EFFECTS OF ACID SUPPRESSING DRUG TREATMENT IN PATIENTS WITH ACUTE PANCREATITIS AND WITH CONCOMITANT CLOPIDOGREL THERAPY

Ph.D. Thesis

Alexandra Demcsák M.D.

Doctoral School of Theoretical Medicine, Faculty of Medicine, University of Szeged, Szeged

Department of Pediatrics and Pediatric Health Center, Albert Szent-Györgyi Health Centre, Szeged

Supervisors:

Imre László Szabó M.D., Ph.D.

Doctoral School of Theoretical Medicine, Faculty of Medicine, University of Szeged, Szeged

Division of Gastroenterology, First Department of Medicine, Medical School, University of Pécs, Pécs

Péter Hegyi M.D., Ph.D., D.Sc., MAE

Institute for Translational Medicine, Medical School, University of Pécs, Pécs

Center for Translational Medicine, Department of Medicine, University of Szeged, Szeged

Division of Gastroenterology, First Department of Medicine, Medical School, University of Pécs, Pécs

Doctoral School of Theoretical Medicine, Faculty of Medicine, University of Szeged, Szeged

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I. PUBLICATIONS RELATED TO THE SUBJECT OF THE THESIS

I.) Demcsák A, Soós A, Kincses L, Capunge I, Minkov G, Kovacheva-Slavova M, et al. Acid Suppression Therapy, Gastrointestinal Bleeding and Infection in Acute Pancreatitis – An International Cohort Study. *Pancreatology* (2020) 20:1323-1331. doi: 10.1016/j.pan.2020.08.009
IF: 3.629 (2019)**, Q1**

II.) Demcsák A, Lantos T, Bálint ER, Hartmann P, Vincze Á, Bajor J,et al. PPIs Are not Responsible for Elevating Cardiovascular Risk in Patients on Clopidogrel – A Systematic Review and Meta-analysis. *Front Physiol* (2018) 9:1550. doi: 10.3389/fphys.2018.01550
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SCIENTIFIC METRICS

Number of publication **related to the subject** of the thesis:	2 (2 first author)
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Q1: 1, Q2: 1, Q3: -, Q4: -	
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Hirsch index:	3
https://scholar.google.com/citations?user=W-lwh2EAAAAJ&hl=en&citsig=AMD79oqbQIqmauGf8IvpJIBqLm2JIxsCZg	
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II. INTRODUCTION

Acid suppressing drugs (ASDs) are the cornerstones in the therapy of diseases in which gastric acid has a causative primary or contributory role. Their indication is to prevent the damage of the gastric mucosa or to propagate its healing. The histamine-2-receptor antagonists (H2-RAs) and proton pump inhibitors (PPIs) are classes of this drug group, and nowadays, PPIs are among the most commonly prescribed drugs. Their usage is constantly increasing, while several studies raised concerns regarding their overprescription without further re-evaluation or termination of these treatments, which could lead to prolonged administration with short- and long-term side effects.

Acute pancreatitis (AP) is an acute inflammatory condition of the pancreas. Its global incidence is 30-100 cases per 100,000 general population per year, and it is one of the most frequent GI causes of hospital admission. There is no specific therapy available for AP, symptomatic and curative treatments are established based on guidelines and the prior experience of the medical staff. Despite the fact that ASDs are routinely administered in clinical practice in the majority of AP cases, strikingly, current national and international guidelines do not include any information on their administration in AP, and there are no well established randomized controlled trials (RCTs) or detailed cohort analyses which would analyze their safety and efficacy. Conventionally, the management of AP patients included nothing by mouth from the time of hospital admission. It was believed that by doing so the inflamed pancreas can rest, because fluid intake or solid nutrients would stimulate exocrine pancreatic functions and promote the release of proteolytic enzymes. During fasting, ASD administration could be a potentially good therapeutic option in patients with AP for the protection of the upper GI mucosa. There are contradictory results in the literature on the beneficial and harmful effects of ASD administration in patients with AP. Such therapy might be beneficial if it decreases severity or mortality; however, acid suppression can be harmful as it might increase the risk for GI infections. Although many international cohort studies were published in AP, few data are available on the use of ASDs, GI bleeding and infection.

