Intra-Abdominal Anaerobic Infections: Bacteriology and Therapeutic Potential of Newer Antimicrobial Carbapenem, Fluoroquinolone, and Desfluoroquinolone Therapeutic Agents

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Intra-abdominal infections are biphasic, synergistic processes with early peritonitis and bacteremia due to aerobes and a later abscess component due to anaerobes. Although Bacteroides fragilis is the most commonly recognized pathogen, other anaerobes, including other members of the B. fragilis–group species, are major components of infection. Anaerobic bacteremia is often associated with an intra-abdominal source. New antimicrobial agents with anaerobic activity are in various stages of development for the therapy of intra-abdominal infections. The in vitro activity and the currently available sparse clinical data are reviewed for a new carbapenem (ertapenem), several fluoroquinolones (trovafloxacin, moxifloxacin, and gemifloxacin), and a desfluoroquinolone (BMS-284756).

Intra-abdominal infection includes a wide variety of markedly different conditions, ranging from primary and secondary peritonitis to intrahepatic infection to diverticulitis, appendicitis, and intra-abdominal abscess, that are lumped together because of anatomical coincidence. The most serious infections are due to viscus inflammation and perforation and almost always involve a mixture of aerobic and anaerobic intestinal flora. The intestinal colonic flora contains $10^{12}$ bacteria/gm of feces, which are predominantly anaerobic, and anaerobic species outnumber aerobes by 1000 to 1. Although anaerobes are ubiquitous commensals and aid in “colonization resistance,” some genera and species are also consummate opportunistic pathogens. Bacteroides fragilis, which accounts for only 0.5% of the normal colonic flora, is recognized as the single most important anaerobic pathogen. Properties that contribute to its virulence include its capsular polysaccharide, adherence potential, piliation, and toxin production. All colorectal surgeons who responded to a survey [1] reported the use of preoperative bowel preparations prior to surgery, and 86.5% add oral and parenteral antimicrobials to the regimen to prevent infection.

Once peritoneal contamination occurs, the body attempts to defend against this invasion via lymphatic clearance, phagocytosis, sequestration of fibrin, and anatomical localization. Anaerobes act synergistically with facultative organisms to induce abscess formation more readily than either mixtures of facultative or anaerobic bacteria alone and may protect the aerobes from phagocytosis and other body defenses. The facultative bacteria also promote the infectious process by lowering the environment’s oxidation-reduction potential and facilitating the growth of anaerobes.

BIPHASIC MIXED INFECTION

Beginning in 1973, Onderdonk et al. [2, 3] developed an animal model to simulate intra-abdominal sepsis. They noted that a biphasic infection developed. After
fetal spillage, peritonitis developed initially and was associated with a predominance of aerobes and Escherichia coli bacteremia and a significant (60%) mortality in untreated animals. Subsequently, abscesses developed in the surviving animals and were associated with a predominance of anaerobes, especially B. fragilis. Using selected antimicrobial probes, they further showed that suppression of the facultative bacteria such as E. coli diminished the early mortality but did not affect abscess formation in survivors. Conversely, suppression of the obligate anaerobes, including B. fragilis, decreased the frequency of abscess formation but did not affect the early mortality. This demonstrated the importance of both the aerobic and anaerobic components of the intra-abdominal infection process.

Physicians must often use both medical and surgical forms of therapy by draining pus, debriding necrotic tissue, and administering appropriate antimicrobials. Clinical criteria that describe patients with acute perforated appendicitis include symptoms >24 h, temperature >38.6°C, and a white blood cell count >14,000 cells/mm³. In a study of 175 patients undergoing appendectomy, Browder et al. [4] noted that “surgeons’ reports significantly underestimated the diagnosis when compared to the pathologists’ report.” Most errors were for appendices categorized as acute, and, when examined by the pathologists, 14% were found to be “suppurative, necrotizing or microscopically perforated.” They found that operative culture results were not predictive of infection and recommended 1 dose of antimicrobial prophylaxis for “all patients undergoing surgery for nonperforative appendicitis.”

