Evaluate of multi-drug-resistant bacteria responsible for chronic urinary tract infections in Al Mahalla regional cities in Egypt

A. Abdallah [1], E. S. Abdelmouty [1], A. N. Emam [2], E. A. Ewais [1], and M. Khedr [1,*]

[1] *Botany and Microbiology Department, Faculty of Science, Al-Azhar University, Cairo, Egypt*
[2] *Refractories, Ceramics and Building Materials Department, National Research Centre (NRC), El Bohouth St., Dokki, Giza 12622, Egypt*

*Corresponding author E-mail: (M. Khedr)

ABSTRACT:

Among chronic bacterial infections in humans is Urinary tract infection particularly when these infections are caused through Multi drug resistant (MDR) isolates which resist various traditional treatment especially antibiotic-based methods. In this article we were isolate then biochemical and molecular identification of most potent MDR isolates cause Urinary tract infection (UTIs) in Egyptian patients' males and females. **So,** in our study we obtained 22 different bacterial isolates which all are gram negative belonging to different four genera based on their growth on Uriselect into E. coli, Klebsiella sp, Pseudomonas sp., and Acinetobacter sp., they have beta-hemolytic activity, beside that they exhibit MDR behavior against twelve antibiotics, according to growth on Macconkey agar, 17 isolates with lactose fermenting (16 Klebsiella sp and one E. coli) and five were non-lactose fermenting isolates. The most potent bacterial isolates were Klebsiella pneumonia A061 and Klebsiella pneumonia A031 with accession numbers OP811040 and OP811041 respectively. Finally, among MDR bacterial isolates, Klebsiella pneumonia is one of the most threat of Urinary tract infection in Egyptian people as they resist traditional treatment with antibiotics.

Keywords: MDR bacteria; UTIs; *Klebsiella pneumonia A061;* Klebsiella *pneumonia A03*.

INTRODUCTION

The urinary system plays a vital role in eliminating waste chemicals from the circulation via metabolic processes. Furthermore, this system is essential for regulating not only blood pressure and volume, but also for ensuring the equilibrium of ions and solutes concentration in the bloodstream [1]. In healthy persons, urine is either sterile or contains minimal levels of pathogens that have the ability to induce sickness [2]. A urinary tract infection (UTI), commonly referred to as bacteriuria, is the condition characterized by the presence of bacteria in the urine. During a clinical disease, the initial bacterial concentration might reach 105 bacteria per milliliter. However, in the context of epidemiology, a substantial quantity of bacteriuria is defined as a minimum of 105 bacteria per milliliter in freshly voided urine. Urinary tract infections (UTIs) are widespread, affecting around 10% of the worldwide population, which amounts to around 150 million persons, on a daily basis. Uropathogenic E. coli (UPEC) is widely acknowledged as the primary causative agent in both severe and moderate urinary tract infections (UTIs) [4]. Urinary tract infections (UTIs) can be classified into several categories depending on their location, including urethritis, cystitis, or pyelonephritis. These infections impact almost 150 million people annually and are linked to high death rates and huge healthcare costs. For example, studies have calculated that the economic consequences of recurring urinary tract infections (UTIs) in the United States surpass $5 billion every year [5,6]. Although the symptoms of urinary tract infections (UTIs) may vary depending on the location of the infection, these illnesses have a negative impact on the patient's relationships, both emotionally and socially, leading to a diminished quality of life [7,8]. Urinary tract infections (UTIs) are classified into two categories: uncomplicated UTIs (uUTI) and complex UTIs (cUTI) [9]. Urinary tract infections (UTIs) commonly affect persons who are in a state of excellent health and do not have any structural or neurological abnormalities in their urinary tract [6]. Complex urinary tract infections (UTIs) are defined by the existence of urinary tract abnormalities that increase the likelihood of infection, such as catheterization or functional and anatomical abnormalities (e.g., obstructive disease). Urinary tract obstruction, urine retention, neurogenic bladder, kidney failure, pregnancy, and stones are the conditions that can cause urinary tract obstruction, urinary retention, neurogenic bladder, kidney failure, pregnancy, and stones [6,10].

The Enterobacteriaceae family is the predominant group of bacteria that are

commonly present in urinary tract infections (UTIs), occurring frequently in both community and hospital environments. The main bacteria accountable for urinary tract infections (UTIs) is uropathogenic Escherichia coli (UPEC) [11,12]. The latter is also the main cause of complicated urinary tract infections (cUTI) [10]. Hospitals have a greater prevalence of antibiotic-resistant Gram-negative bacteria in comparison to samples from the population, such as carbapenemase-resistant Enterobacteriaceae [13]. Urinary tract infections (UTIs) are mostly caused by bacteria, with the presence of other species, such as fungi and viruses, being exceedingly rare.Candida albicans is the primary fungus species accountable for urinary tract infections (UTIs). The main causes of viral urinary tract infections (UTIs) include cytomegalovirus, type 1 human Polyomavirus, and herpes simplex virus [14,15].

