

Microflora of Bile Aspirates and its Antibigram

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ABSTRACT

Objective: To determine the frequency of bacterial infection in patients presenting with Cholelithiasis.

Study Design: Descriptive case-series study.

Place and Duration of Study: This study was carried out in the Surgical Unit Nishtar Hospital, Multan from October 2007 to September 2008.

Materials and Methods: All adults patients aged 20 years and above who were admitted with a provisional diagnosis of cholelithiasis, over a period of one year were entered into the study. The study included 100 patients between the ages of 20 and 60 years.

Results: The 100 patients who presented with cholelithiasis underwent operation and bile was taken for culture and sensitivity. There were 88 women (88%) and 12 (12%) were men. Age was ranging from 20 to 60 years (mean 44.4 years). 56 (56%) were operated laparoscopically while in 44 (44%) open cholecystectomy was done. Out of 100 bile culture 16 (16%) were positive while in 84 (84%) cases no growth cultured.

Conclusion: In patients of cholelithiasis bile may be infected and infection may be causative factor in formation of gall stones. Most of the microorganism were sensitive to cefuroxime axetil and ceftriaxone.

Key words: Bile culture, Cholelithiasis, E.Coli.

INTRODUCTION

Gallstones are abnormal masses of a solid mixture of cholesterol crystals, mucin, calcium bilirubinate, and proteins that have affected people for centuries.¹ Multiple gallstones were found in a mummified Egyptian priestess², but the disease was first described in 1507 by a Florentine pathologist, Antonio Benivenius³.

The Swiss medic Paracelsus viewed gallstones as a consequence of "tartaric" disease. With a prevalence of 10–15% in adults in Europe and the USA, gallstone disease is one of the most common and most expensive to treat of digestive disorders that need admission to hospital⁴. Every year in the USA, more than one million people are newly diagnosed with gallstones, and about 7,00,000 individuals have cholecystectomies⁵. In 1882, in the first open cholecystectomy Langenbuch successfully removed the gallbladder of a 43-year-old man who had had gallstones for 16 years. This technique remained the gold standard therapy for symptomatic gallstones for over a century, although medical treatment with bile acids was first described in the late 19th century⁶.

After a report of complete dissolution of gallstones by bile acids in 1937⁷, oral bile acid litholysis with chenodeoxycholic acid as a method for removing cholesterol gallstones emerged in the 1970s⁸, and litholysis with ursodeoxycholic acid in the 1980s. Extracorporeal shockwave lithotripsy plus oral bile acids for symptomatic gallstones was introduced first in 1986 in Munich⁹.

Later, several studies proved that gallstones recur in 30–50% of cases, 5 years after bile salts therapy or

lithotripsy¹⁰. In 1987, Mouret undertook the first laparoscopic cholecystectomy, which is today the treatment of choice for symptomatic gallstones. In the human gallbladder, three types of gallstones exist, depending on the major constituents: pure cholesterol, pure pigment, and mixed (small amounts of calcium and bilirubin salts).

Pigment stones appear in two major forms: black and brown. Whereas black pigment stones result from haemolysis and consist primarily of calcium bilirubinate, brown pigment stones are associated with infections of the biliary tract (bacterial and helminthic) and are more frequent in Asia or occur after cholecystectomy as de novo common bile duct stones.¹⁸ Cholesterol gallstone disease results from a complex interaction of genetic and environmental risk factors. Infection plays a major role in formation of gall stone and bile may not be sterile in patient with cholelithiasis. The purpose of the study was to determine the microflora of bile aspirates and its antibiogram.

MATERIALS AND METHODS

Patients of Cholelithiasis admitted through out patient department [OPD]. Patients who were about to undergo cholecystectomy were selected from the ward. Ultrasound of abdomen was done on every patient to confirm the diagnosis of cholelithiasis. Informed consent was taken from the patient and patient was briefed about study and confidentiality was maintained. It was also ensured that no risk is involved to the patient in this study. History was taken from the patient about pain right

hypochondrium, dyspepsia, nausea, vomiting, jaundice, fever, weight loss.

Examination of patient was done for pulse, temperature, jaundice, tenderness or mass in right hypochondrium.

Certain basic investigations like complete blood count, random blood sugar, liver function tests, renal parameters and specific investigations like ultrasound of abdomen was done.

A dose of prophylactic antibiotic was given to the patient at the time of induction of anesthesia. Patients were operated under general anesthesia in supine position. After opening/entering the abdomen and confirming the diagnosis bile aspirated from the fundus of gall bladder in 5cc syringe. All specimens were sent to the department of pathology for culture and results were recorded on a proforma.

RESULTS

The 100 patients who presented with Cholelithiasis underwent operation and bile was taken for culture and sensitivity. There were 88 women (88%) and 12 (12%) were men. Age was ranging from 20 to 60 years, mean 44.4 years (Table-1).

