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Stool cultures obtained before liver transplantation are useful for choice of perioperative antibiotic prophylaxis

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Abstract Bacterial infections, especially cholangitis, are still common complications after liver transplantation (LTx). During recent years, multiresistant enterococci have become a nosocomial problem in transplant units. The present prospective study on 26 patients, including 24 patients with chronic liver disease, demonstrated that enterococci were the predominant micro-organism involved in post-LTx bacterial infections. They were cultured in the feces and in other sites of 10 out of 13 (77 %) patients who underwent extensive examinations. Ampicillin-resistant *Enterococcus faecium* strains were isolated in urine or feces of 2 of the 13 patients prior to LTx. Similarly, resistance to ampicillin and gentamicin, the empirically used antibiotics for patients with fever of unknown origin, was found in *E. faecium* strains in 3 and 2 patients, respectively. Moreover, multiresistant *E. faecium* and *E. faecalis* strains were demonstrated in 46 % of the patients in the postoperative period (3 months). However, no vancomycin-resistant enterococci were isolated. The use of antibiotics within 4 months prior to LTx significantly increased the risk of developing ampicillin-resistant bacteria at the time of LTx and of infections with bacteria of enteric origin after LTx ($P = 0.03$ and 0.01, respectively). We conclude that stool and urine cultures performed prior to LTX may be useful for selecting prophylactic antibiotic regimens.

Key words Stool cultures, liver transplantation, antibiotic prophylaxis · Liver transplantation, stool cultures, antibiotic prophylaxis · Antibiotic prophylaxis, liver transplantation, stool cultures

Introduction

Liver transplantation (LTx) is considered to be the treatment of choice for a wide variety of acute and chronic hepatic disorders. During the last decade, more than 40,000 cadaveric liver grafts were transplanted in Europe and the United States [35]. The 1-year survival rate for liver recipients with nonmalignant indications for LTx has increased to over 80 % [38]. These encouraging figures result from advances in organ preservation, immunosuppression, surgical techniques, and better management of complications [29–31]. However, infections such as cholangitis, septicemia, and pneumonia occur in 30 %–35 % of all liver transplant recipients within the 1st year after LTx. This illustrates the need to achieve a balance between the iatrogenic immunosuppression required for survival of the graft and the risk of infectious complications [15, 29].

During the last years, the occurrence of multiresistant enterococci in LTx patients has emerged as a new nosocomial problem [15, 23, 24, 36]. In this prospective study, we investigated the potential role of enteric bacterial flora as a reservoir for enterococcal infections after LTx. The susceptibility of enteric bacteria to various antibiotics and the effect on the intestinal microflora of shortening the duration of antibiotic prophylaxis (ampicillin and cefotaxime) from 5 to 2 days were studied according to an intention to treat analysis.

Materials and methods

Patient demography

Twenty-six liver recipients, 24 of whom suffered from chronic liver disease, who had survived for a minimum of 3 months after LTx were consecutively included in a prospective study. All patients gave their informed consent prior to inclusion in the study. The study was approved by the Ethics Committee at Huddinge University Hospital. Two patient groups were selected to receive antibiotic prophylaxis for 5 or 2 days (Table 1). The groups were comparable with respect to gender, age, and underlying liver disease. The transplantation procedure was performed according to the same protocol [2, 30].

Immunosuppressive therapy

All patients received cyclosporin A (CyA, Sandoz, Basel, Switzerland) combined with azathioprine (AZA) and steroids as basic immunosuppressive therapy (Table 1) [2]. In seven patients, CyA was changed to FK506 (Tacrolimus, Fujisawa, Germany) within 37 days post-LTx due to steroid- and/or OKT3-resistant acute rejection. Episodes of acute rejection were treated with high doses of steroids and, if ineffective, with monoclonal antibodies against CD3 lymphocytes (OKT3; Ortho Pharmaceuticals, N.J., USA) for 7–10 days ($n = 5$) with or without a subsequent change from CyA to FK506.

Antibiotic prophylaxis

The antibiotics used as prophylaxis were the same for all patients, i.e., cefotaxime (Claforan, Hoechst, Frankfurt, Germany) and ampicillin (Doctacillin, Astra Läkemedel, Södertälje, Sweden), 1 g each, q 6 h, i.v. This regimen was used for prophylaxis against wound infections, pneumonia, and early contamination of the abdominal cavity and bile ducts with intestinal bacteria. Prophylaxis was given to 11 patients for 5 days (range 4–7 days) and to 15 patients for 2–3 days. In 5 of the latter patients, the prophylaxis was prolonged for 4–7 days because of perioperative risk factors for infectious complications after LTx. Such risk factors included suspected perioperative aspiration pneumonia, reoperation on day 2 post-LTx, perioperative culture of *Pseudomonas aeruginosa* in the bile, and pre-LTx cultures of hemolytic *Streptococci* from the nasopharynx and of 10^6 *Escherichia coli*/ml from the urine.

