## بسم الله الرحمن الرحيم

قال تعالى:

﴿ وَهَوْقَ كُلَّ خِي عِلْمِ عَلَيْهِ ﴾

حدق الله العظيم الله العظيم الله (76 ) من سورة يوسف

## Dedication

I dedicate this work to my

Parents ...

Teachers ...

sisters and brothers ...

Colleagues ...

Dear and Best Friends ...

And to everyone who helped me to learn new thing.

#### Acknowledgments

Firstly I want to give my greatest thanks to Allah for his gifts which never ends.

I would like to express my gratefulness to my supervisor Dr. Elfadil Mustafa Abass for his advices, comments and patience.

I extend my sincere gratitude to Dr. Khalid Enan for his help and guidance to complete this study and for scientific contributions.

My thanks also extend to Mr. Musab Abd Alrazek and Miss. Suhair Rammadan and to all staff at Research Laboratory and Microbiology Department for their help and advises during the practical work.

Many thanks also go to director of Jabel Aulia Hospital and to the director of the laboratory for allowing collection of specimens.

A special feeling of gratitude to my loving parents, whom have been a great source of motivation and inspiration and to all patients for accepting participation in the study; I wish them quick recovery.

To them, and all I have not mentioned by name, I say a big Thank You.

#### **Abstract**

*P. falciparum* malaria is known to interfere with biology and the specific immunity against EBV and can induce virus reactivation.

This study was carried out to detect Epstein–Barr virus DNA in plasma specimens of children with and without *Plasmodium falciparum* malaria who attended Al-jabel Military Medical Hospital and Jabel Aulia Hospital during the period September to December 2015.

A total of 80 blood specimens were collected and DNA was extracted from plasma of the patients. Epstein-Barr virus DNA was detected using conventional Polymerase Chain Reaction.

The specimens included 50 plasma of children with P. falciparum malaria infection. Their mean age  $7\pm 4$  years among which 31(62%) were males and 19 (38%) were females. Samples included also 30 plasma specimens of children with age matched the malaria patients; mean age  $7.6\pm 4$  years among which 16 were males and 14 were females. Mean of hemoglobin (Hb) of children with P. falciparum malaria infection was  $9.8\pm 2.4$  g/dl and for the children without P. falciparum malaria infection was  $10.4\pm 2$  g/dl.

EBV DNA was detected in 2 specimens of the control group (without P. falciparum malaria, 6.7%), while EBV DNA was not detected among the children with P. falciparum malaria infection (0.0%). The two EBV DNA positive individuals were in the younger age group (1-5 years of age). Analysis showed no relation between EBV infection and patient's age and gender. In addition, there was also no relation between EBV infection, parasitemia, and Hb levels (p. values > 0.2).

These results may indicate absence or low EBV infection among P. falciparum malaria infected children.

#### ملخص الأطروحة

أجريت هذه الدراسة للكشف عن الحمض النووي لفيروس ابشتاين- بار في عينات بلازما لدى الطفال مصابين و غيرمصابين بالملاريا بمستشفى السلاح الطبي بجبل أولياء ومستشفى جبل اولياء خلال الفترة من سبتمبر إلى ديسمبر 2015.

جمعت 80 عينة دم من المرضى وتم استخلاص الحمض النووي من البلاز ما للكشف عن فيروس ابشتاين- بار بواسطة اختبار تفاعل البلمرة التسلسلي.

تضمنت العينات50 عينة بلازما من اطفال مصابين ببلازموديوم فالسيبارم ملاريا. متوسط اعمارهم  $7 \pm 4$  سنوات, 11 (62) ذكور و 11 (63) اناث و 11 عينة بلازما من اطفال مماثلين لهم في العمر غير مصابين ببلازموديوم فالسيبارم ملاريا , متوسط اعمارهم 11 سنوات, 11 (11) ذكور و 11 (11) اناث. متوسط خضاب الدم للاطفال المصابين ببلازموديوم فالسيبارم ملاريا كان 11 عين 11 عين المصابين ببلازموديوم فالسيبارم ملاريا كان متوسط خضاب الدم 11 عين الدم الدم 11 عين المصابين ببلازموديوم فالسيبارم ملاريا كان متوسط خضاب الدم 11 عين الدم 11 عين الدم الدم 11 عين الدم الدريا كان متوسط خضاب الدم 11 عين الدم 11 عين الدم الدريا كان متوسط خضاب الدم 11 عين الدم 11 عين الدم 11 عين الدم وديوم فالسيبارم ملاريا كان متوسط خضاب الدم 11

تم الكشف على الحمض النووي لغيروس ابشتاين- بار في عينتين من الاطفال غير المصابين ببلازموديوم فالسيبارم ملاريا (6.7). بينما لم يتم الكشف عن الحمض النووي لغيروس ابشتاين بار لدى الاطفال المصابين ببلازموديوم فالسيبارم ملاريا (0.0). عينتا المرضى الايجابيتان وجدث في الفئة العمرية الأصغر سنا (1-5 سنوات من العمر).

اظهر التحليل انه لا توجد علاقة بين الاصابة بفيروس ابشتاين- بار, عمر المريض و النوع. إضافة إلى ذلك, لم يكن هناك أيضاً اختلاف ذات دلالة إحصائية بين الاصابة بفيروس ابشتاين- بار, كثافة الطفيل ومعدل خضاب الدم.

