

Natural History of Small Gallbladder Polyps Is Benign: Evidence From a Clinical and Pathogenetic Study

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- OBJECTIVES:** Little is known about the natural history and pathogenesis of small gallbladder polyps (<10 mm, usually of the cholesterol type), particularly in Western populations. It is unclear if these polyps and gallstones represent different aspects of the same disease. The aim of this study was to characterize the natural history and pathogenesis of small gallbladder polyps.
- METHODS:** Fifty-six Caucasian patients with small gallbladder polyps, 30 matched gallstone patients, and 30 controls were enrolled in this 5-year prospective study. Patients underwent a symptomatic questionnaire, abdominal ultrasonography, and ultrasonographic evaluation of gallbladder motility at baseline and yearly intervals for 5 years. Cholesterol saturation index, cholesterol crystals in bile, and apolipoprotein E genotype were also determined.
- RESULTS:** Most patients with polyps (mean size: 5.3 mm) were men (61%), asymptomatic, and had multiple polyps (57%). Polyps did not change in 91% of patients during follow-up. No subject experienced biliary pain or underwent cholecystectomy; four developed gallstones. Cholesterol saturation index was higher in patients with polyps or gallstones than in controls ($P < 0.05$). Cholesterol crystals were more frequent in patients with polyps than in controls ($P < 0.0001$) but less common than in gallstone patients ($P < 0.0001$). Polyps and gallstones were associated with nonapolipoprotein E4 phenotypes.
- CONCLUSIONS:** The natural history of small gallbladder polyps was benign, as no patient developed specific symptoms and/or morphological changes in polyps. Consequently, a “wait and see” policy is advisable in these patients. Polyps have some pathogenetic mechanisms in common with gallstones, but few patients developed gallstones.

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INTRODUCTION

Gallbladder polyps are a frequent finding during ultrasonography and surgical procedures (1). Available evidence indicates that the prevalence of gallbladder polyps is higher in Asiatic (9.7%) than in Western populations (2%) (1–3). Several studies have reported that cholesterol polyps account for the majority (>70%) of gallbladder polyps (1,4) and that they are frequently <8–10 mm in size (1,5). These small gallbladder polyps (SGP) are characterized by an accumulation (either localized or diffuse) of cholesterol esters and triglycerides in the foam cells of the lamina propria of the gallbladder submu-

cosa; thus, the mucosal villi may be swollen, and the surface of the gallbladder mucosa can have a polyp-like appearance. In diffuse forms, the mucosa has a characteristic yellow-spotted or reticular pattern, described as “strawberry gallbladder” (6).

The SGP pathogenesis is still debated (6–9). In fact both impaired uptake of free cholesterol by the gallbladder mucosa (6) and a high level of acyl-coenzyme A: cholesterol acyltransferase activity and normal cholesterol ester hydrolase activity (enzymes involved in cholesterol esterification) in the gallbladder mucosa have been reported (7,8) as possible pathogenetic mechanisms. However, the latter finding was not confirmed in

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a more recent study (9). Moreover, it is still unclear whether SGP and cholesterol gallstones represent two different conditions or different aspects of the same disease. SGP and gallstone disease have different prevalence (2–10% and 10–20%, respectively) (1,10) and different geographic distributions (polyps are more frequent in Eastern countries), and the two conditions rarely occur simultaneously in the same subject. However, they do have some alterations in biliary cholesterol metabolism in common (11), (although some (12,13), but not others (6–8) have documented in SGP a bile supersaturated in cholesterol), whereas there are no data regarding genetic factors, in particular apolipoprotein E (ApoE) polymorphism, which is known to have a central role in the overall regulation of cholesterol metabolism (14,15).

As far as SGP natural history is concerned, information from nonsurgical series are rare and derive only from studies performed in Asia, New Zealand, and Chile (16–18). Few changes in polyp size were reported, and the studies agreed that SGP are generally benign. However, other authors have reported isolated cases of rapid growth and neoplastic transformation in small polyps (19,20). Therefore, guidelines for the overall management of patients with SGP are needed.

The aims of this 5-year prospective study were firstly to evaluate the natural history of SGP and, secondly, to investigate genetic and physicochemical aspects of biliary cholesterol and gallbladder motility in patients with SGP or gallstones compared with control patients.