Nystatin (Mycostatin, Bristol-Myers Squibb, N.Y., USA) was given as prophylaxis against fungal infections, 500,000 units four times daily. It was started when the patient was placed on the waiting list and was given for 3–6 months after LTx [32].

Table 1 Baseline characteristics of patient population, which was divided into two groups and given short-term ($\leq$ 3 days) or long-term ($\geq$ 4–7 days) antibiotic prophylaxis after liver transplantation (*CCS* choledochocholedochostomy, *CJS* choledochojejunostomy)

Characteristics	Short-term prophylaxis ($n = 15$)	Long-term prophylaxis ($n = 11$)
Age in years (median)	38.3 (41.0)	35.0 (37.0)
Sex (M/F)	8/7	5/6
Underlying diagnosis		
Familial amyloidosis	3	3
Budd-Chiari syndrome	1	1
Alpha-1-antitrypsin deficiency	0	1
Primary biliary atresia	2	1
Primary biliary cirrhosis	1	1
Primary sclerosing cholangitis	2	1
Viral chronic active hepatitis	2	1
Hepatocellular carcinoma	1	0
Polycystic liver disease	1	0
Acute liver failure	2	2
Bile duct reconstruction (CCS/CJS)	11/4	9/2
Femoro-axillar venous bypass	12	10
Cold ischemic time in hours (mean)	10.77	10.25
(range)	7.60–13.58	6.63–13.26
Immunosuppression[a]		
CyA + AZA + S	11	6
CyA + AZA + S + OKT3	1	1
FK506 +/− AZA + S	1	3
FK506 +/− AZA + S + OKT3	2	1

[a] Cyclosporin A (CyA), 10 mg/kg per day orally or 3 mg/kg per day i.v. given in combination with azathioprine (AZA), 1.5 mg/kg per day i.v. followed by 1–2 mg/kg per day orally, and steroids (S, prednisolone), tapered from 200 mg to 20 mg/day over a period of 6 days. OKT3 (monoclonal anti-CD3 antibody), 5 mg/day given i.v. for 7–10 days. FK506 (tacrolimus), given in doses of 0.15 mg/kg per day i.v., followed by 0.3 mg/kg orally

The prophylaxis against the protozoal infection *Pneumocystis carinii* pneumonitis consisted of trimethoprim-sulfamethoxazole (Bactrim, F.Hoffmann-La Roche, Basel, Switzerland) in a low dose of 80 mg/400 mg daily for the first 6–12 months [7].

Routine prophylaxis against cytomegalovirus (CMV) infection was not given. However, pre-LTx CMV-seronegative patients receiving a liver graft from a CMV-seropositive donor were given ganciclovir (Cymevene, F.Hoffmann-La Roche, Basel, Switzerland), 5 mg/kg, q 12 h, i.v., for 8–14 days, followed by the oral administration of high-dose acyclovir (Zovirac, Glaxo Wellcome, London, England), 800 mg × 3–4 daily, according to kidney function, for 6–12 weeks post-LTx ($n = 7$).

Clinical criteria for infections

The criteria for the diagnosis of infections were based on the clinical findings and the results of laboratory analyses, as previously presented [2].

Septicemia was defined as clinical symptoms and bacterial growth in paired samples of blood cultures obtained during a single febrile episode (> 38.0 °C).

Cholangitis was diagnosed in a patient with fever (> 38.0 °C) and pathological liver function tests, together with histological signs of cholangitis in the liver biopsy specimen. Cytological evidence (granulocytes and/or intracellular bacteria) of infection in the exteriorized bile and isolation of the same micro-organism in bile and blood were used as additional criteria [17].

Bacterial pneumonia was considered when the following findings were present: an acute onset (developing within 12–24 h) of coughing, malaise, and fever (> 38.0 °C), a localized infiltrate on chest films, and positive bacterial cultures from either the nasopharynx, blood, or bronchoalveolar lavage (BAL) fluid.

Enteritis was defined as an episode of mild-to-moderate diarrhea with an infectious micro-organism or toxin found in the feces.

Pseudomembranous colitis due to *Clostridium difficile* was diagnosed when pseudomembranes or inflammation were visualized with endoscopy in a patient who had diarrhea and had been given antibiotics, combined with the finding of *C. difficile* toxins in the feces.

