

## BACTERIAL ISOLATES FROM EXPRESSED PROSTATIC SECRETIONS IN CHRONIC PROSTATE INFLAMMATION

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**Introduction:** The prostate is an exocrine turbulo-alveolar gland in males. With prostatic inflammation primarily caused by bacterial pathogens, the most significant prostatitis causes involve Gram-positive cocci and Gram-negative rods penetrating the prostatic tissue typically when local immune defences around the prostate are compromised. Clinical symptoms of prostatic inflammation include perineal pain, urethral discharges, dysuria, and recurrent urinary tract infections. Common causative organisms include *Escherichia coli*, *Enterobacter cloacae*, *Pseudomonas aeruginosa*, *Proteus vulgaris*, *Pathogenic Streptococcus pyogenes*, *Staphylococcus aureus* and *epidermidis*, *Enterococcus faecalis* and *faecium* among others. Identifying these bacteria requires culturing on both liquid and agaric nutrient media. The following differential media are required: Endo, StaphiloSel, StreptoSel, PseudoSel, EnterococcoSel, and other. Further biochemical tests, using different carbohydrates, amino acids etc., are necessary to specify bacterial species within samples.

**The aim of the research.** To determine the bacterial species in expressed prostatic secretion (EPS) samples collected during an episode of prostatic inflammation.

**Material and Methods.** We examined the prostate fluid from 46 male participants of reproductive age (18-45 years old) who agreed to take part in the study. We focus on identifying bacterial species and evaluating their quantitative and qualitative presence in expressed prostatic secretion (EPS).

Bacteria were cultured using broth media: (StreptoSel Broth, Lactose-peptone Broth) and semi-solid media (Thioglycolic semi-solid Agar, Trichosel semi-solid Agar), and dense media: (Endo Agar, EnterococcoSel Agar, PseudoSel Agar, Blood Agar, StaphylococcoSel Agar, Wilson-Blair Agar, among others).

All microbiological Agar and Broth were sourced from Becton Dickinson Company, Maryland, USA.

We used carbodic Fuchsin 1%, Methylene blue 1%, Gentian-violet 1% (crystal violet), Safranin 1% solution for bacterial staining.

For microscopy, of samples were Gram-stained with the dye Gentian-violet and Fuchsin for microbiological purposes. The samples were observed under a Microscope with an Olympus digital microphotography camera.

**Results and Discussion.** Most bacterial isolates were from the Enterobacteriaceae family: *Escherichia coli*, *Enterobacter cloacae*, *Klebsiella pneumoniae*, *Proteus vulgaris*, *Proteus mirabilis*, *Proteus morgani*, *Serratia marcescens*. Other specific infective group of bacteria isolated included: *Enterococcus faecalis*, *Enterococcus faecium*, *Pseudomonas aeruginosa*, *Staphylococcus aureus*, *Staphylococcus epidermidis*, *Streptococcus pyogenes*, *Chlamydia trachomatis*, *Candida albicans*, *Trichomonas vaginalis*, *Gardnerella vaginalis*, *Mycoplasma*

*hominis*, *Ureaplasma urealiticum*, Anaerobic bacteria and rarely *Mycobacterium tuberculosis*.

Most chronic prostatitis sufferers were treated with various antibiotic, Therefore, the yeast, *Candida albicans*, was sometimes isolated from expressed prostatic secretion (EPS) samples. In some cases, non-spore-forming, anaerobic, Gram-negative rods from the genera *Bacteroides* and *Fusobacterium*, as well as cocci from the genera *Peptococcus* and *Peptostreptococcus*, and Gram positive non-spore-forming rods, could be isolated. Non-spore-forming anaerobic bacteria are part of the normal microflora of the oral cavity, gastrointestinal tract, and skin, and they can occasionally be isolated from prostatic EPS.

Assessment of prostatic ecology and the relationship between the prostate and its microbial content depends on the local immunity of the sufferer. Therefore, it is crucial to evaluate the morphology of the prostate, including the white blood cell count in EPS, the presence of macrophages indicating inflammation lecithin grains, amyloid bodies, and tubular epithelium. Together, these parameters provide a comprehensive picture of the prostate's condition. Classical microbiological tests on EPS complement morphological assessment, helping to clarify the underlying aethiology of chronic prostatic inflammation. The most common bacterial isolates from EPS are presented in the table.

