WORLD JOURNAL OF PHARMACEUTICAL RESEARCH
SJIF Impact Factor 5.990
Volume 4, Issue 7, 2023-2049. Research Article ISSN 2277– 7105

SCREENING OF MULTI DRUG RESISTANT BACTERIA AND FUNGI FROM MEDICAL DEVICE USED IN URINARY TRACT INFECTION AND INFLAMMATION IN HOSPITAL OF GUNTUR DISTRICT, A.P, INDIA.

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Article Received on
10 May 2015,

Revised on 05 June 2015,
Accepted on 28 June 2015

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ABSTRACT

The ability to stick to materials and promote the formation of biofilm is an important feature of bacteria resulting from participation in the foreign body infections. The fact that staphylococci are the main things related injury medical devices has stimulated a lot of research on the pathogenic mechanisms, which led to significant achievements in our understanding of biofilm formation. And so on, it has been identified on the battery of Staphylococcus virulence factors, and characterized in the past two decades, leading to important information, particularly with regard to bacteria interact with the surface of the transplanted organ or inserted as normal, and live in harmony cons balanced on our skin, and which constitute a key element small plants coetaneous. Put out this medical device objects rarely causes infections. With this, the development needed to study living organism's goals current high living and multiple resistance responsible for community acquired revenues and UIT hospital obtained by catheterization for patients. was observed growth (18) species of bacteria in alleviating serial dilution concentration (10^4) (AVMP 1) were distributed among bacteria (E.coli) and the number (7) and percentage (38.8%), and bacteria Pseudomonas aeruginosa The number (3) and the percentage (16.6%), and bacteria (Proteus mirabilis) was the (2) and percentage (11.1%), and bacteria (Bacillus ceruse) and the number (4) and percentage (22.2%), and bacteria (Citrobacteria freundi) was the (2) and percentage (11.1%), while the observed

growth (15) species of bacteria in serial dilution concentration (10^5) (AVMP 2), distributed among bacteria (Klebsiella pneumonia) and the number (2) and percentage (13.3%), and bacteria (Staph aureus) and the number (5) and the proportion of percentage (33.3%), and bacteria (Staph epidermis) the number (3) and the percentage (20%), and bacteria (Enterococci faecalis) was of (1) and percentage (6.6%), and bacteria (Entrobateria aerogenous) was the (2) and the percentage (3.13%), and bacteria (Bacillus stubilis) was the (2) and percentage (3.13%). It was also noted in this study, the growth of (29) species of fungi which were distributed between Aspergillus Niger and number (6) and percentage (20.6%) yellow and Aspergillus, and the number (8) and percentage (27.5%), and Pencillium prosecution of (5) and the percentage (it was 17.2%), while Candida prosecution of (6) and percentage (20.6%), while the number of Candida albicans (4) and the percentage (13.7%), is also an indication that the medical device also contribute to increase the frequency of microbes.. It was examined antibiotic resistance and (AVMP1, AVMP2) by agar well diffusion method against 10 antibiotics (ampicillin, streptomycin, tetracycline, Chloramphenicol Norfloxacin, aggentomycin, ciprofloxacin, erythromycin, amoxicillin and cefluexcin). Bacterial colony usually occurs in all dilutions ranging from 10^{-1} to 10^{-4} and look like a colony isolated one in the eye of 10^4 mitigation as AVMP 1. And screened for resistance to antibiotics. AVMP1 highly sensitive six antibiotics (such as ampicillin, tetracycline, Norfloxacin, ciprofloxacin, amoxicillin and cefluexacin). While highly resistant to (streptomycin, chloramphenicol, aggentomycin and erythromycin). Bacterial colony usually occurs in all dilutions ranging from 10^{-1} to 10^{-5} and look like a colony isolated one in the eye of (10^5) AVMP2 been isolated in solitary colony 10^5 also ease the tension of the sensitivity of antibiotics examination. However, ciprofloxacin was AVMP2 very sensitive to all the other multiple antibiotics, anti-Lama (erythromycin, amoxicillin and cefluexacin, Ampicillin, streptomycin, tetracycline, chloramphenicol Norfloxacin, aggentomycin). Life was very resistant to antibiotics , according to the literature and isolate any bacteria that show resistance to more than one strain of bacteria can be formulated resistance. Also on our results to determine AVMP1 as multi-resistant bacteria and the drugs AVMP2 of drug-resistant bacteria and one strain. As for antibiotics fungus takes five antifungal (Clotronazole, Terbin, Ivenmycin, Ketoconazole, Fluconazole), and was examined antibiotic resistance against fungi isolated from medical devices that are used in urinary tract infection and sample (Candida prosecutors) in the serial dilution described 10^4 and sample It was Aspergillus prosecutors in mitigation serial 10^5 between the two types resistant to all antibiotics fungal used in this study, either with respect to pencillium mitigation serial in 10^3 has a couple of

antibiotic resistance built are (Ketoconazole and Fluconazole) while three antibiotics sensitive are (Terbin Clotronazole, Ivenmycin), it is assumed that the management of persistent antibiotics to the hospital received patients with urinary tract infection and confirm the catheter young plants resist infection to join the medical device.

KEYWORD: Screening bacteria and fungi from medical device used IN UTI , antibacterial agent and antifungal activity.

INTRODUCTION

Urinary tract infection (UTI) is a common bacterial infection known to affect the different parts of the Urinary tract, both males and females Despite the fact, and have a high risk of UTI that both the genders are susceptible to the infection, women are most vulnerable due to their anatomy and reproductive physiology. Urinary tract infections are infections of the urethra, bladder, ureter, or the kidneys, which comprise the urinary tract. E.coli bacteria cause the majority of UTIs, but many other bacteria, fungi, and parasites may also cause UTIs. Other risk factors for UTIs include any condition that may impede urine flow (e.g., enlarged prostate, congenital urinary tract abnormalities, and inflammation). Patients with catheters or those who undergo urinary surgery and men with enlarged prostates are at higher risk for UTIs. Symptoms and signs of UTI vary somewhat depending on sex, age, and the area of the urinary tract that is infected; some unique symptoms develop depending on the infecting agent. UTIs are diagnosed usually by isolating and identifying the urinary pathogen from the patient; There can be many complications of urinary tract infections, including dehydration, sepsis, kidney failure, and death. If treated early and adequately, the prognosis is good for most patients with a UTI. Although there is no vaccine available for UTIs, there are many ways a person may reduce the chance of getting a UTI. The incidence of urinary tract infections (UTI) Urinary Tract Infection of the most Bacterial infections in humans, especially the urethra and bladder inflammation or at least Germ Bacteruria, and the estimated number of patients of patients with these infections by about 8 million people every year in the world according to the latest Statistics International (Sim., 2001). The incidence of urinary tract infections vehicle Complicated UTI, especially glomerulonephritis Pyelonephritis is the reason for the high mortality rate in the world Mortality Especially when the evolution of the patient's condition to the failure or kidney damage Renal damage, and the latter is now of age and the problems often lead lives that patient did not undergo a kidney transplant (Kidney stretch) (Forfar & Arneils., 1992). Arise urinary tract infection in its early

stages, usually in the urethra as a result of the growth and multiplication of bacteria in. This is called inflammation of the urethra (Uretheritis), and while the injury extends to Mucosa lining of the bladder cause inflammation of the bladder (Cystitis) and this is what is called rheumatoid urinary tract (Lower UTI). And if not treated quickly, these injuries, it extends to infect the upper parts of the urinary system and pelvis, causing inflammation of the kidney pelvis And glomerulonephritis and then kidney damage, which is a serious complication of injuries in the upper urinary tract (Upper UTI). Most of urinary tract infections caused by bacteria, in particular, and in particular members of the family Enterobacteriaceae due to intestinal near the opening of the anal opening of the urethra, making it easier Entry of bacteria into the urinary tract and causing injury, and this is very common in women because of the installation of the urinary system anatomical female, if the palace and the urethra is so close to the source of infection Is the reason for the high rate in women because of the installation of the urinary system anatomical female, if the palace and the urethra is so close to the source of infection Is the reason for the high in women because of the installation of the urinary system anatomical female, if the palace and the urethra is so close to the source of infection Is the reason for the high rate of injuries in females in particular. It may also arise from injury to revive other bacteria, such as Chlamydia and rickettsial and yeasts and fungi (Roberts., 1991; Sim., 2001). And fungal infections come second after a bacterial infection in the urinary tract from hospitals Infections pathogens, Fungal infections of the kidneys and urinary tract occur most commonly as part of systemic fungal infections in patients with underlying immunodeficiency, focal urinary tract infections (UTI) with obstructive lesions, or as a result of indwelling catheters (Herberg J, Pahari A, Walters S, Levin M., 2009). The vast majority of fungal infections of the kidney and bladder result from Candida albicans and Candida spp. (Zarei Mahmoudabadi A, Keradmand AR., 2009) . Prior antibiotic therapy, diabetes, urinary tract pathology and malignancy have been considered as the risk factors in urinary tract candidiasis (Kauffman CA, Vazquez JA, Sobel JD., 2000). The application of urinary tract drainage devices can also trigger the infection (Kauffman CA, Vazquez JA, Sobel JD., 2000). Most patients with candiduria are asymptomatic, and the yeasts merely represent colonization (Herberg J, Pahari A, Walters S, Levin M., 2009). Infected patients may have dysuria, frequency, and suprapubic discomfort, but others have no symptoms. The clinical characteristics of fungal kidney infections depend on whether the disease presents acutely or insidiously. Acute infection of the kidney associated with candidemia is usually bilateral, consisting of multiple micro abscesses in the cortex and medulla. The kidney is the most common organ involved in systemic Candida infections. Chronic Candida kidney

infection is characterized by a sub acute to chronic course from ascending infection. Invasion of the renal parenchyma in such patients tends to involve the renal pelvis and medulla with sparing of the cortex. The kidney is usually the only organ involved and the infection tends to be unilateral. Blazers' and perinephric abscesses can occur (Lehner T., 1964).

