavior was shown against ciprofloxacin (CIP) and cefixime (CFM). *K.pneumonia ESBL* isolates were found to be totally resistant to cefotaxime (CTX), ceftaxidime (CFZ), cefuroxime (CXM) and ceftriaxone (CTR). There were no available data for *K.pneumonia ESBL* isolates sensitivity pattern against imipenem (IMP) and ertapimen (ERT), table (4.8) illustrates these findings.

Table (4.8): Antibiotic sensitivity pattern of Gram-negative bacteria.

Gram-negative bacteria	AK N%	CIP N%	GEN N%	PIT N%	MER N%	IMP N%	ERT N%	CFM N%	CTX N%	CFZ N%	CXM N%	CTR N%
E.coli **(n= 5)**	5 (100%)	1 (20%)	4 (80%)	4 (80%)	3 (60%)	NA	NA	1 (20%)	5 (100%)	4 (80%)	4 (80%)	5 (100%)
E.coli ESBL **(n= 12)**	10 (83.3%)	5 (41.6%)	6 (50%)	8 (66.6%)	10 (83.3%)	10 (83.3%)	10 (83.3%)	4 (33.3%)	3 (25%)	4 (33.3%)	4 (33.3%)	4 (33.3%)
K.pneumonia **(n= 1)**	1 (100%)	1 (100%)	1 (100%)	1 (100%)	1 (100%)	NA	NA	1 (100%)	1 (100%)	1 (100%)	1 (100%)	1 (100%)
K.pneumonia ESBL **(n= 2)**	2 (100%)	1 (50%)	2 (100%)	2 (100%)	2 (100%)	NA	NA	1 (50%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)

Chapter 5

Discussion & Conclusion

5.1- Discussion

Pediatric patients with hematologic malignancies are at higher risk of developing FN. This is due to the dual action of the disease pathophysiology and the aggressive chemotherapy treatment on suppressing the bone marrow in addition to the treatment protocol's long term duration. This exposes the patients to serious life-threatening infections that could be lethal. Those infections may be caused by bacteria, viruses, fungus and many other microbes. This study was conducted to determine the prevalence of FN among pediatric leukemic patients at Huda Al-Masri Pediatric Cancer Department at Beit Jala Governmental Hospital in Bethlehem. In addition to reviewing the patients' clinical profiles, evaluating the microbiology profile of the pathogens that are responsible for FN and assessing their antibiotic sensitivity behavior.

In this study the prevalence of FN was as high as 94% with the mean of 2.5 episodes/patient. The prevalence of FN in hematological oncology patients reported to be as high as eighty percent of the patients [*Klastersky*, 2004], while others reported it to be 86.9% with the mean of episode occurrence per patient is 2.7 [*Li et al.*, 2017]. A Thai study carried out by [*Limvorapitak and Khawcharoenporn*, 2015] found that the incidence of FN was about 14.9% in hematology malignant patients who are even receiving

very intensive chemotherapy, this is as a result for the use of prophylactic G-CSF which has had a significant role in reducing the evidence of FN occurrence. The explanation of the high prevalence of FN in this study may due to not using a prophylactic dose of G-CSF after chemotherapy session. As it is practiced at Huda Al-Masri Pediatric Cancer Department to use G-CSF only when the neutropenia is classified as grade (IV) in which the ANC (< 500 cell/µl). The practice of administering a prophylactic single dose of G-CSF was found to reduce morbidity and mortality among cancer patients. Moreover, it was believed to be cost effective compared to treating FN infections [*Klastersky et al.*, 2016; *Limvorapitak and Khawcharoenporn*, 2015; *Ludwig et al.*, 2019].

The mean number of episodes/ patient in this report is (2.5 ± 1.5), this is nearly similar to the findings documented by [*Li et al.*, 2017] in which it was 2.74 episodes/ patient.

In the current study, the initial temperature at the onset of FN was 39 C°, this is similar to what had been documented by [*Vathana et al.*, 2017]. The median onset of FN after chemotherapy cycle administration was 4.1 days, this is similar to the findings done by [*Park et al.*, 2010] who reported the onset of FN to be 4.3 days after administration of chemotherapy. Rajendranath and co-workers documented it to be 3 days from last chemotherapy [*Rajendranath et al.*, 2014]. However, a delay in the onset of FN since the administration of last chemotherapy to 8.8 days [*Latiff et al.*, 2002; *Vathana et al.*, 2017] and 14 days was also reported [*Badr et al.*, 2016;

Louw et al., 2010]. This study found that the initial ANC at the time of FN onset was 280 (cell/µl), this is somehow similar to what had been reported by [*Vathana et al.*, 2017] who found the initial ANC to be 223 (cell/µl), Badr and his team documented the initial ANC as 225 (cell/µl) [*Badr et al.*, 2016]. Other studies had found the initial ANC to be 300 (cell/µl) [*Rajendranath et al.*, 2014] and others found it to be 100 (cell/µl) [*Louw et al.*, 2010]. The mean duration of fever in the present study is found to last for 2.6 days, while it was documented to be 4 day [*Vathana et al.*, 2017], 5.3 days [*Yilmaz et al.*, 2008], 7days [*Rajendranath et al.*, 2014] and 10 days as found by [*Park et al.*, 2010]. Low hemoglobin level at the time of receiving chemotherapy was found to be a risk factor for FN [*Salar et al.*, 2012], we found that the mean hemoglobin level at the onset of FN was 9.8 (g/dL). The mean duration of neutropenia in the current report is 16.2 days, Bader and coworker found it to be 11.2 days [*Badr et al.*, 2016] while other studies had found it to be 7.3 days [*Vathana et al.*, 2017] and 3 days [*Rajendranath et al.*, 2014]. It has been obvious that the characteristics of FN episodes were variable among studies.

The chemotherapeutic agents included in the induction phase (1A) are: vincristine, steroids, daunarubicin/ anthracycline and L-asparaginase. These agents can induce a complete remission of ALL in the rate of 95%. CNS prophylactic measure is administered in the form of intrathecal (IT) methotrexate. Anticancer medications included in Induction phase (1B) are: cyclophosphamide, mercaptopurine, cytarabine and intrathecal methotrexate. Anti-neoplastic agents used in the reinduction phases are:

vincristine, steroid, high dose cytarabine, methatroxate (IV) with leucoverine, IT methotrexate, cyclophosphamide, L-asparginase, daunarubicine and 6-thioguanine [*Conter et al.*, 2014]. Thus, this very aggressive chemotherapy will cause severe myelosuppression in order to achieve complete remission of ALL cells. Hence, as expected, in this report most of FN episodes were recorded during the induction phases (1A and 1B) by about 40% and FN attacks during the two reinduction phases (first and second reinduction) were reported to be about 33%. These findings were similar to the work done by [*Afzal et al.*, 2009; *Badr et al.*, 2016; *Crawford et al.*, 2004; *Di Blasi et al.*, 2018; *Hassan*, 2009; *Inaba et al.*, 2017; *Lex et al.*, 2001; *Li et al.*, 2017; *O'Connor et al.*, 2014]. However, Kar and his colleagues found that FN episodes occurred more frequently in the reinduction phase [*Kar et al.*, 2017]. While Yilmazand and his team reported the most incidence of FN during the consolidation phase [*Yilmaz et al.*, 2008].

In the current study 54% of the FN episodes were due to non-bacterial origin, while 45% of the FN attacks were as a result of bacterial infections. FUO documented to be 44% of the total FN attacks. This result fits in the range of (13.9-79%) which many recent worldwide studies recorded in describing FUO [*Castagnola et al.*, 2007; *Di Blasi et al.*, 2018; *Hakim et al.*, 2009; *Jeddi et al.*, 2010; *Kar et al.*, 2017; *Lehrnbecher et al.*, 2004; *Lex et al.*, 2001; *N Ozdemir et al.*, 2016].

Confirmed viral infection by laboratory findings in the present document was in the rate of 5%. This is somehow similar to other studies which detected viral infections in range of (1-8%) [*Castagnola et al.*, 2007; *Di Blasi et al.*, 2018; *Lehrnbecher et al.*, 2004; *N Ozdemir et al.*, 2016; *Vathana et al.*, 2017]. Hakim and his team reported viral infection in FN episodes to be as high as 34% [*Hakim et al.*, 2009], the authors explained this high prevalence of viral infections to be due to the modern available diagnostic tools for viruses (antigen detection, virus cultures and molecular diagnostic methods PCR) that they own in their institute.

Confirmed laboratory fungal infections obtained from this report were in the rate of 3.7%, this finding is nearly similar to other surveys, where fungal infections were seen in rate of (0.9-10.4%) [*Cortes et al.*, 2013; *Di Blasi et al.*, 2018; *H El-Mahallawy et al.*, 2005; *H A el-Mahallawy et al.*, 2002; *Meidani et al.*, 2018; *N Ozdemir et al.*, 2016; *Roongpoovapatr and Suankratay*, 2010; *Vathana et al.*, 2017]. In a Malaysian stufy, fungemia accounted around 20% of the FN attacks of positive blood culture [*Latiff et al.*, 2002]. However Hakim and his colleagues had reported no fungal infections in their study, as they referred this for administering prophylactic antifungal agents [*Hakim et al.*, 2009].

