# Quantitative Analysis of Chemical Composition of Gallbladder Stones among Cholecystectomy of Iraqi Patients

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## Abstract

Gallstone disease is one of the major surgical problems in the population; it is probably related to diet, especially excessive consumption of meat. The objective of this study was to determine the quantitative analysis of chemical composition of gallbladder stones cholecystectomised patients. The chemical composition of gallstones from 40 patients (8 males and 32 females) was analyzed. This is a prospective study held in Baquba teaching hospital in the period from 1/10/2012 to 1/1/2013 in which we collected the gallstones for the patients who were performed cholecystectomy for them whether open or laparoscopic. The stones were classified according to their chemical composition as a mixed stones (MS), and examined using stone analysis set (chemical qualitative method) of calcium, magnesium, phosphate, uric acid and oxalate which was used reagent for qualitative determination of main individual components of stones. The results of this study showed the highest incidence of gallstones in the age group 40-49 was 13 cases followed by 11, 8 and 4 cases for age groups 30-39, 50-59, 20-29 and 60-more out of 40 cases respectively. The chemical analysis showed the majority of gallstone were mixed, 37 gallstones contain calcium followed by 31 cases had uric acid, 32 stones contain magnesium, 21 and 32 stones contain oxalate and phosphate respectively. Majority of gallbladder stones showed +2 of chemicals concentration composition for each chemical. 10(25%) gallstones compose of five chemicals, 15(37.5%) gallstones composed of four chemicals, 11(27.5%) gallstones composed of three chemicals 3(7.5%) gallstones composed of two chemicals and only one (2.5%) gallstone composed of one chemical. In conclusion, majority of cases had mixed gallstones that involved five and four of inorganic chemicals of calcium, magnesium and phosphate, highest incidence of gallstones in age group 40-49 years old were 13 cases.

**Keywords:** Chemical analysis, gallstones, calcium, magnesium, phosphate, uric acid and oxalate

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## Introduction

Chemical composition of gall stones is essential for aetiopathogensis of gallstone disease. Cholelithiasis or gallbladder stones are one of the major surgical problems in many hospital admission and surgical interventions (Jaraari et al., 2010). This problem is probably related to obesity, cardiovascular disorders (CVD), metabolic syndrome, and dietary habits, especially excessive consumption of meat, which is known to contain large amounts of cholesterol. Obese individuals with a BMI-30 kg/m2 have 95% cholesterol-dominant gallstones and are at a high-risk for cholesterol stones (CS) (Schafmayer et al., 2006).

A quantitative chemical analysis of total cholesterol, total bilirubin, fatty acids, triglycerides, phospholipids, bile acids, soluble proteins, calcium, sodium potassium, magnesium, copper, oxalate and chlorides as minor components were found in all types of calculi (Chandran et al., 2007). Studies on gallstone composition carried out in different parts of the world indicate a close link with dietary habits and ethnicity (Pundir et al., 2001; Verma et al., 2002; Chandran et al., 2007). Other studies have shown that dietary intake of total calories in the form of carbohydrates and fats were associated with high triglyceride levels in gallstone patients (Lee et al., 2012). Gallstone formation is relatively increased with consumption of dietary fats rich in saturated fatty acids (Jaraari et al., 2010; Tsai et al., 2004).

Gallstones might induce biliary inflammation and cholecystectomy is typically followed by dilation of the bile ducts (Chung et al,. 1990). However, there is few reports on the chemical composition of gallstones in Iraqis though a sizeable population suffers from gallstones. This study describes an extensive quantitative analysis of gallstones, including calcium, magnesium, phosphate, uric acid and oxalate and their association to induce neoplastic changes. Hence, the objective of this study was to determine the chemical composition of gallstones among cholecystectomy patients.

## Methodology

### *Gallstone extraction and Chemical Analysis of Gallstones*

This is a prospective study held in Baquba teaching hospital in the period from 1/10/2012 to 1/1/2013. Expert surgeons from the department of surgery extracted the gallstones during surgery. Initially, the gallbladder from patients was surgically removed whether by using a Laparoscopic Cholecystectomy or open procedure. The gallstones were extracted from the gallbladder and preserved under sterile conditions. Gallstones samples were collected from

40 (8 males and 32 females) patients for this study. The study was approved by the local Scientific and Ethic Committee of the institute and consent was obtained from all patients to use the isolated stones for research purpose.

Stones were examined using stone analysis set (chemical qualitative method) of calcium, magnesium, phosphate, uric acid and oxalate. Gallstones analysis kit was used for qualitative determination of main individual components, which is manufactured by BIOLABO SA 02160, Maizy, France.