MATERIALS and METHODS

Collection of urine specimen

Collected some sample generation of patients with infections of the urinary tract and patients admitted to the lobbies of the urinary tract in both sexes and different ages of the hospital in the city of Guntur There are a number of methods available for the collection of samples of urine for testing laboratory and the methods used in this Study's**.**

1. Midstream Clean Catch Specimen

The sample collection mid-discharge depends on the extent of urinary free sample of microbiological contamination existing in the anterior part of the urethra and urinary obtain a sample sound Untainted Sample (Koneman et al, 1997; Lohr et al., 1996). But in order to avoid external contamination in the sample patient urinary recommended a number of measures he needs to do to in order to reach a proper diagnosis of the situation and to get the best results and these measures are.

1. The patient to wash his hands first and then wash the penis several times As for the female is compelling them to wash the pelvis movement from front to back and not vice versa.
2**.** The patient to urinate a small amount of urine into the toilet and then empties the middle part of the urine into the pot and add sterile and airtight, this procedure ensures rid of the flora in the area of the urethra. With regard to the female, they have to keep lips of the vagina during unloading Labiae mediated fingers for preventing contact With the skin, and with regard to the dimensions of the male part is compelling them non circumcised during that especially children(Lohr et al., 1996; Sim., 2001).

2. Specimen Catheter

This method is used with children or the elderly who suffer from enlarged prostate glands, as well as patients with urinary tract and patients admitted to the hospital in such cases enters the tube the average diameter of the polyethylene into the urethra or bladder for disposal of urine, and this method is optimal in obtaining samples free from external contamination. (Forfar and Arneils., 1992). After obtaining urine samples from the patients were transferred

to the laboratory in a period not exceeding two hours in order to cultivate the appropriate circles.

Urine reagent strips for urinalysis.

Urine Reagent Strips (URS) for Urinalysis are firm plastic strips to which Several Different reagent areas are affixed. Depending on the product being Used, Urine Reagent Strips provide tests for Glucose, Billirubin, Ketones Acetoacetic acid Specific Gravity, Blood, pH, Protein, Urobilinogen, Nitrite, Leukocytes, and Ascorbic Acid in Urine. Test results may provide information regarding the status of carbohydrate metabolism, kidney and liver function, acid-base balance, and bacteriuria. Please refer to the outside box and bottle label for the specific test parameters of the product you are using.Urine Reagent Strips are packaged along with a drying agent in a plastic bottle with a twist-off cap. Each strip is stable and ready to use upon removal from the bottle. The entire reagent strip is disposable. Results are obtained by direct comparison of the test strip with the color blocks printed on the bottle label. No Calculations or laboratory instruments are required.

Cultivation of specimen

Starting the process before the implant could do with centrifugation of the sample for the concentration of bacterial cells in the bottom of the pot (Kunin et al., 1981). Amount taken (0.1) cm 3 of the sample and on the carpeted surface of the central (Nutrient Agar , Agar Agar, MacConkey agar, blood agar) , and then incubated, It must be noted and control the growth in the middle period (24-48 hours).(Sim, 2001; Cappuccino and Sherman., 2002). Movement of colonies developing on the central (MacConkey agar) as a compromise election and then isolate colonies grown in pure habitats amid agar nutritious (Nutrient Agar) until used in the conduct of diagnostic tests. medium used for antibiotic sensitive and resistant bacteria (muller-hinton agar), Medium used for fungi isolation (Czapek Dox Agar, Sabouraud Dextrose Agar) It must be noted and control the growth in the middle period (7 days). Medium used for antibiotic sensitive and resistant fungal (Sabouraud Dextrose Agar).

Biochemical characterization tests

1. Indole Test

Peptone water was inoculated with test organism and incubated at 37ºC for 24 to 48 hours. 5 ml of KOVAC‟s reagent was added along the side of the test tube to form a layer on the top. A positive reaction was indicated by the formation of pink ring at the junction (Steadman R,

Topley N.1998, Shames S R, Auweter S D & Finlay, B. B. , 2009, Rebecca N, Elizabeth M, Blanco M., Alonso M.P et al .,2009).

2. Carbohydrate Fermentation

Pure cultures were inoculated from the agar plates to sugar media and inoculated at 37ºC for 1-2 days. Positive test was shown by acid and gas production by a change in color of the media (pink with indicator) and the gas inside the Durham‟s tube (Steadman R, Topley N.1998, Shames S R, Auweter S D & Finlay, B. B. , 2009, Rebecca N, Elizabeth M, Blanco M., Alonso M.P et al .,2009).

3. Citrate Utilization Test

Test organism was inoculated in Simmons citrate agar and incubated at 37ºC for 2 days. Medium blue with a streak of growth was indicated in citrate utilizing bacteria (positive reaction) (Steadman R, Topley N.1998, Shames S R, Auweter S D & Finlay, B. B. , 2009, Rebecca N, Elizabeth M, Blanco M., Alonso M.P et al .,2009).

4. Urea's Test - Christensen Urea Agar

The test organism was inoculated heavily over the entire slope surface and incubated at 37. A positive reaction was indicated by a pink color of the medium. The alkaline pH produced changes the color of the medium to pink or red (Steadman R, Topley N.1998, Shames S R, Auweter S D & Finlay, B. B. , 2009, Rebecca N, Elizabeth M, Blanco M., Alonso M.P et al .,2009).

5. Triple Sugar Iron Agar Test

TSI agar was stabbed in the center of the boat and streaked on the slope with a needle charged with a single colony of the test organism. The tube incubated at 37 for 24-48 hours. A yellow butt and red slant showed glucose fermentation, yellow butt and yellow Slant showed glucose, lactose and sucrose fermentation. Gas produced was trapped in the medium (Steadman R, Topley N.1998, Shames S, R, Auweter S D & Finlay, B. B. , 2009, Rebecca N, Elizabeth M, Blanco M., Alonso M.P et al .,2009).

6. Methyl Red Test

Test organism was inoculated on glucose phosphate broth and incubated at 37 for 48 hrs. 5 to 6 drops of methyl red reagent was added to the culture. A red color indicated positive reaction. Negative tests were yellow in color. Positive reaction indicated the ability of the

organism to produce and maintain an acid pH (Steadman R, Topley N.1998, Shames S R, Auweter S D & Finlay, B. B. , 2009, Rebecca N, Elizabeth M, Blanco M., Alonso M.P et al .,2009).

7. Voges-Proskauer Test

The test organism was inoculated in glucose phosphate broth and incubated at 37 for 48 hours. Then VP reagent (1ml of 40 % potassium hydroxide 3 ml of 5% alpha napthol in absolute ethanol) was added. The tube was shaken vigorously to ensure maximum aeration. A positive result was indicated by the development of pink color in 2-5 minutes becoming crimson in 30 minutes (. (Steadman R, Topley N.1998, Shames S R, Auweter S D & Finlay, B. B. , 2009, Rebecca N, Elizabeth M, Blanco M., Alonso M.P et al .,2009).

8. Nitrate reduction test

Nitrate reduction tests were done according to Snell, "Snell EE and Wright LD, 1941". In brief, Nitrate broth was prepared (casein Peptone 0.5 gm%, Beef Extract 0.3 gm%, Potassium Nitrate 0.1gm%, Galactose 0.5 mg% and Disodium Phosphate 0.25 mg %) with Durham tubes (gas collector) in it. 0.8 mg% sulphanilic acid (dissolved in 5M acetic acid) and 0.6 mg% alpha naphthole (dissolved in 5M acetic acid) and Zinc dust were simultaneously added to those cultures. Acintobacteria calcoaceticus ATCC 19606 and E.coli ATCC 25922 was taken as negative and positive control strains respectively.

Antibiotic susceptibility testing of isolated *bacteria*

1. Determination of Minimum Inhibitory Concentration

The MIC values of amoxicillin, Ampicillin, Tetracycline, Gentamicin, Chloramphenicol, Streptomycin, Norfloxacillin, Ciprofloxacin, and Impanel, were determined by a broth dilution method using Mueller–Hinton broth (MHB), as recommended by the National Committee for Clinical Laboratory Standards (NCCLS) (NCCLS, 2000). About 5 x 10^{-4} cells in MHB were treated with different concentrations of antibiotics and shaken for 16 h at 37°C. The minimum concentration at which there was no visible turbidity was taken as the MIC of that antibiotic (Chakra borty SP et al. 2011).

