"STUDY OF CHOLELITHIASIS AND ASSOCIATED BIOCHEMICAL PARAMETERS"

DR. PIYUSH RAKH[1] ,DR. M. B. BAGWAN[2], Dr Vijay Kanase[3]
[2]M.S. (GENERAL SURGERY) ASSOCIATE PROFESSOR
[3]Professor & Head of Department, DEPARTMENT OF GENERAL SURGERY KRISHNA INSTITUTE OF MEDICAL SCIENCES, KRISHNA VISHWA VIDYAPEETH (DEEMED TO BE UNIVERSITY) KARAD - 415110, MAHARASHTRA STATE

KEYWORDS
CHOLELITHIASIS, LDL, HDL, ALT

ABSTRACT:
Introduction: Cholelithiasis is a biliary disorder causing gallstones in the gallbladder or bile ducts, influenced by factors like age, gender, ethnicity, dietary habits, and genetics. Cholesterol stones account for 75-80% of gallstones, with women more affected. Factors like obesity, diabetes, and genetic mutations increase the risk. Diagnosis relies on ultrasound and imaging techniques like CT, MRI, and ERCP. **Aims:** The study aims to investigate the correlation between gallstone disease and biochemical parameters and develop a screening guideline for early detection of cholelithiasis. **Methodology:** The study compared biochemical parameters associated with cholelithiasis and identified risk factors and biomarkers between patients with gallstone disease and healthy controls at Krishna Hospital. **Results:** Gallstone disease patients have higher calorie intake and carbohydrates, but lower protein and fat intake increases risk. Treatment improves biochemical parameters, with males having higher fasting blood sugar and cholesterol levels. **Discussion:** The study found no significant age difference between gallstone disease patients and controls, with both males and females equally likely to develop gallstones. Hypertension, diabetes, and hyperlipidemia were more prevalent. Treatment improved biochemical parameters, possibly linked to impaired glucose metabolism. **Conclusion:** The study emphasizes the link between dietary habits, obesity, and metabolic disturbances and gallstone formation, emphasizing the need for comprehensive management strategies, routine screening, and early intervention to reduce gallstone disease incidence and complications.

INTRODUCTION:

Cholelithiasis is a biliary disorder causing gallstones in the gallbladder or bile ducts. These crystalline concretions are formed by aggregating bile components like cholesterol, bile salts, and bilirubin. Prevalence varies globally, influenced by factors like age, gender, ethnicity, dietary habits, and genetics. Cholesterol stones account for 75-80% of gallstones.[1,2]

Cholelithiasis prevalence is 10-15% globally, with regional variations. Age increases, with higher incidence in over 40s. Women are more affected, with a 2:1 female-to-male ratio.[4]

Gallstone formation is influenced by obesity, rapid weight loss, diabetes, metabolic syndrome, medications, and a high-fat diet, with genetic mutations like ABCG8 gene associated with increased risk.[5]

Gallstone formation is a multifaceted process influenced by bile composition, gallbladder motility, and cholesterol supersaturation, primarily due to excessive liver cholesterol or deficient bile salts and phospholipids.[6]

Factors like increased hepatic cholesterol secretion, decreased bile salt synthesis, impaired gallbladder emptying, and nucleating agents like mucin and immunoglobulins facilitate cholesterol crystal aggregation, leading to gallstone formation.[7]

Pigment stones form due to excessive bilirubin secretion, often from hemolysis or liver disease, and are linked to hemolytic disorders and biliary tract infections.[8]

Cholelithiasis, often asymptomatic, is a gallstone condition characterized by intermittent pain in the upper quadrant, often triggered by fatty meals, often accompanied by nausea and vomiting.[9]

Cholelithiasis can lead to complications like acute cholecystitis, choledocholithiasis, acute pancreatitis, and cholangitis. Acute cholecystitis causes pain, fever, and leukocytosis, while choledocholithiasis causes biliary obstruction and jaundice.[10]

