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# THE INHIBITORY EFFECT OF LACTOBACILLI ON UROPATHOGENIC PSEUDOMONAS IN URINARY TRACT INFECTION

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The appearance of multiresistant bacteria in recurrent UTI calls for alternative and advanced medical solutions, one promising alternative is the use probiotics. Lactobacillus is a major part of the lactic acid bacteria group, which inhibits the growth of some harmful bacteria as Uropathogenic *pseudomonas*. So it is effective as probiotic in prevention and treatment of recurrent UTIs in women. The study was conducted on 100 patients in urology outpatient clinics Beni–Suef university hospital, Faculty of Medicine, Beni - Suef University.100patients from the urology outpatient they were suffering from recurrent urinary tract infection, from whom the Midstream urine samples (100 samples) were collected after taking the patient's consent.

*Pseudomonas aeruginosa* was the most 3rd predominant isolated organism followed by *E.coli* (26%) ,as pure isolate in upper UTIs (23%) , while *staphylococcus saprophyticus* was the most predominant isolate in pure culture in lower UTIs (28%).Standard strain (ATTC 1259) was studied for lactic acid production and its inhibitory effect on Uropathogenic *Pseudomonas aeruginosa*.

The inhibitory effect *lactobacillus* Standard strains (ATTC 1259) species on the 15 uropathogenic *Pseudomonas aeruginosa* isolates. All strains of *Pseudomonas aeruginosa* were inhibited by supernatant containing lactic acid of the tested lactobacillus strain. The growth up to the well (no inhibition) the zone is recorded 6 mm.

Lactobacillus acidophilus ATCC L1259 (standard strains) had the mean diameter of inhibitory zone (19.5mm), that mean the ability tested of *lactobacillus acidophilus ATCC L 1259* to inhibit growth of all uropathogenic *Pseudomonas aeruginosa strains.*

**Keywords***: Pseudomonas aeruginosa , lactobacillus, E.coli*

## INTRODUCTION

UTI is one of the most common type of community-acquired, hospital-acquired and recurrent type of infection that occur more commonly in women (Mulvey, 2002).

*Pseudomonas aeruginosa* is the 3rdof organisms in UTI. It possesses specific virulence factors that enable it to colonize and produce symptoms of upper or lower UTIs.

### UTI in women

Lower UTI in women is responsible for significant symptoms, morbidity and loss of life quality. It is estimated that several hundred millions of women suffer from UTI, with costs to health care providers amounting to over six billion annually worldwide. The incidence of uncomplicated UTI in approximately 0-5% episode per year with recurrence rate of between 27% and 48%. UTI is considered a problem in pregnancies affecting around 5% of women, and of those 20% may develop pyelonephritis((Bruce*et al.,* 2009).

Classically, $10^5$ or more colony forming organisms per ml of urine have been regarded as the acceptable bacterial count for UTIs. However, in women, counts of $10^3$ organisms/ml, particularly if associated with irritative bladder symptoms and the presence of increased numbers of white blood cells in the urine are now regarded as indicative of true UTI(Stamm and Raz, 2005).

The usual symptoms of dysuria, frequency of micturition, and occasionally haematuria (particularly terminal) are not always pre nsent, and asymptomatic bacteriuria ($\geq 10^5$/ml) may occur. Asymptomatic bacteriuria tends to increase with age, and may occur in up to 10%–15% of post-menopausal women. Gram negative organisms, particularly *E coli* (up to 85%) are the causative agents in most women suffering from UTI, followed by *Enterococcus faecalis* and *Pseudomonas aeruginosa*(Foxman*et al.,* 2011).

Fifty percentages of women will suffer from at least one urinary tract infection (UTI) during their adult life. UTIs account for around 5% of consultations in General Practice and are the second commonest infection after respiratory infections (DoH, 1998).15% of community use of antibiotics is for UTIs (DoH, 1998).A few women will suffer significant morbidity from the effects of pyelonephritis, an infection of the kidney substance, usually caused by infection ascending from the bladder. Most of those suffering morbidity will have underlying causatory abnormalities. 25% of those who have had one infection will have at least one further infection, and sometimes, multiple recurrences (Stapleton, 1999).Some will develop interstitial cystitis, a debilitating disease of unknown aetiology presumed to originate from recurrent UTI.

Another important type of UTI is the asymptomatic bacteriuria which is defined as the presence of more than100, 000 cfu per ml of voided urine in subjects with no symptoms of UTI and can originate in the bladder or the kidneys. Pregnant and elderly women have the highest rates of incidence of asymptomatic bacteriuria. Treatment is not recommended in the routine practice for asymptomatic bacteriuria, except in pregnant women and individuals undergoing invasive procedures. The microorganisms most frequently found as a cause of asymptomatic bacteriuria are *E. coli*, *P. aeruginosa*, and Gram-positive bacteria such as *Enterococcus* and *S. aureus.* Urinary catheters are a route of entry for bacteria. Between 10 and 20% of hospitalized patients are catheterized. Catheter-associated UTIs account for 40% of all nosocomial infections and are the most common source of Gram negative bacteremia in hospitalized patients. The role of biofilm forming pathogens in catheter-associated UTIs is explained in the present review. The pathogens most frequently found in this type of UTI are *E. coli*, *Proteus*, *Enterococcus*, *Pseudomonas*, *Enterobacter* ,*Serratia ,* and *Candida* spp., being normally acquired exogenously via manipulation of the catheter and drainage device.

UTIs may involve only the lower urinary tract (Cystitis), or it may involve the upper urinary tract (Pyelonephritis). Asymptomatic bacteriuria if the urine contains significant bacterial count but there are no symptoms. UTI is considered complicated if the person has diabetes mellitus, is pregnant or immunocompromised. While UTI is considered uncomplicated if a women is healthy and premenopausal(Colgan, 2006).

*Pseudomonas aeruginosa* is a common gram-negative rod-shaped bacterium that can cause disease in plants and animals, including humans. A species of considerable medical importance, *P. aeruginosa* is a prototypical "multidrug resistant (MDR) pathogen" recognised for its ubiquity, its intrinsically advanced antibiotic resistance mechanisms, and its association with serious illnesses especially nosocomial infections such as ventilator-associated pneumonia and various sepsis syndromes. The organism is considered opportunistic except in immunocompromised individuals, but the organism does produce a range of clinically important infections in the immunocompetent and/or in situations where no pre-existing vulnerability is required e.g. hot tub folliculitis. In all infections produced by *P. aeruginosa,* treatment is dually complicated by the organism's resistance profile, which may lead to treatment failure and/or expose patients to untoward adverse effects from advanced antibiotic drug regimens.In all infections produced by *P. aeruginosa,* treatment is dually complicated by the organism's resistance profile, which may lead to treatment failure and/or expose patients to untoward adverse effects from advanced antibiotic drug regimens. This dilemma is a central clinical problem in the field of antimicrobial resistance.