2. Determination of Minimum Bactericidal Concentration

The MBC value of antibiotics was determined according to Chakra borty SP et al. 2011b. This is an extension of the MIC Procedure. Antibiotics treated bacterial culture, showing growth or no growth in the MIC tests were used for this test. Bacterial culture used for the

MIC test were inoculated onto the Mueller–Hinton agar and incubated at 37°C for 24 hours. Microbial growth or death was ascertained via no growth on Mueller–Hinton agar plate. The minimal concentration of the antibiotic that produced total cell death is the MBC.

3. Testing Susceptibility and resistance from by antibiotics

Antimicrobial susceptibility was determined by inoculated from a two colony) and freshly grown for 24 hours. With this culture, a bacterial lawn was prepared on Mueller-Hinton agar. Use serial dilution sample from (10^1 - 10^9) in test tubes and observe antibiotic susceptibility patterns against 10 antibiotics (amount of antibiotic per capsules in microgram (µg); Ampicillin (250 µg), Tetracycline (250 µg), Gentamicin (250 µg), Streptomycin (150µg), Chloramphenicol (250µg), Ciprofloxacin(250µg), Norfloxacillin(250µg), erythromycin(250mg), amoxicillin (250mg), Cefluexcin(250mg), Antibiotic capsules were obtained commercially from medical hospitals . The diameter of zone of bacterial growth inhibition surrounding the disc (including the antibiotics) was measured and compared with the standard for each drug. This gave a profile of drug susceptibility visa - vis antibiotic resistance.

Testing sensitivity and resistance for fungicide

This test ensures the use of serial dilution in concentration different and (Agar-well- diffusion method) was the measurement of effectiveness or inhibitory ability to five types from fungi isolated from medical device used in urinary tract against to five antifungal (Clotronazole, Fluconazole, Ivermectin, Terbin, Ketoconazole).

1. The preparation fungal inoculation

Depending on the method (10) was prepared pollen transfer part of developing fungal colony in the center (SDA) revitalized using a sterile needle and put it in a sealed tube (Vail) containing 5 ml of the solution Alveoli (Normal saline) solution and shake add fungal spores were (5 × 10).

2. Preparation of antifungal

The preparation of the base solution fungal antibiotics used and the concentration of 10,000 micrograms / ml and by the way (10) having been put 5 ml of material Dimethylsufloxide (DMSO) concentration of 100% in the tube is sealed (Vail) was added to 50 mg of each property and then Shake the solution vigorously using carburetor rotor (Vortex mixer), and this is the basis for the solution concentration of 10,000 micrograms / ml and then diluted

solution was attended by a concentration of 1000 mcg / ml of the base solution for anti-fungal and adding 10 ml of solution (DMSO) concentration of 100% to 1 ml of the basis of the solution concentration of 10,000 micrograms / ml, solution left under room temperature for 30 minutes before use, and after a previous preparations followed the following steps:

1. Taking 0.2 ml of vaccine innate minutes using a sterile pipette (Micropipette) and posted on the middle surface (ESDA) previously record in Petri dishes using a glass rod (Spreader) is a character (L) and sterilized with alcohol and flame. I left the dishes for 30 minutes in order to allow the vaccine and mildew that is absorbed by the middle.
2. Worked five drilling diameter of 5 mm in the middle fertilized by Saqib Corky, one of which represents a control.
3. Add 0.1 ml of fungal antibiotic solution prepared in advance to each hole using a sterile pipette minute (Micropipette), the dishes were incubated 28 degree C for (2-5) days, Qatar has been measuring the inhibition of the growth area (Inhibition zone) in units of a millimeter.

Statistical analysis

Results were analyzed statistically using Chi-square (X2) Qi-square to test the morale of all transactions used in the study at the level of probability 0.01.

RESULTS & DISCUSION

the results of the analysis of urine samples for outpatient Hospital Guntur city, recalling (Robert and his group 1999) to the need for microscopic examination and transplantation laboratory to identify the phenotypic changes and the microstructure of the generation of the patient in order to avoid accidental injuries. Were examined urine samples of the patient to investigate, first, pus cells Pus cells, which is an indication of the injury, as indicated Mahfouz (2002) to note pus cells is evidence of a bacterial infection of the motral races. As that illustrated most isolates microbial were producing cells pus associated with cases of infection and commensurate with the severity of the injury, which ranged between (7-1) cell in the microscopic field to the entire microscopic field coverage (Full High Power Field) has also been investigating the presence of red blood cells RBC deposit in lactation as some samples showed patients to contain the red blood cells that may be considered indicative of the presence of scratch in the urinary tract caused by the presence of a pebble moving inside (Mahfouz, 2002). Our results showed a case of epithelial cells in each lactation belonging to

patients with diabetes samples (100%), since this case is one of the side effects of diabetes [(Web MD, 2005).and also PH rang (4.5 - 8).]

Biochemical tests of Isolates

Two aim clinical isolates (AVMP-1 AND AVMP-2) were identified using standard biochemical tests. Purification of bacterial culture was done using the single colony isolation technique on Nutrient agar containing 10% sodium chloride, exhibited several types of colonies. Isolates were Gram negative and Isolates were Gram positive of gram negative isolates are indole positive. 100% of indole negative isolates were MR positive in serial dilution 10 and negative in serial dilution 10, Nitrate reduction test positive, and Carbohydrate fermentation (Glucose, Lactose, Sucrose) test positive and also positive for carbohydrate utilization with gas production on TSI. , VP test negative, Citrate test positive and Glucuronidase test negative ONPG test negative and Lysine utilization test positive. It was also revealed that 100% of Indole negative isolates were uropathogenic for giving hemolytic activity are all not motile On MacConkey agar hi medium. [Photo -1 and Photo-2]. Enterobacteriaeae (enterics) are Gram-negative bacteria that grow in the intestinal tract of humans and other animals. The IMViC (Indole, Methyl red Voges-Proskauer, and Citrate) tests are frequently employed for identification of this group of as Klebsiella, Enterobacter, and Escherichia coli (Barnes JH et al. 2003). Our study shows 100% of gram negative isolates were Indole negative this is due not to the production of the tryptophan's enzyme by those isolates that can break down the Amino acid tryptophan to indole. When Indole reacts with Para dimethylaminobenzaldehye (Kovac's reagent) a yellow-colored complex is produced**.** Indole negative of those isolates differentiates them from most Klebsiella sp and Enterobacter sp (McFadden JF 2000) Table 1a shows 100% of Indole negative isolates were MR test positive in serial dilution 10 and test negative in serial dilution 10 VP test negative MR-VP media contains glucose and peptone. All enterics oxidize glucose for energy; however the end products vary depending on bacterial enzymes. Our clinical isolates can ferment the glucose in MR-VP media that decreases the pH of the media below 4.4, detected by methyl red indicator which turns the media color cherry red Voges-Proskauer was negative for all indole positive isolates that may be due to lacking of production of acetyl methyl carbinol**.** MR positivity and VP negativity give strong support in favors of E.coli (MacFaddin JF2000) In our study all Carbohydrate fermentation tests (Lactose, Glucose, sucrose, Sorbitol) test positive to sample (AVMP 1, AVMP 2) all Indole positive isolates gave the positive citrate utilization test**.** This is due to metabolism of the citrate compound as

an only source of carbon in the media. So under basic condition Bromthymol blue cannot change the medium color from green to blue. This finding is highly correlated with the finding of (Kanungo S, etal ., 2009). Isolates were oxidase negative that may due non availability of cytochrome c oxidase and therefore cannot utilize oxygen for energy production with an electron transfer chain. Oxides negativity supports that isolates were in the Enterobacteriaeae family (PrescottLMetal.1999).

Photo 1: Standard biochemical tests of clinical isolatesAVMP-1 in serial dilution 10^{-4} collected from urine sample of UTI patient. ND = Tests are not done, + ve = tests are positive, - ve = tests are negative.

