

Impact of Different Static Magnetic Field Forces on Phenotypic and Genotypic Characteristic of Uropathogenic *Escherichia coli*

A Thesis

Submitted to the Council of the Erbil Health and Medical Technical College at Erbil Polytechnic University in Partial Fulfillment of the Requirements for the Degree of Master of Science in Medical Laboratory Technology

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بيني مراكلة الرّحمز الرّحيب مر

[قُلْ هَلْ يَسْتَوِي الَّذِينَ يَعْلَمُونَ وَالَّذِينَ لَا يَعْلَمُونَ إِنَّمَا يَتَذَكَّرُ أُوْلُوا الْأَلْبَابِ]

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I declare that the Master Thesis entitled: (Impact of Different Static Magnetic Field Forces on Phenotypic and Genotypic Characteristic of Uropathogenic *Escherichia coli*) is my original work. I, at this moment, certify that unless stated, all work contained within this thesis is my independent research and has not been submitted for the award of any other degree at any institution except where due acknowledgment is made in the text.

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DEDICATION

This thesis is dedicated to:

- ✤ My mother and memory of my father...
- ✤ My always encouraging, ever faithful brothers and sisters...
- My husband Idries H. Ramaddan and my lovely daughter, Ronya and my sweet son, Rawan...
- ✤ All who taught me, my college, teachers and friends?

Amanj Jamal Azeez

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SUMMARY

The current study aimed to investigate the impact of a static magnetic field (SMF) exposure on uropathogenic *Escherichia coli* colony morphology, cell growth, viability, biochemical characteristics, antibiotic susceptibility and gene expression from urine clinical specimens.

Twenty- five *E.coli* is being isolated clinical samples obtained from urine of patients attended to different hospitals (Erbil, Rizgary, and Rapareen Teaching Hospital in Erbil city/Iraq. All isolates were identified using cultural, morphological, biochemical characteristics, and the using Vitek 2 system for identification.

The magnetic field created manually with the power of (0.04, 0.08, 0.12, 0.16T) and have been measured the force in the Physics Department of the College of Education at the University of Salahaddin in Erbil/ Iraq. The bacterial culture in broth media exposed to different force of magnetic field.

Our findings revealed that exposure to SMF (0.04, 0.08, 0.12, 0.16T) decreased optical density at 620 nm over the course of 24 hours. Also finding exposed bacteria to different magnetic force been altered bacterial biological activity on sugar fermentation and antibiotic sensitivity due to mutation.

In addition, the Vitek 2 system has been used for measuring the antibiotic susceptibility of bacteria against different magnetic fields. After 24 hours of exposure, the minimum inhibitory concentration (MIC) value was calculated. The antibiotics Ciprofloxacin, Trimethoprim/sulfamethoxazole, Ceftazidime, Cefepime and Aztreonam converted from sensitive to resistant compared with negative control (unexposed).

*Escherichia col*i isolates were put through a PCR procedure using the appropriate primer *16SrRNA* to establish their identity as well as other primers *TEM1.CTXM-1 SHV* genes that encode for a multidrug-resistant strain MDR.

The interpretation of the differential expression of the TEM_{I} . CTX_{M-I} , SHV, and I6SrRNA genes under different SMF exposure revealed that the expression level of the I6SrRNA amplification PCR product remained constant throughout the exposure and thus can be used as a reference gene for the observation of the differential gene expression of *E. coli*. Notably, the amplified PCR products of TEM_{I} . CTX_{M-I} , and SHV genes were decreased after different SMF exposure as compared to non-exposed (control) that's lead to increase antibiotic susceptibility. The TEM_{I} . CTX_{M-I} genes were subjected to a genomic study; (Bio Edit V.7.0.5) was used to evaluate the quality of their sequencing data. Utilizing NCBI- BLAST, homology, insertions - deletions, stop codons, and frame shifts were investigated. Laboratory or query sequences were examined and aligned with a second biological sequence to identify a greater degree of similarity and nucleotide variation with other targets.

LIST OF CONTENTS

No.	TITLE	Page
	DECLARATION	II
	SUPERVISOR CERTIFICATE	III
	EXAMININGCOMMITTEE CERTIFICATION	IV
	DEDICATION	V
	ACKNOWLEDGEMENT	VI
	SUMMARY	VII
	LIST OF CONTENTS	IX
	LIST OF FIGURES	XIII
	LIST OF TABLES	XVI
	LIST OF ABBREVIATIONS	XVII
Chapter One		
1	INTRODUCTION	1-4
1	Introduction	1
	Chapter Two	
2	LITERATURE REVIEW	5-28
2.1	Magnetic Field Properties and Their Differential Biological Effects	5
2.2	Dynamic magnetic Field vs. Static Magnetic Field	6
2.3	Earth's Magnetic Field Strength	6
2.4	Cellular Impacts of Static Magnetic Fields (SMFS)	7
2.4.1	Cell Orientation	7
2.4.2	Cell Proliferation/Growth	8
2.4.3	Morphology	9
2.4.4	The Impacts of Static Magnetic Fields and How They Affect Human Bodies	10
2.4.4.1	Magnetic field effects on major blood cells	10
2.4.4.2	Magnetic Field's Influence on Bone Cells	12
2.4.4.3	Neuronal and Brain Influences of Magnetic Fields	14
2.5	Magnetic Therapy Using Static Magnetic Fields	14

2.5.1	Magnetic Resonance Imaging (MRI)	14
2.5.2	SMF Impact on Cancer Cells	15
2.6	Effect of SMF on Animals	16
2.6.1	Static magnetic fields' impacts on bone regeneration	16
2.6.2	Neurons and brain	17
2.7	Static magnetic field impact on microorganisms	17
2.7.1	Impact of Static magnetic field on bacterial cell	17
2.7.1.1	Static magnetic field influence on bacterial growth	18
2.7.1.2	Static magnetic field influence on biochemical	20
	characterization	
2.7.1.3	Static magnetic field influence on Antimicrobial susceptibility	21
2.8	Urinary tract infections (UTIs)	22
2.9	Uropathogenic E.coli	23
2.9.1	UPEC virulence factor	24
2.9.2	Antibiotics Susceptibility of Uropathogenic E. coli	25
2.9.2.1	Extended-spectrum β-lactamases (ESBLs)	25
2.9.2.2	Multidrug Resistance and Uropathogenic Escherichia coli	26
2.9.3	Molecular Characterization of Uropathogenic E.coli	27
Chapter Three		
3	MATERIALS AND METHODS	29-42
3.1	Materials	29
3.2	Methods	30
3.2.1	Culture Media Preparation	30
3.2.1.1	MacConkey Agar Medium	30
3.2.1.2	LB Broth (Luria-Bertani)	31
3.2.1.3	Nutrient broth	31
3.2.1.4.	Brain Heart Infusion Agar	31
3.2.2	Collection of samples	31
3.2.3	Phenotypic identification of isolates	32

3.2.3.1	Colony morphology	32
3.2.3.2	Using the VITEK 2 System for isolate detection	32
3.2.3.3.	Antibiotic Susceptibility Testing	33
3.2.4	Preparation of magnetic field	33
3.2.4.1	Exposed bacteria to the different static magnetic field	33
3.2.5	Bacterial Maintenance and Storage (storing)	34
3.2.6	Molecular analysis of the isolates	34
3.2.6.1	Genomic DNA Extraction	35
3.2.6.2	Determination of DNA Concentration	36
3.2.6.3	Primers and PCR Amplification	36
3.2.6.3.1	Primer Preparations	36
3.2.6.3.2	Detection of 16SrRNA gene of E. coli isolates by PCR	36
3.2.6.3.3	Detection of <i>TEM</i> ₁ gene of <i>E</i> . <i>coli</i> isolates by PCR	37
3.2.6.3.4	Detection of <i>CTX</i> - _{<i>M</i>-1} gene of <i>E</i> . <i>coli</i> isolates by PCR	38
3.2.6.3.5	Detection of SHV gene of E coli isolates by PCR	39
3.2.6.3.6	Detection of Amplified Products by Agarose Gel Electrophoresis	40
3.2.7	Data analysis	41
3.2.8	Statistical Analysis	41
Chapter Four		
4	RESULT	43-101
4.1	Isolation and Identification of <i>E. coli</i>	43
4.2	Exposing Uropathogenic <i>E.coli</i> to Different Static Magnetic Field	44
4.3	Growth Characteristics Curve	44
4.4	Investigation of Magnetic Field on Bacteria Antibiotic Sensitivity	46
4.5	Investigation of Magnetic Field on Biochemical Properties of Bacteria	61
4.6	Molecular Characterizations of isolated E.coli	78
4.6.1	Detection of 16SrRNA gene among E.coli isolates using	78

	PCR Technique	
4.6.2	Detection of CTX_{M-1} gene among <i>E.coli</i> Isolates using PCR Technique	79
4.6.3	Detection of TEM_1 gene among <i>E. coli</i> isolates using PCR Technique	81
4.6.4	Detection of SHV gene Among E. coli Isolates using PCR Technique	82
4.7	Sequence alignment and submission gene to GenBank	85
4.7.1	Molecular sequencing for <i>TEM</i> ₁ gene	86
4.7.2	Molecular sequencing for CTX_{M-1} gene	91
4.7.3	Alignment and detection of variant amino acid	98
4.7.4	Phylogenetic inferences	99
Chapter Five		
5 DISCUSSION 102-108		102-108
5	DISCUSSION	102
Chapter Six		
6	CONCLUSION AND RECOMMENDATIONS	109-110
6.1	CONCLUSION	109
6.2	RECOMMENDATIONS	110
	REFERENCES	R1-R22
	APPENDICES	A1-A4
	پوخته	a I

LIST OF FIGURES

No.	Content	Page
(3-1)	(A):Agarose gel electrophoresis apparatus (B)Thermal cycle PCR machine (C): locally made device static magnetic field	42
(4-1)	Showing Colony morphology of <i>E. coli</i> (A) unexposed to magnetic fields (control), (B) treated to magnetic field 0.04T, (C) treated to 0.08T, (D) treated to 0.12Tand (E) treated to 0.16T.	43
(4-2)	Statistical analysis showing significant differences between optical density (620) nm of <i>E. coli</i> unexposed (Control) vs. optical density (620) nm <i>E. coli</i> under magnetic power (0.04, 0.08, 0.12 and 0.16 T) respectively.	46
(4-3)	Agarose (1%) gel electrophoresis of PCR product of amplified <i>16SrRNA</i> gene of <i>E.coli</i> isolates (amplicon with 1343bp). Group (I-IX) represent number of sample and (0) control unexposed to the magnetic field (1, 2, 3, 4) sample exposed to magnetic power (0.04, 0.08, 0.12 and 0.16 T)) respectively.	78
(4-4)	Agarose (1%) gel electrophoresis of PCR product of amplified <i>16SrRNA</i> gene of <i>E. coli</i> isolates (amplicon with 1343bp). Group (I-IX) represent number of sample and (0) control unexposed to the magnetic field (1, 2, 3, 4) sample exposed to magnetic power (0.04, 0.08, 0.12 and 0.16 T) respectively.	79
(4-5)	Present statical analysis showing significant differences between CTX_{M-1} gene positive for <i>E.coli</i> unexposed vs. CTX_M -1 gene positive for <i>E.coli</i> under magnetic power (0.04, 0.08, 0.12 and 0.16 T) respectively.	80
(4-6)	Agarose (2%) gel electrophoresis of PCR product of amplified CTX_{M-I} gene of <i>E.coli</i> isolate (amplicon with 893bp), group (I-VI) represent number of sample and (0) control unexposed to the magnetic field (1, 2, 3, 4) sample exposed to magnetic power (0.04, 0.08, 0.12 and 0.16T) respectively.	80
(4-7)	Statistical analysis showing significant differences between TEM_1 gene positive for <i>E. coli</i> unexposed vs TEM_1 gene positive for <i>E. coli</i> under magnetic power (0.04, 0.08, 0.12 and 0.16T) respectively.	81
(4-8)	Agarose (2%) gel electrophoresis of PCR product of amplified TEM_1 gene of <i>E. coli</i> isolate (amplicon with 535bp), group (I-IV) represent number of sample and (0) control unexposed to the magnetic field (1, 2, 3, 4) sample exposed to magnetic power (0.04, 0.08, 0.12 and 0.16 T) respectively.	82
(4-9)	Statical analysis showing significant differences between SHV gene	83

	positive for <i>E.coli</i> unexposed vs. <i>SHV</i> gene positive for <i>E.coli</i> under magnetic power (0.04, 0.08, 0.12 and 0.16T) respectively.	
(4-10)	Agarose (2%) gel electrophoresis of <i>SHV</i> gene amplification, L: ladder Group I represent number of samples, (0) control unexposed to the magnetic field (1, 2, 3, 4) sample exposed to magnetic power (0.04, 0.08, 0.12 and 0.16T) respectively (1, 2, 3) positive results (4) negative result.	83
(4-11)	Genotyping of <i>E.coli</i> isolates using Conventional-PCR method through the ESBL gene. The dendrogram was constructed using Conventional-PCR patterns of <i>16SrRNA</i> and ESBL genes (<i>blaCTX_{M-1}, blaTEM₁, and blaSHV</i>) of <i>E.coli</i> isolates. Similarity clustering analysis was performed using the Hierarchical Cluster. The dashed line is a hypothetical line showing ~90% similarity.	84
(4-12)	NCBI blasting pairwise alignment of control samples (unexposed to the magnetic field) sequences query of β -lactamase (<i>TEM</i> ₁) gene with the subject of <i>Escherichia coli</i> in NCBI.	86
(4-13)	NCBI blasting pairwise alignment of sequences for the beta- lactamase (TEM_1) gene in <i>E.coli</i> with a sample exposed to (0.04T) of magnetic field.	87
(4-14)	NCBI blasting pairwise alignment of sequences for the beta- lactamase (TEM_1) gene in <i>E.coli</i> with a sample exposed to (0.08T) of magnetic field.	88
(4-15)	NCBI blasting pairwise alignment of sequences for the beta- lactamase (TEM_1) gene in <i>E.coli</i> with a sample exposed to (0.12T) of magnetic field.	89
(4-16)	NCBI blasting pairwise alignment of sequences for the beta- lactamase (TEM_1) gene in <i>E.coli</i> with a sample exposed to (0.16T) of magnetic field.	90
(4-17)	NCBI blasting pairwise alignment of control samples (unexposed to the magnetic field) sequences query of beta-lactamase (CTX_{M-1}) gene with the subject of <i>Escherichia coli</i> in NCBI.	91
(4-18)	NCBI blasting pairwise alignment of beta-lactamase(CTX_{M-1}) gene sequences from an <i>Escherichia coli</i> sample exposed to (0.04T) of magnetic power	92
(4-19)	NCBI blasting pairwise alignment of the sample exposed to $(0.08T)$ of magnetic power sequences query of beta-lactamase (CTX_{M-1}) gene with the subject of <i>Escherichia coli</i> .	93
(4-20)	NCBI blasting pairwise alignment of beta-lactamase (CTX_{M-1}) gene sequences from an <i>Escherichia co</i> li sample exposed to (0.12T) of	94

	magnetic power	
(4-21)	NCBI blasting pairwise alignment of beta-lactamase (CTX_{M-1})) gene sequences from an <i>Escherichia coli</i> sample exposed to (0.16T) of magnetic power.	95
(4-22)	(A), (B) Multiple protein sequence alignment analysis of $blaTEM_I$ gene among 5 (first sample control and other 4 samples treated to magnetic power (0.04, 0.08, 0.12 and 0.16T) respectively <i>Escherichia coli</i> .	98
(4-23)	(a), (b) Multiple protein sequence alignment analysis of $blaCTX_{-M-1}$ gene among five first sample controls and other 4sample treated to magnetic power (0.04, 0.08, 0.12 and 0.16T) respectively <i>Escherichia coli</i> .	99
(4-24)	Employing Maximum Likelihood with boost strap with red numbers of Mega 11 program show phylogenetic positioning of each mutant of 5 samples (first sample control and other 4sample treated to magnetic power (0.04, 0.08, 0.12 and 0.16T)) respectively samples with similar GenBank sequences $blaTEM_1$ gene among five isolates of <i>Escherichia coli</i> .	100
(4-25)	Employing Maximum Likelihood with Boost strap with red numbers of Mega 11 program show phylogenetic positioning of each mutant of 5 samples (first sample control and other 4 samples treated to magnetic power (0.04, 0.08, 0.12 and 0.16T) respectively samples with similar GenBank sequences $blaCTX_{M-1}$ gene among 5 <i>Escherichia co</i> li.	101

LIST OF TABLES8

No.	Content	Page
(2-1)	Virulence factors of the UPEC	24
(3-1)	Equipment and instruments used in the study	29
(3-2)	Culturing media and chemical substances employed during the investigation	30
(3-3)	The molecular study used commercial kits and materials	30
(3-4)	Present all primers were used in this study	36
(3-5)	Presented primers of 16SrRNA gene with PCR program of <i>E.coli</i> used in this study	37
(3-6)	Presented used primers of <i>TEM</i> ₁ gene with PCR program of <i>E.coli</i>	38
(3-7)	Presented primers of CTX_{M-1} gene with PCR program of <i>E. coli</i> used in this study	39
(3-8)	Presented primers of SHV gene with PCR program of E. coli used in this study	40
(4-1)	Effect of different magnetic field forces on growth rate of Uropathogenic <i>E.coli</i> .	45
(4-2)	Susceptibility test for <i>E.coli</i> result before and after different MG field exposure	47
(4-3)	Presented the result of sugars fermentation metabolism in E.coli	62
(4-4)	GenBank accession no. of both gene $blaTEM_1$ and $blaCTX_{M-1}$ of <i>Escherichia coli</i>	96
(4-5)	Explain the numbers and variant position of nucleotides of each sample are changed with amino acid codons in $blaTEM_1$ and $blaCTX_{-M-1}$ genes of <i>Escherichia coli</i>	97

LIST OF ABBREVIATIONS

Abbreviation	Abbreviation Mean
16SrRNA	16S ribosomal ribonucleic acid rRNA, S=Svedberg sedimentation
	rate measurement
ABI	Applied Bio systems Instruments
AC	Alternating current
AK	Amikacin
AST	Arginine succinyl transferase
astCADBE	operons for arginine catabolism
ATM	Aztreonam
BioEdit	Free biological sequence alignment
BLAST	Basic local alignment search tool
bp	Base pair
°C	Degree Celsius
CARBA	Carbapenems
CAZ	Ceftazidime
CFU	Colony forming unit
CIP	Ciprofloxacin
Cm	Centimeter
СТ	Computed tomography
CTX_{M-1}	Preferential hydrolytic activity against cefotaxime (CTX as its
	acronym, -M from Munich).
DC	Direct current
DEGs	Differentially expressed genes
DNA	Deoxy-ribonucleic acid
DNases	DNases are enzymes that hydrolyze DNA
E .coli	Escherichia coli
ESBL	Extended spectrum beta-lactamases
ExPEC	Extra-intestinal pathogenic Escherichia coli
FASTA	Federal Aviation Science and Technological Association
FEP	Cefepime
G	Gauss (unit for measurement of magnetic field)
g	Gravity
GEN	Gentamicin
glc	Operon involved Glycolate utilization
glp	Glucagon-like peptide
glpFKX	operons responsible for glycerol catabolism
GMF	Geomagnetic field
Hz	Hertz (unit of frequency)

IPM	Imipenem
lacZ	Gen encodes β-galactosidase (LacZ)
LB	Lauria Bertani broth
LEV	Levofloxacin
MDR	Multi drug resistant
MEGA	Molecular evolutionary genetic analysis
MEM	Meropenem
ml	Milliliters
MRI	Magnetic resonance imaging
mT	Millitesla
NCBI	National Center for Biotechnology Information
NCIMB	National Collection of Industrial, Food and Marine Bacteria
NdFeB	Type of Magnets made from combination of Neodymium. Iron,
	Boron. Cobalt
NET	Netilmicin
nm	Nano meter for measuring wave-lengths of light
OXA	Oxacillinasesvariants
PCR	PCR Polymerase chain reaction
PIP	Piperacillin
PMF	Pulse magnetic field)
rpm	Revolutions per minute
SHV	Sulf-hydryl variable active site
SMF	Static magnetic field
SOD	Superoxide dismutase
SoxS	Activator of superoxide stress genes
SXT	Trimethoprim/sulfamethoxazole
т	(Tesla) the international system unit of field intensity for magnetic
I	field
TBE	Tris-EDTA-Borate Buffer
TCA	Citric acid cycle
TEM_1	Temoniera in Greece
TIG	Tigecycline
TZP	Tazobactam/piperacillin
TZP	Piperacillin\tazobactam
UPEC	Uropathogenic Escherichia coli
UTIs	Urinary tract infections
V	Volts
μL	Microliter

Chapter One INTRODUCTION

1. INTRODUCTION

Many experiments have demonstrated that live organisms may respond to Changes in outward magnetic fields, such as the magnetic field of the Earth (Binhi, 2002; Miyakoshi, 2005; Lohmann, 2010). Any recorded physiological reaction of an organism to a magnetic field is accompanied by a complicated sequence of intracellular metabolic changes (Funk *et al.*, 2009).

Since more than 4 billion years ago, the Ground has produced a modest static magnetic field (SMF), often referred to as the geomagnetic field (GMF), with a surface intensity of roughly fifty micro tesla (Le Mouël *et al.*, 2023). As the beginning of living happened after the development of GMF, GMF information may have been employed by microorganisms as a useful natural resource to address the challenges of development. There is growing evidence that GMF can act as signals to assist numerous animals adjust to significant environment modifications and as energy to independently influence metabolic processes (Clites and Pierce, 2017; Mouritsen, 2018). Many animals as insects, turtles, fishes, and birds, may travel thousands of kilometers using GFM as a signal, and magneto therapy is commonly used to reduce pain and repair a variety of illnesses, including diabetes, immunological difficulties, and mental disorders (Markov, 2007; Clites and Pierce, 2017).

Single-cell microorganisms are routinely used to investigate various magnetic biological effects and the molecular processes underlying them. The dynamic intracellular and extracellular aims of SMF as well as study methodologies including SMF intensity and gradients, bacterial strains, culture variables ,treatment time, and may all have an influence on the outcomes of SMF (Hunt *et al.*, 2009;Křiklavová *et al.*, 2014). Understanding the biological impacts of SMF on bacteria will significantly contribute to

enhancing our environment, sanitation, and health, given the pervasive presence of bacteria in our climate and bodies (Li *et al.*, 2022).

E. coli was significantly less viable after 30 minutes of treatment to type of Magnets made from combination of Neodymium, Iron, and Boron. Cobalt (NdFeB) magnets through intensities ranging from 45 mT to 3,500 mT, leading to disruption to the cell surface (Ji *et al.*, 2009). Similarly, a 4-hour exposure with hundred mT homogenous SMF created by an electromagnet significantly inhibited microbial adhesion and ensuing colony formation leading to the breakdown of the cell membrane and the liberation of the cytoplasm from the inner membrane (Bajpai *et al.*, 2012).

According to additional research, *E. coli* cells exposed to two hundred fifty mT SMF had smaller colonies. The disruption of carbon source consumption, notably long-chain fatty acid as well as glyoxylate metabolism, enabled *E. coli* to adapt to the moderately intense SMF, as determined by transcriptomic and metabolomics analyses (Li *et al.*, 2022).

E. coli, an abundant gram-negative bacteria and member of the family *Enterobacteriacea*, is the most common cause of urinary tract infections on a global scale (Halaji *et al.*, 2020). One of the most frequent types of extraintestinal pathogenic *E.coli* (ExPEC) is uropathogenic *E.coli* (UPEC) (Bunduki *et al.*, 2021). *E.coli* is one of the most common causes of septicemia and neonatal meningitis associated with urinary tract infections (UTIs). Bacteria ascending from the periurethral region to the urethra, bladder, and upper urinary tract are the cause of UTIs (Hussein *et al.*, 2022). Colonization of the periurethral region by uropathogenic bacteria is a crucial cause of UTIs (Bunduki *et al.*, 2021).

Certain microorganisms produce ESBLs, which are enzymes with the ability to break down extended-spectrum cephalosporins. Therefore, they are effective against beta-lactam antibiotics such as ceftazidime,ceftriaxone,

cefotaxime, oxyiminocephalosporin, and monobactams (Bradford, 2001; Paterson and Bonomo, 2005). ESBL-producing bacteria target antibiotics include carbapenems and cephamycins. ESBLs are often suppressed by clavulanic acid and tazobactam. *Enterobacteriacea*, in particular, are gramnegative bacteria that possess ESBLs (Hussain *et al.*, 2021).

The main cause of (UTIs) is uropathogenic *E.coli* (UPEC) (Al-Jamei *et al.*, 2019). β -Lactam antimicrobial agents are the most significant family of medications for treating UPEC infections acquired in hospitals and the general public (Muriuki *et al.*, 2022). A clinical problem is the emergence and spread of bacterial resistance to beta-lactam medications, which causes healing failure and recurring infections (Gajamer *et al.*, 2020).

Resistance genes are frequently found on bacterial plasmids, which are moveable DNA pieces that can easily travel throughout bacterial populations and other bacterial species. First discovered in Germany in the 1980s, plasmid-encoded ESBL synthesis provides resistance to most β -lactam antibiotics (King *et al.*, 2012). *TEM*, *CTX-*_{*M-1*} and *SHV* beta-lactamases are among the most prevalent members of the vast family of class (A) β lactamases, which also includes several other uncommon enzymes that frequently display ESBL activity. Genetic procedures, such as polymerase chain reaction (PCR) and sequencing, common techniques for detecting specific β -lactamase genes in bacterial isolates, are the best way to describe and determine the occurrence of a β -lactamase gene (Ahmed *et al.*, 2013).

The aim of this study

- 1- Evaluate the effect of SMF exposure (0.04, 0.08, 0.12 and 0.16T) on the uropathogenic *Escherichia coli* that caused urinary tract infection of human and to determine the growth rate, antibiotic susceptibility against antibiotics, investigation of biochemical properties.
- 2- Detection of *E.coli* identification (*16SrRNA*) genes by using conventional PCR to compare exposed bacterial growth with unexposed as a negative control.
- 3- Differential detection of the genes *bla CTX*_{-*M*-*I*}, *bla TEM*₁, and *bla SHV* by using conventional PCR to compare exposed bacterial growth with unexposed as a negative control.
- 4- DNA sequencing was used for *bla* CTX_{-M-1} and *bla* TEM_1 to compare exposed bacterial growth with unexposed as a negative control.

Chapter Two LITERATURE REVIEW

2. LITERATURE REVIEW

2.1. Magnetic Field Properties and Their Differential Biological Effects

Researchers have long been interested in the influence of low MF intensity on organisms. In 1600, W. Gilbert (1544–1603, England), the inventor of magnetic navigation, wrote about Galen, Plutarch, Ptolemy, and Paracelsus, "Others say that loadstone causes mental disturbance and makes a person gloomy, and sometimes is lethal" (Binhi and Rubin, 2022). Alexander .F. von Middendorf was the forerunner of the academic Studies into the biological impacts of the MF (1815–1894). He claimed that, depending on his findings of migratory birds, "Our sailors of heaven' take on the function of a compass for ships" (Binhi and Rubin, 2022).

The practical research of MFs' biological impacts was connected to medicine. For instance, it is known that German physicians began exploring the medicinal possibilities of MF by using permanent magnets on various body areas as early as the 1750s (ALMazrouei, 2021). Numerous medical professionals from multiple nations attempted to employ artificial direct current (DC), alternating current (AC), and permanent magnets for physiotherapeutic reasons throughout the nineteenth and first half of the twentieth century, according to the research (Binhi and Rubin, 2022).

With the emergence and advancement of millimeter-wave electromagnetic technology, the investigation into the basis of these phenomena started primarily in the 1960s in the Union of Soviet Socialist Republics (USSR) and the United States of America (USA). It was discovered that organisms and plants might be affected by millimeter waves (Chukova, 2015; Finance and Bio, 2021; Binhi and Rubin, 2022).

2.2. Dynamic Magnetic Field vs. Static Magnetic Field

The impacts of magnetic fields on living microorganisms are directly influenced by several variables. Magnetics fields may be categorized as both static magnetic fields (SMF) and dynamic magnetic fields, which may be expanded categorized according to their frequencies (McNaughton, 2022). The phrase static magnetic field is used when the magnetic field's intensity stays constant across time. In contrast, a "dynamic magnetic field" or "timevarying magnetic field" is one in which the strength of the MF differs with time. Most often detected are pulsed magnetic fields (PMFs), which include radiofrequency magnetic fields and alternating current (AC) magnetic fields with 50 Hz as well as 60 Hz power frequencies (Du, 2021). There are four distinct magnetic force strengths: weak, moderate, strong (high), and ultrastrong (ultra-high). According to the geographic spreading of the magnetic force, there are homogeneous and inhomogeneous magnetic fields (Novickij et al., 2014; Amiri et al., 2019). Other researches go into detail about their variations' impacts on biological items. Natural and human-made magnetic fields permeate our environment. The Earth's magnetic fields normally don't go over 100 mT, however artificial magnetic fields used in welding equipment or medical devices can go above this level (Zhang et al., 2017b; Driessen et al., 2020).

2.3. Earth's Magnetic Field Strength

The most common SMF to which everyone is exposed is the geomagnetic field (GMF), which is around 0.5 Gauss/50 μ T (varies depending on location). It is quasi-static, meaning it is partly variable. The geomagnetic field is far weaker than other forms of SMF exposure, but it is essentially ubiquitous and essential for all life on Earth. According to one idea, solar wind might remove the atmospheres of planets without complete global magnetic forces. Many think, for instance, that Mars absences a worldwide

magnetic field; hence, the solar wind has caused Mars' environment to lose water and deteriorate. Researchers think that the magnetic field of Earth (the magnetosphere) protects Earth from the potentially devastating effects of solar wind (Erdmann *et al.*, 2021).

Intriguingly, certain human studies indicate that geomagnetic field could induce neurological and cardiovascular consequences. Burch *et al.* (2008) suggest that GMF exposure can alter melatonin secretion (a hormone made by brain in respond to darkness helps control the body's sleep cycle and regulation of energy metabolism and glucose homeostasis), which of the following is a potential technique for the neurological and cardiovascular impact of changed GMF. As well, Lipnicki (2009) demonstrates that there may be a link between GMF activities and bizarreness in dreams. However, there are also unfavorable results recorded in some studies. Sastre *et al.* (2002) evaluated the impacts of measured variations in the geomagnetic field on fifty human volunteers were studied using electroencephalogram (EEG) and no association was found.

2.4. Cellular Impacts of Static Magnetic Fields (SMFS)

2.4.1. Cell Orientation

First, it was observed that sickled erythrocytes at 0.35 T were perpendicular to the magnetic field (Miyakoshi, 2005). Erythrocytes were oriented with their disk planes perpendicular to the MFs orientation (Ji and Zhang, 2023). This impact on erythrocytes was observable at (1-4T); approximately one hundred percent of cells were oriented. It has been discovered that 1T static magnetic fields may induce the orientation of macromolecules like collagen and animal cell cultures (Miyakoshi, 2005; Miyakoshi, 2006). Furthermore, the magnetic orientation of collagen was utilized to direct exposure of human foreskin fibroblasts to SMFs of 4.0 and 4.7 T(Miyakoshi, 2006). In addition, it has been shown that osteoblast cells are aligned in the lack of collagen when subjected to an eight tesla SMF (Kotani *et al.*, 2000; Miyakoshi, 2006).

Human glioblastoma at 10 T, A 172 cells implanted in collagen gels were aligned perpendicularly to the direction of the SMF (Hirose *et al.*, 2003). After being exposed to a static magnetic field for seven days, A172 cells that were cultivated without collagen failed to exhibit any discernible orientation pattern.

This exploratory study examined the impact of exposure to 0.5 tesla of (SMF) on the adhesion of *E.coli*. Researchers looked examined how well bacteria adhered to glass and glass coated with indium tin oxide (ITO) when subjected to either a parallel or perpendicular magnetic force (vectors of magnetic induction are parallel or comparable to the adhesion surface, respectively) (Txintxurreta *et al.*, 2021). Reference cultures were grown under the same circumstances as experimental cultures, but they were not subjected to a magnetic field. The researcher detected a decline in cell adhesion after treating the cells to a magnetic field. After exposure to a parallel magnetic field, the orientation of bacteria cells altered. Nevertheless, no change in the orientation of bacterial cells was seen when they were exposed to a perpendicular MF (Miyakoshi, 2006; Txintxurreta *et al.*, 2021).

2.4.2. Cell Proliferation/Growth

Changes in the cell cycle, growth factor signaling, and DNA transcription reflect directly on the cell number and viability and provide valuable parameters for detecting alterations in the cell apparatus and estimating the impact of an extrinsic agent on the intracellular system (Albuquerque *et al.*, 2016). Magnetism is believed to induce alterations in cell growth, which should result in the cell entering a state of proliferation and exhibiting increased glycolysis, biosynthesis of lipids, lactate output, and other macromolecule rates (DeBerardinis *et al.*, 2008). The linkage of specific ions

such as magnesium, manganese, calcium, iron, and cofactors to enzymes may be an additional factor that promotes alterations in the cell cycle (*Soetan et al.*, 2010).

The proliferation of three human tumor cell lines cultured in vitro appears to be inhibited by a seven-tesla magnetic field (Maffei, 2022) Changes in the cell cycle and extensive DNA degradation were ruled out as potential factors. These results suggest that the effect of SMF exposure varies by cell type (Ramazi *et al.*, 2023)

In 2012, Li et al. (2012) noticed that after a period of 48 hours of exposure to a 5 mT SMF, the proliferation of human umbilical artery smooth muscle cells (hUASMCs) was considerably reduced compared to the control group. Gioia et al. (2013) After examining the impact of a 2 mT SMF on swine granulosa cells (GCs), it was discovered that after 72 hours of culture, exposed samples' doubling times were dramatically shortened (p 0.05). In 2016, Wang et al. (2016) noticed that cell proliferation was inhibited when adipose-derived stem cells (ASCs) were exposed to 0.5 T SMF for seven days. According to recent studies, human nasopharyngeal cancer CNE-2Z and colon cancer HCT116 cells can both be inhibited by 1 T and 9 T SMFs (Ji and Zhang, 2023).They establish that the Epidermal Growth Factor Receptor /Protein-kinase,TOR/Mammalian-target-of-rapamycin EGFR/AKT /mTOR motioning passageway, which was upregulated in many malignances, was involved in SMF-induced cancer cell proliferation hang-up (Zhang, 2023). SMF-induced effects on cell proliferation was not only cell typedependent, but also dependent on magnetic field intensity as well as cell density (Song et al., 2023).

2.4.3. Morphology

Several studies have shown that SMFs can alter the cellular architecture. In 2003, Pacini *et al.* (2003) discovered that point two tesla SMF altered the

shape of human skin fibroblast cells. Iwasaka and Ueno (2003) in the same year, it was shown that fourteen tesla SMF altered the morphology of assembly of smooth muscle cells, with the colony morphologies expanding along the path of the magnetic flux. Furthermore, cytoskeleton alterations were time -dependent (Chionna *et al.*, 2005). Dini and Abbro (2005) discovered that after 72 hours of exposure to six militesla SMF affected human leukemia U937 cell shape changes and F-actin changes, membrane roughness and large blebs, and decreased expression of certain macrophage surface markers. In 2013, Gioia *et al.* (2013) indicated that Porcine granulosa cells were treated for three days to a 2 mT SMF exhibited changes in cell length, thickness, and actin and alpha-tubulin cytoskeletons.