Urinary tract infection (UTI), in the absence of a catheter in the bladder, was defined as the culture of $\geq 10^4$ bacteria/ml on one occasion in a febrile patient. When a catheter was in situ, UTI was diagnosed on the basis of a growth of $\geq 10^5$ bacteria/ml, together with symptoms of pyrexia (> 38.0 °C) and/or leukocytosis and/or an increased acute phase protein (CRP). Asymptomatic UTI was diagnosed on the basis of a positive urine culture alone, as described above.

Samples and methods

Samples for bacterial cultures of feces and urine were collected for the first time when the patient was evaluated for LTx. These samples were repeated and a additional nasopharyngeal culture was taken before the operation and initiation of antibiotic prophylaxis. Cultures of feces, urine, and drainage fluids were performed weekly during the post-LTx stay in the hospital. If a T-tube or stent was used, bile was collected for 7–10 days, and graft function as well as signs of possible infection were monitored [16].

During febrile episodes (> 38.0 °C), two to three blood samples were drawn for bacterial and fungal cultures and for CMV DNA detection using the nested polymerase chain reaction (nPCR) and virus isolation in leukocytes [6]. A patient presenting with a dry cough, dyspnea, and fever was examined by bacterial and fungal blood cultures, blood gases, chest films, BAL, and bronchial brushing [12]. Urine cultures were also taken every week in the outpatient clinic of the Transplantation Unit.

Thirteen patients, seven of whom had reveived long-term (5-day) prophylaxis and six short-term (2- to 3-day) prophylaxis, were subjected to extensive studies of intestinal microflora; these involved analyses of enterococci and other enteric bacteria in feces, urine, bile, and blood, in addition to routine monitoring of bacterial, fungal, and viral infections. In the remaining 13 patients, the weekly urine and bile samples, and blood cultures when indicated, were analyzed in the Microbiological Laboratory of Huddinge University Hospital. The results of these cultures were included in the overall frequencies of the occurrence of various bacteria and episodes of infections.

Microbiological investigations

Urine cultures for bacteria were performed with the routine methods using quantitative cultures on blood and Cledagar plates (1 µl/agar plate) following incubation at 37 °C for 24 h.

Table 2 Type of bacterial infections related to the duration of prophylaxis in 26 patients 1–3 months after liver transplantation (*UTI* urinary tract infection)

Infection	Short-term prophylaxis ($n = 15$)	Long-term prophylaxis ($n = 11$)	Total
Septicemia	3	2	5
Cholangitis	3	3	6
Pneumonia	0	4	4
Pleuritis	0	2	2
UTI asymptomatic	0 (18)	3[a] (13)	3[a] (31)
UTI symptomatic	5[a]	5[a]	10[a]
Epididymitis	1	0	1
Wound	3	0	3
Enteritis	0	5[b], 2[a]	2[a] (4)
Total episodes	15[a] (33)	21[a] (34)	36[a] (67)
Total episodes/patients	1.0	2.1	1.4

[a] Bacterial infections treated with respect to the total number with positive cultures ( )
[b] *Clostridium difficile*

Feces cultures were performed using 1 g of feces homogenized with a mixer in a 9-ml phosphate buffer. The substrate was then serially diluted in 10-folds up to 10^7, and 0.1 ml of an appropriate dilution was streaked on various nonselective and selective media. The plates were incubated at 37 °C for 24 h. After incubation, the various types of colonies were counted, isolated in pure cultures, and identified using morphological, serological, and biochemical tests [11].

Testing of the susceptibility of bacteria in feces and urine was performed using the disk diffusion method (AB Biodisk, Solna, Sweden).

Statistics

Fisher's exact two-tailed test and Student's t-test were used. A P value below 0.05 was considered significant.

Results

Evaluation of the bacterial cultures and clinical data from the first 3 months after LTx showed that patients ($n = 15$) with the short-term antibiotic prophylaxis (2–3 days) did not have more episodes of bacterial infections than those receiving up to 7 days of prophylaxis ($n = 11$), i.e., a total of 15 vs 21 episodes (Table 2). In the 1st post-LTx month, 12 and 11 bacterial infections occurred in the short- and long-term prophylaxis groups, respectively, while during the 2nd and 3rd months the incidences were 3 and 10 in the same groups. Intestinal bacteria, especially enterococci, were the bacteria most frequently cultured in the urine, bile, and/or blood samples and concomitantly isolated in feces at various infectious epidsodes ($n = 30$): *E. faecium* ($n = 11$), *E. faecalis* ($n = 17$), and *E. durans* ($n = 2$; Table 3).