**RELATIVE FREQUENCY OF ANAEROBES**

B. fragilis is one of the most recognized anaerobic pathogens and is the anaerobe most often isolated from anaerobic bacteremia as well as intra-abdominal infections. Yet it is but one member of a group of isolates that include other virulent pathogens such as Bacteroides thetaiotaomicron, Bacteroides distasonis, Bacteroides vulgatus, Bacteroides ovatus, and Bacteroides uniformis, and each has its own ecological niche. In a study done at 2 community medical centers, Goldstein et al. [5] noted that the B. fragilis group accounted for 34.6% of all anaerobes isolated (relative frequency), of which B. fragilis itself was the most common isolate and accounted for 18.9%. B. fragilis was more likely to be associated with bacteremia and accounted for 46% of intra-abdominal isolates. Although community hospital laboratories do not usually attempt to isolate or identify the majority of anaerobes in clinical mixed specimens because of costs and other considerations, a recent study from 17 countries worldwide on the anaerobic bacteriology of intra-abdominal infections showed similar results [6]. We [6] isolated 1001 anaerobes from 427 clinical specimens that yielded an average of 3 aerobes (range, 0–9) and 2.3 anaerobes (range, 0–13) per specimen. The B. fragilis–group species accounted for 455 (45.5%) of 1001 anaerobes isolated, of which B. fragilis accounted for 134 strains (135/1001, 13.4%) and was present in 31.4% of all intra-abdominal specimens. These results compared favorably with those of a survey of the literature [7] that found an overall mean of 1.2 aerobes and 0.9 anaerobes per patient specimen. Bennion et al. [7] studied 30 patients with gangrenous (n = 12) and perforated (n = 18) appendicitis and found 2.7 aerobes and 7.4 anaerobes recovered per specimen. Escherichia coli was by far the most frequent aerobic isolated and was present in 92% and 78% of gangrenous and perforated appendicitis specimens, respectively. B. fragilis was found in 7 (58%) of 12 cases with gangrenous appendicitis and in 15 (83%) of 18 of cases of perforated appendicitis. However, their specimens included appendiceal tissue, although they excluded the lumen as well as peritoneal fluid and abscess contents. Brook [8] found similar results for children with perforated appendicitis. In their study, Bennion et al. [7, 9, 10] isolated a previously undescribed and distinct anaerobic bacteria, now named Bilophila wadsworthia, in approximately half the patients. Using many of these same cohort of patients, Baron et al. [11] compared the microbiology of acute and complicated appendicitis with several controls and concluded that “some bacteria traverse the intact appendiceal wall prior to perforation and that progressive infection and subsequent tissue damage and necrosis” allows increased genera and species of bacteria to traverse into the peritoneal cavity. Amazingly, “on a population basis, diagnosis of appendicitis has not improved with the availability of advanced diagnostic testing” [12]. Approximately 2 million intra-abdominal procedures are performed each year in the United States. If one estimates that there is a 15% infection rate, including a 4% incidence of abscess formation, it has been extrapolated that the yearly economic impact of abscess alone is >$1 billion [3].

**ASSOCIATED BACTEREMIA**

Anaerobic bacteremia has long been associated with intra-abdominal processes. It accounts for ~4% (0.5%–9%) of all bacteremias or ~1 case per 1000 admissions, with various geographic, demographic, and especially age-related variability [13]. A review of the distribution of species found B. fragilis to account for 17%–42%, B. thetaiotaomicron for 5%–14%, clostridia for 11%–36%, peptostreptococci for 8%–21%, and fusobacteria for 4%–8% of isolates. In a variety of studies, the gastrointestinal tract was found to be the source of 49% of anaerobic bacteremias [14], and an additional 6% had no defined source. Brook [15] evaluated the source of anaerobic bacteremias over a 12-year period from a military hospital and found that the gastrointestinal tract was the principal source of B. fragilis and clostridial bacteremias. Redondo et al. [16]...
reviewed data from 1982–1993 and reported that 69% of the B. fragilis group bacteremias emanated from an abdominal source. Approximately 25% of patients studied underwent surgery of the large bowel, 5% had small bowel surgery, 9% underwent appendectomy, and 9% underwent exploratory laparotomy. In addition, their controlled, matched-pair study showed an attributable mortality of 19.3% with a mortality risk ratio of 3.2 and a 16-day longer hospital stay. Associated risks included diabetes, receipt of immunosuppressive therapy, and mild to moderate renal dysfunction. Claros et al. [17] confirmed the report of Kato et al. [18] that B. fragilis blood culture isolates are more likely to carry the enterotoxin gene than strains isolated from other sources and that the enterotoxin production may contribute to the virulence properties and pathogenicity of B. fragilis strains even in nondiarrheal diseases. A recent multicenter survey of anaerobes isolated between 1998 and 1999 noted that the 51 blood culture isolates were more resistant than the other 505 isolates recovered from other sources [19]. In a prospective study of Bacteroides bacteremia [20], it has been shown that the in vitro activity of the antimicrobial agent selected against a bacteremic strain of B. fragilis “reliably predicts outcome” with a 97% specificity and an 82% positive predictive value.