Not surprisingly, there are several types of study in which no changes in cell shape were identified after SMF exposure. The research by Iwasaka and Ueno (2003) a three-hour exposure of smooth muscle cells to an 8 T magnetic field did not significantly alter cell shape, including membrane constituents, according to the findings. In 2005, Bodega *et al.* (2005) Several time points were used to investigate the responses of main cultures of astroglial cells to one millitesla sinusoidal, static, or mixed magnetic fields. They discovered no significant changes in actin levels. Likewise, the cell type may have a substantial effect on the SMF-induced morphological changes. In 1999, Pacini *et al.* (2003) Researchers showed that a magnetic field force of 0.2 T affected the shape of human neuronal FNC-B4 cells (Neuroblast cell populations (FNC-B4) are generated from fetal olfactory epithelium), but had no effect on mice leukemia or human breast cancer cells.

2.4.4. Human Body Impacts on Static Magnetic Fields

2.4.4.1. Magnetic field effects on major blood cells

In recent decades, blood flow and microcirculation have been intensely researched. Experiments conducted *in vitro* and *in vivo* revealed changes in blood flow in response to MF. (Schuhfried *et al.*, 2005)investigated the impact of a time-varying magnetic field on the microcirculation and temperature change of human volunteers' feet. Twelve healthy male and female volunteers were treated to fields of low dose-low frequency (100 T, 30 hz) and high dose-low frequency (eight point four mile tesla, ten Hz). Individuals were administered treatments for one week at the same time daily. Great joys and dorsum of the foot microcirculation and temperature were assessed every 5 minutes during and 5-10 minutes after exposure. A decline in microcirculation & a temperature drop were noted for both exposure conditions. Nevertheless, no significant changes in blood values were seen. The finding is that exposure did not alter microcirculation in healthy humans.

The blood viscosity was examined by (Haik *et al.*, 2001), they recorded blood flow with a capillary tube of 3 mm in diameter and accounted for changes in blood viscosity. The tube was placed among two MF generators. First, the temporal flow of blood was calculated under the action of gravity and then under the action of SMF. Increasing the field's intensity continuously accelerated the passage of time. The ten tesla of SMF significantly lowered blood flow by thirty present. They hypothesize that this decrease results from increased blood viscosity caused by SMF (Semeano *et al.*, 2022). The torque exerted by the SMF will rise the attachment of plasma particles with red blood cells, increasing blood viscosity (pirkhider Yaba and Ismail, 2019).

Fasshauer *et al.* (2018) investigated the impact of Magnetic Resonance Imaging on DNA double-strand break. They discovered no proof that Magnetic Resonance Imaging induces DNA double-strand breakage. Other study investigated the influence of a time-varying magnetic field (10 tesla, 50 hz) on blood parameters and immune system constituents. Humans were continually exposed to MF for twenty-four hours. Their result indicates that low-frequency magnetic fields have little influence on blood immunity and functioning (pirkhider Yaba and Ismail, 2019). Polycythemia illness blood viscosity was measured using one point five tesla MRI magnetic fields (Kadhim *et al.*, 2016). Utilizing a U-tube viscometer and a mathematical method, the viscosity of blood samples was determined. The duration of the exposure was increased from 1 minute to 21 minutes. Men between the ages of 28 and 48 who were unwell were provided samples. As the magnetic field is raised, the viscosity of the blood decreases. 1 and 15 minutes of exposure produced the greatest change (Kadhim *et al.*, 2016; pirkhider Yaba and Ismail, 2019).

Under the influence of MF, hematological parameters such as RBC, WBC, and PLT counts were examined. Blood cell count abnormalities cause cardiovascular illness. An increase in leukocyte counts correlates with a 65% increase in mortality risk due to ischemic heart illness. Blood viscosity and oxygen delivery are caused by hemoglobin concentration and are associated with ischemic heart disease in males (Maulood and Mahmud, 2016; pirkhider Yaba and Ismail, 2019).

Dasdag *et al.* (2002) evaluated the impact of PMF on the rheological characteristics of blood. Sixteen male wielders (3-4 hours per day, ten years of fin welding expertise) and 14 healthy people (control group) joined the study. All chosen samples were free of chronic conditions and led a healthy lifestyle. The outcome reveals a considerable variance in hematocrit levels. Other blood factors, including red blood cells, WBCs, and platelets, are nearly identical to those of the reference groups (pirkhider Yaba and Ismail, 2019).

2.4.4.2. Magnetic Field's Influence on Bone Cells

Magnetic fields have been used as a substitute or supplemental therapy for osteoarthritis, spinal fusion, and wound healing (Cook *et al.*, 2015). Magneto therapy, which includes static magnetic fields (SMFs), is a straightforward, risk-free, non-invasive method of treating illnesses, the location of a wound, and the source of pain and inflammation. SMFs are clinically applicable

magnetic fields, particularly in the dental area (Sakata *et al.*, 2008). SMFs are produced by rare earth magnets, which are employed in magnetic - retention devices for implants- or toothretained over dentures (Yang *et al.*, 2013), maxillofacial prosthetics following surgery for trauma and malignancy (Aksu *et al.*, 2014), additionally in orthodontic procedures including space closure, molar distalization, intrusion, the traction of affected teeth, and palatal expansion (Sakata *et al.*, 2008). SMFs improved implant stability and reduced bone loss over the first several weeks of healing (Siadat *et al.*, 2012).

Cells exposed continuously to low-intensity SMFs inhibited the growth of human osteoblast cultures (Denaro *et al.*, 2008). Furthermore, SMFs generated by corrosion currents hindered the development of human osteoblasts (Kim *et al.*, 2017). SMFs promotes the osteoblastic differentiation of human osteoblast-such as MG 63 cells and dental pulp cells (Yun *et al.*, 2016). Additionally, we discovered that human bone marrow-derived mesenchyme stem cells (MSCs) increased more rapidly and differentiated into osteogenic cells in vitro when exposed to fifteen -millitesla SMFs. This intensity is relatively low within the intensity range of moderate SMFs (one millitesla to one tesla) (Kim *et al.*, 2015).

Regeneration of the periodontium necessitates the recruitment of progenitor cells that develop into periodontal ligament cells (PDLCs), mineral-forming cementoblasts, and bone-forming osteoblasts (Miyakoshi, 2005, Wang and Qin, 2012). Its effects on PDLCs, cement oblasts, and osteoblasts differentiation are unknown. By applying SMFs to teeth, implants, and other intraoral structures, it is possible to effectively modify the periodontium's cells. In addition, the impact of SMFs (15mT) on osteoblastic and cementoblastic differentiation in osteoblasts, cementoblasts, and PDLCs was examined. its provide a molecular foundation for the osteogenic and cementogenic activities of SMF, which may stimulate bone or cementum

growth throughout bone renewal and in patients with periodontal illness (Kim *et al.*, 2017).

2.4.4.3. Neuronal and Brain Influences of Magnetic Fields

According to (Dileone *et al.* (2017), Parkinson's disease patients who are exposed to Trans cranial static magnetic fields have dopamine-dependent alterations in cortical excitability. Furthermore, the strength of a static magnetic field may affect the firing frequency of neurons (Viudes-Sarrion *et al.*, 2021). Several studies have established that an applied magnetic field alters sensitivity to pain (nociception) and Pain relief (analgesia) (Del *et al.*, 2007). Moreover, a static magnetic forces has been associated to diabetic neuropathy in the clinic (50 mT) (Weintraub *et al.*, 2003; Zadeh-Haghighi and Simon, 2022).

2.5. Magnetic Therapy Using Static Magnetic Fields

2.5.1. Magnetic Resonance Imaging (MRI)

People are increasingly exposed to considerably greater SMFs, such as those produced by hospital MRI scanners, in addition to the small GMF (50T). MRI is considered a safe procedure if the instructions are followed (Sammet, 2016). This technique generates pictures of interior organs by monitoring the reaction of the atomic nuclei of biological tissues to high-frequency radio waves when put in a strong magnetic field. a method that makes use of radio waves, a powerful magnet, and a computer to produce exact photographs of certain body locations. These pictures aid in identifying healthy and unhealthy tissue (Richardson *et al.*, 2005; Katti *et al.*, 2011).

These images are produced using a technique that combines radio waves, a powerful magnet, and a computer to provide accurate images of different bodily areas and make it easier to distinguish between healthy and sick tissue. The images produced by (MRI) of organs and soft tissue are superior than those produced by computed tomography (CT) and x-rays. Magnetic resonance imaging (MRI) makes it easier to see the interior of the bone, the brain, the spine, and joints' soft tissues. often referred to as nuclear magnetic resonance imaging, MRI (Katti *et al.*, 2011; Reda *et al.*, 2021).

A radiation-free imaging technology that studies the structure and function of the body in both health and sickness, it is commonly used for illness detection, diagnosis, and therapeutic monitoring. It focuses on cutting-edge technology that stimulates and detects changes in the protons present in biological tissue-forming water (Fatahi and Speck, 2015; Reda *et al.*, 2021).

Furtherance of the Earth's Weak Magnetic Field (50 T), people are being exposed to much more SMFs, such those from MRI machines in hospitals. The SMF of the MRI system exceeds the earth's magnetic field by a wide margin. Nowadays, most hospital MRI scanners for normal patients range from 0.5 to 3 Tesla or ten thousand to sixty thousand times stronger than the magnetic field of earth (Hartwig *et al.*, 2009; Sammet, 2016).

The safety of MRI has also been studied in the laboratory at the cellular level. In Hsieh *et al.* (2008) indicated that three tesla SMF suppresses human chondrocyte growth in vitro and influences the regeneration of torn knee cartilage in a pig model.

2.5.2. SMF Impact on Cancer Cells

Cancer remains one of the most common reasons of mortality in the industrialized world. Traditional cancer treatments have a number of problems, including those involving surgery, radiation, chemotherapy, and biological therapies. These disadvantages include the tumor's accessibility, the hazard of operation on essential organs, the dissemination of cancer cells throughout the body, and the absence of tumor-cell-specific selectivity. Immunotherapy has been used to heal tiny tumors because its efficacy diminishes in more advanced cancer stages. Multimodal treatment has improved survival chances (Rex *et al.*, 2006).
A recent study has demonstrated that mechanical and magnetic forces may produce physical interactions that might change the shape, function, and fate of cells (Guilak *et al.*, 2009; Zablotskii *et al.*, 2013). Mechanical stress close to the spheroid surface of cancer cells can impede cell proliferation (Cheng *et al.*, 2009). Due to the presence of iron ions, the idea of magnetic behavior is amplified, distinguishing their paramagnetic characteristics from those of healthy cells. Magnetic radial pressure can transform cancer cells to a paramagnetic condition, reducing cancer development (Guilak *et al.*, 2009;Montel *et al.*, 2011).

2.6. Effect of SMF on Animals

2.6.1. Static magnetic fields' impacts on bone regeneration

In addition to promoting new bone formation around implants and during bone healing, SMF also stimulates the production of new bone during bone healing (Puricelli *et al.*, 2006) by creating a bone hollow in the rat femurs and inserting two titanium screws with stainless steel washers, researchers were able to assess the effect of SMF on bone neoformation. In the group with the magnetized washer, significant bone neoformation was shown 15 or 45 days after implant. The researchers discovered that when bone grafts were used to close the surgical gap, SMF boosted both bone neoformation and the fusion of the grafts (Seyfzadeh *et al.*, 2007; Puricelli *et al.*, 2009; Leesungbok *et al.*, 2013). (Seyfzadeh *et al.*, 2007) analyzed that midshaft osteotomy in dogs accelerated bone healing. In addition, Aydin and Bezer (2011) inserted a magnetic rod into the medulla of a rabbit's femur. They studied that a magnetic field helped heal fractures without affecting bone mineral density (BMD).

2.6.2. Neurons and brain

Static magnetic forces are able to impact a variety of brain processes. In a monolayer dissociated cell culture, McLean *et al.* (2008) discovered that a SMF in the 10 mT range reduced the activity potentials of sensory neurons in the somata of adult mouse dorsal root ganglion neurons (Hernando *et al.*, 2020).

Moreover, it has been indicated that treatment with a transcranial SMF across the supplementary motor area can modify both nearby and far-reaching functionally linked cortical networks, which in turn can change resting-state activity and motor manners (Davoudi *et al.*, 2019). Exposure to SMG can influence the generation of a melatonin and a pineal gland and outcome of functional changes in immature cultured rat hippocampus neurons (Zadeh-Haghighi and Simon, 2022).

2.7. Static magnetic field impact on microorganisms

2.7.1. Impact of Static magnetic field on bacterial cell

Static magnetic field (SMF) is a prevalent evolutionary environmental factor for all living organisms. Contrasting the positive effects are findings of growth delay or inhibition (Yang *et al.*, 2023).

Remarkable linear association between the SMF strength and growth inhibition was originate by Kohno *et al.* (2000) when they exposed three distinct bacterial species (*Escherichia coli, Staphylococcus aureus*, and *Streptococcus mutans*) to 30, 60, 80, and 100 mT SMF and saw a proportionate suppression of the growth cultures correlated with the field strength.

The SMF effect is believed to affect the cell's survival and toxicity by modifications to the Ca+2 metabolism, phospholipid bilayer, and enzymatic activity (which involves the scavenger and anti-oxidant response) (Albuquerque *et al.*, 2016). In 2009 Ji *et al.* (2009) *Escherichia coli* cultures

were shown to be less viable after receiving SMF treatment (of several intensities), which was supported by the possibility of cell stress and damage caused by oxygen free radicals.

Zhang *et al.* (2003) established a dose-response association between the magnetic flux density (between 5 and 9 T SMF) and a rise in the frequency of mutations in the superoxide dismutase (SOD)-deficient *E. coli* strain QC774 (Yang *et al.*, 2023).

Oxidative DNA damage serves an essential function in both the ageing process and illnesses caused by environmental stress. Snoussi and coworkers conducted a series of investigations on the expression of outer membrane proteins in *Salmonella* exposed to 200 mT SMF. Snoussi *et al.* (2016) they reported that *Salmonella hadar* exposed to SMF had a differential expression of a total of 11 proteins with changes of more than twofold. Seven of these altered proteins were up-regulated, while four were down-regulated. The proteomic assessment revealed that SMF-exposed *Salmonella hadar* exhibited differential expression of 35 cytosolic proteins, of which 25 were upregulated and 10 were downregulated. In addition, the overexpression of stress response proteins was detected in *Salmonella hadar* exposed to SMF. Carlioz and Touati demonstrated that intense SMF exposures induced the production of a *soxS::lacZ* fusion gene (Yang *et al.*, 2023).

2.7.1.1. Static magnetic field influence on bacterial growth

Bacteria have been studied to investigate how magnetic fields of varied flux densities affect the growth rate and survivability of Microorganisms (Bajpai *et al.*, 2012). Magneto biological impacts on microorganism growth and the essential methods related to the variability, complexity, and inconsistency of previously reported findings (Binhi and Rubin, 2022) Changes in SMF targets, including as intracellular and extracellular targets, intensity and gradients, bacterial strains, treatment period, and culture conditions (Hunt *et al.*, 2009; Křiklavová *et al.*, 2014). Understanding the biological impact of SMF on microorganisms will contribute noticeably to enhancing our environment, sanitation, and health, as bacteria are extensive in our surroundings and bodies (Li *et al.*, 2022).

SMF is separated into four subtypes based on MF strength: mild (1 mT), moderate (1 millitesla to 1 tesla), high (1–5 tesla), and ultra-strong (>5 tesla) (Rosen, 2003). Existing outcomes on the interaction between moderateintensity SMF and prokaryotic microorganisms were inconclusive, positive, negative, or null (Ayrapetyan, 2015; Santos et al., 2017) caused to a disrupted cell surface, 30 minutes of treatment to 45 mT to 3,500 mT NdFeB magnets greatly decreased the viability of *E.coli* (Ji *et al.*, 2009). Similarly, a 4-hour exposure with 100 mT homogenous SMF created by electromagnets greatly hindered bacterial adhesion and subsequent (Bajpai et al., 2012). It was discovered that Gram-negative E.coli is more sensitive to colony formation results from the disintegration of the cell wall and the liberation of the cytoplasm from the inner membrane SMF than Gram-positive S. epidermidis (Bajpai *et al.*, 2012). In rare instances, the prevention of bacterial expansion by SMF was limited to a specific time frame during experiments; this phenomenon is referred to as the biological window influence (Lebkowska et al., 2018). Potenza et al. (2004) discovered that Escherichia coli grew much quicker in 300 mT of SMF than in GMF when cultured in the modified liquid Luria-Bertani (LB) medium with six g/L glutamic acid and four point five g/L NaCl; however no magnetic impact was detected when the traditional LB medium was utilized. In general, it is believed that the stronger the SMF, the less favorable the environment are for microbial growth and viability (Li et al., 2022).

2.7.1.2. Static magnetic field influence on biochemical characterization

Microorganisms are simple unicellular organisms, making them excellent models for studying basic metabolic reactions to magnetic fields. The patterns of metabolites released by *Streptococcus pyogenes* when exposed to magnetic flux densities ranging from 50 to 500 mT varied markedly (Morrow *et al.*, 2007). SMFs of 250–300 mT elicited the maximal release of the majority of metabolites. Hu *et al.* (2009) showed that compared to *Staphylacoccus aureus*, an SMF of 10 T had a substantial impact on *E.coli*, as shown by alterations in the spectral area of Fourier-transform infrared (FTIR) spectroscopy coupled with cluster processing. Under the magnetic circumstances, the nucleic acid, protein, and fatty acid of *E.coli* underwent changes in composition and structure. She *et al.* (2009) Furthermore, it was shown that 3.46–9.92% of the disorder coils in the protein secondary structures of *E.coli* were converted to a–helices by SMF.

SMF's impact on the fermentation process has been studied using measures of biomass and enzymatic action. In 2004 da Motta *et al.* (2004) compared to SMF-unexposed cultures with those treated with 220 mT SMF substantially revealed in the biomass (g/L) of the *S. cerevisiae* strain by 2.5-fold and the concentration of ethanol by 3.4-fold. In cultures that were magnetized, ethanol yield linked with higher glucose consumption. Invertase is enzyme turn sucrose into the non-crystallizable sugar syrup. Taskin *et al.* (2013) found that spores treated to 5mT SMF had the highest invertase action and biomass concentration.

Several investigations give extensive and fresh insight into the molecular processes behind the apparent physiological anomalies in SMF. They demonstrated that cells of *E.coli* exposed to 250 mT SMF exhibited a smaller colony width. The disruption of carbon source metabolism, particularly long-chain fatty acid and glyoxylate digestion, provided a metabolic underpinning

20

for *E. coli* to adapt to the moderately intense SMF, as demonstrated by transcriptomic and metabolomics investigations (Li *et al.*, 2022).

2.7.1.3. Static magnetic field influence on Antimicrobial susceptibility

Antibiotics are antimicrobial drugs employed for the treatment and prevention of bacterial infections. Antibiotics prevent or eliminate bacterial growth (Cao et al., 2020). The antibacterial activity of gentamicin against Pseudomonas aeruginosa was significantly boosted by applying SMF at concentrations between 0.5 and 2 mT (Zhang et al., 2017a). Samarbaf-Zadeh et al. (2006) discovered that a suspension of cephalothin-resistant bacteria that had been given 16 µg/ml of cephalothin and exposed to electromagnetic field duration had its biomass decreased to less than 1/6 of its initial population Stansell et al. (2001) reported that exposure of E.coli to SMF at 4.5 mT considerably raised antimicrobial resistance. Tagourti et al. (2010) indicated that gentamicin's effectiveness against Salmonella hadar was increased by applied to a 200 mT SMF but that other antibiotics active against Enterobacteriacea, such as penicillin, oxacillin, cephalothin, neomycin, amikacin, tetracycline, erythromycin, spiramycin, and chloramphenicol, did not change the diameter of their zones of inhibition. The antibiotic susceptibility test for Staphylococcus auras was conducted at various exposure periods of 2,4,6,8, and 24 hours, which were calculated based on the mode of action. Staphylococcus aureus was susceptible to gentamycin, rifampin, chloramphenicol, ceftriaxone, and tetracycline, but resistant to metronidazole (Albalawi, 2017).

Exposure to radiofrequency electromagnetic fields significantly modified *Enterococcus faecalis'* antimicrobial susceptibility. While the susceptibility of the bacteria decreased considerably after 6 h of exposure, prolonged exposure time (such as 24 h of exposure) increased the bacteria's sensitivity to antibiotics (Mortazavi *et al.*, 2022).

21

2.8. Urinary tract infections (UTIs)

Gram-negative bacteria are the primary cause of urinary tract infections. Infection of the lower and upper urinary tracts constitutes a UTI (Bankar *et al.*, 2021). Cystitis is the term for a disorder of the lower urinary tract (Nashibi *et al.*, 2022). For a more severe progression of the infection, it can extend to the kidneys, which is then considered an upper UTI, and the patient is diagnosed with pyelonephritis (Wagenlehner *et al.*, 2020). Invading bacteria typically originate from the gastrointestinal tract, but they can also be acquired in hospital settings (Organization, 2002). Hospital-acquired UTIs are typically caused by catheters and are therefore classified as nosocomial infection is also defined as an infection that develops 48 hours after a patient's admission and was not visible at the time of admission (Dasgupta *et al.*, 2015). This is comparable to a UTI acquired within the community (Kang *et al.*, 2018).

The incidence of urinary tract infections varies by gender and by age group (Rowe and Juthani-Mehta, 2013). There is a higher occurrence of UTIs in women, with additional risk factors including pregnancy, sexual activity, and age contributing to the frequency of infection; older females (over 65) have twice the incidence rate of UTIs as the rest of the female population (Matuszkiewicz-Rowińska *et al.*, 2015; Medina and Castillo-Pino, 2019) However, this is to be anticipated, as UTIs are more prevalent in the elderly regardless of gender (Rowe and Juthani-Mehta, 2013).

Escherichia coli, *Klebsella pneumoniae*, and Proteus mirabilis are the pathogens that induce urinary tract infections (Erdem *et al.*, 2018). However, there are distinctions between the prevalence of species isolated from hospitals versus community UTIs. *E.coli* remains the most prevalent pathogen in both situations but appears marginally less frequently in hospital cases

(Mancini et al., 2020). Klebsiella species, Enterococcus faecalis, Proteus mirabilis, and Pseudomonas aeruginosa have been found in both community and hospital samples, and although Enterococcus faecalis and Pseudomonas aeruginosa have been recognized in hospital settings, they are uncommon (Mancini et al., 2020).

2.9. Uropathogenic E. coli

Escherichia coli belongs to the family *Enterobacteriacea*, often a harmless commensal of the gut system (Mare *et al.*, 2021). However, some *E. coli* clones have developed the capability to create virulence factors, giving them the potential to spread a number of illnesses to both animals and humans (Croxen *et al.*, 2013; Tanabe *et al.*, 2022).

Two major kinds of pathogenic *E.coli* exist: intestinal strain or diarrhea genic (DEC) and extra intestinal pathogenic *E.coli* (ExPEC), which cause diarrhea and extra intestinal illnesses, newborn meningitis, sepsis, and urinary tract infections (UTIs), respectively (Kaper *et al.*, 2004; Croxen *et al.*, 2013; Santos *et al.*, 2020).

Isolates *of E.coli* causing uropathogenic *E.coli* (UPEC) is the most significant cause of this infection in hospitalized and outpatient individuals globally (Kikuchi *et al.*, 2022).

Consistently, phylogenomic investigations have indicated that the *Escherichia coli* species is highly complex and organized into eight major phylogroups: A, B1, B2, C, D, E, F, as well as the newly-described G. Phylogroup B2 has been ascribed to the vast majority of UPEC isolates, as well as isolates found in other *E. coli* phylogroups (Campos *et al.*, 2020; Flament-Simon *et al.*, 2020).

Uropathogenic *E.coli* is a common cause of cystitis, pyelonephritis, and other UTIs, as well as infectious complications that may lead to acute renal failure in otherwise healthy people and kidney transplant patients.

23

Uropathogenic *E.coli* is the most common bacterium responsible for both mild and severe cases of UTI (Bueris *et al.*, 2007; Kotloff *et al.*, 2013; Gomes *et al.*, 2016; Lima *et al.*, 2019; Duong *et al.*, 2020). Any component of the genitourinary system, such as the urethra, bladder, ureter, renal pelvis, or renal parenchyma, might be affected by this, which could have an effect on people of all ages and demographics (Dias *et al.*, 2016).

2.9.1. UPEC virulence factor

Essential *Escherichia coli* virulence factors can be roughly categorized into two classes: released virulence factor and bacterial cell surface. Most bacterial cell surface virulence factors are fimbriae; particularly kind 1fimbriae and P fimbriae. These fimbriae assist in attachment to the surface of the host cell, tissue invasion (which is crucial to the pathophysiology of UPEC that cause UTIs), biofilm formation, and cytokine production. Furthermore, microbial cell surface virulence factors involve the flagellum, capsular lipopolysaccharide, and outer membrane proteins. Siderophores and hemolysin released virulence factors (Emody *et al.*, 2003; Sarowska *et al.*, 2019). These virulence factors are essential for microbial survival in the urinary tract despite the efficiency of the human immune system (Vagarali *et al.*, 2008).

Virulence factor	Major function				
Capsule	phagocytosis resistance				
Cellulose	Forming a biofilm				
Curli fimbrico	Adhesion, development of biofilm,				
Cummonae	and invasion				
Cutatovic Necrotizing Factor 1	Host cell adhesion, invasion, and				
Cytotoxic Necrotizing Factor 1	apoptosis				
Dr fimbriae	Cell invasion				
F1C fimbriae	Unknown				
Flagalla	E. coli adhesion in the urinary				
	system				
Haemolysin	Damage to tissue and invasion				

Table (2-1)	The UPEC's	virulence factors	(Terlizzi et	al., 2017)
-------------	------------	-------------------	--------------	------------

Iron and zinc acquisition	Nutrition
LPS	Immune response activator
P fimbriae	adhesion with renal epithelial cells
Secrete auto transporter toxins	Tissue damage
Type 1 fimbriae	bladder epithelial cell adhesion

2.9.2. Antibiotics Susceptibility of Uropathogenic E. coli

A significant clinical issue is the ongoing rise of clinical bacterial strains' resistance to antimicrobial agents (Adamus-Bialek *et al.*, 2013). The rise of several antibiotic resistance mechanisms among prevalent human pathogenic *Enterobacteriacea* members raises the alarm and reduces the range of possible therapeutic choices (Boucher *et al.*, 2009). Nonetheless, multidrug-resistant *E.coli* bacteria have also been discovered globally (Pitout and DeVinney, 2017). They can resist multiple types of antibiotics and are referred to as multi-antibiotic-resistant (Bennett, 2008). The National Committee for Medical Laboratory Standards M7-A6 broth microdilution method was used to determine *E.coli* susceptibilities to ampicillin (Sigma Chemical Co., St Louis, MO), sulphamethoxazole/trimethoprim SMX/TMP (Sigma Chemical Co.), nitrofurantoin (Procter & Gamble Inc., Cincinnati, OH), ciprofloxacin (Bayer Inc., Toronto, ON), and levofloxacin (Ortho-McNe) (Zhanel *et al.*, 2006).

2.9.2.1. Extended-spectrum β-lactamases (ESBLs)

Escherichia coli are the most widespread bacterium responsible for urinary tract infections (UTIs). UTIs are commonly controlled with cephalosporins, particularly third-generation cephalosporins (Gallini *et al.*, 2010). These bacteria have changed recently, and through gene exchange, they have developed resistance to several drugs (Lee *et al.*, 2010). One of the methods of resistance is the creation of enzymes such as beta-lactamase and extended-spectrum β - lactamases (ESBLs) that may hydrolyze the beta-lactam ring and inactivate drugs possessing this structural feature (Abbas *et al.*, 2022).

Extended-spectrum β -lactamases (ESBLs) were initially discovered in the 1980s and identified in *Klebsieilla sp, E.coli, Pseudomonas aeruginosa, Serratia marcescens* and other gram-negative bacilli (Morris, 2003; Kiratisin *et al.*, 2008; Cheng *et al.*, 2008).ESBLs can also degrade third- and fourth-generation cephalosporins and monobactams. Beta-Lactamase inhibitors are effective against ESBL-producing strains clavulanic acid, sulbactam, and tazobactam (Bradford, 2001; Giraud-Morin and Fosse, 2003; Pitout *et al.*, 2007). Major reports of ESBL generation are associated with the plasmid genes *bla CTX-M, bla TEM,* and *bla SHV*, which can also harbor other resistance genes (Azap *et al.*, 2010; Gray, 2022).

2.9.2.2. Multidrug Resistance and Uropathogenic Escherichia coli

Alqasim *et al.* (2018) found that all extended-spectrum-beta-lactamase (ESBL) positive *E.coli* specimens are extremely resistant to the first-line antibiotics used to treat urinary tract infections. ESBL-producing UPEC is distinguished by its high level of cross-resistance to many antibiotics., posing a significant clinical problem (Halaji *et al.*, 2020). UTI is related to a higher proportion of ESBL-producing UPEC. In addition, Ali *et al.* (2016) found that most of these bacterial separates are fluoroquinolone- and multi-drug-resistant (MDR), the average degree of UPEC resistance to Various kinds of antibiotics.

According to the research by Reygaert (2018), the synthesis of β -lactamase enzymes by UPEC, which might hydrolyze -lactam ring structure and cause the ring to open, is what confers antibiotic drug resistance (ADR) to β -lactam antibiotics. Outcome, β -lactam medications, including cephalosporin, penicillin, carbapenems, and monobactams, cannot bind to the particular penicillin-binding proteins (PBP). The genes (*bla* genes) frequently found on the plasmid of bacteria, these genes are in charge of coding different types of β –lactamase (Kot, 2019).Extended Spectrum -lactamase (ESBL) generated by *E.coli* provides resistance to extended-spectrum antimicrobial agents, involving penicillin and third- and fourth-generation Cephalosporins (Padmini *et al.*, 2017).

The three categories of ESBL enzymes are ESBLA, ESBLM, and ESBL CARBA. ESBLA contains the most commonly discovered $CTX_{.M}$ enzymes, as well as *SHV* and *TEM* enzymes that can be destroyed by clavulanic acid (Giske *et al.*, 2009; Kot, 2019).

2.9.3. Molecular Characterization of Uropathogenic E. coli

The biological sciences have seen a revolution in recent decades thanks to the power of molecular biology. A new chapter in the characterization of many microorganisms has been opened using molecular techniques. Since late genetic methods have been created to genotype 1980. numerous microorganisms (Katsanis and Katsanis, 2013). Due to their high resolution, these techniques are now commonly utilized for bacterial identification. Detecting any bacteria's genetic makeup by a specific genotyping method can be as unique as a fingerprint (Tshikhudo et al., 2013). The most revolutionary technology is polymerase chain reaction (PCR), which can be utilized efficiently by targeting characteristic deoxyribonucleic acid sequences. PCR is a quick in vitro technique for the enzymatic amplification of specific DNA sequences that raise the number of target sequence copies and the sensitivity of DNA sequence detection. It has also been suggested to build multiplex PCR to recognize common infections, similar to the PCR techniques created to simultaneously detect enterobacteriacea and clinically significant bacteria (Chen *et al.*, 2022). Species-specific recognition of *E. coli* by PCR techniques based on the 16SrRNA genes or their respective functional genes has been discussed previously (Clifford et al., 2012; Franco-Duarte et al., 2019).

UPEC is also related to a high prevalence of the extended-spectrum lactamase (ESBL) gene (Pitout *et al.*, 2005). ESBLs include numerous

27

plasmid-mediated variants, including TEM, OXA, and SHV (Nicolas-Chanoine et al., 2008). Since 2000, a brand-new class of ESBLs known as CTX_{-M} (active on cefotaxime, initially identified in Munich) has appeared (Peirano and Pitout, 2010). Since then, CTX_{-M} lactamases have been the most prevalent ESBL type globally (Cantón and Coque, 2006).CTX.M-15 is now the most pervasive CTX_{M} genotype in the CTX_{M} family (Cantón and Coque, 2006; Peirano and Pitout, 2010). This group of ESBLs has been related to a widespread pattern of antimicrobial resistance to numerous drugs, including like penicillin's, Cephalosporins, drugs monobactams, β-lactam and carbapenems (Rogers et al., 2011; Accogli et al., 2014). Moreover, CTX_M E.coli producing bacteria are often linked with resistance to other wide antibiotic families, involve aminoglycosides, and fluoroquinolone (Rogers et al., 2011). Since it can restrict the therapeutic options used to treat common microbial illnesses like UTIs, the rising prevalence of antimicrobial resistance in UPEC raises serious concerns and underlines the potential of the formation of pan drug resistance in UPEC (Malekzadegan et al., 2018; Alqasim et al., 2018).

Chapter Three MATERIALS AND METHODS

3. MATERIALS AND METHODS

3.1. Materials

The laboratory apparatus and equipment, culture medium, and chemical components, as well as the kits and supplies required for molecular characterizations have been shown in tables (3-1), (3-2), and (3-3), respectively.

Table (3-1) Equipment and instruments used in the study.

Instruments and Equipment	Supplier/Origin	Country
Incubator	Incubator Bc-J800	(China)
Autoclave	Kay Company	(India)
Mini Vortex	Lab genius	(UK)
Hood Safety Cabinet	Pars Azma CO	(Iran)
VITEK system, glass, test tubes, Gram-negative ID and AST Card	BioMerieux	(USA)
Racks	Solar bio life sciences	(China)
Nano Drop (2000)Spectrophotometer	Thermo Fisher Scientific	(USA)
PCR Thermal Cycler Machine	Applied Bio systems™ Veriti ®	(USA)
Refrigerator	Hisense	(China)
Gel Electrophoresis System	Padideh Nogen Pars	(Iran)
UV Trans illuminator	UVP	(UK)
Thermal Shaker Incubator	D.S. Scientific Solutions	India
Refrigerated Bench Top Centrifuge	Nuve	Turkey
Spectrophotometer	mmk	(China)
Eppendrof tubes, Tips	Sterelin Ltd	(UK)
Microwave	Hisense	(China)
Teslometer	PHYWE	(Germany)
Balance	Shimadzu	(India)
Disposable glass and plastic tube	BIOZEK	Netherland
Disposable Petri dishes	Sterilin	(UK)
Sterilized cotton swabs, Urine container	Indiamart	(India)
Rack PCR (0.2ml-1.5ml)	Solarbio life sciences	China
Microcentrifuge Tube, 0.2- & 1.5-mL Polypropylene	Bio Basic	Canada
Adjustable Volume Micropipette	Philip Harris	UK
Micropipette Tips	Accumax	Germany

Table (3-2) Culturing media and chemical substances employed during the investigation.

Culture media	Supplier/Origin	Country
MacConkey agar	Lab M Neogen company	(UK)
Nutrient broth	Lab M Neogen company	(UK)
LB broth (Lauria Bertani broth)	Lab M Neogen company	(UK)
Brain Heart Infusion Agar	Lab M Neogen company	(UK)

Table (3-3) molecular study used commercial kits and materials.