Other bacteria associated with the emergence of severe urinary tract infections (UTIs) include Enterococcus spp., Klebsiella pneumoniae, Staphylococcus aureus, and Pseudomonas aeruginosa. Pathogenic bacteria, including Klebsiella pneumoniae, Staphylococcus saprophyticus, Enterococcus faecalis, Group B Streptococcus (GBS), Proteus mirabilis, Pseudomonas aeruginosa, Staphylococcus aureus, and Candida species, play a significant role in the development of uncomplicated urinary tract infections (uUTIs). The development of UTI is caused by the attachment of certain virulence factors found in bacteria that are present in the urethra. This process entails the colonization of the bladder by pathogens, which gain entry by its extensions, such as flagella and pili. Once a pathogen attaches to the bladder, complex interactions occur between the host and the bacteria, resulting in the progression of the disease to a more severe state [16]. The accumulation of virulence factors is a prominent and noteworthy feature that contributes to the emergence of drug resistance in the urinary tract [17]. MDR bacteria in urine provide challenges for treating urinary tract infections with traditional empirical therapy and increase the overall incidence of illness [18]. The escalating prevalence and swift emergence of multidrug-resistant bacterial illnesses provide a substantial and perilous challenge to public health, particularly in growing populations [19]. The emergence of multidrug-resistant uropathogens has led to an increase in hospital-acquired urinary tract infections worldwide [20]. A considerable proportion of microbial species and isolates are still unknown or have not been fully described [21]. Due to the continuous increase in bacteria multidrug resistance and the limitations of antibiotic therapy, there is an urgent need for the creation of powerful antimicrobial drugs that utilize innovative methods of action [22]. Currently, researchers investigating the manufacturing of nanoparticles are focusing on the advancement of novel methods and substances to produce eco-friendly nanomaterials [23,24]. The focus of this study is on isolating, purifying, and identifying the most potent multi-drug resistant (MDR) bacterial strains responsible for urinary tract infections (UTIs) in the specific region of El Mahalla, Gharbia Governorate, Egypt.

METHODOLOGY

Isolation clinical samples

A total of 66 patients (21 males and 45 females) were selected based on age and had provided urine samples during their visits to the outpatient clinics of the Urology and Nephrology center at Mansoura Authority hospital for a duration of over eight months, from May 2020 to March 2021. The individuals had symptoms indicative with a urinary tract infection (UTI). The patient's age spanned from 15 to 60 years or above. Each patient received a 20 mL sterile screw-capped universal container that had been sterilized and calibrated prior to collecting their clean catch midstream urine. In a duration of six hours, the samples were correctly labeled, delivered to the laboratory, and meticulously analyzed. To impede the growth of bacteria in the urine samples, a dosage of 0.2 mg of boric acid was added to each container. Before collecting samples, all patients were given thorough instructions on how to collect them in a sterile way to avoid contamination of the urethra.

Purification of the Bacterial isolates

Colonies showing distinct growth on any enrichment medium, regardless of form or color, were gathered and streaked once more on agar plates having the identical isolation medium following the incubation period. To guarantee the purity of the colonies, the procedure was carried out multiple times. The isolates were examined both morphologically and by Gram staining under a microscope. Before subjecting the bacterial isolates to the beta hemolysis activity test at a temperature of 40°C, they were first grown on a nutrient agar slant [25].

isolation of Beta-hemolytic strains

To suspend the microbe, a sterile swab or applicator stick is used to transfer enough colonies from a pure culture into a clear plastic (polystyrene) test tube measuring 12 × 75 mm. The bacterium is subsequently dispersed in 3.0 mL of sterile saline, an aqueous solution comprising 0.45%–0.50% NaCl and exhibiting a pH within the range of 4.5 to 7.0. The purified bacterial isolates were analyzed on a blood agar medium containing 5% sheep blood. Each individual sample was inoculated onto sterilized blood agar plates and incubated at a temperature of 37 ◦C for a period of 3 days. The pattern of blood hemolysis was seen as follows: Alpha, Beta, and Gamma are terms used to describe different levels of hemolysis. Alpha indicates partial hemolysis, Beta indicates entire hemolysis, and Gamma indicates no hemolysis. The user's input is the string "[26]".

Antibiotic sensitivity test.

The experiment was done using the disk diffusion technique using Muller Hinton agar plates. We tested twenty-two isolates of beta hemolytic bacteria against various antibiotics including Amikacin, Piperacillin/Tazobactam, Gentamycin, Trimethoprim/Sulfamethoxazole, Ciprofloxacin, Levofloxacin, Ofloxacin, Nitrofurantoin, and Amoxycillin/Clavulanic acid. The antibiotics Ceftazidime, Cefotaxime, and Cefepime are each present in a concentration of 30 µg. Prior to testing, a nutrient-rich broth was utilized to rehydrate each culture. The cultures were adjusted so that the absorbance at 600 nm was precisely 0.5. Next, 250 µl of the bacterial culture was evenly distributed and allowed to absorb on the Mueller Hinton agar. This was done after adding 15 ml of the agar to a sterile Petri dish and allowing it to firm for 10 minutes. Following the placement of lids on the agar plates, three antibiotic plates that were similar were subjected to incubation for a duration of twenty minutes at a temperature of 20 ◦C, followed by an additional twenty-four hours at a temperature of 37 ◦C. The bactericidal capabilities of each antibiotic were tested by calculating the average inhibition zone diameter (mm) based on three measurements.

Automated identification is performed via the biome Rieux VITEK2 system.

Following the application of crystal violet dye, the Gramme positive (GP) and Gramme negative (GN) microorganisms were identified. The process of identifying microbes and determining their susceptibility to antibiotics is conducted using the VITEK 2 compact system. The VITEK 2 is a microbiological system that automates processes using growth-based technologies. The devices are capable of handling colorimetric reagent cards, which are then automatically incubated and interpreted [27].

