

## Anti Microbial Screening of A Siddha Formulation Nandukkal Parpam

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### ABSTRACT

Nandukkal parpam is a siddha formulation indicated mainly for kidney diseases especially kidney stone. This drug was tested for anti microbial property against *Proteus Vulgaris*, *E-cloi*, *Pseudomonas aeruginosa*, *Klebsiella pneumoniae* and *Staphylococcus aureus* by Agar-disk diffusion method and the results obtained from the study showed that NKP has anti- microbial property against *Proteus Vulgaris*, *E-cloi*, *Pseudomonas aeruginosa* and the drug is not effective against *Klebsiella pneumoniae* and *Staphylococcus aureus*.

**Key words:** anti microbial, siddha formulation, nandukkal parpam

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### I. INTRODUCTION

Anti microbial studies are used for drug discovery, epidemiology and prediction of therapeutic outcome. Natural products are still one of the major sources of new drug molecules today. They are delivered from prokaryotic bacteria, eukaryotic micro organisms, plants and various animal organisms. Currently, microbial infections have become an important clinical threat, with significant associated morbidity and mortality which is mainly due to the development of microbial resistance to the existing anti microbial agents. Therefore, methods for anti microbial susceptibility testing and discovering novel anti microbial agents have been extensively used and continue to be developed. Nandukkal parpam is a siddha formulation indicated mainly for kidney diseases especially kidney stone. This drug was tested for anti microbial property against *Proteus Vulgaris*, *E-cloi*, *Pseudomonas aeruginosa*, *Klebsiella pneumoniae* and *Staphylococcus aureus* by Agar-disk diffusion method and the results obtained from the study showed that NKP has anti- microbial property against *Proteus Vulgaris*, *E-cloi*, *Pseudomonas aeruginosa* and the drug is not effective against *Klebsiella pneumoniae* and *Staphylococcus aureus*.

### II. METERIALS AND METHOD

#### Test drug:

The drug Nandukkal parpam (AYUSH approved formulary medication) was selected from **Siddha vaithiya thirattu** which is an approved text to take this medicine for research. The test sample

Nandukkal parpam was purchased from IMCOPS Chennai. The major ingredient of this formulation is Fossil stone crab. This Fossil stone crab is present in nature in sea shores which has got diuretic property. It is used in the treatment of urinary calculus, strangury, chronic ascites, leucorrhoea, gonorrhoea, and phlegm. It is one of the very important seashore minerals in Siddha medicine because of its multiple uses. It is known to the Siddhars, thousands of years ago. It is a kind of stone (T.V. Sambasivampillai, 1931) Nandukkal parpam is taken with honey or hot water; it dissolves the urinary stones and relieves the pain due to urinary stones. Hence this drug is mainly recommended for urinary stones, besides this, this drug is given for bleeding piles and for promoting fertility. Intake of this drug stops kidney failures (IMPCOPS, 2005).

#### Microorganisms:

The bacterial strains *Proteus Vulgaris*, *E-cloi*, *Pseudomonas aeruginosa*, *Klebsiella pneumoniae* and *Staphylococcus aureus* were CLSI (formerly NCCLS-National Committee for Clinical Laboratory Standards, 1999) standard reference strains used for antibacterial susceptibility tests.

#### Disc-diffusion method:

Agar-disk diffusion method is the official method used in many clinical microbiology laboratories for routine anti microbial susceptibility testing. In this method, agar plates are inoculated with standardized inoculums of the test micro organism. Then, filter paper discs (about 6mm in

diameter), containing the test compound at a desired concentration, are placed on the agar surface. The petri dishes are incubated under suitable conditions. Generally, anti microbial agent diffuses into the agar and inhibits germination and growth of the test micro organism and then the diameters of inhibition growth zones are measured.

The organisms tested were Proteus Vulgaris, E-cloi, Pseudomonas aeruginosa, Klebsiella pneumoniae and Staphylococcus aureus (Table.1) and the zone of inhibition was recorded by disk diffusion method on Mueller Hinton Agar media.