**Results**

An interesting finding in this study is that the females are at higher risk of cholelithiasis development than males, in a ratio of female: male (8:1). The results of this study showed the cholelithiasis highest incidence in the age group 40-49 was 13 cases followed by 11, 8 and 4 cases for age groups 30-39, 50-59, 20-29 and 60-more years old out of 40 cases respectively. 10(25%) gallstones compose of five chemicals included calcium, uric acid, magnesium, phosphate and oxalate, 15(37.5%) gallstones composed of four chemicals were calcium, uric acid, magnesium and oxalate, 11(27.5%) gallstones composed of three chemicals of calcium, uric acid and phosphate, 3(7.5%) gallstones composed of two chemicals, which one contain magnesium and uric acid, and other contain magnesium and calcium), and only one (2.5%) gallstone composed of one chemical which is phosphate (Table 1). The chemical analysis showed the majority of gallstone were mixed, 37 gallstones contain calcium followed by 31 cases had uric acid, 32 stones contain magnesium, 21 and 32 stones contain oxalate and phosphate respectively. Majority of gallbladder stones showed +2 of chemicals concentration composition for each chemical (Table 2, Figure 1).

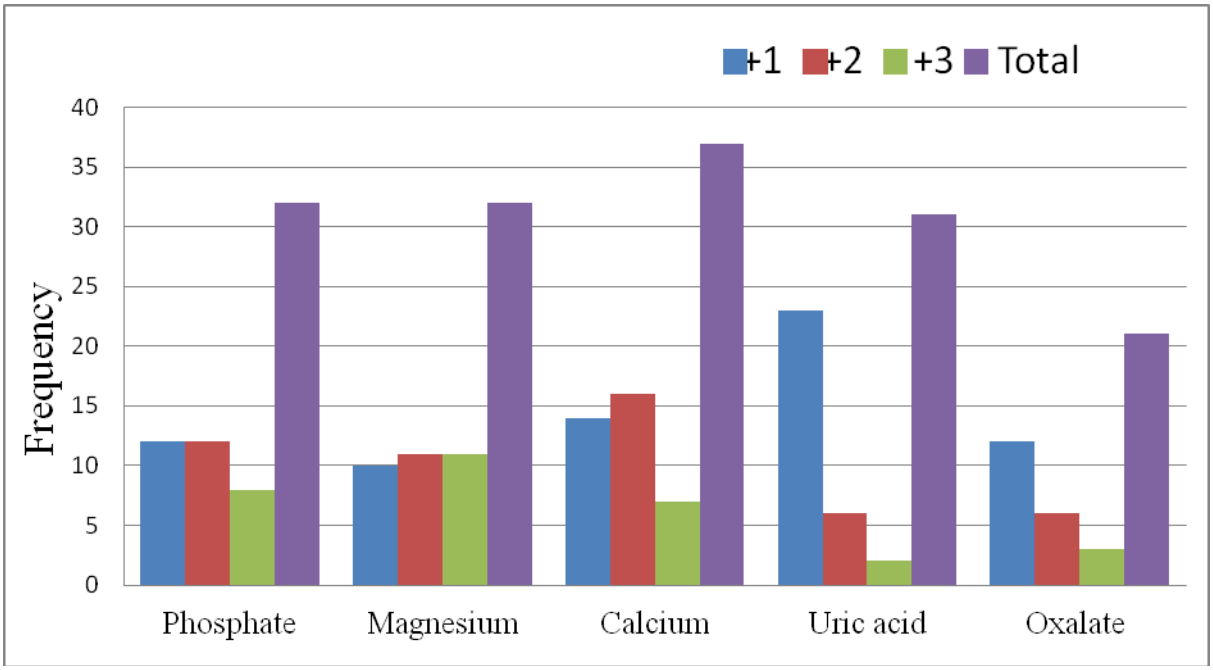
**Table 1: Shows the number of gallstones incidence rate related the gallstones chemical composition**

| NO of cases | Stones' chemical composition | Chemical frequency | |
|---|---|---|---|
| 10 (25%) | Calcium, uric acid, magnesium, phosphate and oxalate | Calcium | 37 |
| 16 (37.5%) | Calcium, uric acid, magnesium and oxalate | Uric acid | 31 |
| 11(27.5%) | Calcium, uric acid and phosphate | Magnesium | 32 |
| 2 (7.5%) | Magnesium and calcium | Oxalate | 21 |
| 1 | Magnesium and uric acid | Phosphate | 32 |
| 1 (2.5%) | Phosphate | | |

**Table 2: Shows the number of gallstones frequency rate related the gallstones chemicals composition of each chemical and patients**

