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Pharmacokinetics and Tissue Penetration of a Single 1,000-Milligram, Intravenous Dose of Metronidazole for Antibiotic Prophylaxis of Colorectal Surgery

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The levels of metronidazole in serum and tissue penetration of metronidazole were studied after prophylactic administration in 11 patients undergoing elective colorectal surgery. A single dose of 1,000 mg given intravenously was administered before surgery. Adequate drug levels in serum (≥MIC for 90% of strains tested [MIC_{90}] for *Bacteroides fragilis*) were found in all patients throughout the procedure. Mean peak (15-min) and last-determined (24-h) metronidazole levels in serum were 28.8 ± 8 and 4.2 ± 1.7 mg/liter, respectively. The β-phase elimination half-life was 9.5 ± 2.3 h, and the clearance and apparent volume of distribution were 57 ± 13 ml/min and 0.7 ± 0.1 liter/kg, respectively. In the colonic wall at surgical anastomosis, tissue metronidazole levels ≥MIC_{90} for *B. fragilis* were found in 91% of patients. In the abdominal wall fat and epiploic fat, tissue metronidazole levels ≥MIC_{90} for *B. fragilis* were found in 40 to 60% of patients at surgical incision and closure. No anaerobic infection occurred during the study.

The use of perioperative antimicrobial prophylaxis is widely accepted to prevent surgical infections in colorectal surgery (1, 8, 10, 20). The antimicrobial agents chosen should be directed against the pathogens most frequently involved in postoperative infections. Since *Bacteroides fragilis* and other anaerobic bacteria are among the main pathogens encountered in infections after colorectal surgery, antimicrobial agents with a spectrum of activity against these organisms are most often advocated (1). Nitroimidazole derivatives, such as metronidazole or ornidazole, are commonly used (1, 4, 7, 10, 11). During colorectal surgery, bacterial contamination occurs primarily in two sites: (i) soft tissues with the risk of abcess or cellulitis and (ii) the abdominal cavity with the risk of peritonitis or intraabdominal abscess. The effectiveness of prophylactic antimicrobial agents depends upon the achievement of adequate activity in the tissues involved by postoperative infection (2). Theoretically, antimicrobial activity should be maintained throughout the procedure, from abdominal incision to closure. For practical reasons, an ideal prophylactic regimen should be as easy as possible to administer, and a single-dose administration is often recommended (6).

The aim of this study was to evaluate whether a single 1,000-mg dose of metronidazole was able to obtain and maintain adequate concentrations (≥MIC_{90} for *B. fragilis*) in serum, abdominal wall fat, colonic wall, and epiploic fat. A recent multicenter susceptibility study demonstrated that the MIC for 90% of strains tested (MIC_{90}) for *B. fragilis* was 2 μg/ml (23).

MATERIALS AND METHODS

This study received the approval of the Ethical Committee of our institution, and all patients gave their informed consent. All patients were scheduled for elective rectal or colonic surgery. None had a history of hypersensitivity to metronidazole or its analogs. All patients had normal renal and hepatic function. At the beginning of the study, the patients underwent a 2-day bowel preparation consisting of a low-residue diet, purgation, and bowel washouts in the evening. Patients did not present any clinical or laboratory signs of infection and were not receiving any antibiotic therapy before surgery.

At the time of administration of anesthesia, patients were given a single dose of 1,000 mg of metronidazole intravenously (i.v.), administered over 30 min via an arm catheter, at a constant flow rate with an automatic pump. No other antimicrobial agent was administered to patients. Blood samples were collected from a central venous line (right internal jugular vein) before metronidazole injection and at 15 (peak concentration) and 45 min and 1, 2, 3, 4, 5, 6, 8, 10, 12, 16, 20, and 24 h (last-determined concentration) after the end of infusion. These samples were centrifuged and the serum was stored at −35°C until time of assay. Tissue samples were also collected from different sites during the surgical procedure: abdominal wall and epiploic fat samples at surgical incision and closure (end of surgical procedure), and colonic wall samples at surgical anastomosis. Simultaneously, additional blood samples were collected. Blood and tissue samples were also stored at −35°C until assay. Tissue samples were rinsed in sterile 10 mmol of morpholinepropanesulfonate (MOPS) buffer, pH 7.0, to eliminate excess blood within minutes of removal.

