Laparoscopic Diagnosis and Treatment of Aortic Vascular Prosthetic Graft Infections in a Porcine Model

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Objectives. To study the feasibility and efficacy of experimental laparoscopy in the diagnosis of aortic graft infection in pigs.

Material and methods. Eight pigs had an aortic tube graft implanted and inoculated with either 5 × 10⁴ or 10⁶ CFU of Staphylococcus aureus ATCC 29213. Laparoscopy was performed after a median of 20 days with debridement and sampling for bacterial culture. Thereafter, the grafts were locally soaked in rifampicin and postoperatively, the pigs received rifampicin and ciprofloxacin orally for two weeks and were then sacrificed.

Results. All pigs developed graft infection. One pig died from severe clostridial septicaemia before laparoscopy could be performed. The remaining pigs had all samples for bacterial culture taken by laparoscopy from the inflamed tissue. The temperature dropped significantly after laparoscopy, and no macroscopic signs of infection presented at autopsy. However, only culture from one pig was without S. aureus at autopsy.

Conclusions. Laparoscopy is a potential diagnostic tool for aortic graft infection and also affords the opportunity to carry out bacteriological sampling and local antibiotic treatment. The efficacy of laparoscopic treatment needs further evaluation.

Introduction

Prosthetic vascular graft infection (PVGI) is one of the most serious complications in vascular surgery.¹,² The current frequency is 1–6%,¹,³ and PVGI is associated with a 12–27% mortality, considerable morbidity, and a 10–15% limb amputation rate in cases involving the infrarenal and femoral arteries.³ Any microorganism can infect a vascular graft, but Staphylococcus aureus is the most frequent pathogen, accounting for 24% to 50% of PVGI, depending on the implant site.¹,³,⁴,⁶

The clinical diagnosis of PVGI can be difficult, especially in the early postoperative period. Air around the graft on CT scans and a positive leukocyte scintigraphy is a common postoperative finding. However, delay in diagnosis and blind antibiotic treatment can lead to severe complications and worse outcome.¹,² The traditional treatments for PVGI are graft removal with extra-anatomic bypass or in situ graft replacement in the contaminated environments combined with months of systemic antibiotic treatment. The management of vascular diseases is developing towards minimal invasive treatments and laparoscopic vascular surgical techniques have been introduced during the last few years.⁷

The aim of this study was to develop an animal model for aortic graft infections, and evaluate whether early PVGI could be diagnosed and managed by laparoscopic debridement and local antibiotic treatment followed by systemic antibiotic treatment (Fig. 1).

Material and Methods

Animal model

Eight female pigs (59–70 kg) were anaesthetized by initially intramuscular injection of a 1 ml mixture consisting of zolamine 5 mg/ml, butorphanol 1 mg/ml, ketamine 10 mg/ml and xylazin 2 mg/ml, followed by continuous intravenous infusion of propofol 10 mg/kg per hour and fentanyl 0.025 mg/kg per hour throughout the operation. At the beginning of the anaesthesia all the pigs received 1.5 g cefuroxime IV. The pigs were placed on their back and the
retroperitoneum was exposed through a 25–30 cm long midline laparotomy under aseptic conditions. After intravenous administration of 5000 IU heparin, the aorta was clamped just below the left renal vein and just above the aortic bifurcation. The infrarenal aorta was replaced with an approximately 5 cm long and 8 mm wide Dacron graft by end-to-end anastomoses using 4-0 polypropylene sutures. The grafts were then contaminated with two different inoculates of *S. aureus*. The first four pigs got $5 \times 10^5$ colony forming units (CFU) *S. aureus* sprayed with a syringe on the graft and $1 \times 10^6$ CFU *S. aureus* was used for the last four. Hereafter, the retroperitoneum and abdominal wall were closed in layers with standard surgical techniques.

During the postoperative period, the pigs were monitored daily, including registration of food and water intake, behavior and measurement of rectal temperatures with an electrical thermometer.

**Bacterial strain**

The *S. aureus* strain ATCC 29213 used in this experiment has been used in several infected animal models, including vascular graft infection studies. The strain was susceptible to several antibiotics including methicillin, oxacillin, gentamicin, ciprofloxacin, rifampicin, vancomycin and cefoxitin, but resistant to penicillin.

**Laparoscopic operation**

Seven pigs underwent laparoscopy 15–26 days (median 20 days) after the initial bacterial inoculation. The pigs were anesthetized as mentioned before and positioned on their right side. A Veress needle was introduced into the abdominal cavity and a pneumoperitoneum was established with insufflation of carbon dioxide, and a pressure of 12–14 mm Hg was maintained. A 30-degree laparoscope was inserted using a 12 mm trocar 5 cm below the left costal margin. Two additional 12 mm trocars for surgical instruments were placed at the supraumbilical and the left paramedian line, respectively. If needed, a 5 mm trocar was positioned in the left flank for assisting instruments. With the right lateral decubitus position the intestine drops to the right side of the abdomen, and adhesions of the bowel with peritoneum and retroperitoneum could be released. The graft was approached retrocolic and circumferentially exposed. Swabs from surface of the graft and perigraft tissues were taken for bacteriological examination. Pus and necrotic tissues surrounding the graft were debrided. After repeatedly rinsing the area with sterile saline solution, 600 mg rifampicin in 10 ml saline solution was poured on the graft and neighbouring area through a silastic tube with a syringe, and the graft was soaked in the antibiotic solution for 10 minutes. Hereafter, the retroperitoneum was carefully closed with a running suture in a way to keep the rifampicin solution around the graft. The ports were removed and the sites were closed in a standard fashion (Fig. 2).