TEST NAME	MR	VP	Citrate	Indole	Glucuronidase	Nitrate	ONPG	Lysine	Lactose	Glucose	Sucrose	Sorbitol
RESULT	+	-	+	-	-	+	-	+	+	+	+	+

Photo 2: ***Standard biochemical tests of clinical isolate AVMP 2 in serial dilution 10^{-5} collected from urine sample of UTI patient. ND = Tests are not done, + ve = tests are positive, - ve = tests are negative.***

TEST NAME	MR	VP	Citrate	Indole	Glucuronidase	Nitrate	ONPG	Lysine	Lactose	Glucose	Sucrose	Sorbitol
RESULT	-	-	+	-	-	+	-	+	+	+	+	+

Infection In Urinary Tract From Used Medicals Device In Hospital

1. Urinary tract infection bacterial infections

Urinary tract infections (UTIs) are among the most common infectious diseases that occur in any society or health care. (Nicole L., 2005). Usually uncomplicated urinary tract infection in healthy adults Accor woman is pregnant, while complicated urinary tract infection (sagging) may Accor sexes and all age groups, and has often either structural or functional abnormalities associated with the urinary tract. One of the strange examples, such as calculus (stones), and the indwelling catheter or other drainage devices, and block and suppress the immune system, kidney failure, and kidney transplant and pregnancy. (P Lichtenberger, Hutton TM., 2008) UTI in the elderly is almost always a complex of enlarged prostate in men with women after menopause and in May interruption who have the last remaining void volume increased year. (Nicole pounds., 2001). And the likelihood of treatment failure and serious complications, particularly the development of antimicrobial resistance, it is more common in the box. Although a wide range of pathogens can cause funds, and Escherichia coli the most common residue; however, this body has acquired resistance even to agents usually this stipulated (Nicole pounds., 1997). In Bo Current search noted that injury UTI is highly prevalent in the community of patient hospital acquired infection catheter patient catheter. This can be due to poor maintenance hygiene in the patient community. % In the presence of high-protein, nitrates and leukocyte also similar to the results of previous studies. It indicates that the ICU community Syndrome patient should prompt commencement of inflammation and lead to a number of administrative and therapeutic problems (Figure 1, Table 5). Associated with a low probability of germ, about 20% in women with little signs or symptoms of UTI and <10% in symptoms residents.5,6 nursing homes in the symptoms of the elderly patient is catheterized, a positive result is less reliable with the presence of pellets White esterase difficulties <50% positive predictive value. However, some experts believe that this detection of nitrite in the patient's symptoms should prompt commencement of treatment (Gopal Rao G, Patel M., 2008), and this leads to a number of administrative and therapeutic problems. Quantitative standard microbiological to determine the bacterial large fitting is generally considered to be at least 108 cfu / L. In some groups are less specific: for men ≥106 cfu / L; and for women with symptoms of urinary tract infection is ≥105 cfu / symptoms germ L.5 (ASB) is common in the elderly, and the high detection of bacteria multidrug-resistant and AVMP2and AVMP1 is also a medical device that indicates the year also contributes to the increased frequency of microbes. With age> to 50% in women and> 35% of men over the age of 80 years. Last participated pathological conditions such as

diabetes or public indwelling catheter also contribute to increasing the frequency of this condition. (Wagenlehner FM, Naber KG, WeidnerW., 2005). Algorithms have been developed to improve the use of antimicrobials for suspected urinary tract infection in a nursing home, in the absence recommends that a minimum of UTI signs, urine should not be cultured BE must antimicrobial indescribable. (Loeb M, Brazil K, Lohfeld L, et al., 2005) has been made and the fact that the call for the performance of US doctors treating asymptomatic bacterial measure is not. (Total PA, B. Patel, 2007) . Sample showed (AVMP1, AVMP2) that have been identified through the screening and macrogen (sequence) in the laboratory department, in the medical device infection microorganisms showed (catheter) used urinary tract infection at a hospital in Guntur district. Sample has been planted (AVMP1) at a concentration of easing (10^{-4}), and the sample (AVMP2) at a concentration of easing (10^{-5}) on nutrient agar and blood Agar and MacConkey Agar showed the current growth sample results of the study (AVMP2, AVMP1) on all materials used. . Showed a sample colony (AVMP1, AVMP2) growth medium MacConkey Agar and fermentation sugar lactose, Sample showed (AVMP1, AVMP2) growth of blood Agar, PURPOSE Blood agar is used for of many types of fastidious bacteria. Isolation and It is also used to differentiate bacteria based on their hemolytic characteristics, especially within the genera The results observed growth in a sample (AVMP1) at a concentration of dilution (10^{4}) Hemolysin enzyme analysis and the emergence of alpha (The greenish zone around the colonies is due to incomplete lysis of red blood each. And beta (The clearing around the growth is due to complete lysis (Of red blood cells). This photograph was taken with transmitted light .It has been observed results growth in the sample (AVMP2) at a concentration of dilution (10^{5}) enzyme haemolysis analysis and the emergence of Alpha (The greenish zone around the colonies is(Due to incomplete lysis of red blood In this sample And beta (The clearing around the growth is due to complete lysis(Of red blood cells).The samples (AVMP1, AVMP2) to see the shape and kind through negative dye cram or positive for the dye Gram, The results showed in both samples positive for the dye Gram and negative dye Gram at 100 X. (photo3 : growth medium nutrient agar (A1 , A2), and growth medium MacConkey Agar(B1,B2),and growth medium blood Agar (C1, C2), under microscopy staining (D1 ,D2).Results sample 10^{4} (AVMP1),And 10^{5} AVMP2 under in table (1,2).

Table 1: Isolation Bacteria from medical devices used in UTI of hospital Guntur AVMP 1 in Serial dilution concentration (10^{-4})

Microorganisms	Number bacteria present in Medicals device at UTI in Serial dilution concentration (10^{-4})	Percentage of bacteria isolation from Medicals device at UTI Serial dilution concentration (10^{-4}) %
E.coli	7	38.8
Pseudomonas aeruginosa	3	16.6
Proteus mirabilis	2	11.1
Bacillus ceruse	4	22.2
Citrobacteria freundi	2	11.1
Total growth	18	100

Table 2 : Isolation Bacteria from medical devices used in UTI of hospital Guntur AVMP 2 in Serial dilution concentration (10^{-5})

Microorganisms	Number bacteria present in Medicals device at UTI in Serial dilution concentration (10^{-5})	Percentage of bacteria isolation from Medicals device at UTI Serial dilution concentration (10^{-5}) %
Klebsiella pneumonia	2	13.3
Staph aureus	5	33.3
Staph epidermis	3	20
Enterococcus faecalis	1	6.6
Entrobateria aerogenous	2	13.3
Bacillus stubilis	2	13.3
Total growth	15	100

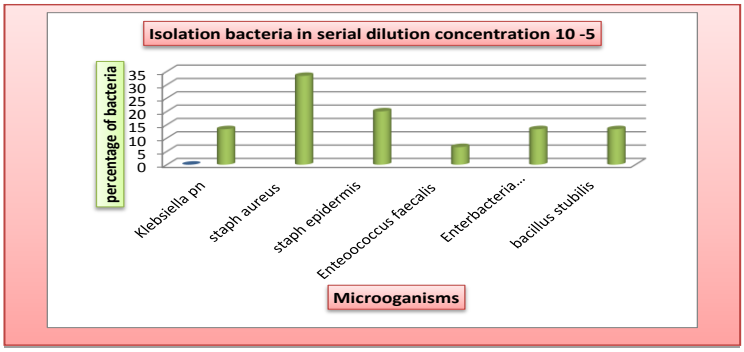


Figure 2: Isolation Bacteria from medical devices used in UTI of hospital Guntur AVMP 2 in Serial dilution concentration (10^{-5})

