Proton Pump Inhibitors and Risk for *Clostridium difficile* Associated Diarrhea

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Increased incidence of *Clostridium difficile* infection (CDI) among in-patients is associated with significant increased mortality, morbidity, and stay in the hospitals. This has occurred despite heightened awareness of the risks of broad-spectrum antibiotics, overall reduction in antibiotic use and increased focus on hospital hygiene. So though the main risk factor for CDI is use of broad-spectrum antibiotics, the use of proton pump inhibitors (PPIs) as a novel potential contributor has been implicated, because of their ability to substantially reduce gastric acid secretion which is an important host defense mechanism in suppressing the ingested *C. difficile* or its spores. Antibiotic disruption of the normal intestinal flora and reduced gastric acidity have been suggested as the risk factors for *C. difficile*-associated diarrhea (CDAD). Based on such assumptions the use of PPIs may be associated with an increased risk of CDAD. While a definite association between PPI use and CDAD has not yet been confirmed, the possibility and such an association however cannot be ruled out at present. Thus among the identified risk factors, the use of PPI is important, previously unrecognized and modifiable risk factors whose use should be carefully evaluated among hospital in-patients receiving antibiotics, especially in those with a diagnosis of *C. difficile* diarrhea. *(Biomed J 2014;37:178-183)*

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Proton pump inhibitors (PPIs) are potent inhibitors of gastric acid production and are the drug of choice for treatment of gastro esophageal reflux disease, bleeding ulcers, stress ulcers and in prophylaxis of ulcer in patients on non-steroidal anti-inflammatory drugs, corticosteroids, patients with head injury, burn patients, and in patients admitted to the intensive care units. They are also indiscriminately used for routine ulcer prophylaxis in patients who do not meet the above - mentioned criteria and even in the absence of clear evidence of a benefit.[1] Moreover, many patients once started on a PPI in a hospital are also frequently continued on these medications for an indefinite period of time even after discharge. Hence their rampant and irrational use increases the risk for several adverse effects like, nosocomial pneumonia, electrolyte imbalances and drug interactions with antimicrobial and antiretroviral agents due to the decreased absorption of the administered drugs. In addition there is also an associated increase in the incidence of colonization by the commensals from the colon to the relatively sterile upper gastrointestinal tract (GI) of patients receiving PPIs,[2] One such commensal is *Clostridium difficile* that can cause super infection resulting in *Clostridium difficile* associated diarrhea CDAD, with the increased use of PPIs.[3] This type of super infection is usually due to disruption of the normal intestinal flora and is a very common adverse effect during treatment with broad spectrum antimicrobial agents.

On the other hand reduced gastric acidity caused by PPIs can also be suggested as a significant independent risk factor for the same.[3] The reason being that the gastric juice is strongly bactericidal for the ingested microorganisms at pH less than 4.0, whereas it is less bactericidal at pH 5.0 and totally ineffective at pH 6.0 and higher.[4] The doses in which the PPIs are commonly used frequently elevate the gastric pH; therefore they emerge as a potential risk factor for CDAD suggesting that hypochlorhydria increases the host susceptibility to bacterial infections.[5]

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Hence such relationship between PPI therapy and CDAD infections was established when it was observed that there was increased survival of the ingested/acquired vegetative forms of *C. difficile* in the gastric contents of patients receiving PPIs. Thus patients receiving PPIs have been found to be 4.17 times more likely to have CDAD as compared to their counterparts,[6] the risk being more when PPI are combined with antimicrobial agents.

C. difficile associated diarrhea

C. difficile is a gram - positive anaerobic, Spore forming bacilli of genus *Clostridium* as seen in Figure 1. It exists in two forms, the vegetative and the spore forms, out of which the vegetative forms are acid- labile, while the spores' are resistant to gastric acid.[7] This microorganism naturally resides in the gut as a commensal in a small percentage of the adult population, while others acquire such spores accidentally, when they are admitted as in -patients in a health care facility. The bacteria are usually transmitted indirectly via contamination through the fecal-oral route or through the spores left on the surfaces. Studies have shown that the rate of acquisition of such spores varies from 13% in patients with hospital stays of up to 2 weeks to 50% in stays longer than 4 weeks.[8]

Accidental ingestion of *C. difficile* can result in either excretion of the bacteria in feces or asymptomatic colonization of the bacteria in the gut of the ingested person, or may result in disease with diarrhea termed as colitis or pseudomembranous colitis (PMN). Though *C. difficile* has been found in approximately 3% of normal adults and in up to 40% of hospitalized patients,[9] only about one - third of them develop colitis, whereas the rest remain asymptomatic where the microbe remains in a metabolically inactive spore form[10] and that it only becomes a problem after such carriers are treated with antimicrobial agents.

