Randomized Double-Blind Clinical Trial of Extended Antibiotic Therapy versus Placebo After Laparoscopic Cholecystectomy for Mild and Moderate Acute Calculous Cholecystitis

(CHART Study, NCT02057679)

Martín de Santibañes MD¹, Juan Glinka MD¹, Pablo Pelegrini MD¹, Fernando A. Alvarez MD¹, Cristina Elizondo MD², Diego Giunta MD², Laura Barcan MD³, Lionel Simoncini⁴, Nora Cáceres Dominguez⁴, Victoria Ardiles MD¹, Oscar Mazza MD¹, Rodrigo Sanchez Claria MD¹, Eduardo de Santibañes MD, PhD¹, Juan Pekolj MD, PhD¹

1. Department of General Surgery, Hospital Italiano de Buenos Aires, Buenos Aires, Argentina
2. Department of Internal Medicine and Statistics, Hospital Italiano de Buenos Aires, Buenos Aires, Argentina
3. Department of Internal Medicine and Infectology, Hospital Italiano de Buenos Aires, Buenos Aires, Argentina
4. Department of Pharmacy & Pharmacology, Hospital Italiano de Buenos Aires, Buenos Aires, Argentina

Correspondence to: Martin de Santibanes MD
Department of Surgery, Division of HPB Surgery, Liver Transplant Unit, Hospital Italiano de Buenos Aires
Juan D. Perón 4190. C1181ACH. Buenos Aires, Argentina.
Tel: +54-11 4981 4501
E-mail: martin.desantibanes@hospitalitaliano.org.ar

Conflicts of Interest and Source of Funding: None of the authors of this manuscript has any direct or indirect commercial financial incentive associated with the publication of this paper. The funding involved in this work has been provided by our institution.

Running head: Randomized Trial of Antibiotic Therapy versus Placebo After Laparoscopic Cholecystectomy for Acute Cholecystitis
**Abbreviations**

MINI-ABSTRACT:
This single-center, prospective, double blind, and randomized trial determines the effect of extended antibiotic therapy on infectious complications in patients with mild and moderate acute calculous cholecystitis undergoing laparoscopic cholecystectomy.

ABSTRACT:
Objective: To determine the effect of extended antibiotic therapy on infectious complications in patients with mild and moderate acute calculous cholecystitis undergoing laparoscopic cholecystectomy.

Background: Acute calculous cholecystitis is the most common complication of cholelithiasis. Laparoscopic cholecystectomy is the gold standard treatment in mild and moderate forms. Currently there is consensus for the use of antibiotics in the preoperative phase of acute cholecystitis. However, the need for antibiotic therapy after surgery remains undefined with a low level of scientific evidence.

Methods: The CHART (Cholecystectomy Antibiotic Randomised Trial) study is a single-center, prospective, double blind, and randomized trial. Patients with mild to moderate acute cholecystitis operated by laparoscopic cholecystectomy were randomly assigned to receive antibiotic (amoxicillin/clavulanic acid) or placebo treatment for 5 consecutive days. Primary endpoint was postoperative infectious complications. Secondary endpoints were as follows: (1) length of hospital stay, (2) readmissions, (3) reintervention and (4) overall mortality.

Results: In the per-protocol analysis, 6 of 104 patients (5.8%) in the placebo arm and 6 of 91 patients (6.6%) in the antibiotic arm developed postoperative infectious complications, absolute difference 0.82 (95% CI, -5.96 to 7.61, p= 0.81). The median hospital stay was 3 days, without significant difference between groups (p= 0.3). There was no mortality. There were no differences regarding readmissions and reoperations between the two groups.

Conclusions: The use of antibiotics in the postoperative period of laparoscopic cholecystectomy for mild and moderate acute calculous cholecystitis is not justified,
since it was not associated with a decrease in the incidence of infectious and other types of morbidity in the present study.

**Keywords:** Acute calculous cholecystitis, laparoscopic cholecystectomy, antibiotic, randomized trial
INTRODUCTION:
The incidence of cholelithiasis in adult population is 10% and acute calculous cholecystitis (ACC) is the most common complication. Acute cholecystitis affects more than 20 million Americans annually, with costs in excess of $6.3 billion, constituting a major health burden that has increased more than 20% in the last three decades. 