Table 3 Frequency of various bacteria cultured from urine, bile, and/or blood and concomitantly isolated in feces at various infectious episodes as related to the duration of prophylaxis in 26 patients 1–3 months after liver transplantation

Bacterial agent	Short-term prophylaxis $n = 15$	Long-term prophylaxis $n = 11$	Total
Enterococcus faecium	5	6	11
Enterococcus faecalis	9	8	17
Enterococcus durans	1	1	2
Streptococcus group C	0	2	2
Streptococcus group G	1	1	2
Alpha streptococci	2	1	3
Staphylococcus epidermidis	19	3	22
Escherechia coli	3	2	5
Enterobacter cloacae	5	7	12
Klebsiella pneumoniae	1	5	6
Pseudomonas aeruginosa	1	1	2
Stenotropomonas maltophilia	2	0	2
Clostridium difficile toxin B	0	4[a]	4[a]

[a] Detected only in feces

Post-transplant bacterial infections and risk factors

Eighteen asymptomatic and 5 symptomatic UTIs occurred in the short-term prophylaxis group compared to 13 and 5, respectively, in the long-term prophylaxis group (Table 2). Eight of 17 episodes of *E. faecalis* found in the urine cultures were associated with asymptomatic UTI. Cholangitis was the second most frequent event, occurring in three patients in each group. Septicemia developed in three patients with short- and in two patients with long-term prophylaxis. Pneumonia ($n = 4$) and pleuritis ($n = 2$) occurred only in patients who had been given long-term prophylaxis. Five patients, all of whom were intended to receive long-term prophylaxis, developed enteritis. *Clostridium difficile* toxin B was detected in the feces of these five patients. One patient had pseudomembranous colitis due to *C. difficile,* concomitantly with a rotavirus infection.

The administration of antibiotics within 4 months prior to LTx proved to be a risk factor for infections due to enteric bacteria after LTx ($P = 0.01$, Fisher's exact test). In contrast, the duration of perioperative antibiotic prophylaxis, choledochojejunostomy for the bile duct anastomosis, or treatment of steroid-resistant rejection with OKT3 and/or FK506 did not increase the risk of enteric bacterial infections post-LTx ($P = 1.00$, $P = 0.35$, $P = 1.00$, respectively, Fisher's exact test). We found no evidence that the duration of LTx, cold ischemic time, anhepatic time, venovenous bypass time, or number of units of erythrocytes transfused influenced the incidence of infections after LTx ($P = 0.56$, $P = 0.62$, $P = 0.61$, $P = 0.79$, $P = 0.76$, respectively, Fisher's exact test).

Co-occurrence of enteric bacteria at various sites

Enterococcus faecium ($n = 6$), *E. faecalis* ($n = 3$), *Escherechia coli* ($n = 1$), *Staphylococcus epidermidis* ($n = 1$), and *Streptococcus* group C ($n = 1$) were concomitantly isolated in the urine and feces of 7 of the 13 extensively studied patients. Furthermore, the same type of *E. faecium* and *Streptococcus* group C were detected in the feces, urine, and blood or bile obtained from one patient with recurrent episodes of cholangitis, pneumonia, and eventually septicemia. In contrast, various biotypes of *E. coli* and *E. faecium* were detected in the feces and urine of two other patients. In the remaining 3 of 13 patients, none of the bacteria cultured in feces was detected in samples obtained from other sites. *Klebsiella pneumoniae* was present in the urine and bile of one patient.

Susceptibility of enteric bacteria to antibiotics used for prophylaxis and factors involved in resistance

We analyzed the susceptibility of the enteric bacteria in the feces and/or urine to perioperative ampicillin prophylaxis. In total, ampicillin-resistant enteric bacteria – *E. faecium* strains ($n = 2$), *K. pneumoniae* ($n = 2$). *E. coli* ($n = 1$) – were present in pre-LTx urine and/or feces cultures of 5 out of 13 (38 %) patients.

Previous (≤ 4 months pre-LTx) antibiotic treatment, or immunosuppressive or cytostatic therapy, given to 5 of the 13 extensively studied patients, was found to be a risk factor for the development of ampicillin-resistant enteric bacteria ($P = 0.03$, Fisher's exact test; Table 4). Chronic liver failure, seen in 11 out of 13 patients, was not associated with the development of ampicillin resistance in enteric bacteria ($P = 0.48$), Fisher's exact test; Table 4).