Molecular identification of isolates of multidrug-resistant bacteria

The Wizard Genomic DNA kit, produced by Promega in Madison, WI, USA, was used to extract the whole genomic DNA of the bacterial isolates being studied, following the manufacturer's instructions. The RW primer (CCAGCCGCAGGTTCCCCT) and the 16Sb FW primer (CGCTGGCGGCAGGCTTAACA) [13] were used as the universal bacterial primers for the 16S rDNA genes. The denaturation step involved 35 cycles at a temperature of 94 degrees Celsius for a duration of 30 seconds. This stage also included the annealing process at a temperature of 55 degrees Celsius for 60 seconds, the extension process at a temperature of 72 degrees Celsius for 90 seconds, and the final extension step at a temperature of 72 degrees Celsius for 180 seconds. The PCR product was amplified and its length, about 1500 base pairs, was verified by submitting 10µl samples to electrophoresis on a 1% horizontal agarose gel containing 0.5 µg/ml ethidium bromide. The gels were examined and recorded under ultraviolet (UV) light [28, 29]. Qiagen Inc., located in Chatsworth, California, employed QIAquick spin columns for the purification of PCR findings. The PCR results underwent sequencing using a Perkin Elmer 377 DNA sequencer and the Dye Deoxy Terminator Cycle Sequencing Kit, as detailed in a recent paper by Perkin Elmer (Foster City, CA) [28]. The identification of known bacterial species was performed by doing a BLAST search of GenBank using 16S rDNA gene sequences that were comparable to those of the isolates. To ascertain the relative distance between each isolate and the specified strain in the blast result, a phylogenetic tree was constructed using the blast result of each isolate. Subsequently, this tree was juxtaposed with the top 10 analogous sequences identified in the NCBI database.

RESULTS

Purification and isolation activity on the test media

Twenty-two isolates of pure single colonies were obtained through serial dilution on

nutrient agar plates which incubated for 16h at 37^{O}C then they primarily identified through Gram reaction, beside microscopic examination for cells shape and cellular arrangement pattern (**Table 1)**. Our isolates belonged to four different genera as indicated by colony pigmentation on Uriselect agar plates after incubation at 37^{O}C for 24h, **(Fig 1).** The twenty-two isolates belonged into four different genera where 16 isolates are *Klebsiella pneumonia*, four isolates belong to *Pseudomonas aeruginosa*, one *Acinetobacter baumannii* and one *E. coli* Isolate (Fig 2).

MacConkey agar plates were classified as our isolates according to lactose fermenting activity to 17 isolates with lactose fermenting (16 *Klebsiella sp* and one *E. coli*) and five were non-lactose fermenting isolates as in Fig 3.

Hemolysis activity of blood

Single colons were cultivated on blood medium for blood hemolysis activity, twenty-two isolates showed β -hemolysis after 24h, Isolates A055 and A059 were the most isolates showed beta-hemolysis clear zone on blood agar (21 and 17mm) respectively followed by A020 and A031 with 16mm for both (Fig. 3, 4).

Antibiotic sensitivity of Beta-hemolytic isolates.

The obtained bacterial isolates were tested against twelve different antibiotics. All isolates were shown MDR phenomenon to the tested antibiotics, minimum resistant isolates were to eight antibiotics (A069) and the maximum resistant was fourteen isolates to all twelve antibiotics as shown in **Fig. 5**. Among them (A031 and A061).

According to one way ANOVA statistical analysis, the most effective antibiotic was Gentamycin, followed by Ofloxacin and Amikacin, then Levofloxacin and Ciprofloxacin as described in Dendrogram that illustrates the complete linkage and correlation coefficient distance between the tested antibiotics as shown in **Fig. 6.**

Molecular Identification of extensive MDR beta-hemolytic isolates

The most extensive MDR isolates were two isolates A031 and A065 which identified through 16S rDNA gene partial sequencing, then compared through NCBI BLASTn with the most related submitted sequences. The data was analyzed, and phylogenetic tree constructed through MEGA11 software as shown in **Fig. 8.** The two isolates were identified as *Klebsiella pneumonia A061* and *Klebsiella pneumonia A031* with accession numbers OP811040 and OP811041 respectively

DISCUSSION

In a study comparing urinary tract infections (UTIs), Abdelrazik *et al.*, [30] found microbial isolates, comprising of 88 gram-negative bacteria, only 8 gram-positive bacteria, and 4 yeast isolates. Among the gram-negative isolates, 80% (70 isolates) exhibited the ability to undergo lactose fermentation, while 20% (17 isolates) did not demonstrate this capability. Two isolates were cultured on blood agar and showed beta hemolysis. Additionally, 22 (65%) of the gram-negative isolates had beta hemolysis, whereas 10 (29%) of the gram-negative isolates showed alpha hemolysis. Among the one hundred isolates, a substantial percentage (72%) exhibited resistance to several drugs, indicating multidrug resistance (MDR) "[31]". The specified order of predominance comprises *Klebsiella pneumoniae*, *Staphylococcus saprophyticus*, *Enterococcus faecalis*, *group B Streptococcus (GBS), Proteus mirabilis*, *Pseudomonas aeruginosa, Staphylococcus aureus*, and *Candida spp*. The primary causative agents causing complicated urinary tract infections (UTIs), ranked by prevalence, include *Enterococcus spp., K. pneumoniae, Candida spp., S. aureus, P. mirabilis, P. aeruginosa*, and GBS. *Uropathogenic Escherichia coli (UPEC)* is the main culprit behind both simple and severe urinary tract infections (UTIs).