It is citrate, catalase, and oxidase positive. It is found in soil, water, skin flora, and most man-made environments throughout the world. It thrives not only in normal atmospheres, but also in hypoxic atmospheres, thus has colonized many natural and artificial environments. It uses a wide range of organic material for food; in animals, its versatility enables the organism to infect damaged tissues or those with reduced immunity. The symptoms of such infections are generalized inflammation and sepsis. If such colonization occur in critical body organs, such as the lungs, the urinary tract, and kidneys, the results can be fatal (Balcht *et al.,* 1994).

**Antibiotic resistance**

One of the most worrisome characteristics of *P. aeruginosa* is its low antibiotic susceptibility, which is attributable to a concerted action of multidrug efflux pumps with chromosomally encoded antibiotic resistance genes (e.g., *mexAB*, *mexXY*, etc.) and the low permeability of the bacterial cellular envelopes.(Poole ,2004).In addition to this intrinsic resistance, *P. aeruginosa* easily develops acquired resistance either by mutation in chromosomally encoded genes or by the horizontal gene transfer of antibiotic resistance determinants. Development of multidrug resistance by *P. aeruginosa* isolates requires several different genetic events, including acquisition of different mutations and/or horizontal transfer of antibiotic resistance genes. Hypermutationfavours the selection of mutation-driven antibiotic resistance in *P. aeruginosa* strains producing chronic infections, whereas the clustering of several different antibiotic resistance genes in integrons favors the concerted acquisition of antibiotic resistance determinants. Some recent studies have shown phenotypic resistance associated to biofilm formation or to the emergence of small-colony variants may be

important in the response of *P. aeruginosa* populations to antibiotics treatment.(Cornelis , 2008).

**Probiotic**

Probiotics are defined as "Live microorganisms which when administered in adequate amounts confer a health benefit on the host" (Hamilton and Bruck, 2003).

The term "probiotics" was first introduced in 1953 by Werner Kollath. Probiotics were defined as microbially derived factors that stimulate the growth of other harmless microorganisms. In 1989, Roy Fuller suggested a definition of probiotics that has been widely used: "A live microbial feed supplement which beneficially affects the host animal by improving its intestinal microbial balance". Fuller's definition emphasizes the requirement of viability for probiotics and introduces the aspect of a beneficial effect on the host(Fuller, 1989).

According to the FAO and WHO guidelines (2001-2002), probiotics used in food must be capable of surviving passage through the gut (they must have the ability to resist gastric juices and exposure to bile and able to proliferate and colonize GIT. In addition, they must be safe and effective, and maintaining their effectiveness and potency for the duration of the shelf-life of the product (Senok*et al.,* 2005).

The spiraling costs of antibiotic therapy, the appearance of multiresistant bacteria and unsatisfactory therapeutic options in recurrent UTI calls for alternative and advanced medical solutions. So far no sufficient means to successfully prevent painful and disabling recurrent UTI has been found. Even though long-term oral antibiotic treatment has been used with some success as a therapeutic option, this is no longer used due to the development of bacterial resistance. One promising alternative is the use probiotics to prevent and treat recurrent complicated and uncomplicated UTI (Borchert*et al.,* 2008).

A study of (Falagas*et al.,* 2006) showed that the use of probiotic is superior to antibiotics for treating recurrent urinary tract infections in women. The intravaginal applications of probiotic consisting of Lactobacillus have proved to be beneficial in limiting the recurrence of UTI by almost fifty percent in UTI susceptible women with minimum side effects.

Recently the use of probiotics in urological diseases such as recurrent UTI or bladder cancer. Probiotics is promising as an alternative or complementary treatment. Probiotics have the potential for a future alternative prevention and treatment strategy in recurrent UTI; especially with emerge of many bacterial resistant strains due to long course of antibiotic and inadequate therapy. They are also potentially prevention for cancer development and progression (Amdekar *et al.,* 2012).

It appears that a given strain of *Lactobacillus* can express several of the known keyfactors that make it able to compete in the urogenital microenvironment. For example, *lactobacilli* can use many mechanisms toadhere to surfaces, such as electrostatic, hydrophobic, hydrophilic,capsular, and fimbrial mechanisms in the urogenital tract, hydrophilic *L. rhamnosus* and hydrophobic *Lactobacillus fermentum* both colonize. Some strains can bind better to uroepithelial cells and inhibit pathogen adhesion (Redondo *et al.,* 2009).

Another characteristic of *lactobacilli* was studied that appeared to be important in conferring probiotic action against uropathogens. Fifteen strains were found to produce biosurfactant. The substance adsorbed to surfaces and inhibited the initial adhesion of *E.coli* by 70%, also the activity was shown to affect a broad range of pathogens the crude substance was analyzed and was found to contain proteins and carbohydrates. The biosurfactant activity is resistant to trypsin and pepsin, and sensitive to amylase and lysozyme, and resistant to heating at 75 $C^{o}$(Velraeds*et al.,* 2011).

The antiadhesive molecules produced by certain lactobacilli hold promise for application to many human sites where pathogens attach, colonize, and confer disease (Heinemann *et al.,* 2012).

So the recurrence rate of UTI can be significantly reduced using one or two capsules vaginally per week for one year, with no side effects or yeast infections. The rate of infection was the same as those found in studies using daily antibiotics for one year. (Andreu, 2011).

The appearance of multiresistant bacteria in recurrent UTI calls for alternative and advanced medical solutions, one promising alternative is the use probiotics to prevent and treat recurrent complicated and uncomplicated UTI (Reid et al., 2003).

*Lactobacillus is* a major part of the lactic acid bacteria group, which inhibits the growth of some harmful bacteria as Uropathogenic *Pseudomonas aeruginosa* So it is effective as probiotic in prevention and treatment of recurrent UTIs in women (Stephanie, 2010).

**The aim of this work**

1. Identify the most common UTI causing microorganisms among female patients in Beni Suef university hospital.
2. Study the antimicrobial sensitivity of the isolated microorganisms and determine the most effective antimicrobial agents.

