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Effects of Short- and Long-course Antibiotics on the Lower Intestinal Microbiome as they Relate to Traveler's Diarrhea

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Abstract

Background—Antibiotics have profound and lasting effects on the lower intestinal (gut) microbiome that can both promote resistance and increase susceptibility to colonization and infection; knowledge of these changes is important to the prevention and treatment of traveler's diarrhea.

Methods—Recent data from epidemiologic and modern metagenomics studies were reviewed in regard to how such findings could inform the prevention and treatment of traveler's diarrhea.

Results—Although it is well recognized that antibiotics increase the risk for *Clostridium difficile* infection, it is less recognized how they predispose patients to typically foodborne pathogens such as *Salmonella* or *Camplyobacter* spp. While these pathogens account for only a fraction of traveler's diarrhea, such predisposition reflects how antibiotic exposure that precedes or occurs during travel may increase the risk for infection with other more common pathogens, even possibly enterotoxigenic *Eschericia coli*, especially in the setting of acquired resistance. Even short antibiotic exposures disrupt the gut microbiome up to a year or more and repeated exposures appear to attenuate recovery from ever occurring. One bacterial phylum that commonly increases in the gut following antibiotics are the proteobacteria including Enterobacteriacea; these are pro-inflammatory and often carry antibiotic resistance genes, the number and diversity of these genes (i.e. the resistome) commonly expands following antibiotics. The gut resistome among healthy community-dwelling adults reflects geographic variability in antibiotic use practices in both humans and food-producing animals as well as possibly the transmission of antibiotic resistance genes through the food supply.

Conclusions—Because antibiotic use among travelers will influence the resistome and thereby promote geographic spread of resistance, it is important that antibiotic use recommendations for travelers be guided by resistance surveillance data as well as a careful assessment of the risks and benefits to both the individual and society.

Introduction

Of over one billion international travelers a year, about 300 million travel from relatively industrialized regions with high levels of sanitation to less industrialized regions where

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travelers' diarrhea occurs in 20–50% [1–3], Among patients with traveler's diarrhea, anywhere from 5% to 62% may take antibiotics despite most cases being mild to moderate in severity.Antibiotics deplete bacterial diversity in the lower intestine (or gut), alter gene expression, select for intrinsically resistant bacteria, and select for new mutations and gene transfers conferring resistance [4]. The depletion of bacterial diversity has attracted probably the most attention in the medical and lay literature as it is associated in animal models with chronic conditions such as compromised immune homeostasis and atopy, as well as deregulated metabolism, obesity, metabolic syndrome, and diabetes. The role of shifts in bacterial diversity and composition in increasing susceptibility to infection and the accumulation of antibiotic resistance is probably less widely understood. The poster-child of an infection with both intrinsic and acquired antibiotic resistance that is widely-accepted to be 'caused' by antibiotics is *Clostridium difficile* infection (CDI). Indeed, the recurrent form of CDI is now frequently managed through microbiome restoration in the form of fecal microbiota transplantation (FMT).

Colonization Resistance and Infection by Pathobionts

Like *C. difficile*, there are a number of other bacterial pathogens that similarly can either only cause disease, or are much more likely to cause disease, after the human or animal host has received antibiotics that disrupt the gut microbiome. Such bacteria can be considered pathobionts: a symbiont that does not normally elicit an inflammatory response but under particular environmentally induced conditions (e.g. factors that disrupt the gut microbiome) has the potential to cause dysregulated inflammation leading to disease [5]. These opportunists stand in contrast to the vast number of true commensal members of the gut microbiota, mostly obligate anaerobes, that are thoroughly avirulent, even in the presence of a severely disrupted microbiome. In addition to *C. difficile*, pathobionts include the typically food-borne pathogens identified in a minority of cases of traveler's diarrhea, for example non-typhoid *Salmonella* spp. [6] and *Camplylobacter* spp.[7]. However, early data suggest that even the much more common bacterial cause of traveler's diarrhea, Enterotoxigenic *Escherichia coli* (ETEC), may also function as a pathobiont, in that there may be unique individual or collective sets of commensal bacteria that protect some humans from disease following challenge [8] and animal models suggest a role for probiotics in the prevention of ETEC diarrhea [9].