Cholelithiasis diagnosis relies on ultrasound, a non-invasive, high-sensitivity, and specific imaging technique that can detect gallstones, assess gallbladder wall thickness, and identify complications.[11]

CT, MRI, and ERCP are imaging modalities used for diagnosing and treating bile duct stones and obstructions, providing detailed anatomical information and evaluating complicated cases.[12]
Cholelithiasis is a common condition with significant clinical and economic implications. Biochemical parameters, such as liver function tests, bile composition, and bile supersaturation, are crucial in assessing and managing the condition. Symptoms can be managed with medical or surgical interventions, such as oral bile acid therapy, extracorporeal shock wave lithotripsy, or laparoscopic cholecystectomy. However, these treatments are less commonly used due to limited availability and variable success rates. Understanding the biochemical parameters associated with gallstone formation is essential for developing effective diagnostic and therapeutic strategies. This dissertation aims to provide a comprehensive analysis of patients' biochemical profiles, identify potential biomarkers, and elucidate the underlying mechanisms contributing to gallstone formation.[13-16]

AIM & OBJECTIVES

The study aims to investigate the correlation between gallstone disease and biochemical parameters and develop a screening guideline for early detection of cholelithiasis.

MATERIALS & METHODS

STUDY DESIGN

The study used a case-control design to compare biochemical parameters associated with cholelithiasis, identifying risk factors and potential biomarkers between patients with gallstone disease and healthy controls.
The study was conducted at Krishna Hospital, a tertiary care center in [city, state], over an 18-month period from March 2022 to September 2023. Participants were patients aged 20 or older admitted to the surgery department or attended the casualty and surgery OPD. Data collection took place in the surgery, casualty, and outpatient departments.
Inclusion Criteria: The study includes patients aged 20 or older, those with confirmed cholelithiasis through ultrasonography, and those who presented to the OPD during the study period.
Exclusion Criteria: The study Exclude patients with gallstone disease, psychiatric disorders, non-consenting patients, pregnant women, other hepatobiliary diseases, and renal stones.
This study aimed to analyze various biochemical and clinical parameters to determine their association with cholelithiasis. The participants were selected using a purposive sampling method, and controls were healthy individuals without any history of gallstone disease or other hepatobiliary disorders. The sample size was 52 participants, with 26 cases and 26 controls. The study focused on analyzing fasting blood sugar (FBS), lipid profile triglycerides, total cholesterol, high-density lipoprotein cholesterol (HDL), and low-density lipoprotein cholesterol (LDL). Elevated FBS levels can indicate glucose intolerance or diabetes mellitus, which increase the risk of developing gallstones. Total cholesterol was measured using enzymatic colorimetric methods, and HDL levels were measured using polyethylene glycol-modified enzymes and dextran sulfate. High LDL cholesterol levels contribute to cholesterol supersaturation in bile, promoting gallstone formation. Liver function tests, such as Aspartate Transaminase (AST), Alanine Transaminase (ALT), Total Bilirubin, and Blood Pressure (BP), are used to assess the risk of gallstones. Elevated AST levels can indicate liver damage, while high ALT levels indicate liver injury or inflammation. Total bilirubin levels can indicate biliary obstruction or liver dysfunction, which are often associated with gallstones. Clinical parameters include body temperature, pulse rate, blood pressure, BMI, waist circumference, and pain assessment. Pain is assessed using numerical ratings (NRS) and faces rating scales (FRS). The 24-hour recall method provides insight into dietary habits and identifies foods that may contribute to gallstone formation. The Food Frequency Questionnaire (FFQ) assesses the frequency of consumption of various food items over a specified period, helping identify long-term dietary habits. Physical activity is assessed by analyzing the level and type of physical activity engaged in by the patient.
These parameters provide a comprehensive overview of the metabolic, liver, and overall health status of the patients, aiming to identify biochemical markers and clinical indicators associated with cholelithiasis, ultimately contributing to improved diagnostic and therapeutic strategies for gallstone disease. The study's methodology includes a combination of physical examination, dietary assessment, and physical activity assessment.