3. Evaluate the role of Lactobacillus, stander species, (as a probiotic agent) in controlling of Pseudomonas aeruginosa under laboratory conditions.
4. Genetic study of Pseudomonas antimicrobial resistance

## PATIENTS, MATERIAL AND METHOD

The study was conducted during the period from January / 2014 to May/ 2015 on 100 patients, from the urology outpatient clinics in Beni-Suef university hospital, faculty of Medicine, Beni-Suef University, they were suffering from recurrent urinary tract infection.
A Midstream urine samples (100 samples) following infection control procedures were collected after taking the patient's consent.
All patients of are females with the same age range (from 19-45) years old, All participants were subjected to:

-Detailed history: the duration of recurrent UTI, operations, instrumentation, history of diabetes mellitus, if under any treatment, especially antibiotics, its duration, dose and type. Participants who were on antibiotics were asked to come one week after the last dose.
-Mid stream voided urine samples were collected during active phase of infection under aseptic conditions in sterile containers then transported to the bacteriological laboratory within half an hour.
-Urine samples were studied bacteriologically as follows:

1. Quantitative viable bacterial counts
2. Culture for isolation and identification of different organisms

The virulence factors of urinary *Pseudomonas aeruginosa* studied was Hemolysin production.
- 15strains of *Pseudomonas* isolated in pure culture from both upper and lower were studied for Hemolysin production.
-The inhibitory effect of the supernatants of lactobacillus isolates on Uropathogenic *Pseudomonas aeruginosa* will be tested in parallel with a standard strains (*Lactobacillus acidophilus ATCC L 1259, Lactobacillus acidophilus CCUG5917 , Lactobacillus delbrueckii subspecies ATCC 7830, and Lactobacillus fermentum ATCC 20049)* obtained from the ATCC Culture Collection by the following tests:
-The inhibitory effects of lactic acid produced by lactobacilli isolates and standard strain on the growth of uropathogenic *Pseudomonas aeruginosa* by the plate-diffusion technique.
-Quantitative assay of lactic acid produced by both isolated lactobacilli strains and the standard strain using a lactic acid dehydrogenase (LDH)commercial test kit.
-The minimum inhibitory concentration (MIC) of lactic acid on *Pseudomonas aeruginosa* growth was determined.
-The bactericidal effect of other inhibitory factors was studied after neutralization of lactic acid by sodium carbonate using plate diffusion technique.

### 3.1 GENETIC STUDY OF ANTIMICROBIAL RESISTANCE OF PSEUDOMONAS

Chromosomal DNA was extracted from each P. aeruginosa isolate by DNA extraction kit (DNA extraction kit (from cell 50 test spin column bioflux): Fermentas, UK) according to manufacturer's instruction. The bacteria were confirmed using the PCR method for nan1 gene of the P. aeruginosa (Strateva T. Microbiological and molecular-genetic investigations on the resistance mechanisms and virulence factors in clinical strains of Pseudomonas aeruginosa,2008).PCR was carried out with 2 µL template DNA, 0.25 µM of each primer (F: 5'-ATGAATACTTATTTTGATAT and R: CTAAATCCATGCTCTGACCC-3'), 0.2 mM deoxyribonucleoside triphosphates, 1X reaction buffer, 2 mM MgCl2 and 1.5 U Taq DNA polymerase (Fermentas) in a total volume of 25 µL. The DNA was amplified using the following protocol: initial denaturation (94 ºC for 5 min), followed by 25 cycles of denaturation (94°C for 35 s), annealing (53°C for 45 s) and extension (72°C for 1 min), with a single final extension of 7 min at 72°C.

## RESULTS

### Investigated patients3.1.

One hindered female patients suffering from recurrent UTIs were enrolled in the present study. Concerning ages, the age were ranges from 19-45 years old with the following distribution: 33 patients were from 19-30 years, 10 from 31-40 years and 57 from 41-49 years, with the mean age 42 years with standard deviation (SD) of ± 4.532 Table (1).

**Table (1):** Age distribution among patients from the studied group.

| The age group | Number | Percentage |
|---|---|---|
| 19 -30 years old | 33 | 33% |
| 31- 40 years old | 10 | 10% |
| 41 – 49 years old | 57 | 57% |
| **Total number** | 100 | %100 |

### 3.2 .RESULTS OF GROUP I (UTI)

Regarding cases of the recurrent urinary tract infections; it was found that 74 /100 (74%) of UTI patients were diagnosed after operation, cesarean section or diagnostic urological methods, while the remaining 26/100 (26%) of UTI patients had no history of hospital admission or any surgical manipulation.

Moreover, 45/100 (45%) were diagnosed as having upper urinary tract infection (pyelonephritis) and 55/100 (55%) were suffering from lower urinary tract infection (cystitis).

The results of quantitative culture of the studied urine samples showed the presence of significant bacteriuria in most case (81%)

### 3.3.ISOLATED ORGANISMS:

Pseudomonas was the most 3rd predominant isolated organism followed by E.coli (26) ,as pure isolate in upper UTIs (23%) , while staphylococcus saprophyticus was the most predominant isolate in pure culture in lower UTIs (28%).
Fifteen strains of *Pseudomonas* isolated in pure culture from both upper and lower were studied for Hemolysin production. Table (2) shows that 11 out of 17 studied strains of urinary *Pseudomonas* (100%) produced β hemolysis on blood agar.

**Table (2):** Virulence factors of urinary Pseudomonas isolated strains.

| Isolated strain | Number of test isolates | Haemolytic activity | % |
|---|---|---|---|
| Pseudomonas | 15 | 15 | 100% |

**Table (3):** The distribution of isolated organisms.

| Isolated organisms | The total No. of (isolated in U-UTI 55) | | | | The total No. of (isolated in L-UTI 45) | | | | Total No.of isolates (U-UTI & L-UTI) (100) | |
|---|---|---|---|---|---|---|---|---|---|---|
| | Pure culture | | Mixed culture | | Pure culture | | Mixed culture | | | |
| | No. | % | No. | % | No. | % | No. | % | No. | % |
| *Pseudomonas aeruginosa* | 8 | 8% | 3 | 3% | 5 | 5% | 2 | 2% | 18 | 18% |
| *E.coli* | 7 | 7% | 10 | 10% | 3 | 3% | 5 | 5% | 25 | 25% |
| *K. pneumoniae* | 3 | 3% | 5 | 5% | 2 | 2% | 2 | 2% | 12 | 12% |
| *Proteus mirabilis* | 2 | 2% | 0 | 0% | 3 | 3% | 0 | 0% | 5 | 5% |
| *Staphylococcus saprophyticus* | 2 | 2% | 13 | 13% | 9 | 9% | 4 | 4% | 28 | 28% |
| *Staphylococcus aureus* | 3 | 3% | 0 | 0% | 0 | 0% | 0 | 0% | 3 | 3% |
| *Enterococci* | 0 | 0% | 3 | 3% | 4 | 4% | 3 | 3% | 10 | 10% |

L: lower UTI.
U:upper UTI.