To protect the GI mucosa, PPIs are administered not only in GI disorders, but to implement secondary prevention in patients with prior cardiovascular (CV) diseases and long term antithrombotic therapy. A combination of antiplatelet drugs is used for the treatment of acute coronary syndrome (i.e., aspirin and thienopyridines) and for the secondary prevention of further CV events. Dual antiplatelet therapy is followed by possible side-effects, such as higher risk for GI bleeding, increasing both mortality and ischaemic complications. To reduce

the risk of GI bleeding in patients with risk factors, PPIs are strongly recommended by international guidelines for these patients. Clopidogrel, a thienopyridine derivative, inhibits platelet aggregation and is commonly used for prevention against CV events. However, the literature consists of contradictory findings on the concomitant use of clopidogrel and PPIs. *In vitro* findings suggested that PPIs reduce the antiplatelet effect of clopidogrel, followed by several clinical studies with contradictory outcomes. A reason for a possible interaction between the drugs is that they are metabolized by similar cytochrome P450 enzymes in the liver. Due to competitive inhibition, PPIs could prevent the formation of the active metabolite of clopidogrel, therefore causing reduced anticoagulant effect and further CV complications. Even though international guidelines are recommending their concomitant administration, there are still contradictory data on them in the literature. A higher risk for CV outcomes was found in several studies, systematic reviews and meta-analyses in patients with clopidogrel on PPI therapy. Generally, whenever observational studies were included, a positive association was described, a higher risk for CV outcomes was found in patients with clopidogrel and PPI therapy. On the other hand, whenever propensity-matched groups were compared the difference between the groups disappeared.

III. AIMS

Therefore, our first aim was to understand the current national and global practice of ASD administration in patients with AP, to investigate their effect on the disease outcome (severity and mortality), and to analyze the efficacy and safety of these drugs in this patient population by evaluating the risk of GI bleeding and GI infection with a retrospective cohort analysis.

Furthermore, our aim was to carry out a precise investigation on the potential CV risks in co-administration of clopidogrel and PPIs with a systematic review of the current literature and a meta-analysis of available data.

IV. MATERIALS AND METHODS

IV.1) Cohort analysis

To assess the worldwide trend of ASD administration in AP patients, an invitation letter was sent out to the members of the International Association of Pancreatology in January 2019 to participate in the present study. The time period of data collection was from January 2013 to December 2018. The study was approved by the Scientific and Research

Ethics Committee of the Medical Research Council in Hungary (TUKEB-22254-1/2012/EKU).

Centers had to provide data on the general demographic data (gender and age of the patient, severity of pancreatitis and mortality); they had to indicate whether the patients received ASDs (PPI or H2-RA) upon admission, during hospitalization and at discharge. Centers had to include data on the signs and cause of GI bleeding. It had to be recorded if a stool culture test (SCT) was performed along with its result. In the case of positive testing, the name of the pathogen had to be included.

The diagnosis of AP was based on the IAP/APA evidence-based guidelines for the management of AP A1 recommendation. At least two from the following three criteria should be confirmed in patients: clinical (upper abdominal pain), laboratory (serum amylase or lipase >3x upper limit of normal) and/or imaging (CT, MRI, ultrasonography). Severity of pancreatitis was determined based on the revised Atlanta classification.

Signs of GI bleeding were provided by each center. These included positive rectal digital examination, macroscopically observed bleeding in the stool, vomit or gastric juice, positive stool blood test, and bleeding verified by an imaging technique. Bleeding cases that occurred in association with endoscopic retrograde cholangio-pancreatography (ERCP) were excluded since administration of ASDs does not have an effect on this type of bleeding. If the cause of the GI bleeding could not be determined, patients were not included in the analyses regarding GI bleeding. The presence of pathogens in the stool verified by laboratory testing was considered GI infections. Non-specific signs such as fever, diarrhea and vomiting without testing were not accepted. The pathogens were identified for each patient.

To identify differences between categorical variables the Chi-square with Fisher's exact test was used. The significance level was set at 0.05. All statistical analyses were performed by using IBM SPSS Statistics for Windows software, Version 25.0 (IBM Corp., Amonk, NY, USA).

IV.2.) Systematic review and meta-analysis

A systematic review of studies was performed accordance with the Preferred Reporting Items for Systematic Reviews and Meta-analysis (PRISMA) Statement. After developing our clinical question and translating it into a well-defined systematic review question based on the PICO format (Patients, Interventions, Comparators and Outcomes), a manual search of medical databases, including PubMed (MEDLINE), Embase and the Cochrane Central Register of Controlled Trials, was performed for human observations using

the following PICO format: P: patients on clopidogrel; I: patients treated with PPI; C: patients without PPI treatment; O: cardiovascular risk. Two independent investigators separately screened the titles and abstracts for eligible studies published from inception to 30 December 2016. The present meta-analysis was registered in the international prospective register for systematic reviews (PROSPERO) under No. CRD42017054316.