Theoretically, all antimicrobials especially the broad-spectrum and antianaerobic -agents pose the greatest risk in altering the normal gut flora. Also some drugs like the fluoroquinolones, clindamycin and third-generation cephalosporins increase the risk of *C. difficile* colonization and overgrowth.[11] One study has found that fluoroquinolones alone were responsible for 55% of the reported *C. difficile* infection (CDI).[12] Other risk factors include advancing age, severe underlying illness, hospitalization, exposure to cytotoxic chemotherapy, and immunosuppressive therapy.[5]

Correlating CDAD with PPIs

Pseudomembranous colitis (PMC) a type of super infection caused by overgrowth of *C. difficile*, has been reported to be increasing in frequency and severity, with an estimated health care costs of over $1.1 billion in the United States each year.[13] This CDI can range in severity from mild or asymptomatic to severe and life - threatening infections, especially in the elderly patients and hospital in-patients.

The reduction in the gastric acidity with PPIs allows increased survival of both the vegetative and spore forms of *C. difficile*. The spores ultimately travel along the small bowel wherein the presence of bile salts[14] and the amino acid glycine[15] as a co-germinant factor, they germinate to their vegetative form and colonize in the lower gastrointestinal tract. In the colon they trigger an inflammatory response with or without the formation of pseudo membranes which may be responsible for the typical characteristic clinical features [Figure 2]. There is subsequent transmission of this microbe to other patients as well.

The use of PPIs has been observed to be associated with a two-fold increase in the risk of CDI[16] for the acid-labile vegetative form has been observed to survive in a PPI- induced raised gastric pH thereby favoring bacterial colonization in the upper G.I tract. So absent or decreased acid secretion due to any reasons can result in inadequate sterilization of ingested organisms and microbial colonization of the normally sterile upper G.I tract.[3] This fact is supported by the finding that when experimental animals were exposed to PPIs the severity of the resulting colitis and the risk of acquiring active CDI were similar to the observations made in affected

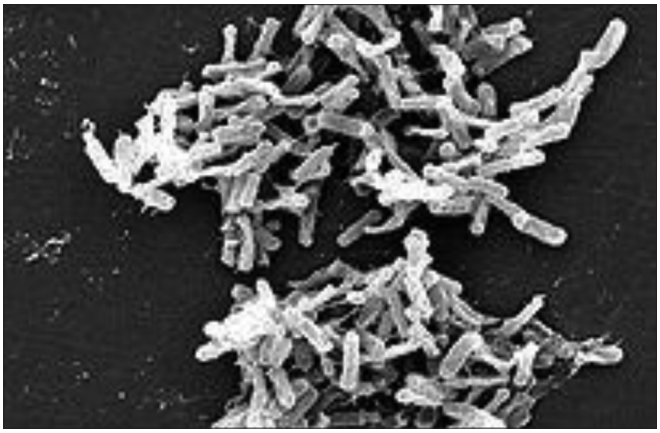

Figure 1: Microscopic picture of *C. difficile.*

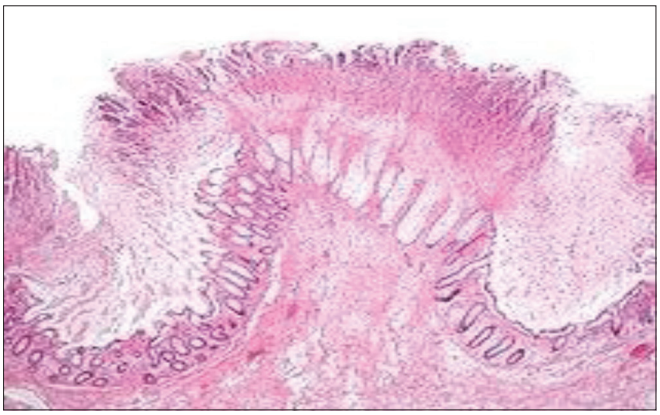

Figure 2: Microscopic picture of pseudomembranous colitis.

