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USE OF LIPID LOWERING AGENTS AND RISK OF GALLSTONE FORMATION, A HOSPITAL BASED CASE CONTROL STUDY

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Abstract

Abstract: Disease of gallbladder is commonly manifested as gallstone and gallbladder cancer. It is a common and costly clinical condition in the adult population of Pakistan. Gallstone is a disease which is unease serious health for human being, and millions of people are affected throughout the world and in Pakistan recently increasing in quantitative number of gallstone cases in all over Pakistan.

Methodology: The sample was constituted of total 370 patients. 100 cases and 270 controls were selected for this study the patient ratio was 1:2. For the purpose of conducting a case-control study, sample was divided into two groups i.e. one for cases and the other for control. Self-administered questionnaire was used to obtain data from each case and control. To minimize the selection bias the sample was collected through random sampling method.

Findings: In the risk factors included age, female gender, and obesity. the median age in women's for gallstone disease was 58 years (68.8%) while for male population it was less then female. current statin users were (4.8%) in control and (5.4%)in cases. the former user of statin was (1.8%) and gallstone disease was (2.3%). This study supported the association between the use of statin and gallstone formation (Erichsen, et al. 2010).The risk of gallstone formation increases with age, change in diet (high calorie food) or use of drugs (lipid lowering agents) being female 87% cases, and being married 78% cases of gallstone were found.

Keywords: BMI, Hypertension, Diabetes Mellitus, Smoking, Obesity.

Introduction

Disease of gallbladder is commonly manifested as gallstone and gallbladder cancer. It is a common and costly clinical condition in the adult population of Pakistan (Stinton et al. 2012). [28] Gallstone is a disease which is unease serious health for human being, and millions of people are affected throughout the world and in Pakistan recently increasing in quantitative number of gallstone cases in all over Pakistan (Hundal and Shaffer 2014) [18]. Epidemiological studies are the best method to accurately determine prevalence of gallbladder stone plus to identify the risky factors and these studies defines the incidence of diseases. Risky factors are found of cholesterol gallstone; advance age, female gender, high blood triglycerol level, high serum level of female sex hormone, sickle cell disease and multiparity. Obesity, rapid loss of weight plus sedentary lifestyle is also found the modified risk factors. Highest rates are reported in Mexico, Bolivia, Chile, and northern Japan. In Chile, gall-bladder cancer is frequently warps and females are the victim of death because of cancer. In Pakistan; cancer of gastrointestinal is more common in women (Kratzer et al. 1999). [21]

Although an accurate reason; the upper right abdominal pain is considered the dominant factor. Mostly patients are suffered from jaundice and many had diagnosed nausea, anorexia, and loss of weight. Least studies are available for the union of gallstone with chronic liver disease in Pakistan is documented in medical literature but no such study has been carried out in Pakistan, but the HCV is endemic in this region. Affected population and the heavy load of patients in the hospitals related to the gallstone with its complications; this simply seems that this is a significant financial burden. The identified components of gallstone provides the information to the physician for investigation and to conclude in depth roots related to gallstone for decision or conclusion whether this gallstone's patient should be therapeutically treated or to treat surgically (Bansal et al. 2014). [4]

Gallstone is the concretion that could be formed in

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biliary tract, and then involved the gall-bladder, gallstones could be one of the predominant and prevalent furthermore expensive gastroentero logical disease which leads towards countless economic expenditure and every country has to bear this burden around the globe. Per anum six lac cholecystectomy is performed in ten to fifteen percent for the adults of America, in addition, it is estimated that 5 billion dollars are being spent per anum for the gallstone's treatment, similarly complications of the surgery consumption is approximately 6.5 billion us dollars (Diehl, 1991) [15]. Pakistani studies reported data is still inadequate, while previous studies has been found cholecystectomy for gallstone which is about 9.03% of Southern Sindh area of Pakistan (Channa et al. 2004) [11]. Multiple studies compared that formation of gallstone has multifactorial characteristics alike "societal background, inherited genes, increasing oldness as well as female' gender which couldn't be customized though nutrition, physical activities, hasty loss of weight plus obesity could changeable".

The gallstone disease is occurring more frequently with increasing of age. Typology of stones are change with increasing age, cool, calm and collected predominantly of cholesterol but in oldness; the stone changes into black pigment stones. Women are twice prone to gallstone then man, especially in fertile years. Multiparty, contraceptive drugs use and estrogen therapy are the risky factors which generate the gallstone (Ko et al. 2008). [20]

In individuals with low caloric diet, weight loss or bariatric surgery are associated with gallstone Formation. Weight losses that exceed from 1.5 kg per week while bariatric surgery is a generator of stone formation. The Metabolic Syndrome comes due to three features; "abdominal obesity, high blood pressure, high fasting", increasing sugar level, triglyceride level as well as reducing level of High Density Lipoprotein. Metabolic syndrome (MS) and diabetes mellitus (DM) are the generator of gall stone diseases. Advanced cirrhosis is a well-known cause of gall stone formation, with an overall prevalence at 25-30%, and it is allied with chronic HEP C plus disease-causing infection in addition non-alcoholic full of fatty liver.

Literature Review

Gallstone disease is one of the most familiar biliary system diseases worldwide in which both inherited and environmental factors have role in its pathogenesis. There has been inadequate research on the downstream events that come about in the gallbladder. The gallbladder has motor function, with 20–30% emptying at 1- to 2-h intervals during the fasting state and 70–80% emptying after stimulation by cholecystokinin (CCK) during at meal. But, the motor function of gallbladder smooth muscle (GBSM) becomes impaired during cholesterol gallstone formation in patients. Among the many factors involved in the pathogenesis of CGD, gallbladder hypo motility and the resultant prolonged stasis of lithogenic bile seem to be the most important.(Chen et al. 2014b). [14]

Gallstones grow inside the gallbladder or biliary tract. These stones can be asymptomatic or symptomatic; only gallstones with symptoms or complications are defined as gallstone disease. Based on their composition, gallstones are classified into cholesterol gallstones, which characterize the major entity, and bilirubin stones. Black pigment stones can be caused by chronic haemolysis. Brown pigment stones typically build up in obstructed and infected bile ducts. Risk factors for gallstones are female sex, age, pregnancy, physical inactivity, obesity and over nutrition. Factors involved in metabolic syndrome increase the risk of developing gallstones and form the basis of most important prevention by lifestyle changes. Diagnosis is mainly based on clinical symptoms, abdominal ultrasonography and liver biochemistry tests. Symptoms often precede the onset of the three frequent and potentially acute complications of gallstones (acute cholecystitis, acute cholangitis and biliary pancreatitis).our future efforts should focus on new preventive strategies to overcome the onset of gallstones in at-risk patients in particular, but also in the universal population.

Gender

The gallstone disease is a common disease of biliary system. Two kinds of problems are related with gallbladder is Gallstone and gallbladder cancer. The frequency of occurrence of gallstone is high in females 'elderly population of Taiwan (25%). Occurrence of gallstone formation is higher two to three times in female then male. pregnancy, childbearing age and sex hormones are major risk factors (Liver and Diabetes 2016). Biliary stone is more associated with female gender ,age ,obesity, blood glucose level and cholecystitis while hepatitis C is associated with male gender (Zhu et al. 2014). [29] The rapid change in life style accompanied by changes in diet and sedentary life style. Prevalence of gallstone disease was high in women (14.8%) as compared to man (12.2%) population, as well with increased age, risk of gallstone formation were high (25.4%) while low (10.5%) in age less than sixty years. patients with diabetes type 2,cardiovascular, obesity, and metabolic syndrome were more risk to development of gallstone (Chen et al. 2014a). [13]
In an history of medicine gall bladder is still remain a common health problem and the common

malignancy of gallbladder .Exact cause of gallstone is unknown while some risk factors are increased age (51 for female and 53 for male), female sex (61 cases), weight loss. the common symptoms were found was pain (92.9%), anorexia (88.88%), nausea and vomiting (40%) the incidence rate was high (7 time in female, 6 time in male), sex ratio (male, female, 1:2.54) And significant difference in bilirubin was seen in male and female participant (Stinton and Shaffer 2012). [28]

The disease of gall bladder as a gallstone or cancer is most common gasto-esophageal reflux disease. The use of iron containing food and diet can increase the heme concentration in the blood and responsible for gallstone formation. A case control study identified that female were 2.7 times more prone to gallstone formation. There was association of diet, social custom and geographical areas. BMI was not influence in gallstone formation. Cholesterol gallstone were (104/459), and (117/459) was mixed gallstone.

The difference of diet between subject with gallstone or without is present. Age, female gender and increased age are prominent risk factors. The risk of gallstone formation increases with age, change in diet (high calorie food) or use of drugs (lipid lowering agents).Being female 87% cases, and being married 78% cases of gallstone were found. Low age for marriage of female has significant for stone formation. The use of oral contraceptives, illiteracy (21.98%), leafy vegetables, and consumption of junk foods, or green chilies (14.5) are the risk factors (Channa et al. 2007). [12]

Three lipid components are contained in bile; cholesterol, phospholipids and bile salt. Cholesterol couldn't dissolve in aqueous media; so mixed micelles are packed with bile salt plus phosphatidylcholine. Whenever the level of cholesterol exceeds in bile which cannot be solubilized by mixed micelles then formation and enhancement of gallstone started. Cholesterol hyper saturation of bile along with gallbladder hypo mobility that promotes nucleation is the key pathophysiological mechanism for gallstone.

Bile's super saturation with cholesterol becomes a cause due to hepatic hyper secretion of cholesterol, phospholipids and sourness salt. Increased level of biliary cholesterol is originated by intestine (dietetic & reabsorbed cholesterol) (Kern, 1995). Mature summation is promoted by the existence of kinematic and pronucleating protein like mucin (Lee, 1981); whereas gallbladder hypo-mobility are also facilitated and formatted by macroscopic gallstones due to cholesterol microcrystal's. Increased age, genders, familial history dietary factors, hormonal factors, sedentary life style and rapid weight loss are directly considered as risky factor of gallstone (Attili et al,. 1997). [3] Infection of gallstone formation has not been well-known.