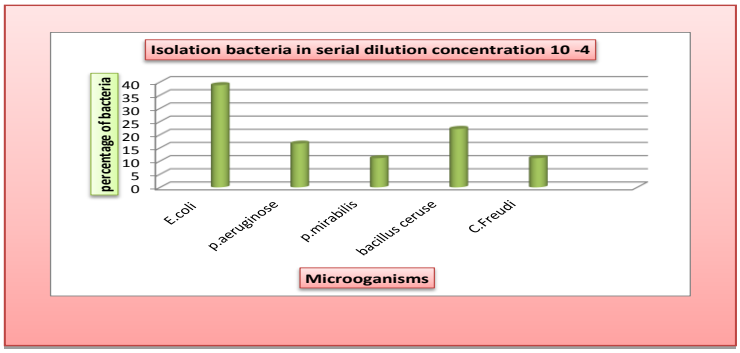
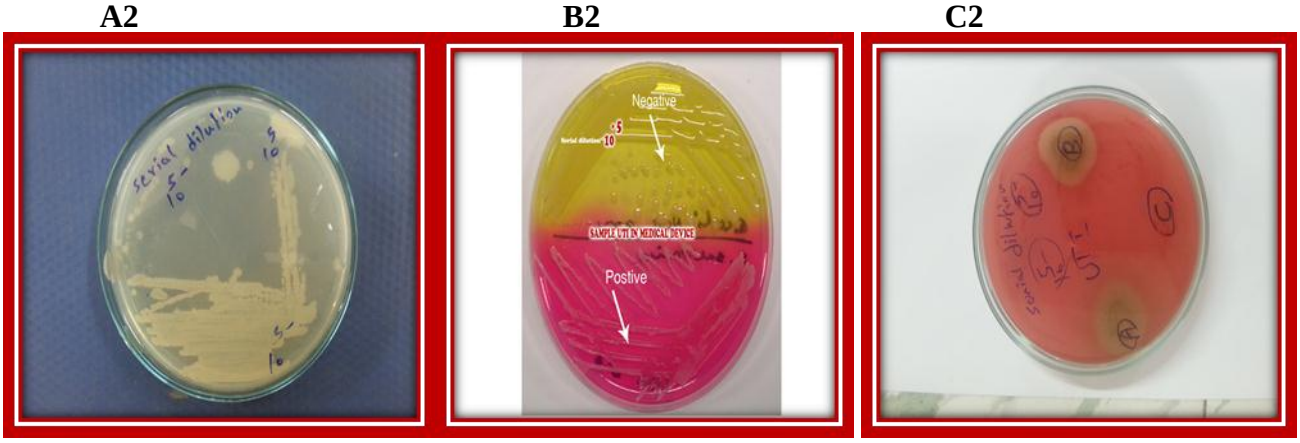
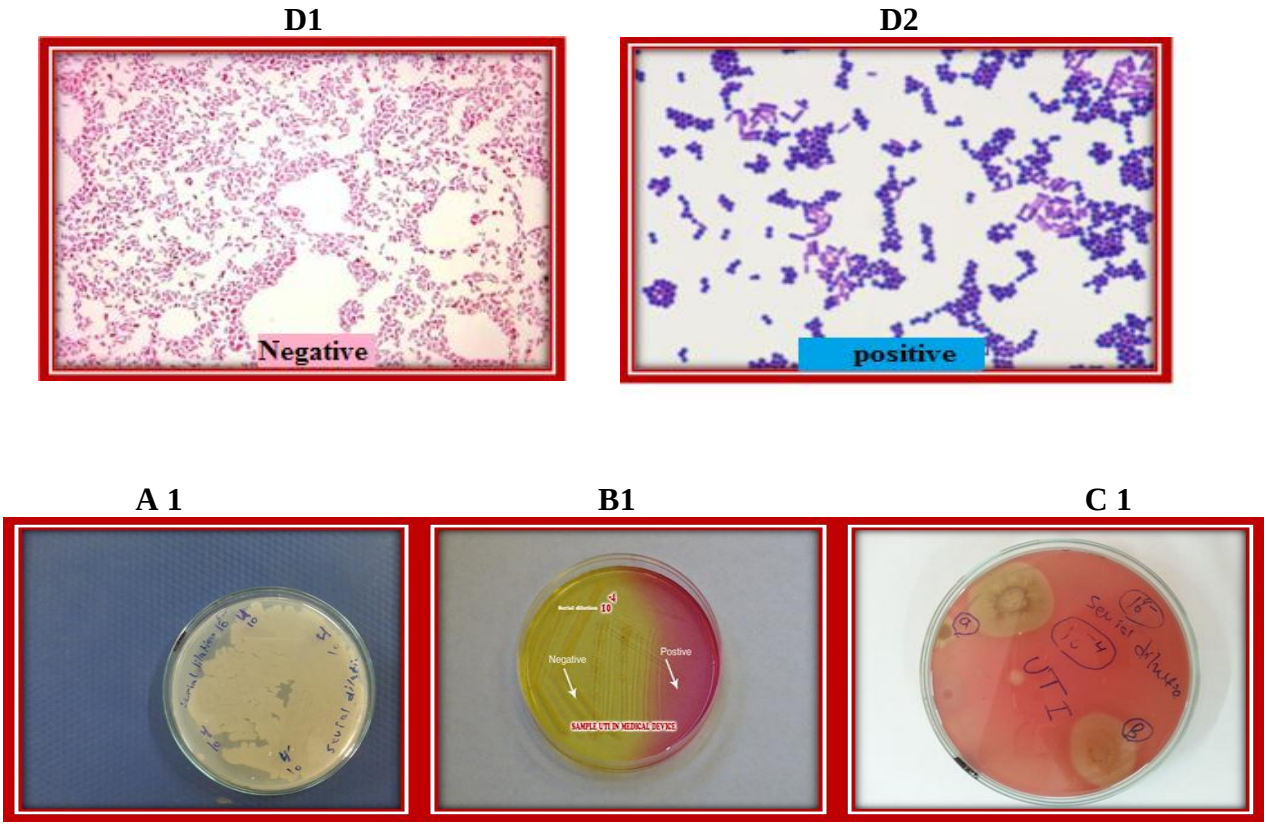


Figure 1: Isolation Bacteria from medical devices used in UTI of hospital Guntur AVMP 1 in Serial dilution concentration (10^{-4})





Fungal infection in urinary tract infections

2. Fungal infection in urinary tract infections

Fungal infections of the urinary tract increases in cases, mainly due to increased use of antibiotics, and hardware and indwelling urinary catheter. (Etienne M et al., 2007) can be symptoms or may remain without symptoms. It has been reported in a range of clinical manifestations range from asymptomatic colonization (the most common), and cystitis, Pyelonephritis, inflammation of the kidneys, and sepsis and the presence of fungi in the blood. (Etienne M et al., 2007) and there is a tendency for drainage structures instead of kidney meat. (Etienne M et al., 2007). It may become Funguria satisfactory depending on host factors, so management relies on the basic state of health of the patient. Candida is the most common fungus that infects the urinary tract. (Etienne M et al., 2007). And the fungus Candida albicans number (10) and percentage (34.3%) if divided between two types The first type is Candidaspp and number (6) and the percentage (20.6) and Candida second egg Oll (4) and proportion (13.7%) in this study, however, it may be them other fungi relationship according to previous anti-fungal treatment.

As for the sex of the second Pencillium, the number of fungi Pencillium growth in this study (5) and proportion (17.2%), Pencillium is a type of fungus everywhere inside the hospital, preferring cool and moderate climates, usually found in any part of the organic material is available. Ppalartmam species of Pencillium are among the well-known representatives of Eurotiales basically live on the organic material biodegradable refundable. Defined templates, it is among the main causes of food spoilage, and especially the species under the genus Pencillium (Samson RA, Seifert KA, Capers AF, for Houbraken JA, and Frisvad JC 2004). Many species produce mycotoxins are highly toxic.

As for the third race were Aspergillus and the number of growth in this study (14), the fungal growth rate in this study (48.1%), and if divided between two types of Aspergillus, the first type AspergillusNigar were a number of developing fungi (8) and proportion (27.5%), type II Aspergillus flavus and the number of developing fungus (6) and the percentage (20.6%), Aspergillus is not a common cause of urinary tract infections fungal (in UTI). Basic Aspergillus infection in the urinary tract is rare enough, even more so in patients who suffer from immune. (SmaldoneMC, et al., 2006) (Bible et al., 2011). Patients who have a history of kidney aspergillum's system. (Bibler MR, et al., 1987).Ten of these men, and it was all the underlying diseases that are predisposed to fungal infection. In patients with weakened immune never, (SmaldoneMC, et al., 2006), (VuruskanH, et al., 2005) . Clogged ureter. (BiblerMR, et al., 1987). Artery colorectal, kidney failure, and sepsis (SmaldoneMC, et al., 2006) has stated (SmaldoneMC, et al., 2006) or the deployment of Aspergillus varying manifestations of disease in the form of balls or Aspergillus Bazuhair. However, mild spastic pain and discomfort, not our patient do not reflect any fever or severe constitutional symptoms. Aspergillus published in aspergillomas prostate or other form of pneumonia (Ludwig M, et al., 2005). It has been reported even in patients who suffer from immune. This can happen because of the use of corticosteroid or surgical intervention. (Ludwig M, et al., 2005). I did not see the signs posted in our patient. Visible passage of balls Aspergillus in the urine is a view of the dramatic symptoms of interesting and frightening for the patient. Rarely, it had previously said in this patient with acute myeloid leukemia. (Turngton KG, et al., 1979). To our knowledge, our patient and immunocompetent host the first to put BazuhairAspergillus. It is recommended that funguria symptoms in patients in hospitals may not be treated as morbidity and mortality rates do not affect. It may be just a disease, such as bacterial index in the elderly have. (Simpson C, et al., 2004) is recommended. It's only treatment when symptoms are funguria or in the case of host factors increase the risk of the

presence of fungi in the blood. It should be tried (Etienne M, et al., 2007). Reduction of risk factors such as indwelling catheter removal and improve the use of antibiotics. In patients with weakened immune, and follow the approach along with the meal topical and systemic breakfast and end urological access to extract, wash and there may be a need debunking. (Irby PB, et al., 1990). However, in immunologically, and surgical treatment is not necessary. In fact, Aspergillus prostate without symptoms has been reported to spread after the eradication of routine transurethral. (Ludwig M, et al., 2005). Aspergillus is said to work more malignantly of Candida in the urinary tract. (Irby PB, et al., 1990) .Has already recommended treatment with amphotericin B. (Khan ZU, et al., 1995). However, it has proven to be an effective treatment Itraconazole even in cases of resistance amphotericin B. (Irby PB, et al., 1990).(Table 3 Figure 3, isolation and percentage fungal from medicals devices used in UTI of hospitals Guntur).and growth fungal in different medium in photo 4 (A1 ,A2, B1,B2 ,C1 ,C2 ,D1 ,D2).