Study inclusion criteria: (1) randomized or observational studies (cohort and case-control studies) carried out either in a retro- or prospective manner; (2) only adult patients (over 18 years); (3) patients receiving clopidogrel treatment; (4) should compare PPI takers and non-PPI takers; (5) we only involved studies that stated exact patient number in the preferred groups (total number of patients, patients who received clopidogrel and PPI, outcome number); (6) human studies; (7) studies should show data for either one or more of the following outcomes: (1) major adverse cardiac event (MACE): composite of cardiac and non-cardiac death, non-fatal myocardial infarction, target vessel failure; (2) myocardial infarction (MI): myocardial infarction or new, definitive major coronarographic defect; (3) CV death: only CV death. Studies published in English were selected.

Numeric and texted data were extracted from the eligible articles, such as study type, study endpoints, number of patients in the study, in PPI and in non-PPI treatment groups, and number of patients who received clopidogrel and data on the study characteristics.

We calculated risk ratio/relative risk (RR) and 95% confidence interval (CI) for CV events (MACE, MI and CV death). Between-study heterogeneity was tested with the I^2 statistic, and it was interpreted according to the Cochrane Handbook for Systematic Reviews and Interventions recommendation. P values of less than 0.05 for relative risks, and p values of less than 0.10 for heterogeneity were considered as indicators of significance. Publication bias was estimated through a visual inspection of funnel plots. All analyses were performed with the Review Manager (RevMan) software, Version 5.3 (Copenhagen: The Nordic Cochrane Centre, The Cochrane Collaboration, 2014).

V. RESULTS

V.1.) Cohort analysis

Data of 17,422 adult patients with AP were collected retrospectively from 59 centers. Upon admission, 23.3% of patients took some kind of ASD, from these patients, 88.3% was admitted with a PPI, 11.3% with a H2-RA, and 0.4% received both kind of ASD. During hospitalization, 86.6% of patients received ASD treatment, 81.8% of these patients had only PPIs, 15.4% had solely H2-RAs and 2.7% had both PPIs and H2-RAs. At the time of

discharge from the hospital, 57.6% of patients were prescribed an ASD, 92.6% of them received prescription for PPIs, 7.3% for H2-RAs and 0.1% for both ASDs.

Acid suppression therapy was associated with more severe AP, and mortality was significantly higher among them compared to those without acid suppression. From the included patients, 817 had GI bleeding. Among these patients, there were significantly more moderate and severe AP cases. In case of GI bleeding, the rate of mortality was significantly higher compared to patients without bleeding. There were significantly more patients suffering from GI bleeding while receiving acid suppressing treatment compared to those who did not. The order of SCTing was associated with more severe AP, and these patients' mortality was significantly higher. From the 1,102 patients who underwent SCT, 28.4% of tested patients had positive results. The most common pathogens causing GI infections were *Clostridium difficile* and the *Klebsiella* species. Among patients with GI infections, we found significantly more moderate and severe cases in patients with positive SCT. In patients with GI infection, the mortality rate was significantly higher compared to the rate in the group tested negative for GI infections. GI bleeding was significantly more frequent in patients with verified GI infection. There was no significant difference in the occurrence of GI infection between patients with or without ASD treatment, therefore we can state that acid suppressing treatment was not associated with higher risk for GI infection.

V.2.) Systematic review and meta-analysis

A total of 27 studies were selected for quantitative analyses. Altogether, we found data for MACE in 23 publications, for CV death in 10, and for MI in 14. Seventeen of them were observational studies, 16 were cohorts, and one was a case-control study. Data from ten RCTs were also collected. As post-hoc analyses of RCTs, in four studies the populations and outcome of our interest were not randomized, therefore, their data were included in the statistical analyses of observational studies. All the studies included were published between 2009 and 2016.

The number of patients involved was 156,823. A total of 63,756 received PPI plus clopidogrel treatment, and 99,910 were in the clopidogrel alone group. Risk of MACE was determined from data from 127,695 patients, MI risk was assessed on the basis of data from 82,330 patients, and risk of CV death was evaluated based on data from 53,905 patients.

Our results showed that the risk of MACE is significantly higher in the PPI group (RR=1.22, 95% CI=1.06–1.39, p=0.004), with considerable heterogeneity across the included studies (I^2=90%, p<0.001). However, separating the data for the RCT studies from that of the

non-RCT studies revealed that a significant association of adverse outcomes (MACE) can only be seen in non-randomized studies (observational studies: RR=1.26, 95% CI=1.09–1.46, p=0.002, I^2=93%, p<0.001; RCTs: RR=0.99, 95% CI=0.76–1.28; I^2=0%, p=0.93), although the heterogeneity remained considerable in the observational group, which might not be relevant in the RCT group. There was no significant effect of concomitant clopidogrel and PPI treatment on CV death (RR=1.21, 95% CI=0.97–1.50, p=0.09). The result from the statistical analysis may represent substantial heterogeneity across the studies (I^2=67%, p=0.001). The risk of MI was significantly higher in the PPI group (RR=1.43, 95% CI=1.24–1.66, p<0.001). The results from the statistical analysis may represent substantial heterogeneity across the studies (I^2=66%, p<0.001).