Table

**Bacteria isolated from EPS in chronic prostatitis**

<b>Enterobacteriaceae family:</b>	
1.	<i>Escherichia coli</i>
2.	<i>Enterobacter cloacae</i>
3.	<i>Klebsiella pneumoniae</i>
4.	<i>Proteus vulgaris</i>
5.	<i>Proteus mirabilis</i>
6.	<i>Serratia marcescens</i>
<b>Other group bacteria and yeast:</b>	
7.	<i>Enterococcus faecalis</i>
8.	<i>Enterococcus faecium</i>
9.	<i>Pseudomonas aeruginosa</i>
10.	<i>Staphylococcus aureus</i>
11.	<i>Staphylococcus epidermidis</i>
12.	<i>Streptococcus pyogenes</i>
13.	<i>Gardnerella vaginalis</i>
14.	<i>Mycoplasma hominis</i>
15.	<i>Ureaplasma urealiticum</i>
16.	<i>Trichomonas vaginalis</i>
17.	<i>Mycobacterium tuberculosis</i>
18.	<i>Chlamydia trachomatis</i>
19.	Non-spore-forming anaerobic rods
20.	<i>Candida albicans</i>

Table shows the bacteria isolated from EPS during chronic prostatitis. *Escherichia coli* is the most leading cause of inflammation in chronic prostatitis accounting for up to 80% of cases, with other bacteria responsible for the remaining

20%. To isolate Enterobacteriaceae family bacteria such as: *Escherichia coli*, *Enterobacter cloacae*, *Klebsiella pneumoniae*, *Proteus vulgaris*, *Proteus mirabilis*, and *Serratia marcescens* we used lactose-peptone Broth with phenol red as an indicator and Endo Agar as a basic Fuchsin

To isolate *Enterococcus faecalis* and *Enterococcus faecium* we used lactose-peptone Broth and Enterococcosel Agar containing the indicator 2,3,5-Triphenyltetrazolium chloride.

To isolate *Staphylococcus aureus* and *Staphylococcus epidermidis* used medium for isolation *Staphylococcus aureus* and *Staphylococcus epidermidis* we used a media containing a hypertonic concentration of sodium chloride (7.5%).

To isolate *Streptococcus pyogenes*, we used Agar with 10% blood serum.

To isolate *Gardnerella vaginalis* we used Blood Agar, where it produces beta hemolysis.

To isolate *Mycoplasma hominis* we used Mycoplasma Broth and Mycoplasma Agar, containing an enzymatic hydrolyzate of bovine heart. Mycoplasma requires a nutrient-rich environment, including 20% horse serum, for optimal growth.

To isolate *Ureaplasma urealiticum* it is necessary to supplement the nutrient media with urea (carbamide) at a concentration of 10.0 mg/L to 50.0 mg/L. Mycoplasma Broth and Mycoplasma Agar can be used for this purpose, provided urea is added to the media.

To isolate *Trichomonas vaginalis* samples must be inoculated TrichoSel Broth and Trichosel semi-solid Agar.

To isolate *Mycobacterium tuberculosis* Lowenstein Jensen medium should be used. It is a selective, egg-based medium designed explicitly for the culture and isolation of Mycobacterium species from clinical specimens.

To isolate *Candida albicans* we used a few media: potato-dextrose Agar, Inhibitory Mold Agar, and Wort Agar.

To isolate non-spore-forming anaerobic rods such as: *Bacteroides*, and *Fusobacterium*, as well as cocci from the genera *Peptococcus* and *Peptostreptococcus* we used Wilson Blair medium (Bismuth sulfite Agar).

To isolate *Chlamydia trachomatis* a bacterium with viral properties, it must be cultured in cell lines sensitive to Chlamydia, such as: NCTC L-929 or McCoy cells. The recommended medium for cultivation is Eagle's MEM supplemented with 10% bovine serum.

**Conclusions.** The microbial content of prostatic EPS was tested using classical microbiological methods in 46 chronic prostatitis sufferers. In most prostatitis cases, bacteria from the *Enterobacteriaceae* family, including *Escherichia coli* were identified as the primary cause of chronic inflammation, accounting for up to 80% of cases. Only others were responsible for the remaining approximately 20% of chronic prostatitis cases.

**Keywords:** prostatic inflammation, bacterial prostatitis, expressed prostatic fluid, EPS.