Table (3) show that *Pseudomonas aeruginosa* was the most 3rd organism isolated form upper UTI sample (18%), followed by *Staphylococcus saprophyticus, E.coli, Klebsiella, Enterococcus, Proteus mirabilis* (28% - 26%-12% - 10% - 5 % - 3%) respectively. The relative frequency of isolate in lower UTI, *staphylococcus Saprophyticus* was the most common organism isolated from lower UTI samples (28%),followed by *E .coli, Klebsiella, Enterococcus, Proteus mirabilis* ( 25% - 12% - 10 % - 5%)respectively.

Activation of standard *Lactobacillus acidophilus* ATTC 1259,*Lactobacillus acidophilus* CCUG5917 subspecies, *Lactobacillus delbrueckii* subspecies ATCC 7830, and *Lactobacillus fermentum* ATCC 20049, by cultured on MRS medium for 48 hrs. at 37 c.

**REGARDING THE AGAR WELL DIFFUSION TECHNIQUE**

Shows the inhibitory effect *Lactobacillus* Standard strains species on the 15 uropathogenic *Pseudomonas aeruginosa* isolates. All strains of *Pseudomonas aeruginosa* were inhibited by supernatant containing lactic acid of the tested *Lactobacillus* strains(*Lactobacillus acidophilus ATCC L 1259 and Lactobacillus acidophilus CCUG5917*) . The growth up to the well (no inhibition) the zone is recorded 6 mm.

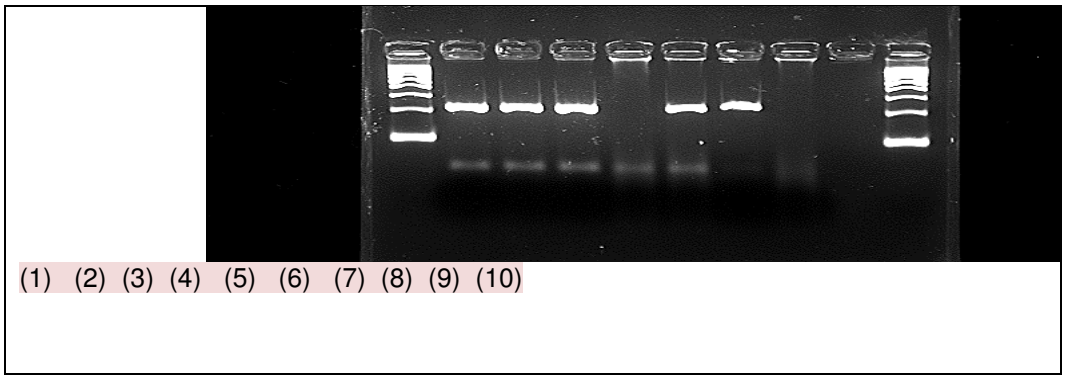
*Lactobacillus acidophilus* ATCC L1259 and *Lactobacillus acidophilus* CCUG5917 (standard strain) had the mean diameter of inhibitory zone (19.5mm and 18. 4 respectively), that mean the ability tested of *Lactobacillus acidophilus* ATCC L 1259 and Lactobacillus acidophilus CCUG5917, to inhibit growth of all uropathogenic *Pseudomonas aeruginosa* strains.

Otherwise *Lactobacillus delbrueckii* subspecies ATCC 7830 and *Lactobacillus fermentum* ATCC 20049, hadn't ability to inhibit Uropathogenic *Pseudomonas aeruginosa* by their lactic acid supernatant as inhibit zone < 6mm

**Analysis of production of O*pr f* gene in *Pseudomonas* strains**

The 15 *Pseudomonas aeruginosa* isolates were subjected to PCR for assessment of *Opr f* gene production, 13 (87%) of the isolates proved to be *Opr f* gene producers. The correlation between the presence of the *Opr f* gene and resistance to penicillin in *Pseudomonas aeruginosa* was statistically significant

Figure. (9): PCR detection of *Opr f* gene. Isolates in lanes 5 and 6were positive, showing bands at 214 kbp. Lane 1 and 10 shows the DNA marker. Lane 2 is positive control. Lane8is the negative control.




**RESULTS OF THE STUDY SHOWED THAT**

1-Significant bacteriuria was found in all urine samples of UTI patients (100%).
2-*Pseudomonas aeruginosa* was the 3rd commonest organisms isolated from the urine samples of UTI patients (49%).
3-As regard hemolysin production, 11 out of 17 strains of urinary *Pseudomonas aeruginosa* (64.7%) produce complete hemolysis on blood agar.
4-Regarding the concentration of lactic acid that was secreted by the standard strains*(Lactobacillus acidophilus ATCC L 1295*) that was 5.64 mg/ml.
5-The minimal concentration of lactic acid that inhibits the growth of Uropathogenic *Pseudomonas aeruginosa* was 2.82 mg/ml.
6-Regarding the result of agar well diffusion technique:
The mean diameter of the inhibitory zone diameter of standard *lactobacillus* strains was +- 19.5 mm.
7-Genetic study of Pseudomonas antimicrobial resistance showed the presence of Opr F gene in resistance to penicillin .

**DISCUSSION**

In the present study, the hemolysin positive pseudomonas was 15 of 15pure isolates (100%) and these isolates were from patients with pyelonephritis and ureteric complications. This result came congruent with that of (Stark, et al., 2003). They found that 95/95 pseudomonas strains that were isolated from a wide range of urine specimens in four hospitals were positive for hemolysin production when grown on sheep blood agar.

Several different epidemiological studies indicate that *Pseudomonas aeruginosa* is a nosocomial pathogen – nosocomial

infections are infections resulting from treatment in a hospital or a healthcare service unit; infections are considered nosocomial if they first appear within 48 hours or more after hospital admission or within 30 days after discharge. There is also evidence suggesting that antibiotic resistance is increasing in recent years in Pseudomonas aeruginosa. According to the Center for Disease Control and Prevention (CDC), the overall incidence of Pseudomonas aeruginosa infections in US hospitals averages about 0.4 percent (4 per 1000 discharges), and the bacterium is the fourth most commonly-isolated nosocomial pathogen accounting for 10 percent of all hospital-acquired infectionsPseudomonas aeruginosa causes urinary tract infections, respiratory system infections, dermatitis, soft tissue infections, bacteremia, bone and joint infections, gastrointestinal infections and a variety of systemic infections, particularly in patients with severe burns, in cancer and in AIDS patients who are immuno-suppressed.