Molecular kits and materials	Supplier/ Origin	Country
DNA Extraction kit	Jena Bioscience	Germany
Primers Synthesis	Integrated DNA	USA
, ,	Technologies (IDT)	
6X DNA Loading Dye	Norgen Biotek	Canada
DNA Ladder	Norgen Biotek	Canada
DNase Free Water	Norgen Biotek	Canada
Agarose Powder	Bio Basic	Canada
TDE Tris Doroto EDTA Duffor	Dionaar	South
TDE THS-BOIAGE EDTA Buller	Biolicei	Korea
PCR Master mix	Promega	USA

3.2. Methods

3.2.1. Culture Media Preparation

The culture medium was prepared according to the manufacturer's instructions and autoclaved for 15 minutes at 121°C.

3.2.1.1. MacConkey Agar Medium

This medium was prepared by suspending 51.5 grams of the medium in one liter of distilled water, heating with frequent agitation and boiling for one minute to dissolve the medium sufficiently, and then autoclaving at 121 $^{\circ}$ C. for fifteen minutes.

3.2.1.2. LB Broth (Luria-Bertani)

LB (Luria-Bertani) Broth is used in molecular genetic studies in a laboratory setting. This broth is nutritionally rich, formulated for the isolation of pure recombinant strains. It was prepared by dissolving 20 g of the medium in liter of distilled water, mixed thoroughly; the pH was adjusted to 7.2 then sterilized by autoclave at 121°C for 15 minutes.

3.2.1.3. Nutrient broth

This medium was prepared by suspending 25 g of the medium in 1000 mL of distilled water, heated with frequent agitation and boiling for one minute to dissolve the medium completely, and then autoclaved at 121°C for 15 minutes.

3.2.1.4. Brain Heart Infusion Agar

The enrichment medium was prepared by putting 49.0 g of Brain Heart Infusion agar in one liter of distilled water and autoclaving it at 121°C for fifteen minutes.

3.2.2. Collection of samples

Seventy five clinical samples were collected aseptically (Garcia, 2010) from patients with symptomatic infections attended to different hospitals in Erbil City (Erbil, Rizgary hospital, Rapareen Teaching Hospitals) during the period from October 2021 to February 2022, then *E coli* bacteria identified by method (VITEK bioMerieux, using VITEK test Testing device: 00014EED3FB) in Rizgary Hospital and Hawler teaching hospital. Specimens were urine samples but one of the samples was taken in Awamedica pharmaceutical Company as reference bacteria samples of E coli National Collection of Industrial, Food and Marine Bacteria (NCIMB 50125). The specimens were immediately streaked onto Blood agar, MacConkey agar plates, and inoculated into Nutrient Broth, after that incubated for 24 hours in an aerobic environment at 37°C.

3.2.3. Phenotypic identification of isolates

Phenotypic identification of the isolates was performed from pure colonies depending on colony morphological, and biochemical characteristics (Forbes *et al.*, 2007) and confirmatory VITEK 2 system using (ID) GN cards.

3.2.3.1. Colony Morphology

Initial identification depended on the colonies' morphological characteristics (colony size, shape, edge, color, odor, and texture)

3.2.3.2. Using the VITEK 2 System for isolate detection

The detection of isolates was also accomplished by biochemical methods in the VITEK system (VITEK bioMerieux, Testing instrument: 00014EED3FB (Pincus, 2006). Identification using the VITEK 2 system was conducted using ID-GN cards in accordance with the manufacturer's guidelines. The ID-GN cards contain biochemical tests, involving sugar assimilation, fermentation, carbon source utilization, decarboxylase tests, and enzymatic activities. The cards were inoculated with a (0.5) McFarland standard suspension of the organism prepared from an (18-20 hours) MacConkey agar plate by means of a vacuum apparatus. The cards were then mechanically sealed before being manually inserted into the VITEK 2 readerinoculation module.

Bacterial isolates were preserved on brain heart infusion agar slants at 4°C for subsequent testing. They were also maintained longer in culture supplemented with 15% glycerol and stored at ⁻ 20°C (Oskouei *et al.*, 2010).

3.2.3.3. Antibiotic Susceptibility Testing

Depending on the manufacturer's recommendations, susceptibility tests were performed on the VITEK 2 system using AST-N417 cards. Among the medications on the AST-N417 (bioMerieux) card were piperacillin (PIP), ceftazidime (CAZ), aztreonam (ATM), levofloxacin (LEV), cefepime (FEP), trimethoprim/sulfamethoxazole(SXT),ciprofloxacin(CIP),tazobactam/piperaci lli-n ((TIG).

The cards were filled with 10^8 CFU/ml of a bacteria inoculum (according to 0.5McFarland) suspension then sealed and read. The antimicrobial susceptibility cards are processed automatically by the VITEK 2 system until the MICs are acquired. The findings were evaluated using VITEK 2 version (08.01) software, and the final results were automatically obtained (Eger *et al.*, 2022).

3.2.4. Preparation of magnetic field

The device used to prepare a static magnetic field applied in our project was a locally made device, where magnet models of the car starter machine were used. The apparatus was checked by a gauss-meter (Gauss meter, Germany) in the Physics Department College of education at the University of Salahaddin in Erbil, Iraq. During application, the system can be adjusted to create different intensities (0.04, 0.08, 0.12 and 0.16 T)) of the magnetic field by changing the used magnetic number or size compared with the standard.

3.2.4.1. Exposed bacteria to the different static magnetic field

The bacterial cultures were grown on a MacConkey agar medium and incubated at 37°C for 24 hours before being harvested by a loop and inoculated in 10 mL of nutrient broth. Different magnetic fields (0.04, 0.08, 0.12 and 0.16 T) were applied to each bacterial culture. The magnetic fields

33

were recorded using a Tesla meter (PHYWE (Germany)) digital (Al-Khaza'leh and Al-fawwaz, 2015).

In this study, (1) mL of bacterial suspension was added to four tubes containing (9) ml of nutrient broth each. Magnetic fields of (0.04, 0.08, 0.12 and 0.16T) were applied to four groups of tubes, respectively. The fifth group served as a negative control, though (no magnetic field was subjected). For 24 hours, all tubes were incubated at 37°C. Through the use of a spectrophotometer, the optical density at 620 nm was measured to assess the impact of various magnetic fields on growth rate (PHYWE (Germany)) (Kamel *et al.*, 2014).

Examination of the impact of various magnetic field forces on growth rate by measuring optical density with the McFarland Turbidity Standards (0.5) method (Ahghari *et al.*, 2020)

3.2.5. Bacterial Maintenance and Storage (storing)

Bacterial isolates were preserved on brain heart infusion agar slants at 4°C for subsequent testing. They were also maintained longer in culture supplemented with 15% glycerol and stored at -20°C (Oskouei *et al.*, 2010)

3.2.6. Molecular analysis of the isolates

For more confirmation of the identity of the isolates, all isolates of *E. coli* were subjected to molecular characterization by detecting specific gene1*6SrRNA* for identification of bacterial pathogen (*E. coli*), and detection of *TEM*, and *CTX*_{*M*-1}, *SHV* genes, respectively, that were responsible for the ESBL genes by using PCR technique. Symbols of the Group represent the number of samples, (0) means unexposed (negative control), and 1, 2, 3, and 4 represent exposed samples to different powers of the magnetics (0.04, 0.08, 0.12 and 0.16 T) respectively.

3.2.6.1. Genomic DNA Extraction

Genomic DNA was extracted from bacterial strains using the DNA extraction kit (Jena Bioscience, Germany) according to the manufacturer's specifications as the followings:

- 1. One milliliter of cultivated cells was transferred to a 1.5-ml micro tube and centrifuged at $15000 \times g$ (11573 rpm) for one minute to harvest the cells; the supernatant was then discarded. The pellet (bacterial colony) was suspended in 300 µl of cell lysis solution for (Cell lysis for Gram-negative bacteria- sample preparation).
- One point Five microliters of *RNase* a solution was added and inverted to mix. The sample was then incubated at 37 °C for 15 to 30 minutes then cooled on ice for 1 minute for (*RNase* treatment).
- 3. One hundred microliters of protein precipitation solution were added, followed by vigorous vortexing for 20-30 seconds and centrifugation at $15000 \times g$ (11573 rpm) for five minutes for (Protein Precipitation).
- 4. Transferred the supernatant to a sterilize 1.5 mL micro tube holding 300 μL of isopropanol >99%. The sample was then gently shaken for one minute. For one minute, the specimen was centrifuged at (11573 rpm) (DNA should be visible as a small white pellet).
- 5. The supernatant was discarded and the tube was drained briefly on absorbent paper. Next 500µl of washing buffer was added, followed by numerous inversions of the tube to wash the DNA pellet. Following that, they were centrifuged for one minute at 15000 ×g (11573 rpm). The ethanol was properly discarded. Finally, the sample was air-dried at room temperature for ten to fifteen minutes for (DNA Precipitation).
- 6. The dried DNA pellet was rehydrated using 50 to 100 μl of DNA hydration solution. The DNA was hydrated by incubating it at 65°C for 60 minutes, and it was then stored at -20°C or -80°C for a long period until it was utilized for PCR amplification for (DNA hydration).

3.2.6.2. Determination of DNA Concentration

A Nano drop spectrophotometer was utilized to measure the concentration and purity of extracted DNA (Thermo Fisher Scientific, USA) at absorbance (260/280 nm), with a ratio of 1.7 to 1.9 for DNA purity and concentration, and in accordance with (Desjardins and Conklin, 2010)

3.2.6.3. Primers and PCR Amplification

3.2.6.3.1. Primer Preparations

Table (3-4) represented all primers of Uropathogenic *E. coli* were used in this study. The primers were provided by Integrated DNA Technologies (IDT) USA in lyophilized form. Since the primer of the *16SrRNA* gene was used to detect the genome of Uropathogenic *E. coli*, while the other three primers were used to detect genes ($TEM_1 CTX_{M-1}$, SHV).

Target genes	Sequence (5'→3')		Amplicon (bp)	Reference	
165mDNA	F	AGAGTTTGATCMTGGCTCAG	1242	(I alrahmi at al. 2020)	
TOSTRINA	R	CGGTTACCTTGTTACGACTT	1545	(Laksiiiii <i>ei al.</i> , 2020)	
CHV	F	TCGGCCTTCACTCAAGGAATG	800	(Wes et al. 2020)	
SHV	R	TCCCGCAGATAAATCACCA	000		
TEM	F	AGGAAGAGTATGATTCAACA	525	(wu <i>et al.</i> , 2020)	
IEM_1	R	CTCGTCGTTTGGTATGGC	333		
CTX- _{M-1}	F	CCGTTTCCGCTATTACAAACCGTTG	202	(7hong at al 2014)	
	R	GGCCCATGGTTAAAAAATCACTGC	695	(Znang <i>et al.</i> , 2014)	

Table (3-4) primers utilized in this study.

3.2.6.3.2. Detection of 16SrRNA gene of E. coli isolates by PCR

The particular primer pairs of the *16SrRNA* gene that were employed to amplify this gene as illustrated in Table (3-5), and the PCR condition of the amplified *16SrRNA* gene was carried out by initial denaturation at 96°C for four minutes, followed by 35 cycles of denaturation at 94°C for thirty seconds, annealing at 57°C for thirty seconds, extension at 72°C for one

minute, and a final extension at 72°C for seven minutes. The *16SrRNA* gene DNA PCR amplicons were estimated and determined by using 1% agarose gel electrophoresis, and 25 μ L of the PCR master mix reaction was prepared in PCR micro tube by adding 12.5 μ L of Master Mix (Promega), 8.5 μ L PCR deionized distilled water, 1 μ L from both of them forward and reverse primers and two μ L of the bacterial genomic DNA extraction from the positive sample as a template. The PCR master reactions of each sample were mixed and centrifuged for a few seconds to spin down the mixture (Lakshmi *et al.*, 2020).

	Primer's c	letail	
Functional category	Primer Sequence (5'-3')	Ampli con size (bp)	PCR Cycling program
16SrRNA	F: AGAGTTTGATCMTGGCTCAG R: CGGTTACCTTGTTACGACTT	1343	96°C-4min: 1cycle 94°C -30s :35 cycles 57°C -30s :35 cycles 72 °C -1min;35cycles 72°C -7 min: 1cycle

Table (3-5) primers of 16SrRNA gene with PCR program of E. coli.

3.2.6.3.3. Detection of *TEM*₁ gene of *E coli* isolates by PCR

The particular primer pairs of the *TEM* gene that were employed to amplify this gene as illustrated in Table (3-6) and the PCR condition of the amplified *TEM* gene was carried out by initial denaturation at 96°C for four minutes, followed by 35 cycles of denaturation at 94°C for thirty seconds, annealing at 57°C for thirty seconds, extension at 72°C for one minute, and a final extension at 72°C for seven minutes. The *TEM*₁ gene DNA PCR amplicons were estimated and determined by using 2% agarose gel electrophoresis, and 25 µL of the PCR master mix reaction was prepared in a PCR micro tube by adding 14.0 µL of Master Mix (Promega), 7.0 µL PCR deionized distilled water, 1 µL from both of them forward and reverse primers and two µL of the bacterial genomic DNA extraction from the positive sample as a template as shown in the table. The PCR master reactions of each sample were mixed and centrifuged for a few seconds to spin down the mixture.

Table (3-6) primers of *TEM*₁ gene with PCR program of *E. coli*.

	Primer'	s detail	
Functional category	Primer Sequence (5′ – 3′)	Amplic on size (bp)	PCR Cycling program
<i>TEM</i> ¹ gene	F: AGGAAGAGTATGATTCAACA R: CTCGTCGTTTGGTATGGC	535	94°C-4min: 1cycle 94°C-30s :35cycles 57°C-30s :35 cycles 72 °C-1min; 35 cycles 72°C- 7min: 1cycle

3.2.6.3.4. Detection of CTX-M₋₁ gene of E. coli isolates by PCR

The particular primer pairs of the *CTX-M*₋₁ gene that were employed to amplify this gene as illustrated in Table (3-7), and the PCR condition of amplified *CTX*-_{*M*-1} gene was carried out by initial denaturation at 96°C for four minutes, followed by 35 cycles of denaturation at 94°C for thirty seconds, annealing at 57°C for thirty seconds, extension at 72°C for one minute, and a final extension at 72°C for seven minutes. The *CTX*-_{*M*-1} gene DNA PCR amplicons were estimated and determined by using 2% agarose gel electrophoresis, and 25 µL of the PCR master mix reaction was prepared in PCR micro tube by adding 14.5 µL of Master Mix (Promega), 7.0 µL PCR deionized distilled water, 0.75 µL from both of them forward and reverse primers and 2µL of the bacterial genomic DNA extraction from the positive sample as a template as shown in the table. The PCR master reactions of each sample were mixed and centrifuged for a few seconds to spin down the mixture. Table (3-7) primers of CTX_{-M-1} gene with PCR program of *E.coli* used in this study

	Primer's deta	il	
Functional category	Primer Sequence (5' – 3')	Ampl icon size (bp)	PCR Cycling program
CTX-M. ₁ gene	F: CCGTTTCCGCTATTACAAACCGTTG R: GGCCCATGGTTAAAAAATCACTGC	893	95°C-4min: 1cycle 94°C -30s :35 cycles 57°C -30s :35 cycles 72 °C _1min; 35 cycles 72°C -7 min : 1cycle

3.2.6.3.5. Detection of SHV gene of E coli isolates by PCR

The particular primer pairs of the *SHV* gene that were employed to amplify this gene as illustrated in Table (3-8) and the PCR condition of the amplified *SHV* gene was carried out by initial denaturation at 96°C for four minutes, followed by 35 cycles of denaturation at 94°C for thirty seconds, annealing at 57°C for thirty seconds, extension at 72°C for one minute, and a final extension at 72°C for seven minutes. The *SHV* gene DNA PCR amplicons were estimated and determined by using 2% agarose gel electrophoresis, and 25 μ L of the PCR master mix reaction was prepared in a PCR micro tube by adding 14.0 μ L of Master Mix (Promega), 7.0 μ L PCR deionized distilled water, 1 μ L from both of them forward and reverse primers and two μ L of the bacterial genomic DNA extraction from the positive sample as a template as shown in the table. The PCR master reactions of each sample were mixed and centrifuged for a few seconds to spin down the mixture.

	Primer's d	etail	
Functional category	Primer Sequence (5′ – 3′)	Ampl icon size (bp)	Cycling program
SHV gene	F: TCGGCCTTCACTCAAGGAATG R: TCCCGCAGATAAATCACCA	800	95°C-4min: 1cycle 94°C -30s :35 cycles 54°C -30s :35 cycles 72 °C _1min; 35 cycles 72°C -7 min: 1cycle

Table (3-8) primers of *SHV* gene with PCR program of *E.coli* used in this study.

3.2.6.3.6. Detection of Amplified Products by Agarose Gel Electrophoresis

The PCR products were evaluated using electrophoresis on a 2% agarose gel dyed with a Safe dye. Two grams of agarose powder was dissolved in 100 mL of Tris-EDTA-Borate Buffer (1× TBE)in the microwave, allowed to cool to 50°C, and then 5 μ L/100 mL of Safe Dye was added (Russell and Sambrook, 2001). The comb was fixed to one end of the tray to create wells for DNA sample loading. The agarose was poured carefully into the tray and allowed to solidify for 30 minutes at room temperature. The comb was then taken from the tray with care. The tray was positioned in an electrophoresis chamber with 1× TBE buffer that covered the gel's surface. Ten milliliters of amplified DNA product was transferred into each well of the agarose gel.

When the power source was turned on and the gel chamber was closed, it operated at 5 Volts/cm. A power source of 45 volts was used for 15 minutes to improve resolution, causing the DNA to leave the wells and migrate 0.5 cm in the direction of the positive electrode. After that, the voltage was raised to 100–135 volts and the electrophoresis was given enough time to complete. The gel was captured using a digital camera after DNA bands were seen using a UV Trans-illuminator (UVP). As a molecular marker, a 100 bp DNA ladder was utilized as molecular marker.

3.2.7. Data analysis

The PCR product of the TEM_1 and CTX_{M-1} genes were sequenced at Macrogene using the Sanger method by South Korean DNA sequence analyzers. Finch TV chromatogram viewer software was used to convert the chromatograms to FASTA format. The ABI file's DNA sequences were manually edited using Bio Edit V7.0.5.NCBI'sBLAST (Basic local alignment search tool) was used to assess the results of sequence editing in order to identify the closest species' homology. The phylogenetic tree was created using the maximum likelihood technique, calculations using Bootstrap with 1000 repetitions, using the Molecular evolutionary genetic analysis (MEGA 11) program (Kumar *et al.*, 2018).

3.2.8. Statistical Analysis

The data are reported as the mean of three independent replicates (Mean \pm SEM), and Prism9.0software was used to perform a one-way analysis of variance (ANOVA) for statistical analysis (Graph Pad Software Inc., La Jolla, CA, USA). If the p-value was less than 0.05, the data were considered statistically significant.





(A)

(B)



(C)

Figure (3-1) (A):Agarose gel electrophoresis apparatus (B)Thermal cycle PCR machine (C): locally made device static magnetic field

Chapter Four RESULTS

4. **RESULTS**

4.1. Isolation and Identification of E. coli

The clinical specimens of urine samples sources, 25 isolates were identified as *E. coli* by using VITEK test method (VITEK bioMerieux), Testing instrument (00014EED3FB) in Rizgary,Hawler Teaching Hospital .The isolates were originally determined to be members of *E. coli* by the pink colony color (lactose fermenting) on the MacConkey agar as figure (4-1). When Gram stain was used to stain the bacteria under a microscope, rod-shaped, gram-negative bacteria were seen.



Figure (4-1) Showing Colony morphology of *E. coli* (A) unexposed to magnetic fields (control), (B) treated to magnetic field 0.04T, (C) treated to 0.08T, (D) treated to 0.12T and (E) treated to 0.16T.

4.2. Exposing Uropathogenic *E. coli* to Different Static Magnetic Field

Magnetic field influence on growth and antibiotic susceptibility of bacteria was confirmed. This to observe the exposure influence of different magnetic fields that is; (0.04, 0.08, 0.12 and 0.16T) for 24 hours on the rate of growth and antibiotic sensitivity of *E.coli*. The bacteria were isolated from the clinical case and identified. Using system acknowledged Vitek 2 system. The susceptibility of the antibiotic of *E.coli* measured. The results exhibited an important logarithm reduction in the number of *E. coli* exposed with different magnetic field the sensitivity of bacteria altered and increase its resistance to the same antibiotic at a long term exposure of 24 hours. Some biochemical tests results showed positive effects of magnetic fields on the biochemical properties.

The results of this investigation relate to the induced changes in the structure and characteristic behavior of *E.coli* caused by exposure to magnetic fields with powers of (0.04, 0.08, 0.12 and 0.16 T) for 24 hours. These findings might be significant for assessing the advantages and risks of exposure to low-level magnetic fields.

4.3. Growth Characteristics Curve

Table (4-1) figure (4-2) represents the evolution of a bacterial strain's absorbance as a function of the time it was exposed to a magnetic field. This figure makes it clear that the exposure times are 24 hours. Reduced absorbance, decrease in the number of cells indicates a circumstance where the bacteria are inhibited

Table (4-1) Effect of different magnetic field forces on growth rate of

	OD 620 nm at 24 hours					
S.NO	Control		Magn	etic force		
	Control	0.04T	0.08T	0.12T	0.16T	
1.	0.992	0.966	0.843	0.825	0.820	
2.	0.990	0.899	0.889	0.855	0.825	
3.	0.995	0.986	0.973	0.933	0.923	
4.	1.028	1.020	0.930	0.922	0.910	
5.	1.030	1.022	1.018	0.995	0.984	
6.	1.022	0.983	0.932	0.922	0.872	
7.	0.937	0.901	0.897	0.791	0.775	
8.	0.970	0.930	0.908	0.889	0.859	
9.	0.983	0.898	0.880	0.858	0.798	
10.	1.017	0.963	0.937	0.848	0.826	
11.	0.400	0.316	0.305	0.206	0.186	
12.	0.948	0.935	0.918	0.907	0.902	
13.	1.055	0.961	0.952	0.899	0.855	
14.	0.992	0.966	0.843	0.825	0.820	
15.	0.990	0.899	0.889	0.855	0.825	
16.	0.995	0.986	0.973	0.933	0.923	
17.	1.028	1.020	0.930	0.922	0.910	
18.	1.030	1.022	1.018	0.995	0.984	
19.	1.022	0.983	0.932	0.922	0.872	
20.	0.937	0.901	0.897	0.791	0.775	
21.	0.970	0.930	0.908	0.889	0.859	
22.	0.983	0.898	0.880	0.858	0.798	
23.	1.017	0.963	0.937	0.848	0.826	
24.	0.984	0.900	0.889	0.820	0.790	
25.	1.055	0.961	0.952	0.899	0.855	
p-value		NS*	0.0005**	0.0000	0.0000	

Uropathogenic E. coli

* S.NO: number of sample isolates, OD=optical density, nm=nanometer



Figure (4-2) statistical analysis showing significant differences between optical density (620 nm) of *E.coli* unexposed (Negative control) vs. optical density (620 nm) *E.coli* under magnetic power (0.04, 0.08, 0.12 and 0.16T) respectively

4.4. Investigation of Magnetic Field on Bacteria Antibiotic Sensitivity

Table (4-2) displayed the antibiotic susceptibility test at various exposure levels (0.04, 0.08, 0.12 and 0.16T) during 24 hours, with action mode estimates. 24 hours after the exposure procedure, specimens that had not been exposed were compared to the minimum inhibitory concentration (MIC)of antibiotics for the various magnetic forces.

Since the sensitivity of bacterial cells and the action of antibiotics has been measured. Bacterial cells became sensitive to a number of antibiotics, including Gentamycin, Trimethoprim/sulfamethoxazole, Piperacillin, and Cefepime. *Escherichia coli* cells changed susceptibility after being exposed to amagnetic field involved Ciprofloxacin,Cefepime,Ceftazidime,Trimethoprim/ sulfamethoxazole.

S.NO	Antimicrobial	Before exposing MG		After exposing MG power 0.04T		After exposing MG power 0.08T		After exposing MG power 0.12T		After exposing MG power 0.16T	
		MIC	Int.	MIC	Int.	MIC	Int.	MIC	Int.	MIC	Int.
	Ceftazidime	16	R	4	R*	4	R*	8	R*	4	R*
	Cefepime	≥64	R	2	R*	2	R*	2	R*	2	R*
	Aztreonam	≥64	R	16	R*	16	R*	16	R*	16	R*
	Trimethoprim/ Sulfamethoxazole	<=20	S	320	R	320	R	320	R	320	R
	Minocycline	25	S	≤1	S	≤1	S	≤1	S	≤1	S
	Amikacin	16	I*	16	S	16	S	16	S	16	S
1	Ticarcillin/Clavulanic acid	≥128	R	≥128	R	≥128	R	≥128	R	≥128	R
L L	Ticarcillin	64	Ι	64	Ι	64	Ι	64	Ι	64	Ι
	Piperacillin	≥128	R	≥128	R	≥128	R	≥128	R	≥128	R
	Gentamycin	≤1	S	≤1	S	≤1	S	<u>≤1</u>	S	<u>≤</u> 1	S
	Tobramycin	≥16	R	≥16	R	≥16	R	≥16	R	≥16	R
	Ciprofloxacin	<u>≥</u> 4	R	<u>≥</u> 4	R	≥4	R	<u>≥</u> 4	R	<u>≥</u> 4	R
	Meropenem	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S
	Piperacillin/Tazobactam	8	S	8	S	8	S	8	S	8	S
	Imipenem	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S
2	Aztreonam	2	R*	≤1	R*	2	R*	≤1	R*	2	R*
	Minocycline	2	S	≤1	S	≤1	S	≤1	S	<=1	S
	Ceftazidime	<u>≤1</u>	S	≤1	S	≤1	S	≤1	S	<=1	S
	Cefepime	<u>≤1</u>	R*	<u>≤1</u>	R*	≤1	R*	<u>≤1</u>	R*	<u>≤1</u>	R*
	Imipenem	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S
	Ticarcillin/Clavulanic acid	≤ 8	S	≤ 8	S	≤ 8	S	≤ 8	S	< 8	S

Table (4-2) Antimicrobial susceptibility test of exposed and unexposed E. coli for different magnetic force

	Ticarcillin	≥128	R	≥128	R	≥128	R	≥128	R	≥128	R
	Piperacillin	≥128	R	≥128	R	≥128	R	≥128	R	≥128	R
	Gentamycin	≤1	S	≤1	S	≤1	S	≤1	S	≤1	S
	Tobramycin	≤1	S	≤1	S	≤1	S	≤1	S	≤1	S
	Amikacin	≤ 2	S	≤ 2	S	≤ 2	S	≤ 2	S	≤ 2	S
	Trimethoprim/ Sulfamethoxazole	≥320	R	≥320	R	≥320	R	≥320	R	≥320	R
	Ciprofloxacin	\leq 0.25	S	\leq 0.25	S	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S
	Meropenem	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S
	Piperacillin/Tazobactam	≤ 4	S	≤ 4	S	≤ 4	S	≤ 4	S	≤ 4	S
	Ceftazidime	16	R	≥64	R	4	R*	16	R	≥64	R
	Cefepime	4	R	≥64	R	2	R*	32	R	32	R
	Minocycline	4	S	8	Ι	4	S	4	S	4	S
	Aztreonam	≥64	R	≥64	R	16	R	≥64	R	≥64	R
	Imipenem	\leq 0.25	S	1	S	32	Ι	0.5	S	0.5	S
	Ticarcillin/Clavulanic acid	≥128	R	32	R	≥128	R	≥128	R	≥128	R
	Ticarcillin	≥128	R	≤128	R	≥128	R	≥128	R	≥128	R
3	Piperacillin	≥128	R	≤128	R	≥128	R	≥128	R	≥128	R
	Gentamycin	≤1	S	≤1	S	≤1	S	≤1	S	≤1	S
	Tobramycin	<u>≤1</u>	S	<u>≤</u> 1	S	<u>≤1</u>	S	<u>≤1</u>	S	<u>≤1</u>	S
	Amikacin	≤ 2	S	≤ 2	S	≤ 2	S	≤ 2	S	≤ 2	S
	Trimethoprim/ Sulfamethoxazole	≤ 20	S	≤ 20	S	≤ 20	S	≤ 20	S	≤ 20	S
	Ciprofloxacin	≥4	R	≥4	R	≥4	R	<u>≥</u> 4	R	≥4	R
	Meropenem	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S
	Piperacillin/Tazobactam	8	S	64	Ι	<=4	S	8	S	16	S
4	Ceftazidime	4	R*	4	R*	4	R*	4	R*	16	R*
	Aztreonam	16	R	16	R*	16	R*	16	R*	16	R*

Piperacillin	≥128	R	
Ticarcillin	≥128	R	
Ticarcillin/Clavulanic acid	≤ 8	S	
Meropenem	≥0.25	S	
Imipenem	≥0.25	S	
Gentamycin	≤1	S	
Tobramycin	≤1	S	
Amikacin	≤ 2	S	
Minocycline	≤1	S	
Ciprofloxacin	≤ 0.25	S	
			1 -

	Ticarcillin	≥128	R	≥128	R	≥128	R	≥128	R	≥128	R
	Ticarcillin/Clavulanic acid	≤ 8	S	≤ 8	S	≤ 8	S	≤ 8	S	≤ 8	S
	Meropenem	≥0.25	S	≥0.25	S	≥0.25	S	≥0.25	S	≥0.25	S
	Imipenem	≥0.25	S	≥0.25	S	≥0.25	S	≥0.25	S	≥0.25	S
	Gentamycin	≤1	S	≤1	S	≤1	S	≤1	S	≤1	S
	Tobramycin	≤1	S	≤1	S	≤1	S	≤1	S	≤1	S
	Amikacin	≤ 2	S	≤ 2	S	≤ 2	S	≤ 2	S	≤ 2	S
	Minocycline	≤1	S	≤1	S	≤1	S	≤1	S	≤1	S
	Ciprofloxacin	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S	\leq 0.25	S
	Cefepime	≤1	R*	≤1	R*	≤1	R*	≤1	R*	≤1	R*
-	Piperacillin	≥128	R	≥128	R	64	R*	64	R*	≥128	R
	Ceftazidime	0.5	R*	1	R*	1	R*	1	R*	1	R*
	Cefepime	0.5	R*	1	R*	1	R*	1	R*	1	R*
	Piperacillin/Tazobactam	≥4	R	≥4	R	≥4	R	≥4	R	≥4	R
	Aztreonam	≤1	R*	≤1	R*	≤1	R*	≤1	R*	≤1	R*
	Imipenem	≤ 0.5	S	≤ 0.5	S	≤ 0.5	S	≤ 0.5	S	≤ 0.5	S
5	Amikacin	≤ 2	S	≤ 2	S	≤ 2	S	≤ 2	S	≤ 2	S
	Gentamicin	≤1	S	≤1	S	≤1	S	≤1	S	≤1	S
	Tobramycin	≤1	S	≤1	S	≤1	S	≤1	S	≤1	S
-	Ciprofloxacin	\leq 0.25	S	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S	\leq 0.25	S
	Meropenem	≤0.5	S	≤ 0.5	S	≤0.5	S	≤ 0.5	S	≤ 0.5	S
	Tigecycline	≤0.5	S	≤ 0.5	S	≤0.5	S	≤ 0.5	S	≤ 0.5	S
	Trimethoprim/ Sulfamethoxazole	≥320	R	≥320	R	≥320	R	≥320	R	≥320	R
6	Piperacillin/Tazobactam	4	S	<u><</u> 4	S	8	S	8	S	8	S
U	Ceftazidime	≤1	R*	≤1	R*	16	R*	≥64	R	<u>≤</u> 1	R*

R*

64

64

R*

≥128

R

≥128

R
	Cefepime	1	R*	1	R*	2	R*	2	R*	2	R*
	Aztreonam	1	R*	1	R*	16	R*	>=64	R*	16	R*
	Imipenem	0.25	S	0.25	S	0.5	S	0.25	S	0.25	S
	Amikacin	2	S	2	S	16	S	16	S	16	S
	Gentamicin	16	R	16	R	≤1	S	≤1	S	≤1	S
	Tobramycin	8	R*	4	S	≥16	R	≥16	R	≥16	R
	Ciprofloxacin	0.25	S	0.25	S	≥4	R	≥4	R	≥4	R
	Ticarcillin	≥128	R								
	Ticarcillin/Clavulanic acid	16	S								
	Meropenem	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S	\leq 0.25	S
	Trimethoprim/ Sulfamethoxazole	≥320	R								
	Piperacillin	≥128	R								
	Minocycline	8	Ι	≤16	R	≤1	S	≤1	S	≤1	S
	Imipenem	1	S	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S
	Meropenem	0.5	S	≤ 0.25	S						
	Ceftazidime	≥64	R								
	Cefepime	≥32	R								
	Piperacillin	≥32	R	≥32	R	<u>≥</u> 32	R	≥32	R	≥32	R
7	Gentamycin	≥16	R								
	Tigecycline	≤ 0.5	S								
	Amikacin	4	S	4	S	4	S	4	S	4	S
	Trimethoprim/ Sulfamethoxazole	≥ 20	R								
	Ciprofloxacin	≥4	R								
	Piperacillin/Tazobactam	≥128	R								
Q	Ceftazidime	≤ 0.12	S	≤ 0.25	S	≤ 0.12	S	≤ 0.12	S	≤ 0.12	S
o	Amikacin	≤1	S	2	S	≤1	S	2	S	≤1	S

	Nitrofurantoin	≤16	S	64	Ι	<u>≤</u> 16	S	<u>≤</u> 16	S	≤16	S
	Amoxicillin/Clavnic acid	4	S	4	S	4	S	4	S	4	S
	Piperacillin/Tazobactam	≤ 4	S	≤ 4	S	≤ 4	S	≤ 4	S	≤ 4	S
	Meropenem	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S
	Imipenem	\leq 0.25	S	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S
	Gentamycin	≤1	S	≤1	S	≤1	S	≤1	S	≤1	S
	Ciprofloxacin	≤ 0.06	S	≤ 0.06	S	≤ 0.06	S	≤ 0.06	S	≤ 0.06	S
	Trimethoprim/ Sulfamethoxazole	≤ 20	S	≤ 20	S	≤ 20	S	≤ 20	S	≤ 20	S
	Cefepime	≤ 0.12	S	≤ 0.12	S	≤ 0.12	S	≤ 0.12	S	≤ 0.12	S
	Cefazolin	≤ 4	S	≤ 4	S	≤ 4	S	≤ 4	S	≤ 4	S
	Piperacillin	32	R*	≤128	R	≤128	R	≤128	R	≤128	R
	Piperacillin/Tazobactam	≤ 4	S	64	Ι	64	Ι	64	Ι	64	Ι
	Ceftazidime	≤1	R*	≥ 64	R	16	R*	16	R*	≥64	R*
	Cefepime	≤1	R*	≥64	R	≥64	R	≥64	R	≥64	R
	Aztreonam	≤1	R*	≥64	R	≥64	R	≥64	R	≥64	R
	Ciprofloxacin	2	R*	≥4	R	≥4	R	≥4	R	≥4	R
	Minocycline	2	S	≤1	S	≤1	S	≤1	S	≤1	S
9	Ticarcillin	≥128	R	≥128	R	≥128	R	≥128	R	≥128	R
	Ticarcillin/Clavnic acid	16	S	16	S	16	S	16	S	16	S
	Meropenem	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S
	Imipenem	\leq 0.25	S	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S	\leq 0.25	S
	Gentamycin	≤1	S	≤1	S	≤1	S	≤1	S	≤1	S
	Tobramycin	≤1	S	<u>≤</u> 1	S	≤1	S	≤1	S	≤1	S
	Amikacin	≤ 2	S	≤ 2	S	≤ 2	S	≤ 2	S	≤ 2	S
	Trimethoprim/ Sulfamethoxazole	≥320	R	≤ 20	S	≤ 20	S	≤ 20	S	≤ 20	S