METHODS

Study design

Among patients attending the ultrasonographic unit of the S. Orsola-Malpighi Hospital between 1999 and 2001, 56 consecutive subjects with an ultrasonographic diagnosis of gallbladder polyps <10 mm in size were enrolled in our prospective study. Patients with concomitant gallstones were excluded, as were subjects with other gastrointestinal or metabolic diseases, and those taking drugs that might affect gallbladder motility. At baseline, all patients underwent abdominal ultrasonography and measurement of gallbladder motility, and completed a specific symptomatic questionnaire. The cholesterol saturation index (CSI) was measured, and the presence of cholesterol crystals (CC) in the bile assessed. ApoE genotype was also determined.

All subjects with SGP were followed up for 5 years, undergoing abdominal ultrasonography and completing the symptomatic questionnaire yearly. Gallbladder motility was measured if a significant change in size or number of polyps was observed, if gallstones developed or if variations in clinical manifestation were observed.

Thirty patients with gallstones, matched for age and body mass index (BMI), were enrolled as a comparator group, and 30 subjects with symptoms of dyspepsia, also matched for sex, age, and BMI, were enrolled as a control group. These subjects underwent ultrasonographic measurement of gallbladder motility and bile evaluation (CSI and presence of CC) at base-

line; in addition ApoE genotype was determined in patients with gallstones.

All subjects were studied as outpatients and provided written informed consent. The protocol was approved by the ethics committee of our hospital.

Abdominal ultrasonography

The criteria for gallbladder polypoid lesions were an immobile hyperechoic image protruding from the gallbladder wall into the lumen, fixed at the wall and without acoustic shadow (21). The number of polyps and the size of the largest polyp were recorded.

Symptom questionnaire

Patients were asked to complete a specifically designed questionnaire about the characteristics of their abdominal symptoms. Details of the questionnaire have been published previously (22). The questionnaire assesses the occurrence of the following symptoms: belching, heartburn, nausea, vomiting, bloated feeling immediately after eating, intolerance to fatty or fried food, bitter taste in the morning, uncomfortable feeling in the right hypochondrium, epigastric discomfort, and abdominal pain. This last symptom was also analyzed in terms of localization, radiation, duration, tolerability, and relation to bowel movements. According to the previous study (22), biliary pain was defined as a pain in the right hypochondrium or epigastrium, radiated to the right shoulder that forced the patient to rest and was not relieved by bowel movements.

Gallbladder motility

Gallbladder motility was evaluated using a validated ultrasonographic technique using the ellipsoid method (23). Briefly, the gallbladder volume was evaluated after 12 h of fasting and at regular intervals after ingestion of a standard liquid test meal (200 ml containing 375 kcal, 17 g fats, 10.4 g proteins, and 10 g carbohydrates). The results are expressed as fasting gallbladder volume and percentage emptying. All measurements were carried out by two experienced sonographers (AC and AL). Inter- and intraobserver variation were both <8%.

Bile cholesterol saturation index and cholesterol crystals in bile

Bile for measurement of CSI and analysis of CC was collected by duodenal intubation during upper endoscopy, after IV administration of 5 μ g cerulein (Farmitalia Carlo Erba, Milan, Italy). CSI was calculated according to the method of Carey and Small (24,25). Fresh unfiltered bile (5 μ l) was examined immediately after collection for the presence of cholesterol monohydrate crystals by polarizing microscopy.

ApoE genotyping

ApoE genotyping was performed according to the method of Dallinga-Thie *et al.* (26). Briefly, genomic DNA was isolated from leukocytes and enzymatic restriction of PCR-amplified DNA was followed by separation on MethaPhor agarose gel to

identify the ApoE isoforms. For analysis, homozygotic (E2/E2, E3/E3, E4/E4) and heterozygotic (E2/E3, E2/E4, E3/E4) genotypes were combined into two phenotype groups: individuals expressing one or two $\epsilon 4$ alleles (ApoE4 group: E4/E4, E4/E3, E4/E2) and individuals without the $\epsilon 4$ allele (nonApoE4 group: E2/E2, E2/E3, E3/E3). Allele frequencies were calculated according to the gene-counting method (26).

Statistical analysis

Results are expressed as mean \pm s.d. Continuous variables were analyzed using one-way analysis of variance and multiple comparison *post hoc* tests. Categorical data were analyzed using the χ^2 -test. A *P* value below 0.05 was considered significant.