*Pseudomonas aeruginosa* infection represents a serious problem in patients hospitalized with cancer, cystic fibrosis, and burns. The case fatality rate in these patients is almost 50 percent.

*Pseudomonas aeruginosa* like other Gram-negative bacteria is difficult to treat with existing antibiotics, but may in addition develop resistance after unsuccessful treatment. Thus, Pseudomonas aeruginosa infections are an increasing threat to the community.

Our study showed that the common age of women with UTI was from 41-49 years old (57%) with a mean age of 42 ± 4.532. Age distribution in our study was consistent with what was reported by Lane and Takha (2011), where 55- 65% of women within the age of 40 - 50 years have had at least one UTI. However, UTIs can affect anyone at any time of life.

*Pseudomonas aeruginosa* causes urinary tract infections, respiratory system infections, dermatitis, soft tissue infections, bacteremia, bone and joint infections, gastrointestinal infections and a variety of systemic infections, particularly in patients with severe burns, in cancer and in AIDS patients who are immuno-suppressed.

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*Pseudomonas aeruginosa* like other Gram-negative bacteria is difficult to treat with existing antibiotics, but may in addition develop resistance after unsuccessful treatment. Thus, Pseudomonas aeruginosa infections are an increasing threat to the community.

Concerning the order of the isolated strains from urine samples in the current study, *Pseudomonas* represented 35% of the isolates followed by *Enterobacteriaceae* (*Proteus mirabilis and Klebsiella species).* This distribution paralleled the finding of Rituparna, (2010), who reported that E.coli was the predominant organism isolated, followed by other Pseudomonas *Proteus mirabilis and Klebsiella* species were identified most frequently.

*Pseudomonas aeruginosa*a particularly dangerous pathogen due to the fact that few antibiotics are effective (a few effective ones are fluoroquinolones, amikacin and gentamicin, and certain broad-spectrum Beta-lactam antibiotics like imipenim). Resistance is due to outer membrane permeablility or mulitdrug efflux pumps, and the ability of it to create new resistant strains on exposure to antibiotics. (Abigail, et al 1994).

Primary selection of potentially effective probiotic strains must be performed through the application of 'in vitro' techniques. Production of antagonistic substances (organic acids, hydrogen peroxide or bacteriocins) against pathogens is a technique that is widely used (Strus et al., 2002).

There are new strategies in prevention and treatment of urinary tract infection, these strategies include the use of probiotics.. Most of probiotic bacteria has the ability to digest lactose and converting it into lactic acid, so lowering the microenvironmental pH. In this group, *Lactobacillus, Enterococcus, Streptococcus, Pediococcus, and Bifidobacteria* are included (Mombelli and Gismondo. 2000).

A group of investigators reported that *Lactobacillus strains* inhibit the growth of Gram-negative pathogenic bacteria. This growth-inhibiting activity has generally been attributed to the fact that Lactobacillus species lower the pH and/or produce lactic acid. For example, strains of *L. acidophilus, L. casei subsp. rhamnosus, and Lactobacillus bulgaricus* inhibited the growth of clinical isolates of uropathogenic pseudomonas, E. coli, and Klebsiella pneumoniae (Forestier, et al. 2001).

In an experimental study, the in vitro testing provided an approach for determination of the ability of lactobacilli to inhibit the growth of some pathogens. L. fermentum completely inhibited pseudomonas colonization of the urinary tract of mice (Nader et al., 1996).

In the present study the amounts of D- lactic acid produced from the standard strain after overnight incubation on MRS broth were analysed enzymically by using a lactic acid dehydrogenase (LDH) commercial test kit. The result showed that the concentration of lactic acid was secreted by the standard strain (*lactobacillus acidophilus ATCC L 1259*) (5.64 mg/ml).

As regarding the MIC of lactobacilli supernatants lactic acid on Uropathogenic *pseudomonas*, showed that the minimal concentration of lactic acid that inhibits Uropathogenic *pseudomonas* growth was 2.82 mg/ml.

In a study conducted by Domitille, et al., (2005), they used a commercial kit to determine the concentration of lactic acid in *lactobacilli* culture supernatants. The inhibitory effect of increasing concentrations of lactic acid (concentrations ranging from 4-5.5 mg/ml) was a dose-dependent killing activity.

Kiňová, et al., (2001) studied the antagonistic activity of *L. acidophilus* against several potential pathogens. They reported that the highest inhibitory activity was detected against *pseudomonas*, with inhibition zone of (20.75 ± 4.90 mm

SD). This result is compatible with the present study as the mean diameter of the inhibition zone of L. acidophilus ATCC L1259 standard strain was (19.5 ± 5.18 mm SD).

A study conducted by Gregor Reid. (2011) showed that the proof of urogenital colonization and protection from infection was obtained from a clinical trial in which 55 postmenopausal women were given one suppository of lactobacilli probiotic weekly for 1 year. The patients were followed up after 2 weeks and then monthly. Their data were included to examine infection rates, and the side effects. The UTI infection rate decreased from 6.0 per previous year to 1.6 (73% decrease) for those given lactobacilli. The viable lactobacillus counts recovered from vaginal swabs increased with therapy, especially for months 7–12 for lactobacilli-treated patients, during which time lower UTI rates were seen.

This clinical trial confirms the beneficial of using *lactobacillus strains* in treatment and prevention of UTI infection.

Based on the findings in this study, it can be concluded that *Lactobacillus strains*(standard strains), are able to inhibit the growth of uropathogens especially *pseudomonas* by the effect of lactic acid with or without other inhibitory substances.

*OprF*, the major outer-membrane protein in *Pseudomonas* has been studied extensively due to its utility as a vaccine component, its role in antimicrobial drug resistance, and its porin function (De´ et al., 1995; Jaouen et al., 2004; Orange1994; Rawling et al., 1995; Worgall et al., 2005). This protein has only been found in this genus (but see Rediers et al., 2004) and may be considered as a diagnostic protein for Pseudomonas sensustricto (de Mot et al., 1994; Vermeiren et al., 1999; Aagot et al., 2001). *OprF* shares C-terminal similarity with *OmpA of Escherichia coli* and thus is a member of the OmpA superfamily of porins(de Mot et al., 1994).