	Ceftazidime	2	R*	≤1	S	16	R	2	S	2	S
	Cefepime	≤ 0.12	R*	≤1	S	≤ 0.12	R*	≤ 0.12	R*	≤ 0.12	R*
	Trimethoprim/ Sulfamethoxazole	≤ 20	S	≥ 320	R						
	Amikacin	≤ 2	S	≤ 2	S	4	S	4	S	4	S
	Ciprofloxacin	0.5	S	0.5	S	1	S	1	S	1	S
	Imipenem	≤ 0.25	S	≤ 0.25	S	0.5	S	\leq 0.25	S	0.5	S
	Piperacillin/Tazobactam	≤ 4	S								
	Aztreonam	≤1	R*								
	Meropenem	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S
	Gentamycin	≤1	S								
	Netlimicin	≤1	S								
	Tobramycin	≤1	S								
	levofloxacin	0.5	S								
	Tigecyclin	≤ 0.5	S								
	Ceftazidime	0.5	S	≤1	R*	≤1	R*	≤1	R*	≤1	R*
	Cefepime	≤ 0.12	S	≤1	R*	≤1	R*	≤1	R*	≤1	R*
	Aztreonam	≤1	S	≤1	R*	≤1	R*	≤1	R*	≤1	R*
	Piperacillin	≥128	R								
	Piperacillin/Tazobactam	≤ 4	S								
11	Imipenem	\leq 0.25	S	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S
11	Meropenem	\leq 0.25	S	≤ 0.25	S						
	Amikacin	≤ 2	S								
	Gentamycin	≤1	S								
	Netlimicin	≤1	S								
	Tobramycin	≤1	S								
	levofloxacin	≥ 4	R								

	Tigecyclin	≤ 0.5	S	≤ 0.5	S	≤ 0.5	S	≤ 0.5	S	\leq 0.5	S
	Trimethoprim/ Sulfamethoxazole	≤ 20	S	≤ 20	S	≤ 20	S	≤ 20	S	≤ 20	S
	Ampicillin	8	S	8	S	8	S	8	S	8	S
	Piperacillin/Tazobactam	4	S	4	S	4	S	4	S	4	S
	Ceftazidime	≤1	S	≤1	S	≤1	S	≤1	S	<u>≤1</u>	S
	Cefepime	≤1	S	≤1	S	≤1	S	≤1	S	≤1	S
	Imipenem	≤ 0.25	S	≤ 0.25	S						
	Norfloxacin	0.5	R*	0.5	R*	0.5	R*	0.5	R*	0.5	R*
	Trimethoprim/ Sulfamethoxazole	≤ 20	S	≤ 20	S	≤ 20	S	≤ 20	S	≤ 20	S
12	Amikacin	≤ 2	S	≤ 2	S	≤ 2	S	≤ 2	S	≤ 2	S
	Gentamycin	≤1	S	≤1	S	≤1	S	≤1	S	≤1	S
	Meropenem	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S
	Ciprofloxacin	≤ 0.25	S	\leq 0.25	S	≤ 0.25	S	≤ 0.25	S	\leq 0.25	S
	Norfloxacin	≤ 0.5	R*	≤ 0.5	R*	≤ 0.5	R*	≤ 0.5	R*	≤ 0.5	R*
	Cefotaxime	≤1	S	≤1	S	≤1	S	≤1	S	≤1	S
	Amoxicillin/Clavulanic Acid	4	S	4	S	4	S	4	S	4	S
	Nitrofurantoin	≤16	S	≤16	S	≤16	S	≤16	S	≤16	S
	Piperacillin	32	R*	≤128	R	≤128	R	≤128	R	≤128	R
	Piperacillin/Tazobactam	\leq	S	64	Ι	64	Ι	64	Ι	64	Ι
	Ceftazidime	≤1	R*	≥64	R	16	R*	16	R*	≥64	R*
	Cefepime	≤1	R*	≥64	R	≥64	R	≥64	R	≥64	R
13	Aztreonam	≤1	R*	≥64	R	≥64	R	≥64	R	≥64	R
	Ciprofloxacin	2	R*	≥4	R	≥4	R	≥4	R	≥4	R
	Minocycline	2	S	≤1	S	≤1	S	≤1	S	≤1	S
	Ticarcillin	>128	R	>128	R	>128	R	>128	R	>128	R
	Tieurennin	_120		_120							

	Meropenem	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S
	Imipenem	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S
	Gentamycin	≤ 1	S	≤ 1	S	≤ 1	S	≤ 1	S	≤ 1	S
	Tobramycin	≤ 1	S	≤ 1	S	≤ 1	S	≤ 1	S	≤ 1	S
	Amikacin	≤ 2	S	≤ 2	S	≤ 2	S	≤ 2	S	≤ 2	S
	Trimethoprim/ Sulfamethoxazole	≥320	R	≤ 20	S						
	Piperacillin/Tazobactam	≤ 4	S	≤ 4	S	8	S	8	S	8	S
	Ceftazidime	≤1	R*	≤1	R*	16	R*	≥64	R	≤1	R*
	Cefepime	1	R*	1	R*	2	R*	2	R*	2	R*
	Aztreonam	1	R*	1	R*	16	R*	≥64	R*	16	R*
	Imipenem	0.25	S	0.25	S	0.5	S	0.25	S	0.25	S
	Amikacin	2	S	2	S	16	S	16	S	16	S
14	Gentamicin	16	R	16	R	≤1	S	≤1	S	≤1	S
	Tobramycin	8	R*	4	S	≥16	R	≥16	R	≥16	R
	Ciprofloxacin	0.25	S	0.25	S	≥4	R	≥4	R	≥4	R
	Ticarcillin	≥128	R	≥128	R	≥128	R	≥128	R	≥128	R
	Ticarcillin/Clavulanic acid	16	S	16	S	16	S	16	S	16	S
	Meropenem	≤ 0.25	S	≤ 0.25	S						
	Trimethoprim/ Sulfamethoxazole	≥320	R	≥320	R	≥320	R	≥320	R	≥320	R
	Ceftazidime	16	R	4	R*	4	R*	8	R*	4	R*
	Cefepime	≥64	R	2	R*	2	R*	2	R*	2	R*
	Aztreonam	≥64	R	16	R*	16	R*	16	R*	16	R*
15	Trimethoprim/ Sulfamethoxazole	≤ 20	S	320	R	320	R	320	R	320	R
	Minocycline	25	S	≤1	S	≤1	S	≤1	S	≤1	S
	Amikacin	16	I*	16	S	16	S	16	S	16	S
	Ticarcillin/Clavulanic acid	≥128	R	≥128	R	≥128	R	≥128	R	≥128	R

	Piperacillin	≥128	R								
	Gentamycin	≤1	S								
	Ciprofloxacin	≥4	R								
	Meropenem	≤ 0.25	S	\leq 0.25	S	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S
	Piperacillin/Tazobactam	8	S	8	S	8	S	8	S	8	S
	Imipenem	≤ 0.25	S	\leq 0.25	S	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S
	Piperacillin	≥128	R	≥128	R	64	R*	64	R*	≥128	R
	Ceftazidime	0.5	R*	1	R*	1	R*	1	R*	1	R*
	Cefepime	0.5	R*	1	R*	1	R*	1	R*	1	R*
	Piperacillin/Tazobactam	≥4	R								
	Aztreonam	≤ 1	R*	≤1	R*	≤1	R*	≤1	R*	≤1	R*
	Imipenem	≤ 0.5	S	<=0.5	S	≤0.5	S	≤ 0.5	S	≤ 0.5	S
16	Amikacin	≤ 2	S								
	Gentamicin	≤ 1	S								
	Tobramycin	≤ 1	S								
	Ciprofloxacin	\leq 0.25	S	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S
	Meropenem	≤ 0.5	S	≤ 0.5	S	≤0.5	S	≤ 0.5	S	≤ 0.5	S
	Tigecycline	≤ 0.5	S	≤ 0.5	S	≤0.5	S	≤ 0.5	S	≤ 0.5	S
	Trimethoprim/ Sulfamethoxazole	≥320	R								
	Aztreonam	2	R*	≤ 1	R*	2	R*	≤ 1	R*	2	R*
	Minocycline	2	S	≤ 1	S						
	Ceftazidime	<u>≤</u> 1	S	≤ 1	S	≤ 1	S	≤ 1	S	<u>≤</u> 1	S
17	Cefepime	<u>≤</u> 1	R*	≤ 1	R*	<u>≤</u> 1	R*	<u>≤</u> 1	R*	≤ 1	R*
	Imipenem	\leq 0.25	S	≤ 0.25	S	≤ 0.25	S	\leq 0.25	S	≤ 0.25	S
	Ticarcillin/Clavulanic acid	≤ 8	S								
	Ticarcillin	≥128	R								

	Piperacillin	≥128	R	≥128	R	≥128	R	≥128	R	≥128	R
	Amikacin	≤ 2	S	≤ 2	S	≤ 2	S	≤ 2	S	≤ 2	S
	Trimethoprim/ Sulfamethoxazole	≥320	R	≥320	R	≥320	R	≥320	R	≥320	R
	Ciprofloxacin	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S
	Meropenem	\leq 0.25	S	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S
	Piperacillin/Tazobactam	≤ 4	S	≤ 4	S	≤ 4	S	≤ 4	S	≤ 4	S
	Ceftazidime	16	R	≥64	R	4	R*	16	R	≥ 64	R
	Cefepime	4	R	≥64	R	2	R*	32	R	32	R
	Minocycline	4	S	8	Ι	4	S	4	S	4	S
	Aztreonam	≥64	R	≥64	R	16	R	≥64	R	≥64	R
	Imipenem	<=0.25	S	1	S	32	Ι	0.5	S	0.5	S
	Ticarcillin/Clavulanic acid	≥128	R	32	R	≥128	R	≥128	R	≥128	R
	Ticarcillin	≥128	R	≥128	R	≥128	R	≥128	R	≥128	R
18	Piperacillin	≥128	R	≥128	R	≥128	R	≥128	R	≥128	R
	Gentamycin	≤1	S	≤1	S	≤1	S	≤1	S	≤1	S
	Tobramycin	≤1	S	≤1	S	≤1	S	≤1	S	≤1	S
	Amikacin	≤ 2	S	≤ 2	S	≤ 2	S	≤ 2	S	≤ 2	S
	Trimethoprim/ Sulfamethoxazole	≤ 20	S	≤ 20	S	≤ 20	S	≤ 20	S	≤ 20	S
	Ciprofloxacin	≥4	R	≥4	R	≥4	R	≥4	R	≥4	R
	Meropenem	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S
	Piperacillin/Tazobactam	8	S	64	Ι	≤ 4	S	8	S	16	S
	Ceftazidime	0.5	S	≤ 1	R*	≤1	R*	≤1	R*	≤ 1	R*
	Cefepime	≤ 0.12	S	≤1	R*	≤1	R*	≤1	R*	<u>≤1</u>	R*
19	Aztreonam	≤1	S	≤1	R*	≤1	R*	≤1	R*	≤1	R*
	Piperacillin	≥128	R	≥128	R	≥128	R	≥128	R	≥128	R
	Piperacillin/Tazobactam	≤ 4	S	≤ 4	S	≤ 4	S	≤ 4	S	≤ 4	S

	Imipenem	\leq 0.25	S	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S
	Meropenem	≤ 0.25	S	\leq 0.25	S	\leq 0.25	S	≤ 0.25	S	≤ 0.25	S
	Amikacin	≤ 2	S	≤ 2	S	≤ 2	S	≤ 2	S	≤ 2	S
	Gentamycin	≤ 1	S	≤ 1	S	≤ 1	S	≤ 1	S	≤ 1	S
	Netlimicin	≤ 1	S	≤ 1	S	≤ 1	S	≤ 1	S	≤ 1	S
	Tobramycin	≤ 1	S	≤ 1	S	≤ 1	S	≤ 1	S	≤ 1	S
	levofloxacin	≥4	R	≥4	R	≥4	R	≥4	R	≥4	R
	Tigecyclin	≤ 0.5	S	≤ 0.5	S	≤ 0.5	S	≤ 0.5	S	≤ 0.5	S
	Trimethoprim/ Sulfamethoxazole	≤ 20	S	≤ 20	S	≤ 20	S	≤ 20	S	≤ 20	S
	Ampicillin	8	S	8	S	8	S	8	S	8	S
	Piperacillin/Tazobactam	4	S	4	S	4	S	4	S	4	S
	Ceftazidime	≤1	S	≤1	S	≤1	S	≤1	S	≤1	S
	Cefepime	≤1	S	≤ 0.25	S	≤1	S	≤1	S	≤1	S
	Iminonom	< 0.25	C	<-0.25	C	< 0.25	2	< 0.25	C	< 0.25	C
	Impenent	≥ 0.23	S S	<=0.23	S S	≥ 0.23	5	≥ 0.23	S	≥ 0.23	3
	Norfloxacin	$\frac{\leq 0.23}{0.5}$	R*	0.5	3 R*	$\frac{\leq 0.23}{0.5}$	R*	0.5	R*	$\frac{\leq 0.23}{0.5}$	8 R*
	Norfloxacin Trimethoprim/ Sulfamethoxazole	$\begin{array}{c} \leq 0.23 \\ \hline 0.5 \\ \leq 20 \end{array}$	8 R* S	<=0.23 0.5 <=20	8 R* S	<u> </u>	8 R* S	<u> </u>	R* S		S R* S
20	Norfloxacin Trimethoprim/ Sulfamethoxazole Amikacin	$\begin{array}{r c} \leq 0.23 \\ \hline 0.5 \\ \leq 20 \\ \leq 2 \end{array}$	S R* S S	<=0.23 0.5 <=20 <=2	S R* S S	<u>>0.23</u> 0.5 <=20 <=2	S R* S S	<u></u>	S R* S S	<u> </u>	S R* S S
20	Norfloxacin Trimethoprim/ Sulfamethoxazole Amikacin Gentamycin	$\begin{array}{r c} \leq 0.23 \\ \hline 0.5 \\ \leq 20 \\ \leq 2 \\ \leq 1 \end{array}$	S R* S S S	<=0.23 0.5 <=20 <=2 <=1	S R* S S S S	<u> </u>	S R* S S S S	0.5 <=20 <=2 <=1	S R* S S S	<u> </u>	S R* S S S S
20	Norfloxacin Trimethoprim/ Sulfamethoxazole Amikacin Gentamycin Meropenem	$ \begin{array}{r} \leq 0.23 \\ \hline 0.5 \\ \leq 20 \\ \leq 2 \\ \leq 1 \\ \leq 0.25 \end{array} $	S R* S S S S S	$ \begin{array}{r} < -0.23 \\ \hline 0.5 \\ < =20 \\ < =2 \\ < =1 \\ \leq 0.25 \\ \end{array} $	S R* S S S S S	$ \begin{array}{r} \underline{>} 0.23 \\ \hline 0.5 \\ <=20 \\ <=2 \\ <=1 \\ \leq 0.25 \\ \end{array} $	S R* S S S S S	$ \begin{array}{r} \underline{<} 0.25 \\ \hline 0.5 \\ <=20 \\ <=2 \\ <=1 \\ \leq 0.25 \\ \end{array} $	S R* S S S S		S R* S S S S S
20	ImperientNorfloxacinTrimethoprim/ SulfamethoxazoleAmikacinGentamycinMeropenemCiprofloxacin	$ \begin{array}{r} \leq 0.23 \\ \hline 0.5 \\ \leq 20 \\ \leq 2 \\ \leq 1 \\ \leq 0.25 \\ \leq 0.25 \end{array} $	S R* S S S S S	$ \begin{array}{r} < -0.23 \\ \hline 0.5 \\ < =20 \\ < =2 \\ < =1 \\ \hline \leq 0.25 \\ \leq 0.25 \\ \end{array} $	S R* S S S S S S S	$ \begin{array}{r} \underline{\leq} 0.23 \\ 0.5 \\ <=20 \\ <=2 \\ <=1 \\ \underline{\leq} 0.25 \\ \underline{\leq} 0.25 \\ \end{array} $	R* S S S S S S	$ \begin{array}{r} \underline{0.23} \\ 0.5 \\ <=20 \\ <=2 \\ <=1 \\ \leq 0.25 \\ \leq 0.25 $	R* S S S S S S		S R* S S S S S S S
20	Imperient Norfloxacin Trimethoprim/ Sulfamethoxazole Amikacin Gentamycin Meropenem Ciprofloxacin Norfloxacin	$ \begin{array}{r} \leq 0.23 \\ \hline 0.5 \\ \leq 20 \\ \leq 2 \\ \leq 1 \\ \leq 0.25 \\ \leq 0.25 \\ \leq 0.5 \end{array} $	R* S S S S R*	$ \begin{array}{r} <=0.23 \\ 0.5 \\ <=20 \\ <=2 \\ <=1 \\ \leq 0.25 \\ \leq 0.25 \\ \leq 0.5 \\ $	S R* S S S S R*	$ \begin{array}{r} \leq 0.23 \\ \hline 0.5 \\ <=20 \\ <=2 \\ <=1 \\ \leq 0.25 \\ \leq 0.25 \\ \leq 0.5 \\ \end{array} $	R* S S S S S R*	$ \begin{array}{r} \underline{<} 0.25 \\ 0.5 \\ <=20 \\ <=2 \\ <=1 \\ \underline{<} 0.25 \\ \underline{<} 0.25 \\ \underline{<} 0.5 \\ \end{array} $	R* S S S S R*	$ \begin{array}{r} \leq 0.23 \\ \hline 0.5 \\ <=20 \\ <=2 \\ <=1 \\ \hline \leq 0.25 \\ \leq 0.25 \\ \leq 0.5 \\ \end{array} $	S R* S S S S R*
20	ImperientNorfloxacinTrimethoprim/ SulfamethoxazoleAmikacinGentamycinMeropenemCiprofloxacinNorfloxacinCefotaxime	$ \begin{array}{r rrrrrrrrrrrrrrrrrrrrrrrrrrrrrrrrrrr$	S S S S S R* S	$ \begin{array}{r} < -0.23 \\ \hline 0.5 \\ <=20 \\ <=1 \\ \leq 0.25 \\ \leq 0.25 \\ \leq 0.5 \\ \leq 1 $	S R* S S S R* S	$ \begin{array}{r} \underline{\leq} 0.23 \\ \hline 0.5 \\ <=20 \\ <=2 \\ <=1 \\ \underline{\leq} 0.25 \\ \underline{\leq} 0.25 \\ \underline{\leq} 0.5 \\ \underline{\leq} 1 \end{array} $	R* S S S S R* S	$ \begin{array}{r} \ \ \ \ \ \ \ \ \ \ \ \ \ \ \$	R* S S S R* S	$ \begin{array}{r} \underline{\leq} 0.23 \\ 0.5 \\ <=20 \\ <=2 \\ <=1 \\ \underline{\leq} 0.25 \\ \underline{\leq} 0.25 \\ \underline{\leq} 0.5 \\ \underline{\leq} 1 \end{array} $	S R* S S S R* S S S S S S S S
20	Imperient Norfloxacin Trimethoprim/ Sulfamethoxazole Amikacin Gentamycin Meropenem Ciprofloxacin Norfloxacin Cefotaxime Amoxicillin/Clavulanic Acid	$ \begin{array}{r rrrrrrrrrrrrrrrrrrrrrrrrrrrrrrrrrrr$	S S S S S S S S S S S S S S S S	$ \begin{array}{r} < -0.23 \\ \hline 0.5 \\ <=20 \\ <=1 \\ \leq 0.25 \\ \leq 0.25 \\ \leq 0.5 \\ \leq 1 \\ 4 $	S R* S S S R* S S S S S S S S S S	$ \begin{array}{r} \underline{\leq} 0.23 \\ 0.5 \\ <=20 \\ <=1 \\ \leq 0.25 \\ \leq 0.25 \\ \leq 0.5 \\ \leq 1 \\ 4 $	R* S S S R* S S S S S S S S S S S S S	$ \begin{array}{r} \underline{\leq} 0.25 \\ 0.5 \\ <=20 \\ <=2 \\ <=1 \\ \leq 0.25 \\ \leq 0.25 \\ \leq 0.5 \\ \leq 1 \\ 4 \end{array} $	S S S S S S S S S S S S S S S S S S		S R* S S S R* S S S S S S S S S S
20	ImperientNorfloxacinTrimethoprim/ SulfamethoxazoleAmikacinGentamycinMeropenemCiprofloxacinNorfloxacinCefotaximeAmoxicillin/Clavulanic AcidNitrofurantoin	$ \begin{array}{r rrrrrrrrrrrrrrrrrrrrrrrrrrrrrrrrrrr$	S S	$ \begin{array}{r} <=0.23 \\ 0.5 \\ <=20 \\ <=1 \\ \leq 0.25 \\ \leq 0.25 \\ \leq 0.5 \\ \leq 1 \\ 4 \\ \leq 16 \\ $	S R* S S R* S S S S S S S S S S S S S S S S S S	$ \begin{array}{r} \underline{\leq} 0.23 \\ 0.5 \\ <=20 \\ <=2 \\ <=1 \\ \underline{\leq} 0.25 \\ \underline{\leq} 0.25 \\ \underline{\leq} 0.5 \\ \underline{\leq} 1 \\ 4 \\ \underline{\leq} 16 \\ \end{array} $	R* S S S S R* S	$ \begin{array}{r} \underline{\leq} 0.25 \\ 0.5 \\ <=20 \\ <=2 \\ <=1 \\ \underline{\leq} 0.25 \\ \underline{\leq} 0.25 \\ \underline{\leq} 0.5 \\ \underline{\leq} 1 \\ 4 \\ \underline{\leq} 16 \end{array} $	S S	$ \begin{array}{r} \leq 0.23 \\ \hline 0.5 \\ <=20 \\ <=1 \\ \leq 0.25 \\ \leq 0.25 \\ \leq 0.5 \\ \leq 1 \\ 4 \\ \leq 16 \end{array} $	S R* S S S R* S S S S S S S S S S S S S S S
20	ImperientNorfloxacinTrimethoprim/ SulfamethoxazoleAmikacinGentamycinMeropenemCiprofloxacinNorfloxacinCefotaximeAmoxicillin/Clavulanic AcidNitrofurantoinPiperacillin	$ \begin{array}{r} \leq 0.23 \\ \hline 0.5 \\ \leq 20 \\ \leq 2 \\ \leq 1 \\ \leq 0.25 \\ \leq 0.25 \\ \leq 0.5 \\ \leq 1 \\ 4 \\ \leq 16 \\ 8 \\ \end{array} $	S S S S R* S R* S R* S R* R* S S R*	$ \begin{array}{r} < -0.23 \\ \hline 0.5 \\ <=20 \\ <=1 \\ \leq 0.25 \\ \leq 0.25 \\ \leq 0.5 \\ \leq 1 \\ 4 \\ \leq 16 \\ 8 $	S R* S S R* S	$ \begin{array}{r} \underline{\leq} 0.23 \\ 0.5 \\ <=20 \\ <=2 \\ <=1 \\ \leq 0.25 \\ \leq 0.25 \\ \leq 0.5 \\ \leq 1 \\ 4 \\ \leq 16 \\ 8 \\ \end{array} $	R* S S S S R* S	$ \begin{array}{r} \underline{\leq} 0.25 \\ 0.5 \\ <=20 \\ <=2 \\ <=1 \\ \leq 0.25 \\ \leq 0.25 \\ \leq 0.5 \\ \leq 1 \\ 4 \\ \leq 16 \\ 8 \\ \end{array} $	R* S S S R* S		S R* S S S R* S

	Cefepime	≤ 0.12	R*	≤1	S	≤ 0.12	R*	≤ 0.12	R*	≤ 0.12	R*
	Trimethoprim/ Sulfamethoxazole	≤ 20	S	≥320	R	≥ 320	R	≥ 320	R	≥ 320	R
	Amikacin	≤ 2	S	≤ 2	S	4	S	4	S	4	S
	Ciprofloxacin	0.5	S	0.5	S	1	S	1	S	1	S
	Imipenem	≤ 0.25	S	\leq 0.25	S	0.5	S	≤ 0.25	S	0.5	S
	Piperacillin/Tazobactam	≤ 4	S	≤ 4	S	≤ 4	S	≤ 4	S	≤ 4	S
	Aztreonam	≤ 1	R*	≤ 1	R*	≤ 1	R*	≤ 1	R*	≤ 1	R*
	Meropenem	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S
	Gentamycin	≤1	S	≤1	S	≤1	S	≤1	S	≤1	S
	Netlimicin	≤1	S	≤1	S	≤1	S	≤1	S	≤1	S
	Tobramycin	≤1	S	≤1	S	≤1	S	≤1	S	≤1	S
	levofloxacin	0.5	S	0.5	S	0.5	S	0.5	S	0.5	S
	Imipenem	1	S	<=0.25	S	<=0.25	S	<=0.25	S	<=0.25	S
	Meropenem	0.5	S	<=0.25	S	<=0.25	S	<=0.25	S	<=0.25	S
	Ceftazidime	≥64	R	≥64	R	≥64	R	≥64	R	≥64	R
	Cefepime	≥32	R	≥32	R	≥32	R	≥32	R	≥32	R
	Piperacillin	≥32	R	≥32	R	≥32	R	≥32	R	≥32	R
22	Gentamycin	≥16	R	≥16	R	≥16	R	≥16	R	≥16	R
	Tigecycline	≤ 0.5	S	≤ 0.5	S	≤ 0.5	S	≤ 0.5	S	≤ 0.5	S
	Amikacin	4	S	4	S	4	S	4	S	4	S
	Trimethoprim/ Sulfamethoxazole	≥20	R	≥20	R	≥20	R	≥20	R	≥20	R
	Ciprofloxacin	≥4	R	≥4	R	≥4	R	≥4	R	≥4	R
	Piperacillin/Tazobactam	≥128	R	≥128	R	≥128	R	≥128	R	≥128	R
	Ceftazidime	4	R*	4	R*	4	R*	4	R*	16	R*
23	Aztreonam	16	R	16	R*	16	R*	16	R*	16	R *
	Piperacillin	≥128	R	64	R*	64	R*	≥128	R	≥128	R

	Ticarcillin	≥128	R	≥128	R	≥128	R	≥128	R	≥128	R
	Ticarcillin/Clavulanic acid	≤ 8	S	≤ 8	S	≤ 8	S	≤ 8	S	≤ 8	S
	Meropenem	\leq 0.25	S	\leq 0.25	S	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S
	Imipenem	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S
	Gentamycin	≤1	S	≤1	S	≤1	S	≤1	S	≤1	S
	Tobramycin	≤1	S	≤1	S	≤1	S	≤1	S	≤1	S
	Amikacin	≤ 2	S	≤ 2	S	≤ 2	S	≤ 2	S	≤ 2	S
	Minocycline	≤1	S	≤1	S	≤1	S	≤1	S	≤1	S
	Ciprofloxacin	\leq 0.25	S	≤0.25	S	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S
	Cefepime	<=1	R*	≤1	R*	≤1	R*	≤1	R*	≤1	R*
	Aztreonam	2	R*	≤1	R*	2	R*	≤1	R*	2	R*
	Minocycline	2	S	≤1	S	≤1	S	≤1	S	≤1	S
	Ceftazidime	≤1	S	≤1	S	≤1	S	≤1	S	≤1	S
	Cefepime	≤1	R*	≤1	R*	<u>≤</u> 1	R*	≤1	R*	≤1	R*
	Cefepime Imipenem	$\frac{\leq 1}{\leq 0.25}$	R* S	$\frac{\leq 1}{\leq 0.25}$	R* S	$\frac{\leq 1}{\leq 0.25}$	R* S	$\frac{\leq 1}{\leq 0.25}$	R* S	$\frac{\leq 1}{\leq 0.25}$	R* S
	Cefepime Imipenem Ticarcillin/Clavulanic acid		R* S S		R* S S		R* S S		R* S S		R* S S
24	Cefepime Imipenem Ticarcillin/Clavulanic acid Ticarcillin	$ \begin{array}{r} \leq 1 \\ \leq 0.25 \\ \leq 8 \\ \geq 128 \end{array} $	R* S R		R* S S R	$ \begin{array}{r} \leq 1 \\ \leq 0.25 \\ \leq 8 \\ \geq 128 \end{array} $	R* S S R	$ \begin{array}{r} \leq 1 \\ \leq 0.25 \\ \leq 8 \\ \geq 128 \end{array} $	R* S R		R* S S R
24	Cefepime Imipenem Ticarcillin/Clavulanic acid Ticarcillin Piperacillin	$ \begin{array}{r} \leq 1 \\ \leq 0.25 \\ \leq 8 \\ \geq 128 \\ \geq 128 \\ \end{array} $	R* S R R		R* S S R R		R* S S R R		R* S R R	$ \begin{array}{r} \leq 1 \\ \leq 0.25 \\ \leq 8 \\ \geq 128 \\ \geq 128 \\ \end{array} $	R* S R R
24	Cefepime Imipenem Ticarcillin/Clavulanic acid Ticarcillin Piperacillin Amikacin	$ \begin{array}{r} \leq 1 \\ \leq 0.25 \\ \leq 8 \\ \geq 128 \\ \geq 128 \\ \leq 2 \\ \end{array} $	R* S R R S	≤ 1 ≤ 0.25 ≤ 8 ≥ 128 ≥ 128 ≤ 2	R* S R R S	≤ 1 ≤ 0.25 ≤ 8 ≥ 128 ≥ 128 ≤ 2	R* S R R S		R* S R R S	$ \begin{array}{r} \leq 1 \\ \leq 0.25 \\ \leq 8 \\ \geq 128 \\ \geq 128 \\ \leq 2 \\ \end{array} $	R* S R R S
24	Cefepime Imipenem Ticarcillin/Clavulanic acid Ticarcillin Piperacillin Amikacin Trimethoprim/ Sulfamethoxazole		R* S R R S R R R R R R R	≤ 1 ≤ 0.25 ≤ 8 ≥ 128 ≥ 128 ≤ 2 ≥ 320	R* S R R S R R R R R		R* S R R R S R		R* S R R S R R R R R R R		R* S R R S R R R R
24	Cefepime Imipenem Ticarcillin/Clavulanic acid Ticarcillin Piperacillin Amikacin Trimethoprim/ Sulfamethoxazole Ciprofloxacin		R* S R S R S R S S S S S S S S		R* S R R S R S R S S S S S S S		R* S R R S R S R S		R* S R S R S R S S S S S S S S S		R* S R R S R S R S S
24	Cefepime Imipenem Ticarcillin/Clavulanic acid Ticarcillin Piperacillin Amikacin Trimethoprim/ Sulfamethoxazole Ciprofloxacin Meropenem		R* S R S R S S S S S S S S S S S		R* S R S R S R S S S S S S S S S S S		R* S R R S R S R S S S S S S S S		R* S R S R S R S S S S S S S S S S		R* S R S R S R S S S S S S S S S
24	Cefepime Imipenem Ticarcillin/Clavulanic acid Ticarcillin Piperacillin Amikacin Trimethoprim/ Sulfamethoxazole Ciprofloxacin Meropenem Piperacillin/Tazobactam		R* S R R S S S S S S S S S S S S S S S S S		R* S R S R S S S S S S S S S S S S S S S		R* S R S R S R S S S S S S S S S S S S S S		R* S R R S S S S S S S S S S S S S S S S S S		R* S R R S S S S S S S S S S S S S
24	CefepimeImipenemTicarcillin/Clavulanic acidTicarcillinPiperacillinAmikacinTrimethoprim/ SulfamethoxazoleCiprofloxacinMeropenemPiperacillin/TazobactamPiperacillin		R* S R S R S S R S R S R S S S R*		R* S R S R S S S S S S R S R R R S S R		R* S R R S R S R S S S S S S S R		R* S R S R S S S S S R S S S S R S R R R		R* S R S R S S S S S R S S S R R
24	Cefepime Imipenem Ticarcillin/Clavulanic acid Ticarcillin Piperacillin Amikacin Trimethoprim/ Sulfamethoxazole Ciprofloxacin Meropenem Piperacillin/Tazobactam Piperacillin		R* S R R S S S S S S S S S S S S S S S		R* S R S R S S R S S S S S S S I		R* S R S R S S S S S S S S S S I		R* S R R S R S R S S R S S S S S I		R* S R R S R S R S S R S S S S I

Cefepime	≤1	R*	≥64	R	≥64	R	≥64	R	≥64	R
Aztreonam	≤1	R*	≥64	R	≥64	R	≥64	R	≥64	R
Ciprofloxacin	2	R*	≥4	R	≥4	R	≥4	R	≥4	R
Minocycline	2	S	≤1	S	≤1	S	≤1	S	≤1	S
Ticarcillin	≥128	R	≥128	R	≥128	R	≥128	R	≥128	R
Ticarcillin/Clavnic acid	16	S	16	S	16	S	16	S	16	S
Meropenem	≤ 0.25	S	≤ 0.25	S	\leq 0.25	S	≤ 0.25	S	\leq 0.25	S
Imipenem	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S	≤ 0.25	S
Amikacin	≤ 2	S	≤ 2	S	≤ 2	S	≤ 2	S	≤ 2	S
Trimethoprim/ Sulfamethoxazole	≥ 320	R	≤ 20	S						

S.NO: number of sample isolates, (MIC) = minimum inhibitory concentration, S= bacterial susceptibility to given antibiotics, R= bacterial resistant to given antibiotics, I=Intermediate, (*) = (AES) modified, meaning Advanced Expert System modification

4.5. Investigation the effect of Magnetic Field on Biochemical Properties of Bacteria

The isolated strains of *E.coli* used in the current investigation were identified using the VITEK test technique showed in table (4-3); the isolated all strains of *E.coli* were SUCCINATE alkalinization (SUCT) test altered after exposed to different power of magnetic field. Also L-LACTATE alkalinization(ILATk) test an apparent recognized characteristic to distinguish among samples and after treatment with (0.04, 0.08, 0.12 and 0.16T) powers of magnetics converted with compared to untreated sample (negative control), however,theTyrosineArylamidase(TyrA)test,alpha-galactosidase(AGAL)test, beta-glucuronidase(BGUR)test, L-Proline A (ProA)test, and O/129 Resistance (O129R) test fermentation converted after treated to magnetic field with compared to control.

Table (4-3) presented the result of sugars fermentation metabolism in E. coli.