RESULTS

Patients

The demographic characteristics of the enrolled patients are shown in **Table 1**. The three groups were similar with respect to sex, age, and BMI. Subjects with SGP were more frequently men (60.7%) in all age classes. In the SGP group, multiple polyps were more frequent than single polyps (57% vs. 43%; not significant; **Table 2**). No differences between subjects with multiple and single polyps were found in terms of sex, age, or BMI (**Table 2**). The mean polyp diameter was 5.3 \pm 1.2 mm; the largest lesion was 8 mm in only two patients (3.5%) and the smallest was 3 mm in four patients (7.1%; **Table 2**). The most frequent size of polyp was 5–7 mm in all age groups (**Figure 1**).

Symptom questionnaire

Evaluation of symptoms by the specific questionnaire showed that patients with SGP did not have specific biliary pain, and there was no difference in nonspecific symptoms between patients with gallstones and the control group. Three patients (10%) with gallstones had experienced biliary pain in the past; the others were asymptomatic.

Gallbladder motility

Patients with SGP had a similar fasting gallbladder volume as the control patients (20.3 \pm 7.1 vs. 20.1 \pm 6.1 ml), but this was lower than in patients with gallstones (26.9 \pm 12.2 ml; *P*<0.05; **Figure 2**). Gallbladder emptying was similar in patients with SGP and control subjects (% emptying: 69.5 \pm 10.1% vs.

Table 1. Demographic characteristics of study subjects

	SGP (n=56)	GS (n=30)	Control subjects (n=30)
Male/female	34/22 (61/39%)	14/16 (47/53%)	16/14 (53/47%)
Age (years)	49 \pm 14	50 \pm 14	48 \pm 13
BMI (kg/m ²)	24.9 \pm 3.1	25.2 \pm 2.7	24.3 \pm 3.0

BMI, body mass index; GS, gallstones; SGP, small gallbladder polyps. None of the parameters were significantly different between the groups.

Table 2. Characteristics of patients with small gallbladder polyps

	Single polyp (n=24)	Multiple polyps (n=32)
Male/female	13/11	21/11
BMI (kg/m ²)	25.1 \pm 3.4	24.7 \pm 2.8
Age (years)	49.1 \pm 16.0	48.3 \pm 14.0
<i>Polyp size (mm)</i>		
Mean size ^a	5.3 \pm 1.3	5.2 \pm 1.2
3 mm, n	2	2
4 mm, n	3	7
5 mm, n	10	10
6 mm, n	5	8
7 mm, n	2	5
8 mm, n	2	0

BMI, body mass index.

None of the parameters were significantly different between the groups.

^aMean size overall: 5.3 \pm 1.2.

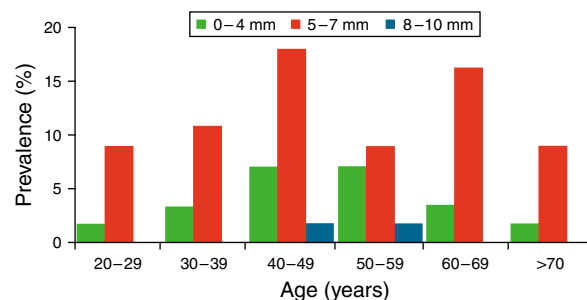


Figure 1. Prevalence of small gallbladder polyps by size and age. Polyps of 5–7 mm were the most frequent in all age groups.

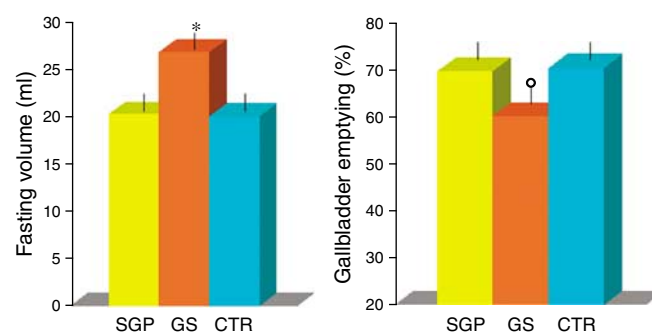


Figure 2. Fasting gallbladder volume and percent gallbladder emptying in patients with small gallbladder polyps (SGP) or gallstones (GS), and control (CTR) subjects. Fasting volume was increased in patients with GS, but not in those with SGP, compared with CTR subjects (**P*<0.05 vs. SGP and CTR). Statistical analysis by analysis of variance (ANOVA, *P*=0.002). Gallbladder emptying was reduced in patients with GS compared with those with SGP or CTR (^o*P*<0.05 vs. SGP and CTR). Statistical analysis by ANOVA (*P*=0.0001).