**Bacteriological sampling from graft and surroundings**

After laparoscopy, the animals received 300 mg rifampicin and 750 mg ciprofloxacin orally twice a day for...
two weeks, and were then sacrificed. The grafts were exposed through a midline laparotomy under aseptic conditions. Signs of graft infection were noted, including the status of perigraft tissue and exudation. A sample of the graft material, tissue around the graft and graft swabs were inoculated on Colombia agar overnight to recover S. aureus.

**Statistical analysis**

The individual mean temperature before and after laparoscopy was compared with Wilcoxon signed rank sum test for paired data. The software used was PEPI (http://www.sagebrushpress.com/PEPI.html). Statistical significance was assigned when p values were less than 0.05.

**Ethics**

The study was approved by the National Animal Research Inspection working for the Danish Ministry of Justice.

**Results**

Seven pigs achieved the planned procedures and survived to the scheduled autopsy. One pig (No. 5) died from clostridial septicaemia and had an autopsy under aseptic conditions the seventh day after initial implantation and inoculation.

Complete laparoscopy with debridement was technically feasible in all seven intended cases; 100% (95% C.I.: 65–100%). The median laparoscopic times were 120 minutes (range, 85–160 minutes), and the median blood loss was 150 ml (range, 50–300 ml). Intestinal perforations occurred twice at the beginning of insufflation due to pronounced peritoneal adhesions (Table 1), and were closed by suturing through the laparoscope.

All the pigs had reduction of behavior and food intake 3–7 days after the initial graft replacement and inoculation operation. Behavior and food intake returned to normal after laparoscopy and systemic antibiotic treatment. The animals No.1 to No. 4 had elevated temperatures 5–10 days before the laparoscopy and 0–4 days after. No. 6 to No. 8 had elevated temperatures 13–21 days before laparoscopy and 3–8 days after. The temperatures declined overall after laparoscopy and were significantly lower than before treatment in six of the 7 pigs (p < 0.05) (Table 2). The median difference of the mean temperature before

<table>
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<tr>
<th>Table 1. Characteristics at laparoscopy</th>
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<td>Pig No.</td>
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+: isolated; ++: multiple small adhesions; +++: extensive. Pig number 5 died before scheduled laparoscopy.

and after treatment was 0.5 degrees (95% C.I.: 0.26–0.71, P = 0.014). During the laparoscopy, all 7 grafts had signs of infection, such as perigraft exudates and necrotic tissues, but no macroscopic signs of infection were present at the autopsies (Table 2).

All the grafts were patent and no enteric erosion and anastomotic pseudoaneurysm were found at laparoscopy or autopsy.

S. aureus was cultured from the graft in 5 of 7 surviving pigs at laparoscopy (Table 3). Both grafts with sterile swabs were macroscopically infected, and one of them harboured S. aureus at later autopsy. Growth of S. aureus at autopsy was noticed in 5 of 7 pigs after 2 weeks antibiotic treatment. Consequently, S. aureus graft infection might have been cleared in 1 out of 6 cases diagnosed by positive culture (17%, 95% C.I.: 0.4–64%), and 2 out of 6 cases diagnosed by macroscopic findings at laparoscopy (33%, 95% C.I.: 6–74%).

**Discussion**

This study demonstrated for the first time that experimental aortic PVGI can be operatively and bacteriologically diagnosed by laparoscopy. Some cases of

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<th>Table 2. Mean body temperatures with standard deviations before and after laparoscopy</th>
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Pig number 5 died before laparoscopy. P < 0.05 is significant.
* The temperatures of 15–18 days before laparoscopy and after were collected and analyzed.
Table 3. Bacteriological and macroscopic findings at laparoscopy and autopsy