Table 3: Isolation fungal from medical devices used in UTI of hospital Guntur

Fungal isolation	Number fungal presents in Medical device used at UTI patient	Percentage of fungal isolation from Medical device used at UTI %
Aspergillus Niger	**6**	**20.6**
Aspergillus flavus	**8**	**27.5**
Pencillium spp	**5**	**17.2**
Candida spp	**6**	**20.6**
Candida albicans	**4**	**13.7**
Total growth	**29**	**100**

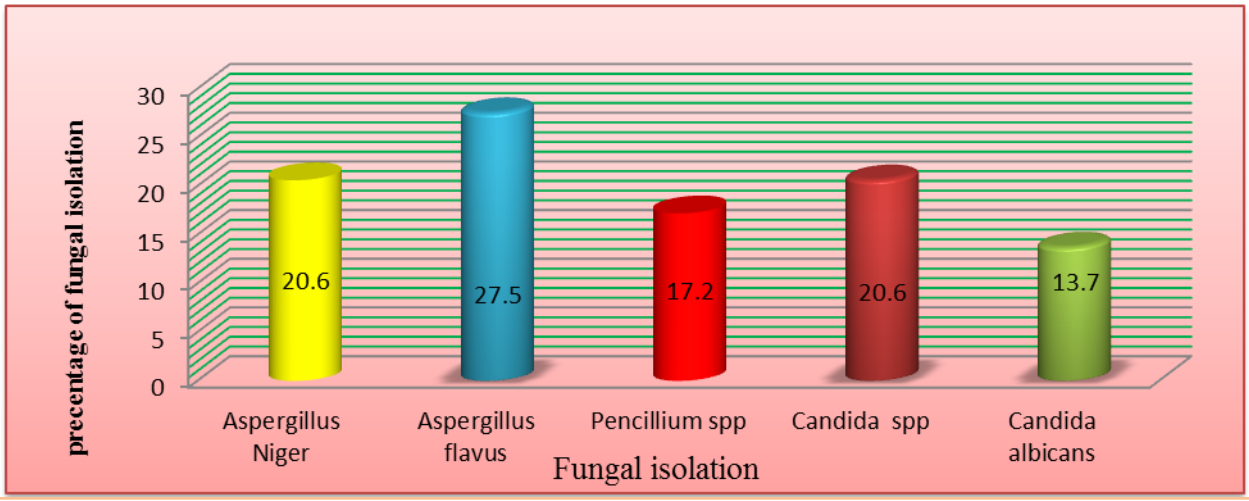
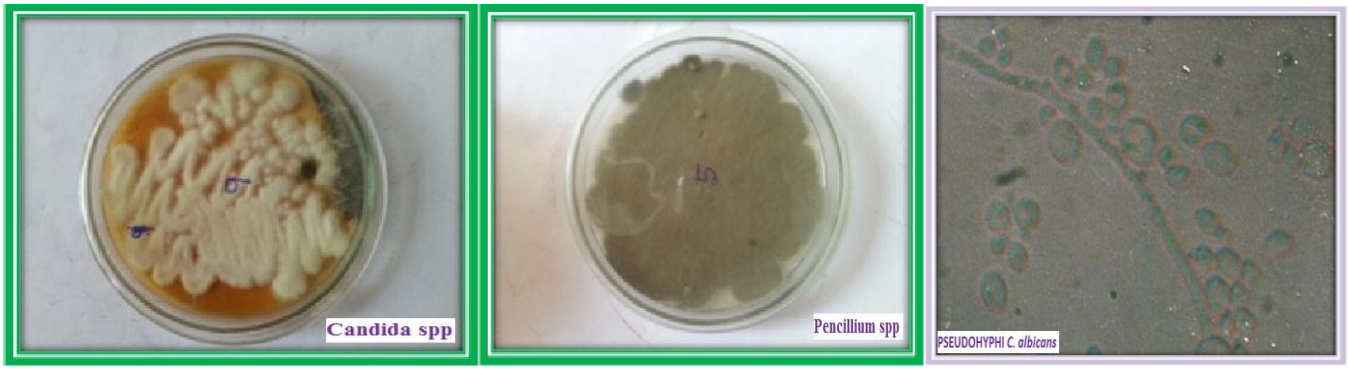


Figure 3: Percentage of fungal isolation from medical devices used in UTI of hospital Guntur

A 1 A2 B1

B2 C1 C2



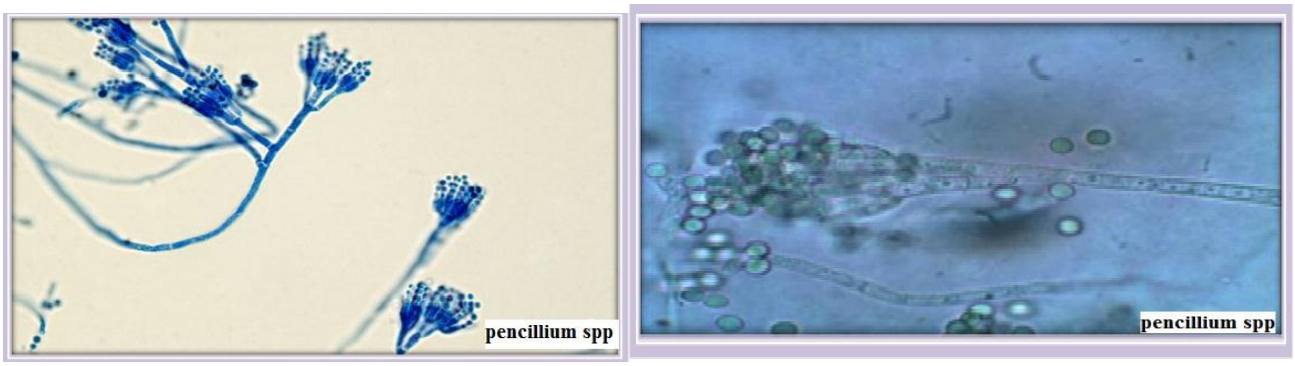
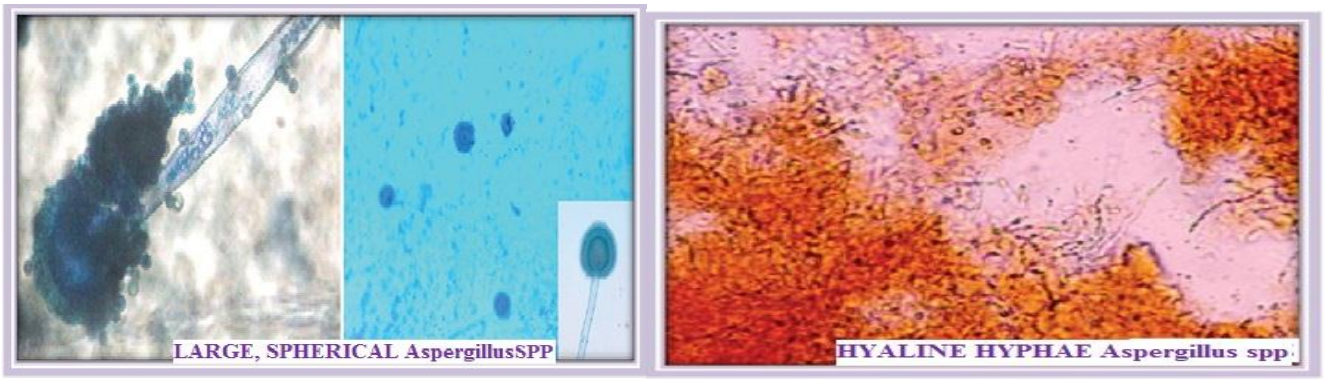


Photo 4: fungal growth in different medium , fungal under microscopy after staining slides



dye lacto phenol. Microscopic examination to test the dye grams have been adopted since found that the yeast Candida oval cells to spherical, oval or rectangular positive for this dye. (Murray et al., 1999)and the formation of germination tube Is a test of the important qualities of the rapid diagnosis of C. albicans was also noted that growth in the emergence of elongated tubular cells produced during the direct examination form. Must either other species, it is. Eggs and C. albicans. Be able to configure the germination tube (Murray et al., 1999). Sex second Pencillium and third sex Aspergillus has appeared after staining dye Lacto phenol, show morphology to fungal, and under show photo graph explains under microscopy after staining

Sensitivity and resistance of microbes to antibiotics

Sensitivity and resistance of bacteria to antibiotics is essential that there is an update of sensitivity and resistance to pathogenic microbes in the urinary tract available antibiotics, which should be based on selection of anti appropriate for the treatment of urinary tract infection in patients with men and pregnant women on the basis of the sensitivity of this test is to determine the sensitivity of nurses in the laboratory pattern. It should also be chosen antibiotic toxic feature, and the weakness of the pregnant woman and the fetus together mother, and that there is no analysis of the state of the pregnant and the fetus the mother of his body cells, and in this study, the sensitivity of the positive and negative isolates for testing grams, and the focus of the serial dilution (10^{4} and 10^{5}) at different concentration. The reason to use this amount of antibiotics to unite the effect of different antibiotics in all isolates, regardless of the class that their isolation, and the aim of the most resistant isolates in preparation for the diagnosis of resistance recipe her genetically and see the size of bacteria resistant to antibiotics and diameters of the inhibition of bacterial isolates region selection isolated from urinary tract infection in patients, and the results indicate the countries of the region inhibition and vital anti resistance screening has been an examination of antibiotic-resistant AVMP1 and AVMP2 by agar well diffusion method against 10 antibiotics (ampicillin, streptomycin, tetracycline, Norfloxacin Chloramphenicol, aggentomycin, ciprofloxacin, erythromycin, amoxicillin and cefluexcin) bacterial colony that usually occurs in all dilutions ranging from 10^{-1} to 10^{-4}, and it appears as it was named one isolated colony in 10^{4} mitigation as AVMP 1 and screened antibiotic resistance. The results shown in (photo 5,plate1, table4,and fig 4), which include either been AVMP1 isolated in serial dilution strain (10^{4}) showed the following antibiotics (amoxicillin, ampicillin, tetracycline , Norfloxacin, ciproflexin and cefluexcin) sensitive direction sample AVMP 1 in concentration ease (10^{4}),

As shown the following antibiotics (streptomycin, chloramphenicol, gentamicin, erythromycin) showed high resistance direction of a sample at a concentration of serial dilution (10^4), either AVMP2 been isolated in serial dilution (10^5) colony solitary strain also to allergies and antibiotic resistance screening. However, the ciproflexin was in the sample AVMP2 extremely sensitive to all other antibiotics as in (photo 6 ,plate 2, table4 and Figure 5).