In this study parasitic infections were about 1.3%. In a Colombian study the incidence for parasitic infections was much higher than the current findings in this thesis as it stated to be 7.1% [*Cortes et al.*, 2013].

In this work, the documented microbially infections' foci was recorded in 92 (57%) of the total FN episodes, this is almost similar to other work [*Lex et al.*, 2001; *Z C Ozdemir et al.*, 2016]. However, these microbially documented infections were much higher than that recorded by Kar's group and Gupta's group where it was reported to be (36% and 22% respectively) [*Gupta et al.*, 2011; *Kar et al.*, 2017]. In this report (BSI) were the most abundant infections. Positive cultures of BSI from peripheral vein blood and indwelling central venous catheter (CVC) accounted for 27% and 22% respectively. While mixed peripheral vein blood and CVC positive cultures were reported to be 14%. These results correlates with many other studies [*Bakhshi et al.*, 2008; *Cortes et al.*, 2013; *Kar et al.*, 2017; *Rajendranath et al.*, 2014; *Rezaee et al.*, 2017; *Roongpoovapatr and Suankratay*, 2010; *Siddaiahgari et al.*, 2014].

The second found foci for positive cultures was obtained from urine samples 13% of the isolates. This matches the findings by other researchers [*Cortes et al.*, 2013; *Kar et al.*, 2017; *Mian*, 2013; *Siddaiahgari et al.*, 2014]. In the current study skin infections were third in occurrence by 9%, followed by other documented infections' sites such as (oral cavity, GIT and positive nasal swabs) (5.4%, 5.4% and 4% respectively). However, Mian reported the sources of infections in his study to be in the order of: catheter related infections, gastrointestinal infections, urinary tract infections, skin and soft tissue infections and lastly lung infections (12.2%, 6.4%, 5%, 5% and 2.1% respectively) [*Mian*, 2013].

In this work the incidence of FN due to bacterial infections was about 45% of the total FN attacks. Gram-positive and Gram-negative bacteria accounted about 62.2 % and 37.8% of the bacterial episodes respectively. These findings correlates with many other studies in which Gram-positive bacteria are predominant in bacterial FN episodes among haematological oncology patients [*Al-Sweedan et al.*, 2012; *Butt et al.*, 2004; *H El-Mahallawy et al.*, 2005; *Lex et al.*, 2001; *Mamishi et al.*, 2005; *Rezaee et al.*, 2017; *Vathana et al.*, 2017]. However, other reports had revealed that Gram-negative bacteria were found to be the most pathogens isolated in FN infections [*Almaziny*, 2014; *Cortes et al.*, 2013; *Gupta et al.*, 2011; *Hassan*, 2009; *Jeddi et al.*, 2010; *Kar et al.*, 2017; *Lakshmaiah et al.*, 2015; *Latiff et al.*, 2002; *C Y Lee et al.*, 2009; *J H Lee et al.*, 2016; *Mian*, 2013; *Rajendranath et al.*, 2014; *Roongpoovapatr and Suankratay*, 2010; *Siddaiahgari et al.*, 2014; *Taj et al.*, 2015; *Zhai et al.*, 2015]. Equal prevalence for both Gram-positive and Gram-negative bacteria was detected by many other groups [*Jeddi et al.*, 2010; *Lakshmaiah et al.*, 2015; *N Ozdemir et al.*, 2016].

The findings reported that among Gram-positive bacteria, *CONS* bacteria were the most isolates in occurrence 62.5%, followed by *S. aureus* and *Micrococcous spp*. (15.2% and 13%) respectively. Those three Gram-positive pathogens are among the skin normal flora. Moreover, in this study they were the most leading cause for (BSI) (isolated from samples of; peripheral vein blood, indwelling central venous catheter (CVC) and mixed peripheral vein blood and CVC), this was similar to what previously stated

[*Al-Sweedan et al.*, 2012; *H El-Mahallawy et al.*, 2005; *N Ozdemir et al.*, 2016; *Rezaee et al.*, 2017; *Roongpoovapatr and Suankratay*, 2010]. However, Almaziny found that among Gram-positive bacteria *S.epidermidis* and *S. aureus* were the most common isolated from urine, blood and pus cultures [*Almaziny, 2014; Rajendranath et al., 2014*]. *Staphylococcus aureus* was documented to be the main Gram-positive pathogen responsible for FN infection followed by *Enterococcus spp*. [*Cortes et al.*, 2013; *Lakshmaiah et al.*, 2015; *N Ozdemir et al.*, 2016; *Taj et al.*, 2015].

This report also showed that Gram-negative bacteria were about 37.5% of the bacterial FN attacks. *E.coli ESBL* was the predominant Gram-negative bacteria 42.9%, it was the main causative microbe for BSI and urine infections. Febrile neutropenia infections with *E.coli* were recorded to be 17.9%, and urine and blood samples were the most common source for *E.coli*. The incidence of *K. pneumonia ESBL* was 7.1% of the Gram-negative infections isolated from peripheral vein blood and urine samples. Urine infection with *K. pneumonia* isolates were about 3.6% of the Gram-negative bacteria. On the other hand, *K. pneumonia* was most common isolated Gram-negative pathogen from blood cultures in an Iranian study [*Rezaee et al.*, 2017]. *Escherichia coli* followed by *Pseudomonas* were the most predominant isolates from urine and blood in a study done by [*Almaziny*, 2014; *Siddaiahgari et al.*, 2014]. However, *E.coli* succeeded by *K. pneumonia* were the main cause for FN infections in hematological malignant patients [*Cortes et al.*, 2013; *Lakshmaiah et al.*, 2015; *Mangaraj et al.*, 2015; *N Ozdemir et al.*, 2016; *Taj et al.*, 2015]. Other scientists stated

that bacteremia with Gram-negative pathogens were most common with *K. pneumonia* followed by *E.coli* [*Al-Sweedan et al.*, 2012; *Rezaee et al.*, 2017]. In a study done at the National Cancer Institute in Cairo /Egypt, found that *Acinetobacter* spp. followed by *Pseudomonas* were the most Gram-negative microbes responsible for BSIs [*H El-Mahallawy et al.*, 2005]. Rajendranath and co-workers found that *Pseudomonas* was the most frequent Gram-negative bacteria detected in FN among leukemic patients [*Rajendranath et al.*, 2014].

The current study reported the susceptibility for the most common Gram-positive isolates *CONS*; were found to be absolutely sensitive to teicoplanin (TEP), vancomycin (VAN), ceftriaxone (CTR) and cefotaxime (CTX). However *CONS* bacteria was found to be (48.1%, 37%, 29.6%, 18.5% and 7.4%) sensitive towards oxacillin (OX), cefuroxime (CXM), meropenem (MER), ampicillin (AMP) and penicillin (PEN) respectively. *S. aureus*, was found to be 100% sensitive for (VAN) and (OX). While it showed 71.4 % sensitivity towards (CTR) and (CTX). *Staphylococcus aureus* sensitivity profiles towards (TEP), (PEN), (MER), (CXM) and (AMP) were (57.1%, 28.6%, 28.6% 14.3% and 14.3%) respectively.

Micrococcus spp. were less sensitive than *CONS* and *S.aureus.* Its sensitivity behavior towards (TEP), (VAN), (CTR), (CTX), (MER), (CXM), (PEN) and (AMP) was as the following (83.3%, 83.3%, 83.3%, 83.3%, 66.6%, 33.3%, 16.6% and 16.6%) respectively. While, there was no records about its sensitivity to (OX). Rezaee found that 88% of *CONS* and 37% of

S. aureus were resistant to (OX) [*Rezaee et al.*, 2017]. Zhai and his team found that, *S. aurues* isolates were 100% sensitive to (VAN), (TEP), linzolide and teigecycline, while it was 50% sensitive to (CIP) [*Zhai et al.*, 2015]. Sigurdardottir's team confirmed that *CONS* isolates were 93%, and 98.8% sensitive to both penicillin G and aminoglycoside [*Sigurdardottir et al.*, 2005]. In another study, *Staphylococcus* bacteria was sensitive to amoxicillin/calvulinic acid combination, Piperacillin/tazobactam combination, carbapenems and to the third generation cephalosporins [*Siddaiahgari et al.*, 2014]. Lakshmaiah *et al*., Taj *et al*., and Rajendranath *et al.*, reported that the Gram-positive Bactria including (*MRSA*) and *Enterococcus ssp*. in their studies were sensitive to vancomycin, eicoplanin, linezolid and levofloxain [*Lakshmaiah et al.*, 2015; *Rajendranath et al.*, 2014; *Taj et al.*, 2015].

Gram-negative bacteria are more vigorous and more virulence than Gram-positive bacteria, that can induce higher rates of morbidity and mortality to human beings causing very serious life-threatening infections. Moreover, Gram-negative bacteria reported to have higher rates of antibiotic resistance, this is due to many factors such as; genetic manipulation within the Gram-negative bacteria as a defensive mechanism against antibiotics and due to the misuse of antibiotics' prescription [*Livorsi et al.*, 2011].

Thus it is of great importance for each hospital to detect the antibiotic sensitivity patterns among both Gram-positive and Gram-negative bacteria in order to set up the appropriate empiric antimicrobial policy.