Patients of gallstone could be treated by lipid-lowering due to cardiovascular. It is appeared and examined that lipid-lowering treatment is the hazardous factor for gallstone-disease because it dissolves the agents; chenodeoxycholic acid (CDCA) plus ursodeoxycholic acid (UDCA) decreases cholesterol saturation of the bile by reducing biliary secretion that's why they are used for gallstone dissolute (Bacharach & Hofmann, 1982). Treatment of chenodeoxycholic acid (CDCA) is highly connected with reduction of hydroxyl-3-methyalglutaryl-co-enzyme A (HMG-CoA) reducates activity (Carulli, De Leon et al. 1980). [10] Moreover, ursodeoxycholic acid (UDCA) formats micelles with cholesterol in the intestine and makes the cause to reduce the engagement of cholesterol (Leiss et al. 1984). These agents have common aspects in the mechanism of action with statin and ezetimibe. This study is a review which made focus on the special effects of extensive and wide-ranging use of lipid lowering drug on the treatment plus prevention related to gallstone, including less discussion is made on the communal use of lipid lowering drugs. Links portrayed between gallstones, vascular in addition to the use of lipid-lowering drug in the above mentioned review.

The situation is different in East Asia while it is totally different in developed countries; however, inflammatory infection is the result of biliary strictures and malignancies. Brown pigment stones are found in Asian population (Tsui, Lam et al. 2011).

Family History and Inherited Genes

Genetic susceptibility is a contributor factor to format the gallstone. Familial studies shed the light that; there relatives have a five times frequency of gallstone patients due to inherited genes. This ratio is higher in monozygotic twins at twelve percent and dizygotic twins at six percent. Formation of stone is found with the multifaceted interfaces (particularly diet-gene interactions) (Horiuchi et al., 1991) [17]. Several genes have several interactions with gallstone. Cholelithiasis is considered a polygenetic disease.

Age

Disease of gallstone formation is common in older population while it's also a complicated obstructive disease in children with blood disorder (hemolytic) was (96%) and (77%) in non-hemolytic patients. the mean age for cholecystectomy were 13 to 18

years old (76%). The comparison of present study with Miltenburg et al., showed that Hispanic child with sever obese was (22% vs. 36%) cholecystectomy in 36 children aged 13 to 18 years was performed (1.8 per year) and gallstone was more common in female children. Female sex hormone also plays role in gallstone formation (Bruno et al., 2011).

Prevalence of gallstone disease is increasing in adults (Lammert & Miguel, 2008). 80% of gallstone is consisting of cholesterol gallstones, while the rest is black/brown pigment stone (Lammert & Miguel, 2008).

Gallstone increases in old age four to ten times and typology of stone is also predominantly due to composed cholesterol then stone formatted into the black pigment stones in addition, further, complication increases with age, which leads towards cholecystectomy (Hllebrant et al., 1998).

Female Hormones

Female is a compelled interaction with the disease of gallstone, particularly in the period of the pregnancy duration. The underlying mechanism lies in female sexual hormones; parity, oral contraceptive usage including estrogen replacement therapy which is a contributive and activist factor headed to cholesterol gallstone formation (Chisholm et al., 1998).Female's sex hormones are adversely influenced by hepatic bile secretion in addition to the gallbladder's function. Estrogen is a cause of escalation the level of cholesterol's secretion and diminishes the bile salt secretion, reduced progestin's act, bile's salt secretion and damaged gallbladder is a serious cause and leads to stasis. Progestin, iron .uses of oral contraceptives which are the risky cause of gallstone; on the other hand, the increased risk is quite modest and not likely to be clinically meaningful (Smit et al., 1996).

During pregnancy; female's sex set of hormone endogenously rises, and biliary sludge is appeared five percent to thirty percent in women. Resolution is become apparent during the post-partum period; while the sludge is disappeared in gallstone (microlithiasis) or vanished in one-third, but again gallstone becomes found five percent (Smit et al., 1995). Additional risk factors which are for stone formation during pregnancy include obesity, reduces HDL, cholesterol and Metabolic Syndrome (Smith et al., 2000).

Obesity and Co-Morbidity

One of the infectious diseases Hepa-c is also responsible for gallstone formation. Its distributed 3% of the world population .male population with younger age gallstone is (0.4%), with HCV infection is more prone to gallstone disease. Hepatitis C is endemic in Pakistan and the complication of gallstone formation can increases the financial burden in Pakistan.2000 of total patient (53.3%) male, having gallstone (0.9%), and female population was (46.6%) ,having gallstone was (13.9%).the age for hepatitis positive patients (63.2%) was 40 years is also concerned (Shah et al., 2014).

The development of gallstone is affecting 15% of population of developed countries. Major risk factors which are modifiable are include, obesity, diabetes, rapid weight loss and sedentary life that can be changed. Obesity and metabolic syndrome are the major factors in increasing prevalence of gallbladder disease. Some recent epidemiological studies shows association between gallstone disease and ischemic heart disease 5.8% of population present with IHD (in men: 3.7%, and in women: 7.3 % (Targher and Byrne 2015).

Obesity is blowing up in frequency of which reaches in the level of epidemic for developed and developing countries similarly in China (Apstein & Carey, 1996). [2] Obesity and centripetal obesity are deep-rooted risks for gallstone ((Mazzella et al., 1992). At least two percentage of morbidly obese individual has been suffered from gallstone. Obesity in oldness; cholelithiasis is at the greatest risk (Miettinen, 1986).Obesity related to the female sex is a greater risk having than before risk which formats the stone. Woman with severe obesity shows an age-adjusted interacted risk of six percent for the development of gallstone as compared to the non-obese controls; their development of gallstone per anum is two percent.

Dyslipidemia, Diabetes Mellitus and the Metabolic Syndrome

Metabolic problem is due to the cholesterol gallstone disease which is correlated with the abnormalities of lipid, diabetes mellitus and adiposity. A low High Density Lipoprotein cholesterol (Botham KM, Bravo, 1995) [7] and hypertriglyceridemia (Miettinen et al., 1996) are carried an increased risk to develop the stones. Similarly there is no interaction between hypercholesterolemia. High homocysteine have correlation with gallstone disease.

Metabolic Syndrome could define with three structures; "abdominal obesity, high blood pressure, high fasting glucose blood level" which is a source of increasing triglyceride levels and reduced High Density Lipoprotein levels (Jones et al., 1993) [19]. Metabolic Syndrome and Diabetes Mellitus are considered the threat for gallstone is linked with stone's complications (Brown et al., 1987) [8]. Insulin resistance is predisposed for cholesterol gallstone' development (Porsch-Ozcurumez et al.,

2001). Hepatic insulin resistance acts improved by hepatic cholesterol secretion, depressing bile salt synthesis.

Rapid Weight Loss

Light caloric diet including bariatric surgery with loss of rapid weight is related with gallstones development in thirty percent to seventy one percent of such individuals (Bodmer et al., 2009). [5] Due to the increasing Bariatric surgery, these stones are shown apparently in the duration of the first six weeks after surgery when weight's loss is started to increase **(**Mathus-Vliegen 2008). Loss of weight is allied gallstones which is typically asymptomatic. Weight fluctuation is a generator of stone formation which is shown in the history of diet.

Diet and Total Parental Nutrition (TPN)

Obesity is led by high caloric intake, when dietary contents are not clear. Diet with high level of cholesterol, fatty acids, carbohydrates and legumes seem to elevator the risk of cholelithiasis. Fats, coffee, fiber, ascorbic acid, calcium plus alcohol become a source of reducing the risk. Western diet which is highly refined but the alarming source is to upsurge the level of cholesterol gallstone. This dietary change shifts pigment of cholesterol stone in Asian countries; that controls cholesterol metabolism (Buhman et al., 2000). [9]

Total Parental Nutrition is a contributive actor to develop the microlithiasis and gallstone plus increases the calculus cholecystitis in ill patients with critical situation. The appearance of biliary sludge comes within five to ten days of fasting. After four weeks of Total Parental Nutrition; TPN develops gallbladder sludge started to come with all evidences after six weeks. Sludge could be resolved within four weeks with an oral intake. Loss of appetite is proven a cause of gallbladder stasis (Angelico, 2000). [1] Additionally, lethal disorders; in which Total Parental Nutrition is frequently required, could affect the enterohepatic cycling of bile acids (Roslyn et al., 1993).

Lifestyle and Status

Social status and gallstones are controversial with each other. Previous cross-sectional study of non-Hispanic refused; that gallbladder disease is by the wrong way round interrelated to socioeconomic status. Socioeconomic status, however, might be an incidental indication of risky factors same alike obesity and other chronic medical conditions. Even the role of smoking for this disease is also not clear (Lee et al., 1998).

Reduce education of physical activity welcomes disease of gallstone while the physical activity prevents cholelithiasis. Endurance exercise averts symptomatic gallstones' development in men (Leitzmann et al., 1999).

Children and Disease of Gallbladder

Gallbladder in peads is found very rare alike 0.1% to 1.0%., black-pigment stone is rarely related to prematurity. Cholesterol stones are communal in children (Peter et al., 2008). Abdominal ultrasonography is found in children according to one explanation. Obesity as pre-dominant; eight percent to thirty percentage of gallstones is explored in children. The common and prevalence of this disease is seen higher in fatty children and adolescents.

Forty (40%) to fifty one (51%) percent of children are found with gallstones pre-dominantly; right upper quadrant abdominal pain is more found (Bogue et al., 2010). [6] Conservative management recommends for these children. Symptomatic cholelithiasis is found with risky complications and explored acute (Kumar et al., 2000) [22]. Laparoscopic cholecystectomy is approximately safe and could be effective for peads. Cholecystectomy with symptomatic gallstones considered the standard for peads (Peter et al., 2008).

Gallbladder (A Cancer)

Cancer of gallbladder is diagnosis at the advance stage, late-time finding and the anatomical features; gallbladder as a low quantity of serosa culminates in a rather dismal prognosis (Lai & Lau, 2008). Survival ratio with advanced stage is six months; five percent ratio of life survival with advance stage is five years (Levy et al., 2001). Potential cure though cholecystectomy is offered by initial gallbladder's cancer (confined to the mucosa). Eighty percent of gallbladder cancers originate from the fundus (sixty percent), body (thirty percent), or neck (ten percent). The basis is genetically or elicited by chronic gallbladder inflammation, even cholelithiasis (Lazcano-Ponce et al., 2001).