Susceptibility of enteric bacteria to empirical treatment of fever and risk factors for developing resistance

The susceptibility of bacteria to the empirical antibiotic treatment used in patients with fever of unknown origin showed similar patterns. Enteric bacteria with resistance to ampicillin and/or gentamicin were found in urine, feces, bile, and/or blood cultures of 6 of the 13 patients (46 %) with fever: *E. faecium* strains ($n = 3$), *E. faecalis, K. pneumoniae* plus *Streptococcus* group C strains ($n = 2$ with resistant strains of each), and *E. coli* ($n = 1$). In 4 out of 13 patients, *E. faecium* strains developed increased resistance against ampicillin and/or gentamicin during the antibiotic treatment.

Antibiotic prophylaxis or treatment within 4 months prior to LTx did not seem to favor the development of enteric bacteria resistant to the empirical treatment of

Table 4 Analysis in 13 liver recipients of various factors that may favor the development of enteric bacterial resistance to antibiotics used prophylactically during liver transplantation (ampicillin), for empirical treatment of fever (ampicillin and gentamicin), or to several antibiotics

Risk factor	Antibiotic resistance		
	Prophylaxis (ampicillin)	Treatment of fever (ampicillin + gentamicin)	Multiresistance
Pre-LTx antibiotics	0.03*	0.10	0.10
Chronic liver disease	0.48	0.46	0.46
Antibiotic prophylaxis ≥ 3 days		1.00	1.00
Choledochojejunostomy		1.00	0.46
Duration of LTx		0.67	0.25
Cold ischemic time		0.58	0.29
Anhepatic time		0.86	0.82
Venovenous bypass time		0.44	0.17
Units erythrocytes transfused		0.64	0.22
OKT3 and/or FK506 treatment		1.00	0.59

* Antibiotics used pre-LTx increased the risk of developing ampicillin-resistant enteric bacteria ($P = 0.03$, Fisher's exact test)

fever with ampicillin (3/5 vs 2/8 patients, $P = 0.29$, Fisher's exact test) or gentamicin (2/5 vs 2/8 patients, $P = 1.00$, Fisher's exact test). Nor did the other possible risk factors analyzed increase the incidence or ampicillin- and/or gentamicin-resistant enteric bacteria (Table 4).

Multiresistant enteric bacteria

Multiresistant *E. faecium* ($n = 5$) and *E. faecalis* strains ($n = 2$) were detected in fecal and urine samples from 6 of the 13 liver recipients (46 %) pre-LTx or within 3 weeks post-LTx. Certain biotypes of these bacteria were susceptible only to gentamicin and vancomycin. However, no vancomycin-resistant enterococci were detected. None of the risk factors analyzed had a significant influence on the development of ampicillin- and/or gentamicin-resistant enteric bacteria in urine, feces, bile, or blood cultures of the 13 extensively studied liver recipients (Table 4).

Discussion

In this prospective study, we demonstrated that the intestinal flora was the reservoir of bacterial infections after LTx, and that enterococci were the most frequently isolated aerobic bacteria. Particularly alarming was the finding of multiresistant *E. faecium* and *E. faecalis* strains in 46 % of the liver recipients. The use of antibiotics before LTx proved to be a risk factor for increased incidence of post-LTx infections caused by the intestinal microflora and increased incidence of ampicillin-resistant enteric bacteria. There was a tendency towards a reduced incidence of bacterial infections in the short-term antibiotic prophylaxis group during the first 3 months after LTx. However, this did not reach statistical significance, probably due to the size of the study population. The incidence of infections seemed to decrease, in particular after the 1st month, in patients receiving the short-term antibiotic prophylaxis.

The incidence of bacterial infections in this investigation was comparable to that of previous reports. Approximately two-thirds of all liver recipients had at least one episode of bacterial infections during the first 3 months after LTx [4, 8, 15, 17, 26, 37, 38]. The high incidence of asymptomatic UTIs may be explained by bacterial colonization or contamination. Enteritis due to *Clostridium difficile* toxin B was associated with prior repeated antimicrobial treatments and the long-term antibiotic prophylaxis during LTx in all affected patients. One patient developed pseudomembranous colitis. Thus, it appears that excessive use of antibiotics may induce *C. difficile* enteritis [19]. Otherwise, cholangitis (23 %), septicemia (19 %), and pneumonia (15 %) were the predominant bacterial infections. These findings are in agreement with those of other studies showing incidences of 3 %–10 %, 15 %–35 %, and 7 %–15 %, respectively [4, 8, 10, 15, 17, 25, 26, 37, 38].