The antibacterial activities of the sample NKP were carried out by disk diffusion method (Bauer et al., 1966). The concentrations of the test compounds were used at the concentration of 100, 200, 300 µg. The target microorganisms were

cultured in Mueller–Hinton broth (MHB). After 24 h the suspensions were adjusted to standard sub culture dilution. The Petri dishes containing Muller Hinton Agar (MHA) medium were cultured with diluted bacterial strain. Disc made of Whatman No.1, diameter 6 mm was pre-sterilized and was maintained in aseptic chamber. Each concentration was injected to the sterile disc papers. Then the prepared discs were placed on the culture medium. Standard drug Ciprofloxacin (5µg) was used as a positive reference standard to determine the sensitivity of each microbial species tested. Then the inoculated plates were incubated at 37° C for 24 hours. The diameter of the clear zone around the disc was measured and expressed in millimetres as its anti-microbial property. The results were depicted in Table.2

**Table.1** Organisms used for Anti-Bacterial Activity

s.no	organisms	Type
1.	Staphylococcus aureus	Gram-positive
2.	Klebsiella pneumonia	Gram-negative
3.	Pseudomonas aeruginosa	Gram-negative
4.	Proteus Vulgaris	Gram-negative
5.	E. Coli	Gram-negative

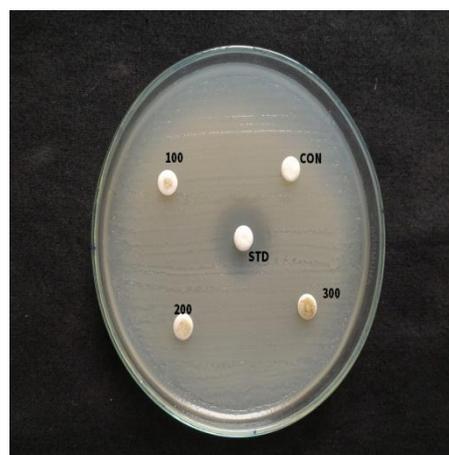
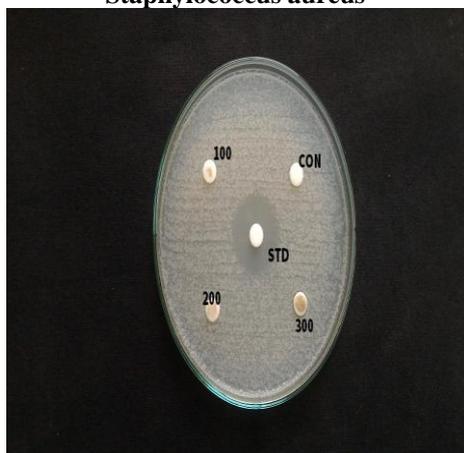
**Table.2** Zone of Inhibition data of Anti-bacterial activity

Sample Code	Klebsiella pneumoniae			Pseudomonas aeruginosa			Staphylococcus aureus			E. Coli			Proteus Vulgaris		
	100 µg	200 µg	300 µg	100 µg	200 µg	300 µg	100 µg	200 µg	300 µg	100 µg	200 µg	300 µg	100 µg	200 µg	300 µg
NKP	-	-	-	-	5	7	-	-	-	1	3	8	-	-	3
Ciprofloxacin (5µg)	19			24			24			24			15		

- = Not active

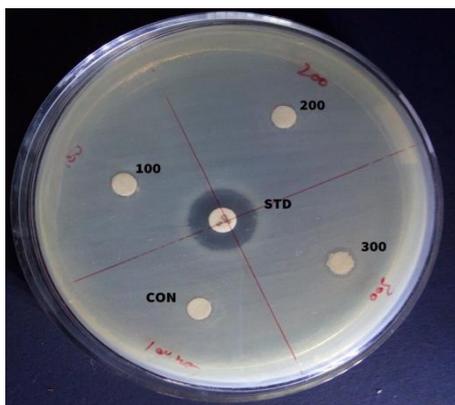
**Anti-Bacterial Evaluation of AO**

**Anti- Microbial Effect of NKP against Staphylococcus aureus**



**Anti- Microbial Effect of NKP against Proteus Vulgaris**

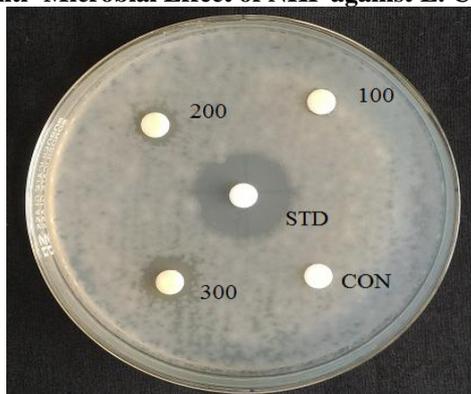
**Anti- Microbial Effect of NKP against Klebsiella pneumonia**