The important role of prior treatment with antibiotics, in this case a large dose of streptomycin, in increasing the susceptibility of mice to intestinal infection with *Salmonella enteriditis* was first recognized in 1964—an effect thought mediated by elimination of certain anaerobes [10]. More recently, in a large combined case-control and source attribution analysis of human salmonellosis cases, Muchinini-Gras et al. showed that recent use of antibiotics had a population attributable risk similar to that of eating raw or undercooked eggs [11]. In a one-year nested case-control study in Denmark the weekly odds ratio (OR) of having received antibiotics prior to onset of salmonella infection, after controlling for Charlson comorbidity index, was about 1.5 for receipt between 13 and 52 weeks, with a sharp increase in odds to above 2.0 over the 2–12 weeks before illness [12]. In another case-control study from Denmark during an era when campylobacter resistance to fluoroquinolones was 21.7%, and to macrolides was 2.3%, Koningstein et al. found the OR

of exposure up to one-year before infection to be 1.48 (95% Confidence Interval [95%CI] 1.38–1.59) for macrolides and 2.42 (95%CI 1.96–2.98) for fluoroquinolones [13]. Interestingly, in contrast to fluoroquinolones, the macrolides appeared protective (OR 0.72 95%CI 0.56–0.92) against campylobacter infection when administered in the month prior to illness—a finding that likely reflects the lower rate of acquired macrolide resistance, the prolonged effective half-life of some of the macrolides, and the fact that these shorter windows of exposure included the 'acute' beginning phase of illness among the cases.

What begins to emerge from these studies is that prior antibiotic exposure increases the overall risk for infection regardless of acquired resistance in the organism and, as resistance to the antibiotic increases in the population, even acute exposure (i.e. that bordering on treatment) may propel rather than retard the infection. Although treatment with an antibiotic to which the offending pathogen is resistant has never been demonstrated to worsen or prolong infection in a randomized controlled study of antibiotics for traveler's diarrhea, neither is it the goal of such studies to examine such a role for antibiotics in these studies—instead special care is taken to study antibiotics in populations where resistance is not a problem. Nonetheless, such potential interaction between rates of acquired resistance and the net benefits vs. harms of antibiotics used in prophylaxis and early treatment of diarrhea may be especially important to consider in travelers' diarrhea. Recent data from CDC's Foodnet and National Antimicrobial Resistance Monitoring Systems suggest a fourfold to 11-fold higher likelihood of fluoroquinolone resistance in recent traveler vs. non-travel related diarrhea depending upon the pathogen (*Camplyobacter* spp., *Salmonella* spp., *Shigella* spp., and shigella-toxin producing *E. coli*) (unpublished data and [14–16]). Moreover, rates of fluoroquinolone resistance in recent travelers range from 11% (*Shigella* spp.) up to 61% (*Campylobacter* spp.).

Epidemiologic data indicating infection risk from antibiotics, in addition to having important implications for how one approaches the prevention and management of traveler's diarrhea, highlight the importance of resistance to colonization and infection afforded by an intact gut microbiome. The mechanisms for such resistance fall into one or more of four broad categories [17]. First, the direct inhibition afforded by production of antimicrobial substances by one or more members of the microbiota against others. Second, the role of the microbiome in maintaining the mucus and mucosal barrier that prevents mucosal attachment and invasion where such processes are necessary in pathogenesis. Third, the role of the microbiome in immune modulation as certain members of the microbiota up- and down-regulate the host immune system in defense against invasive species. Finally, there is nutrient utilization, as members of the microbiota out-compete one another in the ecology of the gut. In addition to these, there may be direct signaling pathways that are maintained by the intact microbiome in a certain state that inhibits a pathobiont. For example, the abundance of primary bile acids are an important signal for the germination of *C. difficil*e from the spore into the actively growing and toxin-producing vegetative form [18]. Meanwhile, secondary bile acids inhibit the growth of vegetative *C. difficile* and bile salts normally undergo conjugation from primary to secondary form by anaerobic bacteria found in the intact gut microbiota. Thus a key metabolic function of the gut microbiome is bile salt metabolism that normally defends against CDI but is disrupted by antibiotics [18]. There

may be other similarly specific mechanisms that lie behind the colonization resistance afforded by the gut microbiome against other pathobionts.