Use of Medicine

The long term use of statin and other lipid lowering agents are also responsible for gall stone formation in the western world. Age is the risky factor, female gender, obesity plus median age of women's for gallstone disease was 58 years (68.8%) while for male population it was less then female. Current statin users were (4.8%) in control and (5.4%) in cases the former user of statin is (1.8%) and gallstone disease was (2.3%). Formation of gallstone and statin has a strong relation which is

reported by this study (Erichsen R et al., 2010) [16]. Uses of some drugs, oral contraceptives, thiazide etc., are also responsible for gallstone formation (Targher and Byrne 2015). The use of oral contraceptives, illiteracy (21.98%), leafy vegetables, and consumption of junk foods, or green chilies (14.5) are the risk factors (Channa et al. 2007). [12]

Post-Operative Complications

The function of gallbladder is bile storage from liver bile discharge from gallbladder to duodenum for food digestion. One of postoperative complication of gastrectomy is gallstone formation. The contractility and systolic function of gall bladder become reduces after gastrectomy. The larger resection of stomach, dissection of lymph node is reported for high incidence of gallstone formation. After gastrectomy the humoral factor of gallbladder is disturbed due to loss of a peptide hormone (cholecystokinin) absence in a result of duodenum dissection (Chen et al., 2014b). [14]

Statement of Problem

Gallstone is a worldwide disease. It is a common and costly clinical condition in the adult population of Pakistan. The best method is epidemiological studies to measure accuracy and determine prevalence of gallbladder stone and to identification the risky factors in a given population under the study. This is a hospital based case-controlled study. While population of the study was comprised upon the patients undergoing gallstone treatment during study specific time duration .Case-controlled method is adopted to give inference the study. The indoor and outdoor of hospitalized patients with gall stone disease, cholecystitis within probable time of study duration in Shalamar hospital Lahore has been selected as cases and against each case two Controls were selected on the basis of with no record of gallstone. The control was match with cases by age, sex and region of residence. The sample was constituted of total of 370 (200 cases and 170 control) patients. Self-administered questionnaire is used to obtain data from teach case and control. To minimize the selection bias the sample is collected through random sampling method. SPSS v.20s.0 is used to analyze the data and explore the association chi-square test was applied among different variables. Current study is made attempt to evaluate the risk of lipid lowering agents in producing gallstones in a hospital based case-control study.

Methodology

Target Population

This was a hospital based case-control study while population of the patients was comprised upon all the patients undergoing gallstone treatment during study's specific time duration.

Study Duration

The study was conducted in a time span of one year 2017.

Study Design

This was a hospital based case-control study design.

Study Location

The study was carried out in a tertiary care hospital named Shalamar Hospital located at Lahore.

Sample Size

The sample was constituted of total 370patients. 100 cases and 270 controls were selected for this study the patient ratio was 1:2. For the purpose of conducting a case-control study, sample was divided into two groups i.e. one for cases and the other for control. Sample was selected on the basis of inclusion and exclusion criteria. Self-administered questionnaire was used to obtain data from each case and control. To minimize the selection bias the sample was collected through random sampling method.

Case Definition

These were the indoor- and outdoor hospitalized patients with gall stone disease, cholecystitis which were diagnosed by ultrasonography positive and clinical findings within probable time of the study duration in shalamar hospital Lahore.

Control Definition

For each case two Controls were selected on the basis of with no record of gallstone by ultrasonography and clinical history of gall stone Disease. The controls were matched with cases by age, sex and region of residence**.**

Inclusion Criteria

These were the patients with gallstone diagnosed with ultrasonography and clinical findings for gallstone treatment in the hospital.

Exclusion Criteria

The participant of study were excluded who were pre-existing cases of cholelithiasis, with cholecystectomy, with liver diseases or bile duct abnormalities before the probable time of study or was not agreed for.

Overall Data Collection Procedure

Informed consents were drawn before collecting of data. The data were collected through pre designed self-administered questionnaire by interviewing each patient with gall bladder of general surgical unit of Shalamar hospital Lahore within probable

time of study, included there address, occupational and geographical risk factors.

Data Analysis and Statistical Designs

Data were analyzed through SPSS v.20 to explore the association chi-square test was applied among different variables. The statistical method to analyzed assessment of risk factor the odds ratio was used to express the associations between variables.

Chi-Square Test

A chi-squared test, also written as test, is any statistical hypothesis test wherein the sampling distribution of the test statistic is a chi-squared distribution when the null hypothesis is true. Without other qualification, 'chi-squared test' often is used as short for Pearson's chi-squared test. The chi-squared test is used to determine whether there is a significant difference between the expected frequencies and the observed frequencies in one or more categories.

Odds Ratio

An odds ratio (OR) is a measure of association between an exposure and an outcome. The OR represents the odds that an outcome will occur given a particular exposure, compared to the odds of the outcome occurring in the absence of that exposure. Odds ratios are most commonly used in case-control studies; however they can also be used in cross-sectional (Szumilas2010).

Details of Demographic and Clinical Factors of Participants

Gender, age, location, marital status, education, occupation, monthly income, past surgery history, previous treatment, history of hypothyroidism, oral contraceptive usage in the past, age at full term pregnancy, current and past lipid lowering agents usage, smoking history, BMI, diabetes mellitus, hypertension, current and past use of oral contraceptives,, use of any homeopathic medicine, number of parity, intake of various type of foods like soft drinks, dairy products, ketchup, water, clinical indications like metabolic syndrome, nausea, vomiting, jaundice, fever, weight loss, abdominal distention, salty sweat, malaise, pain, ultrasonography,and blood investigations are HCV, HBV, AST, cholesterol level, serum Amylase, Alkaline phosphate, ALT, serum Creatinine, serum Bilirubin were also examined during study session for checking the participants' previous and current association and background of the gallstone disease. Details are presented below"

BMI

The body mass index was calculated for the patients undergoing gall stone checkups by using the formula:

BMI= Kg/m^2

Hypertension

A blood pressure reading greater than or equal to 140 mmHg (systolic) or greater than or equal to 90 mmHg (diastolic) and patients on antihypertensive drugs were considered to be hypertension positive.

Diabetes Mellitus

Patients with fasting glucose levels more than 90-110 and random glucose levels more than 110-140 or patients on hypoglycemic drugs were considered to be positive.

Smoking

Patients with active smoking were considered to be positive.

Obesity

Patients with body mass index more than 24.5 were considered to be obese.

Serum Creatinine

Patient with serum Creatinine>200 milimicromol/L preoperatively was considered to be positive.

Pregnancy, Parity and Contraceptives

The female who are expecting a baby and the rank of their expected baby or either they had ever or currently using contraceptive medicines if we're not pregnant at the moment.

Other Laboratory Tests

The other related laboratory tests for gallstone diagnostics were performed on (automated chemistry analyzer) AU 480 fully automatic machine.

Results

This study was based on case-control methodology of data collection and was carried out on a time span of one year approximately in 2017. It was a case-control study and Odds ratio scores were calculated to predict the risk of generation of gallstone among participants who were using lipid lowering agents in Pakistani scenario. The indoor and outdoor of hospitalized patients with gall stone disease, cholecystitis within probable time of study duration in Shalamar hospital Lahore were selected as cases and against each case two Controls were selected on the basis of with no record of gallstone. In this study 1:2 sample sized were achieved and 70 controls were extra added to the study. So therefore the total number of controls is270 and the total number of participants was 370.The control

was matched with cases by age, sex and region of residence**.** The sample was constituted total of 370patients. 100 cases and 270 controls were selected for this study making a ratio of 1:2. The patients were selected through random sampling technique.

Graphical Distribution of Patients Demographics

The patients demographic distributions upon various variables were distributed among cases and control group, following figures were demonstrated on their frequency percentages.

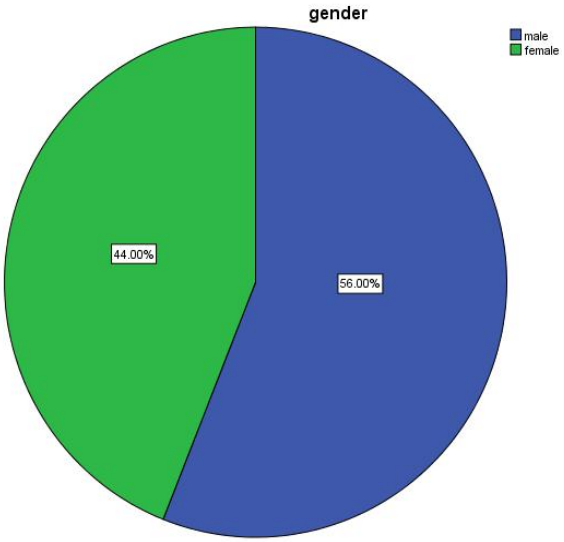

Figure 1: Gender-wise Distribution of Cases

Out of 100 cases 44% were female and 56% were male.

Pre-diagnostic Labs of Control Group

Mean scores and Standard deviation of lab reports on various related blood and urine tests of patients of control groups were calculated. The summary is presented below:

Table 1: Clinical Lab Reports of Control group

Labs	N	Normal	Below Normal/Borderline	Above Normal	Mean	St.Deviation
Hepatitis C	270	215	16	39	1.9148	.44405
Hepatitis B	270	245	10	15	1.9815	.30429
AST	270	96	26	148	2.4519	.66476
Cholesterol level	270	43	34	193	2.5889	.70412
Serum Amylase	270	84	136	50	1.6815	.76787
Alkaline phosphate	270	68	20	182	2.6000	.62417
ALT	270	98	47	125	2.2889	.74541
Serum Creatinine	270	108	63	99	2.1333	.76445
Serum Bilirubin	270	26	116	128	2.0444	.95136

Table 2: Clinical Lab Reports of Cases on Diagnosis of Gallstone

Labs	N	Normal /negative	Below Normal/Borderline	Above Normal/positive	Mean	St.Deviation
Hepatitis C	100	52	19	29	1.9000	.68902
Hepatitis B	100	85	-	15	1.8500	.35887
AST	100	26	4	70	2.6600	.55450
Cholesterol level	100	18	01	81	2.800	.42640
Serum Amylase	100	35	43	22	1.7900	.78232
Alkaline phosphate	100	28	02	70	2.6800	.51010
ALT	100	44	01	55	2.5400	.52068
Serum Creatinine	100	58	36	06	1.7000	.57735
Serum Bilirubin	100	15	69	15	1.4545	.74605

Chi-Square and Odds Ratio Examination on Risk Factors and Outcomes of both Groups

For the purpose of examining differences and Odds of getting gallstone among both control group and cases Chi-square with further odds ratio and risks were calculated on SPSS and summarize odds of cases over control as well.