TABLE 4:

	Zone of inhibition Mm									
Antibacterial agents / Sample	AMP	STREP	TETRA	CHLORO	NORF	AGGENT	CIPRO	ERYTHO	AMOXA	CEFLU
In serial dilution con.10^{-4} µg / mL	**0**	**17**	**0**	**23**	**0**	**15**	**0**	**52**	**0**	**0**
In serial dilution con.10^{-5}µg / mL	**12**	**18**	**19**	**16**	**21**	**20**	**0**	**23**	**12**	**21**

Photo 5: screening of minmum inhibitory concentration of antibiotics (MIC) in serial dilution concentration (10^{-4} µg / mL) AVMP1 to sample bacteria gram positive and negative

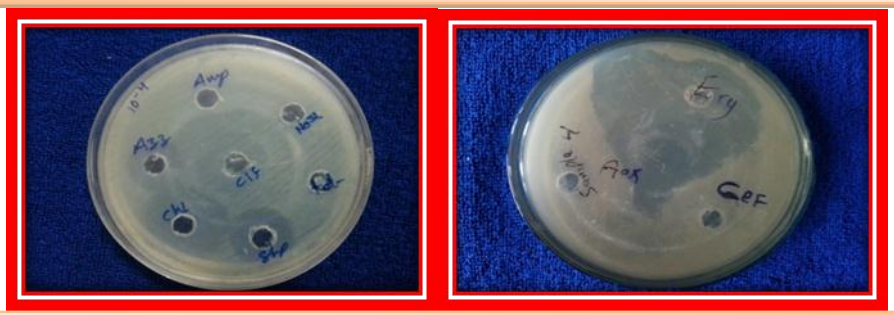
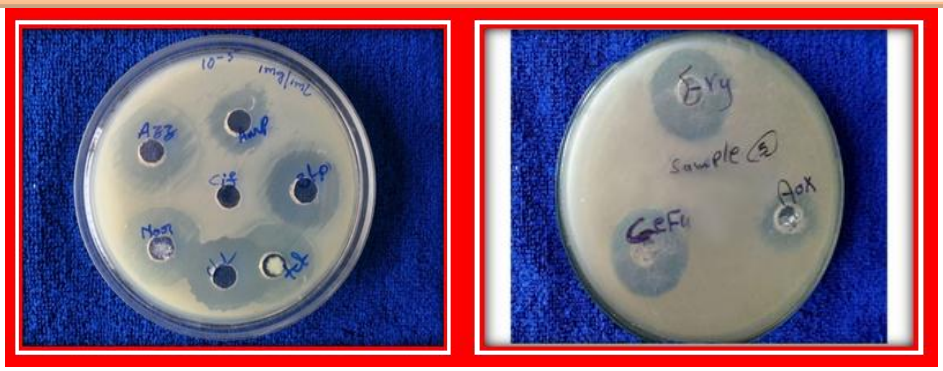
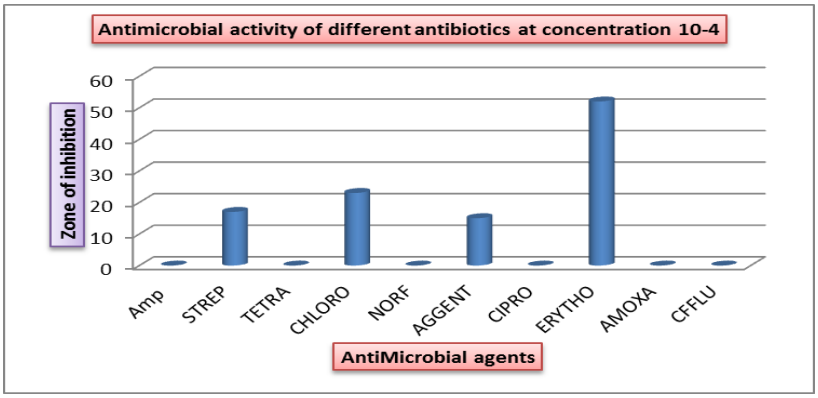
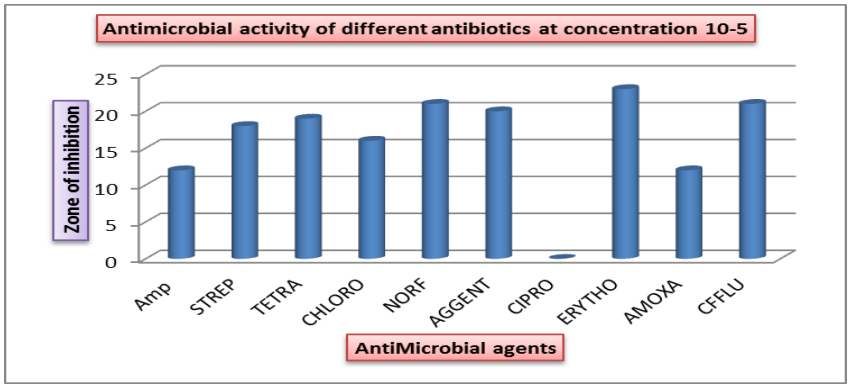


Photo 6 : Screening of minmum inhibitory concentration of antibiotics (MIC) in serial dilution concentration (10^{-5} µg / mL) AVMP2 to sample bacteria gram positive and negative







Sensitive and resistance fungal to antibiotics

Method was used contained in the (1987) World Health Organization for the preparation of antibiotic solutions, and Treasury of antibiotics and the use of solvents (DMSO) dimethyl Sulphoxide to help the melting of these antibiotics for the preparation of Treasury solutions that these solvents do not interfere with the effectiveness of the counter (al-Moussawi, 2003), On this basis, I attended the solvent tablets alone to study the impact on the fungus, it was noted the presence of the solvent effect on fungi isolated from medical devices used in inflammation of the urinary tract in hospitals. The preparation of five antibiotics, a fungal (Terbin, Ivenmycin, Clotrimazole,ketoconazole ,Fluconazole) The ratio of the concentration of antibiotic tablets in ug / ml (Fluconazole 250 , Ketoconazole 250 , Clotronazole 250 , Ivenmycin 150 , Terbin150) and after the piece was measured Qatar mm zone of inhibition for each counter using a ruler. The results show the vast majority of isolates resistant to antibiotics affecting risen relative increase of the concentration of anti We also note that isolates yeast and fungi gave Qatar the inhibition of each Anti different from other areas. The results of the genus Candida in the serial dilution 10^{-1} to 10^{-4} resistant to all antibiotics in the first fungal, and the results shown in (figure 8,9, plate A1,A2, table4), Aspergillus isolated prosecutors in serial dilution 10^{-1} to 10^{-5}, and the results showed the isolation of Aspergillus prosecution direction of the five high-fungal antibiotic resistant to all antibiotics five innate, it

was the results of the first dish direction fungal antibiotics five, and the results shown in (figure 6,7, plate B1,B2, table 5). Pencillium isolated in serial dilution 10^{-1} to 10^{-3} and the results showed isolation pencillium direction of the five fungal antibiotics has been observed by two fungal antibiotic resistance is high (Fluconazole, Ketoconazole) and three of fungal antibiotics are sensitive (Clotronazole, Ivenmycin, Terbin), and the results shown in(figure 10,11, plate C1,C2, table 6),

Table 4: Antifungal activity of antibiotics at concentration serial dilution 10^{-4} **µg / mL** Sample **Candida spp**

	Zone of inhibition Mm				
Antifungal activity / Sample Candida spp	Terbin	Ivenmycin	Clotrimazole	Ketoconazole	Fluconazole
In serial dilution con.10^{-4} µg / mL P1	**12**	**13**	**17**	**14**	**5**
In serial dilution con. 10^{-4} µg / mL P2	**13**	**12**	**18**	**13**	**7**

Table 5: Antifungal activity of antibiotics at concentration serial dilution 10^{-5} µg/ml Sample Aspergillus spp