ANTIMICROBIAL THERAPY

Most clinical laboratories do not perform routine susceptibility testing on anaerobic bacteria, even when isolated from the blood, and rely on published studies and periodic surveys [21]. Although the E-test has made susceptibility testing of individual strains possible [22], it is often reserved until requested by a clinician. As a consequence, the selection of an antimicrobial agent is often empirical. Montravers et al. [23] studied the impact of empirical antimicrobial selection in 100 consecutive cases of postoperative peritonitis from 1987–1992 and found that inadequate empirical selection was associated with a worse outcome and a higher mortality. They also noted that “late changes” in antimicrobial therapy based on culture results “did not affect the outcome when the initial regimen was inadequate.” This suggests that empirical, broad-based effective therapy is essential and that later results could be amended to lesser active agents pending cultural data.

A variety of clinical trials and years of clinical use have established the efficacy of ampicillin-sulbactam, piperacillin tazobactam, imipenem, and meropenem in mixed aerobic anaerobic intra-abdominal infections [24–26]. The continued in vitro activity of these and other agents has been reviewed by Snydman et al. [27], who have published a number of multicenter surveys on the B. fragilis group species, the latest of which covers 1997–2000 and is published in this issue [28]. That report surveys the activity of 14 antimicrobial agents, including 3 cephemycins, clindamycin, chloramphenicol, ertapenem, and 2 fluoroquinolones against 2673 isolates of the B. fragilis group. They note that metronidazole, imipenem, meropenem, ampicillin-sulbactam, piperacillin tazobactam, and ticarcillin clavulanate maintained their excellent anti-anaerobic activity, whereas trovafloxacin and clinafloxacin showed increased geometric mean MICs. Although imipenem resistance due to metallo–β-lactamase has been reported from Japan in the past [29], it does not appear to have spread to the United States or increased in frequency. Hopkins et al. [30] studied the activity of imipenem against 126 anaerobes isolated from intra-abdominal infections and found 1 strain of B. thetaotaomicron resistant to imipenem, 1 strain of B. fragilis that had intermediate susceptibility, and 1 strain that required 4 μg/mL for inhibition. Aldridge et al. [19] reported on a multicenter susceptibility survey of anaerobic isolates collected from 1998–1999, including 346 from abdominal sources, and found all intra-abdominal isolates to be susceptible to piperacillin tazobactam (MIC<sub>90</sub>, 4 μg/mL) and imipenem (MIC<sub>90</sub>, 0.25 μg/mL), whereas ≥99% were susceptible to meropenem (MIC<sub>90</sub>, 0.5 μg/mL), and metronidazole (MIC<sub>90</sub>, 2 μg/mL). They noted increasing resistance to ampicillin-sulbactam (MIC<sub>90</sub>, 8 μg/mL, 92% susceptible), cefoxitin (MIC<sub>90</sub>, 16 μg/mL, 94% susceptible), and clindamycin (MIC<sub>90</sub>, 16 μg/mL, 74% susceptible).