In the present report, *E.coli* were found to be 100% sensitive to amikacin (AK), cefotaxime (CTX) and ceftriaxone (CTR), 80% sensitive to gentamycin (GEN), combination of piperacilline/tazobactam (PIT), ceftazidine (CFZ) and cefuroxime (CXM) 60% sensitive to meropenem (MER), 20% sensitive to both ciprofloxacin (CIP) and cefixime. While there was no available data for its sensitivity pattern against to imipenem (IMP) and ertapimen (ERT). In a study carried out in Pakistan, *E.coli* were found to be sensitive to cefoperazone/salbactam (96%), (AK) (88%), carbapenem (66%) and (PIT) (48%) [*Taj et al.*, 2015]. Another study showed that *E.coli* isolated were 100% sensitive to colistin and teigecycline, 68% sensitive to carbapenem and 37% sensitive to (CTR) [*Mangaraj et al.*, 2015]. Lakshmaiah and his colleagues found that *E.coli* isolates were 100% sensitive to (IMP), while it was found to be resistant to (PIT) [*Lakshmaiah et al.*, 2015]. A Chinese study found that, *E.coli* isolates were sensitive to (MER), (IMP), (PIT), (CFZ) and (CIP) in the following rate: 100%, 100%, 95%, 72.2% and 49.5% respectively [*Zhai et al.*, 2015].

In this thesis, *E.coli ESBL* showed 83.3% sensitivity pattern to (AK), (MER), (IMP) and (ERT), 66.6% sensitive to (PIT), 50% sensitive to (GEN), 41.6% sensitive to (CIP), 33.3% sensitive to (CFM), (CFZ), (CXM), (CTR), and 25% sensitive to (CTX).

Taj and co-workers, found that K.pneumonia isolates were 41% sensitive to (PIT) and 50% to (AK) [*Taj et al.*, 2015]. Mangaraj's group found that *K.pneumonia* isolates were 100% sensitive to colistin and (85.7%)

to teigecyclin, 64.3% sensitive to carbapenems and completely resistant to (CTR) and (PIT) [*Mangaraj et al.*, 2015]. Zhai and co-workers reported that, *K.pneumonia* isolates were sensitive to (MER), (IMP), (PIT), (CIP) and (CFZ) as following: 96%, 96%, 96%, 88% and 75% respectively [*Zhai et al.*, 2015]. On the other hand, the current findings in this study revealed that *K.pneumonia* positive cultures were absolutely sensitive to (AK), (CIP), (GEN), (PIT), (MER), (CFM), (CTX), (CFZ), (CXM) and (CTR). There were no available data for *K.pneumonia* sensitivity for (IMP) and (ERT). However, this may not be conclusive as only one positive culture sample was for *K. pneumonia* in this study.

Moreover, the findings documented that *K.pneumonia ESBL* isolates were totally sensitive (AK), (GEN), (PIT) and (MER). While, *K.pneumonia ESBL* positive cultures were 50% sensitive to (CIP) and (CFM). In addition to that, this study found that *K.pneumonia ESBL* isolates were **completely resistant** to (CTX), (CFZ), (CXM) and (CTR). There were no available data for *K.pneumonia ESBL* isolates sensitivity pattern against (IMP) and (ERT). However, these results may not be representative as this study had included only two *K.pneumonia ESBL* samples.

A study done by Rezaee's group found that the isolated Gram-negative pathogens were sensitive to (AMP), (GEN), (CTR), (CFZ), (AK), (IMP), and (CIP) in the following rate: (61.5%, 66.7%, 37.9%, 16%, 61.5%, 85.2%, and 93.3%) respectively [*Rezaee et al.*, 2017]. Rajendranath's team found that the isolated Gram-negative bacteria were 100% sensitive to

carbapenem, 95.2% sensitive to both (AK) and (PIT) and 33.3% sensitive to (CFZ) [*Rajendranath et al.*, 2014]. [*Siddaiahgari et al.*, 2014] found that Gram-negative isolates (*E.coli, K.pneumonia and pseudomonas*) were sensitive to (PIT), carbapenems and (GEN).

It has been clear that there is world-wide variation in: the causative pathogen for FN and the susceptibility pattern of the pathogen to antibiotics were found to be different between different institutes. This urges the need of each hospital to individualize, determine and evaluate the current causative microbes that are responsible to FN. This should be done along with their microbiology profile. Then-after, each institute should take the step to layout its own appropriate empiric antibiotic therapy irrespective to the international guidelines.

According to the findings Huda Al-Masri Pediatric Cancer Department should specify and reassess the microbiology profile for the bacteria that are responsible for FN infections. Moreover, the hospital should tailor its empiric antimicrobial therapy according to the new findings.

By the year 2017, Beit Jala Governmental Hospital in Bethlehem started to record the antibiogram. The finding of the antibiogram for the years (2017, 2018) was done only for Gram-negative bacteria. In which *E. coli ESBL* sensitivity profile against GEN, CXM, CTX, CFZ was found to be 72%, 53%, 60% and 60% respectively in the year 2017. While it fell to 66%, 39%, 36% and 36% respectively in the year 2018. The antibiogram results for *K. pneumonia ESBL* was reported to be sensitive for the above

mentioned medications in the rate of 78%, 53%, 58% and 58% respectively in the year 2017. While in 2018 it was documented to be 73%, 41%, 33% and 33% respectively. It had been obvious that those Gram-negative bacteria are becoming more resistant to antibiotics, which should alert the infection control body in hospital for the responsibility regarding the misuse of antibiotics and for taking considerations for prophylactic steps for FN, starting from G-CSF prophylactic dose after chemotherapy in addition to medical staff, patients and parents education regarding the self-hygiene and environment hygiene.

5.2- The importance of the study

According to our knowledge, this is the first surveillance study to be conducted in Palestine to:

1. Determine the prevalence of FN among Pediatric Leukemic patients.
2. Identify the causative pathogens that responsible for FN infections.
3. Review the microbiology profile for each pathogen by means of sensitivity patterns towards the antimicrobial agents.

This survey can help Huda Al Masri Pediatric Cancer Department to establish its own empiric antibiotic policy according to the present behavior of the pathogens towards different antibiotics. However, such study should be conducted periodically to monitor and re-evaluate any changes in the

microbes' susceptibility to antibiotics. And thus, according to the obtained results the empiric antibiogram should be reassessed and updated.

5.3- Limitations of the study

This study is retrospective study, that is based on administrative data in the patient's medical records, thus there were some limitations in obtaining the fully detailed clinical and laboratory information. Moreover, there were constraints in the sample size as the study was conducted in a single center in Palestine.

5.4- Recommendations

1. Necessity for administering single dose of 5µg/kg/day subcutaneously of G-CSF within 24-72 hours following the last chemotherapy session.

2. Continuous identification of the pathogens that are responsible for FN should be done in each cancer center in Palestine individually.

3. Periodic evaluation and assessment of the susceptibility pattern for each pathogen.

4. Frequent updating to the empiric antibiotic therapy for the management of FN infections according to the findings.

5. The importance of educating the medical staff, patients and parents for self-hygiene to control infection transmission especially that the three most prevalent Gram-positive bacteria reported from blood samples in this study were skin normal flora.

5.5- Conclusion

ALL pediatric patients are at high risk for FN infections. This study showed the high prevalence 94% of FN among pediatric leukemic patients in Palestine, with a mean average number of FN episode per patient was (2.5 ± 1.5). The incidence of FN episodes due to FUO was 44%, while the incidence of bacterial FN infections was 45%. Gram-positive bacteria were found to be the higher in occurrence among bacterial FN attacks 62.5%,

while Gram-negative bacteria accounted for 37.5%. Among Gram-positive bacteria *CONS* was the predominant pathogen 58.7% that is isolated mostly from blood culture samples. *E.coli ESBL* bacteria were the most abundant Gram-negative microbe (42.9%) that is primarily isolated from blood samples as well.

CONS isolates were absolutely sensitive to (TEP), (VAN), (CTR) and (CTX). However, it is barely sensitive to (AMP) and (PEN). *E.coli ESBL* showed 83.3% sensitivity to (AK), (MER), (IMP) and (ERT). While it showed much lower sensitivity towards (PIT), (GEN), (CIP), (CFM), (CFZ), (CXM), (CTR), and (CTX). According to the current microbiology status, the empiric antibiotic therapy for the treatment of FN should re-evaluated and updated.

References

- Afzal, S., M. C. Ethier, L. L. Dupuis, L. Tang, A. S. Punnett, S. E. Richardson, U. Allen, O. Abla, and L. Sung (2009), ***Risk factors for infection-related outcomes during induction therapy for childhood acute lymphoblastic leukemia***, The Pediatric infectious disease journal, 28(12), 1064-1068.

- Al-Sweedan, S. A., W. Hyajneh, and A. Al-Ostath (2012), ***Patterns of bacteremia in cancer patient with febrile neutropenia at King Abdullah University Hospital-Jordan 2003-2008***, Journal of Pediatric Infectious Diseases, 7(1), 15-20.

- Al Omar, S., L. Nazer, and K. Alkayed (2013), ***A prospective study of febrile neutropenia in pediatric cancer patients in Jordan***, Journal of pediatric hematology/oncology, 35(8), 614-617.