patients.[17] Also it may be assumed that concurrent use of PPIs along with antimicrobials could further increase and aggravate the susceptibility of such patients to CDI. This idea is supported by reports of a possible association between *C. difficile* diarrhea and acid - suppressive therapy and case reports of *C. difficile* diarrhea in patients with pernicious anemia and in patients receiving *Helicobacter pylori* treatment, which combines gastric acid suppressive therapy with antibiotics.[18]

Thus the normal acidity of the stomach is an important host defense mechanism against the ingested pathogens which thereby provides protection against a variety of enteric infections. Decreased gastric acidity is a known risk factor for other infectious diarrheal illnesses as well, such as travelers' diarrhea, salmonellosis and cholera, and because the survival of such microbes is facilitated by higher gastric pH levels.[19]

Evidence based studies suggesting correlation

There have been a number of studies suggesting a possible link between PPIs and an increased risk of CDAD, particularly in vulnerable patients.

1. CDI was increased by 1.7 with once-daily dose and by 2.4-fold with more than a single dose of PPIs, suggesting that the increased risk of CDI is associate`d with the use of PPIs[20]
2. In another cohort-based study, CDAD was evident in 81 (6.8%) subjects among the 1187 hospital in-patients who had received antibiotics along with PPIs[21]
3. In a multivariate analysis, CDAD was significantly associated with use of PPI (adjusted odds ratio [OR] 2.1, 95% confidence interval [CI] 1.2-3.5), receipt of 3 or more antibiotics (OR 2.1, 95% CI 1.3-3.4) and admission to a medical ward (OR 4.1, 95% CI 2.3-7.3)[22]
4. In a case-control study increased incidence of CDAD was associated with female patients (adjusted OR 2.1, 95% CI 1.1-4.0), previous history of renal failure (adjusted OR 4.3, 95% CI 1.5-11.9) or hospital admission (adjusted OR 2.6, 95% CI 1.4-5.2) and with the use of PPI (adjusted OR 2.7, 95% CI 1.4-5.2)[23]
5. Other evidenced-based studies have been presented in Table 1.

Clinical features

Usually this disease is characterized by the presence of typical colonic pseudo membranes and accounts for the most common cause of hospital-acquired diarrhea in developed countries, with an incidence of 0.1-2%.[24] The symptoms are due to the exotoxins of the bacteria, toxin A and toxin B, that are responsible for colonic mucosal injury and inflammation characteristics of PMC. These toxins are glucosyltransferases that target and inactivate the Rho family of the GTPases, resulting in disruption of the cytoskeleton of intestinal epithelial cells by the uridine diphosphate glucose–dependent glycosylation of Rho and Ras proteins that ultimately leads to loosening of the tight junctions which are crucial determinants of barrier function in intestinal epithelia.[25] There also occurs a cascade of inflammatory processes that involves release of destructive leukotrienes and cytokines resulting in tissue damage.

Thus, the symptoms include watery stool, abdominal pain, and fever, which may range from mild to severe form and, in rare cases, can progress to toxic megacolon, which can be complicating and fatal. Uncommon but serious complications include kidney failure, toxic megacolon, bowel perforation, and even death.

Diagnosis depends on the demonstration of *C. difficile* toxins in the stool, along with typical endoscopic finding of pseudo-membranes. However, in patients with underlying Crohn's disease or ulcerative colitis, pseudomembranous changes may not be seen; therefore, typical endoscopic findings of *C. difficile* in such patients may not be present, as the colonic mucosa will reflect only the underlying inflammatory bowel disease.

Mechanisms involved

The normal gastric pH provides a protective host defense mechanism by which the ingested pathogens are killed. By increasing the gastric pH, the PPIs may prevent the gastric content from killing the ingested *C. difficile* spores. But the C difficile spores are resistant to gastric acid, so the mechanism of the reported association between PPI therapy and CDI is not clear.