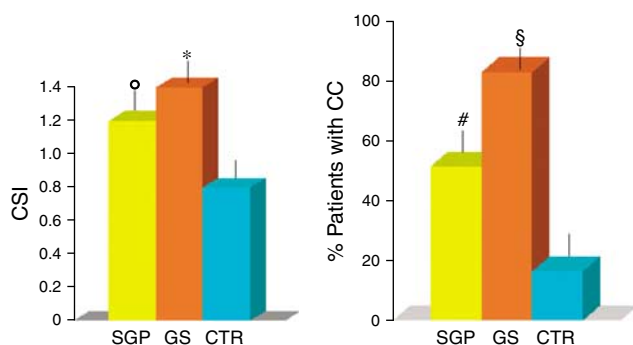


Figure 3. Cholesterol saturation index (CSI) and presence of cholesterol crystals (CC) in patients with small gallbladder polyps (SGP) or gallstones (GS), and control (CTR) subjects. CSI was higher in patients with SGP or GS compared with CTR subjects. (* $P < 0.05$ vs. CTR; χ^2 -test). Patients with SGP had CC more frequently than CTR subjects (# $P = 0.0001$ vs. CTR; χ^2 -test); patients with GS had CC more frequently than patients with SGP and CTR subjects (§ $P = 0.0001$ vs. SGP and CTR; χ^2 -test). Statistical analysis by analysis of variance ($P = 0.0001$).

70.2±9.4%) but it was significantly lower in patients with gallstones (60.0±8.1%; $P < 0.05$ vs. control group). These gallbladder motility characteristics were not significantly different between patients with a single or with multiple polyps.

Bile cholesterol saturation index and cholesterol crystals

CSI was higher in patients with SGP than in the controls (1.2±0.2 vs. 0.8±0.1; $P < 0.05$; **Figure 3**) but was similar to that in patients with gallstones (1.3±0.2). Patients (51.7%) with SGP had CC vs. 16.6% of control patients ($P = 0.0001$); 83.3% of gallstone patients had CC ($P = 0.0001$ vs. SGP and controls). No differences in CSI or presence of CC were found between patients with a single or with multiple polyps.

ApoE genotype

The ApoE4 phenotype was found in 10.8% of patients with SGP, and the non-ApoE4 phenotype in 89.2%. Similar results were found in patients with gallstones: the ApoE4 phenotype was present in 13.3% and the non-ApoE4 phenotype in 86.7%. The distribution of ApoE4 vs. non-ApoE4 phenotypes was not significantly different between patients with SGP or gallstones. **Table 3** shows the distribution of ApoE genotypes in patients with SGP: the most frequent genotype was E3/E3 (75% of cases); 14.2% had the E2/E3 genotype and 11.0% were E4/E3. No other ApoE genotypes were found; in particular none of the subjects carried the homozygote E4/E4 genotype. Gallbladder motility was similar in patients with the different ApoE genotypes. CSI was lower in patients with the E4/E3 genotype compared with the other genotypes, but this difference was not significant.

Follow-up

Three out of fifty-six (5.4%) patients with SGP were lost at follow-up, having relocated to another city; however, during a phone interview they all declared that they had not experienced episodes of biliary pain and had not undergone cholecystectomy.

Table 3. Distribution of apolipoprotein E genotypes in patients with small gallbladder polyps

	E2/E3	E3/E3	E4/E3
<i>n</i> (%)	8 (14)	42 (75)	6 (11)
Male/female, <i>n</i>	6/2	23/19	5/1
Age (years)	50±16	48±13	50±14
BMI (kg/m ²)	25.0±3.2	24.7±2.8	25.1±3.4
Single polyp, <i>n</i> (%)	5 (22)	14 (61)	4 (18)
Multiple polyps, <i>n</i> (%)	3 (9)	28 (85)	2 (6)
CSI	1.18±0.42	1.21±0.45	1.17±0.43
Cholesterol crystals, <i>n</i> (%)	5 (17.4)	20 (68.9)	4 (13.7)
FV (ml)	21.2±11.3	19.2±6.3	22.9±5.9
% Gallbladder emptying	68.3±10.3	69.9±9.5	69.3±7.9

BMI, body mass index; CSI, cholesterol saturation index; FV, fasting gallbladder volume.
No other genotypes were found. None of the parameters were significantly different between the genotypes.