Moreover, a substantial agreement is there among other authors, such as [32, 33; 34], who have confirmed that Klebsiella, Proteus, Pseudomonas, and Enterobacter are often identified as the main causative agents of urinary tract infections in both males and females. Moreover, it is well recognized by [32] that the microbiological origin of urinary tract infections (UTIs) is firmly established, with prevalent pathogens like as E. coli and S. saprophyticus being associated with the prevalence of acute uncomplicated infections in the general population. Singh observed that *Klebsiella, Enterococcus, Proteus Species, Enterobacter, Bacillus*, and *Shigella* are recognized as causative agents of uncomplicated cystitis and pyelonephritis "[33]". The primary strain discovered in this investigation was E. coli 46, which constituted 44% of the isolates. K. pneumoniae was the second most prevalent strain. Among the patients who were examined, 24 individuals (25%) were found to have Pseudomonas aeruginosa, whereas 10 individuals (10%) had

other types of isolates. The remaining isolates were only documented in a limited number of instances. E. faecalis was detected in 2% of the samples, whereas A. baumannii was present in 1% of the samples. These findings align with the outcomes documented by Bennett et al., The investigation done by [35] indicated *E. coli* as the predominant strain. Walsh and Collyns [36] reported the existence of *K. pneumoniae, Proteus vulgaris* is well recognized as the predominant pathogen responsible for urinary tract infections in both males and females [31; 37]. P. mirabilis is the main cause of uncomplicated urinary tract infections, however other bacteria such as *Klebsiella, Pseudomonas, Staphylococcus*, and *Corynebacterium urealyticum* have also been identified as contributors to UTIs in both males and females. In addition, [38; 36] have recorded the participation of some intestinal bacteria in the causation of the clear-cut symptoms of urinary tract infection. The article from [35; 31] has identified *Pseudomonas aeruginosa* as a notable causative agent of uncomplicated urinary tract infections.

There has been a notable increase in the prevalence of microbial infections over the past few decades due to the multidrug resistance (MDR) of the recovered bacterial isolates. The continuous progress of antimicrobial drugs in the treatment of diseases has led to the emergence of resistance among various strains of bacteria. MDR, also known as multidrug resistance, is the term used to describe the capacity of a bacteria to remain unaffected or resistant to antimicrobial medications that it was previously sensitive to, even after they have been given [39]. These robust bacteria possess the capacity to endure assaults from antimicrobial medications, leading to ineffectual therapy. Consequently, this results in the enduring presence and dissemination of infections, accompanied by a notable surge in bacterial resistance. Some notable examples of bacterial resistance include *E. coli's* ability to withstand cephalosporin and fluoroquinolones, *Klebsiella pneumoniae's* ability to resist cephalosporin and carbapenems, Staphylococcus aureus' resistance to methicillin, *Streptococcus pneumoniae's* resistance to penicillin, Nontyphoidal *Salmonella's* resistance to fluoroquinolones, *Shigella sp.'s* resistance to fluoroquinolones, *Neisseria gonorrhoeae's* resistance to cephalosporin, and *Mycobacterium tuberculosis'* resistance to rifampicin, isoniazid, and fluoroquinolones. These drug-resistant strains have a role in causing common infections such urinary tract infections, pneumonia, and bloodstream infections, as well as a significant proportion of infections acquired in healthcare facilities. Moreover, antibiotic resistance is associated with increased mortality rates and significant healthcare costs, while also significantly reducing the effectiveness of antimicrobial medications [42].

CONCLUSION

Most infections of UTIs in our medical urine samples were caused by gram negative strains which belonged to different four genera *E. coli*, *Klebsiella sp*, *Pseudomonas sp.*, and *Acinetobacter sp.*, these isolates have beta-hemolytic activity, beside that they exhibit MDR behavior against twelve antibiotics. The most potent bacterial isolates were *Klebsiella pneumonia A061* and *Klebsiella pneumonia A031* with accession numbers OP811040 and OP811041 respectively

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Table 1: Morphological Identification of bacterial isolates based on Gram reaction and microscopic examination then confirm scientific names through VITK2

Sample code	Gram reaction	Cell description
A020	Gram-negative	Non-motile, and rod-shaped bacteria, capsule lactose-fermenting,
A022		Non-motile, non-fastidious, oxidase-negative, and aerobic Gram-negative coccobacilli
A023		• Rod-shaped, Flagellum one or more, providing motility. Aerobic, non-spore former
A025		Non-motile, and rod-shaped bacteria, capsule lactose-fermenting,
A026		Non-motile, and rod-shaped bacteria, capsule lactose-fermenting,
A027		Non-motile, and rod-shaped bacteria, capsule lactose-fermenting,
A030		Non-motile, and rod-shaped bacteria, capsule lactose-fermenting,
A031		Non-motile, and rod-shaped bacteria, capsule lactose-fermenting,
A055		Rod shaped, non-spore forming, motile with peritrichous flagella or nonmotile
A056		• Rod-shaped, Flagellum one or more, providing motility. Aerobic, non-spore former
A058		Non-motile, and rod-shaped bacteria, capsule lactose-fermenting,
A059		non-motile, and rod-shaped bacteria, capsule lactose-fermenting,
A060		Non-motile, and rod-shaped bacteria, capsule lactose-fermenting,
A061		• Rod-shaped, Flagellum one or more, providing motility. Aerobic, non-spore former
A062		Non-motile, and rod-shaped bacteria, capsule lactose-fermenting,
A063		Non-motile, and rod-shaped bacteria, capsule lactose-fermenting,
A065		non-motile, and rod-shaped bacteria, capsule lactose-fermenting,
A066		Non-motile, and rod-shaped bacteria, capsule lactose-fermenting,
A067		Non-motile, and rod-shaped bacteria, capsule lactose-fermenting,
A068		Non-motile, and rod-shaped bacteria, capsule lactose-fermenting,
A069		• Rod-shaped, Flagellum one or more, providing motility, Aerobic, Non-spore former
A070		Non-motile, and rod-shaped bacteria, capsule lactose-fermenting,