- Almaziny, M. A.-I. (2014), ***Isolation, identification and profile of antibiotic resistance of bacteria in childhood febrile neutropenia*** European Journal of Experimental Biology 4(2), 1-6.

- Alvarnas, J. C., et al. (2015), ***Acute Lymphoblastic Leukemia***, Version 2.2015, J Natl Compr Canc Netw, 13(10), 1240-1279.

- Badr, M., T. Hassan, H. Sakr, N. Karam, D. A. Rahman, D. Shahbah, M. Zakaria, and S. Fehr (2016), ***Chemotherapy-induced neutropenia among pediatric cancer patients in Egypt: Risks and consequence***s, Molecular and clinical oncology, 5(3), 300-306.

- Bakhshi, S., K. S. Padmanjali, and L. S. Arya (2008), **Infections in childhood acute lymphoblastic leukemia: an analysis of 222 febrile neutropenic episodes**, Pediatric hematology and oncology, 25(5), 385-392.

- Borregaard, N. (2010), **Neutrophils, from marrow to microbes**, Immunity, 33(5), 657-670.

- Butt, T., R. K. Afzal, R. N. Ahmad, M. Salman, A. Mahmood, and M. Anwar (2004), ***Bloodstream infections in febrile neutropenic patients: bacterial spectrum and antimicrobial susceptibility pattern***, Journal of Ayub Medical College, Abbottabad : JAMC, 16(1), 18-22.

- Caniza, M. A., C. Odio, S. Mukkada, M. Gonzalez, F. Ceppi, S. Chaisavaneeyakorn, N. Apiwattanakul, S. C. Howard, V. Conter, and M. Bonilla (2015), **Infectious complications in children with acute lymphoblastic leukemia treated in low-middle-income countries**, Expert Rev Hematol, 8(5), 627-645.

- Carroll, W. L., and T. Bhatla (2016), **Chapter 18 - Acute Lymphoblastic Leukemia, in Lanzkowsky's Manual of Pediatric Hematology and Oncology (Sixth Edition)**, edited by P. Lanzkowsky, J. M. Lipton and J. D. Fish, pp. 367-389, Academic Press, San Diego.

- Castagnola, E., et al. (2007), **A prospective study on the epidemiology of febrile episodes during chemotherapy-induced neutropenia in children with cancer or after hemopoietic stem cell transplantation**, Clinical infectious diseasesq: an official publication of the Infectious Diseases Society of America, 45(10), 1296-1304.

- Chessells, J. M., G. Harrison, S. M. Richards, C. C. Bailey, F. G. Hill, B. E. Gibson, and I. M. Hann (2001), ***Down's syndrome and acute lymphoblastic leukaemia: clinical features and response to treatment***, Arch Dis Child, 85(4), 321-325.

- Conter, V., et al. (2014), **Childhood high-risk acute lymphoblastic leukemia in first remission**: Results after chemotherapy or transplant from the AIEOP ALL 2000 study.

- Cortes, J. A., S. Cuervo, C. A. Gomez, D. Bermudez, T. Martinez, and P. Arroyo (2013), **Febrile neutropenia in the tropics: a description of clinical and microbiological findings and their impact on inappropriate therapy currently used at an oncological reference center in Colombia, Biomedica**: revista del Instituto Nacional de Salud, 33(1), 70-77.

- Crawford, J., D. C. Dale, and G. H. Lyman (2004), **Chemotherapy-induced neutropenia: risks, consequences, and new directions for its management, Cancer**, 100(2), 228-237.

- Dale, D. C. (2005), **Advances in the use of colony-stimulating factors for chemotherapy-induced neutropenia**, J Support Oncol, 3(2 Suppl 1), 39-41.
- Dale, D. C. (2009), **Advances in the treatment of neutropenia, Curr Opin Support Palliat Care**, 3(3), 207-212.
- Depasse, J., M. A. Caniza, A. Quessar, M. Khattab, L. Hessissen, R. Ribeiro, S. Cherkaoui, S. Benchekroun, and K. K. Matthay (2013), **Infections in hospitalized children and young adults with acute leukemia in Morocco**, Pediatr Blood Cancer, 60(6), 916-922.
- Di Blasi, R., et al. (2018), **Febrile events in acute lymphoblastic leukemia: a prospective observational multicentric SEIFEM study (SEIFEM-2012/B ALL)**, Annals of hematology, 97(5), 791-798.
- El-Mahallawy, H., I. Sidhom, N. H. El-Din, M. Zamzam, and M. M. El-Lamie (2005), ***Clinical and microbiologic determinants of serious bloodstream infections in Egyptian pediatric cancer patients: a one-year study,*** International journal of infectious diseases: IJID : official publication of the International Society for Infectious Diseases, 9(1), 43-51.
- el-Mahallawy, H. A., I. Attia, N. H. Ali-el-Din, A. E. Salem, and S. Abo-el-Naga (2002), ***A prospective study on fungal infection in children with cancer***, Journal of medical microbiology, 51(7), 601-605.

- Eldomany, R., and N. Abdelaziz (2011), **Characterization and antimicrobial susceptibility of gram negative bacteria isolated from cancer patients on chemotherapy in Egypt**.

- Freifeld, A. G., et al. (2011), **Clinical practice guideline for the use of antimicrobial agents in neutropenic patients with cancer: 2010 update by the infectious diseases society of america, Clinical infectious diseases**: an official publication of the Infectious Diseases Society of America, 52(4), e56-93.

- Ghosn, M., E. Dagher, F. Nasr, G. Chahine, N. Taleb, R. Nasnas, and E. Ghayad (1994), ***Infections in cancer patients with granulocytopenia. Retrospective studies of 59 febrile episodes with hospitalization,*** Le Journal medical libanais. The Lebanese medical journal, 42(3), 117-122.

- Greaves, M. F. (1997), **Aetiology of acute leukaemia**, Lancet, 349(9048), 344-349.

- Gupta, S., M. Bonilla, M. Gamero, S. L. Fuentes, M. Caniza, and L. Sung (2011), ***Microbiology and mortality of pediatric febrile neutropenia in El Salvador***, Journal of pediatric hematology/oncology, 33(4), 276-280.

- Hakim, H., P. M. Flynn, K. M. Knapp, D. K. Srivastava, and A. H. Gaur (2009), ***Etiology and clinical course of febrile neutropenia in children with cancer***, Journal of pediatric hematology/oncology, 31(9), 623-629.

- Hamzeh, F., S. Kanj, and M. Uwaydah (2000), ***Febrile neutropenia in cancer patients in a tertiary care medical center in Lebanon: microbial spectrum and outcome***, Le Journal medical libanais. The Lebanese medical journal, 48(3), 136-142.
- Hassan, B. A. R. (2009), **An explorative study on factors associated with neutropenia onset and severity among cancer patients**.
- Hirmas, N., S. Mubarak, and I. Sultan (2017), ***Patterns of microbial growth in urine cultures in a pediatric hematology/oncology unit over a one-year period: a single institution study***, International Journal of Pediatrics and Adolescent Medicine, 4(3), 95-99.
- Hunger, S. P., and C. G. Mullighan (2015), **Acute Lymphoblastic Leukemia in Children**, N Engl J Med, 373(16), 1541-1552.
- Inaba, H., et al. (2017), ***Infection-related complications during treatment for childhood acute lymphoblastic leukemia, Annals of oncology***: official journal of the European Society for Medical Oncology, 28(2), 386-392.
- Jeddi, R., et al. (2010), **Factors associated with severe sepsis: prospective study of 94 neutropenic febrile episodes,** Hematology, 15(1), 28-32.
- Kanafani, Z. A., G. K. Dakdouki, K. I. El-Chammas, S. Eid, G. F. Araj, and S. S. Kanj (2007), ***Bloodstream infections in febrile neutropenic patients at a tertiary care center in Lebanon: a view of the past decade,*** International Journal of Infectious Diseases, 11(5), 450-453.

- Kanj, S., N. Adel, and G. Araj (2001), **The continued prevalence of Gramnegative infections in febrile neutropenic patients in Lebanon**. Comparison with studies from the Middle East, BMJ (Middle East), 8, 6-13.

- Kar, Y. D., Z. C. Ozdemir, and O. Bor (2017), **Evaluation of febrile neutropenic attacks of pediatric hematology-oncology patients**, Turk pediatri arsivi, 52(4), 213-220.

- Kato, M., and A. Manabe (2018), **Treatment and biology of pediatric acute lymphoblastic leukemia**, Pediatr Int, 60(1), 4-12.

- Katsimpardi, K., V. Papadakis, A. Pangalis, A. Parcharidou, J. P. Panagiotou, M. Soutis, E. Papandreou, S. Polychronopoulou, and S. Haidas (2006), ***Infections in a pediatric patient cohort with acute lymphoblastic leukemia during the entire course of treatment, Supportive care in cancer***: official journal of the Multinational Association of Supportive Care in Cancer, 14(3), 277-284.

- Klastersky, J. (2004), **Management of fever in neutropenic patients with different risks of complications, Clinical infectious diseases**: an official publication of the Infectious Diseases Society of America, 39 Suppl 1, S32-37.