*Oprf* is a multifunctional protein. It is a nonspecific porin permitting passive diffusion of small polar nutrients (Orange, 1994). The *OprF* pore changes its channel size according to the growth conditions, and this could affect outer-membrane permeability in the three major species of the genus *Pseudomonas: P. fluorescens, P. putida and P. aeruginosa* (De´ et al., 1997; Jaouen et al., 2004). *OprF* is also involved in maintaining cell shape and in growth in a low-osmolarity environment (Woodruff & Hancock, 1989 Rawling et al., 1998). Moreover, this protein probably plays a role in adhesion to roots in *P. fluorescens* strains isolated from the rhizosphere (de Mot et al., 1992) and in adhesion to fibronectin in *P. fluorescens* (Rebie`re-Hue¨t, et al., 1999) For P. aeruginosa, it has recently been reported that the binding of human interferon-c to *OprF* results in the expression of the PA-I lectin, a quorum-sensing-dependent virulence determinant (Wu, et al., 2005).

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## CONCLUSION

It can be confirmed that *Lactobacillus stander strains* are able to inhibit the growth of uropathogens especially *Pseudomonas aeruginosa* by the effect of lactic acid with or without other inhibitory substances, *lactobacillus acidophilus ATCC L1259*and*Lactobacillus acidophilus* CCUG5917(standard strains) had the mean diameter of inhibitory zone (19.5mm and 18.4respectively), that mean the ability tested of lactobacillus stander strains to inhibit growth of all Uropathogenic *pseudomonas aeruginosa.*

Genetic study of Pseudomonas antimicrobial resistance showed the presence of *Opr F gene* in resistance to penicillin.

## REFERENCE

Alls, A. Nathan, W. and Jessica, R.( 2011);P. Blueprints emergency medicine (2nd ed. Ed.). Baltimore, Md.: Lippincott Williams & Wilkins. pp. 152.ISBN 9781405104616.

Amdekar, S. Singh, V. and Singh, D.D.(2012); Probiotic therapy: immunomodulating approach toward urinary tract infection. Current microbial. 63 (5): 484-90.

Andreu, A.(2011); Lactobacillus as a probiotic for preventing urogenital infections in women. Rev Med Microbiol, 15 (1): 1-6.

Azzarone, G. Liewehr, S. and O'Connor K. .(2007) Cystitis. Pediatr Rev.2007 Dec; 28(12): 474-6

Balcht , A. and Smith, R. (1994): Pseudomonas aeruginosa: Infections and Treatment. Informa Health Care.1994; pp. 83–84. ISBN 0-8247-9210-6.

Black, J.G. (1996): Microbiology: Principles and Applications. 3rd Edition.Prentice Hall. New Jersey. 227 – 250.

Bollgren, I. and Winberg, J. .(1976) 'The periurethral aerobic bacterial flora inhealthy boys and girls', ActaPaediatrScand, 1976, 65, (1), pp. 74-80.

Borchert D, Sheridan L, Papatsoris A, Faruquz Z, Barua JM, Junaid I, Pati Y, Chinegwundoh F, Buchholz N.(2008); Prevention and treatment of urinary tract infection with probiotics: Review and research perspective Indian J Urol.2008; 24(2): 139–144

Boyle, RJ. Bath-Hextall, FJ. and Leonardi-Bee, J. .(2004) Probiotics for treating eczema. Cochrane Database Syst Rev,2004; (4): CD006135.

Braat, H. Van Den Brande, J. and VanTol, E. .(2004) Lactobacillus rhamnosus induces peripheral hyporesponsiveness in stimulated CD4+ T cells via modulation of dendritic cell function. The American journal of clinical nutrition,2004; 80 (6): 1618–25.

Brady, L.J. Gallaher, D.D. and Busta, F.F. .(2011) The role of probiotic cultures in the prevention of colon cancer. The J. of Nutrition;2011; 130 (2S Suppl): 410S–414S.

Brade, H. Brade, L. and Rietschel, E.T. .(2010) Structure-activity relationships of bacterial lipopolysaccharides (endotoxins). Zentralbl.Bakteriol.Microbiol. Hyg.2010;268:151–179.

Cardieux, P. Burton. J. and Gardiner, G. (2011): Lactobacillus strains the demographic and microbiologic. JAMA; 287: 1940-1.

Chambers, S.T., and Kunin, C.M.(1987). 'Isolation of glycine betaineandprolinebetaine from human urine. Assessment of their role as osmoprotective agents for bacteria and the kidney', J Clin Invest,1987; 79, (3), pp. 731-737.

Chang YH, Kim JK, Kim HJ, Kim WY, Kim YB, Park YH.(2001); Selection of a potential probiotic *Lactobacillus* strain and subsequent *in vivo* studies. Antonie Van Leeuwenhoek.80:193–199.

Chang, Y. Kim, J. Kim, H .and Park Y. .(2012) Selection of Lactobacillus strain and subsequent in vivo studies, Antonie van Leeuwenhoek,2001; 80: 193 – 199.

Clinical and Laboratory Standards Institute. M100–S22 Performance standards for antimicrobial susceptibility testing 22nd informational supplement.Wayne (PA), 2012;CLSI.

Colgan, R. Nicolle, LE. McGlone, A. and Hooton TM. .(2006.(2007)) Asymptomatic bacteriuria in adults. Am Fam Physician.2006; 15, 74(6):985-90.

Conway, PH. Cnaan, A. Zaoutis, T. and Henry, B.V. .(2007) Recurrent urinary tract infections in children: risk factors and association with prophylactic antimicrobials. JAMA, 2007;11, 298(2):179-86.

De Roos N. M., and Katan M.B. .(2000) Effects of probiotic bacteria on diarrhea, lipid metabolism, and carcinogenesis: a review of papers published between. Am J ClinNutr.2000; 71:405-11.

Dicks, L.M. Silvester, M. and Lawson, P.H.(2010). Lactobacillus fornicalis sp. Nov ., isolated from the posterior fornix of the human vagina. International Journal of Systematic and Evolutionary Microbiology (Society for General Microbiology),2010; 50 (3): 1253–8.

Dielubanza E.J. and Schaeffer A.J. .(2011) Urinary tract infections in women. The Medical clinics of North America,2011; 95 (1): 27-41.

Doron, S. I. Hibberd, P. L. and Gorbach, S.L. .(2008) Probiotics for prevention of antibiotic-associated diarrhea. J ClinGastroenterol,2008; 42 (Suppl 2): S58–63.

Friedman, Y. .(2006) Safe use of genetically modified lactic acid bacteria in food. Bridging the gap between consumers, green groups, and industry". Electronic Journal of Bioethanol,2006; 9 (4): E49–55.

Forestier, C., C. De Champs, C.Vatoux, and B. Joly. .(2001) Probiotic activities of Lactobacillus caseirhamnosus: in vitro adherence to intestinal cells and antimicrobial properties. Res. Microbiol.2001; 152:167-173.