S. No.	TEST	Mnemonic	Before treatment	After treatment 0.04T	After treatment 0.08T	After treatment 0.12T	After treatment 0.16T
	L-LACTATE alkalinization	ILATk	-	+	+	+	+
	Tyrosine ARYLAMIDASE	TyrA	-	+	-	-	-
	ALPHA-GALACTOSIDASE	AGAL	-	+	-	+	+
	SUCCINATE alkalinization	SUCT	-	+	+	+	+
	O/129RESISTANCE (comp. vibrio.)	O129R	+	+	+	+	+
	D-MALTOSE	dMAL	+	+	+	+	+
	ORNITHINE DECARBOXYLASE	ODC	-	-	-	-	-
	D-MANNITOL	dMAN	+	+	+	+	+
	D-TREHALOSE	dTRE	+	+	+	+	+
1	LYSINE DECARBOXYLASE	LDC	+	+	+	+	+
1	D-GLUCOSE	dGLU	+	+	+	+	+
	D-MANNOSE	dMNE	+	+	+	+	+
	COUMARATE	CMT	+	+	+	+	+
	FERMENTATION/ GLUCOSE	OFF	+	+	+	+	+
	BETA-GLUCURONIDASE	BGUR	+	+	+	+	+
	GAMMA-GLUTAMYL TRANSFERASE	GGT	-	-	-	-	-
	D-SORBITOL	dSOR	+	+	+	+	+
	L-Proline ARYLAMIDASE	ProA	-	-	-	-	-
	SACCHAROSE/SUCROSE	SAC	-	-	-	-	
	PHOSPHATASE	PHOS	-	-	-	-	
	L-Proline ARYLAMIDASE	ProA	-	-	+	+	+
	SACCHAROSE/SUCROSE	SAC	-	+	+	+	+
	L-LACTATE alkalinization	ILATk	-	-	+	+	+
2	SUCCINATE alkalinization	SUCT	-	-	+	+	+
	Tyrosine ARYLAMIDASE	TyrA	-	-	+	+	+
	ALPHA-GALACTOSIDASE	AGAL	-	-	+	+	+

	BETA-GLUCURONIDASE	BGUR	+	+	-	-	+
	Tyrosine ARYLAMIDASE	TyrA	-	-	-	-	-
	ALPHA-GALACTOSIDASE	AGAL	-	-	-	-	-
	O/129RESISTANCE (comp. vibrio.)	BETA-GLUCURONIDASE BGUR + + + - + Tyrosine ARYLAMIDASE TyrA - <td>+</td>	+				
	D-MALTOSE		+				
	ORNITHINE DECARBOXYLASE		+				
	D-MANNITOL						
	D-TREHALOSE						
2	LYSINE DECARBOXYLASE		+				
4	D-GLUCOSE	dGLU	+	+	+	+	+
	D-MANNOSE	dMNE	+	+	+	+	+
	COUMARATE	CMT	+	+	+	+	+
	FERMENTATION/ GLUCOSE	ETA-GLUCURONIDASE BGUR + + + - - yrosine ARYLAMIDASE TyrA - - - - - PHA-GALACTOSIDASE AGAL - - - - - RESISTANCE (comp. vibrio.) O129R + + + + + D-MALTOSE dMAL + + + + + + D-MALTOSE dMAL + + + + + + D-MANNITOL dMAN + + + + + + D-MANNOSE dTRE + + + + + + + D-GLUCOSE dGLU +<	+				
	BETA-GLUCURONIDASE		+				
3 BETA-GLUCURONIDASE BGUR + + + Tyrosine ARYLAMIDASE TyrA ALPHA-GALACTOSIDASE AGAL O/129RESISTANCE (comp. vibrio.) O129R + + + D-MALTOSE dMAL + + + D-MALTOSE dMAL + + + ORNITHINE DECARBOXYLASE ODC + + + D-MANNITOL dMAN + + + D-TREHALOSE dTRE + + + LYSINE DECARBOXYLASE LDC + + + D-GLUCOSE dGLU + + + COUMARATE CMT + + + FERMENTATION/GLUCOSE OFF + + + BETA-GLUCURONIDASE BGUR + + H- D-DSORBITOL dSOR + + + SUCCINATE alkalinization ILATK - + SUCCINATE alkalinization SUCT - + Tyrosine ARYLAMIDASE TyrA - + ALPHA-GALACTOSIDASE AGAL - + O/129RESISTANCE (comp. vibrio.) O129R + + D-MALTOSE dMAL + + D-MALTOSE dMAL + + COUMARTE CMT - + SUCCINATE alkalinization SUCT - + Tyrosine ARYLAMIDASE TYRA - + ALPHA-GALACTOSIDASE AGAL - + O/129RESISTANCE (comp. vibrio.) O129R + + D-MALTOSE dMAL + + D-MALTOSE MMAL + + BETA-GULCURONIDASE BGUR + + D-MALTOSE MMAL + + D-MALTOSE MM	-	-	-				
	D-SORBITOL	dSOR	+	+	+	+	+
	PHOSPHATASE	PHOS	-	-	+ + + + + + + + + +	-	
	L-LACTATE alkalinization	Image: Sine ARYLAMIDASE TyrA - D-GLUCOSEG	+				
	SUCCINATE alkalinization		+				
	Tyrosine ARYLAMIDASE		+				
	ALPHA-GALACTOSIDASE		+				
	O/129RESISTANCE (comp. vibrio.)		+				
	Tyrosine ARYLAMIDASE TyrA - - - - ALPHA-GALACTOSIDASE AGAL - - - - O/129RESISTANCE (comp. vibrio.) O129R + + + + + D-MALTOSE dMAL + + + + + ORNITHINE DECARBOXYLASE ODC + + + + + D-MANNITOL dMAN + + + + + D-TREHALOSE dTRE + + + + + D-GLUCOSE dGCL + + + + + D-GLUCOSE dGCL + + + + + D-MANNOSE dMNE + + + + + COUMARATE CMT + + + + + FERMENTATION (GLUCOSE OFF + + + + + GAMA-GUTAMYL TRANSFERASE	+					
		-	-				
3	D-MANNITOL	dMAN	+	+	+	+	+
5	D-TREHALOSE	dTRE	+	+	+	+	+
3 CONVITING DECARDOATLASE ODC - D-MANNITOL dMAN + D-TREHALOSE dTRE + LYSINE DECARBOXYLASE LDC + D-GLUCOSE dGLU + D-MANNIOSE dMNE	LYSINE DECARBOXYLASE	LDC	+	+	+	+	+
	+	+	+	+	+		
	D-MANNOSE	dMNE	+	+	+	+	+
	COUMARATE	CMT	+	+	+	+	+
BETA-GLUCOSE O-MANNOSE COUMARATE FERMENTATION/ GLUCOSE BETA-GLUCURONIDASE GAMMA-GLUTAMYL TRANSFERASE D-SORBITOL PHOSPHATASE L-LACTATE alkalinization SUCCINATE alkalinization Tyrosine ARYLAMIDASE ALPHA-GALACTOSIDASE O/129RESISTANCE (comp. vibrio.) D-MALTOSE ORNITHINE DECARBOXYLASE D-MANNITOL D-TREHALOSE LYSINE DECARBOXYLASE D-MANNOSE COUMARATE FERMENTATION/ GLUCOSE BETA-GLUCURONIDASE GAMMA-GLUTAMYL TRANSFERASE	OFF	+	+	+	+	+	
	BETA-GLUCURONIDASE	BGUR	+	+	+	+	+
2 3	GAMMA-GLUTAMYL TRANSFERASE	GGT	-	-	-	-	-

	D-SORBITOL	dSOR	+	+	+	+	+
	L-Proline ARYLAMIDASE	ProA	-	-	-	-	-
	SACCHAROSE/SUCROSE	SAC	-	-	-	-	-
	PHOSPHATASE	PHOS	-	-	-	-	-
	L-LACTATE alkalinization	ILATk	-	+	+	-	+
	SUCCINATE alkalinization	SUCT	-	+	-	+	+
	Tyrosine ARYLAMIDASE	TyrA	-	-	-	-	+
	ALPHA-GALACTOSIDASE	AGAL	-	+	-	-	+
	O/129RESISTANCE (comp. vibrio.)	O129R	+	+	+	+	+
	D-MALTOSE	dMAL	+	+	+	+	+
	ORNITHINE DECARBOXYLASE	ODC	+	+	+	+	+
	D-MANNITOL	dMAN	+	+	+	+	+
	D-TREHALOSE	dTRE	+	+	+	+	+
Λ	LYSINE DECARBOXYLASE	LDC	+	+	+	+	+
4	D-GLUCOSE	dGLU	+	+	+	+	+
	D-MANNOSE	dMNE	+	+	+	+	+
	COUMARATE	CMT	+	+	+	+	+
D-MANNOSEdMNE++COUMARATECMT++FERMENTATION/ GLUCOSEOFF++BETA-GLUCURONIDASEBGUR++GAMMA-GLUTAMYL TRANSFERASEGGTD-SORBITOLdSOR++	FERMENTATION/ GLUCOSE	OFF	+	+	+	+	+
	BETA-GLUCURONIDASE	BGUR	+	+	+	+	+
	-	-	-				
	D-SORBITOL	dSOR	+	+	+	+	+
	L-Proline ARYLAMIDASE	ProA - - - - - SAC - - - - - - PHOS - - - - - - ILATk - + + + - - - SUCT - + + + + + - AGAL - + + - - - - Ol29R + + + + + + - ODC + + + + + + - dMAN + + + + + + - dMAN + + + + + + - dMAN + + + + + + - dGLU + + + + + + - GGT	-				
	SACCHAROSE/SUCROSE	SAC	-	-	-	-	-
	PHOSPHATASE	PHOS	-	-	-	-	-
	L-LACTATE alkalinization	ILATk	+	-	-	-	-
	L-Proline ARYLAMIDASE	ProA	-	-	-	-	+
	SUCCINATE alkalinization	SUCT	+	+	+	+	+
	Tyrosine ARYLAMIDASE	TyrA	+	+	+	+	+
5	ALPHA-GALACTOSIDASE	AGAL	+	+	+	+	+
	O/129RESISTANCE (comp. vibrio.)	O129R	+	+	+	+	+
	D-MALTOSE	dMAL	+	+	+	+	+
	ORNITHINE DECARBOXYLASE	ODC	+	+	+	+	+
L-Proline ARYLAMIDASEProA-SACCHAROSE/SUCROSESAC-PHOSPHATASEPHOS-L-LACTATE alkalinizationILATk-SUCCINATE alkalinizationSUCT-Tyrosine ARYLAMIDASETyrA-ALPHA-GALACTOSIDASEAGAL-O/129RESISTANCE (comp. vibrio.)O129R+D-MALTOSEdMAL+ORNITHINE DECARBOXYLASEODC+D-MANNITOLdMAN+D-TREHALOSEdTRE+LYSINE DECARBOXYLASELDC+D-GLUCOSEdGLU+D-MANNOSEdMNE+COUMARATECMT+FERMENTATION/ GLUCOSEOFF+BETA-GLUCURONIDASEBGUR+COUMARATEProA-FORBITOLdSOR+L-Proline ARYLAMIDASEProA-SACCHAROSE/SUCROSESAC-PHOSPHATASEPHOS-PHOSPHATASEProA-SUCCINATE alkalinizationILATk+L-Proline ARYLAMIDASEProA-SUCCINATE alkalinizationSUCT+Tyrosine ARYLAMIDASETyrA+ALPHA-GALACTOSIDASEAGAL+O/129RESISTANCE (comp. vibrio.)O129R+D-MANNITOLdMAL+ORNITHINE DECARBOXYLASEODC+D-MANNITOLdMAN+	+	+	+	+			

D-TREHALOSE	dTRE	+	+	+	+	+
LYSINE DECARBOXYLASE	LDC	+	+	+	+	+
D-GLUCOSE	dGLU	+	+	+	+	+
D-MANNOSE	dMNE	+	+	+	+	+
COUMARATE	CMT	+	+	+	+	+
FERMENTATION/ GLUCOSE	OFF	+	+	+	+	+
BETA-GLUCURONIDASE	BGUR	+	+	+	+	+
GAMMA-GLUTAMYL-TRANSFERASE	GGT	-	-	-	-	-
D-SORBITOL	dSOR	+	+	+	+	+
SACCHAROSE/SUCROSE	SAC	+	+	+	+	+
PHOSPHATASE	PHOS	-	-	-	-	-
O/129 RESISTANCE(comp. vibrio.)	O129R	+	-	+	+	-
SUCCINATE alkalinization	SUCT	+	-	-	+	-
ALPHA-GALACTOSIDASE	AGAL	+	-	-	+	-
FERMENTATION/ GLUCOSE	OFF	-	-	-	+	-
L-LACTATE alkalinization	ILATk	-	-	-	-	-
Tyrosine ARYLAMIDASE	TyrA	-	-	-	-	-
D-MALTOSE	dMAL	+	+	+	+	+
ORNITHINE DECARBOXYLASE	ODC	-	-	-	-	-
D-MANNITOL	dMAN	+	+	+	+	+
D-TREHALOSE	dTRE	+	+	+	+	+
LYSINE DECARBOXYLASE	LDC	+	+	+	+	+
D-GLUCOSE	dGLU	+	+	+	+	+
D-MANNOSE	dMNE	+	+	+	+	+
COUMARATE	CMT	+	+	+	+	+
BETA-GLUCURONIDASE	BGUR	+	+	+	+	+
GAMMA-GLUTAMYL-TRANSFERASE	GGT	-	-	-	-	-
D-SORBITOL	dSOR	+	+	+	+	+
L-Proline ARYLAMIDASE	ProA	-	-	-	-	-
SACCHAROSE/SUCROSE	SAC	-	_	-	_	-
PHOSPHATASE	PHOS	-	-	-	-	-
L-LACTATE alkalinization	ILATk	-	+	+	+	+

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O/129RESISTANCE (comp. vibrio.)	O129R	_	+	+	+	+
BETA-GLUCURONIDASE	BGUR	+	+	_	_	_
SUCCINATE alkalinization	SUCT	+	+	+	+	+
Tyrosine ARYLAMIDASE	TyrA	+	+	+	+	+
ALPHA-GALACTOSIDASE	AGAL	+	+	+	+	+
D-MALTOSE	dMAL	+	+	+	+	+
ORNITHINE DECARBOXYLASE	ODC	+	+	+	+	+
D-MANNITOL	dMAN	+	+	+	+	+
D-TREHALOSE	dTRE	+	+	+	+	+
LYSINE DECARBOXYLASE	LDC	+	+	+	+	+
D-GLUCOSE	dGLU	+	+	+	+	+
D-MANNOSE	dMNE	+	+	+	+	+
COUMARATE	CMT	+	+	+	+	+
FERMENTATION/ GLUCOSE	OFF	+	+	+	+	+
GAMMA-GLUTAMYL TRANSFERASE	GGT	-	-	-	-	-
D-SORBITOL	dSOR	+	+	+	+	+
L-Proline ARYLAMIDASE	ProA	+	+	+	+	+
SACCHAROSE/SUCROSE	SAC	+	+	+	+	+
PHOSPHATASE	PHOS	-	-	-	-	-
L-LACTATE alkalinization	ILATk	+	-	+	+	+
O/129 RESISTANCE (comp. vibrio.)	O129R	+	-	-	-	-
D-MALTOSE	dMAL	+	-	+	+	+
ORNITHINE DECARBOXYLASE	ODC	+	-	+	+	+
D-MANNITOL	dMAN	+	-	+	+	+
D-TREHALOSE	dTRE	+	-	+	+	+
SUCCINATE alkalinization	SUCT	+	-	+	+	+
LYSINE DECARBOXYLASE	LDC	+	-	+	+	+
D-GLUCOSE	dGLU	+	-	+	+	+
D-MANNOSE	dMNE	+	-	+	+	+
Tyrosine ARYLAMIDASE	TyrA	+	-	+	+	+
COUMARATE	CMT	+	-	+	+	+

	FERMENTATION/ GLUCOSE	OFF	+	-	+	+	+
	BETA-GLUCURONIDASE	BGUR	-	+	+	+	+
	GAMMA-GLUTAMYL TRANSFERASE	GGT	+	-	+	+	-
	D-SORBITOL	IENTATION/ GLUCOSE OFF + - + + + .GUUCURONIDASE BGUR - + + + + + A.GUTAMYL TRANSFERASE GGT + - + + + + RBITOL dSOR + - + + + + + A.GUTAMYL TRANSFERASE GGT - - + + + + RBITOL dSOR + - - + + + + A.GAUACTOSIDASE AGAL - - + + + + + PHATASE PHOS - - +	+				
 FERMENTA' BETA-GLUC GAMMA-GLU D-SORBITOI L-Proline AR ALPHA-GAI PHOSPHATA SACCHARO O/129RESIS' SUCCINATE LYSINE DEC FERMENTA' L-LACTATE BETA-GLUC Tyrosine AR' ALPHA-GAI D-MALTOSI ORNITHINE D-MANNITC D-TREHALC D-GLUCOSE D-MANNOS COUMARAT GAMMA-GI D-SORBITOI L-Proline AR SACCHARO PHOSPHATA SUCCINATE Tyrosine AR' ALPA-GAI D-MANNITC D-TREHALC D-GLUCOSE D-MANNOS COUMARAT GAMMA-GI D-SORBITOI L-Proline AR SACCHARO PHOSPHATA SUCCINATE Tyrosine AR' 10 	L-Proline ARYLAMIDASE	ProA	-	-	+	+	+
	IPERMENTATION OLOCOSE OFF + - + + BETA-GLUCURONIDASE BGUR - + + + GAMMA-GLUTAMYL TRANSFERASE GGT + - + + DSORBITOL dSOR + - + + + L-Proline ARYLAMIDASE ProA - - + + + ALPHA-GALACTOSIDASE AGAL - - + + + PHOS - - + + + + + O/129RESISTANCE (comp. vibrio.) O129R + - + + + SUCCINATE alkalinization SUCT + - + + + FERMENTATION/ GLUCOSE OFF + + + + + FERA-GLUCURONIDASE BGUR - - - - - Tyrosine ARYLAMIDASE TyrA + + + + + + <td>+</td>	+					
	PHOSPHATASE	PHOS	-	-	+	-	-
	SACCHAROSE/SUCROSE	SAC	-	-	-	-	-
	O/129RESISTANCE (comp. vibrio.)	O129R	+	-	+	+	-
	SUCCINATE alkalinization	SUCT	+	-	+	+	+
	LYSINE DECARBOXYLASE	LDC	+	-	+	+	+
	FERMENTATION/ GLUCOSE	OFF	+	+	+	+	-
	L-LACTATE alkalinization	ILATk	-	-	-	-	-
	BETA-GLUCURONIDASE	BGUR	-	-	-	-	-
	Tyrosine ARYLAMIDASE	TyrA	+	+	+	+	+
	ALPHA-GALACTOSIDASE	AGAL	+	+	+	+	+
	D-MALTOSE	dMAL	+	+	+	+	+
0	ORNITHINE DECARBOXYLASE	ODC	-	-	-	-	-
9	D-MANNITOL	dMAN	+	+	+	+	+
	D-TREHALOSE	dTRE	+	+	+	+	+
	D-GLUCOSE	dGLU	+	+	+	+	+
	D-MANNOSE	dMNE	+	+	+	+	+
	COUMARATE	CMT	+	+	+	+	+
	GAMMA-GLUTAMYL-TRANSFERASE	GGT	-	-	-	-	-
	D-SORBITOL	dSOR	+	+	+	+	+
	L-Proline ARYLAMIDASE	ProA	-	-	-	-	-
	SACCHAROSE/SUCROSE	SAC	-	-	-	-	-
	PHOSPHATASE	PHOS	-	-	-	-	-
	SUCCINATE alkalinization	SUCT	-	+	+	+	+
	Tyrosine ARYLAMIDASE	TyrA	-	+	-	-	+
10	ELLMAN	ELLM	+	-	-	-	-
	DOR I <thi< th=""> I <thi< th=""> <thi< th=""></thi<></thi<></thi<>	-					
10	L-LACTATE alkalinization	ILATk	+	+	+	+	+

BETA-GLUCURONIDASE	BGUR	+	+	+	+	+
ALPHA-GALACTOSIDASE	AGAL	+	+	+	+	+
D-MALTOSE	dMAL	+	+	+	+	+
D-MANNITOL	dMAN	+	+	+	+	+
D-TREHALOSE	dTRE	+	+	+	+	+
D-GLUCOSE	dGLU	+	+	+	+	+
D-MANNOSE	dMNE	+	+	+	+	+
COUMARATE	CMT	+	+	+	+	+
GAMMA-GLUTAMYL-TRANSFERASE	GGT	-	-	-	-	-
D-SORBITOL	dSOR	+	+	+	+	+
L-Proline ARYLAMIDASE	ProA	-	-	-	-	-
SACCHAROSE/SUCROSE	SAC	-	-	-	-	-
ADONITOL	ADO	-	+	+	+	+
PHOSPHATASE	PHOS	-	-	-	-	-
L-Proline ARYLAMIDASE	ProA	-	+	+	+	+
L-LACTATE assimilation	ILATa	+	-	+	+	-
BETA-GLUCURONIDASE	BGUR	-	-	-	+	+
L-LACTATE alkalinization	ILATk	+	+	+	+	+
BETA-GLUCURONIDASE	BGUR	+	+	+	+	+
Tyrosine ARYLAMIDASE	TyrA	-	-	-	-	-
ALPHA-GALACTOSIDASE	AGAL	+	+	+	+	+
D-MALTOSE	dMAL	+	+	+	+	+
ORNITHINE DECARBOXYLASE	ODC	-	-	-	-	-
D-MANNITOL	dMAN	+	+	+	+	+
D-TREHALOSE	dTRE	+	+	+	+	+
D-GLUCOSE	dGLU	+	+	+	+	+
D-MANNOSE	dMNE	+	+	+	+	+
COUMARATE	CMT	+	+	+	+	+
GAMMA-GLUTAMYL TRANSFERASE	GGT	-	-	-	-	-
D-SORBITOL	dSOR	+	+	+	+	+
L-Proline ARYLAMIDASE	ProA	-	-	-	-	-
SACCHAROSE/SUCROSE	SAC	-	-	-	-	-
PHOSPHATASE	PHOS	-	-	-	-	-

	L-LACTATE alkalinization	ILATk	-	-	-	-	+
	BETA-GLUCURONIDASE	BGUR	+	+	+	+	+
	Tyrosine ARYLAMIDASE	TyrA	+	-	-	+	+
	ALPHA-GALACTOSIDASE	AGAL	+	+	+	+	+
	D-MALTOSE	dMAL	+	+	+	+	+
	ORNITHINE DECARBOXYLASE	ODC	+	+	+	+	+
	D-MANNITOL	dMAN	+	+	+	+	+
12	D-TREHALOSE	dTRE	+	+	+	+	+
	D-GLUCOSE	dGLU	+	+	+	+	+
	D-MANNOSE	dMNE	+	+	+	+	+
	COUMARATE	CMT	+	+	+	+	+
	GAMMA-GLUTAMYL-TRANSFERASE	GGT	-	-	-	-	-
	D-SORBITOL	dSOR	+	+	+	+	+
	L-Proline ARYLAMIDASE	ProA	-	-	-	-	-
	SACCHAROSE/SUCROSE	SAC	+	+	+	+	+
	PHOSPHATASE	PHOS	-	-	-	-	-
	SUCCINATE alkalinization	SUCT	-	-	+	+	+
	Tyrosine ARYLAMIDASE	TyrA	-	+	-	-	+
	ELLMAN	ELLM	+	-	-	-	-
	5-KETO-D-GLUCONATE	5KG	+	-	-	-	-
	L-LACTATE alkalinization	ILATk	+	+	+	+	+
	BETA-GLUCURONIDASE	BGUR	+	+	+	+	+
	ALPHA-GALACTOSIDASE	AGAL	+	+	+	+	+
	D-MALTOSE	dMAL	+	+	+	+	+
13	D-MANNITOL	dMAN	+	+	+	+	+
15	D-TREHALOSE	dTRE	+	+	+	+	+
	SUCCINATE alkalinization	SUCT	-	+	+	+	+
	D-GLUCOSE	dGLU	+	+	+	+	+
	D-MANNOSE	dMNE	+	+	+	+	+
	COUMARATE	CMT	+	+	+	+	+
	GAMMA-GLUTAMYL-TRANSFERASE	GGT	-	-	-	-	-
	D-SORBITOL	dSOR	+	+	+	+	+
	L-Proline ARYLAMIDASE	ProA	-	-	-	-	-

	SACCHAROSE/SUCROSE	SAC	-	-	-	-	-
	ADONITOL	ADO	-	+	+	+	+
	PHOSPHATASE	PHOS	-	-	-	-	-
	SUCCINATE alkalinization	SUCT	+	-	+	+	+
	LYSINE DECARBOXYLASE	LDC	+	-	+	+	+
	FERMENTATION/ GLUCOSE	OFF	+	+	+	+	-
	L-LACTATE alkalinization	ILATk	-	-	-	-	-
	BETA-GLUCURONIDASE	BGUR	-	-	-	-	-
	Tyrosine ARYLAMIDASE	TyrA	+	+	+	+	+
	ALPHA-GALACTOSIDASE	AGAL	+	+	+	+	+
	D-MALTOSE	dMAL	+	+	+	+	+
	ORNITHINE DECARBOXYLASE	ODC	-	-	-	-	-
14	D-MANNITOL	dMAN	+	+	+	+	+
14	D-TREHALOSE	dTRE	+	+	+	+	+
	D-GLUCOSE	dGLU	+	+	+	+	+
	D-MANNOSE	dMNE	+	+	+	+	+
	COUMARATE	CMT	+	+	+	+	+
	GAMMA-GLUTAMYL-TRANSFERASE	GGT	-	-	-	-	-
	D-SORBITOL	dSOR	+	+	+	+	+
	L-Proline ARYLAMIDASE	ProA	-	-	-	-	-
	SACCHAROSE/SUCROSE	SAC	-	-	-	-	-
	PHOSPHATASE	PHOS	-	-	-	-	-
	O/129RESISTANCE (comp. vibrio.)	O129R	+	-	+	+	-
	L-LACTATE assimilation	ILATa	+	-	+	+	-
	BETA-GLUCURONIDASE	BGUR	-	-	-	+	+
	L-LACTATE alkalinization	ILATk	+	+	+	+	+
	BETA-GLUCURONIDASE	BGUR	+	+	+	+	+
15	Tyrosine ARYLAMIDASE	TyrA	-	-	-	-	-
13	ALPHA-GALACTOSIDASE	AGAL	+	+	+	+	+
	D-MALTOSE	dMAL	+	+	+	+	+
	ORNITHINE DECARBOXYLASE	ODC	-	-	-	-	-
	D-MANNITOL	dMAN	+	+	+	+	+
	D-TREHALOSE	dTRE	+	+	+	+	+

	D-GLUCOSE	dGLU	+	+	+	+	+
	D-MANNOSE	dMNE	+	+	+	+	+
	COUMARATE	CMT	+	+	+	+	+
	GAMMA-GLUTAMYL TRANSFERASE	GGT	-	-	-	-	-
	D-SORBITOL	dSOR	+	+	+	+	+
	L-Proline ARYLAMIDASE	ProA	-	-	-	-	-
	SACCHAROSE/SUCROSE	SAC	-	-	-	-	-
	PHOSPHATASE	PHOS	-	-	-	-	-
	L-Proline ARYLAMIDASE	ProA	-	+	+	+	+
	L-LACTATE alkalinization	ILATk	-	-	-	-	+
16	BETA-GLUCURONIDASE	BGUR	+	+	+	+	+
	Tyrosine ARYLAMIDASE	TyrA	+	-	-	+	+
	ALPHA-GALACTOSIDASE	AGAL	+	+	+	+	+
	D-MALTOSE	dMAL	+	+	+	+	+
	ORNITHINE DECARBOXYLASE	ODC	+	+	+	+	+
	D-MANNITOL	dMAN	+	+	+	+	+
	D-TREHALOSE	dTRE	+	+	+	+	+
16	D-GLUCOSE	dGLU	+	+	+	+	+
	D-MANNOSE	dMNE	+	+	+	+	+
	COUMARATE	CMT	+	+	+	+	+
	GAMMA-GLUTAMYL-TRANSFERASE	GGT	-	-	-	-	-
	D-SORBITOL	dSOR	+	+	+	+	+
	L-Proline ARYLAMIDASE	ProA	-	-	-	-	-
	SACCHAROSE/SUCROSE	SAC	+	+	+	+	+
	PHOSPHATASE	PHOS	-	-	-	-	-
	SUCCINATE alkalinization	SUCT	-	-	+	+	+
	L-LACTATE alkalinization	ILATk	+	-	+	+	+
	O/129 RESISTANCE (comp. vibrio.)	O129R	+	-	-	-	-
	D-MALTOSE	dMAL	+	-	+	+	+
17	ORNITHINE DECARBOXYLASE	ODC	+	-	+	+	+
	D-MANNITOL	dMAN	+	-	+	+	+
16	D-TREHALOSE	dTRE	+	-	+	+	+
	SUCCINATE alkalinization	SUCT	+	-	+	+	+

LYSINE DECARBOXYLASE	LDC	+	-	+	+	+
D-GLUCOSE	dGLU	+	-	+	+	+
D-MANNOSE	dMNE	+	-	+	+	+
Tyrosine ARYLAMIDASE	TyrA	+	-	+	+	+
COUMARATE	CMT	+	-	+	+	+
FERMENTATION/ GLUCOSE	OFF	+	-	+	+	+
BETA-GLUCURONIDASE	BGUR	-	+	+	+	+
GAMMA-GLUTAMYL TRANSFERASE	GGT	+	-	+	+	-
D-SORBITOL	dSOR	+	-	+	+	+
L-Proline ARYLAMIDASE	ProA	-	-	+	+	+
ALPHA-GALACTOSIDASE	AGAL	-	-	+	+	+
PHOSPHATASE	PHOS	-	-	+	-	-
SACCHAROSE/SUCROSE	SAC	-	-	-	-	-
SUCCINATE alkalinization	SUCT	+	-	-	+	-
ALPHA-GALACTOSIDASE	AGAL	+	-	-	+	-
FERMENTATION/ GLUCOSE	OFF	-	-	-	+	-
L-LACTATE alkalinization	ILATk	-	-	-	-	-
Tyrosine ARYLAMIDASE	TyrA	-	-	-	-	-
D-MALTOSE	dMAL	+	+	+	+	+
ORNITHINE DECARBOXYLASE	ODC	-	-	-	-	-
D-MANNITOL	dMAN	+	+	+	+	+
D-TREHALOSE	dTRE	+	+	+	+	+
LYSINE DECARBOXYLASE	LDC	+	+	+	+	+
D-GLUCOSE	dGLU	+	+	+	+	+
D-MANNOSE	dMNE	+	+	+	+	+
COUMARATE	CMT	+	+	+	+	+
BETA-GLUCURONIDASE	BGUR	+	+	+	+	+
GAMMA-GLUTAMYL TRANSFERASE	GGT	-	-	-	-	-
D-SORBITOL	dSOR	+	+	+	+	+
L-Proline ARYLAMIDASE	ProA	-	-	-	-	-
SACCHAROSE/SUCROSE	SAC	-	-	-	-	-
PHOSPHATASE	PHOS	-	-	-	-	-

	O/129 RESISTANCE(comp. vibrio.)	O129R	+	-	+	+	-
	L-LACTATE alkalinization	ILATk	-	+	+	+	+
O/129 RESISTANCE(comp. viL-LACTATE alkalinizationO/129RESISTANCE (comp. viBETA-GLUCURONIDASESUCCINATE alkalinizationTyrosine ARYLAMIDASEALPHA-GALACTOSIDASED-MALTOSEORNITHINE DECARBOXYLAD-MANNITOLD-TREHALOSELYSINE DECARBOXYLASED-GLUCOSED-MANNOSECOUMARATEFERMENTATION/ GLUCOSEGAMMA-GLUTAMYL-TRAND-SORBITOLL-Proline ARYLAMIDASESACCHAROSE/SUCROSEPHOSPHATASEL-LACTATE alkalinizationTyrosine ARYLAMIDASEALPHA-GALACTOSIDASE	O/129RESISTANCE (comp. vibrio.)	O129R	-	+	+	+	+
	BETA-GLUCURONIDASE	BGUR	+	+	-	-	-
	SUCCINATE alkalinization	SUCT	+	+	+	+	+
	Tyrosine ARYLAMIDASE	TyrA	+	+	+	+	+
	ALPHA-GALACTOSIDASE	AGAL	+	+	+	+	+
	D-MALTOSE	dMAL	+	+	+	+	+
	ORNITHINE DECARBOXYLASE	ODC	+	+	+	+	+
	D-MANNITOL	dMAN	+	+	+	+	+
	D-TREHALOSE	dTRE	+	+	+	+	+
	LYSINE DECARBOXYLASE	LDC	+	+	+	+	+
	D-GLUCOSE	dGLU	+	+	+	+	+
	D-MANNOSE	dMNE	+	+	+	+	+
	COUMARATE	CMT	+	+	+	+	+
	FERMENTATION/ GLUCOSE	OFF	+	+	+	+	+
	GAMMA-GLUTAMYL-TRANSFERASE	GGT	-	-	-	-	-
	D-SORBITOL	dSOR	+	+	+	+	+
	L-Proline ARYLAMIDASE	ProA	+	+	+	+	+
	SACCHAROSE/SUCROSE	SAC	+	+	+	+	+
	PHOSPHATASE	PHOS	-	-	-	-	-
	L-LACTATE alkalinization	ILATk	-	+	+	+	+
	Tyrosine ARYLAMIDASE	TyrA	-	+	-	-	-
	ALPHA-GALACTOSIDASE	AGAL	-	+	-	+	+
	SUCCINATE alkalinization	SUCT	-	+	+	+	+
	O/129RESISTANCE (comp. vibrio.)	O129R	+	+	+	+	+
20	D-MALTOSE	dMAL	+	+	+	+	+
20	ORNITHINE DECARBOXYLASE	ODC	-	-	-	-	-
	D-MANNITOL	dMAN	+	+	+	+	+
	D-TREHALOSE	dTRE	+	+	+	+	+
	LYSINE DECARBOXYLASE	LDC	+	+	+	+	+
	D-GLUCOSE	dGLU	+	+	+	+	+
	D-MANNOSE	dMNE	+	+	+	+	+

	COUMARATE	CMT	+	+	+	+	+
	FERMENTATION/ GLUCOSE	OFF	+	+	+	+	+
	BETA-GLUCURONIDASE	ATECMT++IATION/ GLUCOSEOFF++IATION/ GLUCOSEOFF++UCURONIDASEBGUR++IAUTAMYL TRANSFERASEGGTIOLdSOR++ARYLAMIDASEProAROSE/SUCROSESACTASEPHOSTASEPHOSRYLAMIDASETyrA-+RYLAMIDASETyrA-+RYLAMIDASEAGAL-+TE alkalinizationSUCT-+TE alkalinizationSUCT-+TE alkalinizationSUCT-+TOLMAL++NEDCARBOXYLASEODC-TOLdMAN++VECARBOXYLASELDC+SEdGLU++SEdGLU++SEGGTTOL/ONDASEBGUR++ATECMT++ATECMT++ATECMT++ATECMT++ATECMT++ATECMT++ATECMT++ATECMT++ATECMT++ATECMT++ATECMT++ATECM	+	+	+		
21	GAMMA-GLUTAMYL TRANSFERASE	GGT	-	-	-	-	-
	D-SORBITOL	dSOR	+	+	+	+	+
	L-Proline ARYLAMIDASE	ProA	-	-	-	-	-
	SACCHAROSE/SUCROSE	SAC	-	-	-	-	-
	PHOSPHATASE	PHOS	-	-		-	
	L-LACTATE alkalinization	ILATk	-	+	+	+	+
	Tyrosine ARYLAMIDASE	TyrA	-	+	-	-	-
	ALPHA-GALACTOSIDASE	AGAL	-	+	-	+	+
	SUCCINATE alkalinization	SUCT	-	+	+	+	+
	O/129RESISTANCE (comp. vibrio.)	O129R	+	+	+	+	+
	D-MALTOSE	dMAL	+	+	+	+	+
	ORNITHINE DECARBOXYLASE	ODC	-	-	-	-	-
	D-MANNITOL	dMAN	+	+	+	+	+
	D-TREHALOSE	dTRE	+	+	+	+	+
21	LYSINE DECARBOXYLASE	LDC	+	+	+	+	+
41	D-GLUCOSE	dGLU	+	+	+	+	+
	D-MANNOSE	dMNE	+	+	+	+	+
	COUMARATE	CMT	+	+	+	+	+
	FERMENTATION/ GLUCOSE	OFF	+	+	+	+	+
	BETA-GLUCURONIDASE	BGUR	+	+	+	+	+
	GAMMA-GLUTAMYL TRANSFERASE	GGT	-	-	-	-	-
	D-SORBITOL	dSOR	+	+	+	+	+
	L-Proline ARYLAMIDASE	ProA	-	-	-	-	-
	SACCHAROSE/SUCROSE	SAC	-	-	-	-	-
	PHOSPHATASE	PHOS	-	-	-	-	-
	L-LACTATE alkalinization	ILATk	+	-	-	-	-
	L-Proline ARYLAMIDASE	ProA	-	-		-	+
22	SUCCINATE alkalinization	SUCT	+	+	+	+	+
	Tyrosine ARYLAMIDASE	TyrA	+	+	+	+	+
	ALPHA-GALACTOSIDASE	AGAL	+	+	+	+	+