The central role of enterococci in bacterial cultures after LTx may depend on their occurrence in normal intestinal flora and on their increasing occurrence as a nosocomial pathogen [20, 21]. The pathological immune defense caused by chronic liver failure combined with enterococci colonization in the small bowel, bile, and urine may be contributing factors [14, 33]. Reduced Kupffer cell function of the engrafted liver due to ischemic preservation damage and early graft rejection reduce bile production. This enhances the likelihood of various intestinal bacteria invading the bile tree [3, 13]. Furthermore, thrombosis of the hepatic artery with possible ischemic damage of the bile duct epithelium, and bile duct anastomosis without an intact sphincter of Oddi in choledochojejunostomy, have previously been found in our patients and by others to be risk factors for infections and bacteremia due to the intestinal flora [2, 25, 37]. Other known risk factors, such as neutropenia, renal

insufficiency, and previous cytostatic or immunosuppressive therapy [21], are not unusual in LTx patients. Perhaps because of the small number of patients in this study, several of the factors investigated did not reach statistical significance as risk factors for the development of enteric post-LTx infections. A notable finding was that antibiotics often used before LTx for treatment and for prophylaxis, especially against spontaneous bacterial peritonitis, clearly increased the incidence of post-LTx infections caused by intestinal bacteria and the predominance of enterococci. This is in agreement with previous reports that an excessive use of antibiotics is associated with bacteremia caused by *E. faecium* and *E. faecalis* [21, 24].

The finding of multiresistant enterococci strains is alarming. Their occurrence in 46 % of patients already before LTx leads us to believe that resistance may develop during progression of the liver disease itself or that it may be due to antibiotics used to treat spontaneous bacterial peritonitis and other infections before transplantation. Indeed, exposure to antibiotics before LTx proved to be a significant risk factor for the development of ampicillin-resistant enteric bacteria. In contrast, cephalosporines are intrinsically resistant to enterococci. Colonization with enteric bacteria may also occur if patients have previously been admitted to the hospital [36]. In addition, the occurrence of other gastroenterological diseases obviously aggravates the balance of intestinal flora. Many patients with primary sclerosing cholangitis suffer from ulcerative colitis with recurrent episodes of bowel inflammation requiring treatment with steroids [34]. Patients with primary familial amyloidosis have polyneuropathy and gastroenteropathy, with variable bowel activity [28]. In this study, one patient in each of these disease categories was found to have multiresistant enterococci in feces before LTx. These factors ought to be considered when choosing the type of perioperative antibiotic prophylaxis and antimicrobial treatment following LTx.

The presence of *E. faecalis* strains with a high level of gentamicin resistance has proved to be predictive of the loss of synergy between a cell wall-active agent (e.g., penicillin, ampicillin, or vancomycin) and most aminoglycosides [39]. Enterococci with combined gentamicin and ampicillin resistance were also identified in our liver recipients. Furthermore, an increased resistance to these antibiotics was noticed in consecutive weekly feces cultures. The empirical choice of ampicillin and gentamicin for fever of unknown origin therefore seems to be questionable. It would be better to consider the resistance pattern of the intestinal bacteria in cultures when choosing the antibiotic treatment. Therapy with antimicrobial agents that lack enterococcal activity (e.g., cephalosporins and ciprofloxacin) favors bacterial overgrowth and colonization in the small intestine, which can lead to invasive bacterial and fungal infection [1, 22, 37]. This was clearly seen in one patient with fulminant septicemia after recurrent episodes of cholangitis due to multiresistant *E. faecium* and *E. faecalis*, which posed a risk for irreversible liver graft failure. Therefore, these antibiotics should be avoided as long-term prophylaxis against recurrent episodes of cholangitis without knowledge of the antibiotic susceptibility of the actual intestinal microflora. Piperacillin, its derivate with tazobactame and doxycillin, may offer a favorable choice. In addition, new synthetic streptogramin derivates, which are being tested in clinical trials, may prove of value in treating recurrent cholangitis due to multiresistant enterococci [27].

Vancomycin-resistant *E. faecium* has been reported in oncology and LTx patients [5, 9, 18, 23]. No such development of resistance was detected in the present study. Vancomycin should not be used as an empirical therapy for enteritis and septicemia. Such use increases multiresistance in intestinal bacteria. Moreover, it increases the likelihood of resistance by more virulent nosocomial pathogens, such as *Staphylococcus aureus*.