On the other hand, some studies suggest that germination of the ingested spores occurs in the small intestine and is stimulated by the presence of bile salts and glycine.[15]However, the signals triggering the germination of the *C. difficile* spores after ingestion are still not completely understood. But some other studies have stated the role of some unidentified confounding factors which might contribute to the pathophysiology of the disease.[14]

PPI therapy can have a direct effect on the expression of bacterial toxin gene production and regulation in *C. difficile*, which might be the causative factor of CDAD. *C. difficile* expresses various toxins that are responsible for causing damage to the intestinal mucosa. These are toxin A (*tcdA*), toxin B (*tcdB*), and a binary toxin (*cdtB*). The toxins A and B are the major virulence determinants of CDAD that are encoded by the *tcdA* and *tcdB* genes.[26] They disrupt the action of the Ras superfamily of small GTPases by modifying their active site via glycosylation, leading to disorganization of the actin cytoskeleton, which in turn can cause an increase in the permeability of the epithelial layers of the colon that attracts polymorph nuclear cells by chemotaxis to the colonic epithelium. This leads to a host of inflammatory responses that are seen in pseudomembranous colitis.

Table 1: Epidemiological evidences from studies

Year	Authors	Study	Parameters studied	Evidences
2003	Cunningham *et al*.	Proton pump inhibitors as a risk factor for *Clostridium difficile* diarrhoea	Case-control study	*C.difficile* diarrhea was associated with a higher rate of PPI use (OR 2.5, 95% CI 1.5-4.2)
2007	Jayatilaka *et al*.	*Clostridium difficile* infection in an urban medical center: Five-year analysis of infection rates among adult admissions and association with the use of proton pump inhibitors	Trends of incidence of CDAD among adults in an inner-city medical center	The usage of PPI correlated exactly with the increase in CDAD incidence
2008	Choudhry *et al*.	Overuse and inappropriate prescribing of proton pump inhibitors in patients with *Clostridium difficile*-associated disease	Studied PPI prescriptions of 138 hospitalized patients diagnosed with *C. difficile* infection over a 4-month period	Sixty-four percent (88 of 138) of all patients who developed *C.difficile* infections were on PPIs
2008	Aseeri and Schroeder	Gastric acid suppression by proton pump inhibitors as a risk factor for *Clostridium difficile*-associated diarrhea in hospitalized patients	Retrospective case-control study was conducted of all hospitalized patients at a local hospital	Seventy-two (76.6%) cases developed CDAD versus40 controls who did not develop the disease (42.6%) while on PPI
2010	Howell *et al*.	Iatrogenic gastric acid suppression and the risk of nosocomial *Clostridium difficile* infection	Pharmacoepidemiologic cohort study in which a secondary analysis of data collected prospectively on 101,796 discharges from a tertiary care medical center during a 5-year period was performed	The risk of nosocomial *C. difficile* infection increased from 0.3% (95% CI 0.21-0.31%) in patients not receiving acid suppressive therapy to 0.6% (95% CI 0.49-0.79%)
2010	Linsky *et al*.	Proton pump inhibitors and risk for recurrent *Clostridium difficile* infection	The hazard ratio for recurrent CDI, defined by a positive toxin finding in the 15-90 days after incident CDI	Recurrent CDI was more common in those exposed to PPIs than in those not exposed (25.2% vs. 18.5%)
2011	Pohl *et al*.	*Clostridium difficile* infection and proton pump inhibitor use in hospitalized pediatric cystic fibrosis patients	Incidence of *C. Difficile* infection in all pediatric hospital admissions over a 5-year period	*C. difficile* infection is more common in hospitalized pediatric cystic fibrosis patients, although PPI use may or may not be a risk factor for CDAD
2012	Rashid *et al*.	Inappropriate use of gastric acid suppression therapy in hospitalized patients with *Clostridium difficile*-associated diarrhea: A ten-year retrospective analysis	A retrospective analysis of in-patients	In 70% of patients acid suppressive therapy was a major causative factor

Abbreviations: CDAD: *C. difficile*-associated diarrhea; PPIs: Proton pump inhibitors; CDI: *C. difficile* infection; CI: Confidence interval; OR: Odds ratio

An *in vitro* study on the expression of these toxins has reported that at basic pH, there was a greater expression of toxin A, with a 120-fold higher expression when exposed to PPIs. Thus, low pH and PPIs resulted in the greater expression of toxin genes and their positive regulators.

Hence, PPIs might have a *direct* effect on *C. difficile*, perhaps by promoting toxin production or by inducing other virulence behaviors, apart from their effects on gastric pH. Thus PPIs that neutralize gastric acidity and make the medium basic can hence increase expression of the bacterial toxic genes.