- Klastersky, J., J. de Naurois, K. Rolston, B. Rapoport, G. Maschmeyer, M. Aapro, and J. Herrstedt (2016), **Management of febrile neutropaenia: ESMO Clinical Practice Guidelines, Annals of oncology**: official journal of the European Society for Medical Oncology, 27(suppl 5), v111-v118.

- Lakshmaiah, K. C., A. S. Malabagi, Govindbabu, R. Shetty, M. Sinha, and R. S. Jayashree (2015), ***Febrile Neutropenia in Hematological Malignancies: Clinical and Microbiological Profile and Outcome in High Risk Patients***, Journal of laboratory physicians, 7(2), 116-120.

- Latiff, Z., S. Zulkifli, and R. Jamal (2002), ***Risk assessment and microbiological profile of infections in paediatric cancer patients with febrile neutropenia***, MALAYSIAN JOURNAL OF PATHOLOGY, 24(2), 83-90.

- LCCH, E. D. C. S. (2018), **Guideline Management of Fever in a Paediatric Oncology PatientRep**. 1-16 pp.

- Lee, C. Y., P. Y. Chen, F. L. Huang, and C. F. Lin (2009), ***Microbiologic spectrum and susceptibility pattern of clinical isolates from the pediatric intensive care unit in a single medical center- 6 years' experience***, Journal of microbiology, immunology, and infection = Wei mian yu gan ran za zhi, 42(2), 160-165.

- Lee, D. G., S. H. Kim, S. Y. Kim, C. J. Kim, W. B. Park, Y. G. Song, and J. H. Choi (2011), ***Evidence-based guidelines for empirical therapy of neutropenic fever in Korea***, The Korean journal of internal medicine, 26(2), 220-252.

- Lee, J. H., et al. (2016), **Increase in Antibiotic-Resistant Gram-Negative Bacterial Infections in Febrile Neutropenic Children**, Infection & chemotherapy, 48(3), 181-189.

- Lehrnbecher, T., D. Varwig, J. Kaiser, D. Reinhardt, T. Klingebiel, and U. Creutzig (2004), **Infectious complications in pediatric acute myeloid leukemia**: analysis of the prospective multi-institutional clinical trial AML-BFM 93, Leukemia, 18(1), 72-77.

- Lex, C., D. Korholz, B. Kohlmuller, H. Bonig, R. Willers, C. M. Kramm, and U. Gobel (2001), ***Infectious complications in children with acute lymphoblastic leukemia and T-cell lymphoma--a rationale for tailored supportive care, Supportive care in cancer***: official journal of the Multinational Association of Supportive Care in Cancer, 9(7), 514-521.

- Li, M. J., et al. (2017), **Infectious complications in children with acute lymphoblastic leukemia treated with the Taiwan Pediatric Oncology Group protocol**: A 16-year tertiary single-institution experience, Pediatr Blood Cancer, 64(10).

- Lim, J. Y., S. Bhatia, L. L. Robison, and J. J. Yang (2014), **Genomics of racial and ethnic disparities in childhood acute lymphoblastic leukemia, Cancer**, 120(7), 955-962.
- Limvorapitak, W., and T. Khawcharoenporn (2015), **Incidence, Risk Factors, and Outcomes of Febrile Neutropenia in Thai Hematologic Malignancy Patients Receiving Chemotherapy: A 6-year Retrospective Cohort Study**, Asian Pacific journal of cancer prevention: APJCP, 16(14), 5945-5950.
- Livorsi, D. J., E. Stenehjem, and D. S. Stephens (2011), **Virulence factors of gram-negative bacteria in sepsis with a focus on Neisseria meningitidis**, Contributions to microbiology, 17, 31-47.
- Louw, V., J. van der Westhuizen, W. Rautenbach, E. van der Berg, M. Wamelink, and G. Joubert (2010), ***The antibiotic susceptibility of bacteria isolated from blood cultures during episodes of neutropenic fever in patients with acute myeloid leukaemia***, Southern African Journal of Epidemiology and Infection, 25(2), 9-11.
- Ludwig, H., P. Gascon, C. Bokemeyer, M. Aapro, M. Boccadoro, K. Denhaerynck, A. Krendyukov, K. MacDonald, and I. Abraham (2019), **Outcomes of chemotherapy-induced (febrile) neutropenia prophylaxis with biosimilar filgrastim** (Zarzio(R)) initiated "same-day" (< 24 h), "per-guidelines" (24-72 h), and "late" (> 72 h): findings from the MONITOR-GCSF study, Supportive care in cancer : official journal of the Multinational Association of Supportive Care in Cancer, 27(6), 2301-2312.

- Malabagi, A., R. Shetty, M. Sinha, R. Jayashree, Govindbabu, and L. Kuntegowdanahalli (2015), **Febrile Neutropenia in Hematological Malignancies**: Clinical and Microbiological Profile and Outcome in High Risk Patients, 116 pp.
- Mamishi, S., B. Pourakbari, M. H. Ashtiani, and F. B. Hashemi (2005), ***Frequency of isolation and antimicrobial susceptibility of bacteria isolated from bloodstream infections at Children's Medical Center, Tehran, Iran, 1996-2000***, International journal of antimicrobial agents, 26(5), 373-379.
- Mangaraj, J., D. Barkataki, and D. Gogoi Mohan (2015), **Gram Negative Bacilli Causing Blood Stream Infection in Febrile Neutropenic Patients in a Tertiary Care Centre**, 2393-2915 pp.
- Maqsood, S., M. Taj, T. Shah, T. Farzana, and T. Shamsi (2015), C**LINICAL AND MICROBIOLOGICAL PROFILE OF PATHOGENS IN FEBRILE NEUTROPENIA IN HEMATOLOGICAL MALIGNANCIES**: A SINGLE CENTER PROSPECTIVE ANALYSIS.
- Meidani, M., M. Baniasadi, and F. Khorvash (2018), **Prevalence of Fungemia in Pediatric Patients with Febrile Neutropenia**, Advanced biomedical research, 7, 88.
- Mian, M. W. (2013), **Febrile neutropenia in cancer patients; microbiology and clinical outcome of empirical antimicrobial therapy**, Master thesis thesis, 71 pp, Prince of Songkla University Malaysia.

- Ministry of Health, P. H. I. C. P. (2018), Health Annual Report Palestine 2017Rep., 380 pp, Ministry of Health, Palestine.
- Naeem, D., M. Alshamrani, M. Aseeri, and M. Khan (2018), **Prescribing Empiric Antibiotics for Febrile Neutropenia: Compliance with Institutional Febrile Neutropenia Guidelines**, Pharmacy, 6(3), 83.
- Nathan, C. (2006), **Neutrophils and immunity: challenges and opportunities**, Nat Rev Immunol, 6(3), 173-182.
- NHS (2017), **Guideline for the Management of Suspected Neutropenic Sepsis Induced By Cytotoxic Therapy Rep**., 1-22 pp.
- Nirenberg, A., A. Parry Bush, A. Davis, C. R. Friese, T. Wicklin Gillespie, and R. D. Rice (2006), **Neutropenia: state of the knowledge part II, Oncology nursing forum**, 33(6), 1202-1208.
- O'Connor, D., J. Bate, R. Wade, R. Clack, S. Dhir, R. Hough, A. Vora, N. Goulden, and S. Samarasinghe (2014), **Infection-related mortality in children with acute lymphoblastic leukemia**: an analysis of infectious deaths on UKALL2003, Blood, 124(7), 1056-1061.
- Ozdemir, N., G. Tuysuz, N. Celik, L. Yantri, E. Erginoz, H. Apak, A. Ozkan, I. Yildiz, and T. Celkan (2016), **Febrile neutropenia in children with acute lymphoblastic leukemia: single center experience**, Turk pediatri arsivi, 51(2), 79-86.

- Ozdemir, Z. C., A. Koc, and A. Aycicek (2016), **Microorganisms isolated from cultures and infection focus and antibiotic treatments in febrile neutropenic children from Sanliurfa, Turkey**, Turk J Pediatr, 58(1), 47-53.

- Park, Y., et al. (2010), **The suggestion of a risk stratification system for febrile neutropenia in patients with hematologic disease**, Leukemia research, 34(3), 294-300.

- Pillay, J., I. den Braber, N. Vrisekoop, L. M. Kwast, R. J. de Boer, J. A. Borghans, K. Tesselaar, and L. Koenderman (2010), **In vivo labeling with 2H2O reveals a human neutrophil lifespan of 5.4 days**, Blood, 116(4), 625-627.

- Pui, C. H. (1997), **Acute lymphoblastic leukemia**, Pediatr Clin North Am, 44(4), 831-846.

- Rahiala, J., M. Perkkio, and P. Riikonen (1998), **Infections occurring during the courses of anticancer chemotherapy in children with ALL: a retrospective analysis of 59 patients**, Pediatric hematology and oncology, 15(2), 165-174.

- Rajendranath, R., V. K. Balasubramaniyum, V. Vijayakumar, P. Ganesan, and G. S. Tenali (2014), ***Factors predicting outcome in high risk febrile neutropenia in patients receiving intensive chemotherapy for acute sleukemia: A prospective, observational study from South India***, Indian journal of cancer, 51(4), 481-486.

- Rezaee, M. A., B. Abdinia, A. Delpak, and A. Rezamand (2017), ***The Microbiologic Pattern in Pediatric Cancer Patients with Febrile Neutropenia and Bacteremia: A Referral Hospital-Based Study in Northwest of Iran***, Iranian Journal of Pediatrics, 27(2).