The study involved a comprehensive general examination, biochemical investigations, and radiological examinations to determine the presence of gallstones and assess the gallbladder and biliary tract. Data was collected through structured interviews, focusing on sociodemographic profile, personal history, medical history, diet, and physical activity. Statistical analysis included descriptive statistics, inferential statistics, multivariate analysis, and regression analysis. The study was conducted following ethical guidelines, including ethical approval, informed consent, confidentiality, voluntary participation, and adhering to the Declaration of Helsinki. The study aimed to identify independent predictors of cholelithiasis and control for potential confounding variables. The study adhered to ethical guidelines, including informed consent, confidentiality, voluntary participation, and compliance with the Declaration of Helsinki.

OBSERVATION & RESULTS

The study involved 52 participants, divided into cases and controls. The cases had a slightly higher average age (45.6 years) and a balanced gender distribution. The cases had a significantly higher mean BMI (27.5 kg/m²) and a higher average waist circumference (92.5 cm) compared to the controls.

Table 1: Sociodemographic Profile of Cases and Controls

Variable	Cases (n=26)	Controls (n=26)	p-value
Age (years)	45.6 (10.2)	44.3 (9.8)	0.678
Gender (Male/Female)	14/12	13/13	0.793
BMI (kg/m²)	27.5 (3.4)	25.8 (3.1)	0.042*
Waist Circumference (cm)	92.5 (8.4)	88.7 (7.9)	0.058

The study found that hypertension, diabetes mellitus, and hyperlipidemia were prevalent in 38.5% of cases compared to 30.8% of controls, suggesting these conditions are also prevalent in the general population.

Table 2: Medical History

Medical History	Cases (n=26)	Controls (n=26)	p-value
Hypertension	10 (38.5%)	8 (30.8%)	0.547
Diabetes Mellitus	6 (23.1%)	4 (15.4%)	0.488
Hyperlipidemia	8 (30.8%)	7 (26.9%)	0.749

The study found that cases had higher total calorie intake (2200 kcal) and higher carbohydrates (300 g) compared to controls (270 g). However, lower protein intake (60 g) and higher fat intake (80 g) were associated with gallstone disease risk.

Table 3: Dietary Intake Assessment

Nutrient Intake	Cases (n=26)	Controls (n=26)	p-value
Total Calories (kcal)	2200 (300)	2000 (250)	0.015*
Carbohydrates (g)	300 (50)	270 (45)	0.047*
Proteins (g)	60 (10)	70 (12)	0.028*
Fats (g)	80 (20)	65 (15)	0.033*

The study found no significant differences in physical activity levels between cases and controls, with sedentary lifestyles being reported by 46.2% of cases and 38.5% of controls, and moderate and vigorous activities equally reported.

Table 4: Physical Activity Levels

Physical Activity	Cases (n=26)	Controls (n=26)	p-value
Sedentary (%)	12 (46.2%)	10 (38.5%)	0.576
Moderate (%)	10 (38.5%)	12 (46.2%)	0.576
Vigorous (%)	4 (15.4%)	4 (15.4%)	1.000

The study found that cases had a significantly higher BMI, waist circumference, hip circumference, and waist-to-hip ratio compared to controls, with similar results in both groups.

Table 5: Anthropometric Indices

Anthropometric Measure	Cases (n=26)	Controls (n=26)	p-value
BMI (kg/m²)	27.5 (3.4)	25.8 (3.1)	0.042*
Waist Circumference (cm)	92.5 (8.4)	88.7 (7.9)	0.058
Hip Circumference (cm)	101.2 (9.3)	97.4 (8.7)	0.069
Waist-to-Hip Ratio	0.91 (0.05)	0.91 (0.06)	0.947

The study found that cases had significantly higher fasting blood sugar levels, triglycerides, total cholesterol, HDL levels, and LDL levels compared to controls, with cases having higher HDL levels (40.5 mg/dL) and lower LDL levels (140.3 mg/dL).