Determinations of metronidazole concentrations were performed by high-performance liquid chromatography with a Perkin-Elmer apparatus coupled to a spectrophotometric detector (LC 75). Metronidazole was kindly supplied by Specia, Paris, France. Serum samples were added to an equal volume of trichloroacetic acid (0.6 mol/liter), vortexed, and centrifuged (6 min at 3,000 rpm). Samples (20 μl) of the solution were injected onto the chromatography column. The A_{318} was measured. The column used was a

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Bondapak RP 18 reversed-phase column maintained in a temperature-regulated (25°C) oven (LC 100). The mobile phase consisted of acetonitril and KH_2PO_4 buffer, pH 3 (10/90, vol/vol). Tissue samples were ground at ice-cold temperature (0 to 2°C), and 500 μl of trichloroacetic acid (0.6 mol/liter) was added to 0.5 g of tissue. The solution was vortexed, and 2 ml of chloroform was added. After centrifugation (10 min at 3,000 rpm), 20-μl samples of the solution were injected onto the chromatography column. In this study, the lower limit of detection for metronidazole was 0.5 mg/liter for serum samples and 0.5 μg/g for tissue samples and the antibiotic recovery was 93%. Within-day and between-day repeatability was assayed for three different concentrations of metronidazole (5, 25, and 50 μg/ml) with coefficients of variation of 1.5, 1.9, and 0.2%, respectively, for within-day variation and 2.5, 2.5, and 2.4%, respectively, for between-day (3 days) repeatability assays.

Metronidazole concentrations in serum were plotted against time, and individual pharmacokinetic parameters were determined. A two-compartment model was fitted to metronidazole plasma concentration-time data, using a nonlinear least-squares regression program (APIS) (9). The predicted rate constant was used in the calculation of β-phase elimination half-life. Other pharmacokinetic calculations included the apparent volume of distribution and plasma clearance. The area under the plasma drug concentration-time curve from time zero to infinity was calculated by using the log trapezoidal method (24).

Evaluation of metronidazole penetration was performed as follows. For each patient, the ratios of drug concentrations in tissue and in serum were calculated by using metronidazole concentrations in tissue obtained at different periods during the surgical procedure (abdominal wall and epiploic fat concentrations were obtained during incision and closure, and colonic wall concentrations were obtained at anastomosis). Metronidazole concentrations in serum were obtained at the same time.

RESULTS

The patients in this study were 8 men and 3 women, with a mean age of 62 ± 11 years and a mean weight of 63 ± 10 kg.

The metronidazole levels in serum and pharmacokinetic parameters of the study patients were as follows: maximum level (at 15 min), 28.6 ± 8 mg/liter, with a range of 15.8 to 40.1 mg/liter; last-determined level (at 24 h), 4.2 ± 1.7 mg/liter, with a range of 1.7 to 7.7 mg/liter; β-phase elimination half-life, 10.3 ± 1.4 h, with a range of 6.4 to 12.7 h; total body clearance, 1.8 ± 0.16 liter/h · m^2, with a range of 1.0 to 2.8 liters/h · m^2; and apparent volume of distribution, 0.68 ± 0.11 liter/kg, with a range of 0.34 to 0.9 liter/kg. After 24 h, 9 of 11 patients had metronidazole last-determined levels ≥MIC_{90} for *B. fragilis* (2 mg/liter) (Fig. 1). A long plasma elimination half-life was observed in these surgical patients, with wide interindividual variations. The same observations were made regarding the total body clearance and apparent volume of distribution. The metronidazole serum and tissue levels obtained during the surgical procedures are shown in Table 1. At the time of incision (38 ± 7 min after the end of infusion), metronidazole levels were ≥2 μg/g of tissue in 7 of 11 patients for abdominal wall fat and 6 of 11 patients for epiploic fat. At surgical anastomosis (156 ± 40 min after the end of infusion), metronidazole levels ≥2 μg/g of tissue in the colonic wall were found in 10 of 11 patients. At closure (242 ± 70 min after the end of infusion), metronidazole levels were ≥2 μg/g of tissue in 5 of 11 of patients for abdominal wall fat and 4 of 11 patients for epiploic fat. Metronidazole tissue penetration estimated from antibiotic concentrations in tissue and in serum is presented in Table 1.

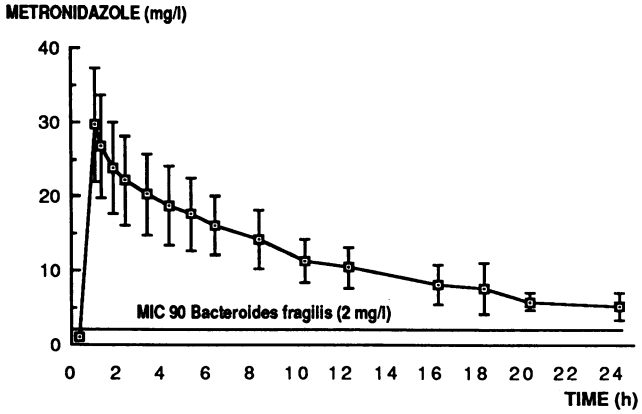

FIG. 1. Metronidazole levels in serum plotted against time. The means ± standard deviations are shown for each point.

One case of perineal aerobic infection (*Pseudomonas aeruginosa*) and one case of aerobic wound infection (*Klebsiella pneumoniae* and *Staphylococcus aureus*) were observed. No cases of anaerobic infection were noted in the study patients.