Figure 1: cultivation of bacterial isolates on Uriselect agar which indicates different genera members according to colonial pigmentation: pink colonies (*E. coli*), Blue (*Klebsiella sp.),* brown (*Pseudomonas sp.),* while creamy white (*Acinetobacter sp.).*

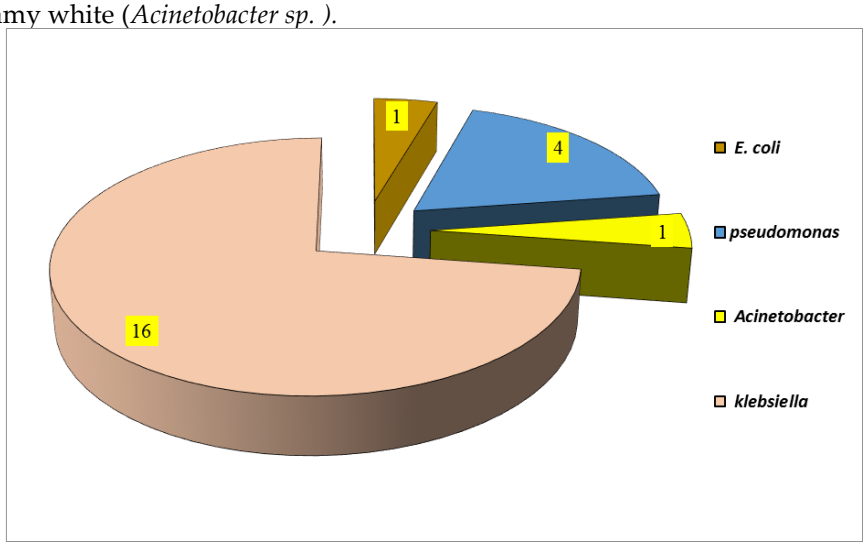

Figure 2: Classification of 22 most potent isolates into different four genera according to Vitek 2.

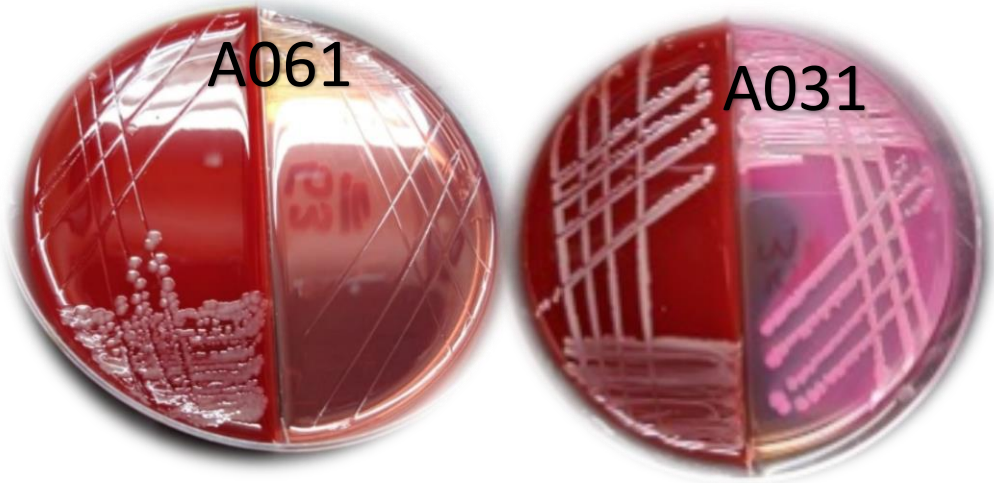

Figure 3: cultivation of bacterial isolates for example (*Klebsiella A061* and *A031*) on blood agar and Macconkey agar for determination of blood hemolysis activity and lactose fermenting isolates.