Among the remaining 53 subjects, none experienced episodes of biliary pain or required cholecystectomy during the 5-year follow-up. In most cases, polyps did not change in size during the observation period; in fact SGP remained unchanged in 48 out of 53 patients (91.0%), decreased in size in 2 patients and increased in size (by about 2 mm) in 3 patients. No additional lesions were observed in any case during the follow-up period. Gallstones developed in four subjects in the SGP group: in two subjects with a single polyp and in two with multiple polyps. All four carried non-ApoE4 alleles. Patients who developed gallstones had a greater, although not significant, BMI than those who remained free of gallstones (27.5±3.4 vs. 24.7±3.1 kg/m²). One patient had a family history of gallstones, one was dieting and two had diabetes. Gallbladder motility was assessed in these patients after the diagnosis of gallstones but there were no significant changes compared with the baseline evaluation.

DISCUSSION

This is the first prospective study of the natural history of SGP in a Western population. Our main results indicate that, as found in studies in other populations, the natural history of SGP is benign, without specific or nonspecific biliary symptoms, thus suggesting that cholecystectomy is not indicated in this condition. Furthermore, we found that SGP have some pathogenetic similarities to gallstones (i.e., the presence of CC in the bile, and a cholesterol-supersaturated bile), but only a few patients developed gallstones. The ApoE4 allele was not the most frequent allele in subjects with SGP and, in our population, was not a predictive factor for gallstone development.

Cholesterol gallbladder polyps are the most common type of gallbladder polyp (>70%) (1,4) and are most frequently smaller than 8–10 mm in size (1,5). In our study, the mean size of polyps

was 5 mm, with only 16% in the range 7–8 mm, and none >8, but <10 mm. We chose to enrol only patients with small polyps (considered to be cholesterol type) (1,5), as the major aim of our study was to evaluate their natural history, and possibly to suggest a practical guideline.

The natural history of polyps is not completely understood, and most available studies are retrospective (1). In our prospective study, we did not observe any significant evolution in either the number or size of polyps. These results are different from those documented in non-European studies on the natural history of polyps. Moriguchi *et al.* (16) reported that 6.7% of polyps increased in size and 2% disappeared during follow-up; Collett *et al.* (17) reported that multiple polyps increased in number, and polyps disappeared in three subjects. In the study by Csendes *et al.* (18), 14 out of 111 patients underwent cholecystectomy because of an increase in the size and number of polyps, and polyps disappeared in about 20% of cases. These results can probably be explained by differences in study design or in the characteristics of the studied populations. Furthermore, detailed information about biliary symptoms was not always reported and putative pathogenetic mechanisms (bile cholesterol, gallbladder motility, or ApoE polymorphism) were not analyzed and there were no control groups.

We did not observe any case of gallbladder cancer during the follow-up in our study; Moriguchi *et al.* (16) observed one case of cancer, which appeared at a different site from the polyps. It is important to note that all polyps in our study remained below 8 mm in size and so no patient required prophylactic cholecystectomy, as suggested by Boulton and Adams (27).

To the best of our knowledge, no paper has prospectively evaluated the clinical aspects of SGP using a specific instrument (questionnaire). Our patients with SGP did not develop specific biliary symptoms during the follow-up period, and there were no differences in the frequency of either specific or nonspecific abdominal symptoms between affected and control patients. In the only prospective study to report data concerning the patients' clinical picture, which did not use a standardized symptoms questionnaire, no symptoms specific for biliary disease, such as cholic pain, acute cholecystitis, or jaundice, were reported (18).

Four (7%) of our patients with SGP developed asymptomatic gallstones during the follow-up, a rate only slightly higher than that observed in the general population (28). Similar results were also found in another study in which the incidence of gallstones in subjects with SGP after 5 years of follow-up was higher than in subjects without SGP at enrolment (9% vs. 2%) (29). This author suggested that the exfoliated polyps could serve as a nidus for stone formation (29). Our secondary objective was to identify the similarities with cholesterol gallstones in terms of pathogenetic mechanisms, as it is still not clear whether SGP and gallstones represent distinct entities or different aspects of the same disease (9,12).