The diagnostic criteria and severity assessment of ACC have been well established in the Tokyo guidelines 2007 and updated in 2013. According to this expert consensus, ACC is classified into three grades: mild, moderate and severe. Laparoscopic cholecystectomy (LC) is the gold standard treatment in mild and moderate forms. Currently there is consensus for the use of antibiotics in the preoperative phase of ACC, with controversies about its usefulness after the surgical treatment has been completed. Recent guidelines suggest that antibiotics should be administered only up to 24 hours after surgery for mild ACC and 4-7 days for moderate or severe forms. It has been suggested that a scheme with, β-Lactam/Inhibitor of β-Lactamase combinations, would be adequate in patients with mild and moderate ACC, according to most frequently isolated germs. Despite this, the need for antibiotic therapy after surgery remains ill defined with a lack of high quality evidence. Hence, we decided to conduct a randomised controlled trial in patients undergoing LC for mild and moderate ACC, randomizing patients to receive antibiotics or placebo after surgery. The primary objective of the present trial was to assess whether antibiotic treatment after LC in mild or moderate ACC reduces the incidence of postoperative infectious complications. The hypothesis was that postoperative antibiotic treatment has no positive impact on patient’s outcome and therefore should not be indicated in this subset of patients.

METHODS
Study design and ethics
The Cholecystectomy Antibiotic Randomised Trial (CHART) is a single-center, randomized, controlled and blind to patient and investigator trial, which compares antibiotic treatment after LC due to mild and moderate ACC versus no antibiotic treatment. The study design has been reported in detail previously. This study was conducted in accordance with the Declaration of Helsinki, Good Clinical Practice Regulations ICH E6, and applicable regulatory requirements. Written informed consent was obtained for all patients and the Hospital Italiano de Buenos Aires
(HIBA) Ethics Committee gave ethical approval to perform this study (N° 2111). The CHART has been registered at Clinicaltrial.gov database (ClinicalTrials.gov, identifier: NCT02057679).

**Study aims**

*Primary endpoint:* Postoperative infectious complications, defined as any infection occurring within the first 30 postoperative days, classified according to the Clavien-Dindo Classification. Any of the following infectious complications were considered: intra-abdominal collections (abscesses, biloma, subphrenic collection or fluid collection from another location within the abdomen), hepatic abscesses, surgical wound infections (trocar insertion site), and extra-abdominal infectious complications such as pneumonia or urinary tract infections.

*Secondary endpoints:* (1) Length of hospital stay: number of days from admission to hospital discharge. (2) Readmission: need of readmission due to postoperative complications that require hospital care (hydration, intravenous antibiotics, percutaneous drainage or surgical treatment). (3) Reintervention: need of surgical treatment under general anaesthesia or percutaneous procedure in complicated patients. (4) Overall mortality: deaths occurring during the first postoperative month.

**Study Population and Study Treatment**

All consecutive patients from February 2014 with a new diagnosis of mild or moderate ACC according to the Revised Tokyo Guidelines admitted to the HIBA were screened for eligibility to be enrolled in the CHART. Patients received parenteral hydration, gastric protection with proton pump inhibitors, analgesics and treatment with ampicillin/sulbactam intravenously every 8 h until surgery, which was carried out within 5 days after admission. Patients were approached for randomized inclusion if they met each of the following *inclusion criteria:* diagnosis of mild or moderate ACC; willingness to participate in the study; ability to understand the nature of the study and what was required of them; men and non-pregnant, non-lactating women between 18 and 85 years of age who undergo early LC. The main *exclusion criteria* were as follows: rejection to participate in the trial or the process of informed consent; hypersensitivity to AMC or lactose (used in placebo); severe ACC; moderate ACC associated with liver and/or gallbladder abscesses, cholangitis or bile peritonitis; intraoperative findings such as liver cancer, liver metastases, common bile duct stones or gallbladder carcinoma; conversion to laparotomy; previous treatment with antibiotics for more than 5 days; active oncological diseases; AIDS; transplanted
patients. If there were no intraoperative criteria for exclusion, patients were randomly assigned to either group of intervention:

- Experimental group: antibiotic treatment after surgery (AG): They received 1000 mg of Amoxicillin/clavulanic acid (AMC) orally every 8 hours for 5 days, immediately after surgery.

- Control group: placebo treatment after surgery (PG): They received 1000 mg of placebo orally every 8 hours for 5 days, immediately after surgery.