1. Analyzing AST level as an indicator of Gall Stone Formation

In this section AST level in the blood was analyzed in terms of past and current exposure of lipid lowering agents among patients, i.e. high AST level among patients exposing to lipid lowering agents depicts an indicator of gall stone formation.

Table 3: Cross-tabulation on Past use of Lipid Lowering Agents vs. AST Level in Control Group

		AST		Total
		Normal	above normal	
Lipid lowering agents in the past	Yes	90	92	182
	%	49.5%	50.5%	100.0%
	No	33	55	88
	%	37.5%	62.5%	100.0%
Total	Count	123	147	270
	%	45.6%	54.4%	100.0%

The above table indicated that 50% of the control patients with past usage of lipid lowering agents have elevated AST level while 62% did not have elevated level of AST.

Table 4: Chi-square Examination on Past Lipid Lowering Agent Usage and AST Level of Control Group

Chi-Square Tests					
	Value	Df	Asymp. Sig. (2-sided)	Exact Sig. (2-sided)	Exact Sig. (1-sided)
Pearson Chi-Square	3.416	1	0.065		
Continuity Correction	2.951	1	0.086		
Likelihood Ratio	3.445	1	0.063		
Fisher's Exact Test				0.069	0.043
Linear-by-Linear Association	3.403	1	0.065		
N of Valid Cases	270				

a. 0 cells (.0%) have expected count less than 5. The minimum expected count is 40.09.
b. Computed only for a 2x2 table

The above chi-square value p= 0.065 indicated that there were no significant association between past usage of lipid lowering agents with that of elevated AST level among control group patients.

Table 5: Odds Ratio and Risk Estimation of Lipid Lowering Agent Past Usage on AST Level of Control Group Patients

Risk Estimate			
		95% Confidence Interval	
	Value	Lower	Upper
Odds Ratio for lipid lowering agents in the past (yes / no)	1.630	0.969	2.743
For cohort AST = normal	1.319	0.970	1.793
For cohort AST = above normal	.809	0.651	1.004
N of Valid Cases	270		

The above table indicated that Odds Ratio value for lipid lowering agent usage for yes over no had OR value = 1.63 i.e. OR > 1; indicating a risk of AST elevation.

Table 6: Cross-Tabulation on Past Use of Lipid Lowering Agents vs. AST Level in Cases Group

			AST		Total
			Normal	above normal	
Lipid lowering agents in the past	Yes	Count	25	44	69
		% within lipid lowering agents in the past	36.2%	63.8%	100.0%
	No	Count	5	26	31
		% within lipid lowering agents in the past	16.1%	83.9%	100.0%
Total		Count	30	70	100

The above table indicated that 63.8% of the cases patients with past usage of lipid lowering agents have elevated AST level while 83.9% did not have elevated level of AST.

Table 7: Chi-Square Examination on Past Lipid Lowering Agent Usage and AST Level of Cases Group

Chi-Square Tests					
	Value	Df	Asymp. Sig. (2-sided)	Exact Sig. (2-sided)	Exact Sig. (1-sided)
Pearson Chi-Square	4.116	1	0.042		
Continuity Correction	3.215	1	0.073		
Likelihood Ratio	4.427	1	0.035		
Fisher's Exact Test				0.059	0.034
Linear-by-Linear Association	4.075	1	0.044		
N of Valid Cases	100				

a. 0 cells (.0%) have expected count less than 5. The minimum expected count is 9.30.The above table indicated a chi-square value i.e. p= 0.042 showed no significant association of past lipid lowering agent usage with that of AST level of cases.

Table 8: Odds Ratio and Risk Estimation of Lipid Lowering Agent on AST Level of Cases Group Patients

Risk Estimate			
		95% Confidence Interval	
	Value	Lower	Upper
Odds Ratio for lipid lowering agents in the past (yes / no)	2.955	1.008	8.662
For cohort AST = normal	2.246	0.949	5.317
For cohort AST = above normal	0.760	0.601	0.962

The above table indicated that Odds Ratio value for lipid lowering agent usage for yes over no had OR value = 2.955, i.e. OR> 1, indicating high chances of developing risk of AST elevation in cases.

Summary of the Odds Ratio of Past Exposure and Prevalence Risk of High AST level

The following table states the Odds of the exposure and the risk of high AST level among the patients of the study.

Table 9: Odds Ratio and Prevalence Risk of High AST level

Risk Estimate			
		95% Confidence Interval	
	Value	Lower	Upper
Odds Ratio for lipid lowering agents in the past among patients (cases / control)	1.81	1.04	3.157
Odds Ratio for lipid lowering agents in the past among cases (yes / no)	2.955	1.008	8.662
Odds Ratio for lipid lowering agents in the past among control (yes / no)	1.630	0.969	2.743

The above table indicated the Odds Ratio for patients (cases / control) equals to 1.81, depicting that OR > 1 indicating that past use of lipid lowering agents are somewhat a risk factor of elevated AST level among patients, while AST level is an indicator of gallstone disease.

Table 10: Cross-Tabulation on Current Use of Lipid Lowering Agents vs. AST Level in Control Group

			AST		
			normal	above normal	Total
Current use of lipid lowering agents	Yes	Count	99	103	202
		% within current use of lipid lowering agents	49.0%	51.0%	100.0%
	No	Count	24	44	68
		% within current use of lipid lowering agents	35.3%	64.7%	100.0%
Total		Count	123	147	270
		% within current use of lipid lowering agents	45.6%	54.4%	100.0%

The above table indicated that 51% of the control patients with current usage of lipid lowering agents have elevated AST level while 64.7% did not have elevated level of AST.

Table 11: Chi-square Examination on Current Lipid Lowering Agent Usage and AST Level of Control Group

Chi-Square Tests					
	Value	Df	Asymp. Sig. (2-sided)	Exact Sig. (2-sided)	Exact Sig. (1-sided)
Pearson Chi-Square	3.859	1	0.049		
Continuity Correction	3.326	1	0.068		
Likelihood Ratio	3.913	1	0.048		
Fisher's Exact Test				0.067	0.034
Linear-by-Linear Association	3.844	1	0.050		
N of Valid Cases	270				

a. 0 cells (.0%) have expected count less than 5. The minimum expected count is 30.98.
b. Computed only for a 2x2 table

The above table indicated a chi-square value i.e. p= 0.049 showed no significant association of current lipid lowering agent usage with that of AST level of control patients.

Table 12: Odds Ratio and Risk Estimation for Current use of Lipid Lowering Agents and AST Level among Control Group

Risk Estimate			
		95% Confidence Interval	
	Value	Lower	Upper
Odds Ratio for current use of lipid lowering agents (yes / no)	1.762	0.998	3.112
For cohort AST = normal	1.389	0.977	1.973
For cohort AST = above normal	0.788	0.631	0.983

The above table indicated that Odds Ratio value for lipid lowering agent usage for yes over no had OR = 1.762, i.e. OR > 1, indicating a risk of AST elevation in control patients.

Table 13: Cross-Tabulation on Current Use of Lipid Lowering Agents vs. AST Level in Cases Group

			AST		
			Normal	above normal	Total
Current use of lipid lowering agents	Yes	Count	27	48	75
		% within current use of lipid lowering agents	36.0%	64.0%	100.0%
	No	Count	3	22	25
		% within current use of lipid lowering agents	12.0%	88.0%	100.0%
Total		Count	30		100

The above table indicated that 64% of the cases patients with current usage of lipid lowering agents have elevated AST level while 88% did not have elevated level of AST.

Table 14: Chi-Square Examination on Current Lipid Lowering Agent Usage and AST Level of Cases Group

Chi-Square Tests					
	Value	Df	Asymp. Sig. (2-sided)	Exact Sig. (2-sided)	Exact Sig. (1-sided)
Pearson Chi-Square	5.143	1	0.023		
Continuity Correction	4.063	1	0.044		
Likelihood Ratio	5.814	1	0.016		
Fisher's Exact Test				0.025	0.018
Linear-by-Linear Association	5.091	1	0.024		
N of Valid Cases	100				

a. 0 cells (.0%) have expected count less than 5. The minimum expected count is 7.50.
b. Computed only for a 2x2 table

The above table indicated a chi-square value i.e. p= 0.023 showed a significant association of current lipid lowering agent usage with that of AST level of cases patients.

Table 15: Odds Ratio and Risk Estimation for Current use of Lipid Lowering Agents and AST Level among Cases

Risk Estimate			
		95% Confidence Interval	
	Value	Lower	Upper
Odds Ratio for current use of lipid lowering agents (yes / no)	4.125	1.130	15.063
For cohort AST = normal	3.000	0.995	9.045
For cohort AST = above normal	0.727	0.582	0.909
N of Valid Cases	100		

The above table indicated that Odds Ratio value for lipid lowering agent usage for yes over no had OR = 4.125, i.e. OR > 1, indicating a high risk of AST elevation in cases patients.

Summary of Odds ratio of Current Exposure and Prevalence Risk of AST level

The following table states the Odds of the exposure and the risk of high AST level among the patients of the study.

Table 16: Odds Ratio and Prevalence Risk of High AST Level

Risk Estimate			
		95% Confidence Interval	
	Value	Lower	Upper
Odds Ratio for current use of lipid lowering agents among patients (cases / control)	2.341	1.132	4.840
Odds Ratio for current use of lipid lowering agents among cases (yes / no)	4.125	1.130	15.063
Odds Ratio for current use of lipid lowering agents among control (yes / no)	1.762	0.998	3.112

The above table indicated the Odds Ratio for patients (cases / control) currently using lipid lowering agents equals to 2.341, depicting that OR > 1 indicating that current use of lipid lowering agents is a risk factor of elevated AST level among patients, while AST level is an indicator of gall stone disease.

2. Analyzing Alkaline Phosphate Level as An Indicator of Gall Stone Formation

In this section Alkaline Phosphate level in the blood would be analyzed in terms of past and current exposure of lipid lowering agents among patients, i.e. high alkaline phosphate level among patients exposing to lipid lowering agents depicts an indicator of gall stone formation.