- Roongpoovapatr, P., and C. Suankratay (2010), ***Causative pathogens of fever in neutropenic patients at King Chulalongkorn Memorial Hospital***, Journal of the Medical Association of Thailand= Chotmaihet thangphaet, 93(7), 776-783.

- Salar, A., et al. (2012), **The need for improved neutropenia risk assessment in DLBCL patients receiving R-CHOP-21: findings from clinical practice**, Leukemia research, 36(5), 548-553.

- Siddaiahgari, S., A. Manikyam, K. A. Kumar, A. Rauthan, and R. Ayyar (2014), ***Spectrum of systemic bacterial infections during febrile neutropenia in pediatric oncology patients in tertiary care pediatric center***, Indian journal of cancer, 51(4), 403-405.

- Siegel, D. A., S. J. Henley, J. Li, L. A. Pollack, E. A. Van Dyne, and A. White (2017), **Rates and trends of pediatric acute lymphoblastic leukemia—United States**, 2001–2014, MMWR. Morbidity and mortality weekly report, 66(36), 950.

- Sigurdardottir, K., et al. (2005), ***A multi-centre prospective study of febrile neutropenia in Norway: microbiological findings and antimicrobial susceptibility***, Scandinavian journal of infectious diseases, 37(6-7), 455-464.

- Stabell, N., et al. (2008), ***Febrile neutropenia in children with cancer: a retrospective Norwegian multicentre study of clinical and microbiological outcome***, Scandinavian journal of infectious diseases, 40(4), 301-307.

- Swati, M., N. Gita, S. Baveja, F. Jijina, and P. Mehta (2010), **Microbial Etiology of Febrile Neutropenia**, 49-55 pp.

- Taj, M., T. Farzana, T. Shah, S. Maqsood, S. S. Ahmed, and T. S. Shamsi (2015), ***Clinical and Microbiological Profile of Pathogens in Febrile Neutropenia in Hematological Malignancies: A Single Center Prospective Analysis***, Journal of oncology, 2015, 596504.

- Tak, T., K. Tesselaar, J. Pillay, J. A. Borghans, and L. Koenderman (2013), **What's your age again? Determination of human neutrophil half-lives revisited**, J Leukoc Biol, 94(4), 595-601.

- Terwilliger, T., and M. Abdul-Hay (2017), **Acute lymphoblastic leukemia: a comprehensive review and 2017 update**, Blood Cancer J, 7(6), e577.

- Vathana, N., S. Thitipolpun, J. Buaboonnam, K. Phuakpet, and K. Sanpakit (2017), ***PREVALENCE OF PATHOGENS IN PEDIATRIC CANCER PATIENTS WITH FEBRILE NEUTROPENIA***, SOUTHEAST ASIAN JOURNAL OF TROPICAL MEDICINE AND PUBLIC HEALTH, 48, 151-160.

- Vossen, M. G., C. Milacek, and F. Thalhammer (2018), **Empirical antimicrobial treatment in haemato-/oncological patients with neutropenic sepsis**, ESMO open, 3(3), e000348.

- Wicki, S., A. Keisker, C. Aebi, K. Leibundgut, A. Hirt, and R. A. Ammann (2008), **Risk prediction of fever in neutropenia in children with cancer**: a step towards individually tailored supportive therapy?, Pediatr Blood Cancer, 51(6), 778-783.

- Xiao, H., L. Xiong, X. Song, P. Jin, L. Chen, X. Chen, H. Yao, Y. Wang, and L. Wang (2017), **Angelica sinensis Polysaccharides Ameliorate Stress-Induced Premature Senescence of Hematopoietic Cell via Protecting Bone Marrow Stromal Cells from Oxidative Injuries Caused by 5-Fluorouracil**, Int J Mol Sci, 18(11).

- Yilmaz, S., H. Oren, F. Demircioglu, and G. Irken (2008), **Assessment of febrile neutropenia episodes in children with acute leukemia treated with BFM protocols**, Pediatric hematology and oncology, 25(3), 195-204.

- Zhai, W., et al. (2015), ***A Prospective Observational Study of Antibiotic Therapy in Febrile Neutropenia Patients with Hematological Malignances from Multiple centers in Northeast China***, International journal of infectious diseases : IJID : official publication of the International Society for Infectious Diseases, 37, 97-103.

Appendix (1)

Fever in Neutropenia Protocol /Huda Al Masri Pediatric Cancer Department

Guidelines for the management of fever and neutropenia in pediatric oncology patients

1-Initial antibiotic therapy

Empiric antibiotic therapy should be administered promptly AFTER OBTAINING CULTURES to all neutropenic patients at the onset of fever.

Cefepime or piperacillin/tazobactam (if abdominal/perirectal pain) may be started initially, and evaluate need for an aminoglycoside and vancomycin (see table below)

Start broad spectrum antibiotics	Ceftazidime: 100mg/kg/day OR Piperacillin/tazobactam or ceftazidime **and** metronidazole (for anaerobic and gram negative coverage in patients with abdominal/perirectal pain) Piperacillin/tazobactam: Infants 1-6 months: 150-300 mg piperacillin component/kg/day IV in divided doses every 6-8 hours; Children > 6 months: 100 mg piperacillin/kg/dose IV q6-8h (max 4 g/dose) Severe penicillin type 1-immediate hypersensitivity: Aztreonam 90-120 mg/kg/day divided every 6-8 hours (max 2 g/dose)* (DO NOT USE IF ALLERGIC TO CEFTAZIDIME) + Vancomycin 15mg/kg/dose IV every 6-8 hours per protocol* Consider aminoglycoside for additional Pseudomonal coverage and metronidazole for anaerobic coverage if patient is having abdominal/perirectal pain

	Penicillin allergy NOT IgE mediated (such as hives, angioedema) consider cefepime
Consider aminoglycoside if: 1. Hypotension or other evidence of cardiovascular impairment 2. Altered mental status 3. Respiratory failure 4. Possible Disseminated Intravascular Coagulation 5. Presence of uncontrolled cancer a. Absence of documented complete remission for patients with leukemia b. Development of new lesions, or enlargement of lesion on chemotherapy, or other failure for patients with solid tumors 6. Gram stain shows gram negative organisms	Gentamicin*: Traditional dosing: 2.5 mg/kg/dose IV every 8 hours Or amikacin 20mg/kg/ day Extended-interval dosing: 7-8 mg/kg/dose IV every 24 h. Patients must meet **ALL** of the following criteria to be eligible for extended-interval dosing: 1. ≥ 3 months of age 2. CrCl > 60 mL/min with stable renal function 3. No evidence of altered volume of distribution (edema, ascites, trauma, shock, obese) 4. NOT critically ill and does NOT have meningitis, enterococcal endocarditis, osteomyelitis, or persistent bacteremia
Consider vancomycin if: 1. Clinically suspect skin or line infection 2. Known colonization with resistant gram-positive organisms (MRSA, penicillin-and-cephalosporin resistant pneumococci) 3. History of prior alpha-hemolytic strep infection 4. Gram stain showing gram-positive bacteria 5. Hypotension or other evidence of cardiovascular impairment 6. Substantial oral or respiratory mucosal damage 7. Fluoroquinolone prophylaxis prior to febrile episode 8. Receipt of high dose cytarabine	Vancomycin* 15mg/kg/dose every 6-8 hours IV (refer to Vancomycin protocol) Or Teicoplanin (targosid) 10 mg/kg/ day

In patients who are responding to initial empiric antibiotic coverage, consider discontinuing aminoglycoside and vancomycin if:

1- Cultures are negative after 48 hours.

2- Patient is afebrile at least 24 hours.

3- There is evidence of bone marrow recovery**.**

2-Antifungals

Any positive fungal culture send sensitivity and consult Peds ID.

AML patients on COG protocol already receiving fluconazole prophylaxis→ With **initial fever spike** increase fluconazole from prophylaxis to treatment doses and start broad spectrum antibiotics. If after 3-5 days of continued fever despite increase in fluconazole and antibiotics, consider changing fluconazole to either voriconazole, Ambisome or caspofungin. Suggest Peds ID consult If a patient is already receiving caspofungin prophylaxis →Continue caspofungin at same doses If patient is unstable, consider adding voriconazole or posaconazole (for better mold coverage.) Suggest Peds ID consult. Call lab for fungal culture results, send serum galactomannan (for aspergillus) If patient is not receiving fungal prophylaxis and is stable consider antifungal therapy (amphotericin b [liposomal] or caspofungin) if: 1. Fever persists after 3-5 days of antibiotic therapy, reassessment does not yield a cause, and patient is expected to have neutropenia for longer than 5–7 more days 2. Pt has history of prior fungal infection 3. Severe mucositis or localized infection suspicious for fungus (i.e. halo sign on CXR)	Amphotericin B (Liposomal) (Ambisome®)*: 5 mg/kg/dose once daily* Or Fluconazole 5 mg/kg /day Any patient from whom Aspergillus species were yielded on culture: Voriconazole: children < 12 y/o, 7 mg/kg IV or PO q12h with no loading dose. For children ≥ 12 y/o, 6 mg/kg IV or PO q12h x 2 then 4 mg/kg IV/PO q12h maintenance.. Adjustment in hepatic disease necessary. Goal trough: 1-5.5 mcg/mL

3-Antiviral

Consider antiviral therapy if there is clinical or laboratory evidence of viral disease, including influenza Send shell vial culture for HSV, CMV Obtain Respiratory film array	Case specific: Varicella: Acyclovir* 500 mg/m^2/dose IV Q8 hrs throughout illness and switched to po Valacyclovir for prophylaxis after completion (COG protocol) Severe RSV and metapneumovirus: Ribavirin 2 grams over 2 hrs three times/day for a minimum of 3-7 days. Ribavirin should be diluted in 100 mL of preservative-free sterile water to a final concentration of 60 mg/mL HSV: acyclovir 250 mg/m2/dose IV q8h. Influenza: > 1 year of age Oseltamivir Wt (kg) dose ≤15 30 mg twice daily for 5 days 16-23 45 mg twice daily for 5 days 24-40 60 mg twice daily for 5 days >40 75 mg twice daily for 5 days CMV: Consider Peds ID consult. Ganciclovir IV (induction) should be initiated.