Patients were randomly assigned using a randomizer provided by the HIBA statistical department. This provided a list with a sequence of numbers from 1 to 300 and each patient was randomly assigned to each treatment group.

The HIBA pharmacy was the only non-blind participant in the study and was in charge of preparing, storing and distributing the medication, also ensuring that medication was used exclusively for the purposes of the study. Each Treatment Pack (TP) had a code that was used to identify which group of treatment modalities the patient was assigned to. Each TP contained capsules for 5-days of treatment. The antibiotic and placebo capsules were packaged and labeled identically.

**Surgical procedure**

The American technique for LC was used, as described previously. Intraoperative cholangiography was used as a routine in all patients after having achieved the “critical view of safety”.

**Safety, tolerability and follow-up**

Any adverse events detected during ambulatory monitoring were recorded and classified according to their severity in mild, moderate and severe and by relationship to study treatment according to the decision of the blinded investigator. Treatment relationship was determined with a reasonable probability that the event might have been caused by treatment. Each patient received a written control to mark the intake of each medication as stipulated. Patients were clinically monitored at an outpatient clinic 7 and 30 days after surgery. They received the telephone number of the investigators before any concern or the need to report any event. Postoperative adverse events were evaluated according to the Clavien-Dindo classification.

**Statistical analysis**

Sample size calculation was based on an expected postoperative infection rate of 3% in the antibiotic group, following the hypothesis that the absence of postoperative antibiotic treatment would not be inferior to the use of antibiotic treatment after LC
for the development of postoperative infections. Assuming a non-inferiority margin of 5%, a one-tailed alpha error of 5% and a power of 80% to reject the null hypothesis, it was estimated that the required sample size was 150 cases in each group. The main analysis was performed according to per-protocol analysis (PP) and secondary according to intention-to-treat (ITT) principle.

Categorical variables are described using percentages. Continuous variables are expressed as mean and standard deviation (SD) for those symmetrically distributed, and the median (interquartile range) for those non-symmetrically distributed. The association between the outcome and the assigned treatment was assessed using the Chi-square test in categorical variables or Fisher's test when appropriate. The risk differences between the two arms were estimated, with their respective confidence intervals (CI) of 95%. P<0.05 was considered significant. The statistical analysis was performed with the STATA software version 14 (StataCorp LP, TX).

**Interim analysis**

Due to the primary and secondary end-points of the protocol, an interim analysis of the results was scheduled, once the 50% of the patients were recruited. A non-related-study investigator presented a report of the interim analysis to the HIBA Ethics Committee and they gave the endorsement for the early suspension of the protocol. Statistical simulations were performed, following the methodology presented by Bratton et al to estimate how many samples would include the non-inferiority limit in their 95% CI if the sample size were reached according to the initial sample size calculation. Two scenarios were considered, one optimistic (equal probabilities to those used for the original sample size calculation) and one more conservative (observed probabilities). In both cases, 1000 samples were generated for both the AG and PG of 150 patients in each branch of each sample, as if the expected sample size had been achieved.

**RESULTS**

**Study participants**

Between February 2014, and March 2017, a total of 314 patients were assessed to participate in the CHART. Finally, 201 patients were randomized, 105 in the PG and 96 in the AG (ITT population). One patient from the PG and 5 from the AG, were excluded from the PP analysis (Fig. 1). No patient was lost during 30-days follow-up. Only 1 patient did not complete all medication intakes and discontinued treatment on
the 4th day due to cutaneous rush in the PG, while 4 patients discontinued medication in the AG. Of these, two patients discontinued on the 2nd day due to digestive intolerance, and the remaining two on the 3rd and 4th day of treatment due to digestive intolerance and cutaneous rash respectively.

**Demographic characteristics**

Characteristics of the study population are summarized in Table 1. No significant differences were found between the two groups. Serious comorbidities were rare in both groups. There were no differences regarding clinical presentation and ultrasound results. Characteristics of the laboratory parameters in the study population are detailed in Table 2. No significant differences were found between the two groups. All patients in the series received preoperative antibiotic treatment until the time of surgery. The mean duration of the surgeries was 90 minutes. Intraoperative cholangiography was performed in 100% of the patients, without bile duct injuries.