VI. DISCUSSION

VI.1.) Effects of acid suppressing drugs in acute pancreatitis therapy

The ASDs are widely used in the clinical practice, not solely for the diseases of the GI tract, but in other conditions, such as CV diseases. Especially PPIs are among the most frequently prescribed drugs with increasing use every year. Even though there are well established indications for a wide array of diseases when and how to conduct treatment with ASDs, several studies were published regarding their overprescription. Data from the cohort analysis supports the worldwide overuse of ASDs, specifically in patients with AP. The number of patients on ASD treatment has increased by 3.7-fold during hospitalization with almost all of the patients receiving some kind of ASD, and more than 50% remained on an ASD after discharge, a more than 2-fold increase relative to the number at admission.

Systemic inflammatory response syndrome is often a complication of severe AP, which leads to high level of inflammatory markers and organ dysfunctions. Patients with severe AP, especially those who require intensive care treatment or mechanical ventilation are prone to develop stress-related acute gastric mucosal lesions, which lesions can range from simple gastritis and erosions to ulceration and bleeding, more than half of patients with AP may develop upper GI ulcers, and the occurrence shows positive correlation with the severity of pancreatitis. Hypersecretion of gastric acid seems to play a major role at the pathogenesis of stress-related acute gastric mucosal lesions. Therefore, in these cases it can be indicated to use prophylaxis for peptic ulcer disease and protection of upper GI mucosa could be a possible indication for ASD administration in AP patients, which could decrease the rate of GI bleeding. Based on our results, not only in case of severe, but also in moderate pancreatitis cases there were significantly more patients receiving ASD treatment. The occurrence of GI

bleeding was associated with higher morbidity and mortality, which increases the length and cost of hospitalization. Investigating its association with ASD treatment, we found that bleeding occurred more frequently in patients on ASD therapy.

ASDs are considered well tolerated and effective, and only rare and mild side effects have been reported in short-term use. However, nonessential long-term ASD treatment can lead to various side effects in spite of their reported good safety profile. Side effects are including elevated risk for GI infections by repressing the gastric acid barrier and altering the microbiome. Notably, *Clostridium difficile* infection has shown strong association with ASD therapy. According to our results, ASD administration did not elevate the risk for GI infections in patients with AP. Even though, there was relatively low number of testing among the included patients, almost 30% of them had GI infections. The most common pathogen was *Clostridium difficile* (60%). In the studied patient population, GI infections have been associated with more severe AP, higher rate of GI bleeding and worse mortality. Therefore, length of hospitalization and the cost of treatment could be worse in patients with GI infections.

VI.2.) Cardiovascular effects of PPI therapy in patients with clopidogrel

A possible interaction between clopidogrel and PPIs came to the fore after an observational study had been performed in 2006, which found clopidogrel activity on platelets was diminished in patients receiving PPI treatment. Clopidogrel, a thienopyridine derivative, inhibits platelet aggregation, and being a prodrug, it requires a two-step oxidative biotransformation intrahepatically, mediated mainly by cytochrome P450 isoenzymes. The active metabolite binds irreversibly to platelet adenosine diphosphate receptor P2Y12, therefore preventing platelet aggregation. All PPIs are extensively metabolized to inactive metabolites mainly via CYP2C19 and CYP3A4 in the liver. The potential interaction mechanism lies in the fact that both clopidogrel and PPIs, in varying degrees, are metabolized by the same cytochrome P450 enzyme. PPIs have the potential to competitively inhibit the metabolism of clopidogrel to its active metabolite, which leads to reduced circulating concentrations of the active compound.

The data on the interactions between clopidogrel and PPIs remain unclear despite the numerous *in vitro* and *in vivo* studies on the subject. The *in vitro* studies have shown that the effectiveness of clopidogrel decreases with simultaneous use of clopidogrel and PPIs, and therefore, the risk for CV events will be elevated. Several possible causative factors may lie behind this phenomenon, one of them is the connected biotransformational route of

clopidogrel and PPIs, or the possible differences in genetic polymorphism of these enzymes. There are several studies, mostly observational ones, whose findings are consistent with these *in vitro* results, showing an elevated risk for CV side-effects in patients on combined clopidogrel and PPI treatment. There is considerable disagreement between the various clinical studies that show no increased risk of CV outcomes.

