

## Detection of bacteria responsible for gallbladder inflammation and gallstones

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### ABSTRACT

**Background:** The most prevalent complication of gallstone is chronic cholecystitis. The aim of the present study was to determine the responsible microorganisms in patients undertook cholecystectomy and determine their antibiotic resistance pattern.

**Materials and methods:** Cholecystectomy was achieved in 100 patients. Collected samples transferred to laboratory in appropriate media, then cultured on selective media to isolate the possible causative bacteria. Antibiotic susceptibility tests were also performed by Kirby-Baur method with a colony in Muller-Hinton agar medium.

**Results:** Totally, 63 samples showed bacterial growth, of which 11 revealed two species of bacteria. A total of 10 gram positive (enterococci and staphylococci) and 63 gram negative (E.coli, klebsiella, aerobacter, pseudomonas, proteus, citrobacter, providencia and acinetobacter) bacteria were isolated.

**Conclusion:** Isolating bacteria and determining their susceptibility to different antibiotics may help physicians to manage cholecystitis and its associated complications.

**Keywords:** Cholecystitis, cholecystectomy, Bacterial contamination.

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### INTRODUCTION

Abnormality or inflammatory diseases of gallbladder usually necessitate cholecystectomy (1,2). Obstruction of cystic duct that is mainly associated with gallstones is the main reason of acute cholecystitis. Gallstones are usually chemically classified as cholesterol or calcium bilirubinate stones. The most prevalent complication of gallstone is chronic cholecystitis occurs in approximately 4% of cases. It affects subjects aged 30-40 years and presents with nausea and vomiting, pain, fever and chills.

Prior investigators have proposed the following causative pathogens in cholecystitis: enterococci 23%, pseudomonas 1%, Salmonella typhi 2-5% and other gram negative bacteria such as E.coli 41.1% (1). Salmonella typhi causes typhoid fever and Salmonella paratyphi (A,B,C) is associate with paratyphoid fevers. Humans as the original source of infection, distribute the organism in the society (2).

In the present study we aimed to detect the responsible microorganisms in patients undertook cholecystectomy and determine their antibiotic resistance pattern.

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## PATIENTS and METHODS

Initial data including age, sex, past medical history and presenting signs and symptoms were gathered by a questionnaire. Cholecystectomy was achieved in 100 patients hospitalized in Sina and Taleghani hospitals. Samples collected by a surgeon and injected into a Stuart transport medium. A section of sample was cultured on Selenite F medium by swap and after 12 to 24 hours incubation in 37°C, was cultured again on selective salmonella-shigella agar (ss) medium. The colonies were studied after 24 hours incubation in 37°C, after which the microorganisms were detected by differential tests. Another section of sample was cultured on tioglycolate medium to detect anaerobic bacteria. To detect other bacteria, the samples were cultured on blood agar and MacKoncky agar incubated in 37°C for 24h. Differential tests such as IMVIC (Indole, Methyl Red, Voges presquare, Citrate), motility, urease and lysin decarboxylase were performed after staining and microscopic observation. We performed antibiotic susceptibility tests by Kirby-Baur method with a colony in Muller-Hinton agar medium. The results were reported as susceptible, resistant, or intermediate, based on the diameter of the clear zone around disks in attention to the antibiotic standard table. The applied antibiotic disks were ampicillin, tetracycline, nalidixic acid, deoxycycline, chloramphenicol, gentamicin, cephtioxim, amikacin, kanamycin, cephalothin, cotrimoxazole and nitrofurantoin

## RESULTS

The study population included 60 females and 40 males with a mean age of 52 years (range, 20-78 years).

Totally, 63 samples showed bacterial growth, of which 11 revealed two species of bacteria. Salmonella species were not detected. A total of 10

gram positive and 63 gram negative bacteria were isolated. The gram positive bacteria included enterococci (8 samples) and staphylococci (2 samples). The gram negative bacteria were E.coli (25 samples), klebsiella (12 samples), aerobacter (10 samples), pseudomonas (9 samples), proteus (3 samples), citrobacter (3 samples), providencia (2 samples) and acinetobacter (1 sample).

Antibiotic susceptibility tests were achieved for enterococci and E.coli isolates and the following data were obtained. E.coli was completely (100%) susceptible to kanamycin, while it was relatively resistant to cotrimoxazole and chloramphenicol (92%). Enterococci were completely (100%) resistant to nalidixic acid, gentamicin, amikacin and kanamycin, however, they were completely (100%) susceptible to tetracycline, deoxycycline and chloramphenicol.

## DISCUSSION

The mean age of patients was 52 years while most of the sufferers (63%) aged 40-60 years. Mean age of cholecystectomy was reported 46 and 51.7 years in two other studies that is in agreement with ours (1,3).

In our study, 37% of samples did not grow any bacteria, however, they could be infected with viruses or fungi. Similarly, Bonnet et al reported viruses as a relatively common causative agent (4). E.coli was detected in 25 samples (39.7% of gram-negative-infected subjects). Tseng et al found E.coli in 57% of their cases (5). In our study, the most prevalent isolated gram positive bacterium was Enterococci (8 of 10 samples). Csendens and Bedirli reported Enterococci as the most prevalent gram positive bacteria (1,6), conversely, another study demonstrated coagulase negative staphylococci as the most prevalent gram positive cocci (3.6%) (7,8). Meanwhile, brucella and aeromonas were also reported (9,10).

The mechanism of increased gallstone formation by bacterial betaglucoronidase has been

proposed by prior researchers. The production of betaglucuronidase by bacteria causes sedimentation in gallbladder and results in gallstones formation.

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