Foster, R.T. .(2008) Uncomplicated urinary tract infections in women. ObstetGynecolClin North Am.2008; 35(2):235-48.

Foxman, B. Barlow, R. and D'Arcy,(2011); H. Urinary tract infection: in a self-reported incidence and associated costs. Ann EpidemiolAntimicrob Agents Chemother; 45 (6): 1751-60.

García JR, Krause A, Schulz S, Rodríguez-Jiménez FJ, Klüver E, Adermann K, Forssmann U, Frimpong-Boateng A, Bals R, Forssmann WG,(2001); Human betadefensin 4: a novel inducible peptide with a specific salt-sensitive spectrum of antimicrobial activity', FASEB J,15, (10), pp. 1819-1821.

Gill, HS. Rutherford, KJ. Cross, ML and Gopal, PK. .(2011) Enhancement of immunity in the elderly by dietary supplementation with the probiotic Bifidobacteriumlactis HN019. Am J ClinNutr.2011; 74(6): 833-839.

Godally, G. Bergsten, G. and Frendéus, B. (2000): Innate defenses and resistance to Gram-negative mucosal infection. Adv. Exp. Med. Biol.; 485: 9–24.

Gorelick, M. and Shaw, K. .(1999) Screening tests for urinary tract infection in children: a meta-analysis Pediatrics,1999, 104(5): 54.

Gould, C.V. Umscheid, C.A. and Agarwal, R.K. .(2010) Guideline for prevention of catheter-associated urinary tract infections. Infect Control HospEpidemiol.2010; 31 (4): 319–26.

G. Reid, A.W.B., and M. Taylor(1995); 'Instillation of Lactobacillus and stimulation of indigenous organisms to prevent recurrence of urinary tract infections', Microecology and Therapy, 23, pp. 32-45.

Guyonnet, D. Chassany, O. and Ducrotte, P. .(2007) Effect of a fermented milk containing Bifidobacteriumanimalis DN-173 010 on the health-related quality of life and symptoms in irritable bowel syndrome in adults in primary care: a multicentre, randomized, double-blind, controlled trial". Aliment.Pharmacol.Ther.2007; 26 (3): 475–86.

Hamilton, M. Professor J M T, Gibson, GR. and Bruck, W. (200)9;Some insights into the derivation and early uses of the word 'probiotic. British J. of Nutrition, (90): 845.

Hamilton-Miller, JM.(2010);The role of probiotics in the treatment and prevention of Helicobacter pylori infection. International Journal of Antimicrobial Agent, s 22 (4): 360–6.

Heinemann, C. van Hylckama, J.E. Janssen, D.B. (2012);Purification and characterization of a surface-binding protein from Lactobacillus fermentum RC-14 that inhibits adhesion of Enterococcus faecalis 1131. FEMS MicrobiolLett. 412:177–180.
Hickson, M. D'Souza, A.L. and Muthu, N.(2007). Use of probiotic Lactobacillus preparation to prevent diarrhea associated with antibiotics: randomised double blind placebo controlled trial,2007; BMJ 335 (7610): 80.
Hickson, M. D'Souza, A.L. and Muthu, N. (2007): Use of probiotic Lactobacillus preparation to prevent diarrhoea associated with antibiotics: randomised double blind placebo controlled trial. BMJ,2007; 335:80.
Hitti, M. (2006). Probiotics May Help Stressed Gut. The American journal of clinical nutrition; 75(8): 1345–65.
Honkinen, O. Jahnukainen, T. Mertsola, J. Eskola, J. and Ruuskanen, O. .(2000) Bacteremic urinary tract infection in children. Pediatr Infect Dis J,2000; 19(7): 630–634.
Hodson EM, Wheeler DM,Wimalchandra D, (2009): Interventions for primary vesicoureteric reflux. Cochrane Database Syst Rev. 18 ;( 3):CD001532.
Hooton, T. M., .(2010) Diagnosis, prevention, and treatment of catheter-associated urinary tract infection in adults: International Clinical Practice Guidelines from the Infectious Diseases Society of America. Clin. Infect. Dis.2010; 50, 625–663.
Huffman, J.W,(1948); .'The detailed anatomy of the para-urethral ducts in the adult human female', Am J ObstetGynecol, 55, (1), pp. 86-101.
Y. K. Lee and S. Salminen. .(2009) 2nd Edition.Ed.Wiley. (2009). Incorporating Probiotics into Food". pg. 60-67. Ross Crittenden, in "Handbook of Probiotics and Prebiotics".
Kasper, DL. Fauci, AS. and Longo, DL. .(2010) Harrison's Principles of Internal Medicine,2010; 16th ed. New York.
Kaye, D. .(1968) 'Antibacterial activity of human urine ', J Clin Invest, 1968, 47, (10), pp. 2374-2390.
Kiessling, G. Schneider, J. and Jahreis, G. .(2012) Long term consumption of fermented dairy products over 6 months increases HDL cholesterol. European Journal of Clinical Nutrition,2012; 56 (9): 843–849.
Kim, H.J. Camilleri, M. and McKinzie, S. .(2003) A randomized controlled trial of a probiotic, VSL#3, on gut transit and symptoms in diarrhoea-predominant irritable bowel syndrome. Aliment PharmacolTher.2003; 17:895-904.
Kingsley, C. Anukam, E.O.O., IjeomaAhonkhai, and Gregor Reid . .(2005) '16S rRNA gene sequence and phylogenetic tree of lactobacillus species from the vagina of healthy Nigerian women', African Journal of Biotechnology,2005, 4, (11), pp. 1222-1227.
Klaenhammer, TR.(2000). Microbiological considerations in selection and preparation of Lactobacillus strains for use as dietary adjuncts. J Dairy Sci,2000; 65:1339–49.
Kunin, C.M., Evans, C., Bartholomew, D., and Bates, D.G. .(2002) 'The antimicrobial defense mechanism of the female urethra: a reassessment',2002 J Urol, 168, (2), pp. 413419.
Kuroda, M. Yamashita, A. and Hirakawa, H. .(2005) Whole genome sequence of Staphylococcus saprophyticus reveals the pathogenesis of uncomplicated urinary tract infection. Proc. Natl. Acad. Sci. U.S.A.2005, 102 (37); 13272–7.
Laleye, L.C. .(1987) Involvement of heterofermentative lactobacilli in development of open texture in cheese, J. Food Prot.1987; 50, 1009.
Land, MH. Rouster, K. and Woods, CR. .(2005) Lactobacillus sepsis associated with probiotic therapy. Pediatrics,2005; 115:178-81.
Lane, D.R. and Takhar, S.S. .(2011); Diagnosis and management of urinary tract infection and pyelonephritis. Emergency medicine clinics of North America, 29 (3): 539-52.
Lauer, E., Helming, C. and Kandler, O. .(1980) 'Heterogeneity of the species Lactobacillus acidophilus (Moro) Hansen and Mocquot as revealed by biochemical characteristics and DNA-DNA hybridization', Zentralbl Bakteriol MikrobiolHyg, 1980, pp. 150-168.
Litwin, MS. And Saigal, CS. (2012): Urologic Diseases in America. US Department of Health and Human Services, Public Health Service, National Institutes of Health, National Institute of Diabetes and Digestive and Kidney Diseases. Washington, DC: US Government Printing Office.2012; NIH Publication No. 07-5512
Ljungh, Å. Wadström, and Torkel. .(2009) Lactobacillus Molecular Biology: From Genomics to Probiotics. Caister Academic Press.ISBN.2009; 978-1-904455-41-7.
Mach, T. .(2006) Clinical usefulness of probiotics in inflammatory bowel diseases. Journal of Physiol and Pharmacol. 2006;57Suppl 9: 23–33. PMID 17242485.
Makarova, K. Slesarev, A. and Wolf , Y. .(2009) Comparative genomics of the lactic acid bacteria. ProcNatlAcadSci U S A.2009; 103 (42): 15611–6.
Marcos, A. Wärnberg, J. and Nova, E. .(2008) Probiotics may protect against food poisoning". European Journal of Nutrition.2008; 43 (6): 381–389.
Marteau, P. Lemann, M. and Seksik, P. .(2006) Ineffectiveness of Lactobacillus johnsonii LA1 for prophylaxis of postoperative recurrence in Crohn's disease: a randomised, double blind, placebo controlled GETAID trial. Gut.2006;55:842-7.1–6.
Mcfarland, LV. (2006) Meta-analysis of probiotics for the prevention of antibiotic associated diarrhea and the treatment of Clostridium difficile disease. Am J Gastroenterol.2006; 101 (4): 812–22.
McGroarty, JA .and Reid, G. .(2005) Coaggregation of urogenital bacteria in vitro and in vivo. CurrMicrobiol.2005; 20:47–52.
Metchnikoff , E. .(1907) Essaisoptimistes. Paris. The prolongation of life. Optimistic studies. Translated and edited by P. Chalmers Mitchell. London: Heinemann.1907..
Modgil, G. and Baverstock A. .(2006) Should bubble baths be avoided in children with urinary tract infections? Arch Dis Child.2006; 91(10):863-5.
Mombelli, B. and Gismondo, MR.(2000); The use of probiotics in medical practice. Int J Antimicrob Agents. 16:531–536.
Moore, K.N. Fader, M. and Getliffe, K. .(2007) Long-term bladder management by intermittent catheterisation in adults and children. Cochrane Database Syst Rev.2007; 17,(4): CD006008.
Moreno, M.R.F. Leisner, J.J. and Tee, L.K. .(2007) Microbial analysis of Malaysian tempeh, and characterization of two bacteriocins produced by isolates of Enterococcus faecium". Journal of Applied Microbiol.2007; 92 (1): 147–157.
Mori, R. Lakhanpaul, M. and Verrier-Jones, K. .(2009) Diagnosis and management of urinary tract infection in children: summary of NICE guidance. BMJ.2009; 25,335(7616):395-7.
Morrison, G., Kilanowski, F., Davidson, D., and Dorin, J. .(2002) 'Characterization of the mouse beta defensin 1, Defb1, mutant