Antibiotic Effects on the Microbiome

Recent advances in next generation sequencing and metagenomics make it possible to build upon epidemiologic data to understand the impact of antibiotics on the risk of infection and antibiotic resistance. Using 16S ribosomal DNA amplification and sequencing, Dethlefsen et al. intensively sampled 3 healthy individuals before, during, and after two 5-day courses of oral ciprofloxacin separated by six months and found that antibiotics profoundly reduced the number of different species and/or genus (i.e. 'Operational Taxonomic Units' [OTUs]) as well as the diversity of those remaining [19]. While good recovery was observed in all three subjects over the six months before and even after the second course, recovery was slower following the second course and one subject did not recover fully following the second dose suggesting the microbiome of some patients may become permanently disabled from repeated antibiotic exposures. Microbiome changes associated with repeated, major shifts in diets fed to mice also indicate that, despite good resiliency following individual insults, there may be attenuated recovery of the microbiome following multiple, sequential insults [20].

The longer term (i.e. up to one year) impacts of antibiotics on the gut microbiome were studied in 30 healthy volunteers including three groups of 10 who were administered a 10-day course of either placebo, ciprofloxacin or clindamycin [21]. Both 16S and culture were used to assess microbiome structure. Most pronounced disruptions were noted on day 11 with statistically significant changes from baseline in 15 OTUs following ciprofloxacin and 21 OTUs following clindamycin. Despite receipt of no additional antibiotics over four months, there remained significant differences from baseline in two different OTUs following exposure to each antibiotic and at 12 months there was still one OTU that was significantly different from baseline following exposure to clindamycin. The authors concluded that it took 4–12 months for the gut microbiome to normalize following either antibiotic and that clindamycin had a more pronounced effect than ciprofloxacin.

One antibiotic-induced bacterial shift associated with an expansion of clinically important forms of antibiotic resistance in the gut is a 'bloom' of the gammaproteobacteria; especially of the family enterobacteriaceae that normally make up less than 1–3% of the gut microbiome [19, 21–24]. In a recent study of the effects of FMT on the resistome, FMT was associated with a reduction in the number of different categories of resistance as well as the number of individual determinants [23]. This contraction in the resistome was associated with an increase in the genus- and species-level diversity of the microbiota overall and a decrease in enterobacteriaceae, reflecting what is likely an increased concentration of antibiotic resistance determinants in this family of organisms. In addition to their role in antibiotic resistance, increases in enterobacteriaceae in the microbiome are associated with inflammation and metabolic syndrome [25]. Thus it is of some import to understand and possibly predict how antibiotic exposures will promote increases in the enterobacteriaceae.

Raymond et al. studied exactly this in 18 healthy subjects, including six controls, who were administered a 7-day course of cefprozil and had their microbiome assessed using deep

sequencing at 0, 7, and 90 days [22]. At baseline (day 0) the subjects were found to broadly fit into four clusters that appeared to be a variation on three previously described enterotypes [26]. Interestingly, all 6 subjects who started out in a single one of these enterotypes had an expansion of *Enterobacter cloacae* complex—a frequent cause of healthcare-associated infections. Meanwhile, there was a decrease in 6 bacterial families across all antibiotic-treated subjects, an increase in *Lachnoclostridium bolteae* among 16 subjects, and an overall expansion of the resistome, especially beta-lactamases that confer important forms of clinically relevant antibiotic resistance.

The 'Perfect Storm' of Antibiotics and Travel

The ability to measure the GI resistome across a sample of subjects from different global geographic regions has shed light on possibly why travelers returning from various regions are at increased risk of colonization with clinically important forms of emerging resistance. Forslund et al showed how country-specific antibiotic use practices impact the human gut resistome by studying the resistome of individuals across several countries; among residents from the United States, Denmark, and Spain specifically they showed higher 'resistance potential' in the gut microbiome based upon a measure of the resistome was associated with more permissive antibiotic use practices in both humans and animals [27]. By combining these data with data from France and Italy, they showed differences in resistance among bacterial isolates collected from meat slaughterhouses were associated with differences in the human gut resistome, suggesting the food supply is an important source of such resistance. These same factors interplay in travelers, with antibiotic use adding to the dysbiosis that is caused by travel itself [28], making them susceptible to expansion of an endogenous and acquired resistome, acquisition of specific resistant bacteria phenotypes, and infection caused by specific diarrhea-causing pathobionts [29].