Based on the several international guidelines, PPIs are recommended for patients with history of upper GI bleeding or with multiple risk factors for GI bleeding who require antiplatelet therapy. The risk reduction achieved by concomitant PPIs might outweigh any potential reduction in the CV efficacy of antiplatelet treatment because of a drug–drug interaction.

In the meta-analysis, our aim was to focus on this discrepancy and to find a possible resolution. Our combined data from all of the studies involved showed that the presence of MACE and MI is significantly higher in the PPI plus clopidogrel patient population, a finding which is consistent with results from previous observational studies. However, in reducing the degree of heterogeneity by creating subgroups based on study design, we also found that this previously experienced risk elevation and heterogeneity will disappear as in other studies. This result is similar to those of previous meta-analyses, where a higher CV risk was found among observational studies without any difference between the clopidogrel plus PPI group and the no PPI group in RCTs. An examination of the results, heterogeneity and risk of bias of the studies involved in our meta-analysis points to the low quality of observational studies, whose results are opposite to those of RCT studies, all proving an acceptance of results from RCT studies showing no enhancement of CV risks due to PPIs.

VII. CONCLUSIONS AND NEW FINDINGS

Hereby, I represented two studies on the safety and efficiency of ASDs in two different disease entities; both works have studied ASDs as a primary or secondary prevention in the therapy of patients with AP and CV diseases.

One of our aims was to investigate the current place of ASDs in patients with AP that we could present in a large AP population first in the literature. Data showed a worldwide unnecessary ASD use in AP patients, even though there is no substantial evidence that ASD treatment is beneficial for the therapy of AP. Thus, we presented their association with higher morbidity and mortality. The cohort analysis is among the firsts to report data on the rate of GI bleeding not related to surgery or ERCP in patients with AP. Based on our data, ASD administration during AP did not increase the risk for GI infections. Taking into consideration

the advice from the American Gastroenterological Association, the benefits of ASDs outweigh their risks if appropriately prescribed, but when there is no indication, modest risks become important because there is no potential benefit. Therefore, the routine administration of ASDs is not recommended in patients with AP if there is no other indication for their administration. Long-term complications could be avoided by re-evaluating the current clinical practices, incorporate recommendations to current guidelines, and by giving detailed plans for patients and their general practitioners how to gradually reduce or leave the ASDs.

Our second aim was to investigate the potential risks of co-administration of clopidogrel and PPIs with a systematic review of the current literature and a meta-analysis. Our results have shown that there is no definitive evidence for any significant association between CV risk elevation and PPI in patients on clopidogrel treatment, based on RCTs. Thus, no definitive evidence exists for an effect on mortality. From this point of view, the previous FDA guidance to use favorable or non-favorable drug combinations does not seem to be relevant by now based on both previous trials and our own analyses. Because PPI induced risk reduction clearly outweighs the possible adverse CV risk in patients with a high risk of GI bleeding, a combination of clopidogrel with PPI should be recommended.

NEW FINDINGS

VII.1.) Effects of acid suppressing drugs in acute pancreatitis therapy

1. We could show the extensive administration of ASD therapy during the course of AP with the majority of patients receiving acid suppressing treatment during hospitalization.
2. ASD administration was associated with more severe AP and higher mortality. Older age and worse than mild pancreatitis increased the chance of starting ASD treatment.
3. Our analysis has shown that low number of patients suffered from GI bleeding while having AP. GI bleeding was associated with more severe AP and higher mortality, and ASD therapy was associated with higher risk for GI bleeding. AP severity carried a 3 times higher probability, and GI infection carried a 2.8 times higher chance for GI bleeding.
4. ASDs during AP did not elevate the risk for GI infection. The most common pathogen associated with ASD therapy was *Clostridium difficile.*
5. According to our data, the routine administration of ASDs is not recommended in patients with AP if there is no other indication for their administration.

VII.2.) Cardiovascular effects of PPI therapy in patients with clopidogrel

1. Based on the results of the meta-analysis, there is no definitive evidence for any significant association between CV risk elevation and PPI in patients on clopidogrel treatment.
2. No definitive evidence exists for an effect on mortality.
3. Heterogeneity and risk of bias of the involved studies points to the low quality of observational studies, whose results are opposite to those of RCT studies. All proving an acceptance of results from RCT studies showing no enhancement of CV risks due to PPIs.
4. Because PPI induced risk reduction clearly outweighs the possible adverse CV risk in patients with a high risk of GI bleeding, a combination of clopidogrel with PPI should be recommended for patients at risk.

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