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<thead>
<tr>
<th>Pig No.</th>
<th>Graft replacement</th>
<th>At laparoscopy</th>
<th>At autopsy</th>
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<tbody>
<tr>
<td></td>
<td>S. aureus inoculation (CFU)</td>
<td>Clinical findings</td>
<td>Bacteriological findings from tissues &amp; swabs</td>
</tr>
<tr>
<td>1</td>
<td>$10^6$</td>
<td>Perigraft exudate + Peritoneal adhesion</td>
<td>No bacterial growth</td>
</tr>
<tr>
<td>2</td>
<td>$10^6$</td>
<td>Perigraft exudate, necrosis + Peritoneal adhesion</td>
<td>S. aureus</td>
</tr>
<tr>
<td>3</td>
<td>$10^6$</td>
<td>Perigraft exudate, pus, necrosis + Peritoneal adhesion</td>
<td>S. aureus</td>
</tr>
<tr>
<td>4</td>
<td>$10^6$</td>
<td>Perigraft exudates, necrosis + Peritoneal adhesion + ++ Growth of Enterobacteria &amp; Non haemolytic streptococci</td>
<td>No growth of S. aureus</td>
</tr>
<tr>
<td>5</td>
<td>$10^6$</td>
<td>Died before laparoscopy</td>
<td>–</td>
</tr>
<tr>
<td>6</td>
<td>$10^6$</td>
<td>Perigraft exudate, pus, necrosis + Peritoneal adhesion ++ (intestinal perforation)</td>
<td>S. aureus</td>
</tr>
<tr>
<td>7</td>
<td>$10^6$</td>
<td>Perigraft exudate, pus, necrosis + Peritoneal adhesion ++ (intestinal perforation)</td>
<td>S. aureus</td>
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<tr>
<td>8</td>
<td>$10^6$</td>
<td>Perigraft exudate, necrosis + Peritoneal adhesion</td>
<td>S. aureus</td>
</tr>
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* BL: before laparoscopy.

Aortic PVGI may be cured by laparoscopic debridement and local antibiotic treatment followed by systemic antibiotic treatment against a susceptible bacterial strain.

The aortic PVGI model in pigs worked quiet well and it seems that the two different quantities of S. aureus gave similar results despite the peroperative cephalosporin treatment.

Most vascular graft infections occur in the early postoperative period (<4 months) and are caused by bacteria such as S. aureus, coagulase negative staphylococci and Gram negative rods.\(^3,4\) Besides clinical observation, the common methods to diagnose aortic graft infection include computed tomography (CT), magnetic resonance imaging (MRI), ultrasound, radionuclide imaging, and surgical exploration. Having high sensitivity and specificity, CT and MRI should be the first examinations.\(^1,2,10\) However, diagnosis of PVGI including identification of the infecting bacteria can be difficult, especially in the early postoperative period. The reasons for this are that most signs and symptoms of intraabdominal graft infection are nonspecific, e.g. fever, elevated white blood cell count and/or CRP. The local alterations of infected perigraft regions are difficult to differentiate from normal changes seen during the first 3 months after graft implantation – even by CT scanning, MRI imaging and radionuclide scanning.\(^1,2,10,11\) If imaging techniques have not been able to provide definite evidence of the presence of graft infection and other sources of infection cannot be identified in a patient with sepsis, surgical exploration of the aortic graft may be performed.\(^12\) This is a true dilemma because it may be performed on seriously ill patients without PVGI.

Laparoscopic intervention is a minimally invasive procedure limiting the operative trauma and reducing the possibility of postoperative ileus, pain and hospitalization.\(^7,12,13,15\) Several articles have been reported on the feasibility of this technique for vascular reconstructions.\(^13–15\) In the present study we have demonstrated that the aortic PVGI diagnosis including bacteriological identification and susceptibility testing can be achieved by laparoscopy. However, the procedure is not without risk, as intestinal perforations occurred in two animals.

It has been reported that 53% of PVGI cases present within the initial thirty postoperative days and 70% within two months.\(^17,18\) However, early debridement, local irrigation with antibiotics, and systemic administration of appropriate antibiotics may make graft removal unnecessary owing to the fact that most patients have patent and intact grafts at this stage.\(^4,18\) Another possibility is antibiotic containment slow-release medium.\(^21\) Likewise, it has been demonstrated that some patients with aortic graft infection can be managed by conservative surgery combined with intensively local and systemic antibiotic treatment.\(^16,19\)

In our model, early aortic PVGI were produced successfully. All pigs’ temperatures except one declined significantly after treatment, and no
macrophocically infectious signs were observed during the autopsy. However, at autopsy the grafts were usually not sterile, which indicates that debridement with a single local antibiotic treatment followed by 14 days of systemic antibiotic treatment is insufficient to eliminate *S. aureus*.

Several studies have demonstrated that rifampicin is highly effective against most gram-positive bacteria in graft infection, especially *staphylococcus*. However, the emergence of resistance to rifampicin is fast when it is used alone. The best prevention of this problem is a combination with another antibiotic, e.g. ciprofloxacin, which has good therapeutic efficacy when it is used alone. The best prevention of this problem is a combination with another antibiotic, e.g. ciprofloxacin, which has good therapeutic efficacy in randomized trials.

In our study, the grafts were still infected after 2 weeks treatment, but not due to antibiotic resistant bacteria. The persisting infection may be as a result of the short treatment period. The period of antibiotic treatment could be prolonged — perhaps up to two months.

In conclusion, we found laparoscopy to be a potentially important diagnostic tool in early PVGI. The necessary lengths of prolonged systemic antibiotic treatment after laparoscopic debridement and local antibiotic treatment need to be studied further.

References


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