Please note: that not all the medication are available at the hospital, thus; please consider just the available agents.

Adapted from:

NCCN Practice Guidelines in Oncology – v.1.2011.

Guidelines for Preventing Infectious Complications among Hematopoietic Cell Transplantation Recipients: A Global Perspective 2009

IDSA Guidelines for Febrile Neutropenia CID2011

Guidelines Developed By: Jennifer Lighter Fisher, MD

Revised by: Liana Mark, Pharm.D, .Sonya Patel, Pharm.D.

Approved by Antimicrobial Stewardship Sub-Committee on: October 11, 2011

Appendix (2)

Data Collection form:

Prevalence and Microbiology Profile of Febrile Neutropenia among Acute Lymphoblastic Leukemic Pediatric Patients

Name: ---------- Gender:---------- Patient ID:----------

Patient (DOB):---------- Age:---------- Place of Residence:--------

File no.: ----------

Diagnosis: ---------- **Stage of disease**:----------

Central line: □ Yes □ No.

Inpatient: □ Outpatient: □.

Chemotherapy used: ---------- Cycle of Chemotherapy: ----------

Last date of chemotherapy: ----------

-

Temperature (fever): ---------- Cº. Date of fever:----------

After how many days does the fever occur from the last chemotherapy: -

---

Duration of fever (days): ----------

Duration of neutropenia (days) :----------

Intensity of neutropenia: □ Prolonged □ Short.

Medication(s) allergy: ----------

Chest X-ray: □ Yes □ No. Date: ----------

Symptoms/ Clinical manifestations: Date: ----------

□ Fever □ Mucositis □ Dyspnea □ Pallor □ Cough

□ Vomiting □ Oral ulcers □ Dysphagia □ Abdominal pain □ Arthralgia

□ Rhinorrhea □ Vaginal discharge □ Others ----------

Vital signs:

Heart rate (HR) ---------- Date:----------

SpO_2: ---------- Date:----------

Blood pressure (BP): systolic:---------- Diastolic: ----------

---------- Date:----------

Site of infection:

□ IV catheter □ GIT □ UT □ Skin & soft tissues □ Lungs

□ Others ---------- □ Unknown.

Microbiology:

Date	Blood culture	Urine Culture	Wound secretions	Others	Sensitive AB	Resistant AB

Laboratory Results:

Creatinin															
BUN															
RBCs															
WBCs															
Platelets															
Hgb															
Neutrophil Granulocytes %															
Neutrophil Granulocyte															
ANC															
Na															
K															
Cl															
Ca															
Lactate															
ALT															
AST															
Bilirubin															
Albumin															
CRP															

Medications:

Type of Medication	**Agent**	**Starting date**	**With holding date**
Prophylactic AB			
Initial AB			
Initial AB			
Initial AB			
Add on AB			
Add on AB			
Add on AB			
Add on Antimicrobial			
Add on Antifungal			
Add on Antiviral			
Others			
Others			
Blood			
Platelets			

Discharge Date: ---------- **Prophylactic agents:----------**

Appendix (3)

Detailed distribution of pathogens that are responsible for FN episodes

Bacterial infection sources (n= 73)		Frequency (%)
Blood/ Peripheral vein	24	33
Blood /Port cath	19	26
Urine	12	16
Mixed Blood (Peripheral vein + Port cath)	10	14
Wound/ Skin	5	7
Throat	3	4

Fungus infection sources (n= 9)		Frequency (%)
Stool	3	33
Mixed blood (Peripheral vein + Port cath)	3	33
Blood/ Peripheral	1	11
Blood/ Port cath	1	11
Oral cavity	1	11

Confirmed Viral infection sources (n= 6)		Frequency (%)
Nasal swab	4	50
Skin	3	37.5
Oral	1	12.5

Parasites source on infection (n=2)		Frequency (%)
GIT	2	100

An-Najah
National University
Faculty of Graduate Studies
Dean's Office

جامعة
النجـــاح الوطنية
كلية الدراسات العليا
مكتب العميد

التاريخ: 2018/5/13

حضرة الدكتورة سماح الجابي المحترمة
منسقة برنامج ماجستير الصيدلة السريرية

تحية طيبة وبعد،

الموضوع: الموافقة على عنوان الاطروحة وتحديد المشرف

قرر مجلس كلية الدراسات العليا في جلسته رقم (358)، المنعقدة بتاريخ 2018/5/10، الموافقة على مشروع الأطروحة المقدم من الطالب/ة رانيا اميل جورج غانم، رقم تسجيل 11558909، تخصص ماجستير الصيدلة السريرية، عنوان الأطروحة:

(مدى الانتشار و التحليل الجرثومي لحالات ارتفاع الحرارة المصاحب لنقص الخلايا الحبيبية المتعادلة (النيوتروفيل) لدى الاطفال المصابين باللوكيميا الليمفاوية الحادة)

(Prevalence and Microbiology Profile of Febrile Neutropenia among Acute Lymphoblastic Leukemic in Pediatric Patients)

بإشراف: أ.د. وليد صويلح

يرجى اعلام المشرف والطالب بضرورة تسجيل الاطروحة خلال اسبوعين من تاريخ اصدار الكتاب. وفي حال عدم تسجيل الطالب/ة للاطروحة في الفترة المحددة له/ا ستقوم كلية الدراسات العليا بإلغاء اعتماد العنوان والمشرف

وتفضلوا بقبول وافر الاحترام ،،،

د. محمد سليمان
عميد كلية الدراسات العليا

نسخة : د. رئيس قسم الدراسات العليا للعلوم الطبية والصحية المحترم
: ق.أ.ع. القبول والتسجيل المحترم
: مشرف الطالب
: ملف الطالب

ملاحظة: على الطالب/ة مراجعة الدائرة المالية (محاسبة الطلبة) قبل دفع رسوم تسجيل الاطروحة للضرورة

فلسطين، نابلس، ص ب 7،707 هاتف: 2345115/، 2345114، 2345113 (09)(972)* فاكسميل 2342907(09)(972)
Nablus, P. O. Box (7) *Tel. 972 9 2345113, 2345114, 2345115 هاتف داخلي (5) 3200
* Facsimile 972 92342907 *www.najah.edu - email fgs@najah.edu

An-Najah
National University
Faculty of medicine
&Health Sciences
Department of Graduate
Studies

جامعة النجاح
الوطنية
كلية الطب وعلوم الصحة
دائرة الدراسات العليا

IRB Approval Letter

Study Title:

"Prevalence and Microbiology Profile of Febrile Neutropenia among Acute Lymphoblastic Leukemic Pediatric Patients"

Submitted by:

Rania Ghanem, Prof. Waleed Sweileh

Date Reviewed:

16th April 2018.

Date Approved:

23rd April 2018.

Your Study titled ***"Prevalence and Microbiology Profile of Febrile Neutropenia among Acute Lymphoblastic Leukemic Pediatric Patients"*** with archived number (12) April 2018 was reviewed by An-Najah National University IRB committee and was approved on 23rd April 2018.

Hasan Fitian, MD
IRB Committee Chairman
An-Najah National University

(970) (09) 2342910 فاكسميل || (970) (09)2342902/4/7/8/14 هاتف || 707 أو 7 ص.ب نابلس

Nablus - P.O Box :7 or 707 | Tel (970) (09) 2342902/4/7/8/14 | Faximile (970) (09) 2342910 | E-mail : hgs@najah.edu

An-Najah
National University
Faculty of Graduate Studies

جامعة
النجـــاح الوطنية
كلية الدراسات العليا

التاريخ : 2018/5/28م

حضرة الدكتورة امل ابو عوض المحترمة
مدير عام التعليم الصحي
وزارة الصحة الفلسطينية

تحية طيبة وبعد ،

الموضوع: تسهيل مهمة الطالبة/ رانيا اميل جورج غانم، رقم تسجيل (11558909)، تخصص ماجستير الصيدلة السريرية

الطالبة/ رانيا اميل جورج غانم، رقم تسجيل 11558909، تخصص ماجستير الصيدلة السريرية، في كلية الدراسات العليا، بصدد إعداد الاطروحة الخاصة به بعنوان:

(Prevalence and Microbiology Profile of Febrile Neutropenia among Acute Lymphoblastic Leukemic in Pediatric Patients)

يرجى من حضرتكم تسهيل مهمتها من اجل جمع معلومات والسماح لها بالاطلاع على ملفات المرضى وعمل مقابلات مع المرضى والاستفسار عن بعض المعلومات حول مرض اللوكيميا الليمفاوية في مستشفى بيت جالا الحكومي / قسم هدى المصري للأورام الدم عند الاطفال، و التابعة لوزارة الصحة الفلسطينية في محافظة بيت لحم.