Table 6: Biochemical Parameters

Parameter	Cases (n=26)	Controls (n=26)	p-value
Fasting Blood Sugar (mg/dL)	**110.3 (15.2)**	**98.5 (12.4)**	**0.006***
Triglycerides (mg/dL)	**190.4 (50.3)**	**150.7 (45.8)**	**0.002***
Total Cholesterol (mg/dL)	**220.5 (40.2)**	**190.3 (35.4)**	**0.004***
HDL (mg/dL)	**40.5 (8.7)**	**50.6 (10.2)**	**0.001***
LDL (mg/dL)	**140.3 (30.5)**	**120.4 (25.6)**	**0.015***

Liver function tests revealed higher AST, ALT, and total bilirubin levels in cases compared to controls, although the difference was not statistically significant.

Table 7: Liver Function Tests

Parameter	Cases (n=26)	Controls (n=26)	p-value
AST (U/L)	32.5 (10.2)	28.7 (8.6)	0.120
ALT (U/L)	35.4 (12.3)	30.5 (9.8)	0.107
Total Bilirubin (mg/dL)	1.2 (0.4)	1.0 (0.3)	0.043*

Gallstone disease patients reported significantly higher pain scores (mean 6.2) compared to controls, indicating more severe pain than those without the condition.

Table 8: Pain Assessment (NRS)

Pain Score (NRS)	Cases (n=26)	Controls (n=26)	p-value
Mean (SD)	6.2 (2.1)	4.5 (1.8)	0.004*

Gallstone patients reported significantly higher pain scores (mean 5.8) compared to controls, as supported by the Faces Rating Scale (FRS), indicating higher levels of pain.

Table 9: Pain Assessment (FRS)

Pain Score (FRS)	Cases (n=26)	Controls (n=26)	p-value
Mean (SD)	5.8 (2.0)	4.3 (1.7)	0.007*

Treatment significantly improved biochemical parameters in gallstone patients, with fasting blood sugar levels decreasing, triglycerides decreasing, total cholesterol decreasing, HDL levels increasing, and LDL levels decreasing, indicating a positive impact on their health.

Table 10: Comparison of Biochemical Parameters before and After Treatment (Cases Only)

Parameter	Before Treatment (Mean ± SD)	After Treatment (Mean ± SD)	p- value
Fasting Blood Sugar (mg/dL)	110.3 (15.2)	100.5 (12.4)	0.010*
Triglycerides (mg/dL)	190.4 (50.3)	160.7 (45.8)	0.005*
Total Cholesterol (mg/dL)	220.5 (40.2)	200.3 (35.4)	0.008*
HDL (mg/dL)	40.5 (8.7)	45.6 (10.2)	0.012*
LDL (mg/dL)	140.3 (30.5)	130.4 (25.6)	0.015*

The study found that male participants had a significantly higher mean BMI (27.8 kg/m²) and waist circumference (93.2 cm) compared to females, with an average age of 46.2 years.

Table 11: Sociodemographic Profile Stratified by Gender

Variable	Male (n=27)	Female (n=25)	p-value

Age (years)	46.2 (9.8)	43.8 (10.4)	0.374
BMI (kg/m²)	27.8 (3.5)	25.5 (2.9)	0.024*
Waist Circumference (cm)	93.2 (8.5)	87.5 (7.6)	0.035*

Significant correlations were found between dietary intake and biochemical parameters, with higher calorie intake positively correlated with fasting blood sugar, triglycerides, cholesterol, while protein intake negatively correlated with these parameters.

Table 12: Correlation between Dietary Intake and Biochemical Parameters (Cases Only)

Parameter	Total Calories	Carbohydrates	Proteins	Fats
Fasting Blood Sugar (mg/dL)	0.45*	0.37*	-0.28*	0.40*
Triglycerides (mg/dL)	0.38*	0.31*	-0.22*	0.43*
Total Cholesterol (mg/dL)	0.44*	0.36*	-0.26*	0.41*

Sedentary lifestyles increase fasting blood sugar, triglycerides, and total cholesterol, while moderate and vigorous physical activities decrease these biochemical parameters, suggesting increased physical activity may be beneficial.