DISCUSSION

Previous studies have established the basic principles of antibiotic prophylaxis in surgical procedures (1, 10, 20), and the main points are as follows. (i) The antimicrobial agent must be present in the involved tissues before surgery allows bacterial contamination. (ii) The drug must attain and maintain a concentration in blood and in tissue high enough to inhibit the growth of contaminating pathogens (5, 20, 22).

In the present study, metronidazole concentrations in serum and different tissues were compared with the MIC_{90}

TABLE 1. Metronidazole penetration into tissue estimated from ratios of antibiotic concentrations in tissue and in serum at different points during surgery

Surgical procedure (mean time [min] ± SD)	Metronidazole concn (mean ± SD) in:				Ratio[a] (mean ± SD)		
	Serum (μg/ml)	Abdominal wall fat (μg/g)	Epiploic fat (μg/g)	Colonic wall (μg/g)	A/S	E/S	C/S
Incision (38 ± 7)	25.1 ± 7.2	2.6 ± 1.4	2.7 ± 2.2		0.1 ± 0.04	0.08 ± 0.04	
Anastomosis (156 ± 40)	19.1 ± 5.8			8.9 ± 3.7			0.42 ± 0.15
Closure (242 ± 70)	17.2 ± 5.7	2.4 ± 2.0	2.6 ± 1.8		0.13 ± 0.04	0.13 ± 0.07	

[a] Abbreviations: A, abdominal wall fat; S, serum; E, epiploic fat, C, colonic wall.

for *B. fragilis*, a pathogen frequently involved in postoperative infections after rectal and colonic surgery. A single i.v. dose of 1,000 mg of metronidazole was given to the patients, and after 24 hours, 10 of 11 patients had drug levels in serum of ≥2 μg/ml. This clearly indicates that adequate drug concentrations in serum were achieved throughout the surgical procedure. High concentrations were also achieved in the colonic wall, and 91% of patients had metronidazole levels ≥MIC_{90} for *B. fragilis*. In the abdominal wall and epiploic fat, lower concentrations were achieved and only 40 to 60% of patients had drug levels in tissue ≥MIC_{90} for *B. fragilis*. Similar drug concentrations in tissue were maintained throughout the surgical procedure and in the abdominal wall fat and epiploic fat. Thus, metronidazole elimination from tissue is a slow process, and relatively stable concentrations in tissue were observed during the surgical procedure. The penetration into fatty tissue relative to drug concentrations in serum was estimated to be from 10 to 13% in the study patients (Table 1). In another study performed under similar clinical conditions, ornidazole demonstrated a higher tissue penetration into fatty tissue (from 23 to 26%) and 84 to 100% of patients had ornidazole levels in tissue ≥MIC_{90} for *B. fragilis* (15). What constitutes optimal tissue levels of antibiotics is poorly understood (2). Many studies define this as a concentration above the MICs for the bacteria, but many examples of effective prophylaxis with antibiotic concentrations in tissue below the MIC and failures of prophylaxis with concentrations above the MIC exist (13, 18, 19). The role of prophylactic antimicrobial agents is to reduce the degree of bacterial contamination during surgery. As a result, it is not necessary to eliminate all bacterial pathogens. When subminimal concentrations to antibiotics are found in tissue, they can produce morphological alterations of bacteria. Thus, the bacterial growth rate can be decreased, which may favorably influence the outcome of infection (14, 25, 26). Clearly, many factors influence the control of operative infections: the discipline of the surgical team, meticulous surgical technique, proper preparation for surgery, and the status of the patient's immune system.

Metronidazole is metabolized by the liver, and the 1-(2-hydroxyethyl)-2-hydroxymethyl-5-nitroimidazole (hydroxy metabolite) has 65% of the bactericidal activity of metronidazole and an equivalent elimination half-life (3). The diffusion of the hydroxy metabolite was not evaluated in this study but might have contributed to the antianaerobic action of the prophylaxis.

Prophylaxis directed only against anaerobic pathogens has been recommended in colorectal surgery (7, 8, 12, 16). In the present study, two cases of infection related to aerobic pathogens were observed. Thus, the coadministration of a second antimicrobial agent active against aerobic bacteria should be considered in this type of surgery. Antibiotics from the cephalosporin or antistaphylococcal penicillin group should be considered in combination with metronidazole (16, 17).

No major modification was seen in metronidazole pharmacokinetics in the study patients. The elimination half-life and the volume of distribution was slightly higher than values observed in normal subjects. No major change in total body clearance was observed (21). In view of these results, the single dose used in this study (1,000 mg given i.v.) can be recommended and represents a simple and convenient technique of administration.

In conclusion, when metronidazole (1,000 mg given i.v.) is given before colorectal surgery, drug levels in blood and colonic wall of MIC_{90} for *B. fragilis* were achieved in most patients. In some patients, lower levels of metronidazole in the abdominal wall and epiploic fat were found. The clinical significance of these observations is not clear. Large groups of patients should be studied to assess the clinical efficacy of such an antimicrobial regimen.

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