Table 17: Cross-Tabulation on Past Use of Lipid Lowering Agents vs. Alkaline Phosphate Level in Control Group

		Alkaline phosphate		Total
		Normal	above normal	
Lipid lowering agents in the past	Yes	56	125	181
	%	30.9%	69.1%	67.04%
	No	32	56	89
	%	37.9%	62.1%	32.96%
Total		89	181	270

The above table indicated that 69.1% of the control patients with past usage of lipid lowering agents have elevated alkaline phosphate level while 30.9% did not have elevated level of alkaline phosphate. Furthermore, those who did not used lipid lowering agents in the past also had (62.1%) above normal level of alkaline phosphate in their blood.

Table 18: Chi-square Examination on Past Lipid Lowering Agent Usage and Alkaline Phosphate Level of Control Group

	Value	Df	Asymp. Sig. (2-sided)	Exact Sig. (2-sided)	Exact Sig. (1-sided)
Pearson Chi-Square	0.792	1	0.374		
Continuity Correction	0.564	1	0.453		
Likelihood Ratio	0.785	1	0.376		
Fisher's Exact Test				0.407	0.226
Linear-by-Linear Association	0.789	1	0.375		
N of Valid Cases	270				

a. 0 cells (.0%) have expected count less than 5. The minimum expected count is 28.79.
b. Computed only for a 2x2 table

The above table indicated a chi-square value i.e. p= 0.792 showed no significant association of past lipid lowering agent usage with that of alkaline phosphate level of control group patients.

Table 19: Odds Ratio and Risk Estimation for Past use of Lipid Lowering Agents and Alkaline Phosphate Level among Control Group

Risk Estimate			
	Value	95% Confidence Interval	
		Lower	Upper
Odds Ratio for lipid lowering agents in the past (yes / no)	0.784	0.458	1.341
For cohort alkaline phosphate = normal	0.851	0.598	1.210
For cohort alkaline phosphate = above normal	1.085	0.901	1.307
N of Valid Cases	270		

The above table indicated that Odds Ratio value for lipid lowering agent usage for yes over no had OR value = 0.784,i.e. OR<1, indicating no association of developing risk of alkaline phosphate elevation in control group patients.

Table 20: Cross-tabulation on Past Use of Lipid Lowering Agents vs. Alkaline Phosphate Level in Cases Group

		Alkaline phosphate		Total
		Normal	above normal	
Lipid lowering agents in the past	Yes	20	49	69
	%	28.9%	71.1%	100%
	No	10	21	31
	%	32.2%	67.8%	100%
Total		30	70	100

The above table indicated that 71.1% of the cases patients with past usage of lipid lowering agents have elevated alkaline phosphate level while 28.9% did not have elevated level of alkaline phosphate.

Table 21: Chi-square Examination on Past Lipid Lowering Agent Usage and Alkaline Phosphate Level of Cases Group

	Value	Df	Asymp. Sig. (2-sided)	Exact Sig. (2-sided)	Exact Sig. (1-sided)
Pearson Chi-Square	0.109	1	0.741		
Continuity Correction	0.009	1	0.925		
Likelihood Ratio	0.108	1	0.742		
Fisher's Exact Test				0.815	0.458
Linear-by-Linear Association	0.108	1	0.742		
N of Valid Cases	100				

a. 0 cells (.0%) have expected count less than 5. The minimum expected count is 9.30.
b. Computed only for a 2x2 table

The above table indicated a chi-square value i.e. p= 0.741 showed no significant association of past lipid lowering agent usage with that of alkaline phosphate level of cases patients.

Table 22: Odds Ratio and Risk Estimation for Past use of Lipid Lowering Agents and Alkaline Phosphate Level among Cases Group

	Value	95% Confidence Interval	
		Lower	Upper
Odds Ratio for lipid lowering agents in the past (yes / no)	0.857	0.343	2.140
For cohort alkaline phosphate = normal	0.899	0.479	1.687
For cohort alkaline phosphate = above normal	1.048	0.788	1.395
N of Valid Cases	100		

The above table indicated that Odds Ratio value for lipid lowering agent usage for yes over no had OR value = 0.857, i.e. OR < 1, indicating no association with developing risk of alkaline phosphate elevation in cases patients.

Summary of the Odds Ratio of Past Exposure and Prevalence Risk of High Alkaline phosphate level

The following table states the Odds of the past exposure and the risk of high alkaline phosphate level among the patients of the study.

Table 23: Odds Ratio and Prevalence Risk of High Alkaline Phosphate Level

Risk Estimate			
		95% Confidence Interval	
	Value	Lower	Upper
Odds Ratio for lipid lowering agents in the past among patients (cases / control)	1.093	0.748	1.595
Odds Ratio for lipid lowering agents in the past among cases (yes / no)	0.857	0.343	2.140
Odds Ratio for lipid lowering agents in the past among control (yes / no)	0.784	0.458	1.341

The above table indicated the Odds Ratio for patients (cases / control) equals to 1.093, depicting that OR > 1 indicating that past use of lipid lowering agents are somewhat a risk factor of elevated Alkaline phosphate level among patients, while Alkaline phosphate level is an indicator of gall stone formation.

Table 24: Cross-Tabulation on Current Use of Lipid Lowering Agents vs. Alkaline Phosphate Level in Control Group

		Alkaline phosphate		
		normal	above normal	Total
Current use of lipid lowering agents	yes	63	138	201
	%	31.4	68.6	74.4
	no	25	43	69
	%	36.2	62.3	25.6
Total		89	181	270

The above table indicated that 68.6% of the control patients with current usage of lipid lowering agents have elevated alkaline phosphate level while 31.4% did not have elevated level of alkaline phosphate. Furthermore, those who are not using lipid lowering agents currently also had elevated level of alkaline phosphate level (62.3%) in their blood samples.

Table 25: Chi-square Examination on Current Lipid Lowering Agent Usage and Alkaline Phosphate Level of Control Group

	Value	Df	Asymp. Sig. (2-sided)	Exact Sig. (2-sided)	Exact Sig. (1-sided)
Pearson Chi-Square	0.678	1	0.410		
Continuity Correction	0.455	1	0.500		
Likelihood Ratio	0.670	1	0.413		
Fisher's Exact Test				0.455	0.249
Linear-by-Linear Association	0.676	1	0.411		
N of Valid Cases	270				

a. 0 cells (.0%) have expected count less than 5. The minimum expected count is 22.25.
b. Computed only for a 2x2 table

The above table indicated a chi-square value i.e. p= 0.678 showed no significant association of current lipid lowering agent usage with that of alkaline phosphate level of control patients.

Table 26: Odds Ratio and Risk Estimation for Current use of Lipid Lowering Agents and Alkaline Phosphate Level among Control Group

	Value	95% Confidence Interval	
		Lower	Upper
Odds Ratio for current use of lipid lowering agents (yes / no)	0.785	0.441	1.397
For cohort alkaline phosphate = normal	0.853	0.587	1.238
For cohort alkaline phosphate = above normal	1.086	0.885	1.331
N of Valid Cases	270		

The above table indicated that Odds Ratio value for current lipid lowering agent usage for yes over no had OR value = 0.785, i.e. OR< 1, indicating no association with the developing risk of alkaline phosphate elevation in control patients.

Table 27: Cross-Tabulation on Current use of Lipid Lowering Agents vs. Alkaline Phosphate Level in Cases Group

		Alkaline phosphate		Total
		normal	above normal	
Current use of lipid lowering agents	yes	22	53	75
	%	29.3%	70.7%	75.0%
	No	8	17	25
	%	32.0%	68.0%	25.0%
Total		30	70	100

The above table indicated that 70.7% of the cases patients with current usage of lipid lowering agents have elevated alkaline phosphate level while 29.3% did not have elevated level of alkaline phosphate. Furthermore, those who are not using lipid lowering agents currently also had elevated level of alkaline phosphate level (68.0%) in their blood samples.

Table 28: Chi-Square Examination on Current Lipid Lowering Agent Usage and Alkaline Phosphate Level of Cases Group

	Value	Df	Asymp. Sig. (2-sided)	Exact Sig. (2-sided)	Exact Sig. (1-sided)
Pearson Chi-Square	0.063	1	0.801		
Continuity Correction	0.000	1	1.000		
Likelihood Ratio	0.063	1	0.802		
Fisher's Exact Test				0.805	0.493
Linear-by-Linear Association	0.063	1	0.802		
N of Valid Cases	100				

a. 0 cells (.0%) have expected count less than 5. The minimum expected count is 7.50.
b. Computed only for a 2x2 table

The above table indicated a chi-square value i.e. p= 0.063 showed no significant association of current lipid lowering agent usage with that of alkaline phosphate level of cases patients.

Table 29: Odds Ratio and Risk Estimation for Current use of Lipid Lowering Agents and Alkaline Phosphate Level among Cases Group

	Value	95% Confidence Interval	
		Lower	Upper
Odds Ratio for current use of lipid lowering agents (yes/ no)	0.882	0.332	2.342
For cohort alkaline phosphate = normal	0.917	0.469	1.793
For cohort alkaline phosphate = above normal	1.039	0.765	1.411
N of Valid Cases	100		

The above table indicated that Odds Ratio value for current lipid lowering agent usage for yes over no had OR value = 0.882, i.e. OR< 1, indicating no association with developing risk of alkaline phosphate elevation in control patients.

Summary of the Odds Ratio of Current Exposure and Prevalence Risk of High Alkaline phosphate level

The following table states the Odds of the current exposure and the risk of high alkaline phosphate level among the patients of the study.

Table 30: Odds Ratio and Prevalence Risk of High Alkaline Phosphate Level

Risk Estimate			
	Value	95% Confidence Interval	
		Lower	Upper
Odds Ratio for current use of lipid lowering agents among patients (cases / control)	1.12	0.752	1.676
Odds Ratio for current use of among patients (yes / no)	0.882	0.332	2.342
Odds Ratio for current use of lipid lowering agents among patients (yes / no)	0.785	0.441	1.397

The above table indicated the Odds Ratio for patients (cases / control) equals to 1.12, depicting that OR > 1 indicating that current use of lipid lowering agents are somewhat a risk factor of elevated Alkaline phosphate level among patients, while Alkaline phosphate level is an indicator of gall stone formation.