In all patients, 56 (56%) were operated laparoscopically while in 44 (44%) open cholecystectomy was done (Table-2).

Out of 100 bile culture 16 (16%) were positive while in 84 (84%) bile culture was negative (Table-3).

Most of bacteria were sensitive to cefuroxime axetil [second generation cephalosporin] and ceftriaxone sodium [third generation cephalosporin].

Table No. 1: Age distribution (n=100)

Age (Years)	No. of patients	%age
20-30	18	18.0
31-40	42	42.0
41-50	26	26.0
51-60	14	14.0

Table No.2: Procedure of surgery (n=100)

Procedure	No. of patients	%age
Laparoscopic cholecystectomy	56	56.0
Open cholecystectomy	44	44.0

Table No.3: Status of patients (n=100)

Status	No. of patients	%age
Positive	16	16.0
Negative	84	84.0

DISCUSSION

Gallbladder disease is the commonest indication for abdominal surgery and is the second most common intra abdominal operation performed in the western

countries¹¹. Gall stones are responsible for more than 95% of biliary tract disease¹².

Different factors have been implicated in the causation of gall stones amongst which infection of the bile is also as important factor. In about 30% of the patients with cholelithiasis, bacteria can be cultured either from the bile or from the wall of the gallbladder. The biliary infection can be caused by any type ranging from aerobic gram positive to gram negative to anaerobic organisms. Aerobic organisms cause 94% of biliary tract infections while anaerobic organisms cause the rest.

Bacteria are commonly found in inflamed gallbladder and in patients with cholelithiasis, whereas evidence suggests that normal bile is sterile¹³. Inflamed gallbladder has markedly altered permeability, which permits absorption of bile acids and movement of inorganic salts into the gallbladder lumen.

The role of excessive cellular debris and increased protein secretion, which occurs in response to inflammation, may be present. Finally, bacterial enzymes effects constituency of bile which may alter its solubility leading to precipitation of bile salts. Most gall stones are composite in nature. Bacteria can be found in most pure stone (i.e. those whose structure consists more than 90% cholesterol). The natural history of gall stones is unknown. It is likely that brown pigment stones can evolve in their chemical composition after termination of the infection process that initiate their formation, and may further develop into either mixed or nearly pure cholesterol stones. It is difficult to ascertain that whether bacterial infection of bile arose from stone formation or vice versa.

Although the exact contribution of bacteria in lithogenesis is not known, it is important for the clinician to realize that most gall stones are likely to be colonized by bacterial biofilm, even though the bile may be culture negative.

Cholecystitis and cholelithiasis are prevalent in certain regions of the world and quite rare at other places. Hence, these are sometimes called South Western American disease¹⁹ and has been reported in 54% of the adults above 21 years of age¹⁴.

A bacterial cause of cholecystitis has been proposed and positive bile cultures have been noted in 46% of patients with acute cholecystitis. In one study from Germany, using molecular genetic methods, bacteria could be found in most pure cholesterol stones (i.e. those whose structure consists of more than 90% cholesterol)¹⁵. It is suggested that bile infection by *E. coli*, in addition to bile stasis, plays a crucial role in the pathogenesis of brown pigment stones¹⁶. Bacterial DNA sequences are usually present in mixed cholesterol, brown pigment, and common bile duct, but rarely in pure cholesterol gallstones¹⁷. Interest has continued to abound in the role of infection in cholelithiasis. Two fallacies, however, exist in this

regard. Firstly, the culture of the organism from the bile at the time of the operation does not necessarily indicate a cause effect relationship between the infective microorganism and lithogenesis, as infection may be secondary to calculus formation. Secondly, the failure to isolate organism from bile also does not indicate that the etiology is unrelated to the infection as it is well-known that organisms which have initiated the stone precipitation may not persist in the viable form in the bile till surgery.

This study shows that this disease is much more common in females as compared to the males. The mean age incidence in this series is 48 years in females and 44 years in males.

Iqbal et al. in 2001 reported maximum number of patients with cholelithiasis between the age of 20 – 30 years with the highest incidence of choledocholithiasis accompanying cholelithiasis in 61–75 years of age¹⁸.

In this series, the positive bile culture was 36%, which is considerably higher than that reported by Yaqin and Sultan¹⁹.

However, more recently Sabir³⁰ has reported an incidence of 16%. Harbi²⁰ in 2001 reported 25% and Csendes reported 46%²¹. Van Leeuwen from Kuwait showed positive bile cultures in 16.4% and 19 different bacterial species were identified²². Guo from China showed the incidence of bacteria to be very high, ranging from 20 to 96%, with an average of 66.7% depending on the kind of gallstone present²³. Therefore, infection is likely to range from 16 to 96 % which corroborates with the present finding.

CONCLUSIONS

In patients of Cholelithiasis bile may be infected and infection may be causative factor in formation of gall stones. E. Coli is most common organism found in bile culture as in many other national and international studies.

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