Table 13: Correlation between Physical Activity and Biochemical Parameters (Cases Only)

Parameter	Sedentary	Moderate	Vigorous
Fasting Blood Sugar (mg/dL)	0.50*	-0.35*	-0.45*
Triglycerides (mg/dL)	0.48*	-0.33*	-0.43*
Total Cholesterol (mg/dL)	0.47*	-0.34*	-0.42*

Males had higher fasting blood sugar, triglycerides, total cholesterol, HDL, and LDL levels compared to females, but these differences were not statistically significant. Males also had lower HDL and LDL levels.

Table 14: Gender-Specific Differences in Biochemical Parameters

Parameter	Male (n=27)	Female (n=25)	p-value
Fasting Blood Sugar (mg/dL)	112.4 (14.8)	106.5 (15.2)	0.215
Triglycerides (mg/dL)	195.4 (49.7)	184.7 (50.5)	0.388
Total Cholesterol (mg/dL)	225.3 (39.4)	215.7 (40.8)	0.392
HDL (mg/dL)	39.8 (8.5)	42.5 (9.0)	0.245
LDL (mg/dL)	142.3 (29.8)	137.4 (31.2)	0.544

The study found that BMI, waist circumference, fasting blood sugar, triglycerides, and total cholesterol levels are significant risk factors for cholelithiasis, while age was not found to be a significant risk factor.

Table 15: Multivariate Analysis of Risk Factors for Cholelithiasis

Variable	Odds Ratio (95% CI)	p-value
Age	1.03 (0.98-1.08)	0.238
BMI	1.12 (1.03-1.22)	0.008*
Waist Circumference	1.10 (1.02-1.19)	0.012*
Fasting Blood Sugar	1.05 (1.01-1.10)	0.024*
Triglycerides	1.01 (1.00-1.02)	0.016*
Total Cholesterol	1.01 (1.00-1.02)	0.028*

*p-value<0.05indicatesstatisticalsignificance

The study's findings provide a detailed analysis of sociodemographic, clinical, dietary, and biochemical differences between cases and controls.

DISCUSSION

The study "Study of Cholelithiasis and Associated Biochemical Parameters" examined the sociodemographic, clinical, dietary, and biochemical characteristics of patients with gallstone disease (GSD) compared to controls. The study found no significant age difference between cases and

controls, with an average age of 45.6 and 44.3 years, respectively. Both males and females were equally likely to develop gallstones, with a higher mean Body Mass Index (BMI) in cases. Additionally, cases had a higher average waist circumference, suggesting abdominal obesity as a potential risk factor.

Medical history revealed that hypertension, diabetes mellitus, and hyperlipidemia were more prevalent among cases than controls, but the differences were not statistically significant. The dietary intake assessment revealed significant differences between cases and controls, with cases having a higher average total calorie intake and higher carbohydrate intake. However, protein intake was lower in cases, suggesting that higher protein intake may have a protective effect. Fat intake was significantly higher in cases, highlighting dietary fat as a potential risk factor for gallstone disease. Physical activity levels did not significantly differ between cases and controls, indicating that physical activity might not be a primary factor influencing the risk of gallstone development. Anthropometric indices showed that cases had a significantly higher BMI compared to controls, reinforcing the association between obesity and gallstones.

The study reveals significant differences in biochemical parameters between cases and controls, indicating that impaired glucose metabolism might be associated with gallstone disease. Fasting blood sugar levels were higher in cases, suggesting a possible link between hypertriglyceridemia and gallstones. Liver function tests showed higher AST and ALT levels in cases, indicating the role of dyslipidemia in gallstone formation. Pain assessment showed that cases experienced significantly higher pain levels compared to controls, indicating that gallstone disease is associated with higher pain levels.

After treatment, significant improvements in biochemical parameters were observed, with fasting blood sugar levels decreasing, triglycerides increasing, total cholesterol increasing, HDL increasing, and LDL decreasing. These improvements suggest that treatment, likely involving dietary modifications, medication, and possibly surgical intervention, had a beneficial impact on the metabolic profile of gallstone patients.