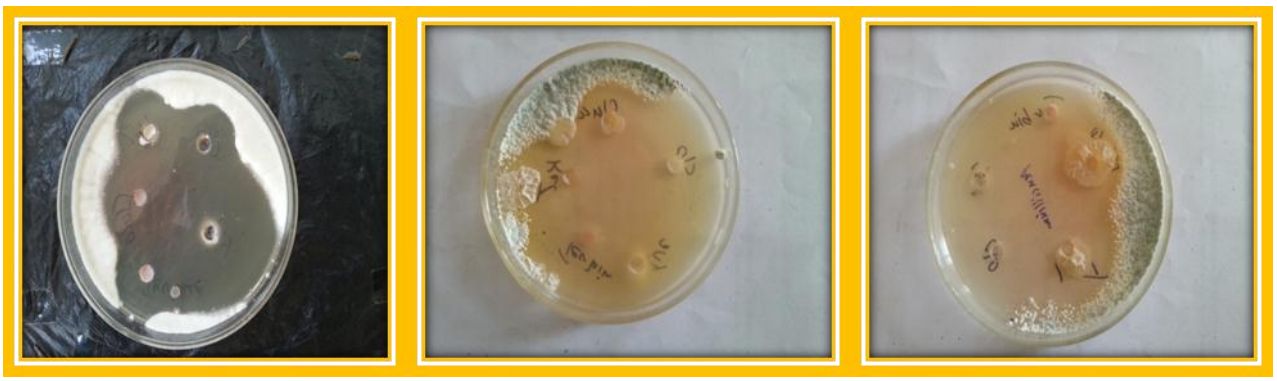
	Zone of inhibition Mm				
Antifungal activity / Sample Aspergillus spp	Terbin	Ivenmycin	Clotrimazole	Ketoconazole	Fluconazole
In serial dilution con.**10^{-5} µg / mL** P1	**25**	**56**	**19**	**21**	**27**
In serial dilution con. **10^{-5}µg / mL** P2	**56**	**44**	**18**	**23**	**30**

Table 6: Antifungal activity of antibiotics at concentration serial dilution 10^{-3} µg / mL Sample

	Zone of inhibition Mm				
Antifungal activity / Sample pencillium spp	Terbin	Ivenmycin	Clotrimazole	Ketoconazole	Fluconazole
In serial dilution con.10^{-3} µg / mL P1	**0**	**0**	**0**	**26**	**22**
In serial dilution con. 10^{-3} µg / mL P2	**0**	**0**	**0**	**36**	**26**

A1 A2 B1

B2 C1 C2



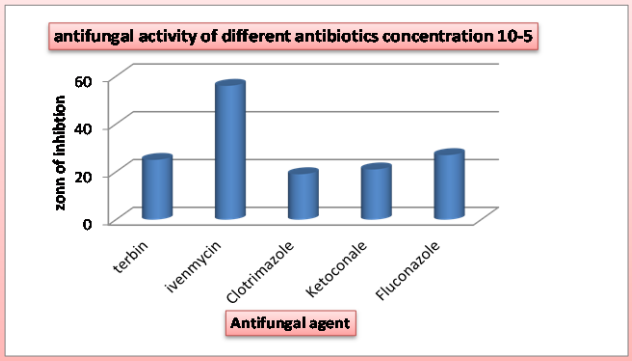


Figure 6: Antifungal activity of antibiotics at concentration serial dilution 10^{-5} µg/ml Sample Aspergillus spp p1

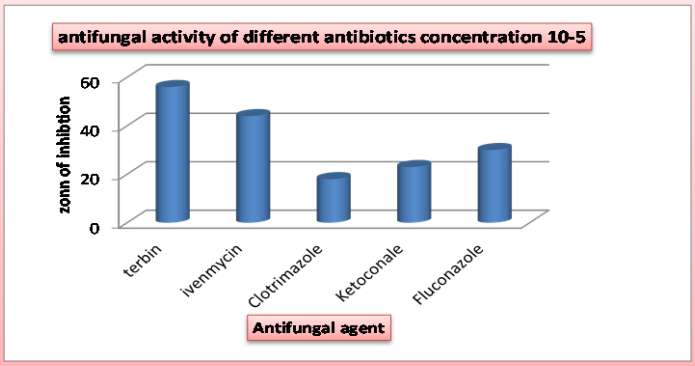


Figure 7: Antifungal activity of antibiotics at concentration serial dilution 10^{-5} µg/ml Sample Aspergillus spp p 2

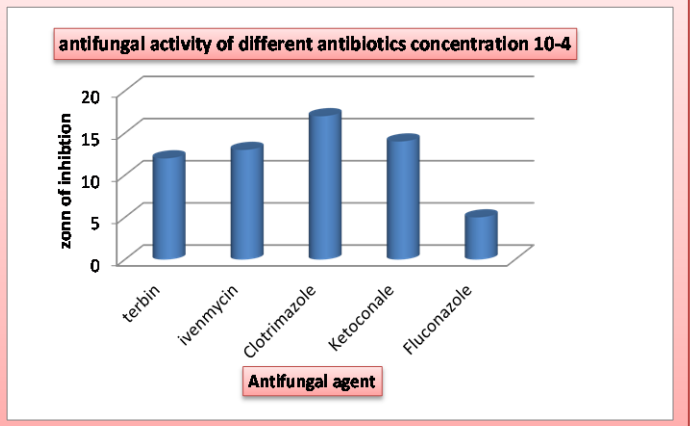


Figure 8: Table 4: Antifungal activity of antibiotics at concentration serial dilution 10^{-4} µg / mL Sample Candida spp

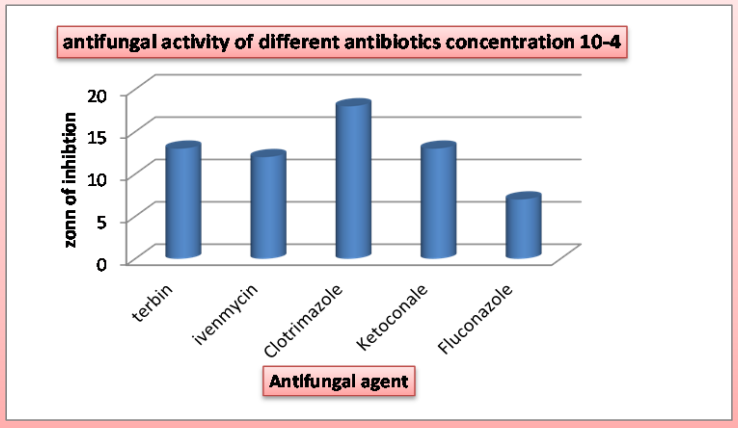


Figure 9: Antifungal activity of antibiotics at concentration serial dilution 10^{-4} µg / mL Sample Candida spp P 2

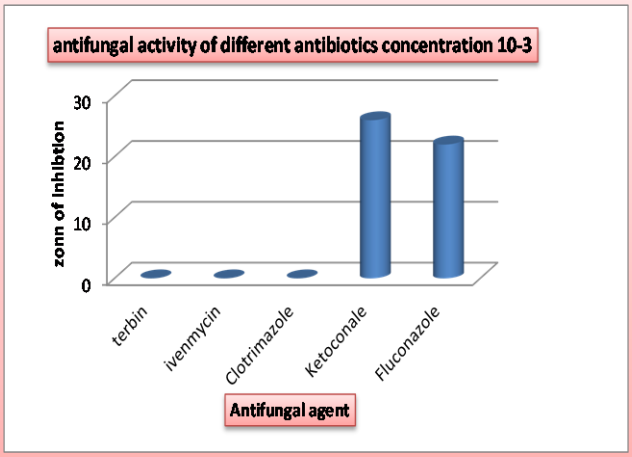


Figure 10 : Antifungal activity of antibiotics at concentration serial dilution 10^{-3} µg / mL Sample pencillium spp P 1

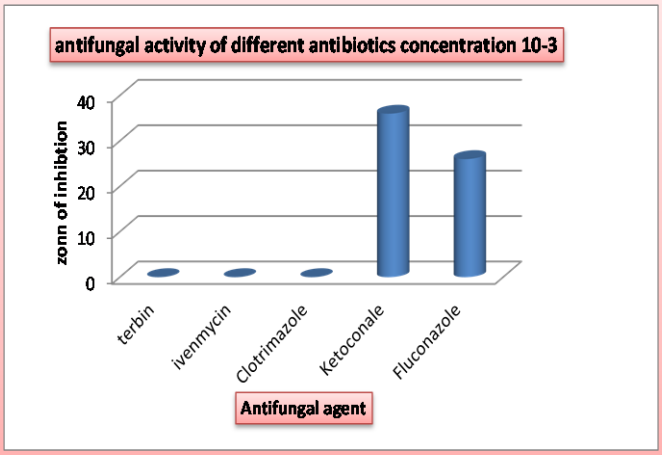


Figure 11 : Antifungal activity of antibiotics at concentration serial dilution 10^{-3} µg / mL Sample pencillium spp P 2

CONCLUSION

The increasing use of indwelling foreign bodies has become essential in modern day clinical practice; however, their use is associated with a multitude of complications, the most common being infection Nowadays, it is generally accepted that the Nowadays, it is generally accepted that the consequences of UTI are substantial, both in terms of morbidity and mortality as well as in terms of financial resources expended. The causative pathogens attach to the surfaces of the medical devices and develop biofilms, leading to an evasion of host defence mechanisms and to aphenotypic resistance to antimicrobial agents. Thus, conservative manage different a role in preventing medical device-related complications, such as metastatic sepsis, yet in most situations It is impossible to eradicate the primary focus Re of.

ACKNOWLEDGEMENT

The authors express gratefulness to Assistant Professor Amrutha V. Audipudi Acharya Nagarjuna University, Department Microbiology for providing the facilities to execute these studies.

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