3 Analyzing Serum Bilirubin level as an indicator of Gall Stone Formation

In this section Serum Bilirubin level in the blood would be analyzed in terms of past and current exposure of lipid lowering agents among patients, i.e. high serum bilirubin level among patients exposing to lipid lowering agents depicts an indicator of gall stone formation.

Table 31: Cross-Tabulation on Past Use of Lipid Lowering Agents vs. Serum Bilirubin Level in Control Group

		Serum Bilirubin		
		normal	above normal	Total
Lipid lowering agents in the past	Yes	92	89	181
	%	50.8%	49.1%	67.0%
	No	48	40	89
	%	53.9%	44.9%	33.0%
Total		140	129	270

The above table indicated that 50.8% of the cases patients with current usage of lipid lowering agents have normal level of serum bilirubin in their blood samples while 49.1% have elevated level that showed merely no difference. Furthermore, those who were not using lipid lowering agents in past (53.9%) also had normal level of serum bilirubin in their blood samples.

Table 32: Chi-square Examination on Past Lipid Lowering Agent Usage and Serum Bilirubin Level of Control Group

	Value	Df	Asymp. Sig. (2-sided)	Exact Sig. (2-sided)	Exact Sig. (1-sided)
Pearson Chi-Square	0.328	1	0.567		
Continuity Correction	0.196	1	0.658		
Likelihood Ratio	0.328	1	0.567		
Fisher's Exact Test				0.604	0.329
Linear-by-Linear Association	0.327	1	0.568		
N of Valid Cases	270				

a. 0 cells (.0%) have expected count less than 5. The minimum expected count is 42.20.
b. Computed only for a 2x2 table

The above table indicated a chi-square value i.e. p= 0.328 showed no significant association of past lipid lowering agent usage with that of serum bilirubin level of controlled patients.

Table 33: Odds Ratio and Risk Estimation for Past Use of Lipid Lowering Agents and Serum Bilirubin Level among Control Group

	Value	95% Confidence Interval	
		Lower	Upper
Odds Ratio for lipid lowering agents in the past (yes / no)	0.861	0.517	1.436
For cohort serum Bilirubin = normal	0.932	0.734	1.183
For cohort serum Bilirubin = above normal	1.082	0.824	1.421
N of Valid Cases	270		

The above table indicated that Odds Ratio value for past lipid lowering agent usage for yes over no had OR value= 0.861, i.e. OR < 1, indicating no association with the developing risk of serum bilirubin elevation in control patients.

Table 34: Cross-tabulation on Past Use of Lipid Lowering Agents vs. Serum Bilirubin Level in Cases Group

		Serum Bilirubin		
		normal	above normal	Total
Lipid lowering agents in the past	Yes	57	12	69
	%	82.6%	17.4%	69.0%
	No	28	3	31
	%	90.3%	9.7%	31.0%
Total		85	15	100

The above table indicated that 82.6% of the cases patients with past usage of lipid lowering agents have normal level of serum bilirubin in their blood samples while only 17.4% have elevated level. Furthermore, those who were not using lipid lowering agents in past also had normal level of serum bilirubin (90.3%) in their blood samples.

Table 35: Chi-square Examination on Past Lipid Lowering Agent Usage and Serum Bilirubin Level of Cases Group

	Value	df	Asymp. Sig. (2-sided)	Exact Sig. (2-sided)	Exact Sig. (1-sided)
Pearson Chi-Square	0.998	1	0.318		
Continuity Correction	0.485	1	0.486		
Likelihood Ratio	1.069	1	0.301		
Fisher's Exact Test				0.381	0.249
Linear-by-Linear Association	0.988	1	0.320		
N of Valid Cases	100				

The above table indicated a chi-square value i.e. p= 0.998 showed no significant association of past lipid lowering agent usage with that of serum bilirubin level of cases patients.

Table 36: Odds Ratio and Risk Estimation for Past Use of Lipid Lowering Agents and Serum Bilirubin Level among Cases Group

	Value	95% Confidence Interval	
		Lower	Upper
Odds Ratio for lipid lowering agents in the past (yes / no)	0.509	0.133	1.951
For cohort serum Bilirubin = normal	0.915	0.781	1.071
For cohort serum Bilirubin = above normal	1.797	0.546	5.919
N of Valid Cases	100		

The above table indicated that Odds Ratio value for past lipid lowering agent usage for yes over no had = 0.509, i.e. OR < 1, indicating no association with the developing risk of serum bilirubin elevation in cases patients.

Summary of the Odds Ratio of Past Exposure and Prevalence Risk of High Serum Bilirubin level

The following table states the Odds of the past exposure and the risk of high serum bilirubin level among the patients of the study.

Table 37: Odds Ratio and Prevalence Risk of High Alkaline Phosphate Level

Risk Estimate			
	Value	95% Confidence Interval	
		Lower	Upper
Odds Ratio for lipid lowering agents in the past among patients (cases / control)	0.591	0.257	1.358
Odds Ratio for lipid lowering agents in the past among cases (yes / no)	0.509	0.133	1.951
Odds Ratio for lipid lowering agents in the past among control (yes / no)	0.861	0.517	1.436

The above table indicated the Odds Ratio for patients (cases / control) equals to 0.591, depicting that OR < 1 indicating that past use of lipid lowering agents has no association with a risk factor of elevated serum bilirubin level among patients, while serum bilirubin level is an indicator of gall stone formation.

Table 38: Cross-tabulation on Current Use of Lipid Lowering Agents vs. Serum Bilirubin Level in Control Group

		Serum Bilirubin		
		normal	above normal	Total
Current use of lipid lowering agents	yes	101	100	201
	%	50.2%	49.8%	74.4%
	No	39	29	69
	%	56.5%	43.5%	25.6%
Total		140	129	270

The above table indicated that 84.0% of the control patients with current usage of lipid lowering agents have normal level of serum bilirubin in their blood samples while only 16.0% have elevated level. Furthermore, those who were not using lipid lowering agents currently also had normal level of serum bilirubin (88.0%) in their blood samples.

Table 39: Chi-square Examination on Current Lipid Lowering Agent Usage and Serum Bilirubin Level of Control Group

	Value	Df	Asymp. Sig. (2-sided)	Exact Sig. (2-sided)	Exact Sig. (1-sided)
Pearson Chi-Square	1.027	1	0.311		
Continuity Correction	0.763	1	0.383		
Likelihood Ratio	1.031	1	0.310		
Fisher's Exact Test				0.329	0.191
Linear-by-Linear Association	1.024	1	0.312		
N of Valid Cases	270				

a. 0 cells (.0%) have expected count less than 5. The minimum expected count is 32.61.
b. Computed only for a 2x2 table

The above table indicated a chi-square value i.e. p= 1.027 showed no significant association of current lipid lowering agent usage with that of serum bilirubin level of control patients.

Table 40: Odds Ratio and Risk Estimation for Current Use of Lipid Lowering Agents and Serum Bilirubin Level among Control Group

		95% Confidence Interval	
	Value	Lower	Upper
Odds Ratio for current use of lipid lowering agents (yes / no)	0.751	0.431	1.308
For cohort serum Bilirubin = normal	0.876	0.684	1.121
For cohort serum Bilirubin = above normal	1.167	0.857	1.588
N of Valid Cases	270		

The above table indicated that Odds Ratio value for current lipid lowering agent usage for yes over no had = 0.751, i.e. OR < 1, indicating no association with the developing risk of serum bilirubin elevation in control patients.

Table 41: Cross-tabulation on Current Use of Lipid Lowering Agents vs. Serum Bilirubin Level in Cases Group

		Serum Bilirubin		
		Normal	above normal	Total
Current use of lipid lowering agents	Yes	63	12	75
	%	84.0%	16.0%	75.0%

	No	22	3	25
	%	88.0%	12.0%	25.0%
Total		85	15	100

The above table indicated that 84.0% of the cases patients with current usage of lipid lowering agents have normal level of serum bilirubin in their blood samples while only 16.0% have elevated level. Furthermore, those who were not using lipid lowering agents currently also had normal level of serum bilirubin (88.0%) in their blood samples.

Table 42: Chi-Square Examination on Current Lipid Lowering Agent Usage and Serum Bilirubin Level of Cases Group

	Value	Df	Asymp. Sig. (2-sided)	Exact Sig. (2-sided)	Exact Sig. (1-sided)
Pearson Chi-Square	0.235	1	0.628		
Continuity Correction	0.026	1	0.872		
Likelihood Ratio	0.245	1	0.621		
Fisher's Exact Test				0.755	0.451
Linear-by-Linear Association	0.233	1	0.629		
N of Valid Cases	100				

The above table indicated a chi-square value i.e. p= 0.235 showed no significant association of current lipid lowering agent usage with that of serum bilirubin level of cases patients.

Table 43: Odds Ratio and Risk Estimation for Current Use of Lipid Lowering Agents and Serum Bilirubin Level among Cases Group

	Value	95% Confidence Interval	
		Lower	Upper
Odds Ratio for current use of lipid lowering agents (yes / no)	0.716	0.185	2.776
For cohort serum Bilirubin = normal	0.955	0.801	1.137
For cohort serum Bilirubin = above normal	1.333	0.409	4.345

The above table indicated that Odds Ratio value for current lipid lowering agent usage for yes over no had = 0.716, i.e. OR <1, indicating no association with the developing risk of serum bilirubin elevation in cases patients.

Summary of the Odds Ratio of Current Exposure and Prevalence Risk of High Serum Bilirubin level

The following table states the Odds of the current exposure and the risk of high serum bilirubin level among the patients of the study.

Table 44: Odds Ratio and Prevalence Risk of High Serum Bilirubin Level

Risk Estimate			
	Value	95% Confidence Interval	
		Lower	Upper
Odds Ratio for current use of lipid lowering agents among patients (cases / control)	0.953	0.429	2.123

Odds Ratio for current use of lipid lowering agents among patients (yes / no)	0.716	0.185	2.776
Odds Ratio for current use of lipid lowering agents among patients (yes / no)	0.751	0.431	1.308

The above table indicated the Odds Ratio for patients (cases / control) equals to 0.953, depicting that OR ≤ 1 indicating that current use of lipid lowering agents is not associated to a risk factor of elevated serum bilirubin level among patients, while serum bilirubin level is an indicator of gall stone formation.