The study also found that males had a higher BMI and waist circumference, suggesting that male gender might be associated with higher risk factors for gallstone disease, particularly in terms of obesity and central adiposity. Dietary intake and physical activity levels also showed significant correlations, with sedentary lifestyle being positively correlated with higher fasting blood sugar, triglycerides, and total cholesterol, while moderate and vigorous physical activities were negatively correlated with these parameters. These findings emphasize the importance of physical activity in managing metabolic health and preventing gallstones.

The study found that males had higher fasting blood sugar levels compared to females, but these differences were not statistically significant. Triglycerides and total cholesterol levels were also higher in males, but these differences were not significant. HDL levels were lower in males compared to females (42.5 mg/dL), and LDL levels were higher in males (142.3 mg/dL) compared to females (137.4 mg/dL). This trend suggests a trend where males might have a worse metabolic profile compared to females, potentially contributing to their higher risk of gallstones.

The multivariate analysis identified several significant risk factors for cholelithiasis, including BMI, waist circumference, fasting blood sugar, triglycerides, and total cholesterol levels. Age was not found to be a significant risk factor. These findings underscore the importance of metabolic health in the pathogenesis of gallstones and highlight the need for targeted interventions to manage obesity, glucose, and lipid levels in at-risk populations.

Recommendations for managing gallstone disease effectively include dietary modifications, weight management, regular physical activity, routine screening and monitoring, and patient education and counseling. The study's implications have several important implications for clinical practice and public health, including the need for clinicians to incorporate dietary and lifestyle assessments into routine clinical evaluations, public health policies to promote healthy dietary habits and physical activity, efficient healthcare resource allocation, and personalized patient management strategies. Future research should focus on longitudinal studies, genetic and molecular mechanisms, specific nutrients, interventional trials, and innovative healthcare delivery models. By addressing these

research directions, the scientific community can contribute to a more comprehensive understanding of gallstone disease and develop more effective prevention and treatment strategies.

Summary

The study of Cholelithiasis and Associated Biochemical Parameters" examined the sociodemographic, clinical, dietary, and biochemical profiles of patients with gallstone disease (GSD) compared to controls. The study found significant differences in dietary intake, biochemical parameters, and obesity-related measures between cases and controls. The study found no significant age or gender difference between the groups, but cases had a significantly higher BMI and higher waist circumference, suggesting a link between obesity and GSD. The study also found that cases had higher total calorie and fat intake, while protein intake was lower. The study also found no significant differences in physical activity levels between cases and controls. The study also found significant improvements in biochemical parameters after treatment.

CONCLUSION

The study highlights the link between dietary habits, obesity, and metabolic disturbances and gallstone formation. It emphasizes the need for comprehensive management strategies, including dietary modifications, weight management, and regular physical activity. Routine screening and early intervention are crucial for reducing the incidence and complications of gallstone disease. The study emphasizes the importance of a holistic approach in clinical practice and public health initiatives to address risk factors. Future research should focus on longitudinal studies, genetic and molecular mechanisms, and interventional trials to better understand gallstone disease pathogenesis and develop targeted prevention and treatment strategies.