O/129RESISTANCE (comp. vibrio.)	O129R	+	+	+	+	+
D-MALTOSE	dMAL	+	+	+	+	+
ORNITHINE DECARBOXYLASE	ODC	+	+	+	+	+
D-MANNITOL	dMAN	+	+	+	+	+
D-TREHALOSE	dTRE	+	+	+	+	+
LYSINE DECARBOXYLASE	LDC	+	+	+	+	+
D-GLUCOSE	dGLU	+	+	+	+	+
D-MANNOSE	dMNE	+	+	+	+	+
COUMARATE	CMT	+	+	+	+	+
FERMENTATION/ GLUCOSE	OFF	+	+	+	+	+
BETA-GLUCURONIDASE	BGUR	+	+	+	+	+
GAMMA-GLUTAMYL TRANSFERASE	GGT	-	-	-	-	-
D-SORBITOL	dSOR	+	+	+	+	+
SACCHAROSE/SUCROSE	SAC	+	+	+	+	+
PHOSPHATASE	PHOS	-	-	-	-	-
L-LACTATE alkalinization	ILATk	-	+	+	-	+
SUCCINATE alkalinization	SUCT	-	+	-	+	+
Tyrosine ARYLAMIDASE	TyrA	-	-	-	-	+
ALPHA-GALACTOSIDASE	AGAL	-	+	-	-	+
O/129RESISTANCE (comp. vibrio.)	O129R	+	+	+	+	+
D-MALTOSE	dMAL	+	+	+	+	+
ORNITHINE DECARBOXYLASE	ODC	+	+	+	+	+
D-MANNITOL	dMAN	+	+	+	+	+
D-TREHALOSE	dTRE	+	+	+	+	+
LYSINE DECARBOXYLASE	LDC	+	+	+	+	+
D-GLUCOSE	dGLU	+	+	+	+	+
D-MANNOSE	dMNE	+	+	+	+	+
COUMARATE	CMT	+	+	+	+	+
FERMENTATION/ GLUCOSE	OFF	+	+	+	+	+
BETA-GLUCURONIDASE	BGUR	+	+	+	+	+
GAMMA-GLUTAMYL TRANSFERASE	GGT	-	-	-	-	-
D-SORBITOL	dSOR	+	+	+	+	+
L-Proline ARYLAMIDASE	ProA	-	-	-	-	-

	SACCHADOSE/SUCDOSE	SAC					
			-	-	-	-	-
	L L ACTATE alkalinization		-	-	-	-	-
	SUCCINATE alkalinization		-	+	+	+	+
			-	+	+	+	+
	ALDUA CALACTOSIDASE		-	+	+	+	+
	ALPHA-GALACIOSIDASE	AGAL	-	+	+	+	+
	0/129RESISTANCE (comp. vibrio.)	UI29R	+	+	+	+	+
	D-MALIOSE	dMAL	+	+	+	+	+
	ORNITHINE DECARBOXYLASE	ODC	-	-	-	-	-
	D-MANNITOL	dMAN	+	+	+	+	+
	D-TREHALOSE	dTRE	+	+	+	+	+
24	LYSINE DECARBOXYLASE	LDC	+	+	+	+	+
27	D-GLUCOSE	dGLU	+	+	+	+	+
	D-MANNOSE	dMNE	+	+	+	+	+
	COUMARATE	CMT	+	+	+	+	+
	FERMENTATION/ GLUCOSE	OFF	+	+	+	+	+
	BETA-GLUCURONIDASE	BGUR	+	+	+	+	+
	GAMMA-GLUTAMYL TRANSFERASE	GGT	-	-	-	-	-
	D-SORBITOL	dSOR	+	+	+	+	+
	L-Proline ARYLAMIDASE	ProA	-	-	-	-	-
	SACCHAROSE/SUCROSE	SAC	-	-	-	-	-
	PHOSPHATASE	PHOS	-	-	-	-	-
	L-Proline ARYLAMIDASE	ProA	-	-	+	+	+
	SACCHAROSE/SUCROSE	SAC	-	+	+	+	+
	L-LACTATE alkalinization	ILATk	-	-	+	+	+
	SUCCINATE alkalinization	SUCT	-	-	+	+	+
	Tyrosine ARYLAMIDASE	TyrA	-	-	+	+	+
25	ALPHA-GALACTOSIDASE	AGAL	-	-	+	+	+
	BETA-GLUCURONIDASE	BGUR	+	+	-	-	+
	Tyrosine ARYLAMIDASE	TyrA	-	-	-	-	-
	ALPHA-GALACTOSIDASE	AGAL	-	-	-	-	-
	O/129RESISTANCE (comp. vibrio.)	O129R	+	+	+	+	+
	D-MALTOSE	dMAL	+	+	+	+	+

ORNITHINE DECARBOXYLASE	ODC	+	+	+	+	+
D-MANNITOL	dMAN	+	+	+	+	+
D-TREHALOSE	dTRE	+	+	+	+	+
LYSINE DECARBOXYLASE	LDC	+	+	+	+	+
D-GLUCOSE	dGLU	+	+	+	+	+
D-MANNOSE	dMNE	+	+	+	+	+
COUMARATE	CMT	+	+	+	+	+
FERMENTATION/ GLUCOSE	OFF	+	+	+	+	+
BETA-GLUCURONIDASE	BGUR	+	+	+	+	+
GAMMA-GLUTAMYL TRANSFERASE	GGT	-	-	-	-	-
D-SORBITOL	dSOR	+	+	+	+	+
PHOSPHATASE	PHOS	-	-	-	-	-

Note: (-) a negative result, (+) a positive result

4.6. Molecular Characterizations of isolated E. coli

All isolates of *E. coli* were undergone PCR assay for more confirmation of the identity of these isolates, using *16SrRNA* gene for detection of *E. coli*, also using CTX_M , *TEM*, and *SHV* genes to determine the occurrence of ESBL producers, ESBL genes (*blaTEM*₁, *blaSHV*, and *blaCTX*-_{*M*-1}), from the process of exposure compared with unexposed (Negative control).

4.6.1. Detection of *16SrRNA* gene among *E.coli* isolates using PCR Technique

Results of molecular identification indicated that all uropathogenic *E. coli* isolates were positive for the presence of *16SrRNA* gene at 1343bp, these isolates were undergone a first run on gel electrophoresis, and the results are shown in Figure (4-3) and (4-4). Generally, all isolates (25) samples of *E. coli* (100%) unexposed and exposed samples of different magnetic powers (0.04, 0.08, 0.12 and 0.16T) demonstrated positive PCR products on gel electrophoresis for *16SrRNA* gen*e* at 1343 bp.



Figure (4-3) Agarose (1%) gel electrophoresis of PCR product of amplified *16SrRNA* gen*e* of *E.coli* isolate (amplicon with 1343bp).Group (I-VI) represent number of sample and (0) control unexposed to the magnetic field (1, 2, 3, 4) sample exposed to magnetic power (0.04, 0.08, 0.12 and 0.16T) respectively.



Figure (4-4) Agarose (1%) gel electrophoresis of PCR product of amplified 16SrRNA gene of *E.coli* isolates (amplicon with 1343bp). Group (VI-IX) represent number of sample and (0) Negative control unexposed to the magnetic field (1, 2, 3, 4) sample exposed to magnetic power (0.04, 0.08, 0.12 and 0.16T) respectively.

4.6.2. Detection of CTX_{M-1} gene among *E.coli* isolates using PCR technique

Outcomes of molecular identification shown in Figure (4-5) indicated that (23) of (25) samples of uropathogenic *E.coli* isolates were positive for the presence of CTX_{M-1} gene at 893bp, these isolates were undergone a first run on gel electrophoresis, and the results are shown in Figure (4-6). Generally, the isolates *E.coli* unexposed (92%) exhibited positive PCR products on gel electrophoresis for the CTX_{M-1} gene at 893bp in an unexposed state, but 16% of the total number, when exposed to magnetic powers (0.04T), became negative also (8%) samples in power 0.08T, 0.12T, and 0.16T became negative PCR products on gel electrophoresis for CTX_{M-1} gene at 893bp.



Figure (4-5) statical analysis showing significant differences between CTX_M gene positive for *E. coli* unexposed vs. CTX_M gene positive for *E. coli* under magnetic power (0.04, 0.08, 0.12 and 0.16T) respectively.



Figure (4-6) Agarose (2%) gel electrophoresis of PCR product of amplified CTX_{M-1} gene of *E.coli* isolate (amplicon with 893bp), group (I-VI) represent number of sample and (0) control unexposed to the magnetic field (1, 2, 3, 4) sample exposed to magnetic power (0.04, 0.08, 0.12 and 0.16T) respectively.

4.6.3. Detection of TEM_1 gene among *E.coli* isolates using PCR Technique

Results of molecular identification shown in Figure (4-7) indicated that (23) of (25) samples of uropathogenic *E.coli* isolates were positive for the presence of the *TEM*₁ gene at 535bp, these isolates were undergone a first run on gel electrophoresis, and the results are shown in Figure (4-8). Generally, all isolates of *E. coli* unexposed (92%) exhibited positive PCR products on gel electrophoresis for the *TEM*₁ gene at 535bp in an unexposed state. 32% of total samples became negative for power (0.08T). 24% of total samples became negative for the *TEM*₁ gene when exposed to powers of magnetic field (0.04T). 8% of total samples became negative in power (0.12T).



Figure (4-7) statistical analysis showing significant differences between TEM_1 gene positive for *E. coli* unexposed vs. TEM_1 gene positive for *E. coli* under magnetic power (0.04, 0.08, 0.12 and 0.16T) respectively.



Figure (4-8) Agarose (2%) gel electrophoresis of PCR product of amplified TEM_1 gene of *E.coli* isolate (amplicon with 535bp), group (I-IV) represent number of sample and (0) Negative control unexposed to the magnetic field (1, 2, 3, 4) sample exposed to magnetic power (0.04, 0.08, 0.12 and 0.16T) respectively.

4.6.4. Detection of SHV gene Among E.coli Isolates using PCR Technique

The results of molecular identification shown in Figure (4-9) show that (23) from (25) samples of uropathogenic *E.coli* isolates were negative PCR products on gel electrophoresis for the *SHV gene* at 800bp. These isolates were undergone run on gel electrophoresis, and the results are shown in Figure (4-10); in general, the result of all isolates of *E.coli* unexposed negative PCR products on gel electrophoresis for *SHV gene* at 800bp. The result was positive for two sample from (25) in powers (0.04, 0.08, 0.12T), but the same sample was negative in power 0.16T.



Figure (4-9) Statical analysis showing significant differences between *SHV* gene positive for *E.coli* unexposed vs. *SHV* gene positive for *E.coli* under magnetic power (0.04, 0.08, 0.12 and 0.16T) respectively.



Figure (4-10) Agarose (2%) gel electrophoresis of *SHV gene* amplification, L: ladder Group I represent number of samples, (0) Negative control unexposed to the magnetic field (1, 2, 3, 4) sample exposed to magnetic power (0.04, 0.08, 0.12 and 0.16T) respectively (1, 2, 3) positive results (4) negative result.

Hierarchial Cluster			Bef	ore t	reati	ment		After treatment 0.04T							After treatment 0.08T							After treatment 0.12T							After treatment 0.16T						
Dendrogram using Ward Linkage	Isolat e No.	ILATk	TyrA	AGAL	SUCT	ProA	SAC	ILATk	TyrA	AGAL	SUCT	ProA	SAC	ILATk	TyrA	AGAL	SUCT	ProA	SAC	ILATk	TyrA	AGAL	SUCT	ProA	SAC	ILATk	TyrA	AGAL	SUCT	ProA	SAC				
Care 13	13	-	-	-	-	-	-	+	+	+	+	-	-	+	-	-	+	-	-	+	-	+	+	-	-	+	-	+	+	-	_				
Case 35	25	-	-	-	-	-	-	+	+	+	+	-	-	+	-	-	+	-	-	+	-	+	+	-	-	+	-	+	+	-	-				
Carl 1	1	-	-	-	-	-	-	+	+	+	+	-	-	+	-	-	+	-	-	+	-	+	+	-	-	+	-	+	+	-	-				
Case JI 10	10	+	+	+	+	-	+	-	+	+	+	-	+	-	+	+	+	+	+	-	+	+	+	-	+	-	+	+	+	-	+				
Case 22 22	22	+	+	+	+	-	+	-	+	+	+	-	+	-	+	+	+	+	+	-	+	+	+	-	+	-	+	+	+	-	+				
Care 5 5	7	-	-	+	+	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	+	+	-	-				
Case II II	19	-	-	+	+	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	+	+	-	-				
(w) in	3	+	+	-	+	-	-	-	-	-	-	-	-	+	+	+	+	+	-	+	+	+	+	+	-	+	+	+	+	+	-				
Con P 19	15	+	+	-	+	-	-	-	-	-	-	-	-	+	+	+	+	+	-	+	+	+	+	+	-	+	+	+	+	+	-				
	11	-	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+				
Cate 0	23	-	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+				
Cat a 10	6	-	+	+	+	-	-	-	+	+	-	-	-	-	+	+	+	-	-	-	+	+	+	-	-	-	+	+	+	-	-				
Case i 8-	18	-	+	+	+	-	-	-	+	+	-	-	-	-	+	+	+	-	-	-	+	+	+	-	-	-	+	+	+	-	-				
Carl D D	8	-	-	-	-	-	-	-	-	-	-	-	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+				
Carl 2-	20	-	-	-	-	-	-	-	-	-	-	-	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+				
Case 14	14	+	-	+	-	-	-	+	+	+	+	-	-	+	-	+	+	-	-	+	-	+	+	-	-	+	+	+	+	-	-				
Case 12 12-	16	+	-	+	-	-	-	+	+	+	+	-	-	+	-	+	+	-	-	+	-	+	+	-	-	+	+	+	+	-	-				
Curl 34-	4	-	-	+	-	-	-	+	-	+	+	-	-	+	+	+	+	-	-	-	-	+	-	-	-	+	-	+	-	-	-				
Can-4 4	2	-	-	+	-	-	-	+	-	+	+	-	-	+	+	+	+	-	-	-	-	+	-	-	-	+	-	+	-	-	-				
Case M 10	12	+	+	+	+	-	-	+	+	+	+	+	-	+	+	+	+	+	-	+	+	+	+	+	-	+	+	+	+	+	-				
Case 9	24	+	+	+	+	-	-	+	+	+	+	+	-	+	+	+	+	+	-	+	+	+	+	+	-	+	+	+	+	+	-				
Case 31 27	9	-	+	+	_	-	+	-	-	+	-	-	+	+	+	+	+	-	+	-	-	+	+	-		_	+	+	+	_	+				
Cut II 11	5	-	+	+	-	-	+	-	-	+	-	-	+	+	+	+	+	-	+	-	-	+	+	-		-	+	+	+	-	+				
Case 3 23	21	-	-	-	-	-	-	+	+	+	+	-	-	+	+	+	+	+	-	+	+	+	+	-	-	+	+	+	+	-	-				
Carl 3 Carl 15	17	-	-	-	-	-	-	+	+	+	+	-	-	+	+	+	+	+	-	+	+	+	+	-	-	+	+	+	+	-	-				

Figure (4-11) Genotyping of *E. coli* isolates using Conventional-PCR method through the ESBL gene. The dendrogram was constructed using Conventional-PCR patterns of *16SrRNA* and ESBL genes ((bla_{CTXM} , bla_{TEM} , and bla_{SHV}) of *E. coli* isolates. Similarity clustering analysis was performed using the Hierarchical Cluster. The dashed line is a hypothetical line showing ~90% similarity.

4.7. Sequence alignment and submission gene to GenBank

The program Bio Edit V.7.0.5 was used to assess the quality of the sequenced data for both genes. Homology, insertions - deletions, stop codons, and frame shifts were examined using NCBI- BLAST, the website was compared and alignment of the laboratory or query sequence with another biological sequence to determine a greater degree of similarity and nucleotide variation with other targets. GenBank Figures (4-12 to 4-16) for the TEM_1 gene and (4-17 to 4-21) for the CTX_{M-1} gene It was WWW-based submission tool featuring wizards that facilitated the submission process. The GenBank database was designed for newly determined and annotated sequence data submitted by the submitter. All sequences have been submitted to GenBank (Table 4-4).
4.7.1. Molecular sequencing for *TEM*₁ gene

Escherichia coli strain GN03344 plasmid p3344-5

Sequence ID: CP095547.1 Length: 131082 Number of Matches: 1

Range 1: 15027 to 1552	V Next I	Match 🔺 Previous Matc	h		
Score	Expect	Identities	Gaps	Strand	
908 bits(1006)	0.0	503/503(100%)	0/503(0%)	Plus/Plus	
CDS: Putative 1 Query	1 1	Q H F R V A L I P TCAACATTTTCGTGTCGCCCTTATTCC	F F A A F	C L P V F A	60
Sbjct CDS:class A broad-sp	15027 4	TCAACATTTCGTGTCGCCCCTTATTCC Q H F R V A L I P	CTTTTTTGCGGCATTT	GCCTTCCTGTTTTTGC C L P V F A	15086
CDS: Putative 1	21	H P E T L V K V K	D A E D Q	L G A R V G	120
Query	61	TCACCCAGAAACGCTGGTGAAAGTAAA	AGATGCTGAAGATCAG	TGGGTGCACGAGTGGG	
Sbjct	15087	TCACCCAGAAACGCTGGTGAAAGTAAA	AGATGCTGAAGATCAG	TGGGTGCACGAGTGGG	15146
CDS:class A broad-sp	24	H P E T L V K V K	DAEDQ	L G A R V G	
CDS: Putative 1 Query	41 121	Y I E L D L N S G TTACATCGAACTGGATCTCAACAGCGG	K I L E S TAAGATCCTTGAGAGT	F R P E E R	180
Sbjct	15147	TTACATCGAACTGGATCTCAACAGCGG	TAAGATCCTTGAGAGT	TTCGCCCCGAAGAACG	15206
CDS:class A broad-sp	44	Y I E L D L N S G	KILES	F R P E E R	
CDS: Putative 1 Query	61 181	F P M M S T F K V TTTTCCAATGATGAGCACTTTTAAAGT	L L C G A	V L S R V D TATTATCCCGTGTTGA	240
Sbjct CDS:class A broad-sp	15207 64	F P M M S T F K V	TCTGCTATGTGGTGCGG LLCGA	V L S R V D	15266
CDS: Putative 1	81	A G Q E Q L G R R	I H Y S Q	N D L V E Y	300
Query	241	CGCCGGGCAAGAGCAACTCGGTCGCCG	CATACACTATTCTCAG	ATGACTTGGTTGAGTA	
Sbjct	15267	CGCCGGGCAAGAGCAACTCGGTCGCCG	CATACACTATTCTCAG	ATGACTTGGTTGAGTA	15326
CDS:class A broad-sp	84	A G Q E Q L G R R	I H Y S Q	N D L V E Y	
CDS: Putative 1	101	S P V T E K H L T	D G M T V	R E L C S A	360
Query	301	CTCACCAGTCACAGAAAAGCATCTTAC	GGATGGCATGACAGTA	AGAGAATTATGCAGTGC	
Sbjct	15327	CTCACCAGTCACAGAAAAGCATCTTAC	GGATGGCATGACAGTAA	AGAGAATTATGCAGTGC	15386
CDS:class A broad-sp	104	S P V T E K H L T	D G M T V	R E L C S A	
CDS: Putative 1	121	A I T M S D N T A	A N L L L	T T I G G P	420
Query	361	TGCCATAACCATGAGTGATAACACTGC	TGCCAACTTACTTCTG/	ACAACGATCGGAGGACC	
Sbjct CDS:class A broad-sp	15387 124	TGCCATAACCATGAGTGATAACACTGC A I T M S D N T A	TGCCAACTTACTTCTG	CAACGATCGGAGGACC T T I G G P	15446
CDS: Putative 1 Query	141 421	K E L T A F L H N GAAGGAGCTAACCGCTTTTTTGCACAA	M G D H V CATGGGGGGATCATGTA	T R L D R W	480
Sbjct	15447	GAAGGAGCTAACCGCTTTTTTGCACAA	CATGGGGGGATCATGTAA	CTCGCCTTGATCGTTG	15506
CDS:class A broad-sp	144	K E L T A F L H N	M G D H V	T R L D R W	
CDS: Putative 1 Query	161 481	E P E L N E A GGAACCGGAGCTGAATGAAGCCA 50	3		
Sbjct CDS:class A broad-sp	15507 164	GGAACCGGAGCTGAATGAAGCCA 15 E P E L N E A	529		

Figure (4-12) NCBI blasting pairwise alignment of Negative control samples (unexposed to the magnetic field) sequences query of β -lactamase (*TEM*₁) gene with the subject of *Escherichia coli* in NCBI.

Sequence ID: CP095547.1 Length: 131082 Number of Matches: 1

Range 1: 15027 to 15529 GenBank Graphics Vext Match 🛦 Previous Match					ch
Score 899 hits(996)	Expect	Identities 501/503(99%)	Gaps 0/503(0%)	Strand Dlue/Dlue	
CDS: Putative 1 Query Sbjct CDS:class A broad-sp	1 7 15027 4	L H F R V A L TCTACATTTTCGTGTCGCCCTT/ TCAACATTTTCGTGTCGCCCCTT/ Q H F R V A L	I S F F A A ATTTCCTTTTTTGCGGC ATTCCCTTTTTTGCGGC I P F F A A	ATTTIGCCTTCCTGTTTTTGC	66 15086
CDS: Putative 1 Query Sbjct CDS:class A broad-sp	21 67 15087 24	H P E T L V K TCACCCAGAAACGCTGGTGAAA IIIIIIIIIIIIIIIIIIIIIIIIIIIIIII	V K D A E D GTAAAAGATGCTGAAGA 	QLGARVG TCAGTTGGGTGCACGAGTGGG 	126 15146
CDS: Putative 1 Query Sbjct CDS:class A broad-sp	41 127 15147 44	Y I E L D L N TTACATCGAACTGGATCTCAAC IIIIIIIIIIIIIIIIIIIIIIIIIIIIIII	S G K I L E AGCGGTAAGATCCTTGA AGCGGTAAGATCCTTGA S G K I L E	S F R P E E R GAGTTTTCGCCCCGAAGAACG IIIIIIIIIIIIIIIIIIIIIIIII	186 15206
CDS: Putative 1 Query Sbjct CDS:class A broad-sp	61 187 15207 64	F P M M S T F TTTTCCAATGATGAGCACTTTT IIIIIIIIIIIIIIIIIIIIIIIIIIIIIIII	K V L L C G AAAGTTCTGCTATGTGG AAAGTTCTGCTATGTGG K V L L C G	A V L S R V D TGCGGTATTATCCCGTGTTGA IIIIIIIIIIIIIIIIIIIIIIIIIIIIIIII	246 15266
CDS: Putative 1 Query Sbjct CDS:class A broad-sp	81 247 15267 84	A G Q E Q L G CGCCGGGCAAGAGCAACTCGGT(R R I H Y S CGCCGCATACACTATTC 	Q N D L V E Y TCAGAATGACTTGGTTGAGTA IIIIIIIIIIIIIIIIIIIIIIIIIIII	306 15326
CDS: Putative 1 Query Sbjct CDS:class A broad-sp	101 307 15327 104	S P V T E K H CTCACCAGTCACAGAAAAGCAT(IIIIIIIIIIIIIIIIIIIIIIIIIIIIIIIIIIII	L T D G M T CTTACGGATGGCATGAC IIIIIIIIIIIIIIIIII CTTACGGATGGCATGAC L T D G M T	V R E L C S A AGTAAGAGAATTATGCAGTGC IIIIIIIIIIIIIIIIIIIIIIIIIIIIIIIIIII	366 15386
CDS: Putative 1 Query Sbjct CDS:class A broad-sp	121 367 15387 124	A I T M S D N TGCCATAACCATGAGTGATAACA 	T A A N L L ACTGCTGCCAACTTACT IIIIIIIIIIIIIIIII ACTGCTGCCAACTTACT T A A N L L	L T T I G G P TCTGACAACGATCGGAGGACC TCTGACAACGATCGGAGGACC L T T I G G P	426 15446
CDS: Putative 1 Query Sbjct CDS:class A broad-sp	141 427 15447 144	K E L T A F L GAAGGAGCTAACCGCTTTTTG GAAGGAGCTAACCGCTTTTTTG K E L T A F L	H N M G D H CACAACATGGGGGATCA CACAACATGGGGGATCA H N M G D H	V T R L D R W TGTAACTCGCCTTGATCGTTG TGTAACTCGCCTTGATCGTTG V T R L D R W	486 15506
CDS: Putative 1 Query Sbjct CDS:class A broad-sp	161 487 15507 164	E P E L N E A GGAACCGGAGCTGAATGAAGCC 	A 509 A 15529		

Figure(4-13)NCBI blasting pairwise alignment of sequences for the betalactamase (TEM_1) gene in *E.coli* with a sample exposed to (0.04T) of magnetic field.

Sequence ID: CP095547.1 Length: 131082 Number of Matches: 1

Range 1: 15027 to 1552	9 GenBar	nk Graphics		Vext Match 🔺 Previous Mate	ch
Score	Expect	Identities	Gaps	Strand	
899 bits(996)	0.0	501/503(99%)	0/503(0%)	Plus/Plus	
CDS: Putative 1 Query	17	L H F R V A L TCTACATTTTCGTGTCGCCCTTA	ISFFA	A F C L P V F A GGCATTTTGCCTTCCTGTTTTTGC	66
CDS:class A broad-sp	4	Q H F R V A L	I P F F A	A F C L P V F A	15680
CDS: Putative 1 Query	21 67	H P E T L V K TCACCCAGAAACGCTGGTGAAAG	V K D A E TAAAAGATGCTGA	D Q L G A R V G AGATCAGTTGGGTGCACGAGTGGG	126
Sbjct CDS:class A broad-sp	15087 24	H P E T L V K	TAAAAGATGCTGA V K D A E	AGATCAGTTGGGTGCACGAGTGGG D Q L G A R V G	15146
CDS: Putative 1 Query	41 127	Y I E L D L N TTACATCGAACTGGATCTCAACA	S G K I L GCGGTAAGATCCT	E S F R P E E R TGAGAGTTTTCGCCCCGAAGAACG	186
Sbjct CDS:class A broad-sp	15147 44	Y I E L D L N	GCGGTAAGATCCT S G K I L	TGAGAGTTTTCGCCCCGAAGAACG E S F R P E E R	15206
CDS: Putative 1 Query	61 187	F P M M S T F TTTTCCAATGATGAGCACTTTTA	K V L L C AAGTTCTGCTATG	G A V L S R V D TGGTGCGGTATTATCCCGTGTTGA	246
Sbjct CDS:class A broad-sp	15207 64	TTTTCCAATGATGAGCACTTTTA F P M M S T F	AAGTTCTGCTATG K V L L C	G A V L S R V D	15266
CDS: Putative 1 Query	81 247	A G Q E Q L G CGCCGGGCAAGAGCAACTCGGTC	R R I H Y GCCGCATACACTA	S Q N D L V E Y TTCTCAGAATGACTTGGTTGAGTA	306
Sbjct CDS:class A broad-sp	15267 84	CGCCGGGCAAGAGCAACTCGGTC	GCCGCATACACTA R R I H Y	S Q N D L V E Y	15326
CDS: Putative 1 Query	101 307	S P V T E K H CTCACCAGTCACAGAAAAGCATC	L T D G M	T V R E L C S A GACAGTAAGAGAATTATGCAGTGC	366
Sbjct CDS:class A broad-sp	15327 104	CTCACCAGTCACAGAAAAGCATC S P V T E K H	TTACGGATGGCAT	GACAGTAAGAGAATTATGCAGTGC T V R E L C S A	15386
CDS: Putative 1 Query	121 367	A I T M S D N TGCCATAACCATGAGTGATAACA	T A A N L CTGCTGCCAACTT/	L L T T I G G P ACTTCTGACAACGATCGGAGGACC	426
Sbjct CDS:class A broad-sp	15387 124	TGCCATAACCATGAGTGATAACA A I T M S D N	CTGCTGCCAACTT/ T A A N L	ACTTCTGACAACGATCGGAGGACC LLTTIGGP	15446
CDS: Putative 1 Query	141 427	K E L T A F L GAAGGAGCTAACCGCTTTTTTGC	H N M G D ACAACATGGGGGA	H V T R L D R W TCATGTAACTCGCCTTGATCGTTG	486
Sbjct CDS:class A broad-sp	15447 144	GAAGGAGCTAACCGCTTTTTTGC	ACAACATGGGGGA H N M G D	TCATGTAACTCGCCTTGATCGTTG H V T R L D R W	15506
CDS: Putative 1 Query	161 487	E P E L N E A GGAACCGGAGCTGAATGAAGCCA	509		
Sbjct CDS:class A broad-sp	15507 164	GGAACCGGAGCTGAATGAAGCCA E P E L N E A	15529		

Figure(4-14)NCBI blasting pairwise alignment of sequences for the betalactamase (TEM_1) gene in *E.coli* with a sample exposed to (0.08T) of magnetic field.

Sequence ID: CP095547.1 Length: 131082 Number of Matches: 1

Range 1: 15027 to 15529 GenBank Graphics Vext Match A Previous Match					h
Score	Expect	Identities	Gaps	Strand	
894 bits(991)	0.0	500/503(99%)	0/503(0%)	Plus/Plus	
CDS: Putative 1 Query	17	Q H F R V A L I TCAACATTTTCGTGTCGCCCTTATT	P F F A A F	C L P V F A	66
CDS:class A broad-sp	4	Q H F R V A L I	PFFAAF	CLPVFA	13000
CDS: Putative 1	21	H P E T L V K V	K D A E D Q	L G A R V G	126
Query	67	TCACCCAGAAACGCTGGTGAAAGTA	AAAAGATGCTGAAGATCAG	TTGGGTGCACGAGTGGG	
Sbjct CDS:class A broad-sp	15087 24	H P E T L V K V	AAAAGATGCTGAAGATCAG K D A E D Q	L G A R V G	15146
CDS: Putative 1	41	Y I E L D L N S	G K I L E S	F R P E E R	186
Query	127	TTACATCGAACTGGATCTCAACAGG	GGTAAGATCCTTGAGAGT	TTTCGCCCCGAAGAACG	
Sbjct CDS:class A broad-sp	15147 44	TTACATCGAACTGGATCTCAACAG	GGTAAGATCCTTGAGAGT G K I L E S	F R P E E R	15206
CDS: Putative 1	61	F P M M S T F K	V L L C G A	V L S R V D	246
Query	187	TTTTCCAATGATGAGCACTTTTAA	AGTTCTGCTATGTGGTGCG	GTATTATCCCGTGTTGA	
Sbjct CDS:class A broad-sp	15207 64	F P M M S T F K	AGTTCTGCTATGTGGTGCG V L L C G A	GTATTATCCCGTGTTGA V L S R V D	15266
CDS: Putative 1	81	A G Q E Q L G R	R I H Y S Q	N D L V E Y	306
Query	247	CGCCGGGCAAGAGCAACTCGGTCG	CCGCATACACTATTCTCAG	AATGACTTGGTTGAGTA	
Sbjct	15267	CGCCGGGCAAGAGCAACTCGGTCGC	CCGCATACACTATTCTCAG	AATGACTTGGTTGAGTA	15326
CDS:class A broad-sp	84	A G Q E Q L G R	R I H Y S Q	N D L V E Y	
CDS: Putative 1	101	S P V T E K H L	T D G M T V	R E L C S A	366
Query	307	CTCACCAGTCACAGAAAAGCATCT	FACGGATGGCATGACAGTA	AGAGAATTATGCAGTGC	
Sbjct	15327	CTCACCAGTCACAGAAAAGCATCT	TACGGATGGCATGACAGTA	AGAGAATTATGCAGTGC	15386
CDS:class A broad-sp	104	S P V T E K H L	T D G M T V	R E L C S A	
CDS: Putative 1	121	A I T M S D N T	A A N L L L	T T I G G P	426
Query	367	TGCCATAACCATGAGTGATAACACT	IGCTGCCAACTTACTTCTG	ACAACGATCGGAGGACC	
Sbjct	15387	TGCCATAACCATGAGTGATAACACT	GCTGCCAACTTACTTCTG	ACAACGATCGGAGGACC	15446
CDS:class A broad-sp	124	A I T M S D N T	A A N L L L	T T I G G P	
CDS: Putative 1	141	K E L T A F L H	N M G D H V	T R L D R E	486
Query	427	GAAGGAGCTAACCGCTTTTTTGCAC	CAACATGGGGGGATCATGTA	ACTCGCCTTGATCGTGA	
Sbjct	15447	GAAGGAGCTAACCGCTTTTTTGCAC	AACATGGGGGGATCATGTA	ACTCGCCTTGATCGTTG	15506
CDS:class A broad-sp	144	K E L T A F L H	N M G D H V	T R L D R W	
CDS: Putative 1 Query	161 487	E P E L N E A AGAACCGGAGCTGAATGAAGCCA	509		
Sbjct CDS:class A broad-sp	15507 164	GGAACCGGAGCTGAATGAAGCCA E P E L N E A	15529		

Figure(4-15)NCBI blasting pairwise alignment of sequences for the betalactamase (TEM_1) gene in *E.coli* with a sample exposed to (0.12T) of magnetic field.