**Anti- Microbial Effect of NKP against Pseudomonas aeruginosa**



**Anti- Microbial Effect of NKP against E. Coli**



### III. DISCUSSION

#### **K.pneumoniae:**

Klebsiella pneumoniae forms urease. It is the 2<sup>nd</sup> most popular member of the aerobic bacterial flora of the human intestine. Klebsiella colonies are large and very mucoid and tend to coalesce with prolonged incubation. K.pneumoniae is present in the respiratory tract and feces of about 5% of normal individuals. K.pneumoniae can produce extensive hemorrhagic necrotizing consolidation of the lung. It

produces urinary tract infection and bacteremia with focal lesions in debilitated patients. Klebsiella sp. Rank among the top 10 bacterial pathogens responsible for hospital- acquired infections. It has become a very important cause of nosocomial infections. It causes pneumonia, urinary infections, other pyogenic infections, septicemia and rarely diarrhoea. K.pneumoniae is a frequent cause of urinary infection. It also causes pyogenic infections such as abscesses, meningitis and septicemia.

#### **Escherichia coli:**

Escherichia coli are part of the normal flora and incidentally cause disease. E.coli and other coliforms account for the large majority of naturally acquired urinary tract infections. Infection may be precipitated by urinary obstruction due to prostatic enlargement, calculi or pregnancy. Four main types of clinical syndrome are caused by E.coli are 1. Urinary infection 2. Diarrhoea 3. Pyogenic infections and 4. Septecemia. The entero toxins produced by E.coli are important in the pathogenesis of diarrhoea. E.coli forms the most common cause of intra-abdominal infections, such as peritonitis and abscesses resulting from spillage of bowel contents. They also cause pyogenic infections in the peri anal area. They are an important cause of neonatal meningitis.

#### **Proteus vulgaris:**

The predominant aerobic bacterial flora present in the large intestine of human beings and animals. Proteus species produce infections in humans only when the bacteria leave the intestinal tract. They are found in urinary tract infections and produce bacteremia, pneumonia and focal lesions in debilitated patients/those receiving intravenous infusions. Proteus mirabilis causes urinary tract infections and occasionally other infections. Proteus species produce urease resulting in rapid hydrolysis of urea with liberation of ammonia. Thus, in urinary tract infections with proteus, the urine becomes alkaline, promoting stone formation and making acidification virtually impossible. The rapid motility of proteus may contribute to its invasion of the urinary tract. Strains of proteus vary greatly in antibiotics for other members of the group are aminoglycosides and cephalosporins.

#### **Pseudomonas aeruginosa:**

Pseudomonas is a group of bacteria that can cause various types of infections. Pseudomonas aeruginosa is the most common disease-causing species, according to the Centers for Disease Control and Prevention (CDC). Pseudomonas can infect any part of the body including the liver, brain, lungs, bones, sinuses and urinary tract.

**Staphylococci:**

Staphylococci produce 2 types of diseases – infections & intoxications. Staphylococci infections are among the most common of bacterial infections and range from the trivial to the fatal.

**Skin & soft tissue:**

Folliculitis, Furuncle (boil), abscess (particularly breast abscess), wound infection, carbuncle, impetigo, paronychia and less often cellulitis.

**Musculoskeletal:**

Osteomyelitis, arthritis, bursitis, pyomyositis.

**Respiratory:**

Bronchopneumonia, lung abscess, empyema, rarely pneumonia.

**CNS:**

Abscess, meningitis, Intra cranial thrombophlebitis.

**Endo vascular:**

Bacteremia, septicaemia, pyemia, endocarditis.

**Urinary:**

Staphylococci are un common in routine urinary tract infections, though they do cause infection in association with local instrumentation, implants or diabetes. Urinary isolates of Staphylococci are to be considered significant even with low colony counts, as they may be related to bacteremia.

**IV. CONCLUSION**

Ciprofloxacin 5 µg was used as the standard anti bacterial agent. From the results of the present study it was concluded that the sample NKP shown anti- microbial property against Proteus Vulgaris, E-cloi, Pseudomonas aeruginosa and the drug is not effective against Klebsiella pneumoniae and Staphylococcus aureus. Nandukkal parpam may have

effective against urinary tract pathogens like E-cloi, Proteus Vulgaris, Pseudomonas aeruginosa ect.

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