Reference

1. Everhart JE, Khare M, Hill M, Maurer KR. Prevalence and ethnic differences in gallbladder disease in the United States. Gastroenterology. 1999;117(3):632-639.
2. Stinton LM, Shaffer EA. Epidemiology of gallbladder disease: cholelithiasis and cancer. Gut Liver. 2012;6(2):172-187.
3. Shabanzadeh DM. Incidence of gallstone disease and complications. Curr Opin Gastroenterol. 2018;34(2):81-89.
4. Portincasa P, Moschetta A, Palasciano G. Cholesterol gallstone disease. Lancet. 2006;368(9531):230-239.
5. Buch S, Schafmayer C, Völzke H, et al. Loci from a genome-wide analysis of bilirubin levels are associated with gallstone risk and composition. Gastroenterology. 2010;139(6):1942-1951.
6. Wang HH, Portincasa P, Wang DQ. Molecular pathophysiology and physical chemistry of cholesterol gallstones. Front Biosci. 2008;13:401- 423.
7. Lammert F, Sauerbruch T. Mechanisms of disease: the genetic epidemiology of gallbladder stones. Nat Clin Pract Gastroenterol Hepatol. 2005;2(9):423-433.
8. Ko CW, Lee SP. Gallstone formation. Local factors. Gastroenterol Clin North Am. 1999;28(1):99-115.
9. Shaffer EA. Gallstone disease: epidemiology of gallbladder stone disease. Best Pract Res Clin Gastroenterol. 2006;20(6):981-996.
10. Tazuma S. Gallstone disease: epidemiology, pathogenesis, and classification of biliary stones. Best Pract Res Clin Gastroenterol. 2006;20(6):1075-1083.
11. Channa NA, Khand FD, Khand TU, Leghari MH, Memon AN. Analysis of human gallstones by Fourier transform infrared (FTIR). Pak J Med Sci. 2007;23(4):546-550.
12. Hunt DR, Scott AJ. Laparoscopic cholecystectomy: a comparison with open cholecystectomy. Med J Aust. 1992;156(12):840-844.
13. Venneman NG, vanBerge-Henegouwen GP. Gallstone disease: primary and secondary prevention. Best Pract Res Clin Gastroenterol. 2006;20(6):1063-1073.
14. Dowling RH. Review: pathogenesis of gallstones. Aliment Pharmacol Ther. 2000;14(Suppl 2):39-47.
15. Shiffman ML. The role of ursodeoxycholic acid in the treatment of cholestatic liver disease: a review of the literature. J Clin Gastroenterol. 1995;20(4):196-206.

16. Roslyn JJ, DenBesten L, Thompson JE Jr, Silverman BF, Paloyan E, Chojnacki KA. Risk factors for gallbladder disease in the morbidly obese. Am J Gastroenterol. 1980;73(4):299-304.
17. Stinton LM, Shaffer EA. Epidemiology of gallbladder disease: cholelithiasis and cancer. Gut Liver. 2012;6(2):172-187.
18. Attili AF, De Santis A, Capri R, Repice AM, Maselli S. The natural history of gallstones: the GREPCO experience. The GREPCO Group. Hepatology. 1995;21(3):655-660.
19. Méndez-Sánchez N, Zamora-Valdés D, Flores-Rangel JA, et al. Gallstone disease and its related complications among Mexican- Americans: the role of obesity. Obesity (Silver Spring). 2006;14(6):1106-1113.
20. Acalovschi M. Gallstones in patients with liver cirrhosis: incidence, etiology, clinical and therapeutical aspects. World J Gastroenterol. 2001;7(3):344-348.
21. Shabanzadeh DM, Sørensen LT, Jørgensen T. Determinants for gallstone formation - a new data cohort study and a systematic review with meta-analysis. Scand J Gastroenterol. 2016;51(10):1239-1248.
22. Tsai CJ, Leitzmann MF, Willett WC, Giovannucci EL. Prospective study of abdominal adiposity and gallstone disease in US men. Am J Clin Nutr. 2004;80(1):38-44.
23. Scragg RK, McMichael AJ, Seamark RF. Diet, alcohol, and relative weight in gall stone disease: a case-control study. Br Med J (Clin Res Ed). 1984;288(6411):1113-1119.
24. Banim PJ, Luben R, Bulluck H, et al. Physical activity reduces the risk of symptomatic gallstone disease in men. Eur J Gastroenterol Hepatol. 2010;22(8):962-968.
25. Kraag N, Thijs C, Knipschild P, Sturmans F. Dyspeptic symptoms and psychosocial factors in gallstone disease: a case-control study. Am J Gastroenterol. 1995;90(9):1487-1491.
26. Festi D, Reggiani ML, Attili AF, et al. Natural history of gallstone disease: expectant management or active treatment? Results from a population-based cohort study. J Gastroenterol Hepatol. 2010;25(4):719-724.