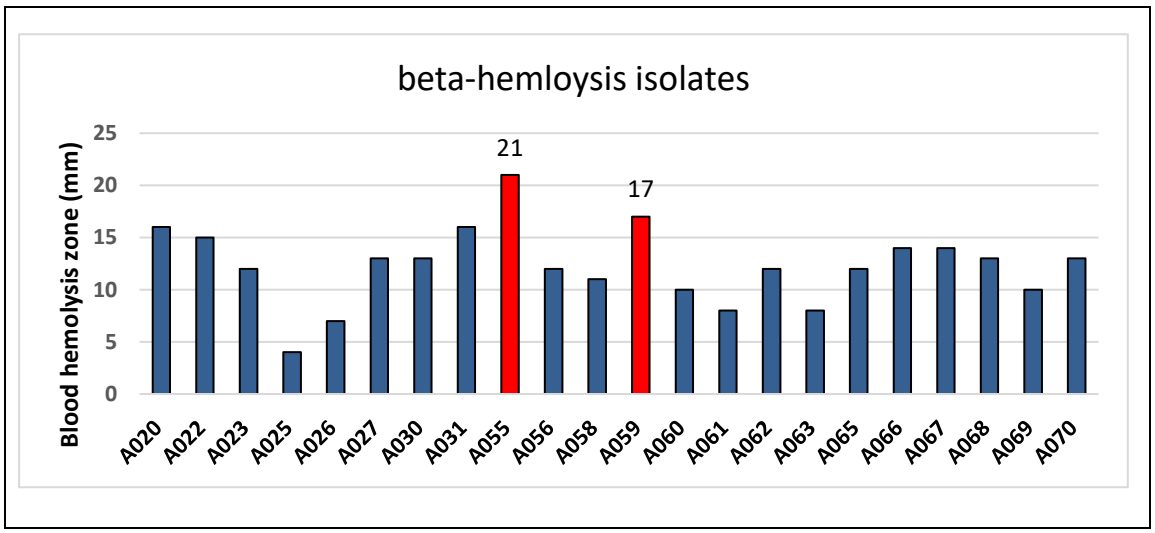

Figure 4: β -hemolysis activity on blood agar of 22 bacterial isolates

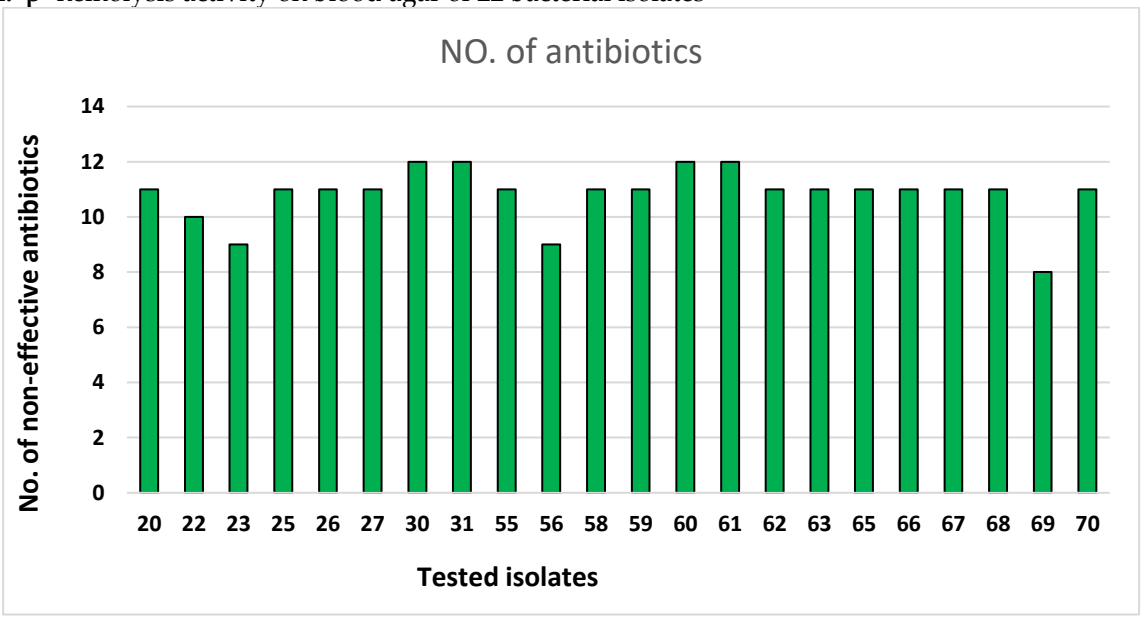

Figure 5: Number of resistant isolates to different antibiotics.

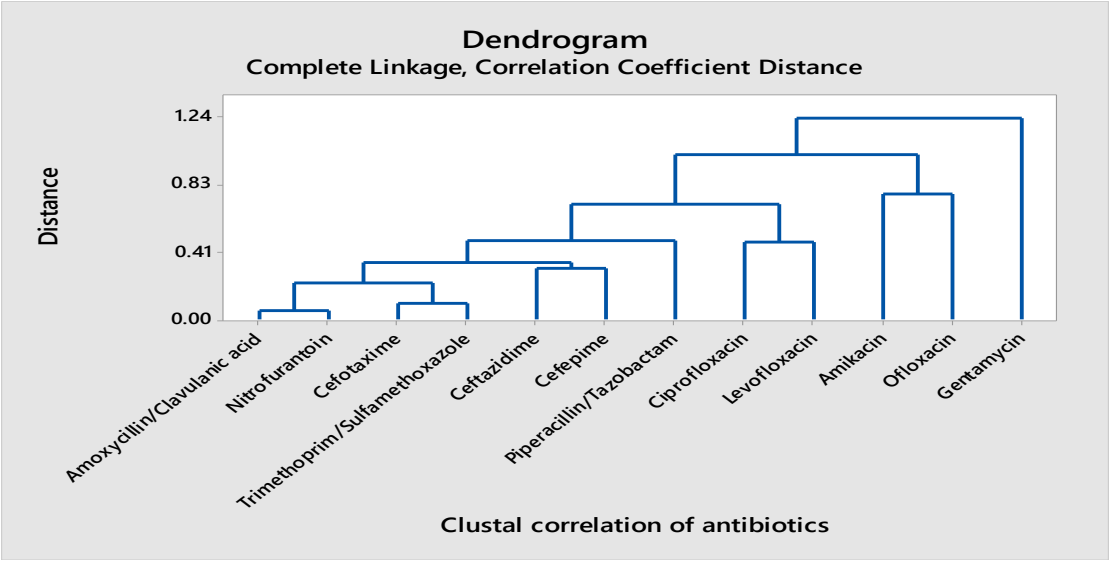

Figure 6: Antibiotic dendrogram based on their results to 22 tested isolates generated by ANOVA statistical software.