| NO of patient | Age/years | Chemical composition of gallbladder stones | | | | | No. of Chemical /patient |
|---|---|---|---|---|---|---|---|
| | | Phosphate | Magnesium | Calcium | Uric acid | Oxalate | |
| 1. | 37 | - | ++ | ++ | + | - | 3 |
| 2. | 50 | + | - | ++ | - | + | 3 |
| 3. | 54 | - | +++ | ++ | - | +++ | 3 |
| 4. | 36 | + | +++ | + | - | - | 3 |
| 5. | 40 | + | ++ | ++ | + | - | 4 |
| 6. | 47 | ++ | + | + | + | + | 5 |
| 7. | 57 | ++ | ++ | + | + | + | 5 |
| 8. | 30 | + | ++ | ++ | + | - | 4 |
| 9. | 35 | - | +++ | - | ++ | - | 2 |
| 10. | 35 | - | ++ | ++ | ++ | - | 3 |
| 11. | 42 | ++ | +++ | ++ | - | - | 3 |
| 12. | 31 | ++++ | + | - | + | - | 3 |
| 13. | 37 | - | - | - | + | ++ | 2 |
| 14. | 29 | +++ | - | ++ | + | + | 4 |
| 15. | 42 | ++ | + | + | + | - | 4 |
| 16. | 20 | + | ++ | + | + | - | 4 |
| 17. | 47 | + | + | ++ | + | +++ | 5 |
| 18. | 48 | + | - | +++ | - | ++ | 3 |
| 19. | 45 | ++ | ++ | + | + | + | 5 |
| 20. | 28 | +++ | - | + | + | + | 4 |
| 21. | 23 | - | + | ++ | + | ++ | 4 |
| 22. | 63 | ++ | ++ | + | + | - | 4 |
| 23. | 41 | ++ | - | +++ | + | + | 4 |
| 24. | 55 | ++ | - | + | + | - | 3 |
| 25. | 42 | + | +++ | ++ | + | - | 4 |
| 26. | 50 | - | ++ | +++ | + | ++ | 4 |
| 27. | 30 | +++ | - | ++ | + | - | 3 |
| 28. | 46 | - | +++ | +++ | + | +++ | 4 |
| 29. | 62 | ++ | ++ | +++ | - | ++ | 4 |
| 30. | 54 | +++ | + | ++ | ++ | + | 5 |
| 31. | 57 | +++ | ++ | + | + | - | 4 |
| 32. | 31 | - | +++ | ++ | | - | 2 |
| 33. | 45 | + | +++ | + | ++ | - | 4 |
| 34. | 40 | +++ | + | +++ | + | ++ | 5 |
| 35. | 35 | +++ | + | +++ | - | - | 3 |
| 36. | 39 | ++ | - | - | - | - | 1 |
| 37. | 48 | ++ | +++ | + | - | - | 3 |
| 38. | 50 | + | +++ | ++ | +++ | + | 5 |
| 39 | 60 | ++ | + | + | ++ | + | 5 |
| 40. | 41 | + | ++ | ++ | +++ | + | 5 |
| 41. | 65 | + | + | + | ++ | + | 5 |
| Total | | 32 | 32 | 37 | 31 | 21 | |

**Figure 1: Shows the number of gallstones frequency rate related the gallstones chemicals composition of each chemical and patients.**




**Discussion**

Gallstone disease is still one of the most common digestive diseases with an overall prevalence ranges from 10% to 15% in adults and most costly digestive diseases requiring hospital admission, since its prevalence. Medical expenses for gallstone treatment exceeded $4 billion in facility charges in 2004 in the United States (Everhart and Ruhl 2009). Cholesterol disease, one of the commonest digestive diseases in western countries, is induced by an imbalance in cholesterol metabolism, which involves intestinal absorption, hepatic biosynthesis, biliary output of cholesterol, and its conversion to bile acids (Agostino et al., 2003).

Since the pathogenesis of gallstones is not clearly understood, its analysis-using chemical and spectroscopic techniques have provided some clues. The results of stones analysis suggested that the chemical composition of varies considerably. This is clear black stone had a higher bilirubin and lower cholesterol content than other type of stone it can be deduced that the bilirubin content of black stones was much higher than that of brown stones in adults (Portincasa et al., 2008). The frequent of metabolic abnormalities such as atherosclerosis,

obesity, metabolic syndrome, and gallstone disease are related to impaired cholesterol homeostasis. The current view that such abnormalities gain clinical relevance only during adulthood and elderly age is dramatically changing. Both genetic and epigenetic studies suggested a very early onset of chronic disease already *in utero*. Starting from these particular metabolic conditions, a better understanding of mechanisms resulting in chromatin remodeling in response to environmental stimuli acting on the epigenome may offer new options for therapy of cholesterol cholelithiasis and better possibilities for primary prevention in subjects at risk (Agostino et al., 2003). The results of this study were agreed with pervious report that mentioned the major of chemical constituent stones is calcium compounds. Chemical composition of gallstones is essential for aetiopathogensis of gallstone disease (Chandran et al., 2007). Mixed stones composed from including calcium, uric acid, magnesium, phosphate and oxalate, this result constant with pervious finding that also mentioned the pure uric acid stones are relatively rare (Morton et al., 2002). Reduced urinary pH could be an important risk factor for uric acid stone formation (Gianfrancesco et al., 2003). In this study, the majority of stones recovered from patients were composed of calcium, oxalate and uric acid. This might be because of a protein (29 kDa protein) which plays a major role in epitaxial deposition of calcium oxalate over uric acid core (Naseem et al., 2007). The results of study agreed with pervious analysis that showed the calcium content was significantly higher in mixed stones. An interesting finding in this study is that the females are at higher risk of cholelithiasis development than males, in a ratio of female: male (7.3:1). This finding is attributed to high percentage of patients with high risk factors such as multiparity, use of oral contraceptives, obesity and family (Taher, 2013).

In conclusion, majority of cases had mixed gallstones that involved five and four of inorganic chemicals of calcium, magnesium, uric acid and phosphate. The highest incidence was 13 gallstones cases in the age group 40-49 years.

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