Sequence ID: CP095547.1 Length: 131082 Number of Matches: 1

Range 1: 15027 to 15529 GenBank Graphics Vext Match 🗼 Previous Match					:h
Score	Expect	Identities	Gaps	Strand	
894 bits(991)	0.0	500/503(99%)	0/503(0%)	Plus/Plus	
CDS: Putative 1 Query Sbjct CDS:class A broad-sp	1 7 15027 4	Q H F R V A L TCAACATTTTCGTGTCGCCCTTA 	I P F F A A TTCCCTTTTTTGCGGCAT IIIIIIIIIIIIIIIIIIIIIIIIIIIIIIIIIIII	F C L P V F A TTTGCCTTCCTGTTTTTGC 	66 15086
CDS: Putative 1 Query	21 67	H P E T L V K TCACCCAGAAACGCTGGTGAAAG	V K D A E D	Q L G A R V G	126
CDS:class A broad-sp	24	H P E T L V K	V K D A E D	Q L G A R V G	15140
CDS: Putative 1 Query Sbict	41 127 15147	Y I E L D L N S TTACATCGAACTGGATCTCAACA IIIIIIIIIIIIIIIIIIIIIIIIIIIIII	S G K I L E GCGGTAAGATCCTTGAGA	S F R P E E R GTTTTCGCCCCGAAGAACG	186 15206
CDS:class A broad-sp	44	YIELDLN	SGKILE	SFRPEER	
CDS: Putative 1 Query	61 187	F P M M S T F TTTTCCAATGATGAGCACTTTTA	K V L L C G AAGTTCTGCTATGTGGTG	A V L S R V D CGGTATTATCCCGTGTTGA	246
Sbjct CDS:class A broad-sp	15207 64	F P M M S T F	AAGTTCTGCTATGTGGTG K V L L C G	CGGTATTATCCCGTGTTGA A V L S R V D	15266
CDS: Putative 1 Query	81 247	A G Q E Q L G CGCCGGGCAAGAGCAACTCGGTC	R R I H Y S GCCGCATACACTATTCTC	Q N D L V E Y AGAATGACTTGGTTGAGTA	306
Sbjct CDS:class A broad-sp	15267 84	CGCCGGGCAAGAGCAACTCGGTC	GCCGCATACACTATTCTC R R I H Y S	AGAATGACTTGGTTGAGTA Q N D L V E Y	15326
CDS: Putative 1 Query	101 307	S P V T E K H CTCACCAGTCACAGAAAAGCATC	L T D G M T TTACGGATGGCATGACAG	V R E L C S A TAAGAGAATTATGCAGTGC	366
Sbjct CDS:class A broad-sp	15327 104	CTCACCAGTCACAGAAAAGCATC S P V T E K H	TTACGGATGGCATGACAG L T D G M T	TAAGAGAATTATGCAGTGC V R E L C S A	15386
CDS: Putative 1 Query	121 367	A I T M S D N TGCCATAACCATGAGTGATAACA	T A A N L L CTGCTGCCAACTTACTTC	L T T I G G P TGACAACGATCGGAGGACC	426
Sbjct CDS:class A broad-sp	15387 124	TGCCATAACCATGAGTGATAACA A I T M S D N	CTGCTGCCAACTTACTTC T A A N L L	TGACAACGATCGGAGGACC L T T I G G P	15446
CDS: Putative 1 Query	141 427	K E L T A F L GAAGGAGCTAACCGCTTTTTTGC	H N M G D H ACAACATGGGGGGATCATG	V T R L D R E TAACTCGCCTTGATCGTGA	486
Sbjct CDS:class A broad-sp	15447 144	GAAGGAGCTAACCGCTTTTTTGC	ACAACATGGGGGATCATG H N M G D H	TAACTCGCCTTGATCGTTG V T R L D R W	15506
CDS: Putative 1 Query	161 487	E P E L N E A AGAACCGGAGCTGAATGAAGCCA	509		
Sbjct CDS:class A broad-sp	15507 164	GGAACCGGAGCTGAATGAAGCCA E P E L N E A	15529		

Figure(4-16)NCBI blasting pairwise alignment of sequences for the betalactamase (TEM_1) gene in *E.coli* with a sample exposed to (0.16T) of magnetic field.

4.7.2. Molecular sequencing for CTX_{M-1} gene

Escherichia coli strain EC488 chromosome, complete genome Sequence ID: <u>CP109874.1</u> Length: 5173170 Number of Matches: 1

Score	Expect	Identities	Gaps	Strand	
1581 bits(1752)	0.0	876/876(100%)	0/876(0%)	Plus/Plus	
CDS: Putative 1 Query	1	M V K K S L I	GCCAGTTCACGCTGATGGCGA	GOCAACCOTCACOCTGTTG	68
Sbjct CDS:extended-spectru	1512294 1	ATGGTTAAAAAATCACTGC	CCAGTTCACGCTGATGGCGAC	GGCAACCGTCACGCTGTTG	1512353
CDS: Putative 1 Query	21 61	TTAGGAAGTGTGCCGCTGT	A Q T A D V C	Q K L A E L	120
Sbjct CDS:extended-spectru	1512354 21	L G S V P L	ATGCGCAAACGGCGGGGCGTAC	Q K L A E L	1512413
CDS: Putative 1 Query	41 121	E R Q S G G I GAGCGGCAGTCGGGAGGCA	R L G V A L I M	T A D N S Q	180
Sbjct CDS:extended-spectru	1512414 41	E R Q S G G	R L G V A L I	CACAGCAGATAATTCGCAA	1512473
CDS: Putative 1 Query	61 181	ATACTITATCGTGCTGATG	E R F A M C S T	CAGTAAAGTGATGGCCGCG	240
Sbjct CD5:extended-spectru	1512474 61	ATACTITATCGTGCTGATG	AGCGCTTTGCGATGTGCAGCAG	CAGTAAAGTGATGGCCGCG	1512533
CDS: Putative 1 Query	81 241	A A V L K K	S E S E P N L I GTGAAAGCGAACCGAATCTGT	AAATCAGCGAGTTGAGATC	300
Sbjct CDS:extended-spectru	1512534 81	A A V L K K	GTGAAAGCGAACCGAATCTGT	AAATCAGCGAGTTGAGATC	1512593
CDS: Putative 1 Query	101 301	K K S D L V AAAAAAATCTGACCTTGTTA	N Y N P I A E A	CHVNGTM GCACGTCAATGGGACGATG	360
Sbjct CDS:extended-spectru	1512594 101	K K S D L V	ACTATAATCCGATTGCGGAAAA	C H V N G T M	1512653
CDS: Putative 1 Query	121 361	S L A E L S	A A A L Q Y S C	N V A M N K	428
Sbjct CDS:extended-spectru	1512654 121	TCACTGGCTGAGCTTAGCG	COCCCCCCCCCCACAGTACAGCG	TAACGTGGCGATGAATAAG	1512713
CDS: Putative 1 Query	141 421	CTGATTGCTCACGTTGGCG	G P A S V T A C	COCCCGACAGCTGGGAGAC	480
Sbjct CDS:extended-spectru	1512714 141	L I A H V G	G P A S V T A	A R Q L G D	1512773
CDS: Putative 1 Query	161 481	E T F R L D I	R T E P T L N 1 GTACCGAGCCGACGTTAAACA	A I P G D P	540
Sbjct CD5:extended-spectru	1512774 161	GAAACGTTCCGTCTCGACC	GTACCGAGCCGACGTTAAACAC	A I P G D P	1512833
CDS: Putative 1 Query	181 541	R D T T S P I	R A M A Q T L I GGGCAATGGCGCAAACTCTGCC	A N L T L G K	600
Sbjct CDS:extended-spectru	1512834 181	CGTGATACCACTTCACCTC	GGGCAATGGCGCAAACTCTGCC	CAATCTGACGCTGGGTAAA	1512893
CDS: Putative 1 Query	201 601	A L G D S Q I	R A Q L V T W J GGGCGCAGCTGGTGACATGGAT	K G N T T G	669
Sbjct CDS:extended-spectru	1512894 201	GCATTOGGCGACAGCCAAC	GGGCGCAGCTGGTGACATGGAT	GAAAGGCAATACCACCGGT	1512953
CDS: Putative 1 Query	221 661	A A S I Q A	GACTGCCTGCTTCCTGGGTTG	G D K T G S	720
Sbjct CDS:extended-spectru	1512954 221	GCAGCGAGCATTCAGGCTG	G L P A S W V V	GOGGGATAAAACCGGCAGC	1513013
CDS: Putative 1 Query	241 721	G G Y G T T I	N D I A V I W ACGATATCGCGGGGGGGGGGGGGGGGGGGGGGGGGGGGG	K D R A P L	780
Sbjct CDS:extended-spectru	1513014 241	G G Y G T T	ACGATATCGCGCGGTGATCTGGCG	K D R A P L	1513073
CDS: Putative 1 Query	261 781	ATTCTGGTCACTTACTTCA	T Q P Q P K A I	AAGCCGTCGCGATGTATTA	840
Sbjct CDS:extended-spectru	1513074 261	ATTCTGGTCACTTACTTCA	CCCASCCTCAACCTAACGCAG	AAGCCGTCGCGATGTATTA	1513133
CDS: Putative 1 Query	281 841	A S A A K I	TCACCGACGGTTTGTAA 870	5	
Sbjct	1513134	GCGTCGGCGGCTAAAATCG	TCACCGACGGTTTGTAA 15	13169	

Figure(4-17)NCBI blasting pairwise alignment of Negative control samples (unexposed to the magnetic field) sequences query of beta-lactamase (CTX_{M-1}) gene with the subject of *Escherichia coli*.

Escherichia coli strain EC488 chromosome, complete genome Sequence ID: <u>CP109874.1</u> Length: 5173170 Number of Matches: 1

Range 1: 1512294 to 1513169 Genilank Graphics View Match					
Score 1567 bits(1737)	Expect 0.0	Identities 873/876(99%)	Gaps 0/876(0%)	Strand Plus/Plus	
CDS: Putative 1 Query	1	M V K K S L	R Q F T L K A	A T A T V T L L CGACGGCCACCGTCACGCTGTTG	60
Sbjct CDS:extended-spectru	1512294		CCAGTTCACCCTGATGG	CGACGGCAACCGTCACGCTGTTG	1512353
CDS: Putative 1	21	L G S V P L			120
Sbjct CDS:extended-spectru	1512354	TTAGGAAGTGTGCCGCTGT			1512413
CDS: Putative 1 Query	41	E R P S G G GAGCGGCCGTCGGGAGGCA	R L G V A L GACTOGGTGTGGCATTGA	I N T A D N S Q	180
Sbjct CDS:extended-spectru	1512414 41	GAGCGGCAGTCGGGAGGCA	GACTOGGTGTGGCATTGA	TTAACACAGCAGATAATTCGCAA INTADNSQ	1512473
CDS: Putative 1 Query	61 181	I L Y R A D	E R F A M C S	S T S K V M A A	240
Sbjct CDS:extended-spectru	1512474 61	ATACTITATCGTGCTGATG	AGCGCTTTGCGATGTGCA	GCACCAGTAAAGTGATGGCCGCG S T S K V M A A	1512533
CDS: Putative 1 Query	81 241	A A V L K K GCCGCGGTGCTGAAGAAAA	S E S E P N I	L L N Q R V E I TGTTAAATCAGCGAGTTGAGATC	300
Sbjct CDS:extended-spectru	1512534 81	GCCGCGGTGCTGAAGAAAA	GTGAAAGCGAACCGAATC	TGTTAAATCAGCGAGTTGAGATC	1512593
CDS: Putative 1 Query	101 301	K K S D L V AAAAAAATCTGACCTTGTTA	N Y N P I A I	E K H V N G T M AAAAGCACGTCAATGGGACGATG	360
Sbjct CDS:extended-spectru	1512594 101	AAAAAAATCTGACCTTGTTA	ACTATAATCCGATTGCGG	AAAAGCACGTCAATGGGACGATG E K H V N G T M	1512653
CDS: Putative 1 Query	121 361	S L A E L S TCACTGGCTGAGCTTAGCG	A A A L Q Y	S D N V A M N K GCGATAACGTGGCGATGAATAAG	428
Sbjct CDS:extended-spectru	1512654 121	TCACTGGCTGAGCTTAGCG S L A E L S	CGGCCGCCGCTACAGTACA	GCGATAACGTGGCGATGAATAAG S D N V A M N K	1512713
CDS: Putative 1 Query	141 421	L I A H V G CTGATTGCTCACGTTGGCG	G P A S V T A	A F A R Q L G D CGTTCGCCCGACAGCTGGGAGAC	488
Sbjct CDS:extended-spectru	1512714 141	L I A H V G	G P A S V T	CGTTCGCCCGACAGCTGGGAGAC	1512773
CDS: Putative 1 Query	161 481	E T F R L D GAAACGTTCCGTCTCGACC	R T E P T L C	N T A I P G D P ACACCGCCATTCCGGGCGATCCG	548
Sbjct CDS:extended-spectru	1512774 161	GAAACGTTCCGTCTCGACC	GTACCGAGCCGACGTTAA	ACACCGCCATTCCGGGCGATCCG N T A I P G D P	1512833
CDS: Putative 1 Query	181 541	R D T T S P CGTGATACCACTTCACCTC	R A M A Q T I	L R N L T L G K TGCGGAATCTGACGCTGGGTAAA	688
Sbjct CDS:extended-spectru	1512834 181	Catataccacttcacctc	R A M A Q T	L R N L T L G K	1512893
CDS: Putative 1 Query	201 601	A L G D S Q GCATTGGGGGGACAGCCAAC	R A Q L V T I	M M K G N T T G GGATGAAAAGGCAATACCACCGGT	668
Sbjct CD5:extended-spectru	1512894 201	GCATTGGGCGACAGCCAAC	GGGCGCAGCTGGTGACAT	GGATGAAAGGCAATACCACCGGT W M K G N T T G	1512953
CDS: Putative 1 Query	221 661	A A S I Q A	GACTGCCTGCTTCCTGGG	V V G D K T G S	720
Sbjct CDS:extended-spectru	1512954 221	GCAGCGAGCATTCAGGCTG	GACTGCCTGCTTCCTGGG	TTGTGGGGGGATAAAACCGGCAGC V V G D K T G S	1513013
CDS: Putative 1 Query	241 721	G G Y G T T GGTGGCTATGGCACCACCA	N D I A V I I	W P K D R A P L	788
Sbjct CDS:extended-spectru	1513014 241	GGTGGCTATGGCACCACCA	ACGATATCGCGGTGATCT	GGCCAAAAGATCGTGCGCCGCTG	1513073
CDS: Putative 1 Query	261 781	ATTCTGGTCACTTACTTCA		A E S R R D V L CAGAAAGCCGTCGCGATGTATTA	840
Sbjct CDS:extended-spectru	1513074 261	ATTELEGTEACTIACTICA	CCCACCTCAACCTAACG	A E S R R D V L	1513133
CDS: Putative 1 Query	281 841	A S A A K I GCGTCGGCGGCTAAAATCG	V T D G L * TCACCGACGGTTTGTAA	876	
Sbjct CDS:extended-spectru	1513134 281	GCGTCGGCGGCTAAAATCG	T D G L	1513169	

Figure (4-18) NCBI blasting pairwise alignment of beta-lactamase (CTX_{M-1}) gene sequences from an *Escherichia coli* sample exposed to (0.04T) of magnetic power.

Escherichia coli strain EC488 chromosome, complete genome Sequence ID: <u>CP109874.1</u> Length: 5173170 Number of Matches: 1

Range 1: 1512294 to 15	13169 Gent	Bank Graphics	W /	inst Match 🔺 Previous Hatch	
Score 1563 bits(1732)	Expect 0.0	Identities 872/876(99%)	Gaps 0/876(0%)	Strand Plus/Plus	
CDS: Putative 1 Query	1	M V K K S L	CCAGTTCACGCTGAAGGC	GACGGCACCCCGTCACGCTGTTG	60
Sbjct CDS:extended-spectru	1512294 1	ATGGTTAAAAAATCACTGC M V K K S L	GCCAGTTCACGCTGATGGC	GACGGCAACCGTCACGCTGTTG	1512353
COS: Putative 1 Query	21 61	L R R V P L TTACGACGTGTGCCGCTGT	ATGCGCAAACGGCGGACGT	Q Q K L A E L ACAGCAAAAAACTTGCCGAATTA	128
Sbjct CDS:extended-spectru	1512354 21	TTAGGAAGTGTGCCGCTGT	ATGCGCAAACGGCGGGACGT	ACAGCAAAAAACTTGCCGAATTA Q Q K L A E L	1512413
CDS: Putative 1 Query	41 121	E R Q S G G GAGCGGCAGTCGGGAGGCA	R L G V A L I GACTGGGTGTGGCATTGAT	N T A D N S Q TAACACAGCAGATAATTCGCAA	190
Sbjct CDS:extended-spectru	1512414 41	GAGCGGCAGTCGGGAGGCA E R Q S G G	GACTOGGTGTGGCATTGAT	TAACACAGCAGATAATTCGCAA N T A D N S Q	1512473
CDS: Putative 1 Query	61 181	I L Y R A D	E R F A M C S	T S K V M A A CACCAGTAAAGTGATGGCCGCG	248
Sbjct CDS:extended-spectru	1512474 61	ATACTTTATCGTGCTGATG	AGCGCTTTGCGATGTGCAG	CACCAGTAAAGTGATGGCCGCG T S K V M A A	1512533
CDS: Putative 1 Query	81 241	A A V L K K GCCGCGGTGCTGAAGAAAA	S E S E P N L GTGAAAGCGAACCGAATCT	L N Q R V E I GTTAAATCAGCGAGTTGAGATC	300
Sbjct CDS:extended-spectru	1512534 81	GCCGCGGTGCTGAAGAAAA A A V L K K	GTGAAAGCGAACCGAATCT S E S E P N L	GTTAAATCAGCGAGTTGAGATC L N Q R V E I	1512593
CDS: Putative 1 Query	101 301	K K S D L V	N Y N P I A E	K H V N G T M AAAGCACGTCAATGGGACGATG	368
Sbjct CDS:extended-spectru	1512594 101	K K S D L V	ACTATAATCCGATTGCGGA N Y N P I A E	ALAGCACGTCAATGGGACGATG	1512653
CDS: Putative 1 Query	121 361	S L A E L S TCACTGGCTGAGCTTAGCG	A A A L Q Y S	D N V A M N K	428
Sbjct CDS:extended-spectru	1512654 121	TCACTOGCTGAGCTTAGCG S L A E L S	CGGCCGCCGCTACAGTACAG	CGATAACGTGGCGATGAATAAG D N V A M N K	1512713
CDS: Putative 1 Query	141 421	CTGATTGCTCACGTTGGCG	GCCCGGCTAGCGTCACCGC	F A R Q L G D GTTCGCCCGACAGCTGGGAGAC	480
Sbjct CDS:extended-spectru	1512714 141	L I A H V G	G P A S V T A	F A R Q L G D	1512773
CDS: Putative 1 Query	161 481	E T F R L D GAAACGTTCCGTCTCGACC	R T E P T L N GTACCGAGCCGACGTTAAA	T A I P G D P CACCGCCATTCCGGGCGATCCG	548
Sbjct CDS:extended-spectru	1512774 161	GAAACGTTCCGTCTCGACC	GTACCGAGCCGACGTTAAA	T A I P G D P	1512833
CDS: Putative 1 Query	181 541	R D T T S P	R A M A Q T L GGGCAATGGCGCAAACTCT	R N L T L G K	608
Sbjct CDS:extended-spectru	1512834 181	CGTGATACCACTTCACCTC	GGGCAATGGCGCGAAACTCT R A M A Q T L	R N L T L G K	1512893
CDS: Putative 1 Query	201 601	A L G D S Q GCATTGGGCGACAGCCAAC	R A Q L V T W GGGCGCAGCTGGTGACATG	M K G N T T G GATGAAAGGCAATACCACCGGT	668
Sbjct CDS:extended-spectru	1512894 201	GCATTGGGCGACAGCCAAC	GGGCGCAGCTGGTGACATG	GATGAAAGGCAATACCACCGGT M K G N T T G	1512953
CDS: Putative 1 Query	221 661	A A S I Q A GCAGCGAGCATTCAGGCTG	G L P A S W V GACTGCCTGCTTCCTGGGT	V G D K T G S TGTGGGGGGATAAAACCGGCAGC	720
Sbjct CD5:extended-spectru	1512954 221	GCAGCGAGCATTCAGGCTG	GACTOCCTGCTTCCTGGGT	TGTGGGGGATAAAACCGGCAGC	1513013
CDS: Putative 1 Query	241 721	G G Y G T T GGTGGCTATGGCACCACCA	N D I A V I H ACGATATCGCGGTGATCTG	P K D R A P L	788
Sbjct CD5:extended-spectru	1513014 241	G G V G T T	ACGATATCGCGCGCGCGATCTG	P K D R A P L	1513073
CDS: Putative 1 Query	261 781	I L V T Y F		E S R R D V L AGAAAGCCGTCGCGATGTATTA	840
Sbjct CDS:extended-spectru	1513074 261	ATTCTCGTCACTTACTTCA	CCCAGCCTCAACCTAAGGC T Q P Q P K A	AGAAAGCCGTCGCGATGTATTA E S R R D V L	1513133
CDS: Putative 1 Query	281 841	A S A A K I GCGTCGGCGGCTAAAATCG	TCACCGACGGTTTGTAA	876	
Sbjct CDS:extended-spectru	1513134 281	GCGTCGGCGGCTAAAATCG	TCACCGACGGTTTGTAA V T D G L	1513169	

Figure (4-19) NCBI blasting pairwise alignment of the sample exposed to (0.08T) of magnetic power sequences query of beta-lactamase (CTX_{M-1}) gene with the subject of *Escherichia coli*

Escherichia coli strain EC488 chromosome, complete genome Sequence ID: <u>CP109674.1</u> Length: 5173170 Number of Matches: 1

Range 1: 1512294 to 1513169 GenBank Graphics V Next Match & Privilous Match					
Score 1535 bits(1702)	Expect 0.0	Identities 866/876(99%)	Gaps 0/876(0%)	Strand Plus/Plus	
CDS: Putative 1 Query	11	M V K K S L	CCAGTTCACGCTGAAGG	CGACGGCACCCCGTCACGCTGTTG	60
Sbjct CDS:extended-spectru	1512294 1		GCCAGTTCACGCTGATGG	CGACGGCAACCGTCACGCTGTTG	1512353
CDS: Putative 1 Query	21 61	L G C V P L	A Q T A D	V Q Q K L A E L TACAGCAAAAAACTTGCCGAATTA	120
Sbjct CDS:extended-spectru	1512354 21	TTAGGAAGTGTGCCGCTGT	ATGCGCAAACGGCGGACG	TACAGCAAAAACTTGCCGAATTA	1512413
CDS: Putative 1 Query	41 121	E R Q S G G I GAGCGGCAGTCGGGAGGCA	R L G V A L GACTGGGTGTGGGCATTGA	I N T A D N S Q TTAACACAGCAGATAATTCGCAA	180
Sbjct CDS:extended-spectru	1512414 41	GAGCGGCAGTCGGGAGGCA	GACTOGGTGTGGCATTGA	TTAACACAGCAGATAATTCGCAA I N T A D N S Q	1512473
CDS: Putative 1 Query	61 181	I L Y R A D ATACTITATCGTGCTGATG	AGCGCTTTGCGATGTGCA	S T S K V M A A GCACCAGTAAAGTGATGGCCGCG	240
Sbjct CDS:extended-spectru	1512474 61	ATACTITATCGTGCTGATG	AGCGCTTTGCGATGTGCA	GCACCAGTAAAGTGATGGCCGCG S T S K V M A A	1512533
CDS: Putative 1 Query	81 241	A A V L K K GCCGCGGTGCTGAAGAAAA	S E S E P N GTGAAAGCGAACCGAATC	L L N Q R V E I TGTTAAATCAGCGAGTTGAGATC	300
Sbjct CDS:extended-spectru	1512534 81	A A V L K K	GTGAAAGCGAACCGAATC	TGTTAAATCAGCGAGTTGAGATC	1512593
CDS: Putative 1 Query	101 301	K K S D V V I	N Y N P I A	E K H V N G T M AAAAGCACGTCAATGGGACGATG	368
Sbjct CDS:extended-spectru	1512594 101	K K S D L V	N Y N P I A	AAAAGCACGTCAATGGGACGATG	1512653
CDS: Putative 1 Query	121 361	S L A E L S C	A A A L Q Y	S D N V A M N K GCGATAACGTGGCGATGAATAAG	428
Sbjct CDS:extended-spectru	1512654 121	TCACTOGCTGAGCTTAGCG	COCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCCC	GCGATAACGTGGCGATGAATAAG S D N V A M N K	1512713
CDS: Putative 1 Query	141 421	CTGATTGCTCACGTTGGCG	GCCCGGCTAGCGTCACCG	A F A R Q L G D CGTTCGCGCGACAGCTGGGAGAC	480
Sbjct CDS:extended-spectru	1512714 141	CTGATTGCTCACGTTGGCG	GCCCGGCTAGCGTCACCG	CGTTCGCCCGACAGCTGGGAGAC A F A R Q L G D	1512773
CDS: Putative 1 Query	161 481	E T F R L D GAAACGTTCCGTCCCGACC	R T E P T L GTACCGAGCCGACGTTAA	N T A V P G D P ACACCGCCGTTCCGGGCGATCCG	540
Sbjct CDS:extended-spectru	1512774 161	GAAACGTTCCGTCTCGACC	T E P T L	ACACCGCCATTCCGGGCGATCCG	1512833
CDS: Putative 1 Query	181 541	R D T T S P C	R A M A Q T	L R N L T L G K TGCGGAATCTGACGCTGGGTAAA	688
Sbjct CDS:extended-spectru	1512834 181	CGTGATACCACTTCACCTC	GGGCAATGGCGCAAACTC	TGCGGAATCTGACGCTGGGTAAA L R N L T L G K	1512893
CDS: Putative 1 Query	201 601	A L G D S Q I	GGGCGCAGCTGGTGACAT	N M K G N T T G GGATGAAAGGCAATACCACCGGT	660
Sbjct CDS:extended-spectru	1512894 201	GCATTGGGCGACAGCCAAC	GGGCGCAGCTGGTGACAT	GGATGAAAGGCAATACCACCGGT N M K G N T T G	1512953
CDS: Putative 1 Query	221 661	A A S I Q A GCAGCGAGCAGTTCAGGCTG	G L P A S W GACTGCCTGCTTCCTGGG	V V G D K T G S TTGTGGGGGGATAAAACCGGCAGC	728
Sbjct CDS:extended-spectru	1512954 221	GCAGCGAGCATTCAGGCTG	GACTOCCTOCTTCCTOOG	TTGTGGGGGGATAAAACCGGCAGC V V G D K T G S	1513013
CDS: Putative 1 Query	241 721	G G Y G T T I GGTGGCTATGGCACCACCA	N D I A V I ACGATATCGCGGTGATCT	W P K D R A P L GGCCAAAAGATCGTGCGCCGCTA	780
Sbjct CDS:extended-spectru	1513014 241	G G V G T T	ACGATATCGCGGTGATCT	GGCCAAAAGATCGTGCGCCGCTG W P K D R A P L	1513073
CDS: Putative 1 Query	261 781	ATTCTGGTCACTTACTTCA		A E S P R D V L CAGAAAGCECTCGCGATGTATTA	840
Sbjct CDS:extended-spectru	1513074 261	ATTCTGGTCACTTACTTCA	CCCAGCCTCAACCTAAGG	CAGAAAGCCGTCGCGATGTATTA A E S R R D V L	1513133
CDS: Putative 1 Query	281 841	GCGTCGGCGGCTAAAATCG	TCACCGACGGTTTGTAA	876	
Sbjct CDS:extended-spectru	1513134 281	GCGTCGGCGGCTAAAATCG	T D G L	1513169	

Figure (4-20) NCBI blasting pairwise alignment of beta-lactamase (CTX_{M-1}) gene sequences from an *Escherichia coli* sample exposed to (0.12T) of magnetic power.

Escherichia coli strain EC488 chromosome, complete genome Sequence ID: <u>CP109874.1</u> Length: 5173170 Number of Matches: 1

Range 1: 1512294 to 1513169 Genetiank Graphics The Next Platter & Previous Match					
Score 1544 bits(1712)	Expect 0.0	Identities 868/876(99%)	Gaps 0/876(0%)	Strand Plus/Plus	
CDS: Putative 1 Query	1	ATGGTTAAAAAATCACTGC	SCCAGTTCACGCTGAAGG	CGACGGCACCCGTCACGCTGTTG	60
Sbjct CD5:extended-spectru	1512294 1		CCAGTTCACGCTGATGG	CGACGGCAACCGTCACGCTGTTG	1512353
CDS: Putative 1 Query	21 61		A Q T A D	V Q Q K L A E L TACAGCAAAAAACTTGCCGAATTA	120
Sbjct CD5:extended-spectru	1512354 21	TTAGGAAGTGTGCCGCTGT	TOCOCAAACOGCOGACO	TACAGCAAAAAACTTGCCGAATTA V Q Q K L A E L	1512413
CDS: Putative 1 Query	41 121	E R Q S G G I	A L G V A L	I N T A D N S Q TTAACACAGCAGATAATTCGCAA	180
Sbjct CDS:extended-spectru	1512414 41	GAGCGGCAGTCGGGAGGCA	ACTOGOTOTOGCATTOA	TTAACACAGCAGATAATTCGCAA I N T A D N S Q	1512473
CDS: Putative 1 Query	61 181	I L Y R A D I	GCGCTTTGCGATGTGCA	S T S K V M A A GCACCAGTAAAGTGATGGCCGCG	240
Sbjct CDS:extended-spectru	1512474 61	ATACTITATCGTGCTGATG	COCTITICCATOTICA	GCACCAGTAAAGTGATGGCCGCG	1512533
CDS: Putative 1 Query	81 241	A A V L K K	S E S E P N STGAAAGCGAACCGAATC	L L N Q R V E I TGTTAAATCAGCGAGTTGAGATC	300
Sbjct CD5:extended-spectru	1512534 81	GCCGCGGTGCTGAAGAAAAA	TGAAAGCGAACCGAATC	TGTTAAATCAGCGAGTTGAGATC	1512593
CDS: Putative 1 Query	101 301	K K S D L V I	Y N P I A	E K H V N G T M AAAAGCACGTCAATGGGACGATG	368
Sbjct CDS:extended-spectru	1512594 101	K K S D L V I	kétátáátécsáttséssá	AAAAGCACGTCAATGGGACGATG E K H V N G T M	1512653
CDS: Putative 1 Query	121 361	S L A E L S A	A A L Q Y	S D N V A M N K GCGATAACGTGGCGATGAATAAG	428
Sbjct CDS:extended-spectru	1512654 121	TCACTGGCTGAGCTTAGCG	GOCCOCCCTACAGTACA	GCGATAACGTGGCGATGAATAAG S D N V A M N K	1512713
CDS: Putative 1 Query	141 421	CTGATTGCTCACGTTGGCG	S P A S V T	A F A R Q L G D CGTTCGCCCGACAGCTGGGAGAC	488
Sbjct CDS:extended-spectru	1512714 141	L I A H V G (SCCCGGCTAGCGTCACCG	CGTTCGCCCGACAGCTGGGAGAC A F A R Q L G D	1512773
CDS: Putative 1 Query	161 481	E T F R L D I	T E P T L	N T A V P G D P ACACCGCCGTTCCGGGCGATCCG	548
Sbjct CDS:extended-spectru	1512774 161	GAAACGTTCCGTCTCGACCO	T E P T L	ACACCGCCATTCCGGGCGATCCG	1512833
CDS: Putative 1 Query	181 541	R D T T S P I	GGGCAATGGCGCAAACTC	L R N L T L G K TGCGGAATCTGACGCTGGGTAAA	600
Sbjct CDS:extended-spectru	1512834 181	COTGATACCACTTCACCTCO	SGGCAATGGCGCAAACTC	TGCGGAATCTGACGCTGGGTAAA L R N L T L G K	1512893
CDS: Putative 1 Query	201 601	A L G D S Q I GCATTGGGCGACAGCCAACC	A Q L V T	W M K G N T T G GGATGAAAGGCAATACCACCGGT	660
Sbjct CDS:extended-spectru	1512894 201	GCATTGGGCGACAGCCAACC	GOCCCACCTGGTGACAT	GGATGAAAGGCAATACCACCGGT W M K G N T T G	1512953
CDS: Putative 1 Query	221 661	A A S I Q A G	S L P A S W	V V G D K T G S TTGTGGGGGGATAAAACCGGCAGC	728
Sbjct CDS:extended-spectru	1512954 221	GCAGCGAGCATTCAGGCTG	ACTOCCTOCTTCCTOGG	TTGTGGGGGATAAAACCGGCAGC V V G D K T G S	1513013
CDS: Putative 1 Query	241 721	G G Y G T T I GGTGGCTATGGCACCACCA	D I A V I ACGATATCGCGGTGATCT	W P K D R A P L GGCCAAAAGATCGTGCGCCGCTA	788
Sbjct CDS:extended-spectru	1513014 241	G G Y G T T I	CGATATCGCCGCTGATCT	GGCCAAAAGATCGTGCGCCGCTG W P K D R A P L	1513073
CDS: Putative 1 Query	261 781	I L V T Y F		A E S P R D V L CAGAAAGCCCTCGCGATGTATTA	840
Sbjct CDS:extended-spectru	1513074 261	ATTCTGGTCACTTACTTCA	CCASCCTCAACCTAAGG	CAGAAAGCCGTCGCGATGTATTA A E S R R D V L	1513133
CDS: Putative 1 Query	281 841	A S A A K I G	T D G L . CACCGACGGTTTGTAA	876	
Sbjct CDS:extended-spectru	1513134 281	A S A A K I	CACCGACGGTTTGTAA	1513169	

Figure (4-21) NCBI blasting pairwise alignment of beta-lactamase (CTX_{M-1}) gene sequences from an *Escherichia coli* sample exposed to (0.16T) of magnetic power.

in a

Table (4-4) GenBank accession no. of both gene *blaTEM* and *blaCTX-M* of *Escherichia coli*.

Bacterial name	Accession No.	Gene name
Escherichia coli	OQ135092	beta lactamase (<i>blaTEM</i> ₁)
Escherichia coli	OQ135093	beta lactamase ($blaTEM_1$)
Escherichia coli	OQ135094	beta lactamase (<i>blaTEM</i> ₁)
Escherichia coli	OQ135095	beta lactamase (<i>blaTEM</i> ₁)
Escherichia coli	OQ135096	beta lactamase ($blaTEM_1$)
Escherichia coli	OQ135097	beta lactamase ($blaCTX$ - $_{M-1}$)
Escherichia coli	OQ135097	beta lactamase ($blaCTX$ - $_{M-1}$)
Escherichia coli	OQ135097	beta lactamase ($blaCTX$ - $_{M-1}$)
Escherichia coli	OQ135097	beta lactamase ($blaCTX$ - $_{M-1}$)
Escherichia coli	OQ135097	beta lactamase ($blaCTX$ - $_{M-1}$)

Table (4-5) Explain the numbers and variant position of nucleotides of each sample are changed with amino acid codons in $blaTEM_1$ and $blaCTX_{-M-1}$ genes of *E. coli*.