In conclusion, antibiotic-induced dysbiosis can last up to one year and repeated insults may attenuate its recovery. These antibiotic effects on the microbiome include a decrease in diversity, expansion of the resistome, loss of keystone species that support complex ecologies, loss of protective species against specific pathogens, promotion of colonization by pathobionts, and promotion of dominance that can be a driver for some types of infection and transmission. Given observations that antibiotic exposures can increase risk of infection by foodborne pathogens, especially in the setting of resistance to a given antibiotic, there is likely to be some proportion of all travelers' diarrhea days of illness that are currently being contributed to, rather than prevented by, either prophylaxis or treatment of traveler's diarrhea with antibiotics. Meanwhile, as demonstrated in studies of antibiotics for traveler's diarrhea, there are clearly days of illness that are currently being prevented, either through effective prophylaxis in high-risk persons or early self-treatment that shortens the course of diarrhea [30]. However, there are other adverse events related to antibiotics that must also be considered, including community-associated *C. difficile* infection that has been reported in antibiotic-exposed travelers [31], as well as other adverse drug events [32]. Focusing just on traveler's diarrhea, the shift from overall benefit to harm caused by antibiotics may be dependent upon both the timing of antibiotic exposure in relation to exposure to the pathobiont (i.e. prior antibiotic exposures appear to uniformly increase risk, except possibly for drugs with effectively prolonged half-life, such as azithromycin) and increased

prevalence of acquired resistance in the pathobionts responsible for travelers' diarrhea (i.e. exposure to an antibiotic to which the pathobiont is resistant is likely to result in net harm regardless of timing before or during acute illness). It is therefore imperative, to both prevent current harm and preserve future antibiotic activity for generations to come, that antibiotic treatment recommendations be guided by surveillance data for resistance as well as careful individual and societal risk-benefit analyses. In addition to the need for ongoing real time surveillance to inform such recommendations, there is a need for additional research into how to better harness our growing understanding of the microbiome to prevent of travelers' diarrhea while simultaneously reducing the development and spread of resistance.

References

1. Kantele A, Laaveri T, Mero S, Vilkman K, Pakkanen SH, Ollgren J, Antikainen J, Kirveskari J. Antimicrobials increase travelers' risk of colonization by extended-spectrum betalactamase-producing Enterobacteriaceae. Clin Infect Dis. 2015; 60(6):837–46. [PubMed: 25613287]
2. Belderok SM, van den Hoek A, Kint JA, Schim van der Loeff MF, Sonder GJ. Incidence, risk factors and treatment of diarrhoea among Dutch travellers: reasons not to routinely prescribe antibiotics. BMC Infect Dis. 2011; 11:295. [PubMed: 22035314]
3. Pitzurra R, Steffen R, Tschopp A, Mutsch M. Diarrhoea in a large prospective cohort of European travellers to resource-limited destinations. BMC Infect Dis. 2010; 10:231. [PubMed: 20684768]
4. Francino MP. Antibiotics and the Human Gut Microbiome: Dysbioses and Accumulation of Resistances. Front Microbiol. 2015; 6:1543. [PubMed: 26793178]
5. Round JL, Mazmanian SK. The gut microbiota shapes intestinal immune responses during health and disease. Nat Rev Immunol. 2009; 9(5):313–23. [PubMed: 19343057]
6. Ayres JS, Trinidad NJ, Vance RE. Lethal inflammasome activation by a multidrug-resistant pathobiont upon antibiotic disruption of the microbiota. Nat Med. 2012; 18(5):799–806. [PubMed: 22522562]
7. Reti KL, Tymensen LD, Davis SP, Amrein MW, Buret AG. Campylobacter jejuni increases flagellar expression and adhesion of noninvasive Escherichia coli: effects on enterocytic Toll-like receptor 4 and CXCL-8 expression. Infect Immun. 2015; 83(12):4571–81. [PubMed: 26371123]
8. Pop M, Paulson JN, Chakraborty S, Astrovskaya I, Lindsay BR, Li S, Bravo HC, Harro C, Parkhill J, Walker AW, Walker RI, Sack DA, Stine OC. Individual-specific changes in the human gut microbiota after challenge with enterotoxigenic Escherichia coli and subsequent ciprofloxacin treatment. BMC Genomics. 2016; 17:440. [PubMed: 27277524]
9. Yang KM, Jiang ZY, Zheng CT, Wang L, Yang XF. Effect of Lactobacillus plantarum on diarrhea and intestinal barrier function of young piglets challenged with enterotoxigenic Escherichia coli K88. J Anim Sci. 2014; 92(4):1496–503. [PubMed: 24492550]
10. Bohnhoff M, Miller CP, Martin WR. Resistance of the Mouse's Intestinal Tract to Experimental Salmonella Infection. I. Factors Which Interfere with the Initiation of Infection by Oral Inoculation. J Exp Med. 1964; 120:805–16. [PubMed: 14247721]
11. Mughini-Gras L, Enserink R, Friesema I, Heck M, van Duynhoven Y, van Pelt W. Risk factors for human salmonellosis originating from pigs, cattle, broiler chickens and egg laying hens: a combined case-control and source attribution analysis. PLoS One. 2014; 9(2):e87933. [PubMed: 24503703]
12. Gradel KO, Dethlefsen C, Ejlertsen T, Schonheyder HC, Nielsen H. Increased prescription rate of antibiotics prior to non-typhoid Salmonella infections: a one-year nested case-control study. Scand J Infect Dis. 2008; 40(8):635–41. [PubMed: 18979601]
13. Koningstein M, Simonsen J, Helms M, Hald T, Molbak K. Antimicrobial use: a risk factor or a protective factor for acquiring campylobacteriosis? Clin Infect Dis. 2011; 53(7):644–50. [PubMed: 21890769]
14. Ricotta EE, Palmer A, Wymore K, Clogher P, Oosmanally N, Robinson T, Lathrop S, Karr J, Hatch J, Dunn J, Ryan P, Blythe D. Epidemiology and antimicrobial resistance of international travel-