About 70% of patients with SGP had CSI above 1, compared with 23% of the control group. Furthermore, 53% of patients with SGP had CC in the bile, which was similar to the proportion in patients with gallstones. These results agree with those of Acalovschi *et al.* (12), Luciano (13), and Chijiwa *et al.* (30), which all compared

patients with SGP and with gallstones, but not with those by Tilvis *et al.* (6), Sahlin *et al.* (7), and Watanabe *et al.* (8), which did not find difference in bile lipid composition between patients with SGP and controls. However, these latter studies agree that CC nucleation time is increased in patients with SGP (6,7,30).

In this study we evaluated ApoE polymorphism, which is known to have a central role in the overall regulation of cholesterol metabolism (14,15), to determine whether there were any genetic differences between patients with SGP and those with gallstones. When we planned the study, the relevance of the *ApoE4* allele was debated, because some authors (31,32), but not others (33), considered it to be the only genetic determinant associated with human gallstone disease. None of our patients with SGP or gallstones had the E4/E4 genotype; this finding is in agreement with that observed by Hasegawa *et al.* (34) in patients with gallstones. The more frequent genotype was E3/E3, whereas only 11% of patients with SGP had the E4/E3 genotype. Our results thus indicate that SGP and gallstones were more frequently associated with non-ApoE4 phenotypes, as recently confirmed by Mella *et al.* (35) in populations at high risk for gallstones.

We did not find significant difference in gallbladder motility between patients with SGP and the controls, whereas, as expected (23), patients with gallstones had impaired gallbladder emptying and an enlarged fasting volume. This indicates that gallbladder motility differs between patients with SGP and gallstones, even though most patients with either SGP or gallstones had a cholesterol-supersaturated bile. Defects in gallbladder motility in patients with gallstones are considered to be secondary events related to cholesterol supersaturation (36). It has been suggested that abnormal cholesterol absorption by the gallbladder wall in the presence of cholesterol-supersaturated bile (37) leads to incorporation of cholesterol into the gallbladder sarcolemma. This in turn impairs gallbladder contraction and relaxation in patients with gallstones (38,39). We have no definitive explanation for the coexistence of normal gallbladder motility and supersaturated bile in patients with SGP. It is possible that SGP represent localized cholesterol accumulation in the submucosa, and the whole sarcolemma is not yet involved. Alternatively, the molecular defect that leads to the incorporation of cholesterol into the sarcolemma may not be present or active in patients with SGP. However, there was no change in gallbladder motility in patients with SGP who developed gallstones during the follow-up.

Our study indicates that the natural history of SGP (in our population <8 mm) is benign, because no changes in the clinical manifestation and/or in the ultrasonographic characteristics were documented, and, as a consequence, there was no indication for an active treatment, mainly cholecystectomy, in any patient. None of the patients developed cancer of the gallbladder. Given the benign natural history, in patients with SGP a "wait and see" policy is advisable, by means of an ultrasonographic and clinical follow-up. A more aggressive management can be performed in case of changes in the patient clinical conditions, i.e., appearance of specific biliary symptoms, or following the ultrasonographic documentation of a change in polyp size/number. Some aspects of biliary cholesterol metabolism are similar in patients

with SGP and those with gallstones; however, only a few patients with SGP developed gallstones, suggesting that additional risk factors are involved, probably relating mainly to lifestyle.

CONFLICT OF INTEREST

Guarantor of the article: Davide Festi, MD.

Specific author contribution: study concept and design: Antonio Colecchia and Davide Festi; acquisition of data: Anna Larocca, Anna Rita Di Biase, Roberta Gualandi; statistical analysis: Maria Letizia Bacchi Reggiani; drafting of the paper: Eleonora Scaioli and Amanda Vestito; Francesco Azzaroli and Patrizia Simoni performed laboratory analyses; critical revision of the paper: Antonio Colecchia and Davide Festi. All authors have approved the final draft.

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Potential competing interests: none.

Study Highlights

WHAT IS CURRENT KNOWLEDGE

- ✓ In Eastern populations small gallbladder polyps (SGP) are mostly benign and of cholesterol-type.
- ✓ No definitive data are available on the frequency of gallstone development in patients with small polyps and their possible pathogenetic correlations.

WHAT IS NEW HERE

- ✓ Also in Western populations SGP are benign and therefore an active treatment, in particular, cholecystectomy, is not required. A “wait and see” therapeutic strategy is preferred.
- ✓ SGPs and cholesterol gallstones have some pathogenetic factors in common. However, the risk of gallstone development in patients with small polyps is not different from that of the general population.

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