In conclusion, we have shown that antibiotics used before LTx significantly increased the incidence of infections and ampicillin resistance of enteric bacteria after LTx. We recommend monitoring the stool/rectal cultures of patients on the waiting list for LTx. This would facilitate the detection of the development of multiresistant bacteria that might cause recurrent infections. In addition, the use of insensitive antibiotics could be avoided.

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References

1. Antalek MD, Mylotte JM, Lesse AJ, Sellick JA Jr (1995) Clinical and molecular epidemiology of *Enterococcus faecalis* bacteremia, with special reference to strains with high-level resistance to gentamicin. Clin Infect Dis 20: 103–109
2. Barkholt LM, Ericzon B-G, Tollemar J, Malmborg A-S, Ehrnst A, Wilczek H, Andersson J (1993) Infections in human liver recipients: different patterns early and late after transplantation. Transpl Int 6: 77–84
3. Brettschneider L, Tong JL, Boose DS, Daloze PM, Smith GV, Huguet C, Blanchard H, Groth CG, Starzl TE (1968) Specific bacteriologic problems after orthotopic liver transplantation in dogs and pigs. Arch Surg 97: 313–322

4. Colonna JO, Winston DJ, Brill JE, Goldstein LI, Hoff MP, Hiatt JR, Quinones-Baldrich W, Ramming KP, Busuttil RW (1988) Infectious complications in liver transplantation. Arch Surg 123: 360–364
5. Edmond MB, Ober JF, Weinbaum DL, Pfaller MA, Hwang T, Sanford MD, Wenzel RP (1995) Vancomycin-resistant *Enterococcus faecium* bacteremia: risk factors for infection. Clin Infect Dis 20: 1126–1133
6. Ehrnst A, Barkholt L, Lewensohn-Fuchs I, Ljungman P, Teodosiu O, Staland Å, Ringdén O, Tydén G, Johansson B (1995) CMV PCR monitoring in leucocytes of transplant patients. Clin Diagn Virol 3: 139–153
7. Elinder CG, Andersson J, Bolinder G, Tydén G (1992) Effectiveness of low-dose cotrimoxazole prophylaxis against *Pneumocystis carinii* pneumonia after renal and/or pancreas transplantation. Transpl Int 5: 81–84
8. George DL, Arnow PM, Fox AS (1991) Bacterial infection as a complication of liver transplantation: epidemiology and risk factors. Rev Infect Dis 13: 387–396
9. Green M, Shlaes JH, Barbadora K, Shlaes DM (1995) Bacteremia due to vancomycin-dependent *Enterococcus faecium*. Clin Infect Dis 20: 712–714
10. Hadley S, Samore HM, Lewis WD, Jenkins RL, Karchmer AW, Hammer SM (1995) Major infectious complications after orthotopic liver transplantation and comparison of outcomes in patients receiving cyclosporin or FK506 as primary immunosuppression. Transplantation 59: 851–859
11. Heimdahl A, Nord CE (1979) Effect of phenoxymethylpenicillin and clindamycin on the oral, throat and faecal microflora of man. Scand J Infect Dis 11: 233–242
12. Heurlin N, Brattström C, Lönnqvist B, Westman L, Lidman C, Andersson J (1991) Aetiology of pulmonary disease in immunocompromised patients. Eur Respir J 4: 10–18
13. Howard TK, Klintmalm GBG, Cofer JB, Husberg BS, Goldstein RM, Gonwa TA (1990) The influence of preservation injury on rejection in the hepatic transplant recipients. Transplantation 49: 103–107
14. Johnson AP (1994) The pathogenicity of enterococci. J Antimicrob Chemother 33: 1083–1089
15. Kibbler CC (1995) Infections in liver transplantation: risk factors and strategies for prevention. J Hosp Infect 30 [Suppl]: 209–217
16. Kobota K, Ericzon B-G, Barkholt L, Reinholt FP (1992) The correlation between cytological pattern in bile and histological findings in liver transplantation. Transplantation 53: 791–795
17. Kusne S, Dummer JS, Sing N, Iwatsuki S, Makowka L, Esquivel C, Tzakis AF, Starzl TE, Ho M (1988) Infections after liver transplantation. An analysis of 101 consecutive cases. Medicine (Baltimore) 67: 132–143
18. Linden PK, Pasculle AW, Manez R, Kramer DJ, Fung JJ, Pinna AD, Kusne S (1996) Differences in outcomes for patients with bacteremia due to vancomycin-resistant *Enterococcus faecium* or vancomycin-susceptible *E. faecium*. Clin Infect Dis 22: 663–670