mouse model', Infect Immun, 2002, 70, (6), pp. 30533060.
Morrow, LE. Kollef, MH. And Casale, TB. .(2010) Probiotic prophylaxis of ventilator-associated pneumonia: a blinded, randomized, controlled trial. Am J RespirCrit Care Med.2010; 182:1058-64.
Norrby, S.R. .(2007) Approach to the patient with urinary tract infection. In: Goldman L, Ausiello D, eds. Cecil Medicine. 23rd ed. Philadelphia, Pa: Saunders Elsevier.2007; chap.306.
Nicolle, L.E. .(2008) Uncomplicated urinary tract infection in adults including uncomplicated pyelonephritis. UrolClin North Am.2008; 35 (1): 1–12
Niedzielin, K. Kordecki, H. and Birkenfeld, B. .(2012) A controlled, double-blind, randomized study on the efficacy of Lactobacillus plantarum 299V in patients with irritable bowel syndrome Pediatrics.2012; 97:1.
Ofek, I. and Doyle, R.J. .(2004) Bacterial adhesion to cells and tissues. New York, London: Chapman and Hall.2004; p. 513–561.
Oksanen, P.J. Salminen, S. and Saxelin, M. (2006) Prevention of travellers' diarrhoea by Lactobacillus GG. Ann Med.2006; 22:53-6.
Oelschlaeger, T.A. Dobrindt. U. and Hacker. J. .(2010) Virulence factors of uropathogens. Curr.Opin. Urol.2010; 12:33–38.
Osterlund, P. Ruotsalainen, T. and Korpela, R. .(2007) Lactobacillus supplementation for diarrhoea related to chemotherapy of colorectal cancer: a randomised study. Br J Cancer.2007; 97:1028.
Ouwehand, A.C. Salminen, S.and Isolauri, E. .(2010) Probiotics: an overview of beneficial effects. Antonie Van Leeuwenhoek.2010; 82 (1–4): 279–89
Islam, M.A. Yun. C.H. Choi, Y.J. and Cho, C.S. .(2010) Microencapsulation of live probiotic bacteria. Journal of Microbiol and Biotechnol.2010; 20 (1367–1377): 1367–1377.
Jack, R. W. Bru de Labanda, E. and de Ruiz HolgadoTagg, J. R. .(2005) Bacteriocins of Gram-positive bacteria. Bacterial.2005; Rev 69, 171–200.
Jepson RG, and Craig JC. .(2008) Cranberries for preventing urinary tract infections. Cochrane Database Syst Rev.2008; 23, (1):CD001321.
Pohl, A.(2007) Modes of administration of antibiotics for symptomatic severe urinary tract infections. Cochrane Database Syst Rev;2007. 17 :( 4):CD003237.
Prakash, S. .(2011) The Gut Microbiota and Human Health with an Emphasis on the Use of Microencapsulated Bacterial Cells.Journal of Biomedicine and Biotechnology;2011..
Reid, G. Bruce, A.W. and Taylor, M. .(2001) ; Instillation of Lactobacillus and stimulation of indigenous organisms to prevent recurrence of urinary tract infections. Microecol Ther; 23:32–45.