associated Campylobacter infections in the United States, 2005-2011. Am J Public Health. 2014; 104(7):e108–14.

15. O'Donnell AT, Vieira AR, Huang JY, Whichard J, Cole D, Karp BE. Quinolone-resistant Salmonella enterica serotype Enteritidis infections associated with international travel. Clin Infect Dis. 2014; 59(9):e139–41. [PubMed: 24973311]
16. Shiferaw B, Solghan S, Palmer A, Joyce K, Barzilay EJ, Krueger A, Cieslak P. Antimicrobial susceptibility patterns of Shigella isolates in Foodborne Diseases Active Surveillance Network (FoodNet) sites, 2000-2010. Clin Infect Dis. 2012; 54(Suppl 5):S458–63. [PubMed: 22572670]
17. McKenney PT, Pamer EG. From Hype to Hope: The Gut Microbiota in Enteric Infectious Disease. Cell. 2015; 163(6):1326–32. [PubMed: 26638069]
18. Theriot CM, Bowman AA, Young VB. Antibiotic-Induced Alterations of the Gut Microbiota Alter Secondary Bile Acid Production and Allow for Clostridium difficile Spore Germination and Outgrowth in the Large Intestine. mSphere. 2016; 1(1)
19. Dethlefsen L, Relman DA. Incomplete recovery and individualized responses of the human distal gut microbiota to repeated antibiotic perturbation. Proc Natl Acad Sci U S A. 2011; 108(Suppl 1): 4554–61. [PubMed: 20847294]
20. Carmody RN, Gerber GK, Luevano JM Jr, Gatti DM, Somes L, Svenson KL, Turnbaugh PJ. Diet dominates host genotype in shaping the murine gut microbiota. Cell Host Microbe. 2015; 17(1): 72–84. [PubMed: 25532804]
21. Rashid MU, Zaura E, Buijs MJ, Keijser BJ, Crielaard W, Nord CE, Weintraub A. Determining the Long-term Effect of Antibiotic Administration on the Human Normal Intestinal Microbiota Using Culture and Pyrosequencing Methods. Clin Infect Dis. 2015; 60(Suppl 2):S77–84. [PubMed: 25922405]
22. Raymond F, Ouameur AA, Deraspe M, Iqbal N, Gingras H, Dridi B, Leprohon P, Plante PL, Giroux R, Berube E, Frenette J, Boudreau DK, Simard JL, Chabot I, Domingo MC, Trottier S, Boissinot M, Huletsky A, Roy PH, Ouellette M, Bergeron MG, Corbeil J. The initial state of the human gut microbiome determines its reshaping by antibiotics. ISME J. 2016; 10(3):707–20. [PubMed: 26359913]
23. Millan B, Park H, Hotte N, Mathieu O, Burguiere P, Tompkins TA, Kao D, Madsen KL. Fecal Microbial Transplants Reduce Antibiotic-resistant Genes in Patients With Recurrent Clostridium difficile Infection. Clin Infect Dis. 2016; 62(12):1479–86. [PubMed: 27025836]
24. Stecher B, Maier L, Hardt WD. 'Blooming' in the gut: how dysbiosis might contribute to pathogen evolution. Nat Rev Microbiol. 2013; 11(4):277–84. [PubMed: 23474681]