Samples	Gene name and position sequences	Variant position	Nucleotide changed	Amino acid changed	Codo n numb er chang ed	GenBank Reference accession number
OQ135092	<i>blaTEM</i> (15027- 15529)	Wild	Wild	Wild type	Wild	CP095547.1
OQ135093	<i>blaTEM</i> (15027- 15529)	15029 15052	A→T C→T	Q→L /Glutamine->Leucine P→S /Proline-> Serine	First 9	CP095547.1
OQ135094	<i>blaTEM</i> (15027- 15529)	15029 15052	A→T C→T	Q→L/Glutamine-> Leucine P→S/ Proline-> Serine	First 9	CP095547.1
OQ135095	<i>blaTEM</i> (15027- 15529)	15505- 15507	TGG→GAA	W→E/Tryptophan>Glutamate	160	CP095547.1
OQ135096	<i>blaTEM</i> (15027- 15529)	15505- 15507	TGG→GAA	W→E/Tryptophan>Glutamate	160	CP095547.1
OQ135097	<i>blaCTX-</i> м 1512294- 1513169	Wild	Wild	Wild	Wild	CP109874.1
OQ135098	<i>blaCTX-_M</i> 1512294- 1513169	1512320 1512331 1512357 1512360	$T \rightarrow A$ $A \rightarrow C$ $G \rightarrow C$ $A \rightarrow C$	$M \rightarrow K/$ Methionine->Lysine $T \rightarrow P$ /Threonine-> Proline $G \rightarrow R$ /Glycine-> Arginine $S \rightarrow R$ Serine -> Arginine	12 16 22 23	CP109874.1
OQ135099	$blaCTX_{-M}$ 1512294- 1513169	1512320 1512421	T→A A→C	$M \rightarrow K$ /Methionine->Lysine $Q \rightarrow P$ / Glutamine-> Proline	12 43	CP109874.1
OQ135100	<i>blaCTX-_M</i> 1512294- 1513169	1512320 1512331 1512360 1512606 1512819 1513106 1513120	$T \rightarrow A$ $A \rightarrow C$ $A \rightarrow C$ $C \rightarrow G$ $A \rightarrow G$ $T \rightarrow A$ $G \rightarrow C$	$M \rightarrow K$ /Methionine->Lysine $T \rightarrow P$ /Threonine-> Proline $S \rightarrow R$ /Serine -> Arginine $L \rightarrow V$ / Glutamine-> Valine $I \rightarrow V$ / Isoleucine -> Valine Silent $R \rightarrow P$ / Arginine -> Proline	12 16 23 105 176 272 276	CP109874.1
OQ135101	<i>blaCTX-_M</i> 1512294- 1513169	1512320 1512331 1512360 1512819 1513106 1513120	$T \rightarrow A$ $A \rightarrow C$ $A \rightarrow C$ $A \rightarrow G$ $T \rightarrow A$ $G \rightarrow C$	$M \rightarrow K/Methionine ->Lysine$ $T \rightarrow P/Threonine ->Proline$ $S \rightarrow R/Serine -> Arginine$ $I \rightarrow V/Isoleucine -> Valine$ Silent $R \rightarrow P/Arginine -> Proline$	12 16 23 176 272 276	CP109874.1

4.7.3. Alignment and detection of variant amino acid

Among 5 summited sequences (first sample control and other 4 samples treated to magnetic power (0.04, 0.08, 0.12 and 0.16T) respectively of *blaTEM*₁ *Escherichia coli* appear totally three different variations in amino acids according to alignment in the program of MEGA version of 11 (is a computer software for conducting statistical analysis of molecular evolution and for constructing phylogenetic trees)Figure(4-22 a and b),there are changed new amino acids in a different position in which codon number one nucleotide A changed to T this led to change Glutamine to Leucine and in codon nine nucleotide C changed to T this led to the change of Proline to Serine also in codon number 160 nucleotide TGG to GAA this led to the change of Tryptophan to Glutamate.

Protein Sequences																															
Species/Abbrv	Δ	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*		*	*	*	*	*	*	*
1. OQ135092		3	G	Ρ	к	E	L	т	А	F	L	Η	N	М	G	D	H	۷	т	R	L	D	R	W	E	Ρ	E	L	N	Е	А
2. OQ135093		3	G	Ρ	K	E	L	т	А	F	L	Н	N	М	G	D	H	۷	т	R	L	D	R	W	E	Ρ	Е	L	N	Е	А
3. OQ135094		3	G	Ρ	ĸ	E	L	т	А	F	L	Н	N	М	G	D	Н	۷	т	R	L	D	R	W	E	Ρ	Е	L	N	Е	А
4. OQ135095		3	G	Ρ	к	E	L	т	А	F	L	Н	N	М	G	D	Н	۷	т	R	L	D	R	Е	Е	Ρ	Е	L	N	Е	А
5. OQ135096		3	G	Ρ	к	E	L	Т	А	F	L	Н	N	М	G	D	Н	۷	т	R	L	D	R	E	E	Ρ	E	L	N	E	A

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pecies/Abbrv		*	*	*	*	*	*	*		*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*	*
0Q135092	Q	H	F	R	۷	А	L	I	Ρ	F	F	A	A	F	С	L	Ρ	v	F	A	H	Ρ	E	т	L	۷	к
OQ135093	L	Н	F	R	v	А	L	I	s	F	F	A	А	F	С	L	Ρ	v	F	A	н	Ρ	Е	Т	L	v	к
OQ135094	L	H	F	R	۷	А	L	1	s	F	F	A	А	F	С	L	Ρ	v	F	A	H	Ρ	E	т	L	۷	к
OQ135095	Q	H	F	R	۷	А	L	I	Ρ	F	F	A	А	F	С	L	Ρ	v	F	A	H	Ρ	E	т	L	۷	к
OQ135096	Q	н	F	R	v	А	L	1	Ρ	F	F	А	А	F	С	L	Ρ	v	F	А	н	Ρ	Е	Т	L	v	к

⁽b)

Figure (4-22) (A), (B) Multiple protein sequence alignment analysis of $blaTEM_1$ gene among 5 (first sample Negative control and other 4 samples treated to magnetic power (0.04, 0.08, 0.12 and 0.16T) respectively *Escherichia coli*.

4.7.4. Phylogenetic inferences

Among of 5 summited sequences (first sample control and other 4sample treated to magnetic power (0.04, 0.08, 0.12 and 0.16T) respectively of *blaCTX*-_{M-1}Escherichia coli appear totally eight different variation amino acid according to alignment in program of MEGA version of 11 (is a software program used for statistical study of molecular evolution and the construction of phylogenetic trees) Figure (4-23 a, and b), there are changed new amino acids in different position in which codon number 12 nucleotide T changed to A this led to change Methionine to Lysine and in codon 16 nucleotide A changed to C this led to change of Threonine to Proline also in codon number 22 nucleotide G to C this led to change of Glycine to Arginine and in codon number 23 nucleotide A to C this led to change of Serine to Arginine and in codon number 43 nucleotide A to C this led to change of Glutamine to Proline and in codon number 105 nucleotide C to G this led to change of Glutamine to Valine also in codon number 176 nucleotide A to G this led to change of Isoleucine to Valine and in codon number 276 nucleotide G to C this led to change of Arginine to proline.

Protein Sequence	5																																						
Species/Abbrv		٠	•		*		•		•		•	•	•	*	•	•						•			•	•	•	•			•		•	• •			*		
1. 00135097	M	۷	ĸ	K	s	L	R	Q	F	Т	L	M	A	T ,	A	T١	1	L	. L	L	G S	V	P	L	Y	A	a 1	A	D	۷	Q	Q	к	LA	E	L	E	R	QS
2. 00135098	М	٧	ĸ	ĸ	s	L	R	Q	F	т	L	ĸ	A	Т	A	T۱	/ 1	L	. L	L	G S	V	P	L	Y	A	a 1	A	D	۷	Q	Q	к	LA	E	L	E	R	PS
3. 00135099	М	v	ĸ	к	s	L	R	Q	F	т	L	к	A	T ,	A	P	/ 1	L	. L	L	RF	۱	P	L	Y	A	a 1	A	D	۷	Q	Q	к	LA	E	L	E	R	QS
4. 00135100	М	v	ĸ	ĸ	s	L	R	Q	F	т	L	ĸ	A	T,	4	P	/ 1	L	. L	L	G	2 V	P	L	Y	A	a 1	A	D	٧	Q	۵	к	LA	E	L	E	R	Q S
5. 00135101	M	٧	ĸ	ĸ	s	L	R	۵	F	Т	L	ĸ	A	г	A	P۱	/ 1	L	. L	L	G	: v	P	L	Y.	A	۵ 1	A	D	۷	Q	۵	ĸ	L A	E	L	Ε	R	QS
Proteir	1 5	Se	20	q	u	e	r	14	-	=	5		_		_			<i>a</i>)										_		_		_		_		_		_	
Species	s//	~	b	b		Y					ų	*		*		*		*		*	1	*	1	•	-	•	-	•	-	•	*		-		*		*	L	*
1. 0Q1	35	0	2	97	7						_	ч		•	•	^	•	1		Р	<pre></pre>	з		>	F	>	F	2		•	Т		-		s		Р		R
2. OQ1	35	0	2	98	8							ч		•	•	^	•	1		Р	1	з		>	F	>	F	٤		•	Т		Т		s		Р		R
3. OQ1	35	0	19	99	Э							ч		•	1	^	•	1		Р	<	3	E	>	F	>	F	٤		•	Т		T		s		Р		R
4. OQ1	35	1	0	00	D							ч		•		~	•	V	r	P	<	з	E	>	F	>	F	٤	D		Т				s		Р		R
5. OQ1	35	1	0	01	1							ч	-	T		~	٠ ·	V	r	Р		з	E		F	>	F	٤	D		Т				s		Р		R

(b)

Figure (4-23) (a),(b) Multiple protein sequence alignment analysis of *blaCTX*- $_{M-1}$ gene among five first sample control and other 4sample treated to magnetic power (0.04, 0.08, 0.12 and 0.16T) respectively *Escherichia coli*.

MEGA 11 program of Phylogenetic analysis with more than 50% automatic program resampling (Boost strap) based on $blaTEM_1$ gene among five samples (first sample was control and other 4 samples treated to magnetic power (0.04, 0.08, 0.12 and 0.16T) respectively of *E. coli* revealed grouping of 5 investigated different mutations on expected lines. Based on sequence divergence similarity data and a constructed phylogeny, it was shown that mutations in five sample sequences belonging to distinct genera were closely related. The unmuting samples of various grouped in one cluster with high similarity of GenBank reference sequences also phylogenetics appeared genetic distance between them were 0.00-0.01 percentage according to GenBank sequences Figure (4-24).



Figure (4-24) Employing Maximum Likelihood with boost strap with red numbers of Mega 11 program show phylogenetic positioning of each mutant of 5 samples (first sample control and other 4sample treated to magnetic power (0.04, 0.08, 0.12 and 0.16T) respectively samples with similar GenBank sequences bla*TEM*₁ among five isolates of *Escherichia coli*.

MEGA 11 programs of Phylogenetic analysis with more than 50% automatic program resampling (Boost strap) based on *blaCTX-_{M-1}* gene among 5 (first sample control and other 4 samples treated to magnetic power (0.04, 0.08, 0.12 and 0.16T) respectively *Escherichia coli* revealed grouping of 5 (first sample control and other 4sample treated to magnetic power (0.04, 0.08, 0.12 and 0.16T) respectively investigated different mutations on expected lines. Based on sequence divergence similarity data and a constructed phylogeny, it was shown that mutations in five sample sequences belonging to distinct genera were closely related. The not mutant specimen several grouped in one cluster with high similarity of into GenBank reference sequences. Also, phylogenetic appeared genetic distance between them was 0.00-0.01 percentage according to GenBank sequences Figure (4-25).



Figure (4-25) Employing Maximum Likelihood with Boost strap with red numbers of Mega 11 program show phylogenetic positioning of each mutant of 5 samples (first sample control and other 4 samples treated to magnetic power (0.04, 0.08, 0.12 and 0.16T)respectively samples with similar GenBank sequences *blaCTX-*_{*M-1*} gene among 5 *Escherichia coli*.

Chapter Five DISCUSSION

5. DISCUSSION

Living organisms may respond to changes in external magnetic fields, especially the Earth's magnetic field, as has often been shown analytically (Zablotskii *et al.*, 2018; Li *et al.*, 2020). Any physiological response to a magnetic field that has been seen involves a complicated series of intracellular metabolic changes (Albuquerque *et al.*, 2016).

Urinary tract infections frequently involve the *E.coli* bacteria, which has a high potential for epidemic spread. There is a rise in the distribution of these bacteria's extended-spectrum beta-lactam antibiotic-resistant strains. The genetic variants and intragenic alterations that code for these resistances are poorly understood. The study aimed to characterize genetic variants and explain the intragenic alterations causing resistance to extend -spectrum beta-lactam antibiotics in sequenced uropathogenic *E.coli*. The polymerase chain reaction was used to find the ESBLs genes. The extended-spectrum - lactamase CTX_{-M-1} gene, encoded by the $blaTEM_I$ gene, dominated the resistance genes discovered using BLAST for sequence similarity and homology on nucleotide sequences (ABE *et al.*, 2021; Hussain *et al.*, 2021).

The study examined the impact of an SMF (0.04, 0.08, 0.12 and 0.16T) on *E.coli* growth, viability, and differential expression of CTX_{M-1} , *TEM*, *SHV*, and *16SrRNA* genes.

Static magnets were used to conduct an experiment on *E. coli* where the biological effects of a static magnetic field were studied. In order to analyze the impacts, Bacteria were cultivated and their growth rate and maximum number were calculated.

In this work, a magnetic field was used to inhibit the growth of *E.coli* under the impact of static magnetic force (0.04, 0.08, 0.12 and 0.16T) respectively, represents the amount of light absorbed by the growing medium harboring *E.coli* bacterium. A higher absorbance denotes that there are more

bacteria in the growing media. Study Results indicated that increasing of static magnetic field reduced the growth rate of bacteria in the growth medium, which supports the prior outcome. This outcome supports that which was found by (Bajpai *et al.*, 2014; Bajpai *et al.*, 2012). These findings show that the effects of a static magnetic field on the rate of bacterial growth are variable on the type of bacteria, which is compatible with the research of (Bajpai *et al.*, 2014).

E. coli's growth was affected by the magnetic field (gram-negative). The results of earlier investigations do not entirely support the impact of static magnetic field on the development of *E.coli* bacteria (Bajpai *et al.*, 2012; Bajpai *et al.*, 2014) indicated that the magnetic field inhibits the growth of the bacterium *E. coli*. Several investigations, including (Haghi *et al.*, 2012), observed that the magnetic fields had no impact on bacterial growth. Al-Khaza'leh and Al-fawwaz (2015) showed that, after 24 hours, providing a static magnetic field with effects of 30, 50, and 80 mT each inhibited the growth of *E.coli* bacteria. It is noticeable that increasing the magnetic field's power inhibits the growth of the *E.coli* bacterium. Strašák *et al.* (2002) showed that the magnetic induction's magnitude varied in the range of 2.7 to 10 mT when the bacteria were exposed to the magnetic field. They reported yet again that the number of bacteria in the exposed culture decreased exponentially.

Alterations in antibiotic sensitivity were seen since resistance. The sensitivity of *E.coli* cells to various antibiotics Ceftazidime, Gentamycin, Minocycline, Trimethoprim/Sulfamethoxazole, Piperacillin, and Cefepime. While other antibiotics changed from sensitive to resistant when exposed to different powers of magnetic field, Ciprofloxacin, Trimethoprim/Sulfamethoxazole, Ceftazidime, Ceftazidime, Cefepime, Aztreonam, and Trimethoprim/Sulfamethoxazole.In the other hand the rate of MIC of antibiotics (minimum inhibitory concentration) converted as compared to the

unexposed MIC of the (negative control). These results corresponded with a study that found that moderate-intensity static fields could lead to an alteration in resistance of *E.coli* and sensitivity. In addition, the study found that the possibility of a magnetic field interfering with the charge on the antibiotic molecule or surface charges of the membrane altering the antibiotic penetration rate may exist (Segatore *et al.*, 2012).

These outcomes were similar to research that showed that static fields of moderate power might altered *E.coli* sensitivity and resistance. Additionally, it was found that there might be a chance that a magnetic field may affect an antibiotic's charge or a membrane's surface charge, changing how quickly the antibiotic penetrated the member (Mousavian-Roshanzamir and Makhdoumi-Kakhki, 2017).

However, the magnetic field may also be associated to other particular processes that aid in bacterial adaptation to new environments. As a result, the bacteria can respond to environmental challenges by stimulating specific inducible systems, such as the DNA repair system, and subsequently destroying processes that increase the variation of genes (Albalawi, 2017).

All of these findings suggest that the application of a static magnetic power has an impact on the drug's mode of action on bacterial cells through inhibition of the formation of cell walls, proteins, nucleic acids, vital enzymes, and changes in barrier function (Karimi *et al.*, 2016; Fu *et al.*, 2023).

Moreover, (Stansell *et al.*, 2001) reported that subjecting bacteria to a medium-strong magnetic field might drastically alter their susceptibility to antibiotics. He also demonstrated that exposing *E.coli* to magnetic fields significantly increased antibiotic resistance.

The biochemical investigation were identified using the VITEK technique the all isolated strains of *E.coli* were SUCCINATE alkalinization (SUCT) test altered after exposed to different power of magnetic field. Also L-LACTATE

alkalinization (ILATk) test an apparent recognized characteristic to distinguish among samples and after treatment with different powers of magnetics converted with compared to untreated sample (control), however, the Tyrosine Arylamidase (TyrA) test, alpha-galactosidase (AGAL) test, beta-glucuronidase (BGUR) test, L-Proline A (ProA) test, and O/129 Resistance (O129R) test fermentation converted after treated to magnetic field with compared to control.

According to the results of certain biochemical investigations, magnetic fields positively affect biochemical characteristics. The impact of the magnetic field on the bacterial enzymes lactose, trehalose, sucrose, mannitol, acetyl-glucosamine, and maltose were observed 24 hours after incubation. The study found that the cellular membrane of the bacterium was affected by the magnet fields (Lister and Horswill, 2014). The intensity of the response cause the change in the internal composition of the cells (Albalawi, 2017).

Transcriptomic investigation showed that the down regulated expression of the *glc* operon, which is important in essential carbon consumption, was closely associated to the poor growth of SMF-exposed bacteria. Two additional operons, *glp FKX* (involved operons responsible for glycerol breakdown) and *ast* CADBE (involved in Arginine catabolism), were also shown to be highly down-regulated in the SMF-treated bacteria in addition to the *glc* operon (Olvera *et al.*, 2009; Shiwa *et al.*, 2020). Since succinct, an intermediary of the TCA cycle and nitrogen are both provided by the *AST* (Arginine succinyl transferase) route, nitrogen, and carbon starvation function to dynamically control the *ast* operon's expression (Förster and Gescher, 2014).

It's significant to note that when Arginine was employed as a nitrogen source, glycerol substantially stimulated the *ast* operon promoter in continuously expanding cells (Förster and Gescher, 2014), and the *glp* and *ast* operons are expressed in a tightly controlled manner. Future research will

need to focus on how SMF coordinately regulates the glyoxylate, glycerate, and *AST* mechanisms (Li *et al.*, 2022)

This result suggests that the inhibitory action of SMF is mediated by alternatively expressed genes (DEGs) that are principally concerned in carbon source utilization (Xie *et al.*, 2022).

Escherichia coli cultures were exposed to different powers of static magnetic forces in order to analyze any alterations that may have been generated in cellular growth and gene expression. A reference housekeeping gene whose expression is unaltered by the applied stress is necessary for this strategy. As a viable reference gene to standardize the differential analysis, the *16SrRNA* gene was selected.

Twenty five of clinical specimens were screened for UTI. It was found technique. By using 2% agarose gel electrophoresis, the amplified products were separated.

The PCR product with gel electrophoreses for all isolated *E.coli* for using primers of *16SrRNA* gene was positive; these results indicated that all uropathogenic *E.coli* isolates were positive for the presence of the *16SrRNA* gene at 1343bp.Its expression was constant during exposure. The result demonstrated that the expression level of the *16SrRNA* the PCR product with gel electrophoreses was %100 (positives) and remained stable during the SMF exposure to different forces it is thus a reference gene for studying the differences in the gene expression of different strains of *E.coli*

PCR result for $TEM_{1,}$ bla $CTX_{-M-1,}$ and blaSHV reduced with compared to different exposures to Magnetic field, that's mean that magnetic field act mutagenic effect leads to mutation of ESBLs genes of *E*.*coli* stains.

The magnetic field acts mutagenic effect leading to the mutation of ESBLs genes of *E.coli* stains or switching off the genes. The findings demonstrate SMF-induced changes in gene detection and cell proliferation as compared to control groups (Overdevest *et al.*, 2011).

Bacteria can undergo mutations, the same as any other type of organism. A mutation is any modification to the DNA nucleotide sequence of the genome. Mistakes in DNA replication, exposure to radiation or foreign substances, and the insertion or deletion of DNA segments are the major causes of mutations (Watford and Warrington, 2017).

A mutation is an irreversible change to a gene's DNA sequence. A gene's DNA sequence mutations can change the protein's amino acid sequence (Kapli et al., 2023). Throughout time, mutations can arise. This happens when external forces like magnetic fields damage DNA or after a cell replicates its DNA mistakenly before multiplying (Vanderstichelen, 2022). There are several types of bacterial mutations, including missense, nonsense, silent, frame shift, lethal, suppressor, and conditional lethal (Kundu, 2022). Mutation is an essential concept in modern biology that causes gene variations. A mutation is a permanent change to the sequence of nitrogenous bases in the molecular structure of DNA. Generally, mutations lead to alterations to the end product specified by a gene (Basu and Essigmann, 2022). In some instances, a mutation can be advantageous if a microorganism acquires a new metabolic activity, or it can be detrimental if a metabolic activity is lost. Mutations can be sporadic or induced by an environmental mutagen. Mispairing is most likely caused by cellular processes such as the Tautomeric shift of bases, oxidative damage to DNA, Depurination, and Deamination, or by "environmental" factors (Mutagens) such as chemicals, radiation, viruses, diet, and lifestyle (Wang et al., 2022). Substitution and deletion or addition of nucleotides is two mechanisms of mutation. There are several types of bacterial mutations, including missense, nonsense, silent, frame shift, lethal, suppressor, and conditional lethal. Methods for detecting these mutations are required for their identification. Classic techniques PCR, gel electrophoresis, gene probes, Southern blotting, DNA sequencing, and DNA microarray are some of these methods (Kundu, 2022).

Mutations cause variations in genes, which are crucial to evolution. A mutation is any variation in DNA sequence that is inherited. Mutation can be either beneficial or harmful. Mutations can occur spontaneously owing to cellular processes or be induced by an environmental mutagen (Ray, 2022). Substitution, deletion, or addition of nucleotides is two mechanisms of mutation. Mutation in bacteria may influence the phenotypic. Mutant detection techniques include molecular approaches. High sensitivity and specificity, and faster than conventional procedures, are suggested (Watford and Warrington, 2017; Sánchez-Romero and Casadesús, 2020).

To identify these mutations, detection methods are required. *E.coli* mutant strains were treated to homogenous static magnetic fields of 500 mT or 3 T. No evidence of enhanced DNA damage was seen in *E.coli* exposed to SMF, even in strains incapable of DNA repair (Hashim, 2016). In vitro, tests showed magnetic field-DNA interactions, mostly point mutations. The magnetic field may directly interact with DNA or increase oxidant radical activity causing disruption in DNA stability (Chow and Tung, 2000). Other examination of the differential expression of the *16SrRNA*, *rpoA*, *dnaK*, *katN*, and SMF genes under SMF exposure (200 mT, 10 hours) revealed that the *16SrRNA mRNA* expression level remained constant. Interestingly, after 10 hours of SMF exposure (200 mT), the *rpoA*, *katN*, and *dnaK* genes' *mRNA*s were overexpressed (El May *et al.*, 2009). However, when cells were exposed to 300 mT, the quantity of 8-hydroxyguanine in extracted DNA was much lower compared to controls, showing that *S. pyogenes* may have some antioxidant defense at this field strength (Yang *et al.*, 2023).

Chapter Six CONCLUSION AND RECOMMENDATIONS

6. CONCLUSION AND RECOMMENDATIONS

6.1. CONCLUSION

- The magnetic field could change colony morphology, and size, decrease the numbers of bacteria the magnetic field could change the susceptibility of bacteria to certain antibiotics and how change bacterial biological activity on sugar fermentation due to bacterial mutation.
- The magnetic forces could be notably obvious by exposing bacterial cells' growth rate will decrease by increasing the static magnetic field. Results indicated that the bacteria's viability decreased with exposure to SMFs for 24 hrs. All isolates exposed to the magnetic field exhibited altered growth rates compared to the negative control of isolated bacteria.
- ✤ In the molecular study using PCR and molecular sequencing, the results revealed that the detection level of the CTX_{M-I} , TEM_I , and SHV genes under a magnetic field decreased, and the rate of PCR positive result converted if compared with the unexposed magnetic fields. The detection level of the *16SrRNA* the PCR product with gel electrophoreses was %100 (positives) and remained stable throughout SMF exposure (0.04, 0.08, 0.12 and 0.16T) and can thus be used as a reference gene for the analysis of the differential gene expression of *E.coli* strains.
- ★ The result of molecular sequencing was done for $(CTX_{M-1} \text{ and } TEM_1)$ and the result indicated silent, point, and protein mutation due to magnetic field effects. The findings give a significant indicator for choosing the ideal settings to increase therapeutic effects in the potential treatment of illnesses and diseased tissues.

6.2. RECOMMENDATIONS

Results are recommended the following:

- Molecular study to detection of virulence genes of *E.coli* exposed to magnetic power involve *pap*, *sfa* genes.
- ✤ Applying molecular methods to detect fermentation genes of *E. coli* under magnetic fields.
- Subsequent investigation of more ESBLs gene samples for sequencing under magnetic fields.
- Future research will need to focus on how SMF coordinately regulates the glyoxylate, glycerate, and AST mechanisms.
- Molecular analysis of the *E.coli* DNA repair gene under magnetic field involves the *RecA/LexA* genes.

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APPENDICES

APPENDICES

Appendix 1. Interpretation of GN (ID) of *E. coli* (BioMerieux, VITEK 2 System.

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Include On the	Confidence: Very good identification										
Analysis Organisms an	d Tests to	Separate:									
Analysis Messages:											
Contraindicating Typi	al Bionet	tern(s)									
Escherichia coli	an biopati	PHOS(81)	BGUR(83)								
			,								
Susceptibility	Card:	AST-N	417 Lot Number	: 0191915504	Expires:	Feb 23, 2023 12:00 (
Information	Status:	Final	Analysis Tin	Analysis Time: 17.93 hours Comple			ted: Feb 10, 2022 05:28 CST				
Antimicrobia		MIC	Interpretation	Aptimicro	bial	MIC	Interpretation				
Amoxicillin/Clavulanic	Acid	4	S	Iminonam	biai	-0.25	e				
Pineracillin/Tazobactam		<= 4	S	Maronanam		-0.25	0				
Cefazolin		<= 4	S	Amikacin		<= 1	5				
Cefuroxime		4	S	Gentamicin		<= 1	S				
Cefuroxime Axetil		4	S	Ciprofloxacin	-	<= 0.06	s				
Ceftazidime		<= 0.12	S	Fosfomycin		- 0.00					
Ceftriaxone		<= 0.25	S	Nitrofurantoin		<= 16	s				
Cefepime		<= 0.12	S	Trimethoprim/		<= 20	S				
- 36				Sulfamethoxazole							
Ertapenem	-	<= 0.12	S								
AES Findings:	Last M	lodified:	Oct 26, 2021 15:	54 CDT Param	eter Set: CI	.SI+Nati	ural Resistance				
Confidence Level:	Consist	ent									
nstalled VITEK 2 Syster MIC Interpretation Guide AES Parameter Set Name	ns Version line: CLSI : CLSI+Na	9.02 M100-S27 atural Resis	(2017) tance	Therapeutic Inte AES Pa	erpretation Guide arameter Last M	eline: NA1 odified: O	FURAL RESISTANCE et 26, 2021 15:54 CDT Page 1 of 2				

Appendix 3. Interpretation of published article (1)

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RESEARCH ARTICLE

Mode of constant magnetic power effects on Escherichia coli viability and antibiotics activities

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ABSTRACT

Background: The region that a magnetic force has an effect on is known as a magnetic field. Normally, two poles of this field are concentrated. Most magnetic objects are made up of a variety of tiny fields known as domains. There are many different techniques that have been published in the literature for using magnetic energy as a diagnostic tool and for treating illnesses in both humans and animals. Aims: To investigate the effects of different levels of static magnetic field on the ultra structure of *Escherichia coli a* bacterium as well as their antibiotics activities changes. Materials and Method: Locally created dipolar static magnetic field with strength 400, 800, 1200, and 1600 Gauss and used. Between July and October 2022, ten patients with urinary tract infections at Hawler Teaching Hospital and Raparren Hospital for Children in Erbil were isolated for E. coli and then identified by Vitek test. Bacterial culture medium in equal amounts of broth was subjected to the magnetic field for 24 hours. Additionally, treated E. coli culture media (Vitek test) was compared with untreated negative control samples in the bacterial growth subculture, which was checked for bacterial population using spectrophotometer and Vitek diagnosis kit depended on response to different types of antibiotics. Results: An recognized bacterial strain known as E. coli was subjected to magnetic field with two poles pressures of (400, 800, 1200, and 1600) Gausses while it was incubated for 24 hours at a temperature of 37°C. Optical density (O.D.) measurements at 620 nm were used. The results showed that the microorganisms' exposure to the magnetic field produced noticeable alterations on response to different types of antibiotics (Ceftazidime, Azetroname, Ceftazidime, Cefepime, Minocyclin, Azetroname, Ticarcillin/ Clavulanic acid, Azetroname, Piperacillin, Ceftazidime, Cefepime, Ciprofloxacin, Tobramycin, Imipenem, Meropenem, Amikacin, Nitrofurantin, Trimethoprim/ Sulfamethoxazole and Gentamycin) and significantly reduced the number of cells in the exposed bacteria as compared to the control. **Conclusions:** We came to the conclusion that due to bacterial mutation, the magnetic field could alter bacterial response to different types of antibiotics and bacterial population.

Keywords: Bacteria; Optical density; Magnetic field, E.coli. Antibiotic sensitivity



a; Optical density; Magnetic field, E.coll. Antibiotic sensitivity This work is licensed under a Creative Commons Attribution Non-Commercial 4.0

International License.

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Appendix 4. Interpretation of published article (2)

Diyala Journal of	Published: 25 December 2022 Doi: 10.26505/DJM.23026860816
	Molecular detection of static
	magnetic field forces on
	uropathogenic Escherichia coli
	number and fermentation
	activities
	Amanj Jamal Azeez (MBCHB) ¹ , Fouad Hussein Kamel (PhD) ² ¹ Department of Nursing, Erbil Technical Medical Institute, Erbil Polytechnic University, Erbil, Iraq ² Department of Medical Laboratory Technology, Erbil Polytechnic University, Erbil, Iraq
	Abstract
OPEN ACCESS Correspondence Address: Fouad Hussein Amel Department of Medical Laboratory technology, Erbil Polytechnic University, Erbil, Irag Email: fouad.kamel@epu.edu.ig Copyright: QAuthors, 2022, College of Medicine, University of Diyala. This is an open access article under the CC BY 4.0: Medicine, University of Diyala. This is an open access article under the CC BY 4.0: Website: https://djm.uodiyala.edu.ig/index.php/djm Received: 16 August 2022 Accepted: 5 October 2022	as a magnetic field. Normally, two poles of this field are concentrated. Most magnetic objects are made up of a variety of tiny fields known as domains. There are many different techniques that have been published in the literature for using magnetic energy as a diagnostic tool and for treating illnesses in both humans and animals. Objective: To examine the influence of varying amounts of static magnetic field on the viability of bacteria and the fermentation of carbohydrates. Patients and Methods: Locally prepared dipolar static magnetic field of strength 400, 800, 1200 and 1600 Gauss were used in this study measured by Teslometer. Escherichia coli isolated from 75 isolates taken from the patient who had the UPE, 25 were identified as E. coli, and only (12) samples of identified E coli specifically detect changes numbers of bacteria and biochemical test, most of the sugars ferment that have be changed and the pOtymerase chain reaction was utilized to identify the 16SrRNA gene (PCR).
	Results: The inhibitory impact of SMF is attributable, according to transcriptomic analysis, to differentially expressed genes (DEGs) predominantly included in carbon source consumption. The bacterial phenotype in SMF is consistently effectively restored by adding To the cultivation conditions, add glycolate or glyoxylate, and mutants losing glycolate oxidase are no longer susceptible to static magnetic field. Conclusion: It was concluded that the magnetic field could notably obvious by exposing bacterial cells growth density will decrease and change bacterial biological activity on sugar fermentation and due to mutation
Introduction	Keywords: Magnetic field, 16SrRNA gene, <i>E.coli</i> , fermentation, PCR
Over a billion years a to produce a weak st	ago, the Earth began (SMF), and known as the geomagnetic field (GMF), with an intensity at the ground of

ئامانجى ئەم توێژينەوەيە بريتىيە لە دياريكردنى كاريگەرى كێڵگە ى موگناتيسى جێگر، يان نەگۆر (SMF)، لەسەر شێوەى مۆڵگەى بەكترياى (E. coli)، كە ھۆكارە بۆ تووشبوون بە نەخۆشى ھەوكردنى رێرەوى ميزەرۆ لە مرۆڤدا.

کیلِّگه موگناتیسییهکان به هیّزهکانی (0.04 و 0.08 و 0.12 و 0.16 T) درووستکراون. هیّزهکه له تاقیگهکانی به شدی فیزیای کولیّژی پهروهردهی زانکوّی صهلاحهدین پیّوراون، بهکتریاکان به هیّزهکانی (0.04 و 0.18 و 0.16 و 10.16 موگناتیسیان بهرکهوت.

ئەنجامەكان دەريانخست:كاريگەرى بەركەوتن لەسەر يەكەكانى درووستكردنى مۆلگەكانى بەكتريا (E. coli) لە ماوەى 24 كاتژميردا كەمبكردووە.

ئەنجامەكان دەريانخست كە گۆرانكارى لە ھەستيارى بەكترياكان بۆ دژە زيندەگيە جياوەزەكانى وەك (Ciprofloxacin, Trimethropim/sulfamethaxazole,Ceftazidime, Cefepime and

ُ Aztreonam) له هستیاری گۆړاوه بو بهرگریکار . دیسانهوه دهرکهوت، که: ئهو بهکتریایانهی هنزی موگناتیسییان بهرکهوت، کاری کردۆته سهر

دیسانهوه دهر کهوب، که: نهو بهکتریایانهی هیری موکنانیسییان بهرکهوب، کاری کردونه سهر کاریگهره زیندهگییهکانی لهسهر تر شاندنی شهکر.

جياكراومكانى بەكترياكە پشكنينى (PCR) يان بۆ ئەنجامدرا ليّكدانەومى دەربرينى جياوازى جينەكانى ھەريەكە لە جينەكانى (*TEM₁.CTX_{M-1}*) ، *I6SrRNA،SHV ،TEM₁.CTX_{M-1}*) ، تويزينەوميەكى جينۆمييان (Sequencing) بۆكرا،بەرنامەي BioEdit V.7.0.5 بەكارھيّىرا بۆ

هملسهنگاندنی کوالیتی داتاکانی ریدخستنهکانیان. به بهکارهینانی NCBI- BLAST هملسهنگاندنی کوالیتی داتاکانی ریدخستنهکانیان. به بهکارهینانی (homology, insertions deletions, stop codons, and frame shifts) لیکولینه ومیان لهسهر کرا بو دهستنیشانکردنی پلهیهکی زیاتری لیکچوون و گورانی نیوکلیوتاید لهگهل ئامانجهکانی تر ئهمهو لیکولینه وهکه به چهندین نهنجامی دیکه گهیشتووه، که له کوتایی تویزینه وهکه دانراون.



کاریگمری هنزه جیاوازهکانی کنِلگه موگناتیسی جنِگرلمسهرسیفهته دیارو بۆماوهیهکانی بهکتریای (Escherichia coli)هۆکار به تووشبونی نهخوشیه کانی ریرهوی میزلدان

هەولىر، كوردستان، عيراق

رممەزان1444هـ نەرۆز 2723ك

ئادار ۲۰۲۳