ERTAPENEM

Ertapenem is a newly licensed parenteral carbapenem that is highly resistant to inactivation by a wide variety of β-lactamases and possesses a broad spectrum of activity [6]. It is not as active as imipenem or meropenem against Pseudomonas aeruginosa. Although clinical studies have been done to establish the efficacy of ertapenem for the therapy of complicated intra-abdominal infections, these studies have, as yet, not been published. Several in vitro studies have supported the activity of ertapenem against anaerobes likely to be isolated from intra-abdominal infections. Goldstein et al. [6] studied ertapenem’s in vitro activity against 1001 anaerobes isolated from intra-abdominal infections from 29 sites in 17 countries worldwide. Ertapenem was active against all isolates, including all members of the B. fragilis group species, with the exception of 20% of Bilophila wadsworthia isolates, 3 isolates of lactobacilli, and 1 strain of Acidaminococcus fermentans. Snydman et al. [27] reported an overall resistance of ≤1% to ertapenem in the 2673 B. fragilis group isolates studied. They did note a clinically insignificant increase in the geometric mean MICs to ertapenem associated with 2.8% of B. distasonis strains being resistant. A recent study by our laboratory [31] on the in vitro activity of ertapenem against 469 less frequently identified anaerobes from 11 genera and 52 species isolated from human infections noted
Trovafloxacin. Trovafloxacin is an extended-spectrum fluoroquinolone with enhanced in vitro activity against anaerobic bacteria [34–36]. Although its use in the United States has been curtailed because of potential hepatotoxicity, it has been studied and used to treat intra-abdominal infections and other surgical indications such as prophylaxis. Citron and Appleman [34] studied trovafloxacin’s in vitro activity against 221 aerobic and 217 anaerobic intra-abdominal isolates, using supplemented Brucella agar, and found it to inhibit 99.3% of strains at ≤2 µg/mL. One strain each of E. coli, Staphylococcus haemolyticus, and a Fusobacterium sp. required >2 µg/mL for inhibition. Spangler et al. [35] used supplemented Wilkins-Chalgren agar and found trovafloxacin to be active in vitro against 487 (99.6%) of 489 anaerobes at ≤2 µg/mL except for 1 strain each of Prevotella bivia and Eubacterium lentum. Wexler et al. [36] found trovafloxacin to be active against 553 strains of anaerobic bacteria at ≤2 µg/mL and that 2 strains of B. distasonis and 2 strains of B. thetaiotaomicron required 4 µg/mL for inhibition. In a multicenter survey, Aldridge et al. [19] noted a 93% susceptibility (MIC90, 2 µg/mL) of 346 intra-abdominal isolates to trovafloxacin. However, Horn and Robson [37] studied 200 clinical isolates of the B. fragilis group found that trovafloxacin (MIC90, 4 µg/mL) was “not as highly active as in other reports” because of resistance in B. vulgatus strains. All of the 20 isolates of B. thetaiotaomicron they tested were susceptible to ≤4 µg/mL (MIC90, 2 µg/mL) of trovafloxacin. Betriu et al. [32] reported that 3.9% of 309 B. fragilis group strains were resistant to trovafloxacin (MIC90, 2 µg/mL), particularly B. uniformis (11%) and B. thetaiotaomicron (6%), whereas all B. distasonis and B. vulgatus strains were susceptible.

Onderdonk [38], using a mixed cecal inoculum, and Thadepalli et al. [39], using a combination of B. fragilis and E. coli, found comparable efficacy of trovafloxacin with that of clindamycin and gentamicin in the therapy of an experimental intra-abdominal abscess model. Stearne et al. [40] compared the efficacy of trovafloxacin in a murine subcutaneous model of mixed infection with B. fragilis and E. coli or B. fragilis and vancomycin-resistant Enterococcus faecium (VREF). They found that, although it was effective for the B. fragilis and E. coli combination, trovafloxacin was ineffective against the VREF, and, in that combination, it also had diminished killing activity of the B. fragilis. Stein et al. [41] studied the serum bactericidal activity of trovafloxacin and found it to have prolonged cidal activity against B. fragilis, C. perfringens, and Peptostreptococcus magnus but not, after the first sampling period, against B. thetaiotaomicron. This diminished cidal activity is suggestive that speciation of B. fragilis group species may have therapeutic implications for patients receiving fluoroquinolone therapy. After its US release, trovafloxacin was widely used in by surgeons but is now relegated to a restricted therapeutic category.

Moxifloxacin. Moxifloxacin is a new 8-methoxyquinolone that has been reported to have in vitro activity against anaerobic bacteria. In vitro studies [42–45] that have used a variety of methods and inocula, as well as a limited genus and species, have noted moxifloxacin to have similar activity as trovafloxacin against anaerobes. Aldrich and Ashcroft [42] found that 91% of 410 anaerobes tested were susceptible to ≤2 µg/mL of moxifloxacin. Bauernfeld [43] noted 20 strains of B. fragilis to have an MIC90 of 2 µg/mL. Elund et al. [44] reported an MIC90 of 1 µg/mL for 50 strains of B. fragilis and 50 strains of Peptostreptococcus and 0.5 µg/mL for 30 strains of C. perfringens. Kleinkauf et al. [45] noted moxifloxacin to have an MIC90 of 1 µg/mL against B. fragilis and Prevotella species. Horn and Robson [37] noted moxifloxacin to have relatively poor activity against 200 B. fragilis group species (MIC90, 8 µg/mL), especially against 22 strains of B. vulgatus (MIC90, 64 µg/mL). Clinical studies are under way to evaluate the efficacy of moxifloxacin in mixed aerobic/anaerobic intra-abdominal infections but have yet to be reported.