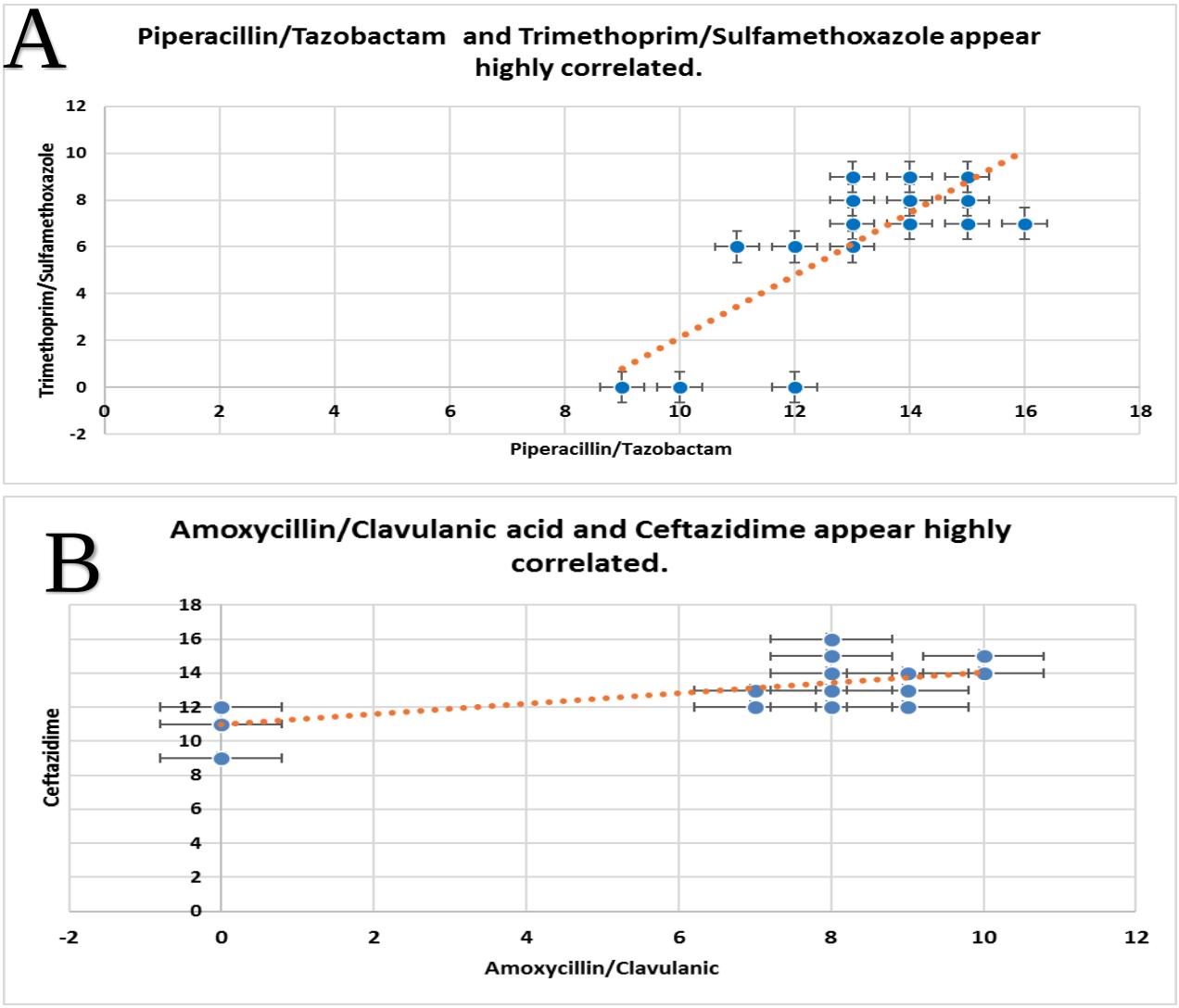

Figure 8: The most correlated antibiotics among twelve tested, there is a high correlation between Piperacillin and Tazobactam, also between Trimethoprim and sulfamethoxazole (A), also there is a correlation between Amoxycillin, Clavulanic acid and Ceftazidime (B).

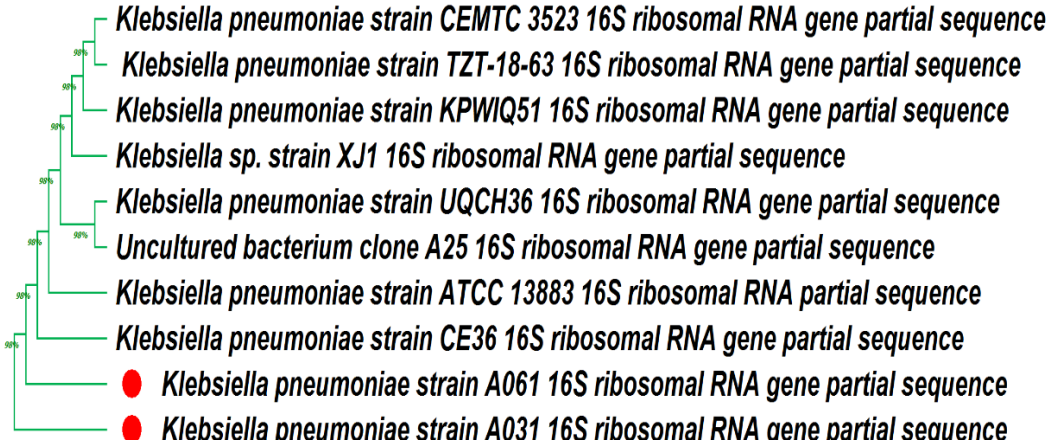

Figure 8: Phylogenetic tree through MEGA11 of *Klebsiella pneumonia A061* and *Klebsiella pneumonia A031* through NCBI BLASTn.