25. Cani PD, Amar J, Iglesias MA, Poggi M, Knauf C, Bastelica D, Neyrinck AM, Fava F, Tuohy KM, Chabo C, Waget A, Delmee E, Cousin B, Sulpice T, Chamontin B, Ferrieres J, Tanti JF, Gibson GR, Casteilla L, Delzenne NM, Alessi MC, Burcelin R. Metabolic endotoxemia initiates obesity and insulin resistance. Diabetes. 2007; 56(7):1761–72. [PubMed: 17456850]
26. Arumugam M, Raes J, Pelletier E, Le Paslier D, Yamada T, Mende DR, Fernandes GR, Tap J, Bruls T, Batto JM, Bertalan M, Borruel N, Casellas F, Fernandez L, Gautier L, Hansen T, Hattori M, Hayashi T, Kleerebezem M, Kurokawa K, Leclerc M, Levenez F, Manichanh C, Nielsen HB, Nielsen T, Pons N, Poulain J, Qin J, Sicheritz-Ponten T, Tims S, Torrents D, Ugarte E, Zoetendal EG, Wang J, Guarner F, Pedersen O, de Vos WM, Brunak S, Dore J, Meta HITC, Antolin M, Artiguenave F, Blottiere HM, Almeida M, Brechot C, Cara C, Chervaux C, Cultrone A, Delorme C, Denariaz G, Dervyn R, Foerstner KU, Friss C, van de Guchte M, Guedon E, Haimet F, Huber W, van Hylckama-Vlieg J, Jamet A, Juste C, Kaci G, Knol J, Lakhdari O, Layec S, Le Roux K, Maguin E, Merieux A, Melo Minardi R, M'Rini C, Muller J, Oozeer R, Parkhill J, Renault P, Rescigno M, Sanchez N, Sunagawa S, Torrejon A, Turner K, Vandemeulebrouck G, Varela E, Winogradsky Y, Zeller G, Weissenbach J, Ehrlich SD, Bork P. Enterotypes of the human gut microbiome. Nature. 2011; 473(7346):174–80. [PubMed: 21508958]
27. Forslund K, Sunagawa S, Kultima JR, Mende DR, Arumugam M, Typas A, Bork P. Country-specific antibiotic use practices impact the human gut resistome. Genome Res. 2013; 23(7):1163–9. [PubMed: 23568836]
28. Youmans BP, Ajami NJ, Jiang ZD, Campbell F, Wadsworth WD, Petrosino JF, DuPont HL, Highlander SK. Characterization of the human gut microbiome during travelers' diarrhea. Gut Microbes. 2015; 6(2):110–9. [PubMed: 25695334]

29. Riddle MS, Connor BA. The Traveling Microbiome. Curr Infect Dis Rep. 2016; 18(9):29. [PubMed: 27447891]

30. Steffen R, Hill DR, DuPont HL. Traveler's diarrhea: a clinical review. JAMA. 2015; 313(1):71–80. [PubMed: 25562268]

31. Neuberger A, Saadi T, Shetern A, Schwartz E. Clostridium difficile Infection in travelers–a neglected pathogen? J Travel Med. 2013; 20(1):37–43. [PubMed: 23279229]

32. Shehab N, Patel PR, Srinivasan A, Budnitz DS. Emergency department visits for antibiotic-associated adverse events. Clin Infect Dis. 2008; 47(6):735–43. [PubMed: 18694344]