19. Lyerly DM (1988) Clostridium difficile: its disease and toxins. Clin Microbiol Rev 1: 1–18
20. Murray BE (1990) The life and times of the enterococcus. Clin Microbiol Rev 3: 46–65
21. Noskin GA, Peterson LR, Warren JR (1995) *Enterococcus faecium* and *Enterococcus faecalis* bacteremia: acquisition and outcome. Clin Infect Dis 20: 296–301
22. Pallares R, Pujol M, Pena C, Ariza J, Martin R, Gudiol F (1993) Cephalosporins as risk factor for nosocomial *Enterococcus faecalis* bacteremia: a matched case-control study. Arch Intern Med 153: 1581–1586
23. Papanicolaou GA, Meyers BR, Meyers J, Mendelson MH, Lou W, Emre S, Scheiner P, Miller C (1996) Nosocomial infections with vancomycin-resistant *Enterococcus faecium* in liver transplant recipients: risk factors for acquisition and mortality. Clin Infect Dis 23: 760–766
24. Patel R, Badley AD, Larson-Keller J, Harmsen WS, Ilstrup DM, Wiesner RH, Steers JL, Krom RAF, Portela D, Cockerill FR, Paya CV (1996) Relevance and risk factors of enterococcal bacteremia following liver transplantation. Transplantation 61: 1192–1197
25. Paya CV, Hermans PE, Washington JA II, Smith TF, Anhalt JP, Weisner RH, Krom RAF (1989) Incidence, distribution and outcome of episodes of infection in 100 orthotopic liver transplantations. Mayo Clin Proc 64: 555–564
26. Paya CV, Wiesner RH, Hermans PE, Larson-Keller JJ, Ilstrup DM, Krom RA, Rettke S, Smith TF (1993) Risk factors for cytomegalovirus and severe bacterial infections following liver transplantation. J Hepatol 18: 185–195
27. Pechére JC (1992) In vitro activity of RP5900 a semisynthetic streptogramin against staphylococci and streptococci. J Antimicrob Chemother 30 [Suppl A]: S15–18
28. Pepys MB (1988) Amyloidosis. In: Samter M, Talmage DW, Frank MM, Austen KF, Claman GN (eds) Immunological diseases, 4th edn, vol 1. Little Brown, Boston, pp 6731–6734
29. Rubin RH (1994) Infection in the organ transplant recipient. In: Rubin RH, Young LS (eds) Clinical approach to infection in the compromised host, 3rd edn. Plenum Medical, New York, pp 629–705
30. Starzl TE, Shunzaburo I, Esquivel CO, Todo S, Kam I, Lynch S, Gordon RD, Shaw BW Jr (1985) Refinements in the surgical techniques of liver transplantation. Semin Liver Dis 5: 349–356
31. Starzl TE, Shunzaburo I, Shaw BW, Gordon RD, Esquivel CO (1985) Immunosuppression and other nonsurgical factors in the improved results of liver transplantations. Semin Liver Dis 5: 334–343
32. Tollemar J, Ericzon B-G (1991) Invasive *Candida albicans* infection in orthotopic liver graft recipients. Incidence and risk factors. Clin Transplant 5: 306–312
33. Triger DR, Wright R (1985) Immunological aspects of liver disease. In: Wright R, Millward-Sadler GH, Alberti KGMM, Karran S (eds) Liver and biliary disease, 2nd edn. Bailliére Tindall, London, pp 215–231
34. Truelove SC, Watkinson G, Draper G (1962) Comparison of corticosteroids and sulphasalazine therapy in ulcerative colitis. BMJ 2: 1708–1711
35. United Network for Organ Sharing (1993) Annual report of the U.S. Registry for Transplant Recipients and the Organ Procurement and Transplantation Network. United Network for Organ Sharing, Richmond VA, and the Division of Organ Transplantation, Bureau of Health Resources Development, Health Resources and Services Administration, U.S. Department of Health and Human Services, Bethesda, MD
36. Wade JJ (1997) *Enterococcus faecium* in hospitals. Eur J Clin Microbiol Infect Dis 16: 113–119
37. Wade JJ, Rolando N, Hayllar K, Philpott-Howard J, Casewell MW, Williams R (1995) Bacterial and fungal infections in liver transplantation. An analysis of 284 patients. Hepatology 21: 1328–1336
38. Winston DJ, Emmanouilides C, Busuttil RW (1995) Infections in liver transplant recipients. Clin Infect Dis 21: 1077–1091
39. Zimmermann RA, Moeöeromg RC Jr, Weinberg AN (1971) Mechanism of resistance to antibiotic synergism in enterococci. J Bacteriol 105: 873–879