Gemifloxacin. Gemifloxacin mesylate is a new fluoronaphthyridone that has been evaluated in vitro against anaerobic bacteria [46–48]. With a presumptive proposed break point of 0.5 µg/mL, a recent review [46] noted gemifloxacin to have high potency against most gram-positive anaerobes such as C. perfringens and Peptostreptococcus spp. Gemifloxacin exhibited only moderate or variable activity against gram-negative anaerobes, with the exception of Fusobacterium nucleatum and Fusobacterium necrophorum. Many but not all B. fragilis strains were inhibited by <0.5 µg/mL of gemifloxacin, whereas isolates of B. thetaiotaomicron, B. distasonis, B. ovatus, and other members of the B. fragilis group species tended to be resistant. Species variability was seen in Prevotella species and Porphyromonas species. Kleinkauf et al. [45] also noted gemifloxacin to have “rather selective anaerobic activity,” with most Peptostreptococcus, Porphyromonas, and Fusobacterium strains susceptible to <0.5 µg/mL of gemifloxacin, whereas B. fragilis had an MIC90 of 4 µg/mL. Although gemifloxacin is currently under clinical investigation for pulmonary and other indications, on
the basis of its activity against B. fragilis, it is unlikely to be effective in the therapy of intra-abdominal infections.

**Desfluoroquinolones.** The desfluoroquinolones are a new group of agents under early development. One such agent is garenoxacin (BMS-284756) (T-3811ME)-(1-cyclopropyl-8-[di-fluoromethoxy]-7-[(1R)-1-methyl-2,3-dihydro-1H-5-isindol-yl]-4-oxo-1,4-dihydro-3-quinolincarboxylic acid methanesulfonate monohydrate), which is a new des-F(6)-quinolone that lacks the 6-position fluorine that characterizes the previous generation of fluoroquinolones. Preliminary data [49, 50] have indicated that this drug has a broad spectrum of antimicrobial activity against most gram-positive and gram-negative aerobes and anaerobes, including certain strains that are resistant to other fluoroquinolones. Studies have focused on more typical isolates, including intra-abdominal pathogens. Fung-Tomc et al. [49] noted 18 strains of B. fragilis to have an MIC<sub>90</sub> of 0.5 µg/mL to BMS-284756, which was the same as that of trovafloxacin and moxifloxacin. However, BMS-284756 also had an MIC<sub>90</sub> of 0.5 µg/mL against 11 strains of B. thetaiotaomicron, compared with 1 µg/mL for trovafloxacin, and 2 µg/mL for moxifloxacin. Hoellman et al. [50] studied the activity of BMS-284756 against 357 recently isolated anaerobes of human origin and found the MIC<sub>90</sub> and MIC<sub>99</sub> to be 0.5 and 2.0 µg/mL, respectively. BMS-284756 had an MIC<sub>90</sub> of 0.5 µg/mL against B. distasonis, 1 µg/mL against B. fragilis and B. thetaiotaomicron, and 2 µg/mL against B. vulgatus and B. ovatus. Only 13 strains required ≥4 µg/mL of BMS-284756 for inhibition: 1 strain each of B. thetaiotaomicron and B. distasonis, 2 of P. bivia, and 8 of Fusobacterium varium. One can expect other desquinolones to be developed by other companies.

Progress on the pathogenesis and therapy of mixed aerobic and anaerobic infections will proceed with the implementation of new molecular microbiological methods for isolation, identification, and susceptibility testing of isolates, as well as the planned and systematic development of new, potent therapeutic agents. It seems clear that specific, active therapy will be required to obtain a good clinical outcome. One can expect the twenty-first century to continue on the “anaerobe odyssey” that Dr. Finegold and his colleagues have led for the past 50 years.

**References**