As evidenced from the above study, the gene expression for toxin A was expressed to a much higher degree in alkaline environments and this effect was exaggerated when PPIs were added to the medium. Further, even transient exposure to an alkali medium in the stomach may enable *C. difficile* to more effectively colonize the colon and promote CDAD by stimulating toxin gene expression.[27]

Additionally, age, antidepressants, length of admission, and admission service all significantly increased the OR for incremental risk.

FDA and ethical issues

On 8 February 2012, the US Food and Drug Administration (FDA) issued a safety announcement on PPIs, which was based on the review report from Adverse Event Reporting System (AERS). Based on such reports, the FDA has alerted the health care professionals and patients that use of PPIs may increase the risk for CDAD in vulnerable subjects.[28]

Although the strength of the association varies widely among the reported studies, most studies have found the risk for CDI to be 1.4-2.75 times higher in patients with PPI exposure than in those without the drug.[29]

So, as stated by the FDA, certain guidelines need to be followed, which suggests that patients should be prescribed with the lowest possible dose and with the shortest duration of therapy with PPIs. They should immediately contact their health care professional and seek care if they are on PPIs and develop diarrhea that does not improve, and a diagnosis of CDAD should be considered in such cases.

The FDA is currently working with manufacturers to include information about the increased risk of CDAD with the use of PPIs in the drug labels.

Management

Discontinuation of the causative drug, and in severe or persistent diarrhea and colitis, oral metronidazole or vancomycin is usually the treatment of choice, but in 20% cases, there is a relapse of diarrhea. Other therapeutic options for CDAD are being developed, and drugs used for other infections are being studied as alternatives to metronidazole and vancomycin.

Nitazoxanide, a nitrothiazolide, acts by interfering with the anaerobic metabolic pathways in *C. difficile*.[30]

Tinidazole, a structural analog of metronidazole, has similar bioavailability and similar *in vitro* activity against *C. difficile* with fewer drug-related adverse effects.[31]

Difimicin, a novel 18-membered macrocycle antibiotic with little or no systemic absorption orally, has been tested well in patients with CDI.[32]

Rifaximin, another non-systemic antibiotic approved by the US FDA for travelers' diarrhea, is currently under evaluation for the treatment of CDAD.[33]

The recently approved fidaxomicin has been found to be equally effective as vancomycin.

Infection control measures, such as wearing gloves when caring for patients with CDAD, have been proven to be effective at prevention, and these work by limiting the spread of *C. difficile* in a hospital setting. In addition, washing with soap and water eliminates the spores from contaminated hands, but alcohol-based hand rubs are ineffective. Bleach wipes containing 0.55% sodium hypochlorite have been shown to kill the spores and prevent transmission between patients.

Conclusion

It may thus be postulated that use of PPIs can significantly contribute to outbreaks of *C. difficile* diarrhea by inadequate sterilization of the ingested organisms[34] resulting in increased numbers of susceptible hosts and carriers in a population.[35]

In the absence of a clear mechanism, the role of PPIs in the pathogenesis of CDI is controversial as some studies have not associated PPIs with *C. difficile* and the mechanisms by which these acid-suppressive medications promote CDI are also not clear.[36] Some studies have shown that PPIs also have some antimicrobial activity,[37] suggesting that a combination of both PPI and antimicrobials can result in a significantly increased risk. So, it may be prudent to withhold the use of PPI in patients on antimicrobials unless there are clear indications for the former.

While a definite association between PPI use and CDAD has not been confirmed, the possibility, however, cannot be ruled out. So, PPIs should be prescribed at the lowest possible dose and for the shortest duration of therapy appropriate to the condition being treated. A diagnosis of CDAD should be considered for any patient who has risk factors for CDAD and also in those who have persistent or severe diarrhea.

Hence, PPI may be an emerging and potentially modifiable risk factor for CDAD, which suggests the importance of vigilance in prescribing a PPI, particularly to patients who are hospitalized, taking multiple antibiotics, and suffering from multiple co-morbidities.

Since CDI is a global problem and the health care costs, morbidity, and mortality associated with CDAD are immense, a comprehensive infection control management rapid response team is recommended for each health care facility of the world.

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