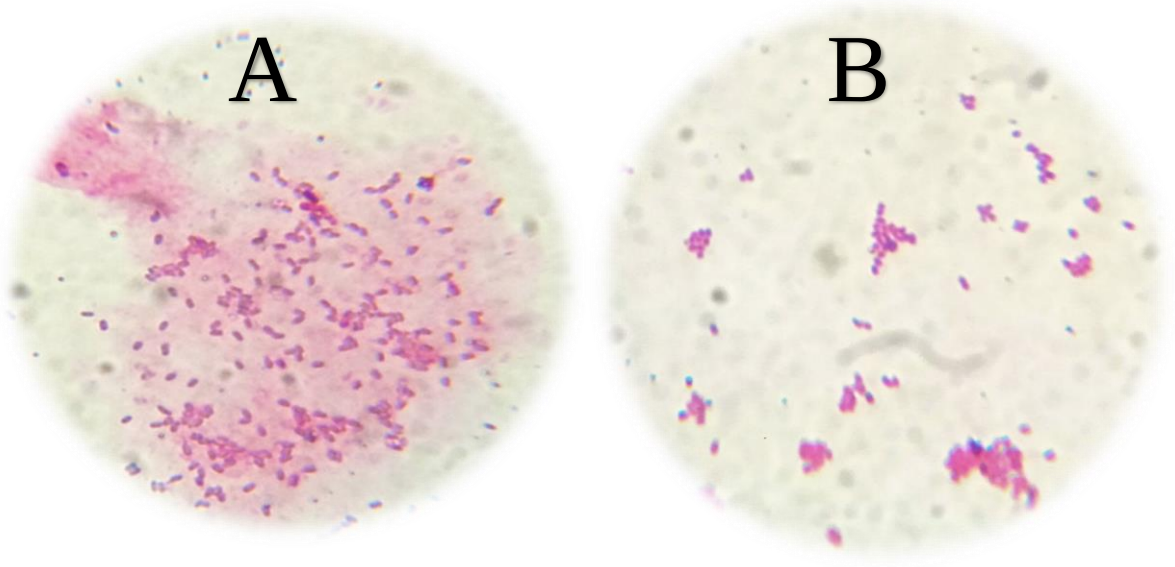

Image 1: Gram stain reaction of two isolates (A) *Klebsiella A061* and (B) *A031*.

تقييم البكتيريا المقاومة للأدوية المتعددة والمسؤولة عن التهابات المسالك البولية المزمنة في مدن إقليم المحلة في مصر

احمد محمد عبدالله [1], اسلام سعيد عبدالمعطي [1], احمد نبيل امام [2], عماد الدين عباس عويس [1], محمد خضر [1,*]

[1] *قسم النبات والميكروبيولوجى، كلية العلوم، جامعة الازهر، القاهرة، مصر*

* البريد الإلكتروني للباحث الرئيسي:

الملخص العربي:

من بين الالتهابات البكتيرية المزمنة في البشر التهاب المسالك البولية، خاصة عندما تكون هذه الالتهابات ناتجة عن عزلات مقاومة للأدوية المتعددة (MDR)والتي تقاوم العلاجات التقليدية المختلفة، خاصة الطرق القائمة على المضادات الحيوية. في هذه المقالة، قمنا بعزل ثم التعرف البيوكيميائي والجزيئي على أكثر العزلات المقاومة للأدوية المتعددة قوة والتي تسبب التهاب المسالك البولية (UTIs) في المرضى المصريين من الذكور والإناث. تم الحصول على عينات بول مختلفة من المرضى الذكور والإناث، واستخدمت كمصدر لعزلات مسببات الأمراض البكتيرية، والتي تم تنقيتها على أطباق الأجار المغذي، ثم زرعت العزلات على أجار الدم لتحديد نشاطها التحللي للدم، ثم تم اختبار جميع هذه العزلات البكتيرية من خلال طريقة انتشار قرص الأجار ضد اثني عشر مضادًا حيويًا مختلفًا، ثم تم التعرف عليها أوليًا من خلال تفاعل جرام ثم من خلالVITEK2 ، تليها التعرف الجزيئي من خلال تسلسل الحمض النووي الريبوزي S.16 . اظهرت النتائج في دراستنا التي من خلالها حصلنا على 22 عزلة بكتيرية مختلفة وكلها سالبة الجرام تنتمي إلى أربعة أجناس مختلفة بناءً على نموها على Uriselect إلى *الإشريكية القولونية*، و*الكليبسيلا*، و*الزائفة*، و*الراكدة*، ولها نشاط تحلل بيتا للدم، إلى جانب أنها تظهر سلوكًا مقاومًا للأدوية المتعددة ضد اثني عشر مضادًا حيويًا. وفقًا للنمو على أجار ماكونكي، 17 عزلة مخمرة للاكتوز (16 *كليبسيلا* وواحدة *إشريكية قولونية*) وخمس عزلات غير مخمرة للاكتوز. كانت أقوى العزلات البكتيرية هي *الكليبسيلا الرئوية *A061 و*الكليبسيلا الرئوية *A031 بأرقام الوصول OP811040 و OP811041على التوالي. **الخلاصة:** *الكليبسيلا الرئوية* المقاومة للأدوية المتعددة هي واحدة من أكبر التهديدات للمسالك البولية في البشر حيث أنها تقاوم العلاج التقليدي بالمضادات الحيوية. لذلك يجب النظر في استراتيجية جديدة للقضاء على هذه المسببات للأمراض البكتيرية.

الكلمات الاسترشادية: البكتيريا المقاومة للعديد من المضادات الحيوية, الالتهابات البكتيرية لمجري البول ,كلبسيلا نومونيا *A061* و *A031*