Discussions

Nearly 20% of every second person have gallstone in developing countries and the cause of gallstones are still not fully known. Gallstones are one of the prominent diseases among gastrointestinal patients in Western countries too (Shaffer, 2006). There are two types of gallstones; they are either pigment gallstones or produced by high cholesterol levels. Risk of generating gallstones is high in females than in males. Previous studies had indicated that females with more parities and use of contraceptive medicines increase the risks of gallstone formation in them. Past studies had also talked about lipid lowering agents to be one of the risk increasing factors for producing gall stones. Current study attempted to evaluate the risk of lipid lowering agents in producing gallstones in a hospital based case-control study.

Findings of the Study

Inferential Findings

A) Risk of Elevated AST Level with past Exposure of Lipid Lowering Agents

The following inferential results are calculated in the study which describes that63.8% of the cases patients with past usage of lipid lowering agents have elevated AST level while 83.9% did not have elevated level of AST. And chi-square value i.e. p= 0.042 showed no significant association of past lipid lowering agent usage with that of AST level of cases. Odds Ratio value for lipid lowering agent usage for yes over no had OR value = 2.955, i.e. OR> 1, indicating high chances of developing risk of AST elevation in cases.

While In this study 50% of the control patients with past usage of lipid lowering agents have elevated AST level while 62% did not have elevated level of AST. The chi-square value p= 0.065 indicated that there were no significant association between past usage of lipid lowering agents with that of elevated AST level among control group Patients. The Odds Ratio value for lipid lowering agent usage for yes over no had OR value = 1.63 i.e. OR > 1; indicating a risk of AST elevation. While Odds Ratio for patients (cases / control) equals to 1.81, depicting that OR > 1 indicating that past use of lipid lowering agents are somewhat a risk factor of elevated AST level among patients, while AST level is an indicator of gall stone disease.

B) Risk of Elevated AST level in Current Exposure of Lipid Lowering Agents

For AST level With Current usage of Lipid Lowering Agents 64% of the cases patients with current usage of lipid lowering agents have elevated AST level while 88% did not have elevated level of AST.T he chi-square value i.e. p= 0.023 showed a significant association of current lipid lowering agent usage with that of AST level of cases patients. While The Cases Odds Ratio value for current lipid lowering agent usage for yes over no had OR = 4.125, i.e. OR > 1, indicating a high risk of AST elevation in cases patients. The AST Level in 51% of the control patients with current usage of lipid lowering agents have elevated AST level while 64.7% did not have elevated level of AST.

The Calculated chi-square value i.e. p= 0.049 showed no significant association of current lipid lowering agent usage with that of AST level of control patients.

While the Odds Ratio value for lipid lowering agent usage for yes over no had OR = 1.762, i.e. OR > 1, indicating a risk of AST elevation in control patients. Odds Ratio for patients (cases / control) currently using lipid lowering agents equals to 2.341, depicting that OR > 1 indicating that current use of lipid lowering agents is a risk factor of elevated AST level among patients, while AST level is an indicator of gall stone disease.

C) Risk of Elevated Alkaline Phosphate Level with past exposure of lipid lowering agents

In This Study 71.1% of the cases patients with past usage of lipid lowering agents have elevated alkaline phosphate level while 28.9% did not have elevated level of alkaline phosphate. The Calculated chi-square value i.e. p= 0.741 showed no significant association of past lipid lowering agent usage with that of alkaline phosphate level of cases patients. And Odds Ratio value for lipid lowering agent usage for yes over no had OR value = 0.857, i.e. OR < 1, indicating no association with developing risk of alkaline phosphate elevation in

cases patients. In this Study 69.1% of the control patients with past usage of lipid lowering agents have elevated alkaline phosphate level while 30.9% did not have elevated level of alkaline phosphate.

The chi-square value i.e. p= 0.792 showed no significant association of past lipid lowering agent usage with that of alkaline phosphate level of control group patients. And Odds Ratio value for lipid lowering agent usage for yes over no had OR value = 0.784, i.e. OR<1, indicating no association of developing risk of alkaline phosphate elevation in control group patients.

While Odds Ratio for patients (cases / control) equals to 1.093, depicting that OR > 1 indicating that past use of lipid lowering agents are somewhat a risk factor of elevated alkaline phosphate level among patients, while alkaline phosphate level is an indicator of gall stone formation.

D) Risk of Elevated Alkaline Phosphate with current exposure of Lipid Lowering Agents

In this Study 70.7% of the cases patients with current usage of lipid lowering agents have elevated alkaline phosphate level while 29.3% did not have elevated level of alkaline phosphate. The Calculated chi-square value i.e. p= 0.063 showed no significant association of current lipid lowering agent usage with that of alkaline phosphate level of cases patients. And Odds Ratio value for current lipid lowering agent usage for yes over no had OR value = 0.882, i.e. OR< 1, indicating no association with developing risk of alkaline phosphate elevation in control patients.

In control group of the study 68.6% of the control patients with current usage of lipid lowering agents have elevated alkaline phosphate level while 31.4% did not have elevated level of alkaline phosphate. And the chi-square value i.e. p= 0.678 showed no significant association of current lipid lowering agent usage with that of alkaline phosphate level of control patients. Odds Ratio value for current lipid lowering agent usage for yes over no had OR value = 0.785, i.e. OR< 1, indicating no association with the developing risk of alkaline phosphate elevation in control patients.

While the Odds Ratio for patients (cases / control) equals to 1.12, depicting that OR > 1 indicating that current use of lipid lowering agents are somewhat a risk factor of elevated alkaline phosphate level among patients, while alkaline phosphate level is an indicator of gall stone formation.

E) Risk of Elevated Serum Bilirubin with past exposure of Lipid lowering Agents

In This Study 82.6% of the cases patients with past usage of lipid lowering agents have normal level of serum bilirubin in their blood samples while only 17.4% have elevated level.

The Calculated chi-square value i.e. p= 0.998 showed no significant association of past lipid lowering agent usage with that of serum bilirubin level of cases patients. And the Odds Ratio value for past lipid lowering agent usage for yes over no had = 0.509, i.e. OR < 1, indicating no association with the developing risk of serum bilirubin elevation in cases patients.

Total number of control 50.8% of the control patients with past usage of lipid lowering agents have normal level of serum bilirubin in their blood samples while 49.1% have elevated level that showed merely no difference. S The calculated chi-square value i.e. p= 0.328 showed no significant association of past lipid lowering agent usage with that of serum bilirubin level of controlled patients.

And the Odds Ratio value for past lipid lowering agent usage for yes over no had OR value= 0.861, i.e. OR < 1, indicating no association with the developing risk of serum bilirubin elevation in control patients. While the Odds Ratio for patients (cases / control) equals to 0.591, depicting that OR < 1 indicating that past use of lipid lowering agents has no association with a risk factor of elevated serum bilirubin level among patients, while serum bilirubin level is an indicator of gall stone formation.

F) Risk of Elevated Serum Bilirubin with current exposure of Lipid Lowering Agents

According to calculated results Shows that 84.0% of the cases patients with current usage of lipid lowering agents have normal level of serum bilirubin in their blood samples while only 16.0% have elevated level. The chi-square value i.e. p= 0.235 showed no significant association of current lipid lowering agent usage with that of serum bilirubin level of cases patients. The calculated Odds Ratio value for current lipid lowering agent usage for yes over no had = 0.716, i.e. OR <1, indicating no association with the developing risk of serum bilirubin elevation in cases patients.

In this study 84.0% of the control patients with current usage of lipid lowering agents have normal level of serum bilirubin in their blood samples while only 16.0% have elevated level. And the chi-square value i.e. p= 1.027 showed no significant association of current lipid lowering agent usage with that of serum bilirubin level of control patients. The Results Shows That Odds Ratio value for current lipid lowering agent usage for yes over no had = 0.751, i.e. OR < 1, indicating no association

with the developing risk of serum bilirubin elevation in control patients.

While the Odds Ratio for patients (cases / control) equals to 0.953, depicting that OR ≤ 1 indicating that current use of lipid lowering agents is not associated to a risk factor of elevated serum bilirubin level among patients, while serum bilirubin level is an indicator of gall stone formation.

Table 45: Conclusion of the Study

The conclusion of the study is depicted in the following table:

Objectives	Hypothesis	Findings
1. To determine the risk factor of gallbladder, the disease	The use of Lipid lowering agents becomes a risk of gallstone in hospitalized patients of Lahore	In this study, AST, alkaline phosphate and serum bilirubin level in blood samples of patients were taken as an indicator of gall stone disease. In this light, • The study found that patients with past as well as current exposure of lipid lowering agents have elevated levels of AST and alkaline phosphate level. • But they don't have elevated levels of serum bilirubin.
2. To investigate and identify the association of risk factors		• There found a risk of elevated AST and alkaline phosphate levels in the blood samples of patients indicating an association of prevalence of gall stone formation with the past and current exposure of lipid lowering agents. • There found no risk of elevated serum bilirubin in the blood samples of patients indicating no association of prevalence of gall stone formation with the past and current exposure of lipid lowering agents.

The elevated risk ratios of AST and alkaline phosphate elevation with the past and current exposure of lipid lowering agents among patients indicate that it could be a cause of gall stone formation. But for the case of serum bilirubin which is also an indicator of gall stone formation, no prevalence could be found. Thus, it can be that there are chances that lipid lowering agents are a risk of gall stone formation among patients yet; further researches must be conducted to explore other factors of risk for the gall stone diseases with reference to demographics and habits of the patients.