هذه النتائج قد تشير إلى غياب أو انخفاض الإصابة بفيروس ابشتاين- بار وسط الاطفال المصابين ببلاز موديوم فالسيبارم ملاريا.

## **Table of contents**

No.	Title	Page
	الآية	No.
	Dedication	
		II
	Acknowledgments	III
	English Abstract	IV
	Arabic Abstract	V
	Tables of contents	VI
	List of tables	IX
	List of figures	X
	List of Abbreviations	XI
	CHAPTER ONE	
	INTRODUCTION	•
1.1	Background	1
1.2	Rationale	2
1.3	Objectives	3
1.3.1	General objective	3
1.3.2	Specific objectives	
	CHAPTER TWO	•
	LITERATURE REVIEW	
2.1	Epstein-Barr Virus (EBV)	4
2.1.1	History of EBV	4
2.1.2	Morphology and genomic organization	4
2.1.2.1	Virion Structure	4
2.1.2.2	Taxonomy and Genome Structure	7
2.1.3	Viral antigen	7
2.1.3.1	Latent phase antigens	7
2.1.3.2	Early antigens	8
2.1.3.3	Late antigens	8

2.1.4	Other properties of EBV	8
2.1.5	Epidemiology of EBV	
2.1.6	Transmission	
2.1.7	Replication of EBV	
2.1.8	Life cycle 1	
2.1.8.1	Establishment and maintenance of EBV latency	12
2.1.8.2	2 Reactivation of EBV from latency 1	
2.1.9	Pathogenesis	14
2.1.10	EBV and host immune response	15
2.1.11	1 Clinical syndrome 1	
2.1.12	2 Laboratory diagnosis	
2.1.13	3 Treatment 1	
2.1.14	14 Prevention and immunization 1	
2.2	Malaria 1	
2.3	Role of malaria in EBV reactivation	20
2.4	Background study	
	CHAPTER THREE	
	MATERIALS AND METHODS	
3.1	Study design	23
3.2	Study area and Study duration	23
3.3	Study population	23
3.3.1	Inclusion criteria	23
3.3.2	Exclusion criteria	23
3.4	Sample size	23
3.5	Sampling technique	23
3.6	Ethical clearance	24
3.7	Data collection	24
3. 7.1	Method of data collection	24
3. 7.2	Study variables	24

3.8	Experiment setting	24
3.1.9	Specimen collection	24
3.10	Laboratory test	24
3.10.1	Laboratory test for confirmation of malaria	24
3.10.2	EBV DNA extraction	26
3.10.3	Polymerase chain reaction for EBV	26
3.10.4	Gel electrophoresis	27
3.10.5	Interpretation of PCR results	27
3.11	Data analysis	27
	CHAPTER FOUR	
	RESULTS	
	Results	28
	CHAPTER FIVE	
	DISCUSSION	
	Discussion	35
	Conclusion 38	
	Recommendations 38	
	REFERENCES	
References 39		39
	APPENDICES	
	Appendices	44

### List of tables

No.	Title of table	Page No.
	Frequency and percentage of EBV DNA among	
Table 4.1	P. falciparum malaria infected and non- infected	31
	children.	
	Frequency of EBV-DNA among P. falciparum	
Table 4.2	malaria infected and non- infected children according	32
	to the age group of Study population	
T. 1.1. 4.2	Frequency of EBV-DNA among the P. falciparum	
Table 4.3	malaria infected and non- infected children according	33
	to gander	
Table 4.4	Frequency and percentage of EBV-DNA among non-	24
	P. falciparum malaria infected children according to	34
	HB levels	

# List of figures

No.	Title of figure	Page No.
Figure 2.1	Epstein-Barr Virus morphology.	6
Figure 2.2	Replication of Epstein-Barr Virus.	11
Figure 2.3	Epstein-Barr Virus latent life cycle.	13
Figure 4.1	Gel electrophoresis for amplification of EBNA1 gene.	30

## **List of Abbreviations**

Abbreviations	Meaning
AIDS	Acquired Immunodeficiency Syndrome
BL	Burkitt's Lymphoma
CMI	Cell mediated immunity
CIDR1a	Cysteine rich interdomain region 1α
CTL	Cytotoxic T lymphocyte
DNA	Deoxyribonucleic acid
eBL	endemic Burkitt's Lymphoma
ELISA	Enzyme Linked Immunosorbent Assay
EBNA	Epstein-Barr nuclear antigen
EBV	Epstein-Barr Virus
EBER	Epstein-Barr Virus encoded mRNA
GC	Gastric Carcinoma
Hb	Hemoglobin
HD	Hodgkin Disease
IM	Infectious Mononucleosis
IARC	International Agency for Research on Cancer
LMP	Latent membrane protein
NPC	Nasopharyngeal Carcinoma
NHL	Non-Hodgkin lymphomas
OHL	Oral Hairy Leukoplakia
PfEMP1	Plasmodium falciparum erythrocyte membrane protein1

P.falciparum	Plasodium falciparum
P. vivax	Plasodium vivax
PCR	Polymerase Chain Reaction
PTLD	post transplantation lymphoproliferative disease
RA	Rheumatoid arthritis
SPSS	Statistical Package of Social Science program
SLE	Systemic Lupus Erythromatosus
TBE buffer	Tris Boric acid EDTA buffer
VCA	Viral Capsid Antigen
WHO	World Health Organization
XLPS	X-linked lymphoproliferative syndrome