وذلك لاستكمال مشروع البحث.

علماً المعلومات سوف تستخدم لاغراض البحث العلمي و لاستكمال مشروع البحث.

شاكرين لكم حسن تعاونكم

واقبلوا فائق الاحترام

د. محمد سليمان شتيه
عميد كلية الدراسات العليا

جامعة النجاح الوطنية
كلية الدراسات العليا
نابلس / فلسطين

فلسطين، نابلس، ص.ب 707، هاتف:/2345115، 2345114، 2345113 (09)(972)* فاكسميل:2342907(09)(972)
Nablus, P. O. Box (7) *Tel. 972 9 2345113, 2345114, 2345115 هاتف داخلي (5) 3200
* Facsimile 972 92342907 *www.najah.edu - email fgs@najah.edu

State of Palestine
Ministry of Health - Nablus
General Directorate of Education in Health

دولة فلسطين
وزارة الصحة- نابلس
الإدارة العامة للتعليم الصحي

Ref.: .....
Date:.....

الرقم: عحد/٨٥٩/٢٠١٨
التاريخ: ٢٠١٨/٦/١٠

الأخ مدير عام الادارة العامة للمستشفيات المحترم،،،

تحية واحترام،،،

الموضوع: تسهيل مهمة طالبة ماجستير – جامعة النجاح

يرجى تسهيل مهمة **الطالبة: رانيا اميل جورج غانم– ماجستير صيدلة سريرية/ جامعة النجاح،** في عمل بحث بعنوان "**مدى الانتشار والتحليل الجزئومي لحالات ارتفاع الحرارة المصاحب لنقص الخلايا الحبيبية المتعادلة (النيوتروفيل) لدى الاطفال المصابين باللوكيميا الليمفاوية الحادة**"، من خلال السماح للطالبة بجمع معلومات من خلال مراجعة ملفات المرضى لجمع معلومات تتعلق بالبحث، وذلك في:

- **مستشفى بيت جالا الحكومي**

علما ان البحث تحت اشراف د. وليد صويلح. كما انه سيتم الالتزام بمعايير البحث العلمي والحفاظ على سرية المعلومات.

مع الاحترام،،،

دولة فلسطين وزارة الصحة الادارة العامة للتعليم الصحي

د. أمل ابو عوض
مدير عام التعليم الصحي

نسخة: عميد كلية الدراسات العليا المحترم/ جامعة النجاح

P.O .Box: 14
Tel.:09-2333901

ص.ب. 14
تلفون: 09-2333901

جامعة النجاح الوطنية
كلية الدراسات العليا

مدى انتشار والتحليل الجرثومي لحالات ارتفاع الحرارة المصاحب لنقص الخلايا الحبيبية المتعادلة (النيوتروفيل) لدى الأطفال المصابين باللوكيميا الليمفاوية الحادة

إعداد
رانيا غانم

إشراف
أ. د. وليد صويلح

قُدمت هذه الأطروحة إستكمالاً لمتطلبات الحصول على درجة الماجستير في الصيدلة السريرية بكلية الدراسات العليا في جامعة النجاح الوطنية في نابلس، فلسطين
2019

مدى انتشار والتحليل الجرثومي لحالات ارتفاع الحرارة المصاحب لنقص الخلايا الحبيبية المتعادلة (النيوتروفيل) لدى الأطفال المصابين باللوكيميا الليمفاوية الحادة

إعداد

رانيا غانم

إشراف

أ.د وليد صويلح

الملخص

الخلفية: قلة الخلايا الحبيبية المتعادلة هي أحد المضاعفات التي تهدد حياة المرضى الذين يعانون من سرطان الدم من نوع اللوكيميا الليمفاوية الحادة. هذه الحالة تنتج عن الفسيولوجيا المرضية للمرض وكآثار جانبية للعلاج الكيماوي للسرطان. الحرارة المصاحبة لنقص الخلايا الحبيبية المتعادلة (FN) هي مؤشر للعدوى بين هؤلاء المرضى الذين يعانون من نقص في المناعة، لذا يكونون أكثر عرضة للخطر بارتفاع معدلات الممراضة والوفيات. إن التدخل الفوري في علاج الإلتهابات المصحوبة بالحرارة و نقص الخلايا الحبيبية المتعادلةFN (infections) عن طريق إعطاء المضادات الحيوية الاولية المناسبة ، هو الأساس في إنقاذ حياة هؤلاء المرضى.

الأهداف: تهدف هذه الدراسة إلى تحديد مدى انتشار الحرارة المصاحبة لنقص الخلايا الحبيبية المتعادلة (FN) بين الأطفال الذين يعانون من سرطان الدم اللوكيميا الليمفاوية الحادة. إضافة إلى التحليل الجرثومي لكل حالات الحرارة المصاحبة لنقص الخلايا الحبيبية المتعادلة (FN) من أجل تحديد نوع الميكروبات المسؤولة عنها . و أيضا تقييم أنماط حساسية هذه الميكروبات للمضادات الحيوية من تنائج زراعة الميكروبات.

المنهجية الدراسة: هذه الدراسة هي دراسة وصفية بأثر رجعي ، أجريت في قسم هدى المصري لأورام الدم عند الأطفال في مستشفى بيت جالا الحكومي في بيت لحم و ذلك خلال الفترة ما بين 2014/1/1 و حتى 2018/6/1. حيث تم استعراض السجلات الطبية من 83 مريضا. و قد تمت مراجعة و تحليل هذه الملفات من حيث الأعراض ، والعلامات الحيوية ، وفحوصات الدم المختلفة،

التأكد من وجود (FN)، إيجابية زراعة الميكروبات، ملف الأحياء الدقيقة وأنماط حساسية الميكروبات للمضادات الحيوية.

النتائج: ثلاثة وستون مريضا تم ادراجهم ضمن هذه الدراسة و قد وجد هنالك ما مجموعه 161 حالة من FN لدى هؤلاء المرضى . وقد كانت الغالبية حالات FN هي غير البكتيرية المشنأ بنسبة 54 ٪. بينما 45 ٪ من هذه الحالات كانت نتيجة لإلتهابات بكتيرية. شكلت البكتيريا إيجابية الجرام 62.5 ٪ من الحالات البكتيرية المنشأ، في حين كانت نسبة البكتيريا سلبية الجرام حوالي 37.5 ٪ من الحالات البكتيرية.

من بين البكتيريا إيجابية الجرام ، كانت *CONS* هي اكثر البكتيريا الممرضة بنسبة 58.7 ٪ ، تلتها *S.aureus* 15.2٪. كلاهما تم عزلها في الغالب من عينات زراعة الدم. لقد وُجد أن *CONS* بكتيريا كانت حساسة بنسبة 100٪ للمضادات الحيوية التالية: تيكوبلانين والفانكومايسين والسيفترياكسون والسيفوتاكسيم. بينما كانت أقل حساسية للأمبيسلين والبنسلين بنسبة 18.5 ٪ و 7.4 ٪ على التوالي.

فيما يخص بالبكتيريا سالبة الجرام ، كانت بكتيريا *E.coli ESBL* أكثر انواع البكتيريا المسببة للأمراض البكتيرية السالبة بنسبة 42.9٪ وقد تم عزلها في الغالب من عينات زراعة الدم ، تلتها بكتيريا *E.coli* بنسبة 17.9٪ وقد عزولت بشكل كبير من عينات البول. أظهرت بكتيريا *E.coli ESBL* حساسية بنسبة 83.3 ٪ اتجاه المضادات الحيوية التالية: الأميكاسين ، الميرونيم ، إيميبينيم والإرتابينيم، بينما كانت حساسة بنسبة 25 ٪ للسيفوتاكسيم.

الأستنتاجات: لقد أظهرت هذه الدراسة ارتفاع في معدل انتشار FN 94 ٪ لدى الأطفال اللذين يعانون من سرطان الدم من نوع اللوكيميا الليمفاوية الحادة. كانت البكتيريا الموجبة للجرام هي الأكثر شيوعا، و قد كانت *CONS* هي اكثرها سببا للإلتهابات البكتيرية الموجبة الجرام، و قد أظهرت حساسية عالية للمضادات الحيوية التيكوبلانين والفانكومايسين والسيفترياكسون والسيفوتاكسيم. بينما

كانت *E.coli ESBL* هي أكثر الميكروبات السالبة للجرام شيوعا إضافة إلى كونها أقل حساسية اتجاه المضادات الحيوية.

توصي هذه الدراسة بضرورة مراجعة و تقييم وتحديث المضادات الحيوية الأولية المستخدمة في علاج حالات (FN) بما يناسب السلوك البكتيري اتجاه المضادات الحيوية.