

بِسْمِ اللَّهِ الرَّحْمَنِ الرَّحِيمِ

قال الله تعالى

قَالُوا سُبْحَانَكَ لَا عِلْمَ لَنَا إِلَّا مَا عَلِمْتَنَا إِنَّكَ أَنْتَ الْعَلِيمُ {
} الْحَكِيمُ

صدق الله العظيم

سورة البقرة الآية 32

Dedication

To my lovely mother

To my great father

To my Daughter

To my Son

To my Family

To my Friends

Acknowledgement

- ❖ All praise and thanks to Allah the Almighty, who blessed me with the courage for preparation and completion of this study.
- ❖ With a great deal of respect I want to thank my supervisor Dr. Humodi Saeed, who spared neither time nor effort in enlightening and helping me.
- ❖ I want to extend my deep thank to Dr. Mogahed, Ustaz. Mansoor and Dr. Mesk Alyaman for their help and support.
- ❖ I express my deepest thanks to U. Mohammed Mosad for his grateful support.
- ❖ This study is honored by the help of the following:-
 - ❖ Mr. Modather, Mr. Muntaser and Miss. Egbal.
 - ❖ I am very grateful to Microbiology department staff that participated in this study.
 - ❖ It is a pleasure to express my respect sincere thanks and gratitude to all test subjects for agreement to participate in this study.

Abstract

This study was carried out in the period from May 2007 - March 2008 to detect the presence of *Klebsiella pneumoniae* in patients suffering from pneumonia. Forty seven sputum samples were collected from patients attended Albangadid Hospital, Alshab Hospital, and National Health Laboratory.

Bacterial DNA was extracted from each sputum specimen using Phenol Chloroform method. Real-time PCR technique was adopted to detect the presence of *Klebsiella pneumoniae* using Real Time PCR.

The result revealed that only three (6.3%) specimens were positive.

The study concluded that the gene probe technique facilitates detection of bacterial pathogens without bacteriological culture.

تم تنفيذ هذه الدراسه فى الفتره من مايو 2007 الى مارس 2008 لاكتشاف الكلبيسيله النزليه . فى مرضى يعانون من الالتهاب الرئوي

جمعت حوالى 47 عينه قشع من مستشفيات الولايه المختلفه وهى مستشفى الباى جدید . ، مستشفى الشعب التعليمى و المعمل ال قومى الصهى

تم استخلاص الحمض النووي لكل عينات القشع بواسطه طریقه فينول كلوروفورم ، ثم استخدمت طریقة التفاعل البلمرى المتسلسل الزمنى لاكتشاف الكلبيسيله النزليه

أوضحت النتائج المتحصل عليها وجود 3 عينات ايجابيه مما يمثل (6.3%) من مجموع العينات التي جمعت .

تخلص هذه الدراسه الى ان طریقه التفاعل البلمرى المتسلسل الزمنى تسهل اكتشاف البكتيريا المرضيه دون الحاجه للمزارع البكتيريه

Table of Contents

No	Contents	page
	الخلاصة	I
	Dedication	II
	Acknowledgement	III
	Abstract (English)	IV
	Abstract (Arabic)	V
	Table of Contents	VI, VII
	List of Tables	VIII
	List of Figures	VIII
	Chapter One: Introduction & literature review	
1.1.	Introduction	1
1.1.1.	Entrance	1
1.1.2.	The genus <i>Klebsiella</i>	1
1.1.2.1.	History	1
1.1.2.2.	Definition	2
1.1.2.3.	Classification	2
1.1.2.4.	Normal habitat	3
1.1.2.5.	Antigenic structure	3
1.1.2.6.	Pathogenicity	3
1.1.2.6.1.	Transmission	3
1.1.2.6.2.	Virulence	4
1.1.2.7.	Disease	5
1.1.2.8.	Epidemiology	5
1.1.2.9.	Mortality/ Morbidity	6
1.1.2.10.	Lab diagnosis	7
1.1.3.	Literature Review	10
1.1.4.	Objective	12
	Chapter Two: Materials and Methods	
2.1.	Study area	13
2.2.	Subjects	13
2.3.	Age groups	13
2.4.	Sample size	13
2.5.	Site of collection	13
2.6.	Sterilization	13
2.7.	Experimental work	13
2.7.1	Collection of Samples	13
2.7.2.	DNA extraction	13
2.7.2.1.	Reagent preparation	13

2.7.2.2.	Method	14
2.7.3.	DNA amplification and analysis	14
2.7.3.1.	Equipments & Reagents	14
2.7.3.2.	Master Mix preparation	15
2.7.3.3.	Plate preparation	15
2.7.3.4.	Amplification	15
Chapter Three: Results		
3.	Results	16
3.1.	Clinical specimens	16
3.2.	DNA extraction	16
3.3.	Detection of <i>Klebsiella pneumoniae</i>	16
Chapter four		
4.	Discussion	22
	References	24
	Appendices	28

List of Tables

Table 1. Distribution of samples according to Hospitals	16
Table 2. Distribution of samples according to gender	17
Table 3. Distribution of samples according to Age	17
Table 4. Plate preparation	17
Table 5. Plate showed the positive specimens	18

List of Figures

Figure 1. Shows plate layout	19
Figure 2. Shows negative control	20
Figure 3. Shows one positive result duplicate	20
Figure 4. Shows one positive result duplicate	21
Figure 5. Shows one positive result duplicate	21