**Primary end-point**

Infectious complications are listed in Table 3. In the PP analysis, 6 of 104 patients (5.8%) in the PG and 6 of 91 patients (6.6%) in the AG developed postoperative infectious complications, absolute difference 0.82% (95% CI, -5.96 to 7.61; p=0.81). Although no difference was found between the two groups, the upper limit of the 95% CI included the non-inferiority margin of 5% that was set in the protocol design. In the ITT analysis, 6 of 105 patients (5.7%) in the PG and 6 of 96 patients (6.2%) in the AG developed postoperative infectious complications, a risk difference of 0.53% (95% CI, -6.03 to 7.10; p=0.87).

**Secondary end-point**

In the PP analysis, 10 patients (9.6%) from the PG had complications, whereas 10 patients (10%) experienced complications in the AG, with absolute difference of 1.37% (95%CI, -7.19 to 9.94; p=0.75). Most of the complications were mild (grade I and II), with no difference between the two groups (p=1) (Table 3). One patient in the AG had a severe complication (IIIb). This patient required a second-look laparoscopy within 24 hours of LC due to an inadvertent small bowel injury, which was solved with an intestinal resection and primary anastomosis. The patient recovered without complications in the distant postoperative period. Throughout the series, 3 patients had to be readmitted. One patient in the PG was hospitalized on the 7th postoperative day with pneumonia and received appropriate antibiotic treatment. Another patient
from the AG had a laparoscopic appendectomy for acute appendicitis on the 27th postoperative day. The third patient was previously mentioned (inadvertent small bowel injury). The overall median hospital stay was 3 days, without difference between the groups compared (p= 0.3).

**Simulation**

First scenario: In all cases the CI included zero. The mean risk differences for the 1000 simulations were 0.00250=0.25% (infection risk AG - infection risk PG). The missing cases were completed with simulated numbers with event probability of 3%, having a CI for the risk difference including the non-inferiority limit of 5%. Considering an event incidence of 3% and reaching the predefined sample size, 52.5% of the samples include the non-inferiority limit.

Second scenario: In all cases the CI included zero. The mean risk differences for the 1000 simulations were 0.00749=0.75% (infection risk AG - infection risk PG). Completing the missing cases with simulated probability event numbers of 0.0576 (6/104) in the PG and 0.0659 (6/91) in the AG, 738 of 1000 samples had a CI for the risk difference that included the non-inferiority limit of 5%. Considering that the observed incidences were maintained till the end of the recruitment when reaching the predetermined sample size, 73.8% of the samples included the non-inferiority limit.

**DISCUSSION**

In this prospective, randomized, blinded to patient and evaluator trial, we compared the use of extended antibiotic therapy versus placebo after LC for mild and moderate ACC. The analysis showed that the absence of extended antibiotics treatment was not associated with an increased risk of infectious complications and other types of morbidity. Moreover, both groups had similar results regarding hospital stay, re-interventions and hospital readmissions.

Annually, ACC generates more than 120,000 cholecystectomies.² Nowadays LC is the gold standard treatment for mild and moderate forms of ACC.² ⁷. There is consensus to establish preoperative antimicrobial therapy on suspicion of infection, based on clinical, laboratory and imaging findings.⁸ Although ACC is one of the most common diseases in general surgery, few studies have assessed the role of antibiotic therapy after LC. Current guidelines propose to administer antibiotics during the postoperative course with a variable time period.⁸ It has been suggested that a β-Lactam/Inhibitor of β-Lactamase combinations monoscheme would be adequate in
patients with mild and moderate ACC without intraoperative complications such as bile peritonitis, cholangitis, gallbladder perforation or abscesses. In relation to these considerations, we decided to use AMC scheme for the group of patients who received postoperative antibiotics treatment, extending it for a period of 5 days. Despite this, we did not observe a reduction in the incidence of postoperative infectious complications. These findings yield similar results to recently published studies. The fact of delivering an adequate preoperative antibiotic treatment to a patient with ACC and then removing the septic focus through a cholecystectomy seems to be a sufficient therapeutic strategy to definitively solve this disease. This change in treatment paradigm leads to a more rational use of antibiotics, following modern infectious policies, reducing bacterial resistance and the incidence of pseudomembranous colitis by Clostridium difficile. Moreover, although the incidence of medication adverse events in the group of patients receiving antibiotics was low, it is well described in the literature.