Previous Findings Contradicting this Study

There are a number of past studies whose results were not in favor of the results of this study in many aspects. Some of such findings are depicted below: One of the studies stated about their current statin users that their adjusted odds ratios associating statin use with the occurrence of gallstone disease were 1.17 (95% confidence interval (CI): 1.06, 1.30) for those who had 1–4 prescriptions, 0.89 (95% CI: 0.80, 0.97) for those who had 5–19 prescriptions, and 0.76 (95% CI: 0.69, 0.84) for those who had ≥20 total prescriptions. In former users, the corresponding adjusted odds ratios were 1.24 (95% CI: 1.11, 1.39),

0.97 (95% CI: 0.86, 1.10), and 0.79 (95% CI: 0.64, 0.97), respectively. The use of other lipid-lowering drugs showed no similar association. In another study a total of 27,035 patients with cholecystectomy and 106,531 matched controls were identified, including 2396 patients and 8868 controls that had statin use. Compared with nonuse, current statin use (last prescription recorded within 90 days before the first-time diagnosis of the disease) was 1.0% for patients and 0.8% for controls (AOR, 1.10; 95% CI, 0.95-1.27) for 1 to 4 prescriptions; 2.6% vs. 2.4% (AOR, 0.85; 95% CI, 0.77-0.93) for 5 to 19 prescriptions, and 3.2% vs. 3.7% (AOR, 0.64; 95% CI, 0.59-0.70) for 20 or more prescriptions. The AORs for current use of statin defined as 20 or more prescriptions were similar (around 0.6) across age, sex, and body mass index categories, and across the statin class. Long-term use of statin was associated with a decreased risk of gallstones followed by cholecystectomy (Bodmer et al, 2009). [5]

Studies that endorse this Study's Findings

Past literature is also equipped with the studies that had associated lipid lowering agents, low cholesterol level and weight loss with that of more risk of gallstone formation. Some of those are enlisted below: Bush et al (1998) claimed that in individuals with low caloric diet, weight loss or bariatric surgery are associated with gallstone formation. Weight losses that exceed 1.5 kg/week following bariatric surgery increase the risk for stone formation.

The long term use of statin and other lipid lowering agents are also responsible for gall stone formation in the western world. In the risk factors included age, female gender, and obesity. the median age in women's for gallstone disease was 58 years (68.8%) while for male population it was less then female. current statin users were (4.8%) in control and (5.4%)in cases. the former user of statin was (1.8%) and gallstone disease was (2.3%). This study supported the association between the use of statin and gallstone formation (Erichsen, et al. 2010) [16]. The risk of gallstone formation increases with age, change in diet (high calorie food) or use of drugs (lipid lowering agents) being female 87% cases, and being married 78% cases of gallstone were found (Channa et al, 2007). [12]

Summary

Disease of gallbladder is commonly manifested as gallstone and gallbladder cancer. It's a common and costly clinical condition in the adult population of Pakistan. Gallstone disease remains a serious health concern for human beings, affecting millions of people throughout the world and in Pakistan as well. Gall stone or cholelithiasis is a common public health problem in Pakistan prevalence is high in female as compare to male. Chronic pain of upper quadrant of abdomen, dyspepsia, nausea, vomiting, fever and jaundice are the clinical features of gallstone disease.

To explore the gallstone formation risks among patients using lipid lowering agents and to investigate the association of risk factors in hospitalized patients in Lahore. The use of Lipid lowering agents becomes a risk of gallstone in hospitalized patients of Lahore.

The case control study had been carried out in a tertiary care hospital. Sample had been selected on the basis of inclusion and exclusion criteria. Self-administered questionnaire was used to obtain data from each case and control. To minimize the selection bias the sample had been collected through random sampling method.100 cases and 270 controls had been selected for this study. The sample ratio for this was 1:2.seventy (70) extra control was included in the study. Data had been analyzed through SPSS v.20.0 .to explore the association chi-square test had been applied among different variables. The statistical method to analyzed assessment of risk factor the odds ratio had been used to express the associations between variables. he outcomes of this study are able to help to estimate the risk of the prevalence of disease. The picture of this study helps for future planning of gallstone preventions and awareness of risk factors to stop gall stone formation and reduce financial burden of disease. The finding also recommends users of lipid lowering agents to use them with caution of risk of gallstone formation.

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Conflict of Interest

There is no conflict of interest between the authors.

References

1. Angelico, M. and Guardia, P.D., (2000). Hepatobiliary complications associated with total parenteral nutrition. Aliment. pharmacol. Ther., 14 (S2): 54-57.
2. Apstein, M. and Carey, M., (1996). Pathogenesis of cholesterol gallstones: a parsimonious hypothesis. Eur. J. of Med.Res., 26(5): 343-352.
3. Attili, A., Capocaccia, R., Carulli, N., Festi, D., Roda, E., Barbara, L., Capocaccia, L., Menotti, A., Okolicsanyi, L. and Ricci, G., (1997). Factors associated with gallstone disease in the MICOL experience. Hepatology, 26(4): 809-818.
4. Bansal, A., Akhtar, M. and Bansal, A.K., (2016). A clinical study: prevalence and management of cholelithiasis. Int. J. Surg., 1(3): 134-139.
5. Bodmer, M., Brauchli, Y.B., Krahenbuhl, S., Jick, S.S., Meier, C.R., (2009). Statin use and risk of gallstone disease followed by

cholecystectomy. Jama, 302(18): 2001-2007.
6. Bogue, C.O., Murphy, A.J., Gerstle, J.T., Moineddin, R. and Daneman, A., (2010). Risk factors, complications, and outcomes of gallstones in children: a single-center review. J. gastroenterol. Nutr., 50(3): 303-308.
7. Botham, K.M. and Bravo, E., (1995). The role of lipoprotein cholesterol in biliary steroid secretion. Studies with in vivo experimental models. J. Lipid Res., 34(1): 71-97.
8. Brown, M.S., Faust, J.R., Goldstein, J.L., Kaneko, I. and Endo A., (1987). Induction of 3-hydroxy-3-methylglutaryl coenzyme A reductase activity in human fibroblasts incubated with compactin (ML-236B), a competitive inhibitor of the reductase. J. Biol. Chem., 253(4): 1121-1128.
9. Buhman, K.K., Accad. M., Novak. S,, Choi. R.S., Wong, J.S., Hamilton, R.L., Turley, S. and Farese, Jr R.V., (2000). Resistance to diet-induced hypercholesterolemia and gallstone formation in ACAT2-deficient mice. Nat. Med., 6(12): 1341.
10. Carulli, N., De Leon, M.P., Zironi, F., Pinetti, A., Smerieri, A., Iori, R. and Loria, P., (1980). Hepatic cholesterol and bile acid metabolism in subjects with gallstones: comparative effects of short erm feeding of chenodeoxycholic and ursodeoxycholic acid. J. Lipid Res., 21(1): 35-43.
11. Channa, N.A., Khand, F.D., Bhanger, M.I. and Leghari, M.H., (2004). Surgical incidence of Cholelithiasis in Hyderabad and adjoining areas (Pakistan). Int. J. Med. Sci., 20(1): 13-17.
12. Channa, N.A., Khand, F.D., Khand, T.U., Leghari, M.H. and Memon, A.N., (2007). Analysis of human gallstones by Fourier Transform Infrared (FTIR). Int. J. Med. Sci., 23(4): 546.
13. Chen, X.J., Li, N., Huang, Y.D., Ren, S., Liu, F., Chen, L., Wang, Y. and Chen, M., (2014a). Factors for postoperative gallstone occurrence in patients with gastric cancer: a meta-analysis. Asian Pac. J .Cancer Prev., 15(2): 877-881.
14. Chen, Y., Kong, J. and Wu, S., (2014b). Cholesterol gallstone disease: focusing on the role of gallbladder. Lab. Invest., 95: 124.
15. Diehl, A., (1991). Epidemiology and natural history of gallstone disease. Gastrointest. Endosc., 20(1): 1-19.
16. Erichsen, R., Froslev, T., Lash, T.L., Pedersen, L. and Sorensen, H.T., (2010). Long-term statin use and the risk of gallstone disease: a population-based case-control study. Am. J. Epidemiol., 173(2): 162-170.
17. Horiuchi, I., Ohya, T., Tazuma, S., Mizuno, T., Takizawa, I. and Kajiyama, G., (1991). Effects of pravastatin (CS-514) on biliary lipid metabolism in patients with hyperlipidemia. Metabolism, 40(3): 226-230.
18. Hundal, R. and Shaffer, E.A., (2014). Gallbladder Cancer: Epidemiology and Outcome. Clinl. Epidemiol., 6: 99.
19. Jones, M., Pandak, W., Heuman, D., Chiang, J., Hylemon, P. and Vlahcevic, Z., (1993). Cholesterol 7 alpha-hydroxylase: evidence for transcriptional regulation by cholesterol or metabolic products of cholesterol in the rat. J. Lipid Res., 34(6): 885-892.
20. Ko, C.W., Beresford, S.A., Schulte, S.J. and Lee, S.P., (2008). Insulin resistance and incident gallbladder disease in pregnancy. Clin. Gastroenterol. Hepatol., 6(1): 76-81.
21. Kratzer, W., Mason, R.A. and Kachele, V., (1999). Prevalence of gallstones in sonographic surveys worldwide. JCU, 27(1): 1-7.
22. Kumar, R., Nguyen, K. and Shun, A., (2000). Gallstones and common bile duct calculi in infancy and childhood. ANZ Journal of Surgery, 70(3): 188-191.
23. Gupta, Ruchika and Agarwal, S.P., (2017). A Comparative Study of Cyber Threats in Emerging Economies. Globus: An International Journal of Management & IT, 8(2); 24-28, ISSN:0975-721X.
24. Agarwal, Nidhi and Kumar, Puneet, (2009). “Role of Information Technology in Education”, AICTE Sponsored National conference on Information Integrity & Supply chain Management Abstracts Proceeding, Book World Publisher, Dehradun, Pp. 18.
25. Agarwal, Nidhi and Kumar, Puneet, (2009). “Reflection on the new Innovations for maximizing the learning in Teacher of Mathematics”. International Journal Educational Herald, 38(2); 41, ISSN: 0974-0732.
26. Kumar, Puneet, (2003). “Weighted average charge for heterogeneous questionnaires and weighed entropy”. An International Journal of Business Research , pp 131-145.
27. Kumar, Puneet, (2008). “A Global Change in Education through Information Technology & Communication”. Gyanodaya : The Journal of Progressive Education’, pp 22-26.
28. Stinton, L.M. and Shaffer, E.A., (2012). Epidemiology of gallbladder disease: cholelithiasis and cancer. Gut Liver, 6(2): 172-187.
29. Zhu, L., Aili, A., Zhang, C., Saiding, A. and Abudureyimu, K., (2014). Prevalence of and risk factors for gallstones in Uighur and Han Chinese. World Journal of Gastroenterology, 20(40): 14942.