Laparoscopic cholecystectomy for ACC is a low to medium complexity surgical procedure, with lower morbidity; mortality and hospital stay rates, when it was compared to the open approach. Several studies have found that early LC in patients with mild and moderate ACC is a safe and effective surgical strategy. Following this therapeutic approach, the majority of the patients in this series were operated within the first 48 hours of admission. Early cholecystectomy has been shown to reduce morbidity, hospital stay, and costs with respect to late cholecystectomy (7 to 45 days) for ACC. The surgical quality standards in this series are equal or higher than those reported in the literature for the treatment of this pathology. In all cases, an intraoperative cholangiography could be performed, without bile duct injuries. The mean duration of the surgeries was 90 minutes, with a 3-day hospital stay, without mortality and a very low reoperation and readmission rate. The overall morbidity reported in the literature ranges from 15-30% and surgical site infection is the most frequent complication. In our study, this incidence was around 5% and was higher than those reported in the literature. A recent meta-analysis reported an incidence of wound infection of 2.7% for early cholecystectomy and 4.1% for late cholecystectomy in acute cholecystitis. This fact could be explained by the strict follow-up of the patients in the present study. In spite of this, the use of antibiotics did not reduce the incidence of surgical site infections, as other studies have shown. In a recent study, Regimbeau et al analyzed a total of 414
patients treated with 2 grams of AMC in the postoperative period of cholecystectomies due to acute cholecystitis, without a decrease in the incidence of infectious complications. Although the study was randomized and included 17 medical centers in France, its main limitations were that there was no comparison with a placebo or a strictly blinded analysis of the results. In addition, the course of antibiotic therapy was nonstandardized, with a variable amount of treatment days. There were also problems in the postoperative follow-up of patients, with a high proportion of protocol violations. On the other hand, both patients operated with conventional surgery (15%) and laparoscopic surgery (with a conversion rate of 10%) were included in the same analysis. These situations could generate doubts in the interpretation of its results. Loozen et al\textsuperscript{22} randomized 156 patients to receive a single preoperative dose of cefazolin (2000 mg) versus antibiotic prophylaxis for 3 days after cholecystectomy (intravenous Cefuroxima 750 mg plus metronidazole 500 mg three times daily). The main conclusion was that standard single-dose antibiotic prophylaxis did not lead to an increase in postoperative infectious complications. However, to demonstrate the non-inferiority of this treatment, a sample size of almost 600 patients would have been necessary. Given the low rate of infection in LC it would be questionable if such a study were necessary.

From a methodological point of view, our study solves many of the problems previously discussed. First, our antibiotic treatment with AMC was compared with a placebo and both the patient and the investigators were blind until the end of the study. The duration of the antibiotic treatment was set for both branches for 5 consecutive days, with a high adherence to the protocol. At the same time, patients were strictly scheduled for clinical controls at 7 and 30 postoperative days, with a complete follow-up of all patients recruited. We found no differences between the results of the primary and secondary end-points raised for the study. Even though the non-inferiority of the placebo compared with antibiotics for development of infectious complications could not be proven, because the non-inferiority margin of 5% finally lied within the 95% confidence interval, wound infection was the most frequent complication of the present series, with the same distribution in both arms of treatment. The clinical relevance of this type of complication for a LC would be debatable, since it is an infection in a small wound, which had no impact in the postoperative evolution of the patients. On the other hand, we estimated the probabilities of including the non-inferiority limits in the confidence intervals of the
risk difference, which ranged between 52.5% and 73.8%. The first scenario is reasonably optimistic for its effect on the standard errors of the difference, while the second scenario is more conservative in the same sense and probably looks more realistic. Finally, other studies used a non-inferiority margin of 11% and the associated wide confidence intervals could have masked a possible difference in postoperative infections between the compared groups. Since the non-inferiority margin is arbitrary set, if we had applied a larger margin for our study, as mentioned above, we would have concluded non-inferiority in our final analysis. The use of antibiotics in the postoperative period of LC for mild and moderate acute cholecystitis seems not justified, since it was not associated with an increased risk of infectious complications and other types of morbidity in the present study. Moreover, both groups compared had similar results regarding hospital stay, reinterventions and hospital readmissions.
REFERENCES


FIGURE LEGENDS

**Figure 1.** Flow of study participants in the CHART trial.